The Surveillance of Creutzfeldt-Jakob Disease in the United Kingdom, 1990 – 2006

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Declaration

I declare that this thesis is of my own composition, and that the research contained within it is my own original work. None of this work has been submitted in support of an application for any other degree or professional qualification.

Michelle Gillies

29th April 2011

Abbreviations

ACDP	Advisory Committee on Dangerous Pathogens
BSE	Bovine spongiform encephalopathy
CCDC	Consultant in Communicable Disease Control
CDC	Center for Disease Control (Atlanta, USA)
CJD	Creutzfeldt-Jakob Disease
CJD IP	CJD Incidents Panel
CMO	Chief Medical Officer
CNS	Central Nervous System
CSF	Cerebrospinal fluid
CT	Computed Tomography
CWD	Chronic Wasting Disease
DH	Department of Health
DWI	Diffusion weighted imaging
ECDC	European Centre for Disease Control
EEG	Electroencephalography
ELISA	Enzyme-linked immunosorbent assay
EU	European Union
FFI	Fatal Familial Insomnia
FLAIR	Fluid attenuated inversion recovery
FRIDA	Frontal Intermittent Rhythmic Delta Activity
FSE	Feline spongiform encephalopathy
gCJD	Genetic CJD
GSS	Gerstmann-Straussler-Scheinker Syndrome
hGH	Human growth hormone
hGnH	Human gonadotrophin hormone
HPA	Health Protection Agency
ICD	International classification of diseases
iCJD	Iatrogenic CJD
LRS	Lymphoreticular system
MBM	Meat and bone meal
MM	Methionine homozygote at *PRNP* Codon 129
MRC	Medical Research Council
MRI	Magnetic resonance imagine
MRM	Mechanically recovered meat
MV	Methionine heterozygote at *PRNP* Codon 129
NCJDSU	National CJD Surveillance Unit
NHS	National Health Service
NPC	National Prion Clinic
NPV	Negative predictive value
NSE	Neuronal specific enolase
ONS	Office of National Statistics
PD	Proton Density
PET	Positron emission tomography
PHS	Public heath surveillance
PIND	Progressive Intellectual and Neurological Deterioration study
PPV	Positive predictive value

PSPr	Protease-sensitive Prionopathy
PSWC	Periodic sharp wave complexes
SaBTO	Advisory Committee on the Safety of Blood, Tissues and Organs
SBO	Specified Bovine Offal Ban
SCIEH	Scottish Centre for Infection and Environmental Health
sCJD	Sporadic CJD
SEAC	Spongiform Encephalopathy Advisory Committee
SOP	Standard Operating Procedure
SPECT	Single photon emission computed tomography
TME	Transmissible mink encephalopathy
TMER	Transfusion Medicine Epidemiological Review study
TSE	Transmissible Spongiform Encephalopathy
vCJD	Variant CJD
UK	United Kingdom
UKBTS	UK Blood Transfusion Services
UKHCDO	UK Haemophilia Centre Doctor's Organisation Study
USA	United States of America
WHO	World Health Organisation
VV	Valine homozygote at *PRNP* Codon 129
95% CI	95% confidence interval

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The altruism of those affect by CJD in their willingness to participate in surveillance is truly remarkable. To all those who agreed to participate over the years I am grateful. I am equally grateful to those who collected the data; to the registrars and research nurses who came before me, and those that I was lucky enough to work alongside.

I dedicate this thesis to my family. To my parents, Neil and Mary Gillies, who have made endless sacrifices to provide me with the opportunities that they never had and whose unwavering love and support has allowed me to pursue those opportunities. Above all, they have shown me what is really important in life. To Pardeep, my soul-mate, and Talvin, my heartbeat, that I have neglected you to complete this thesis, I deeply regret. From now on the weekends are for jumping in muddy puddles. Elephant shoes.

Abstract

Prion diseases are rare, invariably fatal, neurodegenerative diseases, occurring in sporadic, genetic and iatrogenic forms in animals and humans, for which there is no acceptable diagnostic test in life and no effective treatment. In humans the commonest prion disease is Creutzfeldt-Jakob Disease (CJD). Systematic prospective public health surveillance (PHS) of CJD was initiated in the UK in 1990 in response to the detection of a novel prion disease in cattle, bovine spongiform encephalopathy (BSE). The aim of PHS was to detect any change in the clinico-pathological phenotype of CJD that could be attributable to human exposure to BSE. In this thesis I present a series of studies that evaluate various aspects of the PHS of CJD in the UK, 1990 - 2006.

From 1990 to 2006, 2154 suspect CJD cases were referred to the National CJD Surveillance Unit (NCJDSU); 57% had a clinical or neuropathological diagnosis of CJD. Sporadic CJD (sCJD) accounted for the majority of cases. Age adjusted sCJD incidence increased over time in association with an increasing use of CSF 14-3-3 protein for case classification. Genetic prion disease accounted for 9.4% of all cases; 54 iatrogenic CJD cases mediated by recognised routes of transmission were identified. Variant CJD (vCJD), a novel human prion disease, was characterised by the NCJDSU in 1996; by 2006 there had been 165 incident cases in the UK. The primary vCJD epidemic peaked in the UK in 2000 (27 incident cases) and has been in decline since. Secondary transmission of vCJD through the transfusion of labile blood components has been identified, occurring during an asymptomatic phase of illness. The prevalence of asymptomatic vCJD infection in the population is not known. In characterising vCJD and contributing to establishing an aetiological link with BSE, the NCJDSU met a primary aim of PHS. In an evaluation the NCJDSU was found to be flexible, acceptable, sensitive, timely and representative. Falling post mortem rates and an increasing reliance on clinical diagnostic criteria, with evidence of sub-optimal and differential use of investigations to support a diagnosis of sCJD and vCJD are of concern, as is the rising positive predictive value of the system in the face of falling referral rates. NCJDSU operational criteria for the assessment of EEGs for case classification in sCJD were prospectively validated. The sensitivity of EEG was low and specificity high; EEG remains a valuable non-invasive investigation in sCJD if

used in conjunction with other diagnostic tools. With the establishment of systematic prospective PHS the reliance on death certificates to ascertain suspect cases has diminished. The sensitivity and specificity of a death certificate diagnosis of prion disease in the UK are high. Death certificate data provide valid estimates of prion disease mortality in the UK.

Uncertainty around the epidemiology and pathogenesis of vCJD and the emergence of novel atypical prion diseases in animals which pose an as yet unknown threat to human health, provide the imperative to continue systematic prospective PHS of prion disease in humans in the UK for the foreseeable future. The NCJDSU is well placed to achieve this.

Chapter 1. Background to this thesis

Introduction

Prion diseases are a group of rare, invariably fatal, neurodegenerative diseases affecting animals and humans (Table 1). Aetiologically sporadic, genetic and iatrogenic forms of these diseases exist. Prion diseases are also known as Transmissible Spongiform Encephalopathies (TSEs) because of their transmission potential and associated neuropathological features. However not all prion disease are transmissible.(1) Therefore throughout this thesis I will refer to prion diseases rather than TSEs. In humans, the commonest form of prion disease is Creutzfeldt-Jakob Disease (CJD). Systematic prospective CJD surveillance was initiated in the UK in 1990 following the characterization of bovine spongiform encephalopathy (BSE), a novel prion disease in cattle, to which the UK population was widely and involuntarily exposed. The potential threat to human health posed by BSE was, at that time, unknown. In this thesis I report the findings of a number of studies that evaluate various aspects of the surveillance of CJD in the UK from 1990 through 2006.

This thesis is organised into six chapters. In this chapter I provide an overview of the current scientific knowledge in relation to prion diseases, explaining the rationale for the surveillance of prion diseases in humans, based on a literature search strategy. In the second chapter I describe the epidemiology of CJD according to disease subtype in the UK from 1990 through 2006 using data collected by the National CJD Surveillance Unit (NCJDSU). In chapter three, I present the findings of the first ever evaluation of the NCJDSU. A study to prospectively validate the NCJDSU operational criteria for the assessment of electroencephalography (EEG) in case classification of sporadic CJD (sCJD) is described in chapter four. Chapter five is devoted to an examination of the role of death certificates in the surveillance of prion diseases in the UK. Finally, in chapter six I present a general discussion of the findings from these studies, placing them in the context of forthcoming challenges to continued systematic prospective prion disease surveillance in the UK. Each chapter is organized in a standard format: a brief introduction, an outline the of aims and objectives of the chapter, a description of methods used, presentation of results, a bullet point summary of the key findings from the chapter followed by a discussion

and conclusions. Tables and figures are adjacent to the corresponding text. References are listed in a single reference list which precedes the appendices to this thesis.

Table 1 Prion diseases in animals and humans

Animal Prion Diseases
Bovine Spongiform Encephalopathy (BSE)
Chronic Wasting Disease (CWD)
Exotic ungulate spongiform encephalopathy
Feline spongiform encephalopathy (FSE)
Scrapie
Transmissible mink encephalopathy (TME)
Zoo primate spongiform encephalopathy
Human Prion Diseases
Creutzfeldt-Jakob Disease (CJD)
Fatal Familial Insomnia (FFI)
Gerstmann-Straussler-Scheinker Syndrome (GSS)
Kuru
Protease-sensitive Prionopathy (PSPr)
Sporadic Fatal Insomnia (SFI)

Literature review

This literature review comprises of five sections. The first describes the methodology of the literature review. The second provides an overview of prion diseases and their molecular basis. A brief account of prion diseases in animals, focusing on the key issues in relation to human health, follows. In the fourth section prion diseases in humans will be described. In the final section public heath surveillance (PHS) will be defined and the rationale for prion disease surveillance in humans will be explored.

Search strategy

Keyword search syntax was developed in MEDLINE using the following terms: '((Creutzfeldt Ja?ob) AND ('Disease' or 'Syndrome')) or 'CJD' or 'Prion Disease' or 'TSE' or 'Transmissible spongiform encephalopath$'. The strategy was translated, database specific subject headings added, and syntax run in the following electronic databases accessed via the OVID interface: MEDLINE, MEDLINE In- Process & Other Non-Indexed Citations, Cochrane Central Register of Controlled Trials, Cochrane Database of Systematic Reviews, Database of Abstracts of Reviews of Effects, EMBASE and PsycINFO. Articles were limited to those published in the English language; no other limits were set.

The grey literature was examined to identify further, potentially relevant resources. The following search terms were used: 'CJD' or 'TSE' or 'Prion Disease'. Specific resources accessed included: TRIP, ISI Web of Knowledge, National Research Register, Medical Research Council (MRC) Research Register, ReFeR (UK Department of Health Research Findings Electronic Register), Index of Conference Proceedings (accessed via the British Library's Public Catalogue), Dissertation Abstracts, ClinicalTrials.gov, and Intute. A number of additional resources were accessed as outlined in Appendix 1.

Reference lists of selected articles were reviewed and citation checks carried out to identify further potentially relevant studies. All retrieved citations were downloaded using bibliographic software into a database for management (Reference Manager 11). This search was regularly updated throughout the preparation of this thesis to identify critical gaps in the literature and inform the direction of this research. The

final search was carried out on 1st November 2010. In excess of 6,500 documents were retrieved. I screened the titles and abstracts of all documents to determine their relevance in relation to the scope of the literature review. The full text of all potentially relevant material was reviewed.

Prion diseases and their molecular basis

From the mid-1950s prion diseases were considered 'slow virus diseases' due to their long incubation periods and presumed viral aetiology. In the late 1960s the 'protein-only' hypothesis was proposed.(2;3) This signalled a paradigmatic shift in biological theory. The hypothesis suggested that the infectious agent was a self-replicating protein rather than a virus. This was supported by a number of observations, largely based on research conducted using the scrapie agent in sheep or animal models. The transmissible nature of scrapie had been demonstrated by Cuillé and Chelle in 1936.(4) Yet despite extensive research, a virus had not been isolated. The scrapie agent was resistant to ultraviolet and ionizing radiation that would modify any nucleic acid found in viruses.(5) Paradoxically it was sensitive to treatments that denatured proteins such as sodium hydrochloride and proteases.(6) In 1982 Stanley Prusiner and colleagues isolated an infectious fraction from a scrapie infected animal model.(6) The term 'Prion', derived from *pro*teinaceous and *in*fectious, was used to describe this agent. Although initially received with scepticism, Prusiner's theory gained popularity. In recognition of his work he was awarded the 1997 Nobel Prize in Physiology or Medicine.(7)

The current definition of a Prion is a

> "*Proteinatious infectious particle that lacks nucleic acid.*"(7)

According to the 'protein-only' hypothesis the prion protein can exist in a non-pathogenic conformation known as PrP^{c} (Figure 1a) and a pathogenic conformation known as PrP^{Sc} (Figure1b). Of note, PrP^{Sc}, the pathogenic conformation of PrP^{c}, may also be referred to as PrP^{TSE}. PrP^{Sc} is partially resistant to breakdown by proteases and the resistant fragment is designated PrP^{res}. The abnormal disease-related prion protein is typically detected as PrP^{res}; PrP^{res} and PrP^{Sc} are often used interchangeably in the prion disease literature, despite their different specific references. These

technical issues are not directly relevant to this thesis and will not be explored further. PrP^{Sc} will be used throughout this thesis to denote disease-related prion protein.

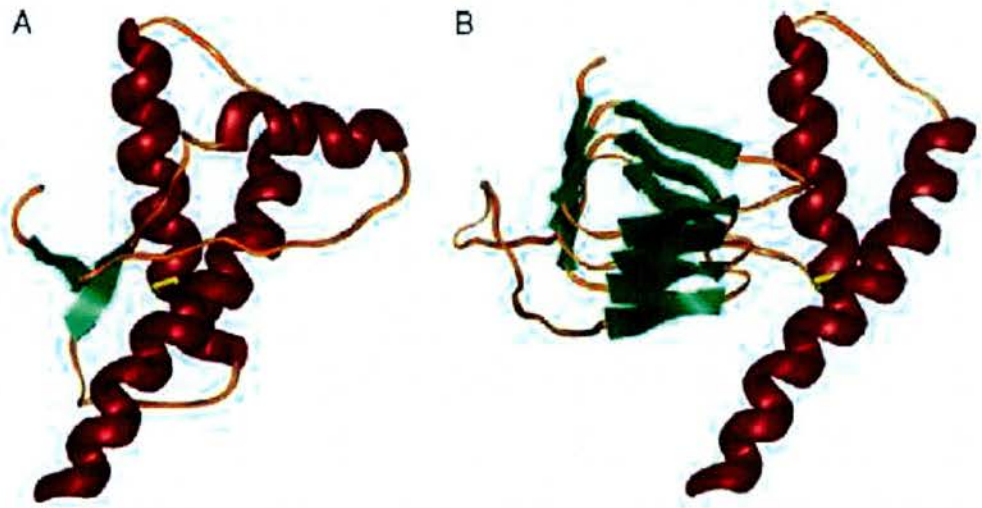

Figure 1 Illustration of the proposed non-pathogenic and pathogenic conformations of prion protein in humans (8)
(A) Non-pathogenic PrP^{c} containing mostly α-helical structure (red ribbons). (B) Pathogenic PrP^{Sc} containing most ß-pleated sheets (green arrows) with a small portion of α-helices (spiral shaped red ribbons).

PrP^{c} is a naturally occurring cellular glycoprotein with a predominantly α-helical structure weighing approximately 35 kD. It is covalently linked to cellular membranes via a glycosylphosphatidylinositol anchor. PrP^{c} is encoded by a single copy gene, named *PRNP*, which is located on the short arm of chromosome 20 (Figure 2).(7) The entire protein coding region is contained within one exon. PrP^{c} is expressed in most cells, but found in high concentrations in neurones. The precise biological function of PrP^{c} is not known although there is growing evidence to suggest a neuro-protective role.(9;10) Transgenic mice without *PRNP* are resistant to prion diseases, implying that PrP^{c} is essential for prion disease development.(11) However such mice appear to have normal life expectancy shedding little light on the normal function of PrP^{c}. PrP^{Sc} is thought to bind to the host PrP^{c} inducing self-replicating conformational change, from a predominantly α-helical structure into a structure consisting of predominantly ß-pleated sheets.(12) The process by which this occurs is not fully understood. In sporadic and genetic prion disease transformational change is thought to occur spontaneously; in acquired prion disease, in response to exposure to an exogenous agent. As a result of conformational change the biochemical properties of PrP^{c} differ

from PrP^{Sc}.(13) Although the change in prion protein conformation is a central molecular event, the actual neuropathogenesis of prion disease is not well understood.

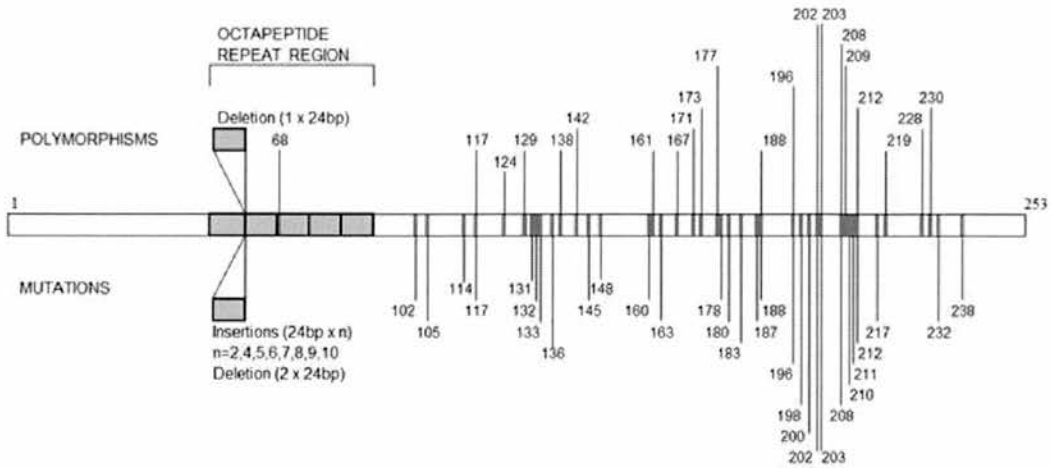

Figure 2 Human prion gene variation showing positions of pathogenic polymorphisms and pathogenic mutations (14)

Prion strains and prion protein typing

The demonstration that distinct clinico-pathological prion disease phenotypes can occur in one species, with distinct and stable experimental transmission properties, has led to the concept that multiple prion strains exist. These disease strains are most convincingly distinguished by their biological properties in living organisms (in terms of incubation periods and neuropathological profiles). In laboratory studies strains can be maintained through successive passages both within (for example from mouse to mouse model) and between (for example from sheep to mouse model) species, but transmission experiments are not the most convenient or rapid way of determining strain variation. Unfortunately, in the absence of a final characterisation of the prion, the molecular underpinning of agent strain variation is not yet understood and there is, therefore, no direct method of agent strain identification. It is hypothesised that the strain-specific characteristics of prion diseases are determined by the structural conformation of PrP^{Sc}. Different protein structures can be studied by differential responses to proteases and differential glycosylation patterns and this 'prion protein typing' has provided a molecular basis to characterise strain behaviour. Western blot has been used to describe different prion protein types according to the size of the proteolytic fragment (ranging from 19 to 21 kDa) and the degree of glycosylation that occurs following proteinase K digestion (the ratio of di, mono and unglycosylated

PrP^{Sc}). These prion protein types have been used as a means of identifying and differentiating different prion diseases (for example sCJD and vCJD) and are regarded by many as being surrogate markers for agent strain. The complexities of prion protein typing and its true relationship to agent strain are beyond the scope of this thesis and will not be discussed further.

The species barrier

The species barrier refers to a difficulty in transmitting an infectious disease between species. In prion disease, this can be illustrated using the example of scrapie, BSE and vCJD. Humans have been exposed to scrapie in sheep for several centuries. There is no evidence of transmission of scrapie from sheep to humans. However there is compelling evidence of transmission of BSE from cattle to humans, the same prion strain having been identified in BSE and vCJD.(15;16) To date all neuropathologically confirmed cases of vCJD in humans have occurred in individuals with a common polymorphic residue at Codon 129 (AG-methionine to GTG-valine, M129V) of *PRNP*, implying that the primary structure of PrP^{c} may be important in determining prion perpetuation. The species barrier has been used to explain the relative rarity of vCJD despite the widespread exposure of the UK population to BSE.(17) The species barrier may not however be absolute so as to prevent disease transmission. There is evidence to suggest that in prion disease, rather than preventing transmission, the species barrier may significantly lengthen the mean incubation period; if sufficiently lengthened disease may not become clinically apparent prior to the death of the infected host.(18;19) In prion disease, after two or three subsequent passages of the infectious agent within species, adaptation can occur and the incubation period may regress back to the previous mean. Prion strain is important in determining effective transmission between species leading some commentators to suggest that the term species barrier should be replaced with "transmission barrier".(17) A number of other factors are known to influence effective transmission including the distribution of tissue infectivity, the route of transmission and the infective dose.

Prion diseases in animals

Prion diseases are known to affect a number of mammals (Table 2).(20) To facilitate in vivo modelling prion diseases have also been transmitted to primates (non-human) and transgenically modified rodents, the latter in an attempt to overcome the species

or transmission barrier. In the section that follows I will briefly describe prion diseases in animals. This section will focus specifically on issues that are directly relevant to human health. Therefore a more detailed account of BSE, the only zoonotic animal prion disease will be provided, with an outline of the control measures initiated in the UK following the characterisation of BSE. In common, there are no definitive ante-mortem diagnostic tests for prion diseases in animals or humans. These diseases are universally fatal; no effective treatments are available.

Table 2 Prion diseases in animals

Year	Animal	Disease
1732	Sheep and goats	Scrapie
1947	Mink (farmed)	Transmissible mink encephalopathy (TME)
1967	Deer and Elk (farmed and wild)	Chronic Wasting Disease (CWD)
1986	Cattle	Bovine Spongiform Encephalopathy (BSE)
1986	Ungulates (nyala, gemsock, eland, large kudu, Arabian orxy, bison, ankole cow)	Exotic ungulate spongiform encephalopathy*
1990	Domestic cats and captive felidae (puma, cheetah, ocelot, tiger and lion)	Feline spongiform encephalopathy (FSE)*
1996	Captive non-human primates (rhesus monkeys and lemurs)	Zoo primate spongiform encephalopathy*

*prion strain indistinguishable from BSE

An overview of prion diseases in animals

Scrapie, the archetypal prion disease, has been reported in the UK since the 18th Century.(21) Much of the research that has informed our current understanding of prion diseases is based on studies of scrapie. Naturally occurring scrapie has been reported throughout Europe and North America. Despite extensive epidemiological investigation, the aetiology and mechanisms of transmission of scrapie are not fully understood. There is a similar paucity of data describing the aetiology and routes of transmission of CWD, a disease of wild and farmed deer and elk, largely confined to North America. Epidemiological studies of TME, which occurs in isolated epidemics among farmed mink again largely confined to the USA, suggest a food borne exposure, although transmission studies to support this are lacking. The animal prion diseases that have emerged in a range of species since 1990 are thought to be BSE related; in some the prion strain has been shown to be indistinguishable from

BSE.(22) BSE is the only animal prion disease that is known to pose a threat to human health.

BSE

In this section I will briefly describe the BSE epidemic and outline a chronology (to 2006) of events in relation to the control measures directly relevant to human health that were instigated in the UK following the detection of BSE. The clinical features of BSE are not directly relevant to this thesis and will not be described.

The epidemiology of BSE in the UK

BSE was described in the UK in 1986, although modelling suggests that the first cases occurred, undetected, in the South of England between 1977 and 1983.(23-25). The epidemic rapidly evolved reaching a peak of 36,680 cases per annum in 1992, equivalent to a rate of 6,636 cases per million bovines aged over 24 months (Figure 3).(26;27) The annual number of cases has consistently fallen since. As of 2009, approximately 184,600 cases of BSE had been confirmed in cattle in the UK.(27) Mathematical modelling suggests that between 1 and 3 million cattle may have been infected with many entering the food chain prior to the onset of clinical symptoms.(28)

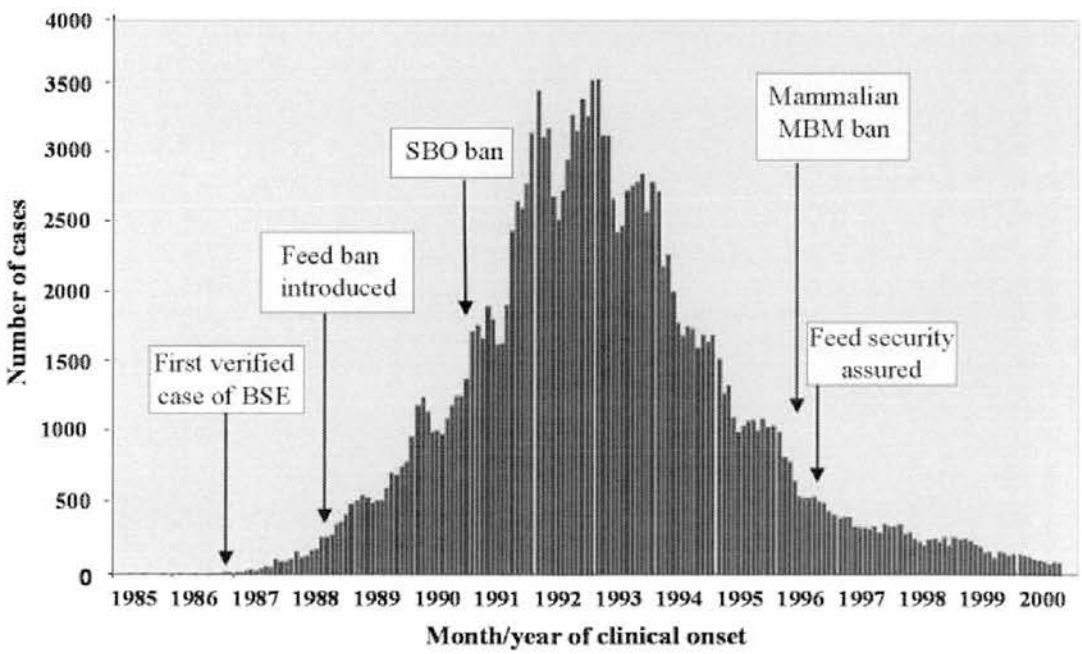

Figure 3 Time course of the BSE epidemic in the UK, 1986-2000, with dates of major precautionary interventions, adapted from Brown et al (29)

The mammalian ban on meat and bone meal in March 1996 extended a 1994 ban for farmed food animal species to include all mammalian species. SBO = specified bovine offal (brain, spinal cord, thymus, tonsil, spleen, and intestines from cattle >6 months of age); MBM = meat and bone meal (protein residue produced by rendering)

BSE worldwide

From 1989 onward BSE cases were reported in most European countries; countries outside Europe such as Japan and Canada were also affected.(27) Small epidemics or isolated cases have been reported in 24 countries. BSE may have been spread through the movement of infected animals between European states which would, in part, explain the scarcity of BSE in non-European countries. The Falkland Islands, Oman and the USA have reported isolated BSE cases in imported animals only. The export of BSE contaminated meat and bone meal (MBM) from the UK to be used in cattle feed may have introduced BSE to indigenous cattle. In countries where BSE has been reported in indigenous cattle, epidemics have differentially matured according to the timing of specific control measures (Figure 4).(30) For example the number of cases peaked in Switzerland in 1995, Portugal in 1999, France, Ireland, Germany and Belgium in 2001 and Spain in 2003. Outside the UK, the incidence of BSE has been greatest in Ireland, France and Portugal; France has the largest cattle population in the European Union (EU).

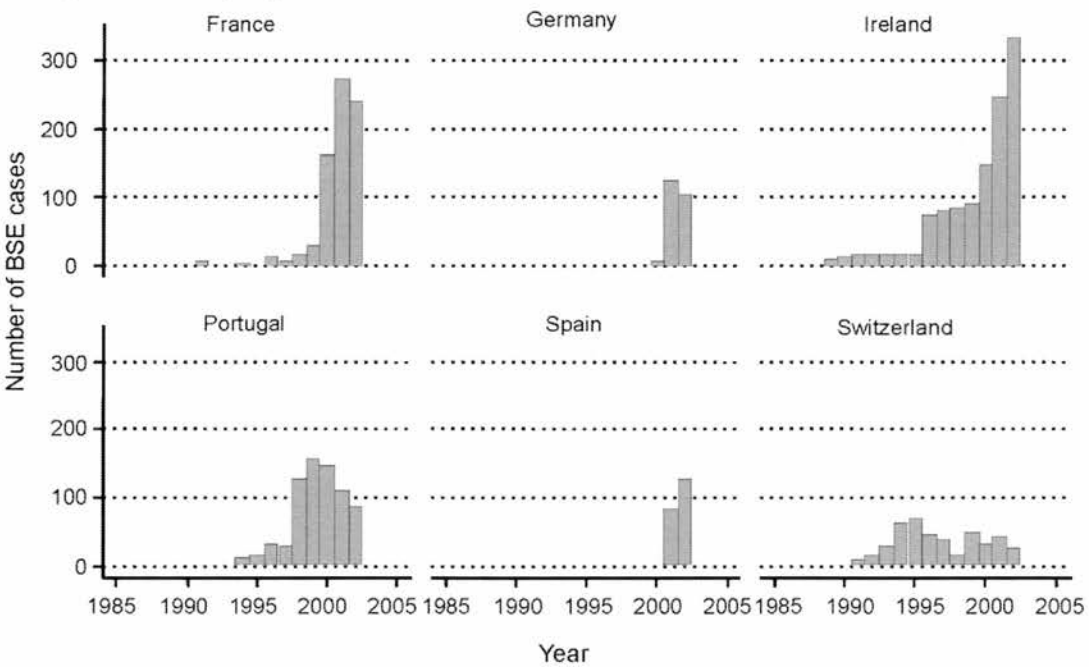

Figure 4 Number of BSE cases in selected European countries by year of onset, adapted from Smith and Bradley. (30)

The origin of BSE

The origin of BSE is unknown. Initially it was suggested that BSE was caused by direct transmission of scrapie, or a scrapie-like agent, from sheep to cattle.(31;32) This hypothesis was founded on a number of observations. Scrapie had been endemic

in the UK for several centuries. The UK had a high ratio of sheep to cattle. The neuropathological appearance of BSE was similar to that of scrapie. The practice of feeding meat and bone meal (MBM) including material from fallen sheep to calves as a protein supplement was common in the UK and could have facilitated oral inoculation of the disease. Critics note that the prion strain in BSE is distinct from all *known* strains of scrapie and that epidemiological or experimental evidence to support the direct transmission of scrapie from sheep to cattle is lacking.

Some commentators believe BSE originated in cattle. Until recently there was no compelling evidence to support this, although in the absence of disease surveillance sporadic BSE might not have been detected if occurring infrequently. In 2004 atypical forms of BSE were identified through active disease surveillance, which involves the testing of asymptomatic animals for disease. It has been hypothesised that sporadically occurring atypical BSE in cattle may be the origin of classical BSE. Other commentators believe that BSE arose as a result of a spontaneous mutation of *PRNP*. Indeed there is recent evidence to suggest that BSE is heritable.(33) Alternative theories regarding the origin of BSE are not currently supported by scientific research.(25) In 2004 SEAC, the Spongiform Encephalopathy Advisory Committee in the UK, stated it

> *"unlikely that the origins of BSE would ever be determined conclusively."*(34)

The origin of the BSE epidemic

Experimental and epidemiological research supports the theory that the BSE epidemic was propagated through the use of MBM in cattle feed. MBM is a high protein supplement that has been fed to cattle, sheep, pigs and chickens in the UK and used in agriculture as a plant fertilizer for several decades. MBM is produced by rendering waste products from several animal species including cattle and sheep. The rendering process separates protein from fat (tallow). The protein component is ground to produce MBM. Tallow is used in certain foods consumed by humans (for example gelatine) and animals (for example pet food). It is also used in pharmaceutical and cosmetic products. Changes in rendering processes in the UK in the decade prior to the emergence of BSE, largely driven by economic factors, resulted in the use of lower temperatures and less solvent.(31) Consequently the BSE agent, once introduced into the rendering process, would not have been inactivated and would

have entered the animal food chain though MBM. Whilst similar changes in the rendering process took place in Europe, albeit to a lesser extent, the UK was the first country to feed MBM to young calves; young calves appear to be more susceptible to BSE.(35) This theory is supported by the observation that an excess number of BSE cases were observed in dairy cattle relative to beef cattle. Dairy cattle in the UK were rapidly, 1 to 2 weeks following birth, weaned onto a diet of milk substitutes and MBM. Experimental studies have shown that less than 1gram of infected cow brain ingested orally is sufficient to transmit BSE to cattle; ½ gram to transmit the disease to sheep or goats.(25) Several cycles of BSE may have occurred before the disease was formally recognised. During this time infected cattle may have entered the human food chain or been recycled and re-entered the animal food chain as MBM, the latter amplifying BSE infectivity in MBM. It should be noted that other factors such as maternal transmission may have contributed to the epidemic, but at a much lower level insufficient to maintain the epidemic.

BSE control measure in the UK

An excellent and contemporary chronology of events in relation to BSE has been produced by The Department for the Environment Food and Rural Affairs.(36) Readers wishing a comprehensive review are directed to this publication. The salient events in relation to human health are summarized in the section that follows.

In 1987 the first case report of BSE appeared in the peer reviewed press. In June 1988 an expert group, the Southwood Working Party, met on the advice of the Chief Medical Officer (CMO). Their remit was to investigate BSE and examine the possible threat to human health posed by this novel prion disease. Shortly thereafter BSE became a notifiable disease. Rapid epidemiological studies implicated MBM in the propagation of BSE. A ban on incorporating ruminant proteins in ruminant feed was announced – the 'ruminant feed ban'. In August of 1988 the Southwood Working Party advised that animals showing signs of BSE should be slaughtered and destroyed (compulsory slaughter). Compensation was paid to farmers at a rate of 50% for confirmed BSE cases and 100% for slaughtered cattle that did not have BSE. This differential compensation scheme may have led to under-reporting of suspect cases and the further entry of infected cattle into the human food chain. In recognition of this full compensation was paid for all suspect cases slaughtered as of February 1990.

A further measure recommended by the Southwood Working Group was the destruction of milk from infected cattle. In December 1988 BSE was designated a zoonosis. The Southwood Report was published in February 1989. The Government accepted all of the Groups' recommendations.

Whilst the Southwood Working Group initially reported that BSE was unlikely to present a significant threat to humans, they recommended an expert committee be formed to advise on research in relation to BSE. The Tyrell Committee was formed in February 1989. In June 1989 the Specified Bovine Offal Ban (SBO) was introduced which banned the use of those categories of offal from cattle that were most likely to be infectious from use in human food based largely on existing knowledge of the pathogenesis and infectivity of scrapie. In 1990 the EU restricted the export of cattle from the UK to member states to cattle under six months old and banned the export of SBO containing material. Nevertheless UK companies continued to export SBO containing materials to outside the EU area. SEAC was formed in April 1900 to provide independent scientific advice on prion diseases in animals and humans. A National CJD Surveillance Unit (NCJDSU) was established the same year with the aim of detecting any change in the clinico-pathological phenotype of CJD in humans that might be attributable to exposure to BSE.

Two important events occurred in 1990. Firstly, it was demonstrated that BSE could be experimentally transmitted within (cattle to cattle transmission) and between species (to pigs via intra-cerebral inoculation and to mice via the oral route). Secondly, Feline Spongiform Encephalopathy (FSE), a novel prion disease affecting a domestic cat in the UK was described, with a prion strain indistinguishable from BSE. Following these events, the SBO ban was extended to all animal feed, including bird and pet food. Despite these control measures, infected cattle continued to be born after the ban in the UK in large numbers, suggesting on-going exposure. Cross-contamination of feed (a restriction on the use of MBM in feed for pigs and poultry was not recommended until March 1996) is thought to have sustained the epidemic and undermined the control measures that had been put in place. In 1992 the use of head meat after the skull had been opened was prohibited to prevent possible contamination from bovine brain; head meat being a common constituent of processed meat products such as pies and sausages. In 1995, The Ministry for

Agriculture, Farming and Food reported that some abattoirs were ignoring the SBO ban. Additional concerns were raised about the possible inclusion of material from the spinal cord in mechanically recovered meat (MRM). In 1995, a ban on the extraction of MRM from the spinal cord of cattle, prohibiting the use of MRM in food for human consumption, was introduced.

In March 1996 a clinico-pathologically distinct form of CJD, vCJD, was described.(37) Shortly thereafter SEAC announced a probable link between BSE and vCJD and the EU banned the export of British beef. More stringent control measures were instigated including a ban on the incorporation of all mammalian protein into animal feed introduced in August 1996 (introduced in EU states in 2001) – 'the Reinforced Feed Ban'. A small number of cases in animals born after this reinforced ban were described. Whilst some may be attributable to maternal transmission, the possibility of residual contamination of feed, or an alternate route of transmission unrelated to feeding, remained. The SBO ban was superseded by the 'Specified Risk Materials' ban which more specifically identified tissues of high infectivity that should be removed from healthy animals slaughtered for human consumption, based on experimental evidence. This included a stipulation that the whole head of cattle over 6 months of age should be treated as tissue of high infectivity. Shortly thereafter the heads of sheep and goats were removed from the food chain as a precautionary measure. In April 1996 the over 30 month rule was instated banning cattle over 30 months of age from the food chain (classical BSE is typically clinically apparent in cattle aged between 4 and 5 years old). These cattle were selectively culled and their carcasses incinerated. The over 30 month rule was superseded in 2005 by cohort and offspring culls in the UK in line with activities in the EU.

The transmission of BSE

In experimental studies BSE has been successfully transmitted to humanized transgenic mice and primates who develop clinical signs and symptoms consistent with vCJD. As previously noted the prion strain in BSE in cattle and vCJD in humans are similar indicating a likely causal association. The prion strain in FSE is indistinguishable from BSE; experimental studies have confirmed transmission of BSE to cats via the oral route. Neither pigs, nor chickens (both exposed to MBM feed) are susceptible to BSE via the oral route. However sheep orally inoculated with

BSE develop a scrapie-like illness. There are no recorded instances of BSE in sheep outside experimental studies. This would however present a realistic threat to human.

Atypical prion diseases: scrapie and BSE

Atypical cases of scrapie and BSE have been ascertained through active surveillance, a process of seeking out cases which may involve testing asymptomatic animals. These cases are clinically and pathologically distinct from classical scrapie and BSE. The epidemiology of these diseases has not been fully characterised due to the small number of cases detected to date, most of which have been detected in asymptomatic healthy animals slaughtered for human consumption, or fallen stock with few clinical signs. Molecular and transmission studies suggest that the prion strain in atypical disease is distinct from that in classical disease. The origin of atypical scrapie and BSE are unknown. Cases appear to be sporadic. It is not clear whether these are entirely novel prion diseases or previously unrecognised diseases which are now being detected through active surveillance. Interestingly in transmission studies an evolution toward the classical BSE strain occurs when transgenic mice are inoculated with atypical BSE. This has led some commentators to suggest that atypical BSE may have been the origin of the BSE epidemic.(38;39) It should be considered that if sporadically occurring atypical BSE was the origin of the BSE epidemic, eradication of BSE may not be possible although existing control measures should prevent a further epidemic in cattle. Of concern, the molecular subtype of atypical BSE has been shown to be similar to molecular subtypes described in sCJD in humans.(40;41) To date there have been no published accounts of the transmissibility of atypical BSE by oral inoculation. A comprehensive assessment of the risk to human health from these atypical prion diseases is not available, however in 1997 SEAC considered that in light of existing control measures for classical BSE,

> "*the risk of spread to other cattle, sheep and goats is likely to be very low, assuming as with classical BSE, environmental transmission is negligible.....the risk to human health is likely to be very low to negligible.*"(42)

The surveillance of prion diseases in animals

The principal aims of prion disease surveillance in animals are to monitor trends in the occurrence of disease and evaluate the effectiveness of control measures. The public health imperative for this relates to the threat to human health posed by BSE

and the as yet un-quantified threat to human health posed by atypical prion diseases in cattle, sheep and goats. As previously noted BSE-related prion diseases have been identified in a number of species. There is a theoretical risk of transmission of BSE to small ruminants. To date this has not been described outside laboratory studies. However in laboratory studies the pathogenesis of BSE in sheep differs from that in cattle such that it has been suggested that the current approaches to testing animals slaughtered for human consumption in the UK might not identify sheep asymptomatically infected with BSE.(43) If BSE were to emerge in sheep, this would pose a threat to human health. It should also be considered that novel animal prion disease may be identified through disease surveillance.

Disease surveillance may be passive or active. Passive surveillance involves the reporting of suspect (by definition symptomatic) cases to the authorities. Active surveillance involves seeking out cases. Passive surveillance of BSE was carried out from the point of recognition of the first case in 1986. This involved a process of farmers and/or veterinarians reporting suspect BSE cases to the authorities. In 1988 BSE was made a notifiable disease meaning that there was a legal requirement for owners to notify the authorities of a *suspect* case. In 1994 the laboratory diagnosis of prion disease in an animal of any species became notifiable. In 2001 under EU legislation it became a legal requirement to inform the authorities of *suspect* prion disease in any species of animal. Scrapie has been notifiable in the UK since 1993.

Active surveillance of BSE has been carried out in the UK since 1999 (2001 in the EU). In the UK active surveillance currently involves the testing of all fallen stock and cattle slaughtered for emergency (sick cattle) or entry into the food chain aged over 48 months, for BSE. Rapid post mortem diagnostic tests have allowed testing for BSE in animals fit for human consumption prior to entry into the human food chain. Active surveillance of scrapie has been carried out in the UK since 2002. Surveillance activities include annual sheep abattoir and fallen stock surveys; the former may identify asymptomatic disease and the latter symptomatic disease.

The rapid fall in the number of BSE cases 3 – 4 years after the institution of control measures in the UK (Figure 3) and the evolving epidemics elsewhere (Figure 4) lend epidemiological evidence to support the hypothesis that MBM was the vector for the

BSE epidemic. Control measures are however expensive to maintain. As the BSE epidemic draws to a close there will be increasing pressure to relax or remove these measures. On-going disease surveillance in animals will be crucial in monitoring the impact that such changes might have on trends in disease occurrence.

Prion diseases in humans

Despite the rarity of prion diseases in humans a number of issues have led to intense public, political and scientific interest in this area: (1) the lack of a practical and acceptable ante-mortem diagnostic test (2) lengthy incubation periods during which an individual may be infectious but asymptomatic (3) the transmissibility of the disease from human to human and animal to human during an asymptomatic stage (4) the resistance of prion proteins to decontamination (5) the lack of an effective treatment in the context of an invariably fatal illness. This section is organised into five major sub-headings. The first three will, in turn, examine sporadic, acquired and genetic prion diseases in humans (Table 3). The fourth, briefly reviewing therapeutic approaches to human prion diseases, applies to all human prion diseases irrespective of aetiology. A final sub-heading will review measures to reduce human to human transmission of prion diseases in the UK.

Table 3 Human prion diseases according to aetiology

Sporadic	
	Creutzfeldt-Jakob Disease (sCJD)
	Sporadic Fatal Insomnia
	Protease-Sensitive Prionopathy (PSPr)
Acquired	
	Kuru
	Variant Creutzfeldt-Jakob Disease (vCJD)
	Iatrogenic Creutzfeldt-Jakob Disease (iCJD)
Genetic	
	Genetic Creutzfeldt-Jakob Disease (gCJD)
	Fatal familial Insomnia (FFI)
	Gerstmann-Straussler-Scheinker Disease (GSS)

Diagnostic criteria for the classification of human prion diseases are outlined in Appendix 2 and will be referred to throughout this thesis.

Sporadic prion diseases

This section will focus on sCJD which accounts for up to 85% of all human prion diseases. Two further forms of sporadic prion disease in humans are recognised. These will be briefly described at the end of this section.

Creutzfeldt-Jakob Disease (CJD)

The term Creutzfeldt-Jakob Disease was first introduced in 1922 to describe five cases published in two separate case reports by German neuroscientists, HG Creutzfeldt and AM Jakob. Of the original cases described by Creutzfeldt and Jakob a retrospective analysis has shown that only two of five would have met the current diagnostic criteria for sCJD.

Epidemiology

Worldwide the incidence of sCJD is reported to vary from 0.44 to 1.61 per million persons per year.(44) Due to the rapid clinical course and universal fatality of sCJD, mortality rates are commonly used as a proxy measure for incidence. Most countries with mature surveillance systems report an increase in sCJD mortality over time.(44-47) This has largely been attributed to improved case ascertainment mediated through formalized disease surveillance, improved access to and use of diagnostic technologies and an increased awareness of prion diseases among the public and health care professionals.(44;45;48) As a notable exception, sCJD mortality rates in the USA have remained stable over time despite an increase in surveillance activity.(49) This raises the possibility, however remote, that the increase in mortality in other countries is attributable to increasing exposure to an unknown exogenous risk factor.

Studies from Europe, North America, Japan and Australia have consistently reported sCJD mortality peaking in individuals aged 60 – 79 years, and falling thereafter.(47;48;50-53) In 2005, Ladogana *et al* reported an age-specific sCJD mortality rate of less than 1 per million persons per year in those aged under 50 years, rising to 6–7 per million persons per year in the age group 70–79 years and falling to 2–5 per million persons per year in those aged 80 years and over, using pooled data from 11 international collaborators from the EUROCJD group.(44) Commentators have suggested that this finding may be explained by under-ascertainment of sCJD cases in the very elderly.(48) This hypothesis is supported by a recent increases in

age-specific mortality rates observed in association with increased surveillance activity.(52;54;55)

sCJD is reported to affect men and women in proportions consistent with the age and sex structure of the population under study. A recent study by Holman and co-workers reported no apparent sex difference in sCJD mortality rates in those aged under 60 years of age, but a slightly higher sCJD mortality rate in men relative to women aged over 60 years of age (49); a finding that has been duplicated elsewhere.(44) No socioeconomic gradient in sCJD has been demonstrated. However studies from the USA consistently report an excess of cases among white relative to black populations.(49) This may be due to ascertainment bias. Racial differences in access to health care have been documented. However racial differences in disease occurrence may also reflect genetic susceptibility/resistance factors. For example, Plaitakis *et al* reported an excess of cases of sCJD in Crete where the incidence rate was five times higher than expected.(56) The authors demonstrated a higher than expected proportion of the local population had a susceptible *PRNP* Codon 129 genotype (methionine homozygote).

Spatio-temporal clusters of sCJD have been described. In some cases investigation has revealed that the disease has a genetic rather than sporadic aetiology such as in Slovakia,(57) Chile,(58) Israel,(59) France,(60) Italy(61) and Japan.(62) In others, exhaustive investigation has failed to reveal an explanation for the higher than expected number of cases. Such clusters have been reported in France,(63;64) England,(65;66), Japan,(67) the USA (68) and Australia.(69;70) In the absence of an alternative explanation it has been proposed that enhanced surveillance in geographically defined areas may have led to apparent clustering.(70;71)

Risk factors

The cause of sCJD is not known. Putative risk factors have been investigated through case control studies conducted in America, Japan and Europe. Given the temporal and spatial distribution of cases some commentators have suggested that an environmental exposure is unlikely.(72) Studies to date have largely focused on risk factors that might indicate possible case to case transmission or zoonotic spread from animal to human. Case control studies examining putative risk factors for sCJD are summarised

in Table 4. Due to the rarity of sCJD, the multiplicity of putative risk factors and the potential latency period between exposure and disease development, case control studies are an appropriate epidemiological tool for exploring aetiology. However, there are a number of limitations to this study design that should be considered in interpreting these results. The most significant limitation is that case control studies are prone to bias, a systematic error in the design, analysis or reporting of a study that leads to incorrect conclusions being drawn. This may be information bias (systematic difference in the way that information on exposure and/or outcome is assessed between cases and controls) or selection bias (controls are not representative of cases with respect to all factors except outcome). Case control studies are always retrospective, both exposure and outcome have occurred at the time of data collection, therefore conclusions regarding temporality between an exposure and outcome cannot be drawn. Finally sCJD is a rare condition. Most studies have had small sample sizes and therefore limited statistical power. Consequently studies demonstrating an association between an exposure and sCJD often have wide confidence intervals reflecting uncertainty as to the true measure of association and can be difficult to interpret. A number of approaches have been taken to attempt to increase sample sizes and statistical power. Several studies recruited more than one control per case. It can be difficult recruiting additional controls particularly in studies employing a matched design whereby cases and controls are matched on key variables such as age, sex or residency. Alternative approaches include collaborative studies (73-75) and the pooling and re-analysis of data from published studies.(76) Due to the rarity of sCJD and the diverse geographical spread of cases, even in a comparatively small geographically area such as the UK, national case control studies are extremely time consuming and expensive to carry out; standardising methodologies across countries is extremely challenging.

Table 4 Summary of case control studies examining putative risk factors for sCJD

Setting	Study population	Data Collection	Factors associated with sCJD
USA (77) 1966-1973	38 Definite CJD 76 controls (38 relatives; 38 age & sex matched friends of case)	Structured interview	Eating raw seafood
Japan (78) 1975-1977	60 Definite or probable CJD 103 Age & sex matched controls (47 spouses; 56 neighbours)	Structured interview	Surgery within 5 years of onset; mechanical injury
USA (79) 1970-1981	26 Definite CJD 140 Age & sex matched controls (18 family;22 hospital controls)	Structured interview	Injury/surgery to head, neck or face; trauma to other body parts; ocular tonometry
UK (80) 1980-1984	92 Definite or probable CJD 184 hospital controls (92 neurological; 92 medical)	Structured interview	Herpes zoster in adult life; family history of dementia
USA(81) 1984-1995	636 underlying cause of death coded on death certificate as CJD 3180 controls, cause of death coded as non-neurological	Death certificates	Butcher or worked in office of a physician 9 other occupations association with CJD
Europe (74) 1993-1995	405 Definite or probable CJD 405 Age & sex matched hospital controls	Structured interview	Family history of dementia; eating raw meat; frequent exposure to leather products; exposure to fertilizer containing hoof and horn
Europe (75) 1993-1995	405 Definite or probable CJD 405 Age & sex matched hospital controls	Structured interview	No medical risk factors
Europe (73) 1993-1995	405 Definite or probable CJD 405 Age & sex matched hospital controls	Structured interview	Ear piercing in females; psychiatric visits; gynaecological procedures; 'other' operations†
Australia(82) 1970-1997	241 Definite or probable CJD 784 Age, sex and residency matched population controls	Structured interview	Any surgery; lived or worked on farm or market garden for >10 years

† any operation other than neurologic, eye, ear, gallbladder, gastrointestinal, and gynaecologic operations, tonsillectomy, and appendectomy
* Other surgery including stitches to skin, nose/throat, growth/cyst/mole removal, plastic surgery

Table 4 cont'd. Summary of case control studies examining putative risk factors for sCJD

Setting	Study population	Data Collection	Factors associated with sCJD
UK(83) 1990-2002	510 Definite or probable CJD 432 Age and sex matched controls (226 hospital; 106 population)	Structured interview	No association with ophthalmic surgery
UK (84) 1998-2006	431 Definite or probable CJD 454 Population controls matched by age cohort, sex and residency	Structured interview	Any surgery (lifetime); 'other' surgery (lifetime)*
UK (85) 1990-2006	857 Definite or probable CJD 454 Population controls matched by age cohort, sex and residency	Structured interview	No medical risk factors
Japan (86) 1999-2008	753 Definite or probable CJD 210 Age stratified hospital controls	Structured interview	No association blood transfusion or surgical procedures
Denmark & Sweden(87) 1987-2003	167 Definite or probable CJD 3059 controls (835 age, sex & residency matched; 2224 unmatched)	Hospital Discharge Data	Any major surgery within 20 years, including surgery on peripheral vessels, digestive system, spleen, female genital organs
Switzerland(88) 2001-2004	69 Definite or probable CJD 224 Age matched controls (69 general practice; 155 population)	Structured interview	Travel abroad; work in animal laboratory; invasive dental treatment; orthopaedic surgery; ophthalmological surgery (>1980); ate kidney
Germany(89) 1993-2005	685 Definite or probable CJD 659 Age & sex matched controls (434 hospital; 225 population)	Structured interview	Family history of dementia; ApoE4 allele frequency and carriage

† any operation other than neurologic, eye, ear, gallbladder, gastrointestinal, and gynaecologic operations, tonsillectomy, and appendectomy
* Other surgery including stitches to skin, nose/throat, growth/cyst/mole removal, plastic surgery

As can be seen in Table 4, very few putative risk factors are consistently associated with sCJD. Seven studies found an association between surgical intervention and sCJD, although the timing (from within 5 years of symptom onset to lifetime surgical history) and type of procedure ('any', head, neck, face, ophthalmological, orthopaedic, gynaecological, peripheral vascular, gastrointestinal) varied significantly.(73;78;79;82;84;87;88) These findings may in part be explained by bias (control selection and assessment of exposure). Two studies have however reported a dose-response effect which would provide additional evidence to support an association.(82;87) Whilst unrecognised contamination occurring during surgical procedures may represent a route of transmission of sCJD, due to the inherent limitations of these studies firm conclusions cannot be drawn. Indeed a recent re-analysis of data from six case-control studies reporting the relationship between sCJD and surgery stated that

> "*variation in the type of control subjects used and in exposure assessment in case-control studies may partially explain conflicting data regarding the association between surgery and CJD.*"(90)

Medical risk factors of trauma or physical injury were identified in two studies (78;79) although the sample sizes in these studies were small and confidence intervals wide. A subsequent pooled analysis of American, Japanese and UK studies found no association.(76) Positive family history of dementia in a first degree relative was reported more common in sCJD cases than controls in three studies.(74;80;89) Some of these studies have included genetic prion disease cases and the possibility of differing Codon 129 genotype distributions between cases and controls was not adequately explored.(74) More recently a comprehensive German study confirmed that sCJD cases were more likely than controls to report a family history of dementia; this could not be explained by the inclusion of genetic prion disease cases or the distribution of *PRNP* Codon 129 genotyping.(89) An excess of ApoE4 allele carriers was reported in sCJD cases with a family history of dementia, although the association between ApoE4 and sCJD is the subject of intense debate. It is noteworthy that family history of dementia was self-reported and may be subject to reporting bias as the relatives of individuals with sCJD may be more likely to recall and report a positive family history than healthy control subjects. There are isolated

reports of sCJD occurring in spousal couples,(91) siblings and co-workers,(92) but little evidence of case to case transmission through social contact.

Given interest in a possible aetiological association between animal prion diseases and sCJD, several studies have examined diet, exposure to animal products and occupation. Isolated associations with exposure to leather products, living or working on a farm or market garden, working in an animal laboratory or as a butcher and use of hoof and horn fertilizer have not been reproduced.(81) The study by Cocco *et al* using death certificate data to ascertain both cases and controls identified nine occupations in seven industries associated with sCJD in addition to working as a butcher and working in the office of a physician.(81) In the absence of a biologically plausible hypothesis for the relationship between these occupations and sCJD and a lack of supporting epidemiological evidence these associations are likely to be spurious associations that have arisen due to multiple comparisons. Many studies report inverse relationships between sCJD and various exposures. For example Zerr *et al* reported a reduced likelihood of sCJD cases reporting any surgery when compared to controls (odds ratio (OR) 0.68 (95%CI 0.48 – 0.98).(75) This is unlikely to represent a protective effect. The authors re-analysed their data according to control selection and found that hospital control reported exposure to medical and surgical interventions more frequently than non-hospital (population) controls thereby biasing their results toward the null.

Clinical features

Non-specific prodromal symptoms, such as weight loss, lethargy, disordered sleep, headache, depression or anxiety may be reported at onset. It is not clear whether these initial symptoms relate specifically to sCJD or are reported as a result of recall or reporting bias.(80) In most cases a rapidly progressive global dementia follows, although the clinical presentation may vary. Other recognised clinical presentations include a pure cerebellar onset (the Brownell-Oppenheimer variant) and cortical blindness (the Heidenhain variant). As the clinical picture progresses signs and symptoms reflect global neurological involvement. Dementia is present during the course of the clinical illness in 97% of cases, cerebellar signs in 87% and myoclonus in 81%.(93) Extra-pyramidal and pyramidal features are reported to occur in 74%

and 55% of cases respectively. Visual symptoms ranging from blurred vision, visual hallucinations and visual field defects to cortical blindness are reported in over half (58%) of all cases. Pain is rarely reported. Seizures occur in around 12% of cases, typically in the later stages of illness. Akinetic mutism dominates the terminal phase of illness. Most patients succumb to aspiration or dependant pneumonia a median of 5 months (mean 7.3 months) after symptom onset.(94) A survival advantage has been reported in association with a number of sociodemographic characteristics (female, young age at onset) and diagnostic features (*PRNP* Codon 129 heterozygosity, CSF 14-3-3 positivity and Prion Protein Type 2).(94)

Diagnostic criteria

Diagnostic criteria for sCJD were first proposed by Masters *et al* in 1979.(95) These included clinical features (progressive dementia and at least two of myoclonus, visual or cerebellar disturbance, pyramidal or extra-pyramidal dysfunction, or akinetic mutism) and typical electroencephalography (EEG) findings. In 2000 a cerebrospinal fluid (CSF) biomarker of neuronal injury, CSF 14-3-3 protein, was added to the clinical criteria.(96;97) The case definition of sCJD applied by the WHO for surveillance purposes at the time of data collection for this thesis is outlined in Appendix 2.(98) These criteria have a sensitivity and specificity of 92% and 71% respectively.(99) Recently, acknowledging the role of magnetic resonance imaging (MRI) in the diagnosis of sCJD, a change to the diagnostic criteria has been proposed.(99) The data collected in this thesis pre-date these developments therefore the published criteria referred to in Appendix 2 will be applied throughout this thesis.

Differential diagnoses

The differential diagnosis of sCJD is wide, encompassing a range of neurological and psychiatric conditions (Table 5). The most common differential diagnosis is Alzheimer's Disease following a rapid course.(1;100;101) In younger patients encephalitic processes are more commonly found. A less frequently, but potentially treatable differential diagnosis, is Hashimoto's encephalitis.

Table 5 Clinical differential diagnosis of sCJD, adapted from Zerr et al (93)

Frequent	*Rare*
Alzheimer's Disease	Parkinson's Disease
Lewy Body Dementia	Psychiatric disease
Inflammatory diseases of the CNS	Multi-system atrophy
Vascular / hypoxic encephalopathy	Frontotemporal Dementia
Corticobasalar Degeneration	Huntington Chorea
	Hashimoto's encephalopathy
	Paraneoplastic Encephalitis
	Lymphoma
	Intracerebral tumour or metastasis
	Wernicke-Korsakow syndrome
	Hydrocephalus

Investigations that support a diagnosis of sCJD

In the section that follows I will focus on investigations of proven value in sCJD. These include EEG, MRI, CSF 14-3-3 protein, *PRNP* Codon 129 genotyping, full sequencing of *PRNP* for mutations and neuropathological studies (ante-mortem brain biopsy and autopsy). Routine haematological and biochemical biomarkers are typically normal in all forms of prion disease.

Electroencephalogram (EEG)

The EEG was first recognised as being of diagnostic value in sCJD in 1954 (102) and incorporated into diagnostic criteria in 1979.(95) The characteristic features associated with sCJD on EEG are periodic sharp wave complexes (PWSC) (Figure 5). PSWC are usually generalized although lateralized or focal complexes are recognised. Lateralized complexes often evolve into typical bilateral PSWC. Criteria for the quantitative assessment of EEG in suspect sCJD were proposed by Steinhoff and Knight and adopted by the WHO for surveillance purposes (Table 6).(98) These criteria have not been prospectively validated in large scale studies and for several reasons the use of these criteria in the UK and elsewhere has been limited. For example in the UK there is significant variation in access to and use of digital

recording for EEGs. As a result the assessment of EEG for disease surveillance purposes remains largely subjective (personal communication R. Knight).

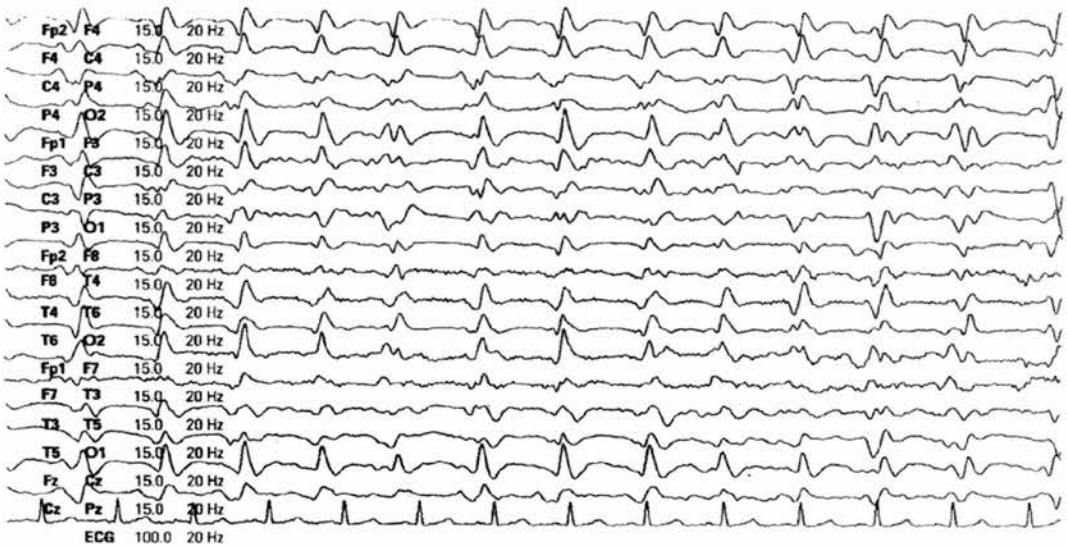

Figure 5 EEG recording showing a 'typical' EEG with generalised PSWC in a case of sCJD (14)

Table 6 Criteria for the quantitative assessment of EEG in suspect sCJD (98)

Strict periodic activity
- Variability of inter-complex intervals is <500 ms
- Continuous periodic activity for at least one 10-second period
Bi or tri-phasic morphology of periodic complexes
Duration of majority of complexes ranges from 100 to 600 ms
Periodic complexes can be generalized or lateralized but not regional or asynchronous

The EEG often evolves over the course of the clinical illness in sCJD.(103) In the early stages the EEG may show non-specific background slowing or FIRDA-like activity (Frontal Intermittent Rhythmic Delta Activity). Subsequent EEGs may show PSWC. In the advanced stages of disease PSWC reduce and may disappear. The timing of this investigation is therefore crucial. An EEG too early or too late in the course of the clinical illness may not detect typical findings; serial EEGs maximise the diagnostic yield. Approximately two thirds of sCJD cases develop a typical EEG at some point in the course of their clinical illness. In a series of 150 confirmed sCJD cases, Steinhoff *et al* reported that 64% developed a typical EEG, first recorded a

mean of 3.7 months (SD 3.1) after onset and recorded 2.3 months (SD 3.4) before death.(104)

The sensitivity, specificity and positive predictive value (PPV) of a typical EEG in sCJD are 58% – 66%, 74% – 91% and 93% - 95% respectively.(96;97;104;105) Much of the data on the diagnostic value of EEG in sCJD has been published by the German CJD Surveillance Unit. This group recently reported a fall in the sensitivity of EEG from 66% (1996 – 2000) to 32% (2001 – 2003).(106) The authors attributed this to the increasing use of CSF 14-3-3 protein which has led to suspect sCJD cases being referred to the surveillance unit at an earlier stage, prior to the onset of PSCW on EEG. Similar findings have been reported elsewhere in Europe.(107)

Collins *et al,* in a study of 2083 neuropathologically confirmed sCJD cases, demonstrated that the sensitivity of EEG varied according to molecular subtype.(97) In addition, the likelihood of a typical EEG increased with age and decreased with disease duration (disease duration <6 months more likely to have a typical EEG than duration > 6 months). The authors also showed that a typical EEG was more likely in the last, compared to the first, third of illness.

The EUROCJD group reported temporal trends in the use of investigations to support a diagnosis of definite or probable human prion diseases across 11 countries from 1993 through 20002.(107) EEG was more commonly undertaken than MRI or CSF 14-3-3 protein examination; up to 80% of definite or probable prion disease cases underwent EEG examination at some point in the courses of their clinical illness. However the trend was toward a fall in the number of cases undergoing EEG examination over time, mirrored by a rise in CSF 14-3-3 protein examinations. As a result, the annual proportion of patients meeting the WHO diagnostic criteria as a probable case of sCJD based on EEG and clinical features alone fell from 95% in 1993 to 3% in 2002.

PSWC are not pathognomonic of sCJD and have been demonstrated in patients with alternate diagnoses as outlined in Table 7.

Table 7 Differential diagnosis of PSWC-like pattern on EEG

Neurodegenerative	Alzheimer's Disease, Lewy Body Dementia
Vascular	Vascular Dementia, Acute Cerebral Thrombosis or Emboli,
Neoplastic	Cerebral Neoplasm
Inflammatory/	Encephalitis especially Herpes Simplex Encephalitis,
Infectious	Subacute Sclerosing Panencephalitis
	Multiple Cerebral Abscesses, AIDS Dementia
Metabolic	Hepatic encephalopathy, MELAS, hyperammonaemia, hypoxia,
	hyperparathyroidism, hypo and hypernatraemia, hypoglycaemia
Toxic	Baclofen, mianserin, metrizamide, lithium toxicity,
	phencyclidine (angel dust), ketamine, barbiturate
Other	vCJD (late stage)

One small study from the German CJD Surveillance Unit examined inter-observer variation in the reporting of EEGs using the objective criteria described in Table 6 to assess EEGs.(105) Two reviewers blindly assessed 68 EEGs from 29 suspect sCJD cases (15 sCJD cases and 14 non-cases diagnosed using the WHO clinical criteria that include assessment of EEG; of note the diagnosis was not neuropathologically confirmed in any of the non-cases). The authors report a kappa statistic of 0.95, indicating almost perfect agreement between reviewers, a sensitivity of 67% and specificity of 86%. In a follow up study Steinhoff and colleagues presented a comprehensive review of EEG data collected by the same unit from 1996 through 2000.(104) All EEGs in this study were objectively scored using the same criteria by a single reviewer. Sensitivity, specificity, positive and negative predictive values of 64%, 91%, 95% and 49% (without associated 95% confidence intervals) were reported. Intra-observer variation was not assessed in this series. Indeed, there are no published reports of intra-observer variation in the assessment of EEG, despite seminal studies, such as that by Steinhoff *et al*, utilizing a single reviewer to assess all EEGs.(104) A validation study of objective or subjective criteria for the evaluation of EEG for case classification in sCJD should include an assessment of both intra and inter-observer variation. This is a critical gap in the literature.

Given potential variation in subjective EEG reporting and the importance of EEG in the diagnostic criteria for sCJD, it is essential that as many EEGs as possible are

reviewed by PHS systems. Large studies reporting surveillance data, such as that by Collins *et al,* do not report the proportion of EEGs that were examined by a member of the surveillance team.(97) Nor do they provide information on the number of suspect cases referred to the PHS system that underwent EEG examination. Yet, these data are required to determine the completeness of case ascertainment by PHS systems.

Cerebrospinal fluid (CSF)

As with EEG, lumbar puncture examination to obtain cerebrospinal fluid (CSF) is an extremely common investigation undertaken in the investigation of subacute encephalopathy. In the appropriate clinical context, the detection of CSF 14-3-3 protein, a physiological cellular protein released in large quantities following neuronal injury, is a useful diagnostic test for sCJD (Figure 6). The detection of CSF 14-3-3 protein by immunoblot was incorporated into the WHO diagnostic criteria for sCJD in 2000. By 2000 the proportion of definite or probable sCJD cases undergoing CSF 14-3-3 protein testing was comparable to the proportion undergoing EEG (over 90%).(107)

The reported sensitivity and specificity of CSF 14-3-3 protein are between 43 - 100% and 84 - 100% respectively.(96;97;108-117) The sensitivity and specificity are known to vary according to a number of factors. Cases with younger age at onset and longer illness duration (>6 months) are more likely to have a negative examination.(97;109;110;118) Molecular subtype is an important determinant of positivity; CSF 14-3-3 protein has a higher sensitivity in MM1, MV1, VV1 and VV2 molecular subtypes than in the MM2 or MV2 subtypes.(97;109;110;116-118) Unlike EEG examination the timing of this investigation in relation to the clinical course of the disease does not influence the probability of a positive result.(97;118)

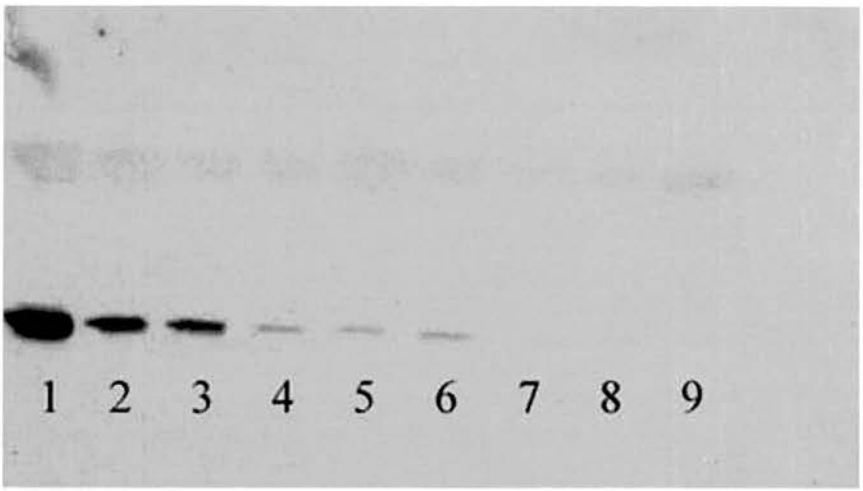

Figure 6 Western Blot of CSF 14-3-3 protein (14)
Lanes 1, 2, 3 are positive for CSF 14-3-3; Lanes 4, 5, 6 are weakly positive for CSF 14-3-3; Lanes 7, 8 and 9 are negative for CSF 14-3-3 Lanes 1 and 2 are from two patients with sCJD; Lanes 3 and 4 are from patients who have had a stroke; Lanes 5, 6, 7, 8 and 9 contain CSF samples from patients who do not have CJD. In the UK weakly positive CSF 14-3-3 results are not considered to be supportive of a diagnosis of sCJD.

Various studies have reported sensitivities using different methods of detecting CSF 14-3-3 protein. The most widely applied method is Western Blot (Figure 6), which requires a qualitative assessment of the result by an experienced biomedical scientist and produces sensitivities and specificities ranging from 84 - 100% and 60 – 100%.(96;97;109-115) Other quantitative methods including ELISA (Enzyme-linked immunosorbent assay), capture essay and sICMA (sandwich immunochemiluminometric assay) have slightly lower sensitivities and specificities. (116)

A positive CSF 14-3-3 protein test result has been reported in a number of conditions, although most are distinguishable clinically from sCJD (Table 8).

Other brain specific proteins have been explored as diagnostic tools in sCJD, either alone or in combination with CSF 14-3-3 protein, although as yet none have been incorporated into the diagnostic criteria. Tau, phosphorylated Tau and NSE (neuronal specific enolase) are markers of neuronal damage whilst S-100b is a marker of astrocytic gliosis. The most promising in sCJD is Tau which reportedly has a sensitivity and specificity approaching that of CSF 14-3-3 protein.(113;119;120)

Table 8 Differential diagnosis of a positive CSF 14-3-3 protein examination

Neurodegenerative	Alzheimer's Disease, Lewy Body Dementia, Frontotemporal Dementia, Corticobasilar Degeneration
Vascular	Vascular Dementia, Acute stroke including subarachnoid haemorrhage
Neoplastic	Carcinomatous Meningitis from small-cell lung cancer, Paraneoplastic Encephalopathy, Glioblastoma
Inflammatory/ Infectious	Viral encephalitis especially Herpes Simplex Encephalitis, Subacute Sclerosing Panencephalitis Multiple Cerebral Abscesses, AIDS Dementia
Metabolic	Hepatic encephalopathy, hyperammonaemia, hypoxia, hyperparathyroidism, hypo and hypernatraemia, hypoglycaemia, MELAS
Autoimmune	Hashimoto Encephalopathy
Toxic	Barbituates
Other	Post-ictal (epilepsy)

Other CSF abnormalities have been described in sCJD. In a European collaborative study of 450 neuropathologically confirmed sCJD cases, Green and co-workers describe an elevated total protein concentration of greater than 0.6 g/L as the most common abnormality of the CSF, affecting around 10% of sCJD cases.(121) The total protein concentration was greater than 1g/L in approximately 1%. The prevalence of oligocolonal bands in the CSF was 4.4%, which is less than the reported prevalence in the general population. A white cell count of greater than 5 cells/µl was extremely uncommon. The authors concluded that significantly elevated white cell counts and total protein concentrations would suggest a diagnosis other than sCJD.

Whilst an extremely valuable investigation, CSF 14-3-3 protein requires a lumbar puncture which is an invasive test. In addition to being expensive, CSF 14-3-3 protein assays require experienced and highly skilled biomedical scientists to perform analyses and interpret results. This investigation may therefore not be widely available in some countries. In the UK for example CSF 14-3-3 protein testing is available through the NCJDSU, not local or regional laboratories. Requests for this investigation are received from across the UK and the service is provided

free at the point of access. Centralisation of the service ensures quality control and location of the service within the PHS unit ensures rapid recognition of suspect cases of sCJD. The UK CSF 14-3-3 protein service also acts as a WHO reference centre and processes samples from countries which do not readily have access to this resource.

Magnetic Resonance Imaging (MRI)

Until recently the principal value of neuroimaging in sCJD was to exclude a potentially treatable differential diagnosis. Most suspect sCJD cases undergo a computed tomography (CT) scan of the brain. CT is usually normal, although in the late stages of disease (6 months or more after symptom onset) cerebral atrophy is seen in up to 20% of cases.(122) Decreased metabolism and changes in cortical blood flow have been reported in small case series using positron emission tomography (PET) and single photon emission computed tomography (SPECT) respectively. These findings have not been validated in large scale prospective studies and the diagnostic utility of such imaging modalities is unclear. There is however significant interest in the use of SPECT scanning in certain molecular subtypes of sCJD in which MRI is of limited value, for example the MM2 subtype.(123)

MRI is the neuroimaging modality of choice in CJD, irrespective of aetiological subtype. A number of MRI sequences are of value in sCJD. Initially T2-weighted and proton density (PD) weighted images were favoured.(124;125) These have largely been superseded by the more sensitive sequences of fluid attenuated inversion recovery (FLAIR) and diffusion weighted imaging (DWI).(99;106;126;127) DWI is considered the most sensitive of these sequences, followed by FLAIR and T2 weighted imaging.(127;128)

Characteristic findings on MRI include bilateral hyper-intense signal in the putamen and caudate (although asymmetrical involvement of the corpus striatum can occur) and so called 'cortical ribboning' denoting high signal in the cerebral cortex (Figure 7). (99;124;126;129) Occasionally features suggestive of the 'pulvinar sign,' hyper-intensity of the pulvinar nucleus relative to the anterior putamen, have been reported.

A recent study by Zerr *et al* reported the greatest diagnostic accuracy of MRI (in the appropriate clinical context) when high signal was detected in both the caudate nucleus and putamen, or in two of the following cortical regions: parietal, temporal or occipital.(99) Over 80% of definite or probable sCJD cases had evidence of these changes on FLAIR or DWI imaging in this European collaborative study. In an earlier study European collaboration examining 1063 neuropathologically confirmed sCJD cases, just over a third had evidence of basal ganglia high signal on MRI scanning; this study did not however consider cortical high signal, only basal ganglia changes.(97) In a small cases series (n=26) Shiga and co-workers reported cortical high signal only in 42% of definite or probable sCJD cases, basal ganglia high signal only in 13% and high signal in both in 46%.(126)

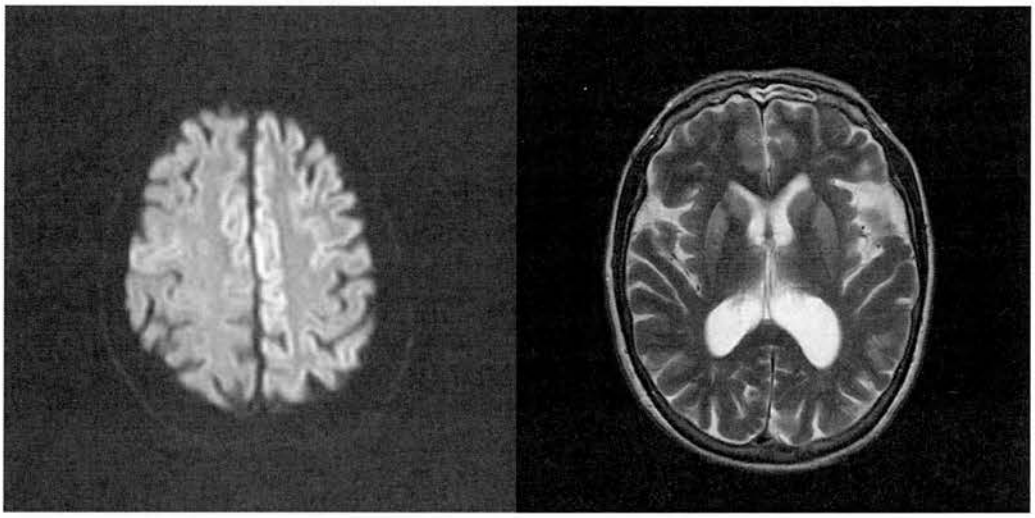

Figure 7 MRI showing cortical (left) and basal ganglia (right) high signal on DWI sequences in sCJD (14)

The use of MRI has increased over time reflecting increasing availability of this investigation and a growing recognition of its diagnostic value in suspect sCJD. The proportion of definite or probable sCJD cases undergoing MRI examination increased from approximately 15% in 1993 to around 70% in 2002 in the multi-centre EUROCJD collaborative study by Pedro-Cuesta *et al.*(107)

MRI changes have been observed as early as 3 weeks following symptom onset and prior to the detection of EEG abnormalities.(126;130) Unlike CSF 14-3-3 protein and

EEG, MRI findings are not associated with age at onset or disease duration.(97) MRI appears to be of value in rarer molecular subtypes of sCJD. Meissner *et al* evaluated MRI scans from 211 pathologically confirmed sCJD cases from 12 countries, correlating MRI findings to molecular subtypes.(127) Basal ganglia high sign was most commonly reported in the MM1, MV2 and VV2 subtypes, cortical ribboning in the MM2, MV1 and VV1 subtypes and thalamic high signal in the MV2 and VV2 subtypes.

It is difficult to determine the sensitivity and specificity of MRI in sCJD for a number of reasons. The diagnostic technology has evolved rapidly and as previously noted the sensitivity and specificity of this investigation varies according to the MRI sequences used; in many studies these were not optimized. Moreover, the diagnostic value of cortical high signal has only recently been recognised. Early studies reported basal ganglia high signal only.(97;106) The most recent study to report the sensitivity and specificity of MRI defined a positive scan as one showing high signal in the caudate nucleus and putamen or high signal in two of three cortical regions (parietal, occipital or temporal) on DWI or FLAIR sequences.(99) The authors reported a sensitivity and specificity of 83%. In this study the addition of these MRI criteria to the current WHO diagnostic criteria improved the overall sensitivity of the diagnostic criteria from 92% to 98% without compromising specificity (71% both current and proposed criteria). The additional cases detected through MRI scanning were of the rare molecular subtypes, such as VV1 and MV2, in which EEG and CSF 14-3-3 protein are of limited diagnostic value. Based on this evidence the authors have called for MRI criteria to be added to the current WHO diagnostic criteria for sCJD. At the time of writing this had not yet occurred although appeared imminent.

Basal ganglia high signal has been reported in a number of conditions, most of which are clinically distinguishable from sCJD (Table 9).

Table 9 Differential diagnosis of basal ganglia high signal on MRI

Neurodegenerative	Alzheimer's Disease, Lewy Body Dementia, Frontotemporal Dementia, Mitochondrial disease (Leigh's Disease), Huntington's Disease
Vascular	Vascular Dementia
Neoplastic	Carcinomatous Meningitis from small-cell lung cancer, Paraneoplastic Encephalopathy, Glioblastoma
Inflammatory/ Infectious	Lymphocytic encephalitis, Progressive multifocal leucoencephalopathy, viral encephalitis Subacute Sclerosing Panencephalitis, AIDS Dementia, Multiple Sclerosis
Metabolic	Hepatic encephalopathy, hypoxia, hypoglycaemia, Wilson's Disease
Autoimmune	Steroid responsive encephalitis associated with autoimmune thyroiditis
Toxic	Carbon Monoxide poisoning
Other	Depression, Schizophrenia

MRI findings can be difficult to interpret. Basal ganglia changes are often missed, especially in elderly populations. Schroter *et al* reported that characteristic changes in the basal ganglia are missed in up to 80% of images if these are not specifically sought by a neuroradiologist.(124) This is consistent with earlier findings from Zeidler *et al* in the UK.(131) Two large studies have reported intra and inter-observer variation in the examination of MRI imaging in suspect sCJD cases. Tschampa *et al* examined 442 MRI scans from 193 consecutive suspect sCJD cases referred to the German Surveillance Unit between 2001 and 2003.(106) Three reviewers independently assessed the scans. The overall kappa statistic for inter-observer agreement was 0.53 indicating moderate agreement; when only scans considered diagnostic were considered this rose to 0.66 indicating substantial agreement. Almost two thirds of definite or probable sCJD cases had evidence of basal ganglia high signal on MRI in this series and the sensitivity, specificity and positive predictive values of these MRI findings varied from 60 – 71%, 82 – 90% and 94 – 96% respectively between the three reviewers. Intra-observer agreement was not reported in this study but was examined in a recent study by Zerr *et al* who examined 436 MRI scans from suspect sCJD cases referred to 12 European surveillance centres between 1998 and 2007.(99) Five neuroradiologists blindly reviewed one scan per patient. A kappa statistic of 0.64, indicating substantial agreement was reported for

intra-observer variance; whilst kappa statistics varying from 0.45 (moderate agreement) to 0.64 (substantial agreement) were reported for inter-observer variance.

Genetic analysis

The phenotype of genetic disease may be clinically indistinguishable sCJD. The principal indication to test for a mutation of *PRNP* in suspect sCJD is to exclude genetic disease. The EUROCJD consortium report that genetic analysis was performed on approximately 64% of all definite or probable cases of human prion disease referred to national disease surveillance units; mutations were detected in 8.5%.(107) These figures have been relatively stable across time (1993-2002).

The *PRNP* polymorphic residue at Codon 129 determines susceptibility to sCJD.(132) In European populations the Codon 129 allelic distribution is as follows: methionine homozygote 37%, methionine heterozygote 51% and valine homozygote 12%.(133) However in European studies over 70% of sCJD cases are methionine homozygote.(134) Codon 129 genotype influences age at onset, disease duration and clinical phenotype. Across Europe an estimated 70% of all definite or probable human prion disease cases ascertained by national surveillance units participating in the EUROCJD consortium underwent genotype analysis; this figure is invariant over time (1993–2002).(107)

Neuropathology

Neuropathological examination is required to reach a definite diagnosis in all prion diseases. Neuropathological material can be obtained ante-mortem through brain biopsy or at post mortem following death.

Brain biopsy

Brain biopsy is an invasive investigation that involves removal of a small area from the non-dominant frontal cerebral cortex under general anaesthesia. Associated with serious complications and non-diagnostic in over 40% of cases, brain biopsy is reserved for suspect cases in which a treatable differential diagnosis is considered likely.(135) Heinemann *et al* reported the neuropathological findings in all suspect CJD cases referred to the German Surveillance Unit (1993-2005) that underwent brain biopsy (n=26).(135) Biopsy was non-diagnostic in 42% (n=11). In a further

42% (n=11) sCJD was confirmed, although almost half (n=5) of these individuals met the WHO diagnostic criteria as a probable case of sCJD prior to biopsy. A potentially treatable diagnosis was identified in just 3 (12%) suspect cases (vasculitis, chronic encephalitis and progressive encephomyelitis with rigidity and myoclonus) although none of these patients improved clinically following treatment. Between 10-20% of dementias are considered 'reversible.'(136) However one study has shown that less than half of all 'reversible' dementias improve with treatment and only around 10% fully reverse with treatment.(136) Beyond clinical considerations, there are further issues in relation to the need for appropriate facilities and the destruction or decontamination of surgical instruments following brain biopsy that may determine the availability of this investigation.

Post mortem examination

In the absence of a non-invasive ante-mortem diagnostic test post mortem examination following death is extremely important in suspect CJD. In the UK an estimated 70% of suspect CJD cases undergo post mortem examination,(72) however post mortem rates are falling.(137) Views toward this practice may have changed over time meaning that relatives are less likely to consent to, and clinicians may be more reluctant to request, post mortem examination. There have been several highly publicised issues relating to informed consent and organ retention in the UK in recent years which may have negatively influenced public opinion.

Pedro-Cuesta *et al* reported significant international variation in rates of post mortem in countries participating the EUROCJD project (1993-2002).(107) For example Slovakia reported autopsy rates of 100% whilst Spain, Germany and Italy reported rates of less than 60% (definite or probable sCJD cases). In the UK the rate fluctuated at around 80%. It is noteworthy however that this study did not report autopsy rates in *all* suspect cases referred to surveillance systems over time. Only around half of all suspect cases referred to surveillance systems meet the WHO diagnostic criteria for definite or probable disease.(138) In Japan, the rate of post mortem examination among all suspect prion disease cases referred to their surveillance system is quoted as approximately 25%.(139) In Germany 38% of all

suspect sCJD cases (n=358) referred to the surveillance system over a three year period (1993–1996) underwent autopsy; 49% of cases that met the WHO diagnostic criteria as a probable case of sCJD in life, 39% who met the criteria as a possible case in life and just 18% that did not meet the diagnostic criteria.(100) As previously noted practical issues relating to the examination of neuropathological material in suspect CJD may limit the use of post mortem examination, for example access to appropriate facilities and infection control issues.(140)

Neuropathological features

Macroscopic changes in sCJD are confined to the central nervous system. On gross examination cerebral or cerebellar atrophy may be observed. Microscopically, a classical triad of features are found: spongiform degeneration, astrocytosis and neuronal loss (Figure 10). Spongiform change may be localized or widespread. Amyloid plaques composed of PrP may be detected in up to 10% of cases. Definitive diagnosis requires immunocyochemical detection of PrP^{Sc} (Figure 8).

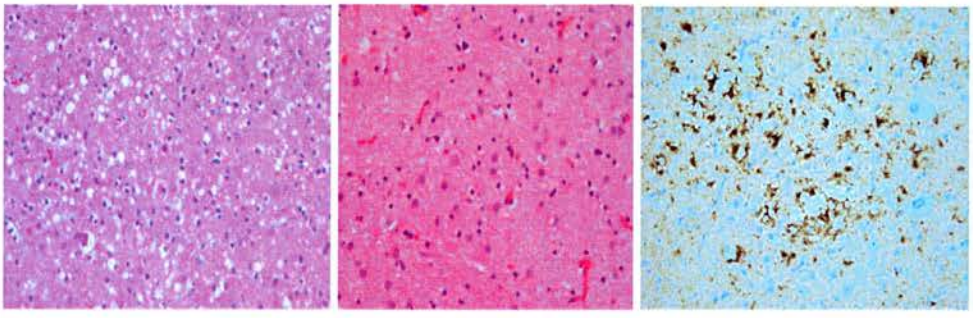

Figure 8 Microscopic and immunocytochemical features of sCJD (14)
The MM1 subtype of sCJD is characterised by microvacuolar spongiform change in the cerebral cortex, composed of small relatively uniform rounded vacuoles in the neuropil. Haematoxylin and eosin (H&E) stain (left); Severe neuronal loss and gliosis in the MM2 (thalamic) subtype of sCJD is a typical neuropathological finding, most evident in the medial and anterior thalamic nuclei. There is relatively little spongiform change present in the affected areas. H&E stain (centre). Immunocytochemistry for prion protein in the MM2 (cortical) subtype of sCJD shows widespread perivacuolar accumulation. A fine background granular/synaptic pattern of abnormal prion protein accumulation is also evident (right).

Molecular subtyping

Molecular subtype, the Codon 129 genotype in association with PrP^{Sc} prion protein typing, influences the clinico-pathological phenotype in sCJD.(141) In sCJD two major PrP^{Sc} prion protein types have been described: Type 1 (21 kDa non-glycosylated PrP^{Sc}) and Type 2 (19 kDa non-glycosylated PrP^{Sc}).(142) *PRNP* Codon 129 genotype and PrP^{Sc} prion protein type have been combined to produce a

molecular classification for sCJD that includes 6 major subgroups: MM1, MM2, MV1, MV2, VV1 and VV2 which explain much of the phenotypic variability in sCJD.(133;143;144) The clinical and neuropathological features described in Parchi *et al*'s classification are outlined in Table 10.(133) As can be seen from Table 10 several disease subgroups correlate directly with well-defined clinical presentations of sCJD such as that MM1 and VV2 subgroup. Other subgroups, such as the MM2 and VV2, correlate with what would be considered 'atypical' presentations of sCJD, rare presentations, early age at onset and/ or long illness durations. The MM1 and MV1 molecular subtypes have been grouped together in this classification because phenotypically they are indistinguishable.

Two alternate classifications exist. Hill *et al* describe three prion protein types in sCJD: Type 1, 2 and 3 (a fourth is described found uniquely in vCJD).(145) In the Hill classification Type 1 PrP^{Sc} is approximately 0.5 kDa higher than Type 1 in Parchi and all cases are methionine homozygote. Broadly speaking Hill Type 1 and 2 appear to correlate with Parchi Type 1, and Hill Type 3 (and 4) with Parchi Type 2. Zanusso *et al*'s classification is similar however these authors identified two groups of MM1 cases distinguishable based on the pH sensitivity of PrP^{Sc}; clinically the pH sensitive group have a shorter disease duration than the pH insensitive group.(146) Whilst heterogeneity in clinical phenotype in the MM1 subgroup is recognised, differences in PrP^{Sc} pH sensitivity have not been consistently reproduced.(143) In summary, both Hill and Zanusso are similar to Parchi except the former argue for a further sub-grouping of MM1 cases based on PrP^{Sc} and clinical phenotype, although this view is not supported by the current literature.(143)

A more pressing issue undermining the Parchi classification is the co-existence of two PrP^{Sc} types. In 12 – 44% of sCJD cases Type 1 and Type 2 PrP^{Sc} are found concurrently.(147) Some commentators suggest that these estimates may be significantly lower than the true figures;(148) others argue that methodological issues such as examining a small number of cases and bias in case selection, examining a limited number of samples from each case and the areas of the brain that have been sampled (focal areas of mixed protein type have been described) and the

Table 10 Molecular and phenotypic features of sCJD subtypes adapted from Parchi et al (133)

Sporadic CJD subtype	Previous Classification	Percentage of cases (%)	Duration (months)	Clinical Features	Neuropathological Features
MM1 or MV1	Myoclonic, Heidenhain	70	3.9	Rapidly progressive dementia, early and prominent myoclonus, typical EEG; visual impairment or unilateral signs at onset in 40% of cases.	"Classic CJD" distribution of pathology; often prominent involvement of occipital cortex; "synaptic type" PrP straining; one third of cases show confluent vacuoles and perivaculor PrP staining.
VV2	Ataxic variant	16	6.5	Ataxia at onset, late dementia, no typical EEG in most cases.	Prominent involvement of subcortical, including brain stem nuclei; in neocortex spongiosis is often limited to deep layers; PrP staining shows plaque-like, focal deposits, as well as prominent perineuronal staining.
MV2	Kuru-plaques variant	9	17.1	Ataxia in addition to progressive dementia, no typical EEG, long duration (>2 yr) in some cases	Similar to VV2 but with presence of amyloid-kuru plaques in the cerebellum and more consistent plaque-like, focal PrP deposits
MM2-thalamic	Thalamic variant	2	15.6	Insomnia and psychomotor hyperactivity in most cases, in addition to ataxia and cognitive impairment, no typical EEG.	Prominent atrophy of the thalamus and inferior olive (no spongiosis) with little pathology in other area; spongiosis may be absent or focal and PrP^{sc} is detected in lower amount than in the other variants.

Table 10 cont'd. Molecular and phenotypic features of sCJD subtypes, adapted from Parchi *et al* (133)

Sporadic CJD subtype	Previous Classification	Percentage of cases (%)	Duration (months)	Clinical Features	Neuropathological Features
MM2-cortical	Not established	2	15.7	Progressive dementia, no typical EEG.	Large confluent vacuoles with perivacuolar PrP staining in all cortical layers; cerebellum is relatively spared.
VV1	Not established	1	15.3	Progressive dementia, no typical EEG.	Severe pathology in the cerebral cortex and striatum with sparing of brain stem nuclei and cerebellum; no large confluent and very faint synaptic PrP staining.

use of novel technologies that do not produce consistent results, may have led to an overestimation of the co-existence of Type 1 and Type 2 PrP^{Sc}.(147) In the largest study to date, examining samples from 200 consecutive cases, Parchi *et al* estimated that PrP^{Sc} Types 1 and 2 coexist in approximately 35% of sCJD cases.(147) In this study mixed protein types occurred more frequently in the MM genotype than the MV or VV genotypes. In such cases the MM1 clinical phenotype predominated, although exceptions were noted. These results are not consistent with smaller studies that have variably reported the coexistence of PrP^{Sc} Types 1 and 2 occurring most frequently in MM or MV,(149) MV (150) and MV or VV genotypes (151), with associated variation in the predominant clinical phenotype. Some of the variation between studies may be explained by the methodological limitations previously highlighted and a systematic approach is required to ensure comparability of studies. An updated nomenclature proposed by Parchi *et al* based on their recent study has been reproduced in Table 11.(147)

Collins *et al* reported diagnostic sensitivities across molecular subtypes in analysis of 743 definite sCJD cases.(97) The distribution of molecular subtypes was as follows: MM1 60%, MM2 4%, MM1/2 4%, MV1 5%, MV2 10%, MV1/2 1.%, VV1 2%, VV2 14% and VV1/2 1% (1/2 indicating mixed protein types). MM1 patients were older than other subtypes at disease onset and had the shortest median illness duration whilst cases with MM2 or MV2 subtype had the longest median illness duration. Cases with an MM1 molecular subtype were more likely to have a typical EEG than others. CSF 14-3-3 protein positivity was most likely in the MM1 and VV2 subgroups whilst the VV2 subgroup was most likely to have MRI consistent with sCJD. Unfortunately the study lacked statistical power to adequately address clinical phenotype in mixed protein cases.

Increasingly complex molecular classifications have been developed. Their relation to the clinical and neuropathological phenotype is unclear. This area is becoming increasingly uncertain.

Table 11 Nomenclature and classification of sCJD subtypes, adapted from Parchi et al (147)

Nomenclature[a]	Percentage[b]	Distinctive histopathological features
Pure subtypes		
MM/MV1, VV2, MV2K; MM/MV2C, MM2T, VV1	65	Previously established (see Table 10)
Mixed subtypes		
MM/MV1+2C	26	As in MM/MV1 but with clusters of large vacuoles associated to perivacuolar and coarse PrP deposition mainly in cerebral cortex or thalamus.
MM/MV2C+1	2	As in MM/MV2C but with synaptic-type PrP staining in the molecular layer of the cerebellum.
VV2+1	3	Virtually indistinguishable from VV2.
MV 2K+1	1	Virtually indistinguishable from MV2K.
MV 2 K+C	3	As in MV2K but with clusters of large vacuoles associated to perivacuolar and coarse PrP deposition mainly in cerebral cortex.
MM 2T+C	<1	As in MV2T but with clusters of large vacuoles associated to perivacuolar and coarse PrP deposition mainly in cerebral cortex.

[a] It is largely based on Codon 129 *PRNP* genotype, which can be either methionine (M) or valine (V) and the PrP^{Sc} type (1 or 2 according to Parchi et al(133)). Since both MM2 and MV2 groups are associated to 2 distinct phenotypes, these are further defined with a third parameter (capital letter) referring to distinctive histopathological features: K kuru type amyloid plaques, C predominant cortical pathology with confluent vacuoles and perivacuolar PrP staining, T prominent thalamic pathology with atrophy; [b] Percentage of total consecutive sCJD cases (n = 200) investigated

Other sporadic prion diseases

Two further sporadic prion diseases have been described in humans.

At the time of writing nine pathologically confirmed cases of Sporadic Fatal Insomnia had been reported in the literature.(152) The clinical phenotype in Sporadic Fatal Insomnia differs from Fatal Familial Insomnia (FFI), a genetic prion disease, in age of symptom onset and disease duration only.

Protease-sensitive prionopathy (PSPr) was characterised by Gambetti *et al* in 2008.(153) This novel prion disease has a non-specific phenotype and the investigations typically of value in other forms of human prion disease, such as EEG, MRI and CSF 14-3-3 protein, are of limited utility. In the Gambetti series 3% (11) of all sCJD cases referred to the National Prion Disease Pathology Surveillance Center in Ohio (USA) between 2002 and 2006 were identified as being PSPr cases. Further cases have been identified following publication of this series, including two cases in the UK and one in the Netherlands.(154;155) These data underscore the importance of clinical and pathological disease surveillance and the need for high levels of case confirmation in suspect sCJD cases and atypical dementias.

Acquired prion diseases

Acquired prion diseases have arisen as a result of the transmission of infection from human to human (iCJD and Kuru) and from animal to human (vCJD).

Iatrogenic CJD (iCJD)

Over 400 cases of iCJD, attributable to the transmission of sCJD via health care associated interventions, have been reported worldwide.(156) The global distribution of iCJD is shown in Table 12.(156) The first case of iCJD was reported in 1974 in the recipient of a corneal transplant in the USA. sCJD was confirmed following autopsy in both recipient and donor. Two further cases were reported several years later in individuals that had undergone electrocorticography for intractable epilepsy also in the USA. The electrodes used in both procedures had previously been implanted in a patient that died of pathologically confirmed sCJD. The electrodes had been disinfected and sterilized between uses. These same electrodes were

inserted into the frontal lobes of a primate who later developed clinical and neuropathological features of sCJD thus confirming the route of transmission. Further iatrogenic transmissions of sCJD via neurosurgical instrumentation have been reported in the UK and France. There have been no new reports of transmission via depth electrode or through the use of contamination of neurosurgical instrumentation for three decades.(156)

Table 12 Global distribution of iCJD adapted from Brown et al (156)

	Surgical procedures				Pituitary Hormone Therapy	
	Dura mater	**Surgical instruments**	**EEG needle**	**Corneal transplant**	**Growth hormone**	**Gonadotrophin Hormone**
Argentina	1					
Australia	5				1	4
Austria	2					
Brazil					1	
Canada	4					
Croatia	1					
France	13	1			107	
Germany	8			1		
Holland	2				1	
Ireland	1					
Italy	4					
Japan	123					
New Zealand	2				6	
Qatar					1	
South Africa	1					
Spain	10					
Switzerland	1		2			
Thailand	1					
UK	7	3			51	1
USA	3			1	26	
Total	**196**	**4**	**2**	**2**	**194**	**5**

Additional possible single cases after corneal transplant or keratoplasty (not included in the table) occurred in Japan, the UK, and the USA. † Brazil and New Zealand human growth hormone (hGH) was prepared in the USA; Qatar hGH was prepared in France. Additional possible single cases due to hGH (not included in the table) occurred in The Netherlands, Scandinavia, and New Zealand.

Cadaveric-derived dura mater grafts

Cadaveric-derived dura mater grafts account for almost half of all cases of iCJD, with half of these reported in Japan.(156) The first case of iCJD related to the use of a cadaveric-derived dura mater graft was reported in 1987. Most cases have been associated with the use of the Lyodura graft produced prior to 1987 by one manufacturer; a small number of cases have been reported in the recipients of Tutoplast grafts produced in Germany. Grafts were produced by pooling dura from different donors; tissue had not been treated according to current recommendations regarding decontamination. Synthetic dura mater grafts are now available commercially.

Cadaveric-derived pituitary hormones

The first case of cadaveric-derived human growth hormone (hGH), used to treat children with growth hormone deficiency since the late 1950s, was reported in 1985. Further cases followed, predominantly in France, the UK and USA; in all cases exposures were pre-1985. Isolated cases of iCJD related to the use of cadaveric-derived pituitary gonadotrophin (hGnH) have been reported in Australia. Pituitary hormone was produced in batches with each containing up to 2000 pituitary glands. In all cases of iCJD associated with the injection (intramuscular or subcutaneous) of cadaveric-derived pituitary hormones the decontamination procedures adopted were not stringent enough to meet current recommendations. In the UK and the USA the young age at treatment onset and prolonged duration of treatment appear to be risk factors for developing iCJD among recipients.(156) In France all exposures occurred during a two year window (1983-1985) suggesting significant contamination of product during this period.

Clinical features

Table 13 outlines the clinical features of iCJD.(156) Reports suggest that investigations including EEG, CSF 14-3-3 protein and MRI are consistent with sCJD.(156) An excess of *PRNP* Codon 129 methionine homozygote cases are reported relative to the population distribution of this genotype, suggesting a genetic susceptibility. The polymorphism at *PRNP* Codon 129 appears to have little influence over the clinical phenotype, the exception being in the French hGH cases in whom a shortened incubation period is associated with *PRNP* Codon 129

heterozygosity.(157) The route of exposure does appear to influence clinical phenotype. Cerebellar signs are prominent in peripheral routes of exposure. Neuropathologically cases are generally indistinguishable from sCJD with some variability in the distribution of spongiform change, neuronal loss and astrocytosis. In hGH cases cerebellar disease is prominent with pronounced cerebellar atrophy and PrP positive amyloid plaque formation. Plaques are also seen in the spinal cord.

Table 13 Clinical and pathological features of iCJD, adapted from Brown et al (156)

Mode of infection	**Agent entry into brain**	**Mean incubation period (range)**	**Clinical presentation**
Corneal transplant	Optic nerve	18 – 320 months	Dementia/cerebellar
Stereotactic EEG	Intracerebral	16 – 20 months	Dementia/cerebellar
Neurosurgery	Intracerebral	12 – 28 months	Visual/dementia/cerebellar
Dura mater graft	Cerebral surface	16 – 23 years	Cerebellar (visual/dementia)
Growth hormone	Haematogenous (?)	4 – 36 years	Cerebellar
Gonadotrophin	Haematogenous (?)	12 – 16 years	Cerebellar

Kuru

The first accounts of kuru were published in 1957 by Gajdusek and Zigas.(158) Within a decade the epidemiology of the disease had been characterised. Kuru is a subacute neurodegenerative disease occurring in a geographically defined area in the Eastern Central Highlands of Papua New Guinea.(159) It has been suggested that the origin of kuru may have been a single human case of sCJD.(160) The epidemic was propagated by mortuary rituals in which women and children ate the brains and internal organs of deceased relatives. The proscription of ritual cannibalism in the mid-1950s interrupted transmission and curtailed the epidemic. No cases of kuru have been documented in individuals born after 1959. Together with age at onset, these data can be used to estimate the incubation period of kuru. The shortest incubation period is estimated to be 5 years, the longest 56 years, with a mean of 12 years.(17;161) The clinical phenotype of kuru is uniform. The disease presents with a pure cerebellar syndrome. Cognitive impairment is late and mild. Death typically occurs 12 months (range 6 to 36 months) following onset.(162) The *PRNP* Codon 129 genotype is known to influence both susceptibility and incubation period in

kuru.(161;163;164) Methionine homozygosity at Codon 129 is associated with shorter incubation periods and a rapid demise; heterozygosity is associated remarkably long incubation periods. Female survivors of the kuru epidemic are mostly heterozygotes suggesting that heterozygosity conferred some degree of protection.(165) Recently a study by Mead *et al* detected a novel polymorphism at Codon 127 (G127V) in a large number of susceptible *PRNP* Codon 129 methionine homozygote women who lived in the region during the kuru endemic.(166) This polymorphism is believed to confer resistance to kuru. The polymorphism was not found in kuru cases and has not been detected in other populations. The authors concluded that this is evidence of

> *"a complex selection event in the Fore population at PRNP during the kuru epidemic".*(166)

The significance of kuru

For a period kuru was considered of historical interest only. Following the emergence of vCJD there was renewed interest in kuru. Kuru is the only known epidemic of human prion disease, sustained through human to human transmission of an exogenous infectious agent via a peripheral route (oral inoculation).(167) Infection resulted in a surprisingly consistent clinical phenotype. Although incubation periods varied markedly these were typically long (up to and beyond 50 years). More recently evidence of genetic selection in the closed population exposed to kuru has emerged. Detailed study of the clinical phenotype, incubation periods, routes of transmission and factors determining genetic susceptibility/resistance have provided valuable insights that have informed policy and practice in relation to both human and animal prion diseases.

Variant CJD (vCJD)

In 1995 the NCJDSU identified a number of CJD cases in young individuals (aged at death <30 years old) presenting with an unusual clinico-pathological phenotype. By April 1996 the first ten cases of 'new variant' or simply 'variant' CJD as it would be known were characterised in an article published in the *Lancet* medical journal, and an aetiological link to BSE in cattle, proposed.(37)

Epidemiology

In the UK the primary vCJD epidemic peaked in 2000 (27 incident cases and 28 deaths) and has been in decline since (Figure 9).(46) As of the 1st November 2010, 221 definite or probable cases of vCJD had been reported worldwide; 174 in the UK (Table 14).(46) Outside the UK the greatest number of cases have been in France, where the annual number of incident vCJD cases peaked at 6 per annum in 2005/06, and declined thereafter.

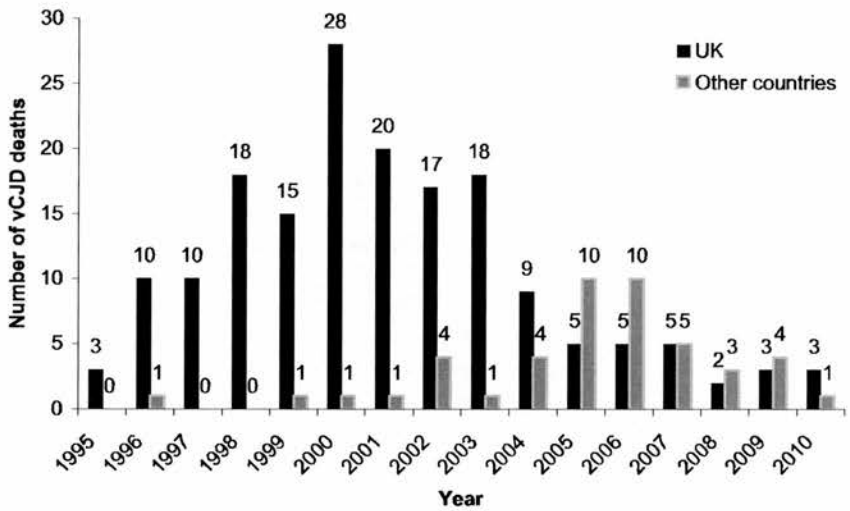

Figure 9 Annual number of vCJD deaths worldwide, 1995 to 2010 (46)
Accurate as of 1st November 2010

A number of other countries have experience isolated cases of vCJD (Table 14). In many, the affected individuals spent time in the UK during the BSE epidemic or were reportedly exposed to UK derived beef products from this era.(168) Comparative studies of vCJD cases from the UK and France suggest that a common agent is responsible for cases in both locations; phenotypically cases are indistinguishable with the exception of age at onset (French cases were on average 8 years older at symptom onset than UK cases).(169)

Table 14 Total number of incident vCJD cases worldwide, 1995 - 2010 (46)

Country	Number of incident cases (alive)	Number of incident cases due to secondary transmission
United Kingdom	174 (4)	3
France	25	0
Spain	5	0
Ireland	4	0
Netherlands	3	0
USA	3	0
Portugal	2	0
Italy	2 (1)	0
Canada	1	0
Japan	1	0
Saudi Arabia	1	0

Accurate as of 1st November 2010

Three vCJD cases in the UK have been attributed to secondary transmission via the transfusion of labile blood components (red blood cells, fresh frozen plasma and platelets, plasma derivatives such as Factor VIII, immunoglobulin or albumin).(154) For disease surveillance purposes these cases are considered vCJD cases not iCJD cases. They will therefore be addressed in this section of the thesis. There are no reported episodes of secondary transmission of vCJD outside the UK.

Mathematical modelling techniques have been used to predict the size of the vCJD epidemic. Initial estimates were wide, ranging from 70 to 136,000 deaths in the UK attributable to vCJD, reflecting uncertainty around the epidemiology and transmission potential of the disease.(170;171) The most recently published model (2010) considers *known* susceptibility/resistance factors and *known* primary and secondary routes of vCJD transmission. This model has estimated that an additional 390 (95% credibility interval 84 – 3000) incident vCJD cases will emerge in the UK between 2010 and 2079.(172) The authors concluded stating

> *"even in the absence of any further control measures, we do not find self-sustaining epidemics."(172)*

A slight male preponderance of vCJD cases has been noted.(154) The median age at symptom onset is 26 years (range 12 – 74 years); this has not changed significantly over time.(154) The reason for an excess incidence in younger age groups is not clear. It has been argued that differential dietary exposure alone is not sufficient to explain this finding.(173) An examination of vCJD deaths in the UK according to birth cohort revealed significantly different epidemic curves in those born in or before the 1970s compared to those born in the 1980s.(154) There were no deaths prior to 1999 in the latter cohort. This may indicate a greater susceptibility in the very young following exposure or reflect age dependant differences in incubation periods.(173) There have been no cases of vCJD born after 1989 (the year of the SBO ban).

An excess of vCJD cases in the 'North' of the UK relative to the 'South' has been reported.(154) As of 2008, the rate of vCJD (according to place of residence as of January 1991) was 4.42 per million population in the North, compared to 2.92 in the South. The North was defined as Scotland, North of England, Yorkshire and Humberside and the North West of England whilst the South was defined as Wales, West and East Midlands, East Anglia, the South West and South East of England. Extensive examination of the geographical distribution of cases in the UK has revealed a single cluster of 5 cases in the Leicestershire area, attributed to local butchery practices.(174)

Risk factors

There is compelling evidence from experimental transmission studies to support an aetiological link between BSE in cattle and vCJD in humans.(15;85;175;176) The only large case control study to examine risk factors in vCJD was published by the NCJDSU in 2006.(85) Examining 136 definite or probable vCJD cases and 922 general population controls, the study found an increased likelihood of reporting consumption of beef and beef products (likely to contain mechanically recovered meat) and chicken in vCJD cases compared to controls (> once per week *vs.* < once per week). No other robust evidence of medical, surgical (including dental), occupational, animal exposure or dietary risk factors were identified. The limitations of case control studies have been noted. Other risk factors for vCJD appear to be

young age and *PRNP* Codon 129 genotype (all definite or probable vCJD cases to date have been methionine homozygote at *PRNP* Codon 129). It is unclear whether *PRNP* Codon 129 genotype is genuinely risk factor, or whether vCJD in non-methionine homozygotes has yet to emerge due to an extended incubation period. A recent case report in the *Lancet* identified a possible case of vCJD in a Codon 129 heterozygote who died without post mortem.(177) Whilst the polymorphism at Codon 129 is considered the most important genetic risk factors for vCJD other candidate loci have recently been identified that may confer susceptibility/resistance and warrant further investigation.(178)

Secondary transmission via blood transfusion

To date there have been no conclusive reports of vCJD secondary transmission via health care associated procedures other than the transfusion of labile blood components. Three cases of vCJD attributable to the transfusion of labile blood components have been identified through the Transfusion Medicine Epidemiological Review study (TMER) in the UK.(179) All three individuals received non-leucocyte deplete red blood cells from asymptomatic individuals who developed vCJD between 17 months and 42 months after donating blood. The recipients developed symptoms of vCJD 6 ½ years to 8 years, 4 months post transfusion. Transfusion related vCJD cases have been phenotypically indistinguishable from other cases of vCJD.

To date the TMER study has identified 66 recipients of vCJD implicated labile blood components from 18 donors.(180) A retrospective case note review examined the medical case notes of 33 deceased recipients and found no evidence that any further recipients expressed clinical signs or symptoms suggestive of vCJD during life.(181) Only four of these recipients survived greater than 5 years post-transfusion. Tissue was not available to examine for evidence of asymptomatic infection in any of the deceased recipients however evidence from the kuru epidemic suggests that the minimum incubation period is likely to be 4 ½ years.

Abnormal PrP^{Sc} has been detected in the lymphoreticular system (LRS) of two further recipients of vCJD implicated labile blood components in the UK. Both were neurologically normal at the time of death; both were methionine heterozygote at the *PRNP* Codon 129. These cases have been termed 'asymptomatic vCJD infections.'

The first case, identified through the TMER study and reported in 2004, died of unrelated causes 5 years after receiving a vCJD implicated transfusion.(19) Neuropathological examination at autopsy was normal however PrP^{Sc} was detected in the spleen and a cervical lymph node. PrP^{Sc} has been detected on a single specimen taken from the spleen at autopsy of a second individual, a haemophiliac who had received pooled plasma products, red blood cells and underwent numerous invasive medical procedures including endoscopy.(182) This case was ascertained through a collaborative prospective surveillance study between the UK Haemophilia Centre Doctor's Organisation (UKHCDO) and NCJDSU. There are a large number of uncertainties around the pathogenesis of vCJD and the significance of asymptomatic vCJD infection. These issues will be discussed in greater detail later.

Clinical features

Behavioural change (withdrawal, apathy, aggression), psychiatric symptoms (ranging from emotional lability to psychosis) and/or painful sensory symptoms predominate at onset.(183-185) Many patients are referred to a psychiatrist rather than neurologist for initial investigation. Neurological signs are not present until a median of 6.25 months after symptom onset. Neurological signs include global cognitive impairment, ataxia and movement disorder (myoclonus or choreoathetosis). In the terminal stages the clinical picture is similar to sCJD; patients are usually akinetic and mute, some develop cortical blindness. Death occurs a median of 14 months (range 9 – 35) following symptom onset. The median age at death is 29 years (range 19 - 41).

Diagnostic criteria

The clinical picture in vCJD has been remarkably consistent. This facilitated the rapid development of clinico-pathological diagnostic criteria, despite the novelty and rarity of the condition. WHO diagnostic criteria were introduced in 1998 based on characterization of the initial cases. These were amended in 2002 to include tonsil biopsy and later a footnote added in relation to EEG findings (Appendix 2).(98) In the most comprehensive and contemporary series to date, Heath *et al* (2010) described the sensitivity and specificity of diagnostic criteria for vCJD in 106 neuropathologically confirmed cases ascertained by the NCJDSU from 1995 through 2004 and 45 pathologically confirmed non-cases.(186) The study reported the

sensitivity, specificity, positive and negative predictive values of the diagnostic criteria (met the diagnostic criteria as a probable case during life) to be 83% (75 – 90), 100% (92 – 100), 100% (96 – 100) and 71% (59 – 82) respectively. Twelve pathologically confirmed cases of vCJD did not fulfil the diagnostic criteria on the basis of insufficient clinical features.

Differential diagnoses

The study by Heath *et al* reported the final outcome of 99 suspect vCJD cases that met the WHO diagnostic criteria as a possible vCJD case at some point in the course of their clinical illness. The majority (n=83) were classified as definite or probable vCJD cases, three remained possible vCJD cases and a further eight were classified as sCJD cases. In three cases an alternate neuropathological diagnosis was reached (Alzheimer's Disease, viral encephalitis and subacute sclerosing panencephalitis). In one case a clinical diagnosis of Wilsons disease was reached and in the final case a formal clinical diagnosis was not reached but the patient spontaneously improved.

Investigations that support a diagnosis of vCJD

Routine haematological and biochemical investigations are typically normal in vCJD. Transient abnormalities of liver function tests are reported in up to half of all cases although this may not be disease specific. EEG is less useful in suspect vCJD than in suspect sCJD. The EEG is often normal in early disease, progressing to show non-specific abnormalities during the late stages of illness. In isolated cases PSWC have been described on EEG in the terminal stages.(98) CSF examination is typically normal.(121) The sensitivity of CSF biomarkers in vCJD is low; CSF 14-3-3 protein has a sensitivity of 40% (95%CI 30 – 50), Tau 24% (16 – 35), s100b 62% (51 – 72) and NSE 24% (10 – 45).(109) Phosphorylated Tau has shown some promise in discriminating between vCJD and other forms of dementia, including sCJD, but not sufficient to warrant inclusion in the diagnostic criteria.(187)

The section that follows will focus on diagnostic investigations of proven value in vCJD, which are those investigations included in the WHO diagnostic criteria. It is noteworthy that vCJD is a relatively new entity and therefore there are fewer studies in this area than for sCJD which has been extensively studied.

MRI

The neuroimaging modality of choice in vCJD is MRI scanning. Neuroimaging using CT scanning is typically normal or occasionally shows generalised atrophy. Other imaging modalities such as SPECT scanning show non-specific changes.

Two case reports of hyper-intensity in the pulvinar nucleus of the thalamus were published in 1996.(188;189) The following year Zeidler *et al* noted high signal in the posterior thalamus in two of the first 14 cases of vCJD reported in the UK (on T2 and PD weighted MRI sequences).(183) The MRI findings correlated with pathological changes found in the thalamus. In an update of this study published in 2000, Will *et al* describe bilateral high signal in the posterior thalamus in 77% of definite or probable vCJD in the UK (n=35).(184) More recently Heath *et al* report characteristic findings in 91% of vCJD cases in the UK (106 confirmed cases from 1995-2004).(186) The so called 'pulvinar sign' has been incorporated into the WHO diagnostic criteria.(98) This describes

> "*a characteristics distribution of symmetrical hyper-intensity of the pulvinar nucleus (posterior nucleus) of the thalamus (relative to the grey matter of the anterior putamen and normal cerebral cortex)*" (Figure 10).(98)

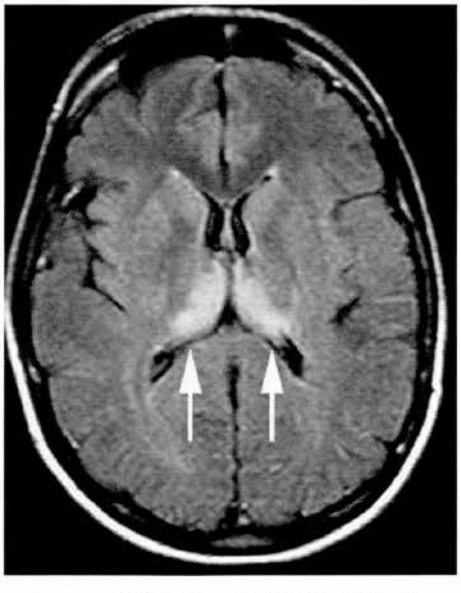

Figure 10 The Pulvinar sign on MRI in vCJD (FLAIR sequence) (14)

In a case-control study examining 36 pathologically confirmed vCJD cases and 57 controls the sensitivity and specificity of these radiological changes were 78% (60 –

90) and 100% (94 – 100) respectively (intra-observer reliability extremely high; kappa statistic >0·8).(190) This study examined T2 and PD weighted images. A subsequent study examining 86 pathologically confirmed vCJD cases also found that the sensitivity and specificity of the pulvinar sign in vCJD was extremely high.(191) The most sensitivity sequences were FLAIR followed by PD and T2 weight images. Very few DWI images were available for review although the value of this sequence has since been recognised. Other radiological features less commonly seen on MRI in vCJD include high signal in the dorsomedial thalamic nuclei ("hockey stick" sign), periaqueductal grey matter, caudate head and deep white matter.(191) Cerebral atrophy is rarely seen.

The differential diagnoses of thalamic high signal on MRI scanning are outlined in Table 15.(98) Of note two reports of the classical pulvinar sign in sCJD have been published.(192;193) Other reports in vCJD indicate that the pulvinar sign may disappear with disease progression.(191) Given the novelty and rarity of vCJD extensive studies reporting the timing of MRI changes in relation to disease course have not been published.

Table 15 Differential diagnosis of thalamic high signal on MRI scanning (98)

Pulvinar and dorsomedial nuclei high signal	*Thalamic high signal excluding pulvinar*
sCJD	Carbon monoxide poisoning
Benign intracranial hypertension	Japanese Nipositu encephalitis
Cat-scratch disease	Wernicke encephalitis
Alpers syndrome	Bithalamic glioma
Post-infectious encephalitis	Thalamic infarction

Genetic analysis

The principal use of genetic analysis in vCJD is to exclude genetic disease. To date all definite or probable vCJD cases have been *PRNP* Codon 129 methionine homozygotes.

Tonsil biopsy

Despite uncertainty regarding the pathogenesis of vCJD, early recognition of the presence of PrP^{Sc} in the tonsils of patients with vCJD, but not other forms of prion

disease, both ante- and post mortem led to the inclusion of this investigation in the diagnostic criteria.(194) The initial case series was based on tonsil samples from nine vCJD cases, all of which tested positive; there were no positive results in the control or 'other' CJD groups.(194) These data suggest a high sensitivity and specificity but the numbers are very small. A positive tonsil biopsy elevates the diagnostic classification from possible to probable, however examination of neuropathological material is still required to reach a definitive diagnosis. In the study by Heath *et al* just 15 of the 106 neuropathologically confirmed cases of vCJD underwent tonsil biopsy.(186) Tonsil biopsy was positive in all but one case. The procedure requires general anaesthetic and is associated with risks including haemorrhage and sepsis. Moreover a negative result does not definitively exclude vCJD and does not assist in excluding alternate diagnoses such as sCJD.(195)

Neuropathology

Brain biopsy can be used to reach a definitive diagnosis in life and exclude potentially treatable differential diagnoses but is not, as previously discussed, without risk. More commonly pathological material is obtained at post mortem. Issues relating to both post mortem examination and brain biopsy have been discussed previously and will not be revisited here.

Neuropathological features

vCJD is neuropathologically distinct from other forms of human prion disease.(37) In all forms of human prion disease spongiform change, neuronal loss and astrocytosis are present. In vCJD these changes are most marked in the basal ganglia and thalamus. Similarly, neuronal loss and astrocytosis are most prominent in the posterior thalamus, correlating with MRI findings. The most defining feature however is the presence of 'florid plaques' in the cerebral and cerebellar cortical grey matter. These are "abundant kuru-type fibrillary PrP plaques often surrounded by a halo of spongiform change."(98) In addition to florid plaques, small plaques are seen clustered within the cerebral and cerebellar cortex, unrelated to spongiform change. Of note other tissues, the dorsal root and trigeminal ganglia, the retina, optic nerves and substantia gelatinosa of the spinal cord also stain positive for abnormal prior protein. The pathological features of vCJD are shown in Figure 11. Beyond the

central nervous system, abnormal prion protein is also detectable in LRS tissue including the appendix, tonsils, spleen and lymph nodes.

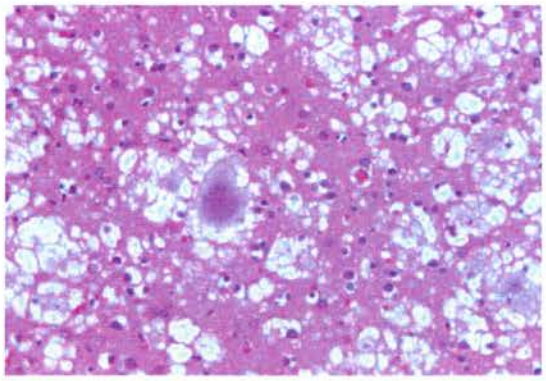

Figure 11 Pathological features in vCJD (14)
The characteristic pathological feature in vCJD is the florid plaque (centre) composed of large radiating fibrils of amyloid with a dense core and paler periphery, surrounded by a halo of spongiform change. Multiple smaller plaques are present elsewhere in this image and there is severe neuronal loss with accompanying astrocytosis. H&E stain.

Molecular subtyping

A single prion strain has been described in vCJD which shares features in common with the strain characteristics found in BSE. vCJD is characterised by a Type 2B prion protein with a predominantly diglycosylated band.

Genetic prion diseases

Genetic prion diseases account for between 10-15% of all human prion diseases.(196) Genetic prion disease is caused by disease specific mutations in *PRNP*, inherited in an autosomal dominant pattern. Mutations can be point or insertion. In the absence of neuropathological material, the diagnosis of a genetic prion disease requires the presence of a *PRNP* disease causing mutation or a positive family history in an individual with appropriate clinical features (Appendix 2). The criteria thus recognise the ever increasing number of *PRNP* mutations that have been characterised and that up to half of all cases of genetic prion disease report no significant family history.(98;196) The penetrance of some mutations is variable; individuals with the mutation do not necessarily develop the disease, or development of the disease may be, for example, age dependent.(196) There is no evidence of genetic anticipation.

Genetic disease is broadly considered in three phenotypes: genetic CJD (gCJD), Gerstmann-Straussler-Scheinker Disease (GSS) and Fatal Familial Insomnia (FFI). In practice these groupings include diverse clinic-pathological phenotypes. The haplotype, the pathogenic mutation in association with a polymorphic residue (usually at Codon 129 but other polymorphism have been described including Codon 171 (N/S), 219 (E/K), the deletion of one 24-bp octapeptide repeat and 12 other silent polymorphisms) determines the clinical phenotype. Perhaps the most significant aspect of genetic prion disease is that it can be clinically and pathologically indistinguishable from sCJD. Detailed accounts of the clinical phenotypes associated with specific haplotypes can be found in a number of manuscripts.(98;152;197-200) I would direct an interested reader to these resources.

The geographical distribution of genetic prion diseases is significantly different from other human prion diseases. Overall the incidence of genetic prion disease was reported to be 0.17 per million population in countries in the EUROCJD consortium from 1999 through 2002 with two thirds of cases accounted for by genetic CJD.(196) Clusters of gCJD cases associated with a specific mutation have been reported in Israel, Chile, Italy and Slovakia.(196) In the example of Slovakia 70% of all human prion diseases were aetiologically genetic (largely attributable to a single mutation, the E200K mutation) and the annual mortality rate from genetic prion disease was 1.1 per million population.(196) This compares to 6.6% and 0.2 per million population in the UK. Some mutations are reported exclusively in geographically defined populations and are exceptionally rare, affecting one kindred or a single individual.

Treatment of human prion disease

Whilst a number of agents have been studied, to date no treatments of proven value have been identified in human prion disease. A 2008 systematic review identified 34 primary research studies examining 15 drugs.(201) Just one randomised control study was identified. This suggested a slowing of cognitive decline associated with the use of flupirtine. The remaining studies were case series; 20 studies examined

one patient, three studies reported on more than ten patients. The latter studies examined the use of quinacrine, mepacrine and pentosan polysulphate with mixed results. Primary research studies were limited by poor study design and inadequate reporting. Research in this area is further hindered by the heterogeneity of the populations studied, including genetic prion diseases, vCJD and sCJD cases, and the diversity of the clinical phenotype within each group.

Measures to reduce the risk of secondary transmission of prion disease

This section will review the control measures that have been put in place to reduce the risk of human to human transmission of prion disease in the UK. Control measures to reduce the risk of animal to human transmission in the UK were explored in the section on prion diseases in animals and will not be revisited here. A number of issues are important with respect to prion diseases that must be considered when discussing control measures to reduce the risk of iatrogenic transmission: (1) long incubation periods during which an individual may be asymptomatically infected and infectious (2) lack of a valid diagnostic test to detect asymptomatic infection (3) resistance of prion protein to routine decontamination procedures. In the section that follows I will review some of the key issues and uncertainties relating to asymptomatic vCJD infection, before examining specific control measures relating to human prion diseases in the UK.

Asymptomatic vCJD infection

Abnormal PrP^{Sc} has been detected in the LRS of neurologically normal individuals that received vCJD implicated labile blood components. These individuals are considered to have 'asymptomatic vCJD infection'. However the pathogenesis of vCJD is poorly understood. It is not known for example whether all asymptomatic but infected individuals are infectious, whether all infected individuals will develop disease or, whether a sub-group will remain asymptomatic but infectious carriers or whether a further sub-group will clear the disease without ever developing symptoms. A number of synonyms have been used to describe these states including pre-clinical, sub-clinical and pre-symptomatic disease. For consistency throughout this thesis I will use the term asymptomatic infection to refer to these synonyms. The

prevalence of asymptomatic infection and potential for asymptomatic but infected individuals to transmit vCJD are important parameters for public health planning, including the establishment and maintenance of control measures.

Prevalence studies

A number of studies have been undertaken to inform the public health response to the possibility of a health care associated secondary epidemic of vCJD. The aim of these studies has been to estimate the prevalence of asymptomatic infection in the population. Of note, in these studies the detection of PrP^{Sc} is considered evidence of asymptomatic infection without implying infectiousness.

Hilton *et al* retrospectively examined 11,247 appendix and 1,427 tonsil samples from histopathology departments across the UK.(202) Samples, from the 1961 – 1985 birth cohort, were anonymised prior to testing. In addition, appendix samples from vCJD patients, either at autopsy or surgery prior to symptom onset, were examined. PrP^{Sc} was detected in three appendix samples, two of which were from asymptomatic individuals that subsequently developed vCJD giving a prevalence of 292 per million population (95% CI 60 – 853). This study retrospectively examined paraffin embedded tissue samples. In such studies frozen tissue is unavailable thereby limiting the study to histological examination (immunohistochemistry) and precluding biochemical testing (immunoblotting).

Frosh *et al,* prospectively examined 2000 consecutive tonsillectomy specimens using immunohistochemistry and immunoblotting.(203) No samples tested positive. Given the rarity of vCJD this study is likely to be underpowered due to the small sample size, an issue compounded by the fact that almost half of the study population were in a birth cohort unlikely to have had substantial dietary exposure to BSE contaminated food products.

More recently Clewley and colleagues examined 63,007 tonsil pairs electively removed and stored in the National anonymous tissue archive for Scotland and England between 2004 and 2008.(204) Using both biochemical and histological diagnostic technologies there were no positive results in the study. The overall

prevalence of disease related prion protein was 0 per million population (0 – 59); in the 1961 – 1985 birth cohort the prevalence was 0 per million population (0 – 289). The authors offered three possible explanations for the discrepancy between this and the previous study by Hilton *et al*: the sensitivity of the tests (in the absence of a definitive diagnostic test to confirm infectivity), the representativeness of the sample and the natural history of prion protein infectivity.

The pathogenesis of vCJD is not fully understood. It is not clear at what stage in the disease process PrP^{Sc} might be detected and in which tissues. The LRS has been chosen for these studies as PrP^{Sc} has been detected in appendix and tonsil tissue in both in vCJD cases and asymptomatic individuals that received vCJD implicated transfusions of labile blood components. Whilst PrP^{Sc} has been detected by both immunoblotting and immunohistochemistry in the tonsil tissue of vCJD cases it has never been detected in an asymptomatic individual. Animal studies would however suggest that PrP^{Sc} does accumulate in this tissue and can be detected prior to symptom onset.(205;206) The sensitivity and specificity of any test will be influenced by the distribution of prion infectivity in tissue during incubation and the timing of testing. The significance of a positive result is also unclear. In animal studies PrP^{Sc} has been cleared following inoculation.(207) It is not clear therefore whether the detection of PrP^{Sc} in tissue removed from an individual will mean that the individual in question will develop vCJD, and if so when. Thus far studies in this area have used unlinked anonymised samples. There are complex ethical issues to be considered in carrying out linked or named studies in the face of such uncertainties and in the context of an untreatable and invariably fatal disease.

Asymptomatic vCJD infection in non-methionine homozygote Codon 129 genotypes

One further issue worthy of discussion is the detection of PrP^{Sc} in asymptomatic individuals with a non-methionine homozygote Codon 129 genotype both in the anonymised studies described above and through disease surveillance (TMER and UKHCDO) in the UK. This finding implies that non-methionine homozygotes are susceptible to vCJD. This is congruent with animal studies that have shown a species barrier in transmission of BSE to human transgenic mice, but efficient transmission

of vCJD between human transgenic mice.(208) A gradient of efficiency according to *PRNP* Codon 129 genotype (MM to MV to VV) was reported although crucially all genotypes were susceptible. *PRNP* Codon 129 genotype determined clinico-pathological phenotype. Codon 129 methionine heterozygotes or valine homozygotes were likely to remain in an infectious asymptomatic stage extending beyond their natural life span. These data support the hypothesis that a significant but as yet un-quantified population of asymptomatic but infectious individuals may exist; individuals that may donate blood or other tissues and undergo invasive medical procedures. If novel routes of vCJD transmission emerge, the possibility of a self-sustaining secondary epidemic of vCJD will become increasingly real.

Control measures in the UK

The transmission of sCJD, vCJD and genetic prion disease will be addressed here. In this section I will refer to an "at increased risk" of CJD group. This is a group of asymptomatic individuals who have been informed that they are "at increased risk" of developing CJD as a result of their medical or family history (Table 16).(209) They are considered at risk for public health purposes. They have been informed of their status and advised not to donate blood or organs and in turn, to inform health care providers prior to any medical intervention such that appropriate precautions can be taken to minimise the potential for further transmission, in the event that they have asymptomatic infection. There are over 6,500 individuals in this group, the majority of whom are patients with bleeding disorders that received UK sourced plasma products between 1980 and 2001.

Table 16 Individuals "at increased risk" of developing CJD in the UK (209)

1.	Individuals with a pathogenic mutation of the *PRNP* gene
2.	Individuals with a blood relative with a pathogenic mutation of the *PRNP* gene
3.	Individuals with ≥ 2 blood relatives affected by human prion disease
4.	Recipients of human derived hormone products
5.	Individuals that underwent intradural neurosurgical or spinal procedures pre-1992
6.	Individuals identified by the CJD Incidents Panel as having undergone surgery with instruments previously used on someone who has gone on to develop CJD or become "at increased risk" or developing CJD
7.	Individuals who have received an organ or tissue from a donor infected with CJD or "at increased risk" of CJD
8.	Individuals who have been identified prior to high risk surgery as having received blood or blood components from ≥ 80 donors since January 1980
9.	Individuals who have received blood from someone who has developed variant CJD
10.	Individuals who have donated blood to someone who subsequently developed variant CJD
11.	Individuals who have received blood from someone who has given blood to a patient that subsequently developed variant CJD
12.	Individuals treated with certain implicated UK sourced plasma products between 1980 and 2001

A number of factors are considered in determining the risk of human to human iatrogenic transmission, including the distribution of PrP^{Sc} in tissue and bodily fluids, the route of transmission, the dose of infectivity and the agent. As previously noted, the detection of PrP^{Sc} does not necessarily infer infectivity, nor does its absence exclude it. In general high levels of PrP^{Sc} are assumed to represent a greater risk of infection. These data have been used to inform an assessment of the relative risk associated with various procedures. Tissues are classified as high, medium or low risk. Central nervous system tissues are considered high risk in all prion diseases. Ophthalmological procedures on the anterior segment of the eye (cornea, iris, ciliary body and lens) are considered medium risk and procedures in the posterior segment of the eye (retina, retinal pigment epithelium, choroid, subretinal fluid and optic never) high risk. PrP^{Sc} is found in higher levels in vCJD in the peripheral nervous system, LRS and alimentary tract than in other prion diseases; these tissues are considered a medium risk in vCJD.(210)

Cadaveric-derived human dura mater grafts and pituitary hormones

The use of cadaveric-derived human pituitary hormone was banned in the UK in 1985; the use of cadaveric-derived human dura mater grafts in 1992. Incident cases of iCJD via these routes of transmission continue to be reported due to prolonged incubation periods; some are retrospectively identified through case reviews applying emergent diagnostic technologies. It would be anticipated that these forms of iatrogenic disease will rapidly disappear given interruption to the route of transmission, in much the same way as has been observed with the kuru epidemic.

Organ donation

There no recorded episodes of iCJD attributed to the transplantation of body organs. Organ donation is prohibited in individuals dying of dementia or suspect prion disease. Individuals designated "at increased risk" of CJD are requested not to donate organs or other bodily fluids.

Surgical procedures

Where possible single use instruments are recommended for invasive medical procedures involving tissues of high to medium infectivity in suspect prion disease cases or individuals "at increased risk" of CJD.(209) Where this is not possible, instruments may be quarantined and re-used exclusively on the index case. Alternatively instruments are destroyed. No special precautions are required for low infectivity tissues or bodily fluid.

A number of general preventative measures have been implemented. There has been significant investment to improve decontamination facilities although the cost effectiveness of this is unknown.(211) Efforts have been made to track all surgical instruments and avoid migration of instruments between surgical sets, a measure that is cost effective.(211) In 2001 all re-usable tonsillectomy surgical instruments were withdrawn and single use instruments recommended for all patients undergoing this procedure. However reports rapidly emerged of increasing surgical complications related to these single use instruments, in particular haemorrhage due to ineffective diathermy, and this decision was swiftly reversed. In 2006 (updated 2008) the National Institute for Health and Clinical Excellence in the UK issued further guidance on reducing the risk of iCJD transmission via invasive medical

procedures.(211) This recommended that all accessories for neuroendoscopies should be single use, but that there was no evidence of cost effectiveness for single use instruments for other procedures including neurosurgery, ophthalmological surgery, tonsillectomy and endoscopy. The guidance further stipulated that single use instruments should only be used where they were of a comparable quality to re-useable instruments.

In instances of possible iatrogenic exposure through health care associated procedures, cases are reviewed by the CJD Incidents Panel (CJD IP) who investigate and advise on further action.

Dentistry

Uncertainties remain regarding the risks associated with dental procedures, particularly endodontic treatment which involves contact with dental pulp.(212) A survey of decontamination procedures in dental surgeries in Scotland identified serious short comings in the practice of cleaning and decontaminating dental equipment.(213) Whilst studies in humans have not detected infectivity in the oral cavity, in animal models vCJD infectivity has been detected in the oral cavity in both the symptomatic and asymptomatic stages of disease.(214) In light of these data single use instrument were recommended for use in endodontic procedures (files and reamers) in 2007.(215) Further advice issued in 2009 stated

> *"Other instruments or device types for which a reliable cleaning regime is not available should also be considered for replacement by single-use types or the single use of reprocessible types."*(216)

Blood transfusion and donation

There is no compelling evidence to suggest that sCJD or genetic prion diseases are readily transmissible from human to human through the transfusion of labile blood components. By contrast, evidence from basic science and epidemiological studies suggest that vCJD is transmissible through the transfusion of labile blood components. In the absence of a blood test to detect infectivity in humans much of the evidence from this area is based on animal models of infectivity. It is not clear how reflective these studies are of the pathogenesis of vCJD in humans. Animal models suggest that infectivity in the blood is low relative to levels in the brain,

during the incubation period and following symptom onset.(217) Infectivity in blood is distributed evenly between plasma and leukocytes, with very low or absent levels in red blood cells or platelets.(218) Steps taken in the UK to reduce the risk of secondary transmission via blood products include leukodepletion which is thought to reduce the risk by up to 40% (Table 17). (219;220) Prion reduction filters have been developed to reduce infectivity in labile blood components. These have shown promise in animal studies but their utility in humans is unknown.(221)

Table 17 Measures taken to reduce the risk of secondary transmission of vCJD through the transfusion of blood and blood products in the UK (219;220)

Year	Measure
1997	Recall and discard labile blood components and plasma derivatives from donors who subsequently developed vCJD
1998	Importation of plasma destined for fractionation from non-UK sources (fully implemented October 1999)
1998	Leukoreduction of labile blood products (fully implemented Autumn 1999)
2003	Importation of fresh frozen plasma (FFP) for recipients born after January 1st 1996 (fully implemented 2004)
2004	Permanent exclusion of whole blood donors who received a transfusion of blood components after January 1st 1980 in the UK
	Permanent exclusion of blood donors who have received a transfusion of blood component or plasma derivative from the UK after January 1st 1980
	Importation of FFP for recipients aged < 16 years old
	Permanent exclusion of donors who received a transfusion of blood components or plasma derivatives anywhere in the world after January 1st 1980
2005	Permanent exclusion and notification of donors whose donations have been transfused to recipients who later developed variant CJD
	Progressive replacement of platelet pools with apheresis (single-donor) platelets. Apheresis platelets recommended for children < 16 years old

note these measures cover the period examined in the studies in this thesis 1990 - 2006

Screening for PrP^{Sc}

Considerable effort has gone into the development of a blood test to detect PrP^{Sc} in humans. Such a test could be used to screen all blood and organ donations for example. A number of issues would need to be considered prior to the introduction of a universal screening programme. There are major deficiencies in our understanding

of how PrP^{Sc}, if detected, relates to infectivity and the likelihood of developing clinical disease. A simple, safe, validate and acceptable screening test is required. The test would need to have a high sensitivity (ability to detect true positives) and specificity (ability to detect true negatives). Establishing these qualities in the absence of a gold standard test to detect PrP^{Sc} is challenging. Whilst sensitivity and specificity are properties of a test, the PPV, that is the likelihood that an individual with a positive test result will have the disease, is determined by the prevalence of disease in the population being screened. The population prevalence of asymptomatic PrP^{Sc} infection is unknown, but thought to be low. Applying a screening test in this context is likely to result in a large number of false positive results. In most screening programmes an individual with a positive result from screening would be offered a diagnostic test. However a simple, safe, validated and acceptable diagnostic test in this context does not exist and given the uncertainties surrounding the natural history of asymptomatic PrP^{Sc} infection the interpretation of a diagnostic test would be problematic. There are wider issues relating to whether it is necessary to inform screened individuals of abnormal results given the natural history of asymptomatic infection is poorly understood and in the absence of an effective treatment. The likely consequence of screening in this context will be the identification of an increasing number of individuals designated 'at increased risk' of CJD with all the inherent implications for these individuals, their families and the health care service. In short, the costs (financial, physical or psychological) of screening to the individual, health service and society, should not outweigh the public health benefits.

Public Health Surveillance

In this section I will define public health surveillance (PHS), briefly outline the rationale for, and challenges in, PHS in relation to human prion diseases and describe the evolution of PHS system for human prion diseases in the UK and beyond.

Public Health Surveillance

Public Health Surveillance (PHS) has been defined as

> "*the on-going, systematic collection, analysis, and interpretation of data (e.g., regarding agent/hazard, risk factor, exposure, health event) essential to the planning, implementation, and evaluation of public health practice, closely integrated with the timely dissemination of these data to those responsible for prevention and control.*"(222)

A number of factors are considered when determining whether a condition merits PHS. These include the frequency of the disease (incidence or prevalence), the severity (case-fatality), the preventability, the transmissibility or potential for an outbreak to occur, the costs associated with the condition and the level of public interest and media attention.

The rationale for PHS of human prion diseases

The term 'prospective' will be used to refer to real-time surveillance. Intermittent CJD surveillance studies were conducted both prospectively and retrospectively in England and Wales from 1970 and retrospectively in Scotland and Northern Ireland from 1980. In the UK continuous systematic prospective PHS of CJD was instigated in 1990 as recommended by both the Southwood Working Party and the Tyrell Committee. The primary aim of systematic prospective CJD surveillance in the UK was to detect any changes in the clinico-pathological phenotype of CJD that might be attributable to exposure to BSE. It should be noted that the term CJD has historically been used as an umbrella term to describe a range of phenotypically and aetiologically diverse prion diseases, including for example sCJD and GSS. The term 'prion disease' is now more frequently used in this context to reflect our advancing knowledge of this heterogeneous group of diseases.

In 1996 the NCJDSU characterised a clinico-pathologically distinct form of CJD, vCJD, which would later be aetiologically linked to BSE in cattle. A WHO

consultation the same year examined the public health issues relating to human and animal prion diseases.(223) At this time just ten cases of vCJD had been characterised and cases were confined to the UK. Whilst the most likely explanation for the emergence of vCJD was exposure of the UK population to BSE, scientific evidence to support this hypothesis was lacking. The epidemiology and natural history of vCJD were yet to be determined, the size and distribution of the primary vCJD epidemic un-quantified and the potential for a secondary epidemic unknown. An opportunity to address many of these uncertainties through PHS existed. Global surveillance of all forms of CJD was recommended based on a number of principles. An aetiological link with BSE in cattle seemed likely. Indigenous and imported BSE had been reported outside the UK indicating that a wider population may have been exposed to BSE and potentially at risk of developing vCJD. Knowledge about the clinical phenotype of vCJD was based on the experience of just ten cases. The possibility of a clinical phenotype indistinguishable from sCJD remained. Many developed countries had mature PHS systems for CJD in place and this experience was considered crucial in detecting a novel disease phenotype.

The social and political climate in the UK

The demonstration of a novel and universally fatal disease in humans attributed to BSE (a disease attributed to intensive farming practices), to which the UK population had been widely and involuntary exposed for a decade, in the face of repeated reassurances from the scientific and political communities, generated considerable controversy. Public anger and mistrust were tangible and fuelled by a sensationalist media who dubbed vCJD "human mad cow disease". An urgent need for the development of national and international public health policy supported by robust scientific evidence and facilitating effective communication of the risk to restore public confidence, was recognised.

The relationship between PHS in animal and human prion diseases

Whilst I have focused here on PHS of human prion diseases, the aetiological link between vCJD and BSE in cattle underlines the importance of concurrent PHS for animal prion diseases. The need for PHS of animal prion diseases is of increasing importance for two reasons. Firstly, there have been calls for the relaxation of the

control measures instigated during the BSE epidemic. On-going surveillance is essential to ensure that BSE does not re-emerge if control measures are removed or relaxed. Secondly, atypical forms of prion disease in animals are being described with increasing frequency. The risk that these atypical diseases may present to human health has not yet been quantified. Issues relating to the surveillance of prion diseases in animals have been reviewed and will not be examined further.

Methods of PHS

PHS can be passive or active. In passive surveillance cases are ascertained by direct referral (without prompting) or through other mechanisms without prior solicitation from the PHS system. For example a clinician aware of the PHS system might refer a suspect case without prompting, or vital statistics such as death certificates might be used to monitor trends in mortality. This approach has a number of advantages. It is (relatively) inexpensive, can be useful if covering a large geographical area with a disparate population and if consistent methods are applied over time can provide valuable information to assess temporal trends in disease occurrence. There are however a number of caveats that should be considered. This approach often relies on routinely collected data which may have extremely limited clinical information and a low sensitivity for detecting cases. Depending on the objectives of the PHS system such data may not be fit for purpose. The representativeness of the data should also be considered. For example voluntary notification of suspect cases by clinicians or other health care professionals may result in an excess of notifications from those with a specialist interest in the area and a dearth of referrals from others. This bias can lead to under-ascertainment or simulate clustering of cases.(70)

In active surveillance cases are actively sought by the PHS system. For example, by contacting health care professionals and reminding them to notify the PHS system of suspect cases, or conducting special surveys in targeted populations. These approaches are likely to produce more detailed information, be more sensitive methods of ascertaining cases, and subject to less bias. However as would be anticipated active surveillance is more expensive, and both labour and time intensive.

In practice PHS systems often use both active and passive surveillance. For example, cases may be ascertained through unsolicited direct referral from any health care professional but the PHS may regularly contact a targeted group of health care professionals reminding them to refer suspect cases. Periodic enhanced active surveillance in specific high risk groups might be carried out in time limited and externally funded studies based on research priorities. The approach adopted will depend upon the objectives of the PHS system and the resources available.

Strategies in the PHS of prion diseases

Three commonly used strategies to ascertain suspect cases include direct referral, specialist surveys and the use of routine data.(98) Each will be addressed in turn and the relative merits of each approach evaluated. The approach ultimately adopted by the surveillance system will depend upon a number of factors including the epidemiology of the disease, existing infrastructure and reporting mechanisms, the population under study (including the size and geographical distribution), the aims of objectives of the PHS system and the resources available. In practice most countries employ multiple and overlapping method of case ascertainment in the surveillance of CJD.

Direct referral

The PHS system receives direct referrals of confirmed or suspect cases from health care professionals and/or members of the public. This may be an entirely passive process in which unsolicited referrals are received. Alternatively the PHS system may issue frequent reminders to specific professional groups such as neurologists, neuropathologists and neurophysiologists, encouraging referral of all suspect cases. The professional groups targeted are typically those that have an increased likelihood of encountering cases based on the clinical illness.(72;98) Perhaps unsurprisingly reports received by this group are considered one of the most sensitive and specific strategies of case finding. Reports received from other health care professionals or the public are considered less sensitive.(72;98) Whilst assumptions have been made regarding the sensitivity and specificity of referrals made to PHS systems, contemporary data to support these assumptions are lacking. Prion diseases are rare with a diverse clinic-pathological phenotype. The assessment of clinical features and

investigations that support a diagnosis of prion disease requires skill and expertise. In this context the willingness of the PHS system to advise on the investigation and management of a suspect case may encourage direct referral of suspect cases.

The WHO recommend that the number of referrals received by a prion disease PHS system should exceed the number of confirmed cases by a factor of 2 or more to increase the likelihood of ascertaining cases.(98) If the surveillance system is not meeting this target, enhanced contact with referrers may be necessary. Many prion disease PHS systems have broad referral criteria reflecting the lack of a single, acceptable diagnostic test for prion disease in life in the context of a diverse disease phenotype, and a core objective of detecting novel prion disease. For this reason many systems also endeavour to clinically review suspect cases and/or any associated investigations. The ability of a PHS system to review a high proportion of non-cases will be determined in part by the resource available to operate the system. It should be considered that the number of direct referrals to a PHS system and hence the ratio of cases to non-cases referred to and reviewed by the system may be vulnerable to changes in awareness about prion diseases among the public and health care professionals in addition to the prevailing political agenda. For example in the UK following the characterisation of vCJD and intense media coverage a significant increase in the number of direct referrals received by the NCJDSU occurred which resulted in a change in the ratio of cases to non-cases being referred.

Many surveillance systems are centralised due to the rarity of prion diseases and the expertise required in the assessment and interpretation of surveillance data. Increasingly diagnostic services such as neuropathology or CSF 14-3-3 protein testing are offered by PHS systems. These services are valuable in ascertaining suspect cases not referred to the PHS system directly.(224)

Specialist Surveys

This approach involves enhanced active surveillance in sub-groups that are considered to be at greater risk of human prion disease than the general population. An obvious example would be enhanced surveillance in the family members of an

individual known to have a pathogenic *PRNP* mutation or individuals known to have received cadaveric-derived hGH during a specified time period.

In the UK a number of special surveys have been carried out. The Health Protection Agency (HPA) in the UK maintains a database of all individuals who have been informed that they are "at increased risk" of CJD for public health purposes; a second database containing information on individuals considered to be at low or uncertain risk of iCJD is also maintained. This latter group have not been informed of their status. This database facilitates rapid identification and comprehensive long-term follow up of individuals potentially exposed to CJD through medical interventions. A complementary prospective cohort study, The National Prion Monitoring Cohort, co-ordinated by the National Prion Clinic (NPC) is currently recruiting patients diagnosed with, or at high risk of, all forms of human prion disease. In addition to contributing to surveillance activities, research of this nature offers an invaluable opportunity to document the natural history of human prion disease and explore diagnostic and therapeutic opportunities. However such studies are expensive, require high levels of participation and low rates of attrition to ensure adequate statistical power to provide meaningful results.

Three further UK based studies are worthy of mention. The TMER study is a collaborative study between the UK Blood Transfusion Services (UKBTS) and the NCJDSU.(179) The UKBTS are notified of definite or probable sCJD, vCJD and genetic prion disease cases ascertained by the NCJDSU. The UKBTS determine whether the case has received a blood transfusion or is a blood donor. If the case has been a recipient, the donor is in turn traced; if the case has been a donor, the recipient is in turn traced. The aim of this study is to identify episodes of transfusion transmitted infection. Through this study four episodes of transfusion transmitted vCJD have been identified (one asymptomatic); no episodes of transfusion transmitted sCJD or genetic prion disease have been identified.

Active surveillance among individuals with congenital or acquired haemophilia who received UK sourced plasma products between 1980 and 2001, was initiated in 2001

in a collaborative study between the UKHCDO and the NCJDSU.(182) This study involves the prospective and retrospective examination of tissue removed from the LRS or central nervous system during surgical procedures in life or at autopsy following death in patients with haemophilia in the UK. Informed consent is required for tissue to be examined, either provided by the patient, or their next of kin if the patient is deceased. The success of the study will be determined, in part, by participation rates. To date, tissue from 17 neurologically normal haemophiliac patients has been examined. PrP^{Sc} has been detected in the spleen of one deceased patient.

Finally, the Progressive Intellectual and Neurological Deterioration (PIND) study uses an existing surveillance network among highly motivated paediatricians to identify vCJD in children.(225) This strategy is effective for a number of reasons. The existing paediatric surveillance system has extremely high participation rates. Referrals to the surveillance system are made prospectively, while patients are alive, therefore the identification of a potential case facilitates rapid public health action if required. Operationally clinical information on all PIND cases is reviewed by a panel of expert paediatric neurologists in an attempt to reach a diagnosis. This step is extremely important because the rate of post mortem among children with PIND is surprisingly low. Finally, the clinical phenotype of vCJD in children is not well described. To date six vCJD cases in children have been ascertained through this study.

This is not an exhaustive account of enhanced surveillance efforts, rather an outline highlighting some of the strategies adopted to improve surveillance in specific sub-populations in the UK. Integral to the success of these strategies is the co-operation of a diverse range of agencies external to the PHS system and of course the remarkable willingness of patients and their significant others to participate in surveillance.

Routinely collected data

Readily available and low cost, routinely collected and collated morbidity (hospitalisation) and mortality (death certificates) data can be used in PHS. Death

certification by a clinician is a statutory requirement therefore coverage is universal. These data are used for official purposes and are generally current. The rapid clinical course in sCJD and universal fatality of the condition means that mortality data are a reasonable proxy measure for incidence; this is not necessarily so for other prion diseases. Suspect cases must be deceased before they will be identified therefore prompt public health action is not facilitated by this method of case finding. In addition only individuals recorded by the certifying doctor as having died of CJD or as having a co-morbid diagnosis of CJD that contributed to but did not directly cause death will be identified. Some studies utilising death certificates examine only the underlying cause of death, that is the condition that led directly to death.(47;51) Others consider multiple causes of death, including the underlying cause of death and any co-morbid conditions that may have contributed but not led directly to death.(50)

The information recorded on death certificates is routinely coded using the International Classification of Diseases (ICD) 9 (pre-1996) and 10 (post-1996). The accuracy of death certificates in identifying prion disease cases therefore depends upon the accuracy of both the diagnosis (as made by the certifying clinician) and the accuracy of ICD coding. The USA consider the periodic analysis of death certificates to be the most "*systematic and cost effective*" method of disease surveillance in CJD.(50) A number of studies examining temporal trends in CJD mortality using routinely collected death certificate data have been published.(47;49;51;226;227) Advocates of the use of death certificates in surveillance usually quote a 1995 manuscript by Davanipour *et al* which examined 69 neuropathologically confirmed sCJD cases reporting that 80% were identified by death certificate review; a false positive rate of 8.3% was noted. The authors concluded that the examination of death certificates was a reliable and sensitive method of case finding compared to alternative strategies (direct referral from neuropathologists and review of hospital records). Other studies have produced less convincing evidence of the utility of death certificates. In Italy, Conti *et al* compared data from the National CJD Surveillance Service to official death records. The authors reported misclassification of CJD status by death certificates in up to 50% of cases from 1996 to 1999. In the UK, Will *et al* found that just two thirds of cases certified as dying from sCJD met the WHO

diagnostic criteria as a definite, probable or possible cases of sCJD in the 1980s and early 1990s; use of death certificates as the sole method of case ascertainment would have resulted in 22 – 28% of definite or probable sCJD cases being missed.(72)

In the UK, review of death certificates is used as a "safety net" to maximise case ascertainment and as a method of follow up of non-cases referred to the NCJDSU.(72) To date no studies examining the changing sensitivity of death certificates over time, or the impact of age on the sensitivity of death certificates in the surveillance of prion disease have been published. It might be expected for example that the sensitivity of death certificates would be greater in younger patients who may have been more thoroughly investigated and have undergone post mortem examination, than in older patients. It is questionable whether a system reliant solely on examination of vital statistics would have detected vCJD and very unlikely that such a system would have been able to rapidly characterise the condition to facilitate prompt public health action.

In the UK, virtually all elective and emergency health care is provided free at point of access by the National Health Service (NHS). Data are routinely collected on all episodes of hospital care in the UK. Given the clinical course of illness associated with prion disease, the majority of suspect cases will, at some stage in their clinical illness, be hospitalised and may therefore be detected in an examination of hospitalisation records. Elsewhere organisational and structural differences in access to and use of health care services may profoundly influence case finding using hospitalisation data. Cultural differences in health seeking behaviour may exist between or within counties resulting in certain groups, for example socioeconomically deprived or ethnic minority groups, being less likely to engage with medical services. This should be considered in the interpretation of hospitalisation data.

There are a number of other weaknesses of hospitalisation data. As for death certificate data, hospitalisation data usually record a variable number of discharge diagnoses for each episode of hospital care. These discharge diagnoses are then ICD

coded. As for death certificate data, the accuracy of these data is determined by the accuracy of both the diagnoses and the ICD coding. To the best of my knowledge, there are no published data specifically addressing the accuracy, sensitivity, specificity and completeness of hospitalisation data in the surveillance of prion disease.

The use of routinely collected data is advantageous because it is readily available, inexpensive and often contemporary. It is important to consider whether the data available are fit for purpose and whether the limitations of the data (accuracy, sensitivity, specificity, coverage, potential biases) are acceptable.

Compulsory reporting of prion disease

In some countries, for example Australia, Austria, Ireland, France, Germany and Sweden, prion diseases are notifiable, there is a statutory requirement for clinicians to report prion disease cases to the PHS system. In other countries, such as the UK, there is no legal obligation for health care professionals to report cases; the reporting of cases to the PHS system relies upon the co-operation of health care professionals and the public. Compulsory reporting of prion diseases may be adopted as a means of maximising case ascertainment, monitoring trends in disease and facilitating prompt public health action. A key issue that must be considered is the definition of a 'case'. For example in Australia notification is required if there is a strong clinical suspicion of CJD whilst in Austria there is a compulsory requirement to inform the PHS system of neuropathologically confirmed cases only. An unintended effect of the former approach to compulsory reporting might be that clinicians defer notification of suspect cases to the PHS system until case confirmation is available. Indeed following the introduction of compulsory notification in Slovakia, referrals to the surveillance system fell.(24) The decision by authorities in the UK not to make CJD a notifiable disease on the grounds that this might reduce the number of suspect cases referred to the PHS system, particularly atypical cases that did not fulfil the diagnostic criteria, was vindicated by the BSE enquiry.(24)

A final point to consider is that of compulsory autopsy. In Austria for example neuropathological examination of all suspect prion disease cases is mandatory.

Whilst such an approach results in high levels of case confirmation it may not be culturally acceptable. In the UK there is no legal requirement for a suspect prion disease case to undergo autopsy examination on expiration, unless instructed to do so by a coroner or in Scotland a procurator fiscal.

The challenges of PHS of human prion diseases

The detection of human prion diseases through PHS is difficult for a number of reasons. Human prion diseases are exceptionally rare. In surveillance systems employing direct referral, case ascertainment requires a high level of co-operation from health care professionals, patients and their significant others. The clinical and neuropathological phenotype of human prion disease is diverse therefore prion disease may not be considered as a differential diagnosis in life. This is compounded by a lack of a simple and acceptable ante-mortem diagnostic test. Diagnosis in life requires the application of diagnostic criteria based on clinical features and supportive investigations (Appendix 2). Specialist expertise is required in assessing clinical features and conducting and interpreting investigations. An excellent example of this is the EEG in sCJD. Whilst objective criteria have been adopted by the WHO for the assessment of EEG for case classification in sCJD, these have not been prospectively validated and due to practical issues around the application of these criteria the assessment of EEG in many European countries including the UK remains largely subjective. The interpretation of EEG in the hands of a general neurophysiologist may be very different from the interpretation of the same EEG in the hands of an expert in the field of prion disease. To ensure adequate clinical and diagnostic expertise centralization of PHS is often necessary. This is expensive and may be logistically difficult if the PHS system covers a large geographical area.

Central to PHS is the requirement for a case definition of the event under study. A primary aim of the NCJDSU was the detection of a change in the clinico-pathological phenotype of CJD that might be attributable to BSE; vCJD was detected without a case definition.(37) The first case definition of vCJD was based upon the characterization of first ten cases ascertained by the NCJDSU.(223) Indeed it is only in the last year, 15 years after the first case, than the diagnostic criteria of vCJD have been validated.(186) In sCJD diagnostic technologies have evolved rapidly,

including CSF 14-3-3 protein, MRI, genetic analysis and molecular subtyping, necessitating regular review and updating of diagnostic criteria. The completeness of case ascertainment will, to an extent, be influenced by the quality of diagnoses in suspect CJD cases referred to the PHS system which in turn is dependent upon the introduction and application of these diagnostic technologies in clinical practice. The PHS system must be responsive to rapid transfer of scientific research into clinical practice.

Neuropathological examination following death of a suspect case referred to the PHS system is essential to achieve high levels of case ascertainment and a pre-requisite for identifying novel disease phenotypes which may not meet pre-defined diagnostic criteria for known disease phenotypes. In some countries the post mortem rate among hospitalised patients is extremely high. In the UK rates of post mortem, as previously noted have fallen in recent years, which may threaten the activities of the PHS system.

As demonstrated in the preceding pages, much of the progress that has been made in understanding human prion diseases has come directly from the study of animal prion diseases or indirectly from the use of animal models to study human disease. An effective PHS system will work collegiately with basic scientists in veterinary and human medicine, clinicians, electrophysiologists, neuroradiologists, neuropathologists, epidemiologists and public health specialists to translate scientific research into clinical practice and in turn disease surveillance. Data derived from the PHS system will be used to develop and evaluate national and international policy. Finally, in some countries, such as the UK, the PHS system interfaces directly with the general public, patients and their significant others, advising on treatment and care in suspect cases and communicating risk in lay terms.

One final challenge of PHS of prion diseases is the interpretation of surveillance data. Year to year fluctuations in the incidence of disease are extremely difficult to interpret due to the rarity of the disease. Temporal data must be interpreted with caution and consideration given to other factors (clinical practice, diagnostic

advances, public awareness, media attention, political will) that may have contributed, directly or indirectly, to any observed change. Direct comparison of national with internationally collected surveillance data can assist greatly in interpreting temporal trends in disease occurrence, but only if these data are equally robustly collected and valid.

International PHS systems in human prion diseases

National CJD PHS systems and disease registers had been operating throughout Europe since the 1970s.(54) In the early 1990s in response to the BSE epidemic several European countries including France (1992), Italy, Germany and the Netherlands (all 1993) established systematic prospective CJD surveillance systems. A number of international collaborative surveillance and research projects have been undertaken. These will be described in the section that follows.

EUROCJD

In 1993 an international research and surveillance project, EUROCJD, including seven collaborators (Austria, France, Germany, Italy, the Netherlands, Slovakia, Spain and the UK) received EU funding.(54) The aim of the study was to produce comparable international data describing the epidemiology of CJD in Europe. In 1996 the network expanded to include Switzerland and non-European collaborators in Australia and Canada. The same year a survey of the surveillance methodologies used in EU member states identified significant variation in the application of diagnostic criteria and reporting between countries, thereby threatening the comparability and hence utility of surveillance data.(228) The study suggested that harmonisation of PHS methodologies would facilitate collective and comparative examination of international data. The first steps recommended were the application of common diagnostic criteria, the creation of a minimum dataset for reporting and the use of internationally agreed surveillance methods. Common diagnostic criteria were adopted, a minimum dataset defined and strategies for conducting disease surveillance described in the 2003 'WHO Manual on the Surveillance of Human TSE'.(98) The EUROCJD network has continued to expand, now including 34 countries worldwide each providing data from their national surveillance systems

(Figure 12). In turn the aims of the group have evolved to focus on the detection and characterization of vCJD and other forms of novel prion disease in humans.

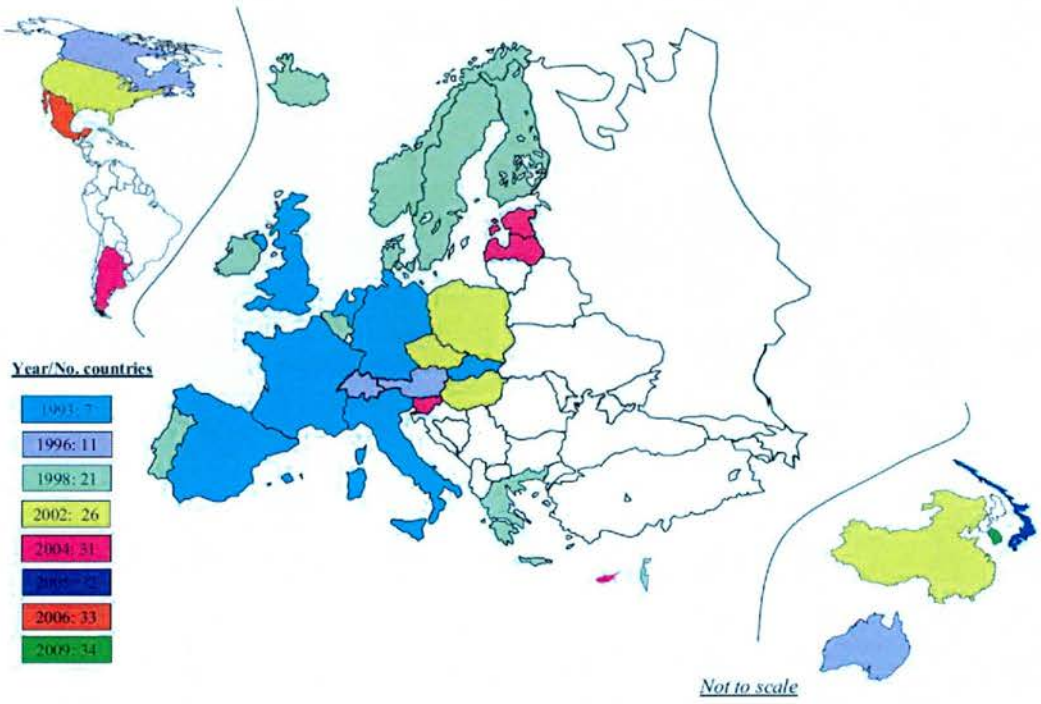

Figure 12 European CJD Surveillance 1993 – 2009, adapted from Will (229)

NEUROCJD

In 1998 a further surveillance network, NEUROCJD, was formed. Members included Belgium, Denmark, Finland, Greece, Iceland, the Republic of Ireland, Israel, Norway, Portugal, Sweden and the UK. The principal aim of this group was harmonization of PHS methodologies. With the exception of the UK all member states had a population of less than 12 million people. Some of the difficulties in carrying out surveillance of a rare disease in a large country with a geographically dispersed population have been discussed. There are also significant challenges faced by smaller countries. In a country such as Iceland for example with a population of a little over 300,000 it may be several years before a single case of sCJD (incidence approximately 1 per million population) is detected by the PHS system. Yet significant financial resource and expertise is required to operate a PHS system, the sustainability of which is questionable. The NEUROCJD network offered the

possibility for small countries to collaborate to share diagnostic and public health expertise and set research priorities. As for the EUROCJD network, members of this consortium retained autonomy over the operating characteristics of their PHS systems but common standards were adopted for the application of diagnostic criteria, diagnostic methodologies and reporting of surveillance data. Of note, the NEUROCJD network is no longer in operation.

SEEC-CJD

In 2001 a further collaborative surveillance network was established covering central and eastern European countries and China, with similar aims to the EUROCJD project.

Operational characteristics of international PHS systems

Beyond brief accounts of national PHS systems included in the methods sections of published studies that report the epidemiology of prion disease in specific populations, data describing and comparing the operating characteristics of prion disease PHS systems are surprisingly sparse. I am aware of two studies, both unpublished, describing the operating characteristics of selected members of the EUROCJD and NEUROCJD groups.(230;231) Data from these studies is reproduced in Table 18. The primary data collected by these studies was different therefore a comprehensive picture of the operational characteristics of international prion disease PHS systems is not available. However, a number of general observations can be made. Firstly, with one exception all PHS systems were centrally operated. In common, most countries utilized more than one approach, epidemiological, neuropathological and/or neurological, to surveillance. In almost three quarters of countries there was a statutory obligation to report prion disease cases to the PHS system. Analysis from the NEUROCJD network suggests that, contrary to previous evidence, no significant change in reporting of cases occurred in countries in which prion diseases became notifiable during the period covered by the study.(230) In general the criteria for referral to the PHS systems were poorly described; for many referral of 'all suspect cases' were the only stated criteria. As previously noted there are difficulties in setting strict criteria for referral to a prion disease PHS system given the diverse clinico-pathological phenotype and the aim of

Table 18 Operating characteristics of prion disease PHS systems in EUROCJD (1998) and NEUROCJD countries (1997 - 2004), adapted from Pedro-Cuesta et al (231) and Sanchez-Juan (230)

Country	Nature of PHS	Notifiable?	Criteria for notification and methods of case ascertainment	Suspect cases seen in life?
Australia[E]	Epidemiology/ Neuropathology	Yes	Referral criteria not specified Death certificate, Hospital records, Special surveys, Laboratory (CSF 14-3-3 protein), Other	No
Austria[E]	Neurology/ Epidemiology/ Neuropathology	Yes	Referral criteria not specified Special surveys, other	Yes
Belgium[N]	Epidemiology/ Neuropathology	No	Direct referral all suspect cases	No
Denmark[N]	Epidemiology/ Neuropathology	Yes	Direct referral all suspect or definite cases, Death certificates	No
Finland[N]	Neurological/ Neuropathology	Yes	Direct referral all suspect cases, Death certificates, Hospital records	Partial
France[E]	Epidemiology	Yes	Laboratory (CSF 14-3-3 protein), other	No
Germany[E]	Epidemiology/ Neurology/ Neuropathology	Yes	Direct referral all suspect cases, Laboratory (CSF 14-3-3 protein), other	Yes
Greece[N]	Neurological/ Neuropathology	Yes	Not known	Partial
Iceland[N]	Neurological/ Neuropathology	No	Not known	Yes
Ireland[N]	Neurological/ Neuropathology	Yes	Direct referral all suspect cases	Partial
Israel[N]	Epidemiology/ Neuropathology	Yes	Direct referral all suspect cases, Death certificates, Hospital records, Neurogenetics laboratory	Partial

[N]=NEUROCJD; [E]=EUROCJD; *regional service, all other services centralised

Table 18 cont'd. Operating characteristics of prion disease PHS systems in EUROCJD (1998) and NEUROCJD countries (1997 - 2004), adapted from Pedro-Cuesta et al (239) and Sanchez-Juan (238)

Country	Nature of PHS	Notifiable ?	Criteria for notification and methods of case ascertainment	Suspect cases seen in life?
Netherlands[E]	Epidemiology / Neurology	No	Direct referral all suspect cases, Laboratory (CSF 14-3-3 protein)	Yes
Norway[N]	Epidemiological	Yes	Direct referral all suspect or definite cases, Death certificates	No
Portugal[N]	Epidemiology/ Neuropathology	Yes	Direct referral all suspect cases	No
Spain[E*]	Epidemiology/Neuropathology	No	Referral criteria not specified. Death certificates, hospital records, laboratory (CSF 14-3-3 protein)	No
Sweden[N]	Epidemiology	Yes	Not specified	No
Switzerland[E]	Epidemiology/Neuropathology	Yes	Referral criteria not specified Laboratory (CSF 14-3-3 protein)	No
UK[N,E]	Epidemiological / Neurological / Neuropathology	No	Direct referral all suspect cases, Death certificates, Special surveys, Laboratories (CSF 14-3-3 protein, genetics)	Yes

[N]=NEUROCJD; [E]=EUROCJD; *regional service, all other services centralised

many PHS systems to detect novel or atypical prion diseases. The application of diagnostic criteria at referral might result in such cases being missed. However broad criteria might result in a high proportion of suspect cases being referred which do not have prion disease with the PHS system being unable to respond. There was significant variation in the strategies adopted to ascertain cases between systems, although most employed multiple approaches to case ascertainment including direct referral, review of routine data and/or laboratory reports. Finally only half of all PHS systems reviewed suspect cases in life. Whilst reviewing suspect cases in life is considered by many to be the most sensitive approach to surveillance, the cost and logistical difficulties associated with this may be prohibitive. Lack of clinical review of suspect cases might be expected to affect levels of case ascertainment given that the diagnostic criteria, in the absence of neuropathological confirmation, require assessment of the presence of core clinical features in addition to supportive investigations. However mortality rates for sCJD are remarkably consistent between countries over time, ranging from 0.53 – 1.7 per million population. Moreover countries in which suspect cases are not reviewed by a neurologist have ascertained incident vCJD cases.(46) To fully interpret these data rates of post mortem examination among suspect cases referred to the PHS systems would be required. Unfortunately these data were not available for all countries. Within the NEUROCJD consortium post mortem rates ranged from 40% to 100%. Where post mortem rates are high this may result in an apparent excess of cases simply because the diagnosis is confirmed in individuals that do not meet the diagnostic criteria.(54) In countries where post mortem rates are low obtaining clinical data on as many suspect cases as possible is extremely important.

These data confirm significant international variation in operating characteristics of prion disease PHS systems. However, irrespective of the methodological approach to surveillance adopted, mortality rates from surveillance data appear broadly consistent. The application of common diagnostic techniques, diagnostic criteria and standardised reporting have ensured that the data reported are fit for purpose.(46) On-going monitoring to ensure that this continues to be the case as PHS systems expand and mature, and new PHS systems emerge, is crucial. Timely and robust

international comparative data are vital in the interpretation of changes in disease occurrence or clinico-pathological phenotype that may occur nationally.

Evaluation of PHS systems

A vital, yet often overlooked, step in PHS is evaluation. Evaluation can be used to determine whether the PHS system is fit for purpose and meeting its stated objectives. Periodic evaluation of PHS systems is recommended to ensure that surveillance is both efficient and effective.(222) Detailed guidelines on the evaluation of PHS systems have been produced by the Centre of Disease Prevention and Control (CDC).(222) These guidelines outline the attributes of the PHS system, including simplicity, flexibility, data quality, acceptability, sensitivity, predictive value positive, representativeness, timeliness, and stability, that should be considered in evaluation. It is remarkable that despite the considerable investment in global prion disease PHS, the significant changes that have occurred over the two decades in which systematic prospective prion disease PHS has been in place and the political and public health imperative of ensuring that surveillance is robustly conducted, published examples of evaluations of prion disease PHS systems are vanishingly rare.

Two evaluations of the EUROCJD network (one unpublished) have been produced.(231;232) An evaluation of the EUROCJD network was carried out by the European Centre for Disease Prevention and Control (ECDC) in 2007.(232) The only published account of this evaluation is a summary report which fails to describe the evaluation methodology or provide a detailed account of the results of the evaluation. The summary report concluded by recommending improved reporting of epidemiological data collected through surveillance, improved database management with the developing of standard operating procedures (SOPs) to support this, improved communication with national epidemiologists, and collaboration with non-EU surveillance systems. An earlier, unpublished, evaluation of the EUROCJD network by Pedro-Cuesta *et al* (2003) applied the 1998 CDC methodology for the evaluation of PHS systems. The challenges of examining system attributes such as sensitivity and PPV in the absence of a gold standard ante-mortem test and variable post mortem rates were highlighted. The authors found significant variation in rates

of referral of suspect prion disease cases to PHS systems among the young (< 50 years old), with greater referral rates in countries that had previously reported a high incidence of specific prion disease (for example genetic prion disease). International variation in the quality of diagnosis based on the use of diagnostic technologies (including post mortem, CSF 14-3-3 protein and genetic analysis) in sCJD cases that met the diagnostic criteria, and delays in reporting, were also noted.

Two further evaluations of national surveillance systems have been carried out. Robotin *et al* evaluated the Australian Surveillance System in 2002 applying the CDC criteria.(233) Recommendations to improve the sensitivity and timeliness of the PHS system were made. It was not possible to fully assess certain aspects of the surveillance system as vCJD has never been detected in Australia. More recently in 2008 the Canadians undertook an evaluation of their National Prion Disease Program (1998 – 2008); PHS is one of three core activities of this program.(234) This external evaluation used both quantitative and qualitative approaches to examine the relevance, success, design and delivery of the Prion Disease Program. The study did not examine the system attributes described by the CDC therefore direct comparison with the Australian study is not possible. The evaluation noted that whilst the PHS system appeared be delivering well with a high level of satisfaction among the health care professionals that used the system a number of areas could be improved. These included the development of SOPs to ensure consistency in operating procedures when staff changed, entry of epidemiological data onto a database to facilitate analysis and dissemination of findings beyond reporting of the minimum monitoring dataset, and improved engagement with more remote areas within Canada. One further issue not raised in this evaluation but of importance should be highlighted. It has been said that

> *"the strength of an evaluation depends on the ability of the evaluator to assess these characteristics [the attributes of the surveillance system] with respect to the system's objectives."(235)*

The formal objectives of the Canadian surveillance system were not clearly defined when this evaluation took place. Indeed the authors of the evaluation reported that a

> *"vision for the CJDSS [CJD Surveillance system] appears to be developing,"* and that *"there is limited awareness of the formal mission and objectives by PDP[Prion Disease Program] managers and staff."*(234)

These evaluations are unlikely to be applicable to the UK. The PHS systems differ from that in UK with respect to their methodologies and objectives. The system attributes that are crucial to the success of each of these national or indeed international collaborative PHS systems, will differ from those in the UK. The CDC note that an evaluation must

> *"consider those attributes that are of the highest priority for a given system and its objectives."(222)*

The Australian evaluation for example was unable to adequately assess the flexibility of the surveillance system in response to vCJD because they had not detected a vCJD case. This is significantly different from the UK which has experienced the greatest number of vCJD cases worldwide.

The need for an evaluation of the NCJDSU

Systematic prospective CJD surveillance has been carried out in the UK since 1990. During this time a novel human prion disease attributed to BSE in cattle has been characterised and previously unrecognised routes of human to human transmission of prion disease identified. Significant advances in diagnostic technology have led to the revision of the diagnostic criteria applied by the NCJDSU in case classification. Operationally the NCJDSU has responded to these changing demands. There has never been an evaluation of the CJD surveillance system in the UK. There is an urgent need for such a study.

Summary

- Prion diseases are rare, invariably fatal neurodegenerative diseases affecting animals and humans for which there is no effective treatment and no acceptable or practical ante-mortem diagnostic test.
- Aetiologically sporadic, genetic and acquired forms of disease exist.
- The infectious agent is thought to be a pathogenic isoform (PrP^{Sc}) of a normal cellular protein (PrP^{c}), the precise biological function of which is unknown.
- The binding of PrP^{Sc} to PrP^{c} results in self-replicating conformational change.
- Multiple prion strains are thought to exist, determined by their structural conformations and glycosylation patterns, and distinguished by their biological properties, including incubation periods and neuropathological profiles.
- The true nature of prions and the neuropathogenesis of prion disease are not well understood.
- BSE in cattle is the only animal prion disease known to be zoonotic.
- The origin of BSE is unknown; the epidemic in the UK was propagated through intensive farming practices. Control measures interrupted the epidemic, but not before the widespread exposure of the UK population to BSE.
- In humans the commonest form of prion disease is CJD.
- Systematic prospective PHS of CJD was initiated in the UK to identify any change in the clinico-pathological phenotype of CJD attributable to BSE.
- In 1996 the NCJDSU in the UK characterised vCJD, a novel human prion disease aetiologically linked by epidemiological and transmission studies to BSE.
- The vCJD primary epidemic has been smaller than many feared and in decline in the UK since 2000. However with the potential for exceptionally long incubation periods (> 50 years) this may continue at a low level for many years to come.
- There has been clear evidence of genetically determined susceptibility/resistance and/or genetically determined incubation periods in vCJD.
- Secondary transmission of vCJD through the transfusion of labile blood components, a novel route of transmission, has been identified with transmission occurring during an asymptomatic stage. To date no further routes of transmission have been identified as yet.

- Given the widespread exposure of the UK population to BSE there is potential for a self-sustaining secondary vCJD epidemic if a large population of asymptomatic but infectious individuals exist.
- Population prevalence estimates of asymptomatic vCJD infection in the UK have ranged from 0 to 853 per million population, although the studies that produced these estimates have significant methodological limitations.
- The natural history and pathogenesis of vCJD is poorly understood. The significance of detecting abnormal prion protein in these studies is unknown.
- Despite scientific uncertainty public health measures have been instigated to minimise the risk of human to human transmission of vCJD.
- The success of control measures in preventing human to human and animal to human transmission of prion disease can only be determined by on-going monitoring of epidemiological trends in disease through PHS.
- Robust comparative data are vital to interpret national PHS data.
- The surveillance of all forms of CJD has led the recognition of an increasingly diverse clinico-pathological phenotype of sCJD and genetic prion disease; genetic prion disease may be clinically indistinguishable from sCJD.
- Emergent diagnostic technologies have been incorporated into increasingly sensitive and specific clinical diagnostic criteria, although an acceptable and practical ante-mortem diagnostic test remains elusive.
- The relationship between molecular genetics and clinico-pathological phenotype is becoming increasingly complex and uncertain in sCJD.
- Despite intensive epidemiological study the underlying aetiology of sCJD remains unknown; determining the risk of secondary transmission is challenging although novel routes of transmission have not emerged in three decades.
- Through active surveillance atypical animal prion diseases have been described. The threat they pose to human health is as yet unknown however, recent evidence suggests a possible link between atypical prion disease and some sCJD subtypes.
- There is a clear rationale for the on-going systematic prospective PHS of prion diseases in the UK.
- Periodic evaluation of the PHS system is crucial to ensure that the system meets its objectives. This forms the basis of this thesis.

Aims and objectives of this thesis

In this thesis I will evaluate various aspects of the surveillance of human prion diseases in the UK from 1990 through 2006. Consideration was given to this overall aim and the critical gaps in the evidence base identified from the literature review. This aim was in turn translated into the following objectives:

1. To describe the epidemiology of prion disease in the UK according to disease subtype using surveillance data collected by the NCJDSU, from 1990 through 2006

2. To evaluate the NCJDSU using CDC guidelines for evaluating PHS systems as a standard for assessing the performance of the system (1990 – 2006)

3. To prospectively validate the operational criteria for the assessment of EEG in the surveillance of suspect sCJD in the UK (2005 – 2006)

4. To evaluate the use of death certificates in the surveillance of prion disease in the UK (1990 – 2006)

Some general definitions

A number of definitions will be applied throughout this thesis.

- 'Suspect case' will denote an individual referred to the NCJDSU as a suspect prion disease case. This does not make any inference about case classification.
- Unless stated otherwise, the term 'case' will refer to a definite or probable prion disease case as defined in the WHO diagnostic criteria (Appendix 2).
- Unless stated otherwise, the term 'non-case' will refer to a suspect case that does not fulfil the WHO diagnostic criteria as a possible (or greater) case.
- Unless otherwise stated case classification refers to case classification at the time of data censoring.
- Where reference has been made to a specific aetiological subtype of prion disease this will be defined, for example sCJD will be used to describe sporadic CJD. The term 'prion disease' will be used to denote all forms of human prion disease.
- The following abbreviations will be used throughout: sCJD to denote sporadic CJD, vCJD to denote variant CJD and iCJD to denote iatrogenic CJD and gCJD to denote genetic CJD.
- Where the term 'CJD' appears without identification of an aetiological subtype this will also denote all forms of human prion disease.
- The term genetic prion disease will encompass all forms of genetic prion disease including GSS, FFI and genetic CJD.

Chapter 2. The epidemiology of prion disease in the UK, 1990 – 2006

Introduction

In chapter 1, I outlined the epidemiology and diagnostic features of prion disease according to aetiological subtype using a literature search strategy. There have been numerous developments in the 16 years since systematic prospective PHS of CJD was initiated in the UK. These include, but are not limited to, the identification of a novel human prion disease, the identification of novel routes of disease transmission, the characterisation of an increasingly diverse clinico-pathological phenotype of known prion disease, emergent diagnostic technologies and the incorporation on these technologies into clinical diagnostic criteria. In this chapter I will use data collected by the NCJDSU from 1990 through 2006 to describe the epidemiology and diagnostic features of prion disease in the UK according to disease subtype. These data will provide essential context for the forthcoming chapters evaluating various aspects of disease surveillance in the UK.

Aim

The aim of this chapter was to describe the epidemiology and diagnostic features of prion disease in the UK using data collected by the NCJDSU from 1990 through 2006.

Methods

Data collection

All suspect prion disease cases referred to the NCJDSU between 1st May 1990 and 31st December 2006 were followed for a minimum of two years until 31st December 2008. An electronic search of the NCJDSU minimum monitoring dataset was carried out to identify all prion disease 'cases,' at the time of data censoring (as previously defined). The paper-based NCJDSU case record of each case was examined by hand and the following information extracted: sex, date of birth, date of death, date of symptom onset, clinical presentation, case classification, disease subtype (sporadic variant, iatrogenic or genetic), date of referral to NCJDSU and referral source. The

number, result and date of EEG, MRI and CSF 14-3-3 protein examinations were collected. In addition, details of genetic analyses, brain biopsy and post mortem examination were collected. In genetic prion disease cases, a recorded family history of prion disease in a first degree relative was extracted from the cases note. For iCJD cases, the route and date of exposure were recorded. Data extracted from case records were anonymised and entered onto a password protected database maintained on a desk top computer.

Definitions

Age, in years, was calculated at symptom onset, at referral to the NCJDSU and at death. Unless otherwise stated, age was treated as a continuous variable. Clinical presentation was determined by symptoms at onset and treated as a categorical variable. For sCJD this was based on criteria developed by Knight and Will (Appendix 3).(230) A 'typical clinical presentation' was considered as a clinical presentation of RPD, the Heidenhain variant or a cerebellar onset. An 'atypical clinical presentation' was defined as any presentation other than RPD, the Heidenhain variant or a cerebellar onset. For vCJD clinical presentation was described in one of 3 categories: neurological, psychiatric or both (neurological and psychiatric) characterizing the most prominent clinical features at onset (Appendix 4).(185) The source of referral was taken as the individual who initially contacted the NCJDSU to discuss the case; this was treated as a categorical variable.

All EEGs and MRI scans undertaken during the course of the clinical illness were requested for review by the NCJDSU. When available EEGs were reviewed by one of two senior neurologists (RGW, RK). EEG's were classified using a five point order categorical scale ranging from normal to 'typical'. According to the WHO diagnostic criteria a 'typical' EEG can be used to change the classification of a possible sCJD case to a probable sCJD case. In practice in the UK EEGs that are considered highly suggestive but not entirely typical may also be used for case classification based on the judgement of the reviewing clinicians. The operational criteria employed in the UK for the assessment of EEG for case classification will be discussed in greater detail in chapter 4.(98) For the purposes of this study an EEG that was used for case classification will be referred to as a 'typical'. Where the EEG

was unavailable for review by the NCJDSU, the EEG classification reported by the local clinician was used. The time from symptom onset to first typical EEG was calculated by subtracting the date of symptom onset from the date of first typical EEG. This was measured in months.

All MRI scans, where available to the NCJDSU, were reviewed by one of two designated neuroradiologists (DC, DS). Where unavailable, the local report was used. In sCJD the MRI was considered positive if there was evidence of high signal in the caudate nucleus and putamen or high signal in the striatum. In vCJD the MRI was considered positive if the 'pulvinar sign' was present, defined as

> *"a characteristic distribution of symmetrical hyper-intensity of the pulvinar nucleus (posterior nucleus) of the thalamus (relative to the grey matter of the anterior putamen and normal cerebral cortex)."*(98)

Negative scans did not meet these criteria. As noted in chapter 1 over time the optimal sequences for detecting changes consistent with a diagnosis of sCJD and vCJD on MRI scanning have been clarified, as have the specific features on MRI. This study examined unselected surveillance data prior to and following these developments. In this thesis I considered whether an MRI scan had been undertaken, not the specific sequences used, and if so whether the features on MRI were consistent with a diagnosis of sCJD or vCJD based on the above definitions. The time from symptom onset to first positive MRI scan was calculated by subtracting the date of symptom onset from the date of first positive MRI scan. This was measured in months.

The National CSF 14-3-3 protein Reference Laboratory has been located in the NCJDSU since 1997, although this investigation has been widely available in the UK since 1996. The CSF 14-3-3 protein examination is either positive or negative. The test can yield a 'weak positive' result. For the purposes of disease surveillance such tests are considered negative results. A test may be requested but not processed. This can arise because, for example, the sample is heavily blood stained or has been inappropriately stored. Under these circumstances the test result may be invalid and the laboratory will not process the sample. In such cases the test was recorded as not being undertaken. The time from symptom onset to first positive CSF 14-3-3 protein

examination was calculated by subtracting the date of symptom onset from the date of first positive CSF 14-3-3 protein examination. This was measured in months.

The results of genetic analysis, *PRNP* Codon 129 genotyping and full sequencing of *PRNP* for mutations, if performed, were recorded. Molecular subtyping according to the Parchi classification where available was described.(147) The date and result of tonsil and brain biopsies undertaken during life were recorded. The time from symptom onset to positive biopsy was calculated by subtracting the date of symptom onset from date of biopsy. This was measured in months. Post mortem examination was recorded. Illness duration was measured by subtracting date of symptom onset from the date of death. This was measured in months.

'Atypical sCJD cases' were defined as sCJD cases that were: aged under 50 years old at symptom onset, had an illness duration of 1 year or more and/or had a clinical presentation other than RPD, Heidenhain variant or cerebellar onset.

Statistical analyses

Data were cleaned and coded using the definitions outlined above. Cases were examined according to disease subtype: sCJD, vCJD, iCJD and genetic prion disease. For each disease subtype descriptive summary statistics were produced overall, and by year of referral. Where data were normally distributed this was presented as mean (standard deviation); skewed data were presented as median (inter-quartile range unless otherwise stated). Univariate parametric tests of association between key variables including age, sex, illness duration and year of referral were carried out (t tests, Chi^2 tests); where the assumptions of these tests were violated, non-parametric equivalents were used (Fisher's exact test, Wilcoxon Ranksum test, Kruskal Wallis test). Chi^2 tests for trend (or non-parametric equivalents where appropriate) were used to compare proportions over time. The sensitivity of diagnostic investigations (such as EEG, MRI and CSF 14-3-3 protein) was calculated as the proportion of cases (according to disease subtype) that had a positive test result among those cases (according to disease subtype) that underwent the investigation. The rate of post mortem, presented as a percentage, was calculated as the number of cases undergoing post mortem each year divided by the number of deaths that year.

The NCJDSU has a limited remit in relation to the clinical surveillance of iCJD and genetic prion disease, collecting a minimum reporting dataset on these cases only. For these disease subtypes analyses were limited to basic descriptive statistics. The NCJDSU is responsible for clinico-pathological surveillance of sCJD and vCJD therefore more detailed analyses were carried out for these diseases.

Incidence and mortality rates

Annual age and sex-specific incidence and mortality rates of sCJD and vCJD were calculated using denominator data from mid-year population estimates in the UK. Incident cases were defined by year of referral to the NCJDSU; mortality by year of death. Age standardised incidence and mortality rates of sCJD and vCJD were calculated using data from the 2001 Census data and the direct method. A joinpoint regression model was fitted to estimate the annual percentage change (APC) in age-adjusted and age-specific sCJD and vCJD incidence and mortality rates in men and women, and to detect time points at which a significant change in the overall trend occurred. To select the best-fitting model Bayesian Information Criterion (BIC) was used. A maximum of three join points were allowed for each estimate. A corresponding 95% confidence interval (CI) was calculated for each APC estimate.

Crude and adjusted case-fatality

Crude and adjusted case-fatality was calculated from the date of symptom onset. For sCJD crude case-fatality at 6 months and 1 year was calculated as the proportion of all cases that were dead at each time point; for vCJD case-fatality was calculated at 1 and 2 years reflecting the longer median survival in this disease. Median survival was calculated using time from symptom onset to death. Log rank tests were used to test for differences in survival experience by age group, sex, clinical presentation, year of onset and molecular subtyping for sCJD and age group, sex, clinical presentation and year of onset for vCJD. A Cox proportional hazards model, a regression method for survival data, was used to analyse case-fatality. The Cox model estimates hazard ratios and 95% confidence intervals. A hazard is the rate at which an event occurs, in this case death. The hazard ratio is the hazard in one group relative to the hazard in a comparison group, for example the hazard of death in the youngest relative to the oldest age group, or the hazard of death in men relative to

women. In modelling case-fatality adjustment was made for age group, sex, year of onset and molecular subtype for sCJD. Molecular subtype was selected rather than clinical presentation because the literature indicates that molecular subtype determines clinical phenotype.(147) For vCJD, analyses were adjusted for age group, sex, year of onset and clinical presentation (among those tested to date all vCJD cases have the same molecular subtype). Where a case was known to be alive, survival time was censored at 31st December 2008 (minimum of 2 years follow up). The proportional hazards assumption was tested using Schoenfeld residuals and was met for each of the models at each time point.

Missing data

For all dates, where the month and year were available this was used and the mid-point of the month, the 15th, imputed. Where the day was missing but the start or end of the month had been specified the 1st and 30th respectively were imputed. Where the day and month was missing these data were excluded from analyses. In such instances the number of observations that analyses are based on is noted. In calculating crude and adjusted case-fatality date of onset was missing for 14 cases (all sCJD). These were excluded from further analyses. These cases did not differ significantly with respect to age (P=0.297) or sex (P=0.702) from all other cases.

With the exception of regression analyses, all other analyses were carried out using STATA Version 10 (Stata Corp. College Station, Texas, USA). Regression analyses were carried out using Joinpoint Regression Program (Version 3.4.3). A level of statistical significance of 0.05 was used throughout.

Results

In total 1228 prion disease cases were ascertained by the NCJDSU between 1st May 1990 and 31st December 2006; 935 (76.1%) definite and 293 (23.9%) probable cases. Overall, according to disease subtype, 893 (74.9%) were sCJD, 165 (13.5%) vCJD, 116 (9.4%) genetic prion disease and 54 (4.4%) iCJD cases. Over time there was a significant change in the disease subtypes ascertained by the NCJDSU (P<0.001) as can be seen in Figure 13.

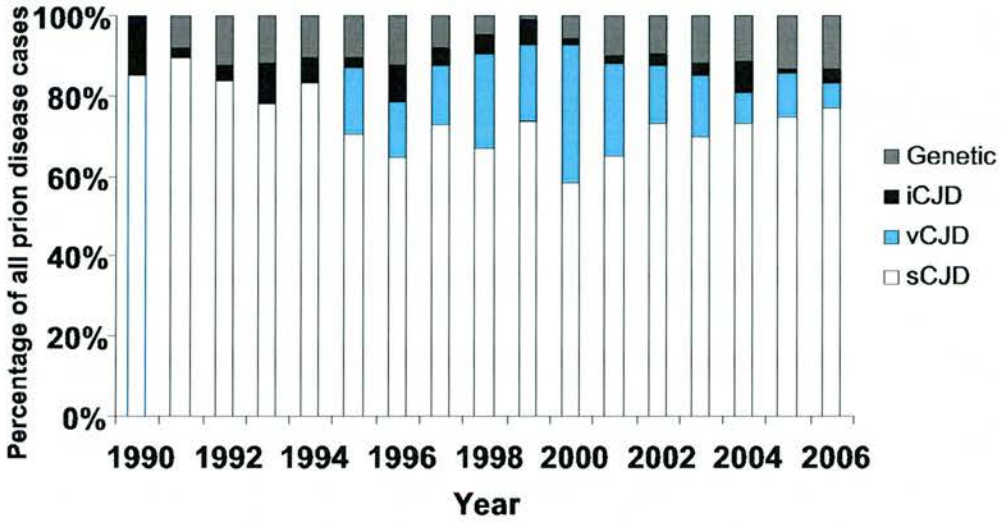

Figure 13 Distribution of disease subtypes of all prion disease cases ascertained by the NCJDSU, 1990 – 2006

Sporadic CJD

From 1st May 1990 to 31st December 2006, there were 893 incident sCJD cases in the UK, 689 (77.2%) definite and 204 (22.8%) probable sCJD cases (Table 19). In total 432 (48.4%) cases were in men; 337 (49.1%) definite and 95 (46.6%) probable cases. There was no significant difference in the proportion of definite or probable cases according to sex (P= 0.346). However there was a significant reduction in the proportion of all cases for which a neuropathological diagnosis was available over time, from 88.2% (15) in 1990 to 61.5% (40) in 2006 (P<0.001). The median age at symptom onset was 67.1 years (IQR 60.6 – 74.2). This did not vary according to sex (P=0.476) or case classification (P=0.223). The youngest sCJD case was 15.6 years

old at symptom onset, the oldest 94.9 years. There was a significant increase in median age at onset over time, from 60.9 years (59.2 – 67.8) in 1990 to 69.8 years (61.0 - 77.2) in 2006 (P= 0.008). The median illness duration was 4.3 months (2.7 – 7.9). This did not vary according to sex (P=0.126) or over time (P=0.370). Cases were most frequently referred by a neurologist (n=590, 66.1%), followed by a neuropathologist (n=133, 14.9%) (Figure 14).

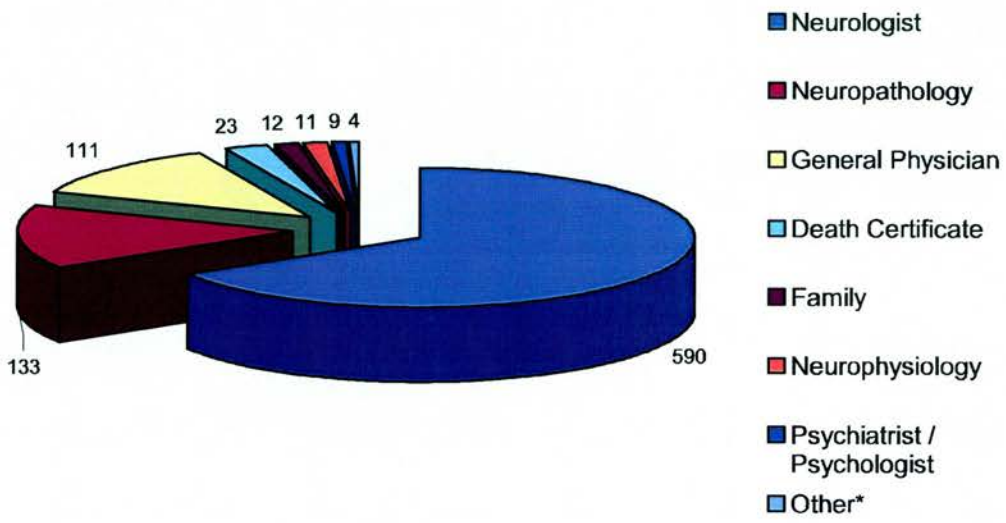

Figure 14 Source of referrals of sCJD cases ascertained by the NCJDSU, 1990 – 2006

Table 19 Characteristics of sCJD cases ascertained by the NCJDSU, 1990 – 2006

Year of referral	Cases n	Definite cases n (%)	Male n (%)	Median Age at Onset years (IQR)	Median Illness Duration months (IQR)	Post Mortem n (%)	Brain Biopsy n
1990	17	15 (88.2)	7 (41.2)	60.9 (59.2 – 67.8)	4.3 (2.1 – 5.7)	15 (88.2)	0
1991	34	30 (88.2)	13 (38.2)	63.2 (58.0 – 67.7)	4.1 (2.4 – 7.9)	29 (85.3)	1
1992	47	40 (85.1)	20 (42.6)	67.3 (62.4 – 75.1)	3.4 (2.4 – 6.0)	40 (85.1)	1
1993	39	33 (84.6)	19 (48.7)	66.2 (61.7 – 74.9)	3.9 (2.9 – 7.0)	31 (79.5)	4
1994	55	48 (87.3)	20 (36.4)	68.5 (61.6 – 74.9)	4.2 (2.6 – 6.3)	47 (85.5)	1
1995	33	29 (87.9)	14 (42.4)	63.4 (59.0 – 72.8)	5.2 (3.2 – 10.9)	29 (87.9)	0
1996	42	38 (90.5)	21 (50.0)	67.2 (59.1 – 74.2)	3.6 (2.6 – 6.7)	37 (90.2)	1
1997	63	59 (93.7)	29 (46.0)	65.5 (58.7 – 74.4)	5.1 (3.0 – 11.8)	54 (85.7)	5
1998	58	48 (82.8)	31 (53.5)	65.1 (58.4 – 72.9)	4.2 (2.5 – 7.5)	47 (81.0)	2
1999	62	46 (74.2)	35 (56.5)	65.1 (54.0 – 74.1)	4.3 (2.8 – 9.2)	45 (72.6)	3
2000	49	41 (83.7)	23 (46.9)	68.2 (63.5 – 74.5)	4.8 (3.2 – 7.5)	40 (81.6)	3
2001	59	46 (78.0)	29 (49.2)	69.1 (63.3 – 74.3)	5.2 (2.5 – 8.1)	46 (78.0)	1
2002	76	55 (72.4)	32 (42.1)	67.3 (62.6 – 71.7)	4.9 (3.0 – 10.4)	51 (67.1)	4
2003	75	43 (57.3)	40 (53.3)	67.6 (61.2 – 74.0)	4.4 (2.5 – 7.5)	41 (54.7)	4
2004	57	39 (68.4)	26 (45.6)	65.9 (59.5 – 74.2)	4.7 (2.8 – 7.5)	39 (68.4)	0
2005	62	39 (62.9)	37 (59.7)	71.5 (62.9 – 78.2)	4.3 (3.1 – 6.8)	39 (62.9)	1
2006	65	40 (61.5)	36 (55.4)	69.9 (61.0 – 77.1)	4.2 (2.8 – 6.9)	38 (60.3)	3
All	893	689 (77.2)	432 (48.4)	67.1 (60.6 – 74.2)	4.3 (2.7 – 7.9)	668 (75.1)	34

Incidence rates

Age and sex-specific incidence rates

Age-specific incidence rates of sCJD in men and women are shown in Figure 15. In both men and women the incidence of sCJD in those aged under 50 years old was low. Incidence rates rose thereafter to peak in men and women aged 70 – 79 years, before falling in those over 80 years of age. In men there was no statistically significant difference between incidence rates in those aged 60 – 69 years (rate 3.75 (95% CI 3.19 – 4.32) per million), 70 – 79 years (rate 4.15 (3.43 – 4.86) per million) and 80 years and over (rate 2.74 (1.83 – 3.64) per million). In women incidence rates in those aged 60 – 69 years (rate 3.26 (2.76 – 3.77) per million) were comparable to incidence rates in those aged 70 – 79 years (rate 3.73 (3.14 – 4.32) per million). However the decline in incidence rates in those aged 80 years and over was significant (rate 1.57 (1.10 – 2.03) per million.) The 95% confidence intervals for age-specific rates in men and women overlapped in each age group indicating that overall there was no significant difference in age-specific rates according to sex.

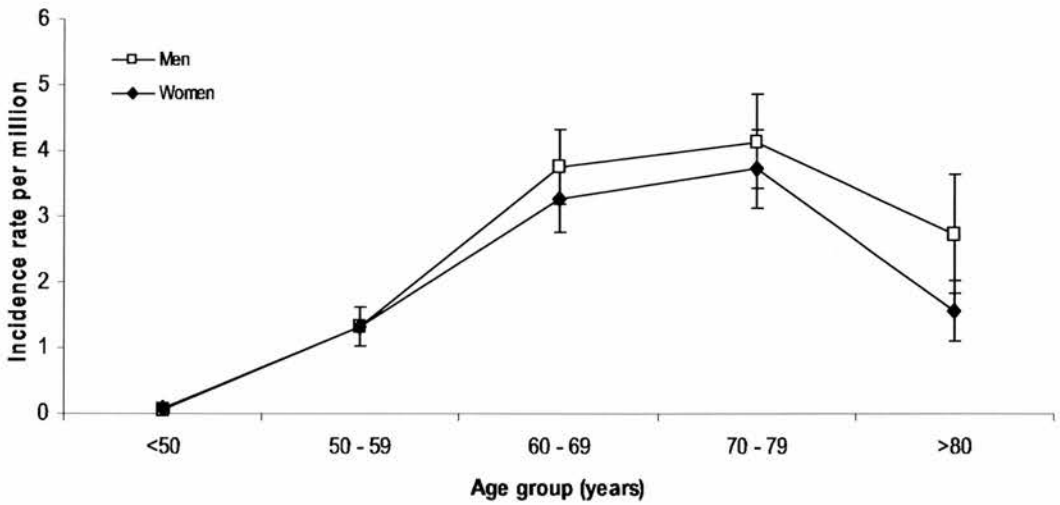

Figure 15 Age-specific sCJD incidence rates according to sex, 1990 – 2006

Temporal trends in age and sex-specific incidence rates

Over time the only statistically significant increase in the sCJD incidence rate was observed in men aged 70 – 79 years old, in whom the annual percentage change (APC) in incidence rate was 8.87% (4.54 – 13.38). In this group the incidence rate

increased from 0.60 (0.00 – 1.78) per million in 1990 to 7.75 (3.83 – 11.67) per million in 2006. In women of the same age a non-significant increase in the incidence rate from 0.83 (0.00 – 1.99) per million in 1990 to 4.23 (1.61 – 6.86) per million in 2006 was observed. Similar non-statistically significant increases in sCJD incidence were observed in men aged 60 – 69 years (from 1.11 (0.00 – 2.36) per million to 4.52 (2.06 – 6.97) per million), men 80 years and over (from 0.00 per million to 5.30 (0.65 – 9.95) per million), women aged 60 – 69 years (from 1.64 (0.20 – 3.07) per million to 2.95 (1.02 – 4.87) per million) and women aged 80 years and over (from 0.00 per million to 2.85 (0.35 – 5.34) per million). There was no significant increase, clinically or statistically, in the incidence of sCJD in men or women under 60 years of age.

Age standardised incidence rates

Age standardised incidence rates of sCJD in men and women are shown in Figure 16. In men, the age standardised rate rose from 0.27 (0.07 – 0.47) per million in 1990 to peak at 1.35 (0.93 – 1.77) per million in 2003 and has remained stable since. At the end of the study period, 2006, the incidence rate per million was 1.18 (0.79 – 1.56) per million. In women the age standardised rate in 1990 was 0.33 (0.13 – 0.54) per million. The rate peaked at 1.45 (1.02 – 1.88) per million in 2002. At the end of the study period, 2006, the rate was 0.94 (0.60 – 1.28) per million.

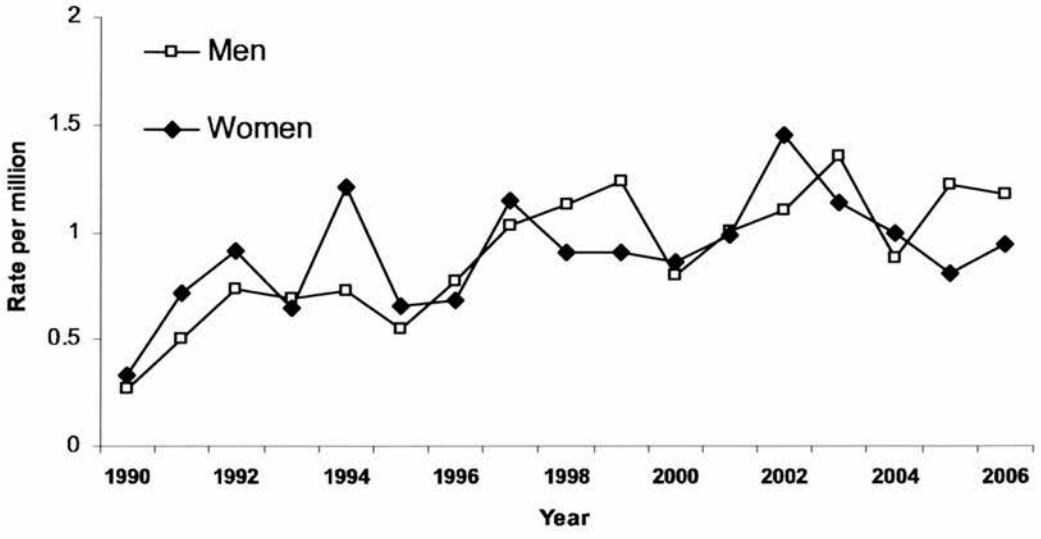

Figure 16 Age standardised incidence rate of sCJD in men and women, 1990 – 2006

Temporal trends in age standardised incidence rates

There was a statistically significant increase in the age standardised incidence of sCJD in men and a non-significant increase in women from 1990 through 2006. In men, the APC in incidence was 5.20% (2.62 – 7.83). In women, this was estimated to be 2.57% (-0.38 – 5.60).

Clinical presentation

The majority of sCJD cases, 61.6% (550), presented with a RPD, 11.5% (103) with a cerebellar onset and 5.4% (48) with the Heidenhain variant (Figure 17). Almost a fifth, 173, sCJD cases had an 'atypical clinical presentation', defined as a presentation other than RPD, cerebellar onset or Heidenhain variant. There was no significant difference in presentation according to sex (P= 0.653). Clinical presentation was not specified for 19 (2.1%) sCJD cases. Over time there was no significant change in the proportion of sCJD cases that had an atypical presentation at symptom onset (P=0.185).

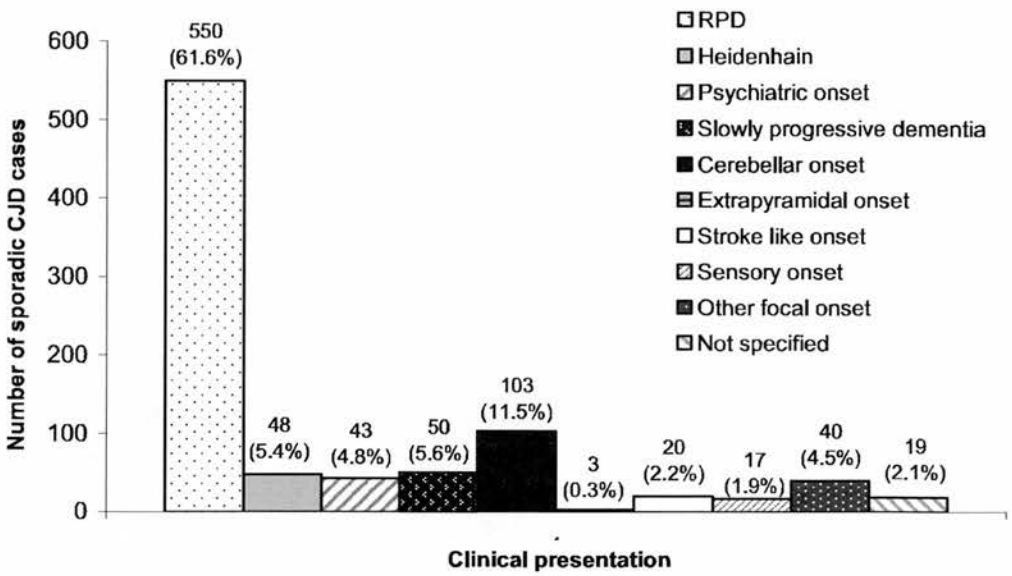

Figure 17 Distribution of clinical presentations in sCJD cases, 1990 – 2006

Investigations to support a diagnosis of sCJD

In this section the use of EEG, MRI, CSF 14-3-3 protein, brain biopsy, *PRNP* Codon 129 genotyping, full sequencing for *PRNP* mutations and post mortem examination in the investigation of sCJD cases will be described.

EEG

Overall 808 (90.5%) sCJD cases underwent at least one EEG examination during the course of their clinical illness (Figure 18). There was no significant change in the proportion of sCJD cases undergoing EEG examination over time (P= 0.779). Overall the median number of EEGs undertaken was 1 (range 1 – 5). A non-significant fall in the median number of EEGs from 2 (1 – 4) in 1990 to 1 (1 – 4) in 2006 was observed (P=0.088). Over a third, 302 (37.4%), of the sCJD cases that underwent EEG examination had a typical EEG. There was significant year to year variation in the proportion of patients with a typical EEG over the study period (Figure 19). This ranged from a high of 59.4% (29) in 1994 to 23.0% (12) in 2001. The overall trend however was toward a fall in the proportion of sCJD cases with a typical EEG over time from 50.0% (8) in 1990 to 33.3% (19) in 2006 (P<0.001). Overall, the median time from symptoms onset to typical EEG was 2.3 (1.3 – 3.5) months (n=301). Over the study period the median time from symptom onset to

typical EEG fell from 2.2 (1.1 – 3.4) months in 1990 to 2.0 (1.4 – 2.9) months in 2006 (P= 0.044).

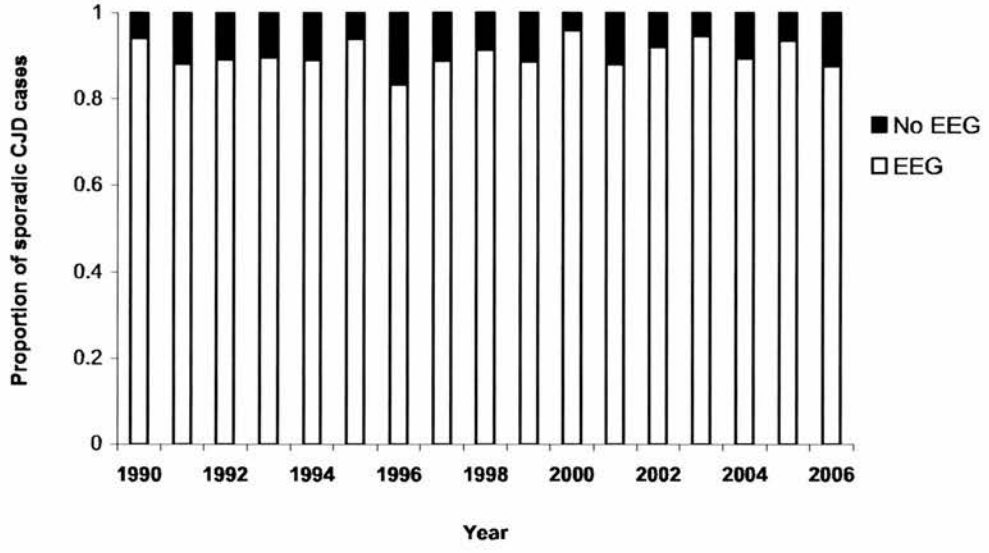

Figure 18 Proportion of sCJD cases undergoing at least one EEG examination, 1990 – 2006

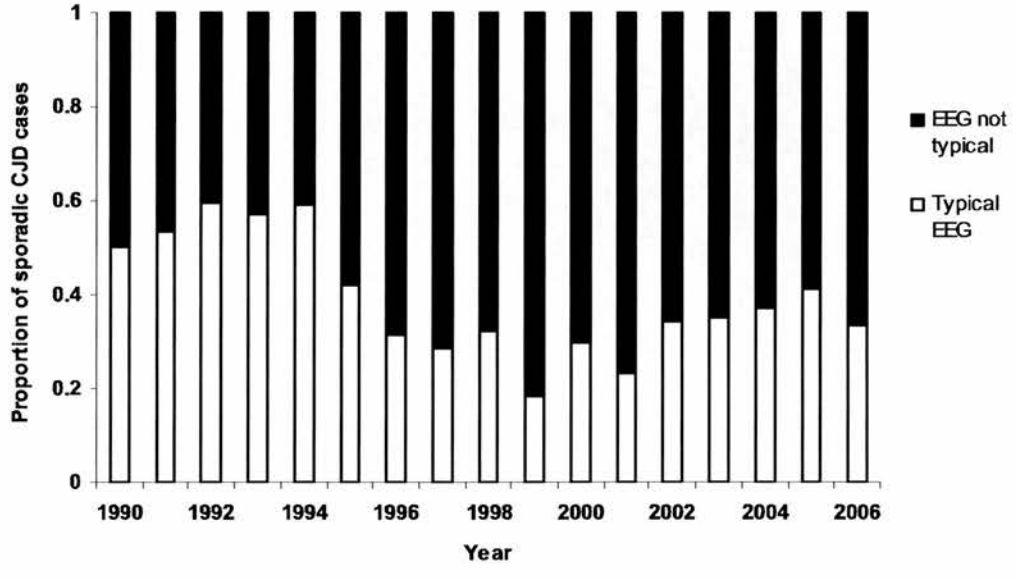

Figure 19 Proportion of sCJD cases with a typical EEG among sCJD cases that underwent at least one EEG examination, 1990 – 2006

MRI

Overall 596 (66.7%) sCJD cases underwent at least one MRI examination (any sequence) during the clinical course of their illness (Figure 20). The proportion of sCJD cases undergoing at least one MRI examination increased significantly over time, from 11.8% (2) in 1990 to 84.6% (55) in 2006 (P<0.001). Overall the median number of MRI undertaken in the investigation of sCJD cases was 1 (range 1 – 3); this increased over time from 1 (range 1 – 1) in 1990 to 2 (range 1 – 3) in 2006 (P<0.001). Radiological changes consistent with sCJD were seen on MRI scans in 201 (33.8%) sCJD cases that underwent MRI examination (Figure 21). The proportion of sCJD cases with a positive MRI scan increased significantly over time from 0% (0) in 1990 to 45.5% (25) in 2006 (P<0.001). Overall, the median time from symptoms onset to characteristic MRI scan was 3.8 (2.2 – 6.5) months (n=201). This was invariant over time (P= 0.568).

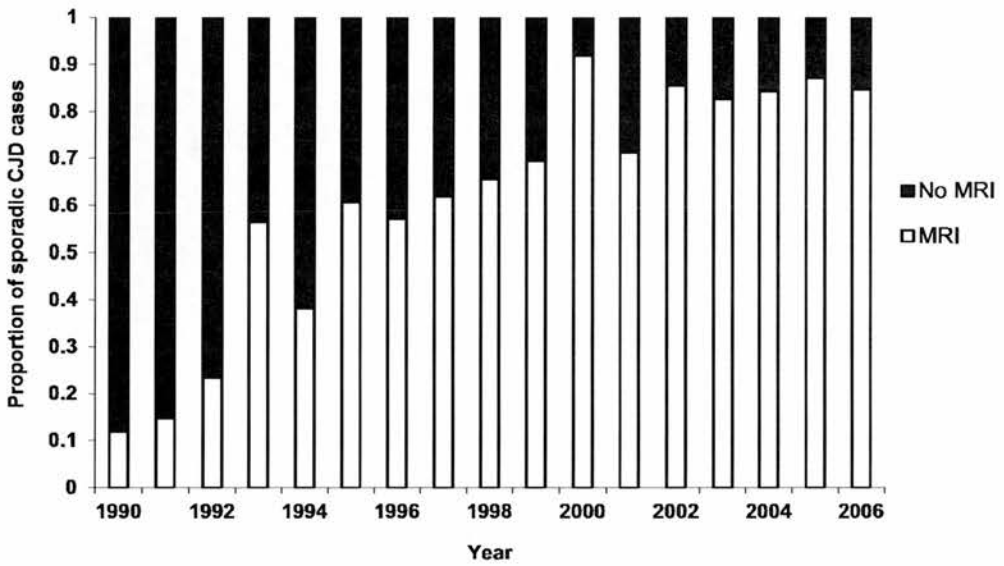

Figure 20 Proportion of sCJD cases undergoing at least one MRI examination, 1990 – 2006

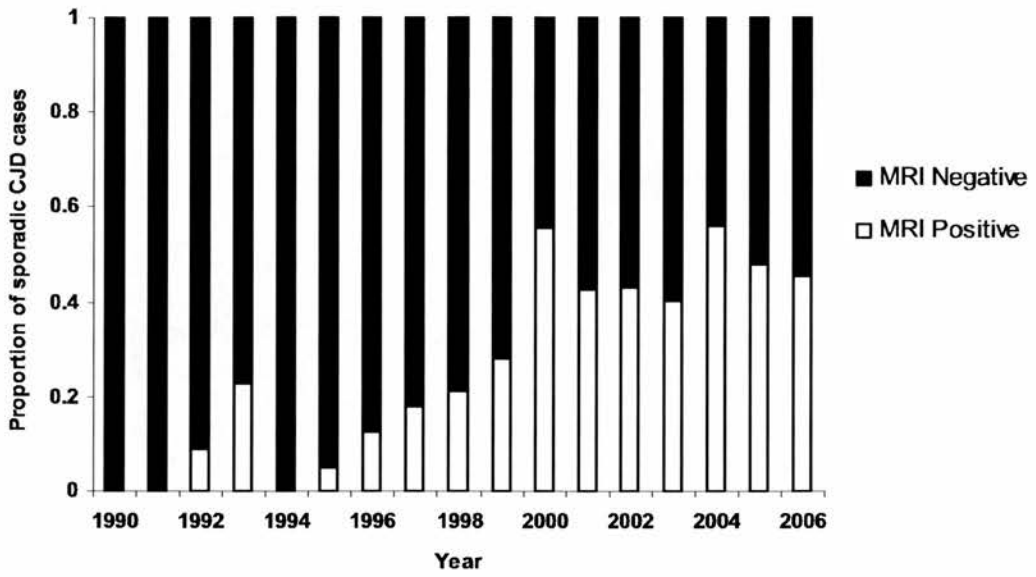

Figure 21 Proportion of sCJD cases with an MRI scan that had changes consistent with sCJD among sCJD cases that underwent at least one MRI examination, 1990 - 2006

CSF 14-3-3 protein (limited to 1996 onwards)

Almost two thirds of all sCJD patients, 431 (64.8%), underwent at least one CSF 14-3-3 protein examination following the introduction of the investigation in the UK in 1996 (Figure 22). The proportion of sCJD cases undergoing CSF 14-3-3 protein examination increased from 35.7% (15) in 1996 to 73.8% (48) in 2006 (P<0.001). Overall the median number of CSF 14-3-3 protein examinations was 1 (range 1 – 3). This was invariant over time (P=0.564). CSF 14-3-3 protein was positive in 83.7% (361) of all sCJD cases undergoing this investigation. There was year to year variation in the proportion of sCJD cases with a positive CSF 14-3-3 protein examination, from a high of 93.3% (14) in 1996 to a low of 68.4% (26) in 2000, but no significant trend over time (P=0.237) (Figure 23). Overall, the median time from symptoms onset to positive CSF 14-3-3 protein examination was 2.9 (1.9 – 5.3) months (n=351). This was invariant over time (P= 0.694).

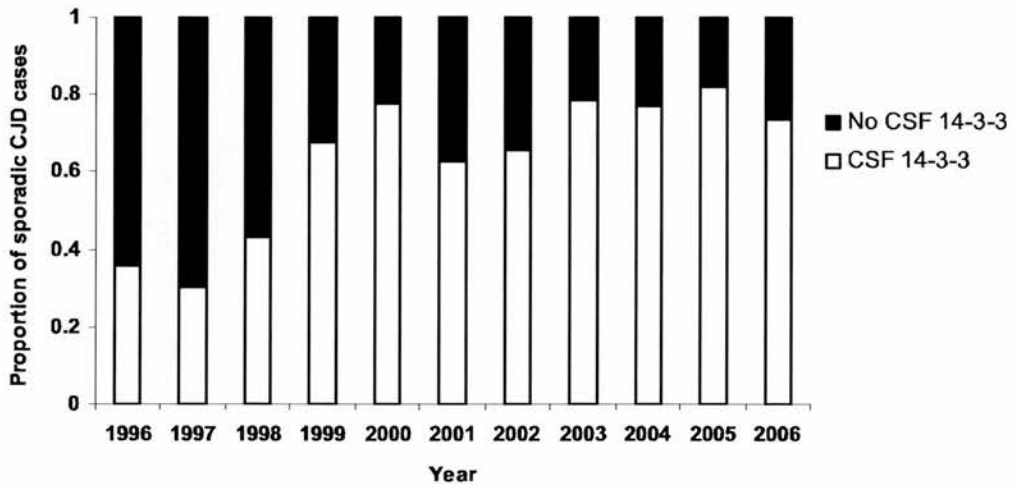

Figure 22 Proportion of sCJD cases undergoing at least one CSF 14-3-3 protein investigation, 1996 – 2006

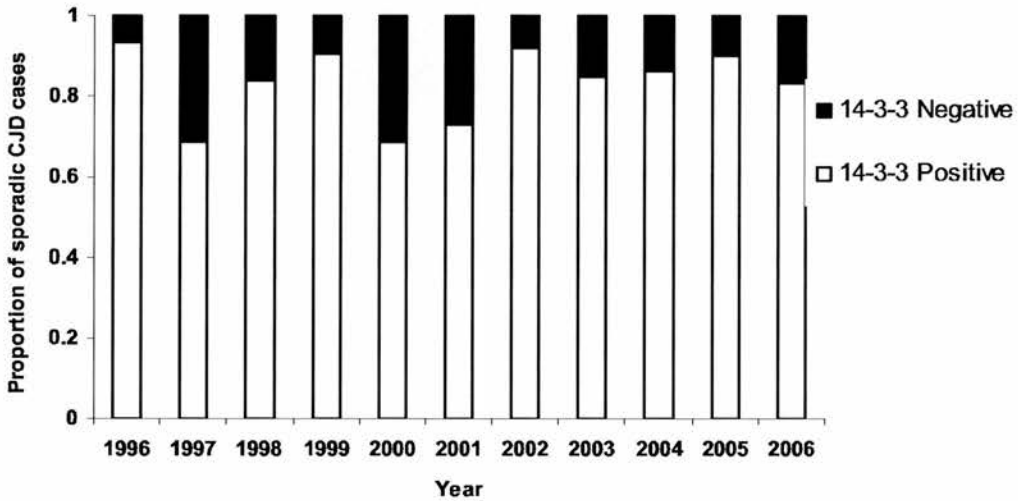

Figure 23 Proportion of sCJD cases with a positive CSF 14-3-3 protein examination among those undergoing CSF 14-3-3 protein examination, 1996 – 2006

Over time the relative importance of EEG and CSF 14-3-3 protein examination in case classification has changed (Figure 24). The proportion of probable sCJD in whom the diagnosis was based solely on EEG examination fell over the study period following the introduction of CSF 14-3-3 protein. Of note, whilst CSF 14-3-3 protein

was not formally introduced into the WHO diagnostic criteria until 2000, this test was widely available in the UK from 1996. Possible sCJD cases ascertained by the NCJDSU from 1996 through 2000 that had a positive CSF 14-3-3 protein were retrospectively classified as probable sCJD cases for disease surveillance purposes following the change in diagnostic criteria.

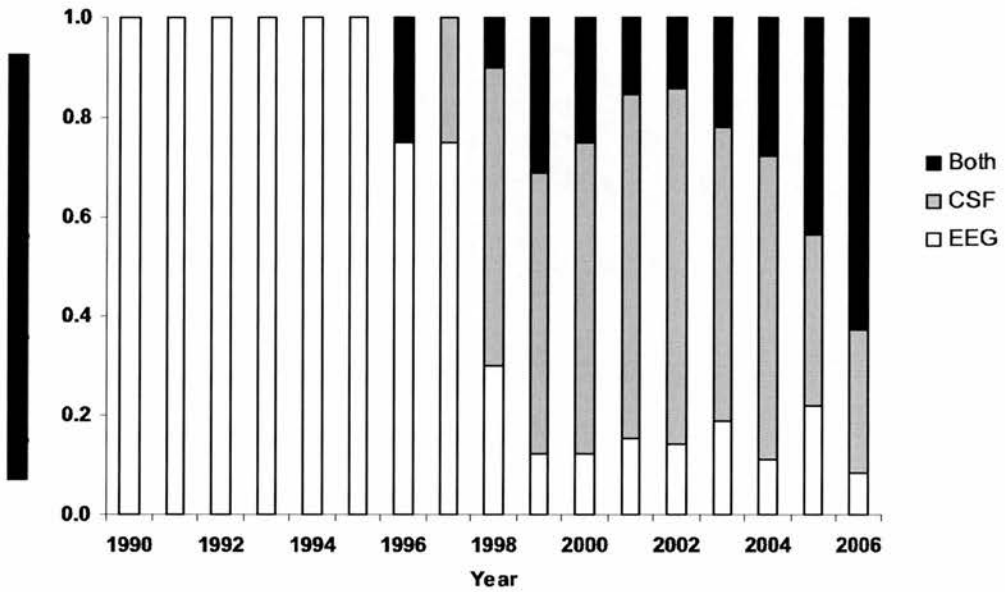

Figure 24 Proportion of probable sCJD cases meeting the WHO diagnostic criteria based on EEG, CSF 14-3-3 protein or both, 1996 – 2006

PRNP Codon 129 Genotyping

Genetic analysis for polymorphism at Codon 129 of *PRNP* was available for almost two thirds, 580 (65.0%), of sCJD cases (Figure 25). There was no significant difference in the proportion of sCJD cases undergoing *PRNP* Codon 129 genotyping over the study period (P=0.111). The distribution of *PRNP* Codon 129 genotypes among sCJD case ascertained over the study period is shown in Figure 26. The majority of sCJD cases, 375 (64.7%), were methionine homozygous (MM), 103 (17.8%) valine homozygous (VV) and 102 (17.6%) heterozygous (MV). There was a non-significant reduction in the proportion of sCJD cases with the MM genotype, with an increase in the proportion of sCJD cases with MV and VV genotypes, over time (P=0.138).

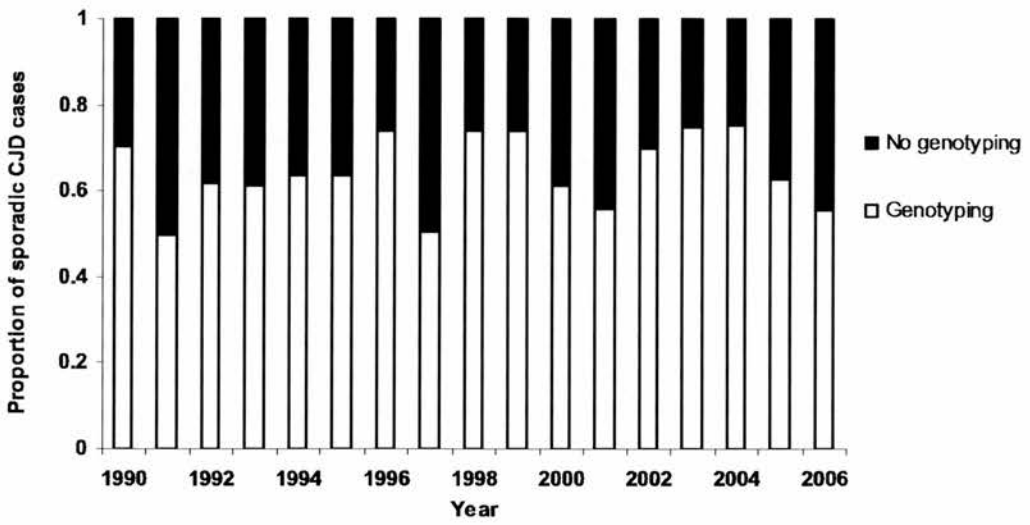

Figure 25 Proportion of sCJD case for which *PRNP* Codon 129 genotyping was available, 1990 – 2006

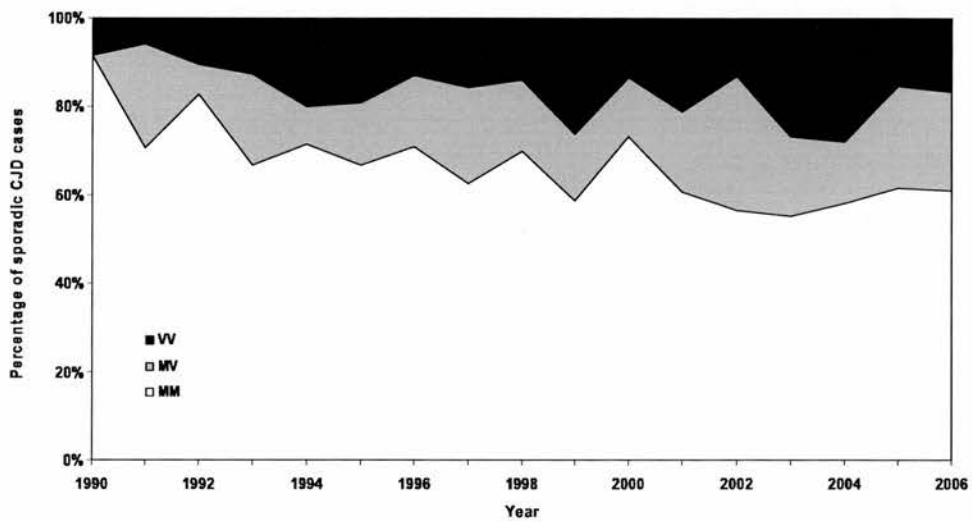

Figure 26 Distribution of *PRNP* Codon 129 genotypes, 1990 – 2006

Full sequencing for mutations of PRNP

Genetic prion disease may be clinically and neuropathologically indistinguishable from sCJD and only an estimated 50% of genetic prion disease cases report a family history of prion disease. Testing for mutations of *PRNP* is important in ensuring that genetic prion disease and sCJD cases are correctly classified. Half of all sCJD cases,

451 (50.5%) underwent genetic testing to exclude a mutation of *PRNP*. The proportion of sCJD cases undergoing genetic testing to exclude a mutation of *PRNP* decreased over time from 70.6% (12) in 1990 to 24.6% (16) in 2006 (P<0.001), as shown in Figure 27.

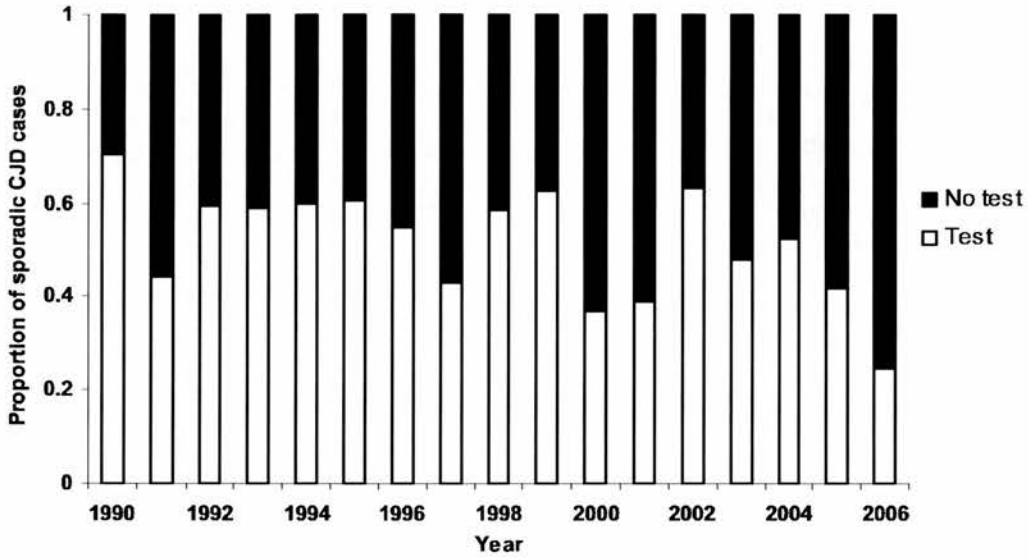

Figure 27 Proportion of sCJD cases undergoing genetic testing for a mutation of *PRNP*, 1990 – 2006

Post mortem and brain biopsy

As of 31st December 2008, three neuropathologically confirmed sCJD cases referred to the NCJDSU between 1st May 1990 and 31st December 2006 were still alive. Overall 75.1% (668) of deceased sCJD cases underwent post mortem examination. Over time there was a statistically significant reduction in the percentage of sCJD cases undergoing post mortem examination, from 88.2% (15) in 1990 to 60.3% (38) in 2006 (P<0.001) (Figure 28). Overall just 3.8% (34) of all sCJD cases underwent brain biopsy in life. This was diagnostic in almost three quarters (n=25); the remaining 9 sCJD cases in whom brain biopsy was non-diagnostic underwent post mortem examination following death.

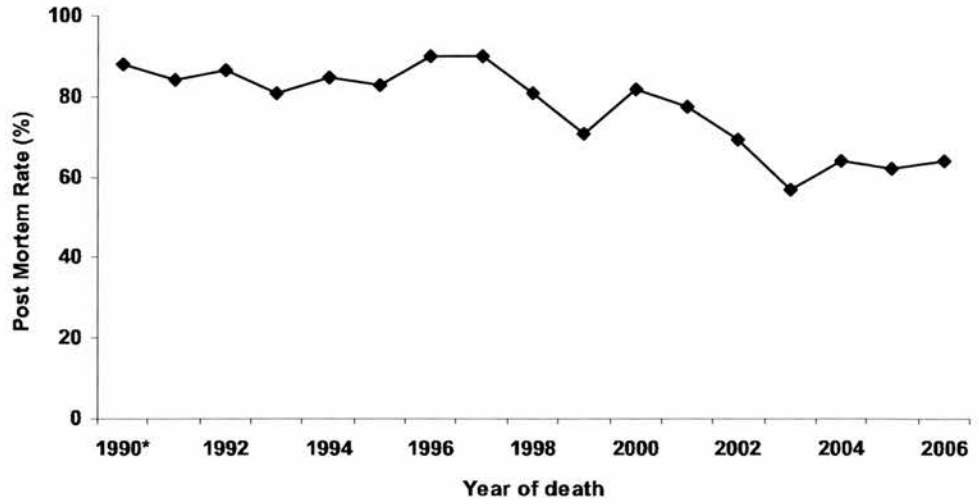

Figure 28 Rate of post mortem examination in sCJD cases according to year of death, 1990 – 2006

PrP^{Sc} protein typing

PrP^{Sc} protein typing was carried out on 301 (43.7%) sCJD cases for which neuropathological material was available following brain biopsy or post mortem examination (Figure 29). Over time the proportion of sCJD cases for whom PrP^{Sc} protein typing was available increased significantly (P<0.001).

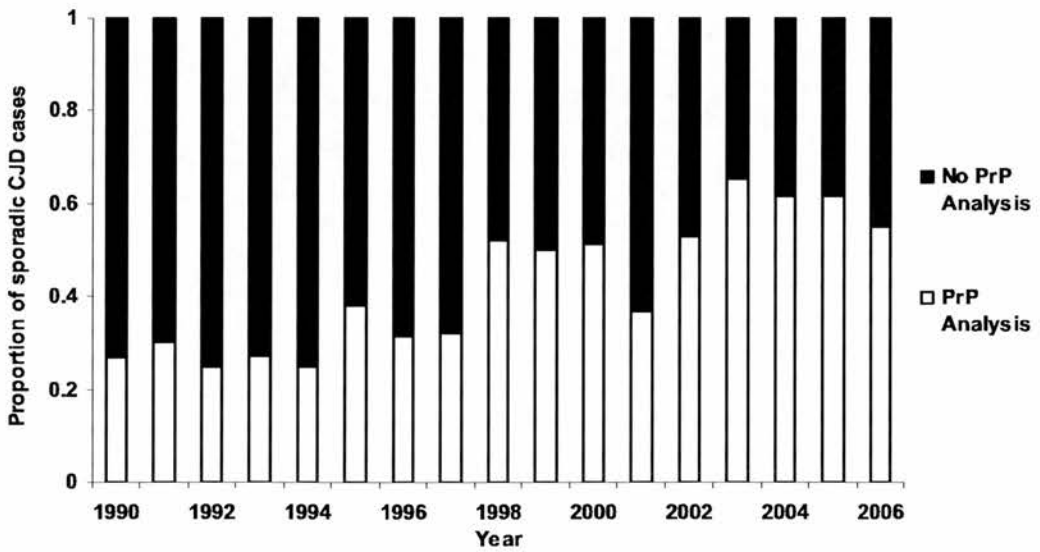

Figure 29 Proportion of sCJD cases in whom PrP^{Sc} protein typing was available among sCJD cases for which PrP^{Sc} protein typing was carried out, 1990 – 2006

The distribution of PrP^{Sc} protein type among sCJD cases for whom PrP^{Sc} protein type analysis was available is shown in Figure 30. In total 189 (62.8%) sCJD cases were PrP^{Sc} Type 1, 62 (20.6%) PrP^{Sc} Type 2 and 50 (16.6%) PrP^{Sc} mixed Type 1/Type 2.

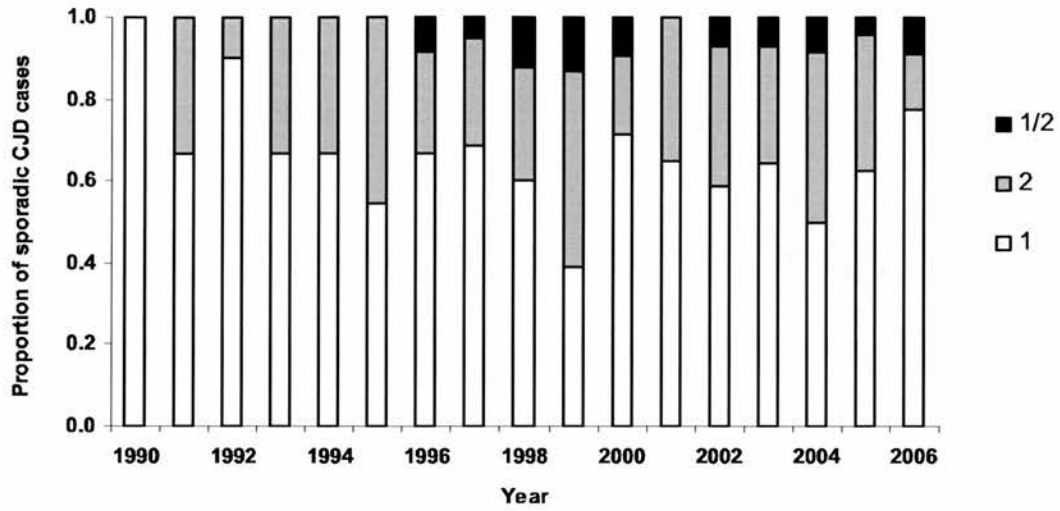

Figure 30 Distribution of PrP^{Sc} protein type in sCJD cases for whom prion protein typing was available, 1990 - 2006

Molecular subtyping

Molecular subtyping was available for 299 sCJD cases in whom PrP^{Sc} protein type and *PRNP* Codon 129 genotyping was known (Figure 31). The most common molecular subtype was MM1, followed by MV2 and VV2. The characteristics of sCJD cases according to molecular subtype are outlined in Table 20. The median illness duration was shortest in sCJD cases with the MM1 and VV1/2 subtype and longest in those with the MM2 and MV1/2 subtype. EEG was most frequently typical in the MM1 subtype and least frequently in the VV subtypes. CSF 14-3-3 protein was most commonly positive in the MM1 subtype, least frequently in the MV2 and MM2 subtypes. MRI scanning was most commonly positive in the VV subtypes. Over time there was no significant change in the molecular subtype of sCJD cases ascertained by the NCJDSU (P=0.991).

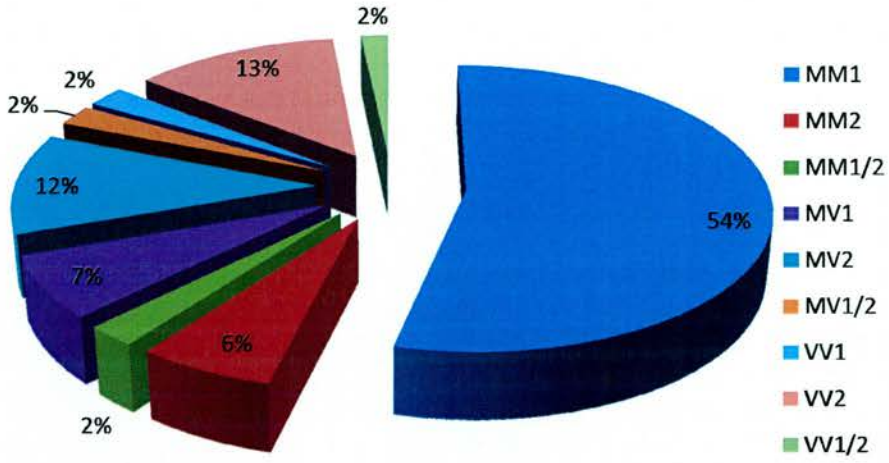

Figure 31 Molecular subtyping of sCJD cases for whom *PRNP* Codon 129 genotyping and PrP^{Sc} protein typing was available, 1990 - 2006

Table 20 Characteristics of sCJD cases according to molecular subtyping, 1990 – 2006 (n=299)

	MM1	MM2	MM 1/2	MV1	MV2	MV 1/2	VV1	VV2	VV 1/2
Number (% of total)	161 (53.8)	19 (6.4)	7 (2.3)	21 (7.0)	34 (11.4)	7 (2.3)	7 (2.3)	38 (12.7)	5 (1.8)
Male, n (%)	76 (47.2)	10 (52.6)	5 (71.4)	12 (57.1)	16 (47.1)	4 (57.2)	5 (71.4)	21 (55.3)	3 (60.0)
Median Age,	68.2	53.2	63.6	74.0	65.9	63.1	55.6	67.9	65.0
Years (IQR)	(61.3 - 75.9)	(48.9 - 60.6)	(61.5 - 72.3)	(61.1 - 77.9)	(60.6 - 72.6)	(61.5 - 65.8)	(41.9 - 68.0)	(57.5 - 72.0)	(63.2 - 74.0)
Median Illness									
Duration,	3.1	15.8	9.0	4.7	11.5	16.8	8.6	6.2	3.9
months (IQR)	(2.2 - 5.3)	(8.7 – 23.9)	(5.7 - 12.4)	(2.5 - 8.3)	(8.4 - 17.2)	(6.8 - 30.0)	(4.6 - 29.4)	(4.3 - 8.6)	(3.5 - 5.2)
Typical EEG*	81 / 145	3 / 17	1 / 6	7 / 17	1 / 32	1 / 7	0 / 7	0 / 33	0 / 5
Positive CSF 14-3-3									
protein*	70 / 76	3 / 10	2 / 2	9 / 10	12 / 16	1 / 5	1 / 2	17 / 18	4 / 4
Positive MRI*	32 / 108	5 / 17	1 / 4	3 / 15	9 / 24	2 / 6	2 / 5	14 / 30	2 / 5

*Numerator is the number of sCJD cases undergoing at least one EEG, at least one CSF 14-3-3 protein examination or at least one MRI scan that had a positive result and denominator the number of sCJD cases undergoing at least one of the aforementioned investigations. Data have been presented in this way to aid interpretation as the absolute number of sCJD cases undergoing some of these investigations is extremely small

Sensitivity of diagnostic investigations in sCJD (limited to 1996 onward)

The sensitivity of EEG, MRI, CSF 14-3-3 protein and brain biopsy examinations in sCJD cases that underwent these investigations is shown in Figure 32. Whilst MRI was not at the time of writing included in the WHO diagnostic criteria for sCJD it has been included here in recognition of its value in sCJD. CSF 14-3-3 protein examination was the most sensitive of investigations over this period, being positive in 83.9% (95% CI 80.0 - 87.2) of sCJD cases. The sensitivity of brain biopsy was 70.6% (55.3 – 85.9), MRI scanning 39.6% (35.4 – 44.0) and EEG examination, 31.6% (27.9 - 35.4).

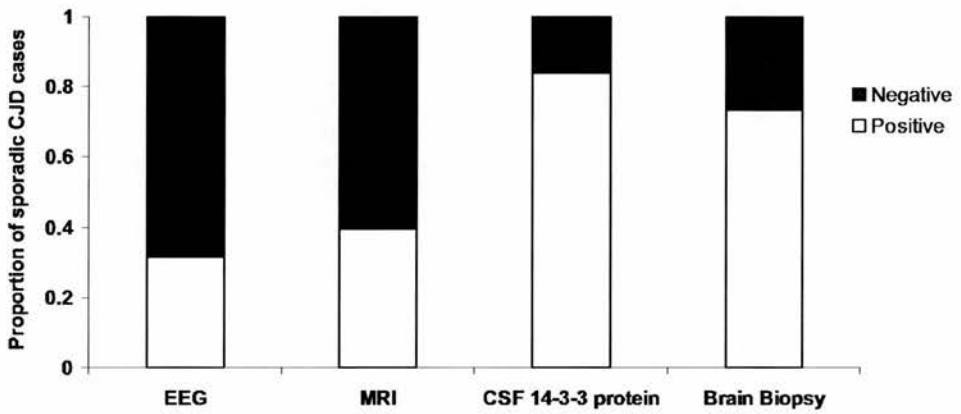

Figure 32 Overall sensitivity of EEG, MRI, CSF 14-3-3 protein and brain biopsy examinations in sCJD cases, 1996 – 2006

sCJD cases with negative EEG, MRI and CSF 14-3-3 protein examinations

From 1996 through 2006, 24 pathologically confirmed sCJD cases underwent at least one EEG, at least one MRI and at least one CSF 14-3-3 protein examination, and *all* three diagnostic investigations (EEG, MRI *and* CSF 14-3-3 protein) were negative. This represents 4.9% of all pathologically confirmed sCJD and 3.6% of all sCJD cases (definite or probable) ascertained over this ten year period. Men accounted for 13 (54.7%) of these sCJD cases. Compared to sCJD cases in whom one or more of the aforementioned investigations was positive, sCJD cases in this group were younger (median age 59.9 (52.1 – 67.4) years *vs.* 67.4 (60.7 – 74.0) years, P< 0.001), more likely to have an atypical clinical presentation (P<0.001), had a longer median illness duration (16.6 (8.2 – 21.1) months *vs.* 4.2 (2.7 – 7.3) months, P< 0.001), were

more likely to undergo brain biopsy during life (P=0.011) and post mortem examination following death (P=0.004). However, there was no statistically significant difference in *PRNP* Codon 129 genotype between the groups (P= 0.084). Of these 24 sCJD, four (16.7%) underwent brain biopsy in life; this was diagnostic in two (50%) cases.

Atypical sCJD Cases

'Atypical sCJD cases' were defined as sCJD cases aged under 50 years old at symptom onset, that had an illness duration of 1 year of more and/or had a clinical presentation other than RPD, Heidenhain variant or cerebellar onset; all other sCJD cases were considered 'typical'. Over a quarter, 28.8% (256) sCJD cases were considered atypical, of which 114 (45.5%) were male. Atypical sCJD cases included 173 sCJD cases with an atypical clinical presentation, 45 sCJD cases under 50 years of age at symptom onset and 140 sCJD cases with an illness duration of over 1 year (groups not mutually exclusive) (Figure 33). Figure 34 shows the proportion all sCJD accounted for by atypical sCJD cases per year. There was no significant change in the proportion of all sCJD cases accounted for by atypical sCJD cases over time (P=0.118). The diagnosis of sCJD was pathologically confirmed in 223 (87.1%) atypical sCJD cases. The sensitivity of EEG (25.9% *vs.* 41.8%, P<0.001) and CSF 14-3-3 protein (68.9% *vs.* 89.4%, P<0.001) examination but not MRI scanning (31.0% *vs.* 34.9%, P=0.324) was lower in atypical sCJD cases compared to typical sCJD cases. Atypical sCJD cases were more likely than typical sCJD cases to undergo brain biopsy during life (9.0% *vs.* 1.7%, P<0.001) and post mortem examination following death (80.5% *vs.* 72.5%, P<0.001). Relative to typical sCJD cases there was an excess of *PRNP* Codon 129 heterozygotes (MV) among atypical sCJD cases (P<0.001). Atypical sCJD cases were most frequently of the MM1 (34) molecular subtype, followed by MV1 (26), MV2 (19) and MM2 (16) molecular subtypes. A greater than expected number of atypical sCJD cases had MM2, MV1, MV2 and MV 1/2 mixed protein molecular subtypes (P<0.001).

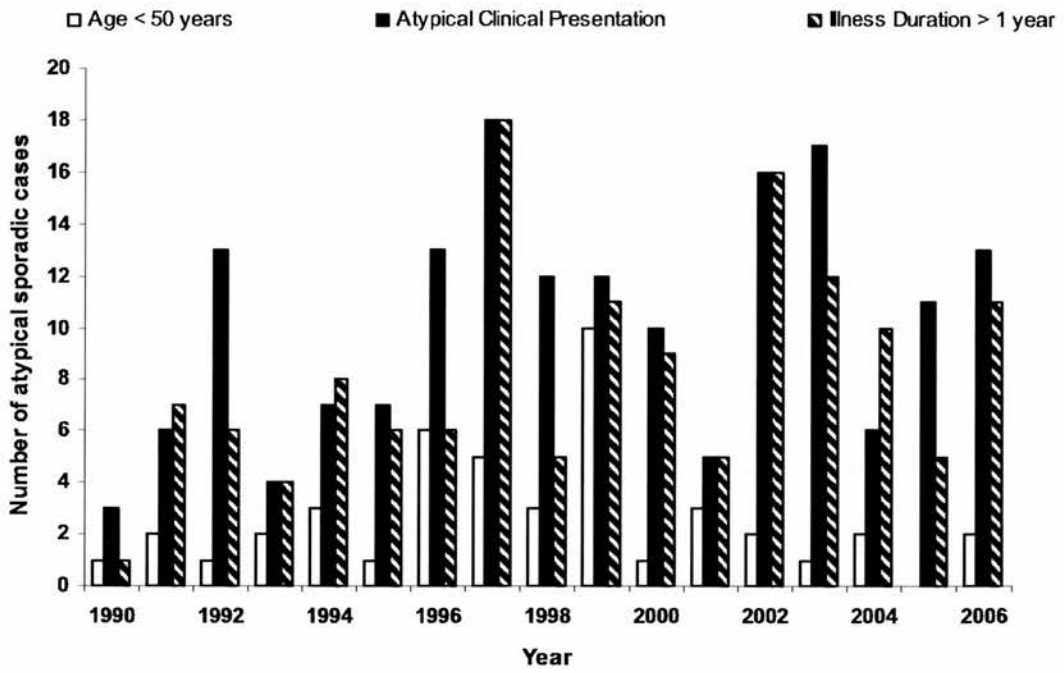

Figure 33 Number of 'atypical sCJD cases' according to year, 1990 - 2006

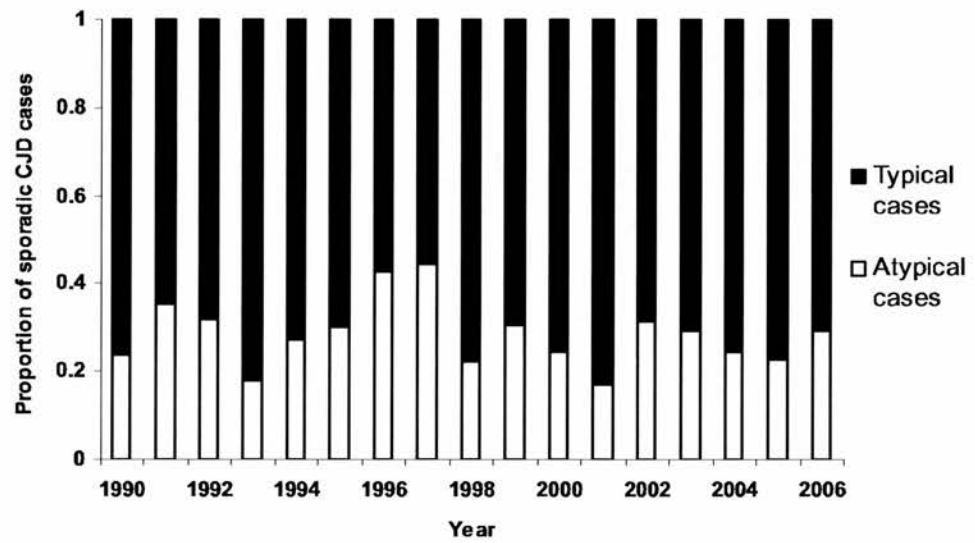

Figure 34 Proportion of all sCJD cases accounted for by 'atypical sCJD cases', 1990 – 2006

sCJD cases aged under 50 years old at onset

In total 45 sCJD cases were aged under 50 years old at symptom onset, of whom 20 (44.4%) were male. Compared to sCJD cases aged 50 years and over at onset, sCJD

cases aged under 50 years of age at onset were more likely to have an illness duration of over 1 year (40.0% *vs.* 14.4%, P<0.001) and more likely to have an atypical clinical presentation at symptom onset (47.7% *vs.* 18.3%, P<0.001). The sensitivity of EEG (18.2% *vs.* 38.5%, P=0.007) and CSF 14-3-3 protein (69.6% *vs.* 84.6%, P=0.008) examination but not MRI scanning (35.9% *vs.* 33.6%, P=0.797) was lower in sCJD cases aged under 50 years old at onset compared to sCJD cases aged 50 years and over at onset. sCJD cases aged under 50 years old at onset were more likely to undergo brain biopsy during life (13.3% *vs.* 3.3%, P<0.001) but not post mortem after death (P=0.637) than sCJD cases aged 50 years and over at onset. *PRNP* Codon 129 genotype distribution did not vary according to age at onset (P=0.267). However a greater than expected number of sCJD cases aged under 50 years old at onset had the MM2 (5 / 26), MV1 (7 / 26) or VV1 (7 / 26) molecular subtype (P<0.001). A neuropathologically confirmed diagnosis was available in the majority, 86.7% (39) of sCJD cases aged less than 50 years old at onset. The clinical details of the six sCJD cases aged less than 50 years old at onset that did not have a neuropathological diagnosis are shown in Table 21. There is little evidence from the clinical presentation or summary of diagnostic investigations to suggest that any of these sCJD cases were in fact misclassified vCJD cases.

Table 21 Clinical details of sCJD cases under 50 years of age at onset that did not have a neuropathological diagnosis

Case	Clinical Presentation	EEG	MRI	CSF 14-3-3 protein	Illness Duration (months)
1	RPD	Typical	N/A	N/A	4.3
2	RPD	Typical	Negative	N/A	2.7
3	Cerebellar	Typical	Negative	N/A	2.4
4	Stroke-like	Not typical	Positive	Positive	5.5
5	Other focal	Not typical	Positive	Positive	8.4
6	RPD	Not typical	Positive	Positive	4.7

N/A not carried out

Case-fatality

Crude case-fatality

Crude case-fatality at 6 months and 1 year was 62.7% (560) and 84.3% (753) respectively. Median survival overall was 4.3 (IQR 2.7 - 7.9) months; this was invariant over time (P= 0.370). Median survival did not vary according to sex (P= 0.126). Median survival fell with increasing age, from 10.5 (4.6 - 20.9) months in sCJD cases under 50 years of age to 3.0 (1.9 – 4.6) months to sCJD cases 80 years and over (P<0.001). Figure 35 shows the survival time from symptom onset, by age group. Median survival varied by clinical presentation (P<0.001). sCJD cases with an atypical clinical presentation had a median illness duration of 9.2 (4.2 – 20.9) months, compared to sCJD cases with a typical clinical presentation, 3.8 (2.6 – 6.6) months (Figure 36). The shortest median survival was seen in sCJD cases with a stroke-like onset, 2.4 (1.9 – 3.9) months; the longest for sCJD cases with a slowly progressive dementia, 23.2 (16.3 – 30.0) months (Table 22). According to *PRNP* Codon 129 genotyping the median survival was 3.3 (2.4 – 5.7) months for methionine homozygotes, 9.3 (6.2 – 16.2) months for methionine heterozygotes and 6.2 (4.3 – 8.7) months for valine homozygotes (P<0.001). As previously noted illness duration varied according to molecular subtype.

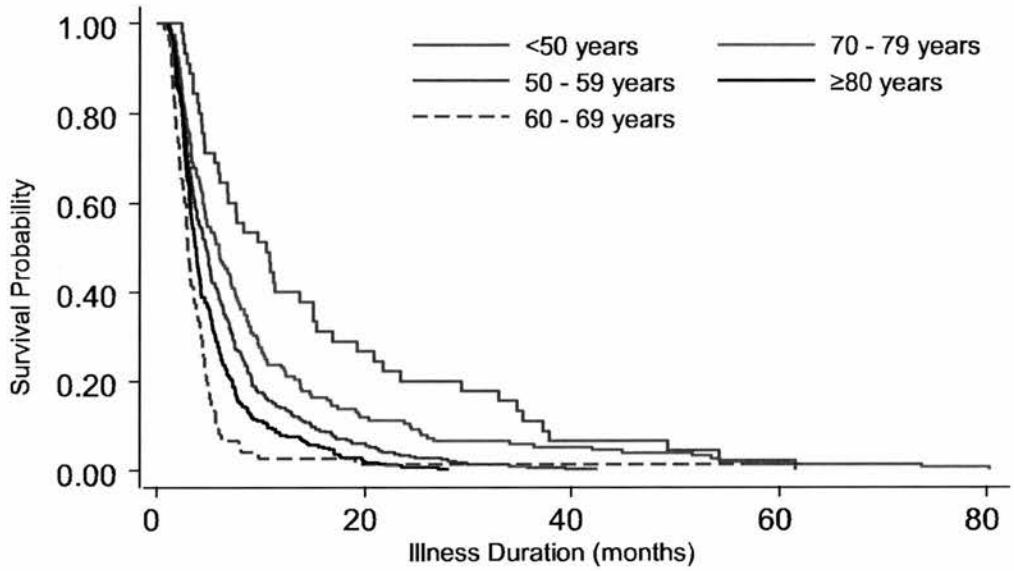

Figure 35 Kaplan Meier estimates of survival (months) in sCJD cases, 1990 – 2006, according to age group

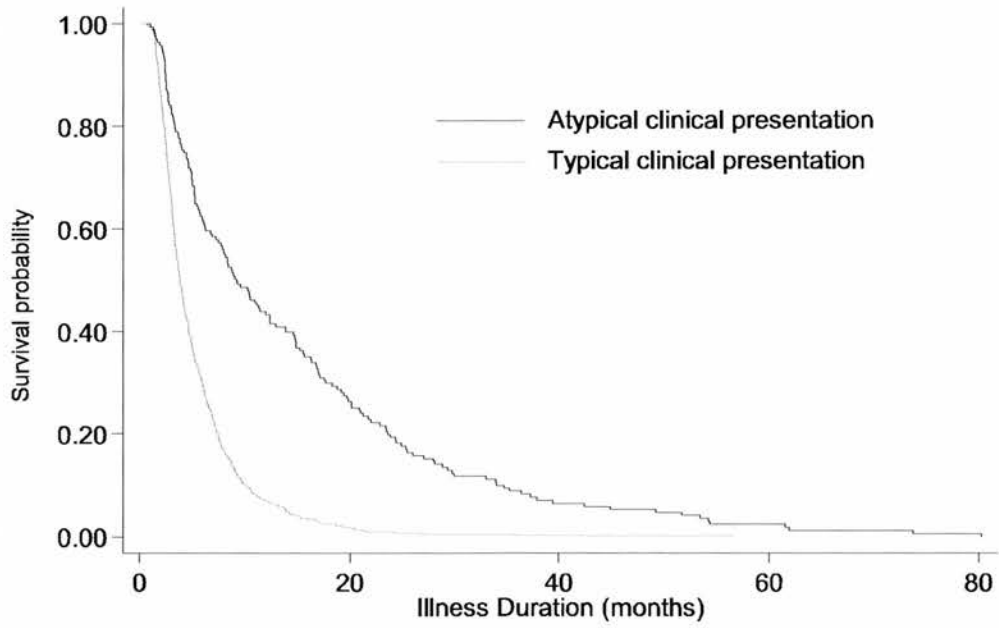

Figure 36 Kaplan-Meier estimates of survival (months) in sCJD cases, 1990 – 2006, according to clinical presentation (typical or atypical)

Table 22 Median Survival (months) in sCJD cases in the UK, 1990 – 2006, according to clinical presentation

Clinical Presentation	Number of sCJD cases	Median Survival (months)
Rapidly Progressive Dementia	548	3.7 (2.5 – 6.4)
Heidenhain Variant	48	3.0 (2.4 – 4.2)
Psychiatric Onset	42	7.1 (5.0 – 11.0)
Slowly Progressive Dementia	50	23.2 (16.3 – 30.0)
Cerebellar Onset	103	5.2 (3.5 – 8.7)
Extra-pyramidal Onset	3	14.8 (3.0 – 16.8)
Stroke-like Onset	20	2.4 (1.7 – 3.4)
Sensory Onset	17	7.5 (4.8 – 12.4)
Other Focal Onset	39	6.7 (2.7 – 16.2)
Onset Missing	6	4.4 (2.3 – 9.8)

Adjusted case-fatality

Case-fatality was modelled adjusting for age group, sex, year of onset and molecular subtype. The hazard of death six months after illness onset increased with age (Table 23). For example, at six months the hazard of death was seven times greater in sCJD cases aged 80 years and over at onset relative to those aged less than 50 years old at onset, Hazard Ratio (HR) 7.21 (2.69 – 19.31). The hazard of death did not vary according to sex and did not improve over time (at either six month or 1 year end-points). Following adjustment for age group, sex and year of onset the hazard was lower in sCJD cases with the MM1 molecular subtype relative to all other molecular subtypes, at both six month and1 year end-points.

Table 23 Hazard ratios for death (and 95% Confidence Intervals) at 6 months and 1 year after symptom onset in sCJD cases from the UK (1990 – 2006), adjusted for age group, sex, year of onset and molecular subtype

		Hazard Ratios of death (95% CI)	
		6 months after onset	**1 year after onset**
Age group	**<50 years**	1.00 (Reference group)	1.00 (Reference group)
	50 - 59 years	2.19 (0.84 – 5.72)	1.93 (1.03 – 3.58)
	60 – 69 years	2.823(1.12 – 7.12)	2.57 (1.41 – 4.67)
	70 – 79 years	4.19 (1.66 – 10.60)	3.71 (2.02 – 6.81)
	≥ 80 years	7.21 (2.69 – 19.31)	6.06 (3.03 – 12.10)
Sex	**Men *vs.* Women**	0.80 (0.60 - 1.06)	0.85 (0.67 - 1.07)
Molecular subtype	**MM1**	1.00 (Reference group)	1.00 (Reference group)
	MM2	0.10 (0.02 – 0.42)	0.16 (0.07 – 0.35)
	MM 1 / 2	0.21 (0.05 – 0.87)	0.28 (0.11 – 0.68)
	MV1	0.31 (0.20 – 0.50)	0.33 (0.23 – 0.47)
	MV2	0.04 (0.02 – 0.15)	0.15 (0.09 – 0.24)
	MV 1 / 2	0.12 (0.02 – 0.85)	0.10 (0.02 – 0.40)
	VV1	0.39 (0.12 – 0.96)	0.51 (0.37 – 0.71)
	VV2	0.34 (0.20 - 0.56)	0.47 (0.32 - 0.69)
	VV 1 / 2	0.83 (0.34 - 2.03)	0.85 (0.68 - 1.07)
Year (onset)	**(per year)**	0.98 (0.95 – 1.01)	1.00 (0.97 – 1.03)

sCJD mortality rates

Age and sex-specific mortality rates

Age-specific sCJD mortality rates in men and women are shown in Figures 37. As would be anticipated given the high case-fatality and short illness duration, these closely mirrored sCJD incidence rates. In both men and women sCJD mortality rates in those aged under 50 years old were low, rising to peak in men and women aged 70 – 79 years, before falling in those over 80 years of age. In men there was no statistically significant difference between sCJD mortality rates in those aged 60 – 69 years (rate 3.73 (3.17 – 4.29) per million), 70 – 79 years (rate 4.15 (3.43 – 4.86) per million) and 80 years and over (rate 2.74 (1.83 – 3.64) per million). In women sCJD mortality rates in those aged 60 – 69 years (rate 3.24 (2.74 – 3.74) per million) were comparable to sCJD mortality rates in those aged 70 – 79 years (rate 3.68 (3.10 – 4.27) per million). However the declined in sCJD mortality rates in those aged 80 years and over was significant (rate 1.57 (1.10 – 2.03) per million.) The 95% confidence intervals of age specific rates in men and women overlapped in each age group indicating that overall there was no significant difference in age-specific sCJD mortality rates according to sex.

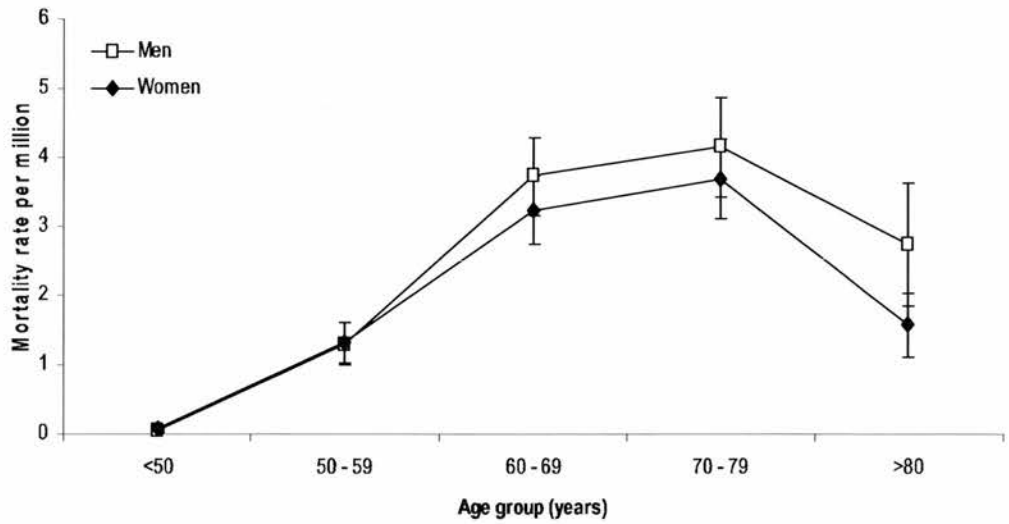

Figure 37 Age-specific sCJD mortality rates according to sex, 1990 – 2006

Temporal trends in age and sex-specific mortality rates

Temporal trends in sCJD mortality rates closely resembled temporal trends in sCJD incidence rates. There was no clinically or statistically significant change in sCJD mortality in men and women under 60 years of age. A clinically, but not statistically significant increases in sCJD mortality rates in men aged 60 – 69 years and 80 years and over and women over 60 years of age was observed. Finally a statistically significant increase in the sCJD mortality rate in men aged 70 – 79 years was observed with an APC in the sCJD mortality rate of 9.47% (4.62 – 14.54).

Age standardised mortality rates

Age standardised sCJD mortality rates in men and women are shown in Figure 38. In men, the sCJD mortality rate was 0.27 (0.07 – 0.47) per million in 1990, rising to peak at 1.36 (0.94 – 1.77) per million in 2003 and stabilising thereafter. At the end of the study period, 2006, the sCJD mortality rate was 1.27 (0.87 – 1.67) per million. In women, the sCJD mortality rate rose from 0.33 (0.13 – 0.54) per million in 1990 to peak at 1.29 (0.88 – 1.69) per million in 2002 and stabilised thereafter. At the end of the study period, 2006, the sCJD mortality rate was 0.90 (0.57 – 1.23) per million.

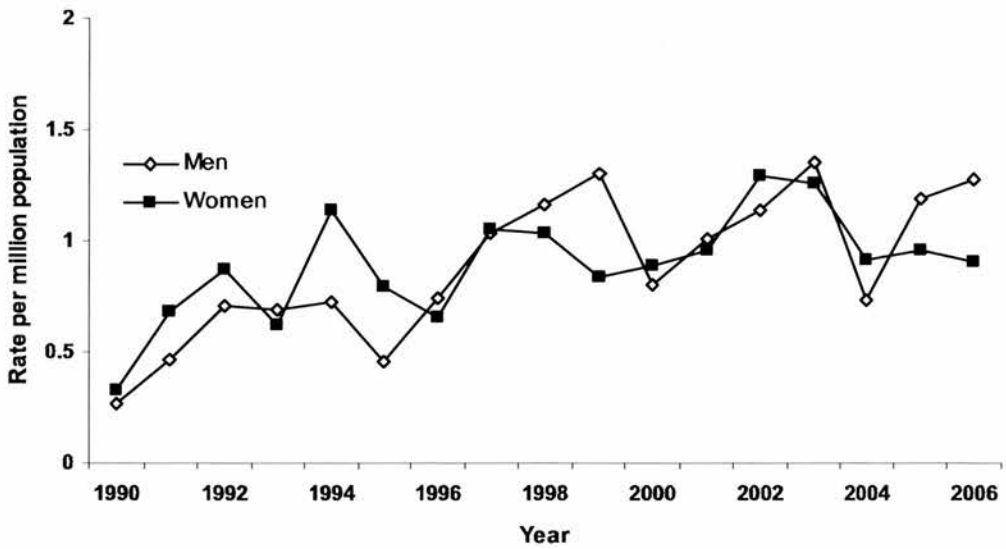

Figure 38 Age standardised sCJD mortality rates in men and women, 1990 – 2006

Temporal trends in age standardised mortality rates

Following adjustment for age there was a statistically significant increase in sCJD mortality rates in both men and women across the study period. In men from 1990 through 2006 the APC in the sCJD mortality rate was 5.47% (2.45 – 8.58). In women the APC in sCJD the mortality rate over the same period was 2.82% (0.21 – 5.50).

Variant CJD

From 1st May 1990 to 31st December 2006 there were 165 incident cases of vCJD in the UK, 115 (70.0%) definite and 50 (30.0%) probable vCJD cases (Table 24). In total, 92 (55.8%) vCJD cases were in men including 67 (58.3%) definite and 25 (50.0%) probable vCJD cases. There was no significant difference in the proportion of definite or probable cases according to sex (P= 0.326). The median age at symptom onset was 26.6 years (IQR 20.7 – 33.3). The youngest vCJD case was 12.6 years old at symptom onset; the oldest 74.4 years old. Age at symptom onset did not vary according to sex (P=0.096) or change over time (P= 0.953). Of note all vCJD cases ascertained by the NCJDSU over this period were born before 1989.

The majority of vCJD cases, 89.1% (143), were referred to the NCJDSU by a neurologist. As of 31st December 2008, 3 probable vCJD cases referred to the NCJDSU prior to 31st December 2006 were known to be alive; 2 men (aged 17.6 and 18.4 years at symptom onset) and one woman (aged 17.3 years at onset). These individuals had illness durations of 74.5, 87.5 and 70.5 months respectively at the time of data censoring.

Table 24 Characteristics of vCJD cases ascertained by the NCJDSU according to year of referral, 1990 -2006

Year of referral	Cases, n (%)	Definite cases, n (%)	Male, n (%)	Age at Onset, Years (IQR)	Median Illness Duration, months (IQR)	Post Mortem, n (%)	Tonsil Biopsy, n (%)
1995	8 (4.8)	8 (100)	2 (25.0)	28.5 (23.1 - 29.3)	14.4 (11.1 - 20.3)	3 (100)	0
1996	9 (5.5)	9 (88.9)	6 (66.7)	28.7 (24.4 - 32.9)	14.0 (12.1 - 23.5)	8 (80.0)	0
1997	13 (7.9)	12 (92.3)	3 (23.1)	22.5 (18.6 - 31.4)	16.7 (12.3 - 24.9)	7 (70.0)	4 (30.8)
1998	20 (12.1)	18 (90.0)	12 (60.0)	24.6 (21.5 - 34.7)	13.9 (11.5 - 16.2)	15 (83.3)	7 (35.0)
1999	16 (9.7)	16 (100)	9 (56.3)	27.7 (15.9 - 32.9)	14.5 (9.1 - 18.3)	15 (100)	0
2000	29 (17.6)	24 (82.8)	20 (69.0)	27.3 (23.7 - 31.2)	11.6 (10.8 - 14.1)	24 (85.7)	2 (6.9)
2001	21 (12.8)	5 (23.8)	10 (47.6)	25.5 (19.9 - 32.8)	13.8 (10.9 - 20.0)	13 (65.0)	8 (38.1)
2002	15 (9.1)	7 (46.7)	10 (66.7)	26.4 (18.6 - 37.1)	15.0 (11.2 - 17.2)	5 (29.4)	5 (33.3)
2003	16 (9.7)	7 (43.8)	10 (62.5)	27.4 (21.6 - 37.2)	14.8 (13.3 - 16.8)	8 (44.4)	3 (18.8)
2004	6 (3.6)	3 (50.0)	4 (66.7)	32.6 (25.2 - 33.8)	9.8 (8.1 - 15.1)	3 (33.3)	2 (33.3)
2005	7 (4.2)	4 (57.1)	3 (42.9)	29.1 (22.2 - 33.1)	13.4 (12.1 - 33.1)	3 (60.0)	2 (28.6)
2006	5 (3.0)	3 (60.0)	3 (60.0)	31.3 (26.1 - 33.6)	11.5 (11.8 - 16.2)	2 (40.0)	2 (40.0)
All	165 (100)	115 (70.0)	92 (55.8)	26.6 (20.7 - 33.3)	13.8 (11.1 - 17.6)	109 (67.3)	35 (21.1)

vCJD incidence rates

Age and sex-specific incidence rates

Age specific vCJD incidence rates in men and women are shown in Figure 39. In men, the vCJD incidence rate increased from 0.17 (0.08 – 0.25) per million in those aged 19 years and under, to peak at 0.95 (0.56 – 1.34) per million in those aged 25 – 29 years, before falling to 0.13 (0.07 – 0.18) per million in those aged 35 years and over. The 95% confidence intervals for vCJD incidence rates in the age groups 20 – 24 years, 24 – 29 years and 30 – 34 years overlapped indicating that there was no statistically significant difference in between groups. In women, the vCJD incidence rate increased from 0.19 (0.09 – 0.27) per million in those aged 19 years and younger to peak at 0.86 (0.47 – 1.25) per million in those aged 20 – 24 years before falling to 0.06 (0.03 – 0.09) per million in those aged 35 years and over. As for men, there was no significant difference in the vCJD incidence rate in women aged 20 – 24 years, 25 – 29 years and 30 – 34 years.

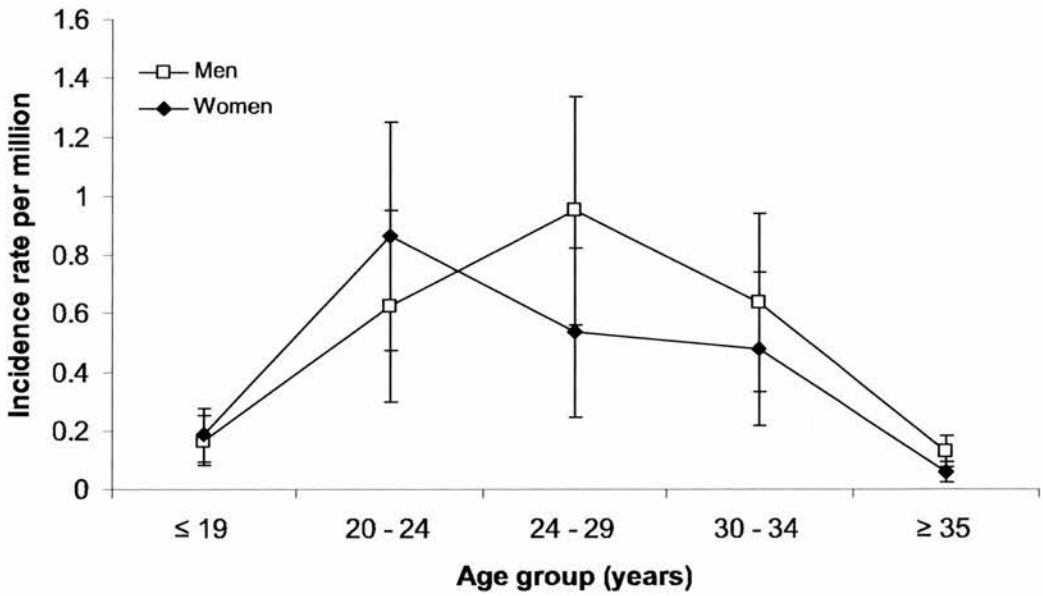

Figure 39 Age-specific vCJD incidence rates according to sex, 1995 – 2006

Temporal trends in age-specific incidence rates

There was no statistically significant increase in vCJD incidence rates over time according to age group in men or women. However, due to the small number of incident vCJD cases in each age group and sex the analyses were underpowered. A statistically non-significant but clinically relevant increase in vCJD incidence rates was observed in men age 20 – 24 years in whom the vCJD incidence rate increased from 0 per million in 1995 to peak at 2.85 (0.35 – 5.34) per million in 2000 before falling to 0.63 (0.30 – 0.95) per million in 2006. A similar trend was observed in men aged 25 – 29 years in whom the vCJD incidence rate increased from 0 per million in 1995 to 2.98 (0.60 – 5.36) per million in 2000 before falling to 0.95 (0.56 – 1.34). In women a non-significant but clinically relevant increase in the vCJD incidence rate from 0 per million in 1995 to 2.25 (0.04 – 4.45) per million in 1997 in the age group 20 – 24 years was observed.

Age standardised incidence rates

Age standardised vCJD incidence rates in men and women are shown in Figure 40. In men the age standardised vCJD incidence rate rose from 0.07 (0.03 – 0.17) per million in 1995 to peak at 0.69 (0.39 – 1.00) per million in 2000, before falling to 0.11 (0.01 – 0.24) in 2006. In women, the vCJD incidence rate rose from 0.15 (0.03 – 0.27) per million in 1995 to 0.36 (0.13 – 0.54) in 2001 thereafter falling to 0.06 (0.00 – 0.15) per million in 2006.

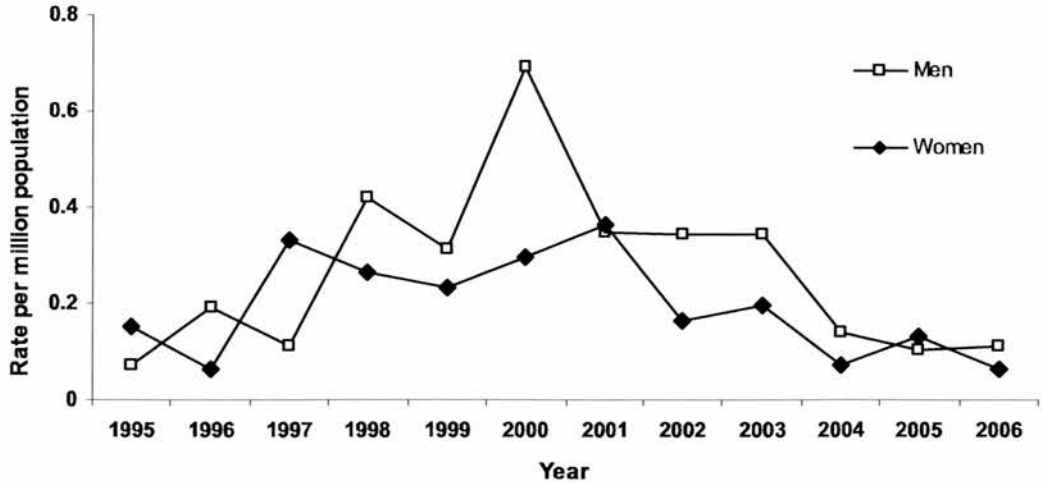

Figure 40 Age standardised vCJD incidence rate in men and women, 1995 – 2006

Temporal trends in age standardised incidence rates

Following adjustment for age there was a statistically significant increase in the vCJD incidence rate in men between 1995 and 2000. The APC in vCJD incidence rate over this period was 42.60% (3.40 – 96.70). Between 2000 and 2006 the vCJD incidence rate fell with an APC of -26.24% (-41.90 – -6.37). In women, there was no statistically significant change in the vCJD incidence rate between 1995 and 2006 (APC -3.73 (-14.45 – 8.33)).

Clinical presentation

Over half of all vCJD cases, 86 (52.1%), presented with psychiatric symptoms at onset, 52 (31.5%) with neurological symptoms (predominantly sensory disturbance), and 27 (16.4%) with mixed psychiatric and neurological symptoms. Clinical presentation varied by sex, with an excess of men presenting with psychiatric onset relative to women (P= 0.034). There was no statistically significant difference in clinical presentation with respect to age group (P=0.934) and no change in clinical presentation over time (P=0.354).

Diagnostic investigations

In this section the use of EEG, MRI, CSF 14-3-3 protein, tonsil biopsy, brain biopsy, *PRNP* Codon 129 genotyping, full sequencing for *PRNP* mutations and post mortem examination in the investigation of vCJD cases will be described.

EEG

All vCJD cases underwent at least one EEG examination during the course of their clinical illness; 153 (92.7%) cases underwent multiple examinations (range 1 – 3). The typical EEG described in sCJD (PSWC) was not found in any of the EEG examinations undertaken.

MRI

The majority of vCJD cases, 163 (98.2%), underwent at least one MRI examination during the clinical course of their illness; 49 (29.7%) underwent multiple examinations (range 1 - 3). Radiological changes consistent with vCJD were seen on the MRI of 141 (85.5%) vCJD that underwent this investigation (any sequence). Eighteen (11.0%) vCJD cases did not have signs consistent with vCJD on MRI scanning. In two vCJD cases MRI scanning was reported as 'equivocal' and in a further two vCJD cases the images were degraded due to movement artefact. Overall, the median time from symptoms onset to characteristic MRI scan was 8.1 (5.9 – 10.5) months (n=140). This was invariant over time (P= 0.095).

CSF 14-3-3 protein (limited to 1996 onward)

The majority of vCJD cases, 126 (80.3%) underwent at least one CSF 14-3-3 protein examination. There was no significant change over time in the proportion of vCJD cases undergoing CSF 14-3-3 protein examination (P=0.115). CSF 14-3-3 protein was positive in 53 (42.1%) vCJD cases. Overall, the median time from symptom onset to positive CSF 14-3-3 protein examination was 8.6 (6.9 – 10.3) months. This was invariant over time (P= 0.225).

Tonsil and brain biopsy

In total 35 (21.2%) vCJD cases underwent tonsil biopsy. Over time the there was a significant increase in the proportion of vCJD cases undergoing tonsil biopsy (P=0.023). vCJD cases that underwent tonsil biopsy did not differ significantly from those who did not with respect to age (P=0.094), sex (P=0.561) or clinical

presentation (P=0.340). Evidence of PrP^{Sc} was found in 33 (85.7%) vCJD cases that underwent tonsil biopsy. In one vCJD case tonsil biopsy was 'equivocal' and in the final vCJD case tonsil biopsy was negative for PrP^{Sc}. The median time from symptom onset to positive tonsil biopsy was 10.7 (8.3 – 12.7) months. Interestingly, of this group the majority, 29 (82.9%) had features on MRI examination that supported a diagnosis of vCJD prior to tonsil biopsy and hence were already classified as a probable vCJD cases. Tonsil biopsy contributed to case classification in just 6 (17.1%) of the vCJD cases that underwent this examination.

Ten (6.1%) vCJD cases underwent brain biopsy during life. These patients did not differ significantly from others with respect to age (P=0.881), sex (P=1.000) or clinical presentation (P=0.607). Interestingly, all but one of these vCJD cases had MRI features consistent with a diagnosis of vCJD and met the WHO diagnostic criteria as a probable vCJD case prior to brain biopsy. Brain biopsy was diagnostic in six of the ten vCJD cases; the remaining four cases all underwent post mortem examination on expiration.

PRNP Codon 129 Genotyping

PRNP Codon 129 genotyping was available for 149 (90.3%), vCJD cases, with no significant change over time in the proportion undergoing this investigation (P=0.226). All vCJD cases undergoing this examination were methionine homozygote at *PRNP* Codon 129.

PRNP mutation testing

Genetic testing to exclude a *PRNP* mutation was carried out on 128 (80.6%) vCJD cases. There was no significant change in the proportion of vCJD cases undergoing *PRNP* mutation testing over time (P=0.153).

Post mortem examination

As of 31st December 2008, three patients with probable vCJD referred to the NCJDSU prior to 31st December 2006 were still alive. Of the 162 deceased vCJD cases, 109 (67.3%) underwent post mortem examination. The rate of post mortem examination in vCJD cases varied over time (P<0.001) (Figure 41). Initially the rate was very high. A gradual decline in post mortem rates from 1999 (100%) through

2002 (24%) occurred, during which the number of tonsil biopsies carried out was high. Post mortem rates have fluctuated from 46% - 60% since 2002.

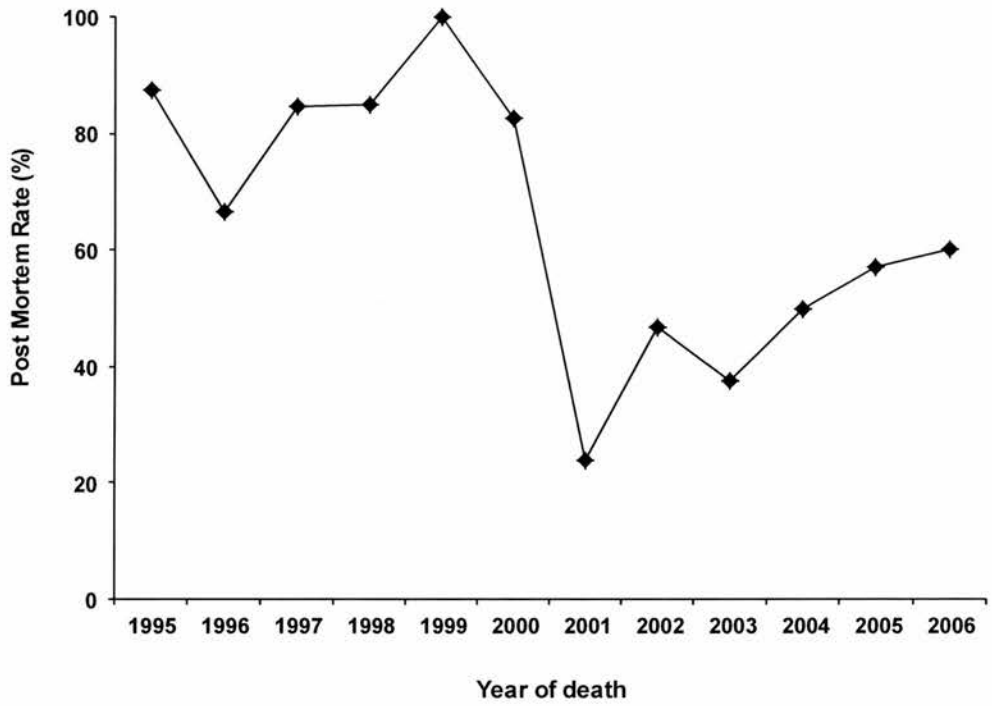

Figure 41 Post mortem rate according to year of death in vCJD cases, 1995 – 2006

PrP^{Sc} protein typing

PrP^{Sc} protein typing was carried out on 75 (64.7%) vCJD cases for which pathological material was available following tonsil biopsy, brain biopsy or post mortem examination. In all cases the PrP^{Sc} isotype was 2B.

Sensitivity of diagnostic investigations in vCJD

Figure 42 shows the proportion of all vCJD cases that underwent at least one MRI scan, tonsil biopsy or brain biopsy and had a positive result from these investigations respectively, from 1996 and 2006. Tonsil biopsy was the most sensitive of all investigations, with a sensitivity of 94.3% (86.6 – 1.00), followed by MRI scanning, 86.5% (81.3 – 91.7) and finally brain biopsy, 60.0% (29.6 – 90.4).

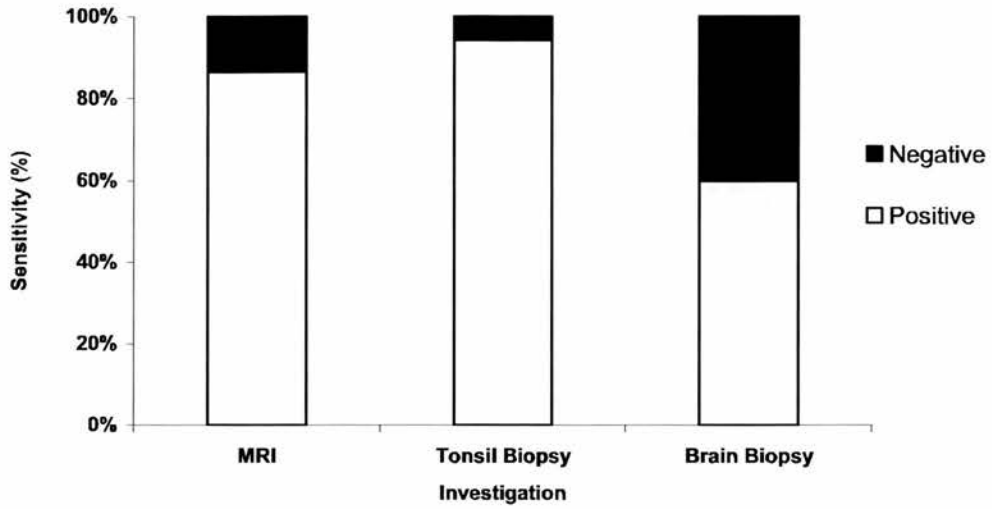

Figure 42 Sensitivity of diagnostic investigations in vCJD, 1996 - 2006

Definite vCJD cases with negative investigations

Between 1996 and 2006, nine definite cases of vCJD had a negative or non-contributory MRI scan and did not undergo tonsil biopsy. Men accounted for 4 (44.4%) of these cases. There was no difference in age (P=0.384), sex (P=0.357) or clinical presentation (P=0.649) between this group and all other vCJD cases. Just one of nine these vCJD cases underwent brain biopsy in life. This was diagnostic.

Case-fatality

Crude case-fatality

Crude case-fatality at 1 and 2 years was 35.2% (58) and 86.7% (143) respectively. Median survival overall was 13.8 (IQR 11.1 - 17.6) months. This was invariant over time (P= 0.949). The median survival was greater in women, 15.7 (11.3 - 20.9) months than in men, 13.2 (10.8 - 16.6) months (P= 0.026). Median survival varied by age group as seen in Figure 43 (P<0.001). Cases in the youngest age group, aged 19 years and under had the longest median survival at 18.2 (13.6 – 32.6) months; cases aged 25 – 29 years had the shortest median survival at 12.0 (10.0 – 16.5) months (P<0.001). Median survival did not vary according to clinical presentation (P=0.211).

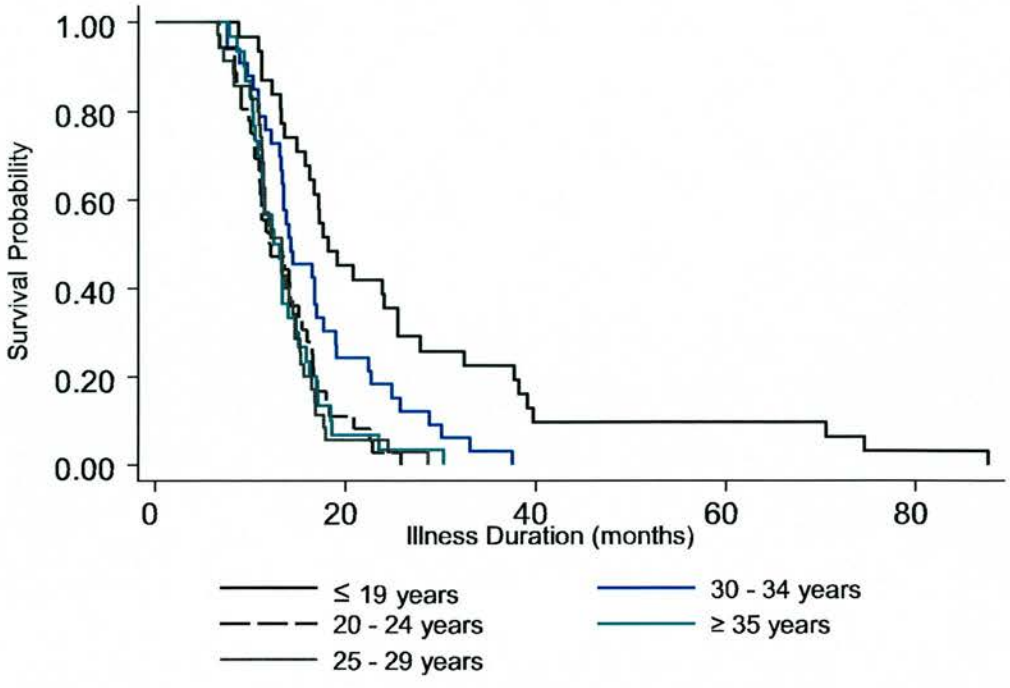

Figure 43 Kaplan-Meier estimates of survival (months) in vCJD cases, 1995 – 2006, according to age group

Adjusted case-fatality

The hazard of death at 1 year increased with age (Table 25). Relative to cases aged 19 years and under at symptom onset, the hazard of death at 1 year was almost five times higher in cases aged 25 - 29 years at symptom onset,, HR 4.57 (1.53 – 12.62). The hazard of death increased with age in all groups, relative to the youngest age group. Following adjustment for age group and year of symptom onset, the hazard of death at 1 year did not vary according to sex or clinical presentation at onset. There was no change in the hazard of death at 1 year over time, HR 1.02 (0.93 – 1.10). Similar trends were observed for adjusted case-fatality at 2 years.

Table 25 Hazard ratios for death (95% Confidence Interval) in vCJD cases at 1 and 2 years after symptom onset, adjusted for year of symptom onset, sex and age group

		Hazard Ratios (95% CI)	
		1 year	2 years
Year (onset)	Per year	1.02 (0.93 - 1.11)	1.06 (1.00 - 1.12)
Age group	**≤ 19 years**	1.00 (Reference)	1.00 (Reference)
	20 – 24 years	2.17 (0.65 - 7.23)	2.00 (1.10 - 3.62)
	25 – 29 years	4.57 (1.53 - 12.62)	3.40 (1.92 - 6.02)
	30 – 34 years	3.96 (1.34 - 12.11)	3.44 (1.90 - 6.20)
	≥ 35 years	4.04 (0.40 - 1.22)	3.45 (1.92 - 6.17)
Sex	**Men *vs.* Women**	0.70 (0.40 - 1.22)	0.71 (0.50 - 1.01)
Clinical Presentation	**Psychiatric onset**	1.00 (Reference)	1.00 (Reference)
	Neurological onset	1.10 (0.59 – 2.08)	0.88 (0.60 – 1.30)
	Both	1.76 (0.86 – 3.60)	0.90 (0.55 – 1.48)

vCJD mortality rates

Age-specific mortality rates

As for sCJD, trends in mortality from vCJD closely mirrored trends in vCJD incidence (Figure 44). The age-specific vCJD mortality rate increased from 0.14 (0.07 – 0.22) per million in men aged 19 years and under, to peak at 0.95 (0.56 – 1.34) per million in men aged 25 – 29 years, before falling to 0.12 (0.07 – 0.17) per million in men aged 35 years and over. In women, the age-specific vCJD mortality rate increased from 0.16 (0.08 – 0.25) per million in those aged 19 years and under, to peak at 0.82 (0.44 – 1.19) per million in those aged 20 – 24 years old, before falling to 0.06 (0.03 – 0.09) per million in those age 35 years and over.

Temporal trends in age-specific mortality rates

Over time there was no statistically significant increase in vCJD mortality rates in either sex or any age group. As for temporal trends in vCJD incidence rates this is likely to be in part due to the small sample size leading to insufficient statistical power to detect a change in the trend. In men a non-statistically significant but clinically relevant increase in vCJD mortality rates was observed in those aged 25 - 29 years, in whom the incidence rate increased from 0 per million in 1990 to 2.48 (0.31 – 4.66) per million in 2000 before falling to 0.95 (0.56 – 1.34) per million in

2006. In women a similar trend was observed in vCJD cases aged 20 -24 years in whom the mortality rate increased from 0 per million to 2.89 (0.36 – 5.43) per million in 1998 before falling to 0.82 (0.44 – 1.19) per million in 2006.

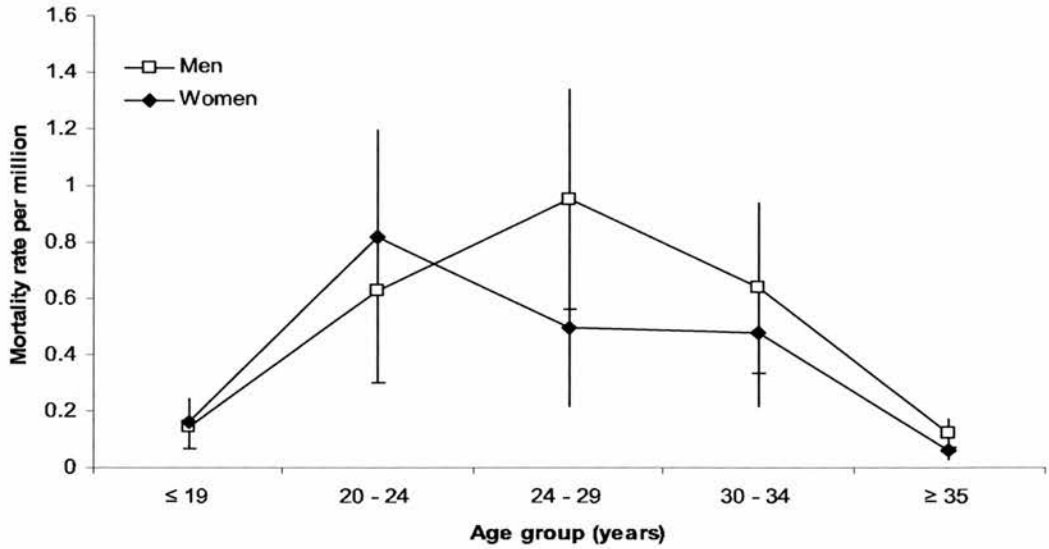

Figure 44 Age-specific vCJD mortality rates in men and women, 1995 – 2006

Age standardised mortality rates

Age standardised vCJD mortality rates in men and women are shown in Figure 45. In men, the age standardised vCJD mortality rate rose from 0.07 (0.03 – 0.17) per million in 1995 to peak at 0.52 (0.26 – 0.78) per million in 2000, before falling to 0.15 (0.00 – 0.29) in 2006. In women, the rate rose from 0.03 (0.00 – 0.08) per million in 1995 to 0.46 (0.21 – 0.71) in 2000, before falling to 0.03 (0.00 – 0.09) per million in 2006.

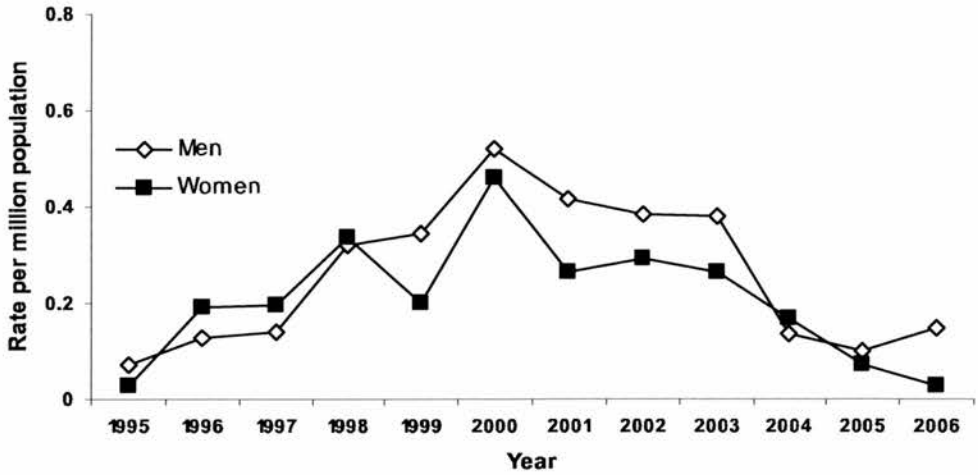

Figure 45 Age standardised vCJD mortality rates in men and women, 1995 – 2006

Temporal trends in age standardised mortality rates

In men there was a statistically significant increase in vCJD mortality rates between 1995 and 2000, with an APC of 47.12% (13.87 – 90.09). From 2000 through 2006, the vCJD mortality rate in men declined significantly such that the APC was -21.34% (-33.47 - -7.00). In women there was no statistically significant change in the vCJD mortality rate over the study period (APC 1990 – 2006: -2.55% (-15.56 – 12.45)).

vCJD cases attributed to the transfusion of labile blood components

Between 1st May 1990 and 31st December 2006, four vCJD cases (one asymptomatic) transmitted through the transfusion of labile blood components, were ascertained by the NCJDSU. All were the recipients of non-leukodeplete red blood cells that had been donated by individuals who subsequently developed vCJD. The median time from blood donation to donor symptom onset was 18.3 (16.6 – 29.7) months. For disease surveillance purposes, these cases are considered vCJD, rather than iCJD cases. They have therefore been included in the figures already presented under the section on vCJD. This group are however of special interest because they have acquired vCJD through a previously unrecognised route of transmission. Accordingly, the epidemiological characteristics of the group will be described in

this section. Due to small numbers comparison with the entire cohort of vCJD cases is not possible and this section will be limited to descriptive epidemiology only.

All three symptomatic vCJD cases were in men. The median age at symptom onset was 68.8 (31.3 – 74.2) years. Clinical presentations varied: one case presented with RPD, one with sensory features and the third with psychiatric symptoms. The median incubation period from transfusion to symptom onset was 7.8 (6.5 – 8.3) years. All three vCJD cases underwent EEG and MRI examinations during the course of clinical illness; none underwent CSF 14-3-3 protein examination. EEGs were unremarkable in all three cases. The MRI scan was consistent with a diagnosis of vCJD in one case only. These findings were first observed 4.7 months after symptom onset, in the final phase (last quarter) of the clinical illness. The remaining two cases also underwent MRI examination in the advanced stages of illness however imaging was not consistent with vCJD in either case. The median duration of illness was 11.5 (10.3 – 13.3) months. All cases underwent post mortem examination; in addition one case underwent tonsil biopsy in life from which PrP^{Sc} was detected. In all cases the molecular subtype was MM 2B.

The asymptomatic case occurred in an individual that was methionine heterozygote at *PRNP* Codon 129. PrP^{Sc} was detected in the spleen of this individual at post mortem 5 years following transfusion of the implicated labile blood components. This individual died from non-neurological cases and had no signs of symptoms suggestive of vCJD in life.

Iatrogenic CJD

In the UK, acquired CJD cases attributed to the use of cadaveric-derived human pituitary hormones, cadaveric-derived dura mater grafts and the transfusion of labile blood components have been ascertained by the NCJDSU. vCJD cases attributable to the transfusion of labile blood components are considered vCJD cases for disease surveillance purposes, not iCJD cases. These were addressed in the previous passage and will not be revisited here. This section will focus on iCJD acquired via other routes of exposure. The distribution of iatrogenic cases in the UK annually according to route of exposure is shown in Figure 46 below.

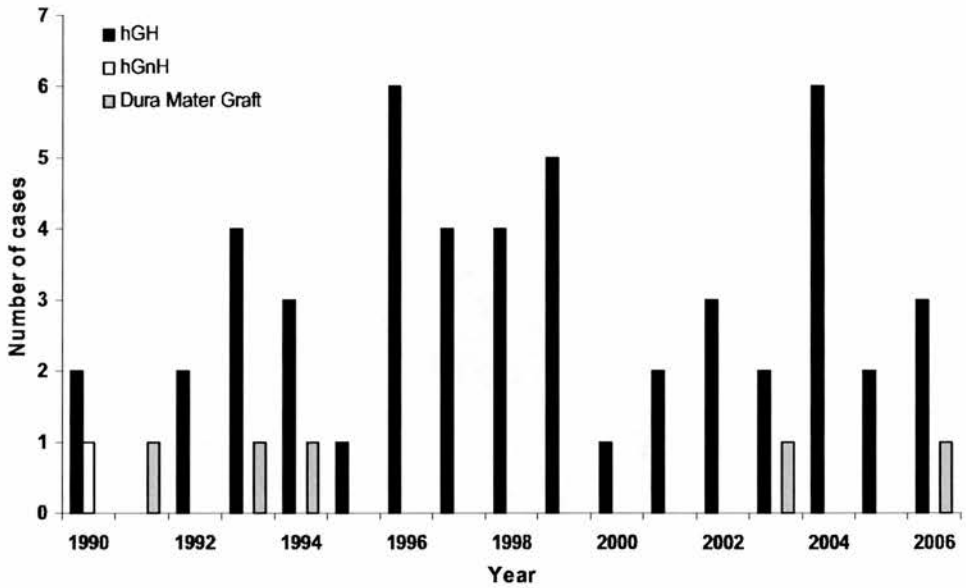

Figure 46 iCJD cases in the UK according to year and route of exposure, 1990 - 2006

Cadaveric-derived human pituitary hormones recipients

In total, 49 cases of iCJD in the recipients of cadaveric-derived human pituitary-derived hormone have been ascertained by the NCJDSU from 1st May 1990 through 31st December 2006; 41 definite and 8 probable cases. One iCJD case arose as a result of administration of human gonadotrophin hormone (hGnH). The remainder as a result of the administration of human growth hormone (hGH). The clinical

phenotype in hGH and hGnH cases is similar. Therefore in the analyses that follow these cases were grouped together and are referred to as hGH cases.

Men accounted for over half, 30 (61.2%) of all hGH-related iCJD cases. The mean age at symptom onset was 31.0 (SD 6.4) years. This did not vary by sex (P=0.862). The majority of cases, 41 (83.7%) had a cerebellar onset; 7 (14.3%) presented with psychiatric symptoms at onset. Median illness duration was 9.3 months (IQR 7.1 – 17.8). This did not vary by sex (P=0.231). Cases were administered hGH from 1968 through 1985, for a median of 6.0 (4.0 - 9.0) years. The mean age at first exposure was 10.1 (SD 5.0) years and last exposure 16.3 (4.4) years. The mean incubation period (from the midpoint of hGH administration) was 16.9 (4.3) years (range 10 – 27 years). The minimum and maximum mean incubation periods were 13.9 (4.3) years with a range of 5 – 17 years, and 19.3 (5.7) years with a range of 7 – 23 years, respectively. As of 31st December 2008, 48 cases were known to be deceased.

During the course of the clinical illness 35 hGH-related iCJD cases were known to have undergone EEG examination; none showed changes that would be considered typical of sCJD (PSWC) (Table 26). Of the 34 hGH-related iCJD cases known to have undergone MRI examination, 8 (23.5%) had features were consistent with sCJD. Characteristic MRI findings were reported a median of 9.5 (7.5 – 13.5) months after symptom onset. CSF 14-3-3 protein was positive in over half, 13 (56.5%) of the hGH-related iCJD cases known to have been tested. CSF 14-3-3 protein was first positive a median of 5.0 (2.7 – 6.2) months after symptom onset.

Table 26 Investigations undertaken in hGH-related iCJD cases, 1990 - 2006

	Number of cases undergoing investigation	Number of positive tests	Sensitivity of test, % (95%CI)
EEG	35 / 36	0 / 35	0
MRI	34 / 38	8 / 34	23.5% (9.3 – 37.8)
CSF 14-3-3 protein	23 / 49	13 / 23	56.5% (36.3 – 76.8)

Codon 129 genotyping was known in 63.4% (31) of hGH-related iCJD cases, of whom 1 (3.2%) case was methionine homozygotes, 16 (51.6%) were heterozygotes and 14 (45.2%) valine homozygotes. Median illness duration was significantly longer in Codon 129 heterozygotes, 16.3 months (13.3 – 20.8), than valine homozygotes, 6.3 months (5.3 – 7.9) (P<0.001). There was no significant difference in mean incubation period according to Codon 129 genotype (P=0.310). Post mortem examination was performed in 38 (80%) hGH-related iCJD cases; brain biopsy, in life, in three.

Cadaveric-derived dura-mater graft recipients

Over the study period there were 5 cases of iCJD, 4 definite and one probable case, in dura-mater graft recipients. Two occurred in men. The median age at symptom onset was 44.6 (33.6 – 46.7) years. Of the five cases, four presented with a cerebellar syndrome, the fifth isolated sensory symptoms. All had received a Lyodura graft. In four of the five, the date of grafting was known. This was between 1983 and 1987. The median incubation period from dura-mater grafting to symptom onset was 7.5 (range 7.0 – 15.0) years. During the course of the clinical illness all cases underwent serial EEG examinations. In three cases the EEG was considered typical for sCJD, showing PSWC. The median time from symptom onset to typical EEG was 2.6 (1.4 - 3.8) months. Three cases underwent MRI scanning; one had radiological features consistent with sCJD at 0.3 months after symptom onset. Only one case underwent CSF 14-3-3 protein examination; this was negative. All five cases were deceased. The median duration of illness was 6.2 (5.1 – 10.5) months. Post mortem examination was carried out in four of the five cases. All cases (n=3) for whom prion protein type and *PRNP* Codon 129 genotype was known, were molecular subtype MM1.

Genetic prion disease

The NCJDSU was informed of 116 genetic prion disease cases from 1st May 1990 to 31st December 2006; 42 (36.2%) cases of Gerstmann-Straussler-Scheinker Disease (GSS), 5 (4.3%) cases of fatal familial insomnia (FFI) and 69 (59.5%) cases of genetic CJD (gCJD). Genetic prion disease accounted for 9.4% of all prion disease cases in the UK over this period. The diagnosis was neuropathologically confirmed in 71 (61.2%) cases. As of 31st December 2008, 20 genetic prion disease cases referred to the NCJDSU over the study period were known to be alive. Among the deceased, the rate of post mortem examination was highest in FFI cases (100%); 25 (71.4%) GSS cases and 40 (58.0%) gCJD cases were known to have undergone post mortem examination respectively. The characteristics of genetic prion disease cases are outlined in Table 27. A causative mutation was identified in 102 (87.9%) cases; all FFI cases, 35 (83.3%) GSS cases and 62 (89.9%) gCJD cases. Over half, 67 (58.3%) of all genetic prion disease cases were known to have a family history of prion disease; 2 (40.0%) FFI cases, 24 (57.1%) of GSS cases and 41 (59.4%) gCJD cases.

GSS

There was a preponderance of women with GSS (female: male ratio of 2:1). Overall the median age at symptom onset in GSS cases was 43.9 (37.5 – 55.2) years and median illness duration 39.0 (29.0 – 72.6). In the UK the most common mutation in GSS cases was the P102L mutation, accounting for 61.9% of all GSS cases. There was no statistically significant difference in median age at onset (P=0.1750) or median illness duration (P=0.1936) by causative mutation in GSS patients.

FFI

All FFI cases were caused by the D178N mutation. In FFI cases the median age at symptom onset was 49.3 (39.0 – 60.7) years and median illness duration 12.1 (9.2 – 20.8) months.

Genetic CJD

Octapeptide repeat insertion mutations accounted for 43 (62.3%) of all gCJD cases. Overall median age at symptom onset was 43.9 (37.5 – 55.2) years, with median

illness duration of 20.9 (5.9 – 93.0). With an increasing number of repeats, the age at symptom onset fell (P=0.0012). E200K mutations accounted for 19 (27.5%) of all gCJD cases. The median age at symptoms onset in cases was an E200K mutation was 60.1 (51.6 – 66.0) years with median illness duration of 4.4 (2.5 – 8.6) months. In total 16 (84.2%) gCJD cases with an E200K mutation were initially referred to the NCJDSU as a case of suspect sCJD and 14 (73.7%) were visited by a NCJDSU neurologist in life.

Table 27 Characteristics of genetic prion disease cases referred to the NCJDSU, 1990 – 2006

Disease	Causative Mutation	Number (alive)	Male, n (%)	Median Age Onset, Years (IQR)	Median Age at Death, Years (IQR)	Median Illness Duration, Months (IQR)	Know FH, n (%)	Onset date known, n
GSS	*All*	42 (7)	15 (35.7)	43.9 (37.5 - 55.2)	48.1 (44.8 - 57.5)	39.0 (29.0 - 72.6)	24 (57.1)	24
	P102L	26 (2)	8 (30.8)	51.1 (40.3 - 57.8)	54.8 (45.5 - 59.9)	43.1 (28.5 - 78.1)	16 (61.5)	15
	A117V	6	2 (33.3)	42.2 (41.9 - 44.0)	45.1 (44.0 - 46.8)	29.7 (25.0 - 34.5)	3 (50.0)	4
	P105L	1 (1)	0	35.2	-	-	0	1
	Q212P	1 (1)	0	37.2	-	-	0	1
	S132I	1	0	-	64.8	-	0	0
	Unspecified	7 (3)	5 (71.4)	36.1 (27.9 - 44.3)	42.9 (39.0 - 47.6)	81.0 (29.0 - 132.9)	5 (71.4)	2
FFI	**D178N**	5	2 (40%)	49.3 (39.0 - 60.7)	50.6 (40.7 61.6)	12.1 (9.2 - 20.8)	2 (40.0)	4
Genetic CJD	*Insertion - All*	43 (12)	24 (55.8)	42.9 (32.4 – 53.2)	49.4 (44.3 – 58.8)	20.9 (5.9 – 93.0)	32 (74.4)	25
	96bp	5	4 (80.0)	65.4 (56.8 – 67.7)	66.6 (57.0 – 67.9)	2.7 (2.6 – 14.9)	1 (20.0)	5
	120bp	4	3 (75.0)	55.8 (49.4 – 62.1)	64.7 (62.9 – 66.5)	107.6 (9.6 – 205.6)	3 (75.0)	2
	144bp	27 (10)	13 (48.2)	35.5 (31.7 – 42.9)	44.9 (39.1 – 48.0)	48.0 (20.7 – 101.3)	23 (85.2)	15
	168bp	1	0	-	32	-	1 (100)	1
	240bp	1 (1)	0	-	-	-	1 (100)	0
	Unspecified	5 (1)	4 (80.0)	49.6 (28.7 – 58.6)	48.2 (37.6 – 58.8)	54.3 (2.4 – 106.3)	3 (60.0)	3
	Point Mutations							
	E200k	19	12 (63.2)	60.1 (51.6 - 66.0)	60.4 (52.0 - 66.7)	4.4 (2.5 - 8.6)	7 (36.8)	17
	Y163STOP	1 (1)	0	-	-	-	Not known	0
	D178N (V)	1	1 (100)	56.2	57.4	14.4	0	1
	D167G	1	0	68.7	70.1	16.9	Not known	1
	V201I	1	0	64.4	64.5	0.9	0	1

Summary of key findings

- 72.7% of all prion disease cases ascertained in the UK from 1990 through 2006 were sCJD, 13.5% vCJD, 9.4% genetic prion disease and 4.4% iCJD.
- sCJD incidence and mortality peaked in men and women aged 70 – 79 years.
- Age standardised sCJD mortality rates increased over time in men and women.
- A trend toward increasing sCJD mortality was observed in men and women aged over 60 years old; this was significant in men aged 70 – 79 years old only.
- Overall median survival in sCJD was 4.3 months (invariant over time).
- Rates of case confirmation fell over time (sCJD and vCJD).
- EEG was commonly used in the assessment of sCJD cases although the sensitivity of this investigation was low and had fallen over time.
- Over time CSF 14-3-3 protein examination was increasingly used to support a diagnosis of sCJD in sCJD cases.
- Overall, fewer than half of all sCJD cases underwent genetic testing to exclude a *PRNP* mutations; in 2006 just one if every four sCJD cases.
- From 1990 through 2006, 165 vCJD cases were ascertained in the UK including 4 vCJD cases attributed to the transfusion of labile blood components.
- The primary vCJD epidemic peaked in 2000 (2001) in men (women) and has been in decline since.
- Median age at symptom onset in vCJD was 26.6 years (invariant over time).
- Median survival in vCJD cases was 13.8 months (invariant over time). Relative to older age groups, survival was greatest in those aged $\leq$ 19 years old.
- All vCJD cases tested were *PRNP* Codon 129 methionine homozygote.
- From 1990 through 2006, 54 iCJD cases were ascertained in the UK; 48 hGH-related, 1 hGnH-related and 5 in cadaveric-derived dura mater grafts recipients.
- Fewer than expected hGH-related iCJD cases were *PRNP* Codon 129 MM.
- 60% of genetic prion disease was accounted for by gCJD, 36% GSS and 4% FFI.
- A causative mutation was identified in over 90% of genetic prion disease cases.
- A quarter of all gCJD cases were attributable to the E200K mutation.
- In 40% of genetic prion disease cases no significant family history was reported.

Discussion

In this chapter I described the epidemiology of CJD in the UK according to disease subtype using data collected over 16 years of prospective surveillance in a country of almost 62 million people. This study examined 1,228 prion disease cases, including 935 (76.1%) with a neuropathologically confirmed diagnosis. A discussion of the key findings from this chapter follows.

sCJD

Trends in incidence and mortality rates

One of the most striking findings from this study is the consistent increase in age standardised sCJD mortality rates in men and women in the UK from 1990 through 2006. Due to the rarity of the sCJD minor statistical fluctuations in disease occurrence can cause an apparent excess of cases. This underscores the need for national longitudinal monitoring of temporal trends in disease occurrence using systematically collected data and the importance of having robust international data for comparison. Several countries with established systematic prospective disease surveillance systems, including Germany,(48) France,(236) Italy,(52) Switzerland,(237) Japan,(47) Canada(44) and Australia(44;238) have reported similar findings. In most countries a sustained increase in sCJD incidence/mortality has been attributed to improved case ascertainment as a result of systematic prospective surveillance and increasing awareness of all forms of CJD.(44;45;48) Diagnostic advances such as CSF 14-3-3 protein examination may also have contributed to these trends.(48;96;239) In the UK, CSF 14-3-3 protein is the most sensitive investigation used to support a diagnosis of sCJD and an increasing proportion of probable sCJD cases are meeting the clinical diagnostic criteria based on this investigation. Molecular subtyping has provided significant insights into the diverse clinico-pathological phenotype of sCJD which may have resulted in ascertainment of rare subtypes.(240) In Switzerland for example a two fold increase in the incidence of sCJD between 2000 and 2001 was attributed to an increase in case ascertainment of the rare MV2 subtype; an increase in sCJD incidence was accompanied by a significant change in the age and sex distribution of cases.(237) In the UK there has been a non-significant increase in sCJD cases with an MV or VV

genotype over time. This may then be evidence of improved ascertainment of rarer sCJD subtypes by the NCJDSU. However there has been no significant change in the proportion of 'atypical cases' over the study period as would be expected if this increased had been significant. These data suggest that whilst increasing ascertainment of rarer subtypes may have contributed to the overall trends in incidence/mortality, they are unable to fully explain them.

An examination of age-specific trends in sCJD incidence/mortality provides some additional insight to these data. In keeping with the published literature incidence/mortality rates were low in those under 50 years of age, increasing to peak in those aged 70 – 79 years of age, before falling in those aged 80 years and over.(44;47;48;50-52;238) This trend was seen in both men and women. A decline in sCJD incidence/mortality in those aged 80 years and over of age is incongruent with other neurodegenerative diseases but well described in sCJD. If sCJD is a truly sporadic disease then a continuous increase in the number of cases with increasing age would be expected.(241) Most commentators attribute an age-related fall in sCJD incidence/mortality to systematic under-ascertainment of cases in the very elderly. In the present study there was no significant difference in incidence/mortality rates for sCJD in men aged 60 – 69 years, 70 – 79 years or 80 years and over. In women there was a significant decline in incidence/mortality in the very elderly (80 years and over) compared to those aged 70 – 79 years. Indicating that in the UK there may be evidence of differential under ascertainment in sCJD cases according to sex. Over time there were clinically but not statistically significant increases in sCJD incidence/mortality in men and women over 60 years of age. The only statistically significant increase in sCJD incidence/ mortality occurred in men aged 70 – 79 years of age. These data suggest that in the UK the increase in sCJD incidence/mortality has occurred as a result of increased ascertainment of sCJD cases in men and women over 60 years of age with the greatest increase over time in men age 70 – 79 years. It remains possible that increasing incidence/mortality rates are attributable to increasing exposure to an unknown exogenous risk factor for sCJD. However exhaustive case-control studies have not provided compelling evidence of a putative risk factor for sCJD and the demonstration of this phenomenon in different

population over time using surveillance data collected by different methodologies make this possibility remote.

Is it possible that the apparent increase in incidence of, and mortality from, sCJD in the UK is actually due to misclassification of vCJD cases? This seems unlikely for a number of reasons. Firstly, the increase in incidence of, and mortality from, sCJD has not solely been confined to the UK, but observed in countries which have not reported BSE, such as Australia.(44;238) Secondly, in the UK and elsewhere the clinical phenotype of vCJD has been remarkably consistent and efficiently detected through routine surveillance. Thirdly, an examination of age-specific rates reveals that the sCJD incidence and mortality rates in men and women under 60 years of age are low and have not increased over time as would have been expected if these cases were due to vCJD; just 5% (45) of sCJD cases were aged less than 50 years old at symptom onset. In keeping with the published literature sCJD cases under 50 years of age were less likely to have a classical sCJD clinical course and supportive investigations including PSWC on EEG and a positive CSF 14-3-3 protein examination, than sCJD cases aged 50 years and over at symptom onset. Reassuringly, the diagnosis was neuropathologically confirmed in the majority, 87%, of sCJD under 50 years of age at onset; of the 6 for which a neuropathological diagnosis was unavailable, none had clinical features or investigations that would support a differential diagnosis of vCJD. Fourthly, as previously noted there has been no significant change over time in the proportion of 'atypical' sCJD cases which would be expected if these cases were vCJD. Finally, there has been a non-significant decrease in the proportion of sCJD cases with a methionine homozygote genotype over time; were these cases attributable to vCJD a rise in the proportion of cases with the methionine homozygote genotype would be expected.

Diagnostic investigations in sCJD

Falling rates of case confirmation

Definitive diagnosis of CJD requires examination of neuropathological material obtained from brain biopsy in life or autopsy following death. Overall a neuropathological diagnosis was available 77.2% (689) sCJD cases; 664 from autopsy examination following death and an additional 25 from brain biopsy in life.

Brain biopsy in life is rare and was undertaken in just 3.8% (34) of all sCJD cases, although in this population the sensitivity was high; brain biopsy was diagnostic in 73.5% (25) of cases. The remaining 9 cases with non-diagnostic brain biopsies all underwent post mortem examination following death. Post mortem rates in sCJD cases in the UK fell over time. A number of factors may have contributed to this including the willingness of clinicians to request an autopsy, the willingness of relatives to consent to an autopsy and the availability of appropriate facilities to carry out the examination.(137) An increasingly diverse clinical spectrum of sCJD has been recognised and the value of investigations such as EEG, MRI and CSF 14-3-3 protein in specific molecular subtypes of sCJD, clarified.(97) The inclusion of CSF 14-3-3 protein examination in the WHO diagnostic criteria has increased the sensitivity of a clinical diagnosis of sCJD to over 90%; MRI scanning has also made a significant contribution, particularly in atypical cases, although not yet incorporated into the diagnostic criteria.(99) From a disease surveillance perspective it is essential to ensure that a high proportion of suspect sCJD cases undergo post mortem examination following death. This maximises case ascertainment and provides valuable information on differential diagnoses and the diagnostic value of investigations, such as CSF 14-3-3 protein and MRI scanning, to inform clinical practice and research in this area. However the relatives of a sCJD cases may be reticent to consent to post mortem examination, and clinicians to ask for this, if it has been possible to reach a clinical diagnosis in life with a high degree of certainty. Thus an unintended consequence of the increasingly sensitive diagnostic criteria for sCJD may have been a fall in case confirmation rates.

EEG

The value of EEG in sCJD has been recognised for a number of decades and this investigation is generally available in local and certainly regional centres throughout the UK. In the UK the EEG remains the supportive diagnostic investigation most commonly utilised in sCJD cases; consistently over 90% of sCJD cases underwent at least one EEG during the course of their clinical illness. This is in contrast to international trends which show a reduction in the use of EEG in sCJD cases over time.(107) However the median number of EEGs undertaken per case in the UK has fallen over time. A corresponding reduction in the sensitivity of EEG, from 50% in

1990 to 33.3% in 2006 was observed. The timing of EEG examination in sCJD is important. EEG findings often evolve such that serial examinations may be required before PSWC are detected. It is likely that the fall in the number of EEGs carried out per case has contributed to the falling sensitivity of EEG in the UK over time. The introduction of CSF 14-3-3 protein as a viable diagnostic alternative to serial EEG examinations may have contributed to this. In Germany, Tschampa *et al* described the sensitivity and specificity of EEG in sCJD (definite or probable sCJD cases) as 32% and 94% respectively using data collected by the German Surveillance Unit between 2001 and 2003.(106) The estimated sensitivity in the study by Tschampa *et al* was comparable to that in the present study. Tschampa *et al* attributed the falling sensitivity of EEG over time in Germany (previous reports placed the sensitivity of EEG at 64% from 1996 through 2000 (104)) to suspect sCJD cases being referred to the surveillance unit earlier in the disease process, prior to the development of PSWC on EEG. They hypothesised that this has arisen as a result of the increasing use of CSF 14-3-3 protein examination to support a diagnosis of sCJD, although evidence to support this hypothesis was not provided. If this were the case in the UK it might be expected that the median time from symptom onset to first positive CSF 14-3-3 protein examination would have fallen over time. However this has not happened despite the increasing use of CSF 14-3-3 protein examination in the investigation of sCJD cases. Data from the UK suggest that CSF 14-3-3 protein and for that matter MRI scanning are being used in addition to EEG examination in the investigation of sCJD cases, with the latter investigations are being pursued in preference to serial EEGs. Overall, the median time from symptom onset to first typical EEG was shorter than the median time from symptom onset to first positive CSF 14-3-3 examination or first positive MRI scan.

CSF 14-3-3 protein

In this study CSF 14-3-3 protein was the most sensitive supportive diagnostic investigation in sCJD cases. The use of CSF 14-3-3 protein in the investigation of sCJD cases has increased significantly over time. Increasingly CSF 14-3-3 protein is contributing to the diagnosis of probable sCJD cases. Nevertheless, CSF 14-3-3 protein is less frequently undertaken in the investigation of sCJD cases in the UK when compared to other countries.(107) This may be a reflection on clinical practice

in the UK. CSF 14-3-3 protein requires lumbar puncture examination. Lumbar puncture examination is commonly undertaken in the investigation of subacute encephalopathy. However CSF 14-3-3 protein examination may not be considered at the time of lumbar puncture examination (this test in not available locally in the UK, only nationally) and the CSF sample obtained may not be appropriate for CSF 14-3-3 protein examination at a later date. If a diagnosis of sCJD has been made based on clinical features and a supportive EEG it may be clinically inappropriate to repeat a lumbar puncture examination solely for CSF 14-3-3 protein examination. Moreover in a moribund patient in whom consent has already been granted for post mortem examination on expiration and a treatable differential diagnosis has been excluded, lumbar puncture examination may be considered cruel and unnecessary.

MRI scanning

The value of MRI in sCJD has been reinforced recently and there have been renewed calls for the inclusion of MRI in clinical diagnostic criteria for sCJD.(99) Consistent with other European countries, MRI is increasingly being used in the UK in the investigation of sCJD.(107) MRI scanning is increasingly available in local centres although anecdotally this may be less available in the UK than elsewhere (personal communication R. Knight). The sensitivity of MRI in the present study (33.8% overall) was lower than published reports, the most recent of which by Zerr *et al* described the sensitivity of MRI using pooled data from definite or probable sCJD cases ascertained by the EUROCJD consortium as 83%. It should be noted however that the data in the present study span 16 years of prospective surveillance over which time the features on MRI scanning that are considered consistent with a diagnosis of sCJD have been defined and the optimal imaging sequences identified. The study by Zerr *et al* was limited to an examination of DWI or FLAIR imaging sequences only. The surveillance data analysed in the present study reflect routine clinical practice and are therefore unselected images many of which were not taken using the recently agreed optimal imaging sequences. This is likely to explain the lower sensitivity that I report. It is not clear whether an examination of only DWI and FLAIR images using contemporary data from the surveillance system in the UK would produce comparable results as the study by Zerr *et al*. Finally, MRI is known to be of particular value in the rarer subtypes of sCJD, especially the MV1, MV2 and

VV1 subtypes. The distribution of Codon 129 genotypes and molecular subtypes in the present study is comparable to that in the study be Zerr and colleagues, therefore the lower sensitivity that we report is unlikely to be attributable to the population under study.

Full sequencing for PRNP mutations

Worryingly, only half of all sCJD cases underwent testing to exclude a mutation of *PRNP*. This proportion fell significantly over time such that in 2006 just one in four sCJD cases underwent full sequencing of *PRNP*. Genetic prion disease may present in a clinical syndrome that is indistinguishable from sCJD (for example E200K mutation) and a significant proportion of genetic prion disease cases do not report a family history of prion disease. It is possible that genetic prion disease cases have been misclassified as sCJD. The proportion of all prion disease cases accounted for by genetic disease was almost 10%, fluctuating from 5% to 13% annually. Despite only a quarter of sCJD cases undergoing *PRNP* mutation testing in 2006, almost 13% of all prion disease cases ascertained by the NCJDSU in 2006 were aetiologically genetic. It is unlikely then that a significant degree of misclassification of cases has occurred. In the UK most prion disease cases are incapacitated at the time of diagnosis and consent to undertake genetic testing is provided by the next of kin, who is often a first degree relative. The diagnosis of genetic prion disease has significant implications for a patients relatives' – the penetrance of some mutations is variable, some individuals that have the mutation will not develop the disease but for those that do there is currently no disease modifying therapy available for this universally fatal condition. This may, in part, explain low levels of genetic testing.

Molecular subtyping

Molecular subtyping was available for a third of all sCJD cases. This figure is consistent with estimates from the EUROCJD network.(97) Type 1 and 2 PrP^{Sc} was found to co-exist in 6.5% of all cases which is considerably lower than the literature suggests with estimates varying from 12% to 44%. There are methodological limitations of the studies in this area. Many have examined small samples with significant bias in case selection and methodological variation in the diagnostic technologies used and approach to sampling of tissue. The present study utilises data

collected in routine surveillance. Neuropathological tissue was reviewed by the NCJDSU pathologist (JI) in the majority, 297, of the cases for which molecular subtyping was available. However, retrieval of brain tissue is carried out in centres across the UK and whilst guidance on tissue sampling is available this may not be followed. This is likely to have contributed to the low estimate of mixed protein subtypes in the UK. Indeed Collins *et al,* examining data collected by the EUROCJD consortium in routine surveillance from 1992 to 2002, also found type 1 and type 2 PrP^{Sc} co-existed in just 6% of the 743 sCJD cases they studied.(97) The distribution of molecular subtypes I found was in keeping with that reported by Collins *et al.*(97) The most common subtype was MM1 followed by MV2 and VV2. The clinical phenotype was also in keeping with this study. For example, MV1 cases were the oldest at disease onset and MM2 and VV1 the youngest whilst illness duration was shortest in the MM1 subtype and longest in the MM2, MV2 and MV 1/2 subtypes. MM1 cases were the most likely to have a typical EEG, VV2 cases a positive MRI scan and the sensitivity of CSF 14-3-3 protein was over 90% in the MM1, MM 1/2, MV1, VV2 and VV 1/2 subtypes. Over time there has been no significant change in the distribution of molecular subtypes in cases ascertained by the NCJDSU.

Survival in sCJD cases

Illness duration is commonly used as an indicator of clinical phenotype in sCJD. It was considered important in this study to determine whether median survival had changed over the study period and to examine the predictors of survival in this population. Overall median survival from sCJD was 4.3 months. Increased survival from sCJD has been reported in those with young age at onset, women, Codon 129 methionine heterozygotes, cases with a positive CSF 14-3-3 protein and Type 2 PrP^{Sc}.(94) Following adjustment for age group, sex, year of symptom onset and molecular subtype, there was an excess risk of death at 6 months and 1 year in individuals aged 70 – 79 years and 80 years and over, compared to those aged under 50 years at symptom onset. This age related effect has been documented previously and may relate to co-morbidity contributing to death.(94) There was no effect of sex on survival at either 6 months or 1 year. Previous studies in this area have produced conflicting accounts of the effect of sex on survival.(94) Following adjustment for age group, the hazard of death at 6 months remained highest in the MM1 and VV1/2

subtypes and at 1 year the MM1, VV2 and VV1/2 subtypes. There has been no significant change in the distribution of molecular subtypes of sCJD cases in the UK and no significant therapeutic advances over the study period. It is perhaps unsurprising then that there has been no significant increase in median survival in sCJD cases over time in the UK.

vCJD

vCJD was characterised in the UK in 1996.(37) Systematic prospective surveillance provided longitudinal data characterising the clinico-geno-pathological profile of all prion diseases in the UK from 1990. These data were essential in confirming that vCJD was indeed a novel prion disease with a distinct clinico-pathological phenotype. International surveillance data were equally important in determining that this phenomenon was unique (initially at least) to the UK. A case definition and diagnostic criteria based on these early cases was rapidly developed and subsequently adopted by the WHO for disease surveillance purposes.(242) Despite intensive systematic prospective surveillance efforts worldwide vCJD has been identified in only 11 countries outside the UK; 80% of all cases worldwide have occurred in the UK.(46) Unsurprisingly given the rarity of the disease and the concentration of cases in the UK, few data external to those collected by the NCJDSU, are available for comparison. Indeed most published literature describing the epidemiology of vCJD has been based upon cases ascertained by the NCJDSU. The present study is therefore one of the most comprehensive accounts of the epidemiology and diagnostic aspects of vCJD available.

The significance of age at onset

In contrast to sCJD (67.1 years (60.6 – 74.2)), the median age at symptom onset in vCJD cases was 26.6 years (20.7 – 33.3). Interestingly, this has not changed over time. All the vCJD cases ascertained by the NCJDSU were born prior to 1989, the year that the SBO ban was introduced. This suggests that the SBO ban was effective in preventing further exposure of the population to BSE. It would be anticipated that, with time, the median age at symptom onset in vCJD cases would increase. The fact that this has not occurred suggests that age related factors may influence the incubation period of the disease; vCJD cases that were younger at the time of

exposure experience longer incubation periods than vCJD cases that were older at the time of exposure. The possibility of dietary exposure to BSE through MRM between 1989 and 1996 remains. Overall the incidence of vCJD was greatest in men aged 25 – 29 years and in women aged 20 – 24 years. Dietary exposure alone cannot explain the age-related variation in vCJD incidence.(173) This observation suggests that younger age groups are more susceptible to BSE for a given level of exposure. The mechanisms that might mediate this effect are as yet unknown. It has been suggested that the development of Peyer's Patches in the gut, or a factor closely related to this, is a major determinant of age-related susceptibility.(143;243)

Survival in vCJD cases

Median illness duration was significantly longer in vCJD (13.8 (11.1 – 17.6) months) than sCJD (4.3 (2.7 – 7.9) months). Crude median survival was significantly longer in women than men in vCJD. However following adjustment for year of symptom onset, clinical presentation and age group, this survival advantage was no longer present. The hazard of death at one and two years was related to age such that relative to those aged 19 years and under, older cases had a greater risk of death at each time point. The reason for this is unclear. Individuals aged less than 35 years old would not be expected to have significant co-morbidity that might contribute to premature death. Differential recall of date of symptom onset according to age may have contributed to this. Often symptoms are non-specific in early disease and the majority of cases present with psychiatric symptoms. It is possible that the date of onset is more accurately identified and recalled by the relatives of young cases who may still be living at home with their parents and in full time education, compared to those over 20 years of age who may be living independently and working. Age related factors appear to influence susceptibility to vCJD and also incubation period, therefore it is conceivable that age related factors might also influence illness duration, although they do not appear to influence other aspects of the clinical phenotype. In general, it is recognised that neurodegenerative brain disease in youth is associated with longer illness duration. This observation may not therefore be specific to vCJD.

The primary vCJD epidemic

The primary epidemic of vCJD in the UK has been described in great detail. The present data confirm published accounts.(37;183;184;186;244) The vCJD epidemic peaked in 2000 in men, 2001 in women and has been in decline since. Significant uncertainties around the parameters of the primary epidemic remain however. The incubation period of vCJD is unknown. At the time of writing, incident cases continue to emerge in the UK, albeit in small numbers. The incubation period can be estimated to be up to and in all likelihood beyond, 22 years (from the SBO ban in 1989). Experience from Kuru suggests that incubation periods of up to and beyond 50 years are possible. All vCJD cases to date have been methionine homozygote at Codon 129. The epidemiology of prion disease in humans suggests that other genotypes will also be susceptible but that the clinical phenotype, including incubation period, may vary. Combined, these data suggest that the primary epidemic may continue, at a low level, for many years to come. Recent modelling work predicted an additional 100 (11 – 220) incident vCJD cases from the primary epidemic.(172)

The clinico-pathological phenotype in non-methionine homozygotes

The clinical phenotype of vCJD has been remarkably consistent over time. The majority of cases present with early psychiatric symptoms, many develop painful sensory symptoms over the course of their illness, cerebellar signs and movement disorders are prominent and cognitive decline is universal. The diagnostic criteria for vCJD, developed following characterisation of the first 10 cases and amended in 2002 in recognition of the value of tonsil biopsy, were only formally evaluated and validated in 2010.(186) Heath *et al* report the sensitivity, specificity and positive predictive value of the diagnostic criteria for vCJD to be 83.0% (74.5 – 89.6), 100% (92.1 – 100) and 100% (95.9 – 100) respectively.(186) The clinical phenotype of vCJD presenting in genotypes other than methionine homozygote, is unknown. In 2008 a clinical diagnosis of 'possible' vCJD was reported in a methionine heterozygote.(177) Investigations did not conclusively support a diagnosis of vCJD although the clinical phenotype in this suspect case was indistinguishable from vCJD. Post mortem examination was not undertaken following death therefore diagnostic confirmation is unavailable in this case. It should be noted that between

1996 and 2006, 5 suspect cases met the clinical criteria as a 'possible' case of vCJD that had alternate diagnoses – one Alzheimer's Disease, one Wilsons Disease, one Viral Encephalitis, one subacte sclerosing panencephalitis and one case that improved clinically although a clinical diagnosis was not reached.

Experience from other prion diseases suggest that the clinical phenotype of vCJD may vary according to *PRNP* Codon 129 genotype. In turn, diagnostic sensitivities, including the diagnostic criteria, may also vary according the *PRNP* Codon 129 genotype. It is not clear then whether vCJD cases in methionine heterozygotes or valine homozygotes would be identified as suspect CJD cases and referred to the NCJDSU. This highlights the importance of neuropathological examination in the surveillance of all forms of CJD. Of the original cases described by Will *et al* in the seminal case series, 30% were referred to the NCJDSU as a result of examination of neuropathological material alone.(37) These cases would not otherwise have been ascertained by the NCJDSU. In the UK post mortem rates in vCJD (and other prion diseases) are falling which may have a significant impact on the ability of the PHS system to detect vCJD in non-methionine homozygote genotypes. It should be considered that the pathological phenotype of vCJD in non-methionine homozygote genotypes may also differ from that in methionine homozygotes. In this context, molecular subtyping using Western Blot examination and transmission studies, linked to neuropathology will be central to surveillance activities.

The secondary vCJD epidemic

The identification of a novel human prion disease aetiologically linked to BSE in cattle was a primary aim of CJD surveillance in the UK and elsewhere. The iatrogenic transmission of vCJD through the transfusion of labile blood components has provided the imperative to continue PHS of CJD in the context of declining primary epidemics of vCJD in humans and BSE in cattle. It is likely that there was widespread exposure of the UK population to BSE. The prevalence of asymptomatic vCJD infection in the population of the UK is unknown. However there is clearly potential for transmission whilst in an asymptomatic phase of illness and non-methionine homozygote genotypes appear susceptible. The detection of PrP^{Sc} in peripheral tissues in vCJD raises the theoretical risk of transmission through surgery,

principally ophthalmological, neurological, gastrointestinal and dental surgery. Progress is being made toward the development of a blood test to detect PrP^{Sc} in humans, although there are significant scientific and ethical issues that will need to be addressed before this could be introduced as a screening tool at population level. In the interim there is a clear rationale to continue public health surveillance of all forms of CJD.

iCJD

There have been over 400 cases of accidental transmission of sCJD through medical or surgical interventions worldwide.(156) In the UK iCJD accounts for a little over 4% of all prion diseases in humans. Only France and Japan have reported a greater number of iCJD cases.(156)

CJD attributable to cadaveric-derived human pituitary hormones

iCJD in the UK is largely attributable to the administration of cadaveric-derived human pituitary hormones; the majority of cases as a result of the administration of hGH and a single case as a result of the administration of hGnH. An estimated 95% of all hGH-related iCJD cases worldwide have occurred in France, the UK and USA.(156) Approximately 30,000 children worldwide have been treated with hGH. In the UK an estimated 1 in every 100 recipients develops iCJD.(162) Cadaveric-derived hGH was withdrawn in the UK in 1985 however incubation periods are exceptionally long, known to range from 4 years to 36 years. The maximum incubation period observed in the present study was 23 years. It would be anticipated then that cadaveric-derived hGH related cases will continue to be reported in the UK in the foreseeable future.

Consistent with published reports, the majority of the UK cases presented with a progressive cerebellar syndrome at onset.(156) Genotyping was available in almost two thirds of cadaveric-derived hGH iCJD cases. Assuming those a case for which genotyping was unavailable were all methionine homozygotes, this genotype would account for fewer than 40% of all cadaveric-derived hGH iCJD cases. This figure remains lower that published accounts from the USA and France but is close to the population distribution of *PRNP* Codon 129 genotype in the UK. The NCJDSU

collate a minimum dataset on cadaveric-derived hGH iCJD cases but is not directly responsible for disease surveillance in this group. It is unlikely however that there has been systematic under-ascertainment of cadaveric-derived hGH iCJD, or differential ascertainment according to *PRNP* Codon 129 genotype. An accurate and up to date register of cadaveric-derived hGH recipients in the UK exists. This group are reviewed by Professor Michael Preece's team at the Institute of Child Health in London. Communication between this team and the NCJDSU is excellent. The clinical phenotype in cadaveric-derived hGH iCJD cases is fairly consistent, irrespective of *PRNP* Codon 129 genotype. It is unlikely that a classical clinical picture in an individual known to be at risk would not be detected and reported to the NCJDSU. The finding that the distribution of cadaveric-derived hGH iCJD cases in the UK differs from the distribution of iCJD cases outside the UK is entirely novel and warrants further investigation.

One case of iCJD attributable to cadaveric-derived hGnH was ascertained in this study. This is exceptionally rare. Worldwide just 5 cases of hGnH have been reported.(156) The remaining 4 cases were reported in Australia with the last case occurring over a decade ago. Use of cadaveric-derived hGnH ceased in the UK in 1985. The maximum reported incubation period was 16 years, although this was based on a small number of cases. It is unlikely then that further cases of cadaveric-derived hGnH iCJD will emerge in the UK.

CJD attributable to cadaveric-derived dura mater grafting

A small number of iCJD cases attributable to cadaveric-derived dura mater grafting have been observed in the UK in individuals that received Lyodura grafts between 1983 and 1987. In one case the date of grafting was not known. This individual died in 2005. The minimum incubation period in this case is 22 years. The longest recorded incubation period in iCJD via this route of exposure is 25 years.(245) The clinical phenotype in iCJD cases attributable to cadaveric-derived dura mater grafting was similar to classical sCJD which is entirely consistent with the literature in this area.

Genetic prion disease

Overall genetic prion disease accounted for 9.4% of all prion disease cases ascertained by the NCJDSU from 1990 through 2006. This figure is consistent with pooled data from the EUROCJD consortium which estimated genetic prion disease to account for 9.4% of all prion disease cases ascertained in 10 countries in Europe, Australia and Canada from 1993 through 2002 (Slovakia excluded as over 70% of CJD cases in Slovakia are attributable to genetic prion disease).(196)

The majority of genetic prion disease cases were accounted for by gCJD, 36% by Gerstmann-Straussler-Scheinker Disease (GSS) and less than 5% by Fatal Familial Insomnia (FFI). A causative mutation was identified in almost 90% of genetic prion disease cases; the remainder were classified based on reported family history. In total 16 mutations of the *PRNP* gene, 10 point mutations and 6 octapeptide repeat mutations were identified, of which two were novel point mutations that had not previously been described (Y163STOP and D167G). Worldwide over 50 mutations of the *PRNP* gene have been described although many are exceptionally rare and limited to small geographical areas, a single family and in some cases a single individual.(152)

Over half, 62% (43), of all gCJD cases were attributable to an insert mutation, the commonest of which was the 144bp insert mutation. The E200K mutation accounted for the majority (over 80%) of missense point mutations but only a quarter of gCJD cases. This is contrary to published reports from other countries in which the E200K mutation is consistently found to be the commonest mutation in gCJD.(152;196;246) As previously noted the clinical phenotype in genetic prion disease may be indistinguishable from sCJD. Therefore there may be under-ascertainment of gCJD cases in the UK because *PRNP* mutation testing is not undertaken in all cases referred to the NCJDSU. The clinical phenotype and causative mutations identified in both GSS and FFI cases were consistent with published reports.(152)

Almost 40% of all genetic prion disease cases had no known family history of prion disease. This is also in keeping with previous reports.(196) It is interesting to note

that a family history of prion disease was more frequently reported in mutations associated with a long clinical illness and least frequently reported in mutations associated with a short clinical illness and a clinical phenotype suggestive of sCJD, such as the E200K and 96bp insert mutations. There are a number of reasons that a family history of prion disease may not be reported including a lack of knowledge of family history and non-paternity. The penetrance of some mutations is incomplete, and some mutations may arise spontaneously. The large proportion of genetic prion disease cases that do not report a family history of prion disease highlights the importance of undertaking *PRNP* mutation testing in suspect prion disease cases.

Strengths and limitations

This study examined longitudinal data prospectively and systematically collected using standardised and reliable methods in the UK over a 16 year period. This is therefore one of the most comprehensive accounts of the epidemiology of prion disease according to disease subtype, produced to date. There are a number of limitations that should be considered. Minimal data were available on iCJD and genetic prion disease cases limiting analyses. Analyses of all disease subtypes included both definite and probable cases. Probable cases have met the diagnostic criteria based on clinical features and supportive diagnostic criteria. The sensitivity of the diagnostic criteria has increased over time but is not 100%. These data may include a small number of individuals that met the diagnostic criteria but did not have prion disease and exclude a small number of individuals that did not met the diagnostic criteria but did have prion disease. Nevertheless, this approach is internationally adopted and was therefore considered appropriate for this study.

Conclusions

The most significant finding over this period was the identification of a clinico-pathologically distinct human prion disease, vCJD. The primary vCJD epidemic was smaller than initially feared and has been in continued declined since 2000. There is evidence of genetic susceptibility in vCJD, with all cases to date occurring in the methionine homozygote genotype. Uncertainties exist as to the susceptibility and incubation period in other genotypes and the associated phenotypic expression of

disease in these groups. The transmission of vCJD via blood transfusion is a major public health concern providing the imperative to continue prion disease surveillance for the foreseeable future. The incidence of sCJD increased over the study period, most likely attributable to improved case ascertainment through surveillance activities linked to diagnostic advances. Declining autopsy rates, in both sCJD and vCJD, and a significant fall in the proportion of sCJD cases undergoing *PRNP* mutation testing over time, are of concern, with evidence of possible under-ascertainment of genetic prion disease cases in the UK as a result of the latter. The potential for distinct clinico-pathological forms of vCJD to emerge in individuals with non-methionine genotypes, argues for continued clinico-geno-pathological surveillance with broad referral criteria, high autopsy rates and examination of atypical cases at the molecular level.

These data support on-going systematic prospective PHS of prion disease in the UK. However they also provide evidence to suggest that the PHS system is under-performing in areas. This warrants further investigation. In the chapter that follows I report the findings from the first ever evaluation of the NCJDSU.

Chapter 3. An evaluation of the NCJDSU in the UK, 1990 – 2006

Introduction

In this chapter I report the findings of the first evaluation of the NCJDSU in the UK. The importance of periodic evaluation of PHS systems and the paucity of published studies evaluating prion disease PHS systems were outlined in Chapter 1; the need for such a study in the UK was confirmed in the preceding chapter.

Aims and objectives

The aim of this chapter was to carry out the first evaluation of the NCJDSU in the UK, applying an established framework for the evaluation of PHS systems. The overall aim of the evaluation was to provide the first in-depth examination of the operational characteristics, activities and outputs of the NCJDSU in relation to the systems objectives.

Specific objectives of the evaluation related to the attributes of the PHS system that I considered being the most important for the system to meet its objectives:

1. To assess the sensitivity of the PHS system.
2. To determine the ability of the NCJDSU to respond to changing demands over time (flexibility).
3. To examine the quality of surveillance data produced by the NCJDSU (data quality).
4. To explore the willingness of patients, relatives and health care providers to participate in surveillance (acceptability).
5. To examine the timeliness of surveillance activities and outputs.
6. To consider the relevance and value of activities and outputs from the NCJDSU (usefulness).

Methods

This evaluation applied guidelines published by the Centers for Disease Control and Prevention (CDC), Atlanta and the World Health Organization (WHO).(222) This approach first requires a detailed description of the public health importance of the condition under surveillance. An outline of the PHS system, including the aims and objectives of the system, the operational procedures, and resource, both financial and personnel, required to operate the system, follows. Finally, specific attributes of the PHS system are examined in detail including: simplicity, flexibility, data quality, acceptability, sensitivity, positive predictive value, representativeness, timeliness, stability and usefulness.

Data sources

Data collected by the NCJDSU were used to provide credible evidence against which the attributes of the system could be assessed. All suspect cases of prion disease referred to the NCJDSU between 1st May 1990 and 31st December 2006 followed for a minimum of two years until 31st December 2008 at which point data were censored. The following information was extracted from the NCJDSU's electronic minimum monitoring: sex, date of birth, date of referral to NCJDSU, date of death, case classification, disease subtype (sporadic, variant, iatrogenic, genetic), country from which referral came. Two further cohorts were examined in greater detail.

'Selected years' cohort

This cohort consisted of all suspect prion disease cases referred to the NCJDSU at three-yearly intervals (1991, 1994, 1997, 2000, 2003 and 2006). For this cohort the NCJDSU paper-based case note was examined by hand and the following information extracted: sex, date of birth, date of death, date of symptom onset, clinical presentation, case classification, disease subtype, date of referral to NCJDSU, referral source, date first sought medical attention, date first admitted to hospital (if admitted), and date first reviewed by a neurologist (if reviewed). The number, result and date of EEGs, MRI scans and CSF 14-3-3 protein examinations were collected. Details of genetic analyses, tonsil biopsy, brain biopsy and post mortem examination were recorded. Whether the NCJDSU had centrally reviewed available EEGs, MRI scans and pathological or neuropathological material was

determined. In addition whether the NCJDSU had clinically examined the suspect case, interviewed relatives of the suspect case and/or reviewed medical records from primary and secondary care, was assessed. If a risk factor questionnaire had been completed the number of missing or blank responses was assessed. The highest case classification reached by a suspect case based on clinical and neuropathological information was determined for all suspect cases; for those that met the diagnostic criteria as a possible (or greater) case at any stage, details of NCJDSU follow up and the final clinical and/or pathological diagnosis where reached, were recorded. Finally, a search of key variables from the NCJDSU electronic minimum monitoring dataset was carried out to determine the degree of missing data for the entire cohort. In a sub-group from this cohort, sCJD and vCJD cases for whom multi-source data were complete, the accuracy of key variables from the minimum monitoring dataset was checked against data available from the multiple sources. Data from this cohort were used in most of the analyses that follow. The term 'selected years' will alert the reader to the use of data from this cohort. Of note where analyses were limited to suspect vCJD cases only, the selected years examined were 1997, 2000, 2003 and 2006.

Not referred in life cohort

This cohort consisted of all sCJD and vCJD cases ascertained by the NCJDSU between 1st May 1990 and 31st December 2006 that were deceased at the time of referral to the NCJDSU. For this cohort the NCJDSU paper-based case note was examined by hand and in addition to the information described above, whether CJD had been suspected in life (if so why), the date that referral to the NCJDSU was first suggested (if known) and the highest classification reached in life based on clinical not neuropathological information, was extracted. Data from this cohort were used to examine the sensitivity of the PHS system.

Statistical analysis

All data were anonymised and entered onto three separate password protected databases maintained on a desk top computer. Data were cleaned and coded using the definitions applied in the previous chapter. In most cases multiple analyses using different metrics were carried out to evaluate each system attribute. The metrics

selected are described in the results section. There follows a general description of the statistical techniques applied to analyse these data. Where data were normally distributed this was presented as mean (standard deviation); skewed data were presented as median (inter-quartile range). Univariate parametric tests of association between key variables (t tests, Chi^2 tests); where the assumptions of these tests were violated, non-parametric equivalents were used (Fisher's exact test, Wilcoxon Ranksum test, Kruskal Wallis test). Chi^2 tests for trend (or non-parametric equivalents where appropriate) were used to compare proportions over time. Age standardised rates of referral of suspect prion disease cases according to disease subtype were calculated using denominator data from the 2001 Census data (direct method). A joinpoint regression model was fitted to estimate the APC in age adjusted referral rates overall and according to disease subtype and to detect time points at which a significant change in the overall trend occurred. To select the best-fitting model Bayesian Information Criterion (BIC) was used. A maximum of three join points were allowed for each estimate. A corresponding 95% CI was calculated for each APC estimate. To examine representativeness, age-specific rates of referral of suspect sCJD and age-specific sCJD incidence rates for (definite or probable cases) in men and women in Scotland, England, Wales and Northern Ireland were calculated using mid-year population estimates for each year. Indirect standardization was used to calculate a standardised referral ratio and standardised incidence ratio for each country, relative to England.

All analyses were carried out using STATA Version 10 (Stata Corp. College Station, Texas, USA). Regression analyses were carried out using Joinpoint Regression Program (Version 3.4.3). A level of statistical significance of 0.05 was used throughout. Note for analyses involving dates missing data were treated in the same way as outlined in the previous chapter.

Results

Description of the surveillance system

The rationale for prion disease PHS was outlined in chapter 1 and the epidemiology of prion disease in the UK according to disease subtype, from 1990 through 2006, described in chapter 2. I shall therefore move directly to a description of the PHS system in the UK prior to an examination of specific attributes of the PHS system.

Systematic prospective surveillance of CJD was initiated in the UK in May 1990 in response to publication of the Report of the Working Party on Bovine Spongiform Encephalopathy (Southwood Committee). The initial aim of the NCJDSU was to identify a change in the clinic-pathological phenotype of CJD that could be attributable to BSE; this was realized in 1996. In 2006, the objectives of the NCJDSU were to

> *"monitor characteristics of CJD, specifically sCJD and vCJD, to identify trends in incidence rates and to study risk factors for the development of disease."(247)*

Population under surveillance

The NCJDSU collect and report data on *all* suspect prion disease cases referred to the NCJDSU that are resident in the UK at the time of symptom onset. The UK includes four countries, Scotland, England, Northern Ireland and Wales covering an area of approximately 242,514 square kilometres. England is the largest country within the UK; approximately 83.8% of the population of the UK live in England, 8.4% in Scotland, 4.9% in Wales and 2.9% in Northern Ireland. In 1990 the population in the UK was 57.2 million. This increased by 0.3% per annum to 60.6 million in 2006. Until mid-1999 this was driven by an increase in births and reduction in deaths; post 1999 by immigration. Almost half of the population, 49%, are men. In common with most industrialised countries the population is ageing. Overall life expectancy increased from 73.4 years to 77.3 years in men, 78.9 years to 81.5 years in women, from 1990 through 2006. In the UK primary and secondary health care is provided to all citizens, free at point of access, by the National Health Service (NHS). Funding for public expenditure on health is provided by the UK Treasury (revenue largely generated through taxation).

Case definition

Suspect prion disease cases are classified according to internationally agreed criteria (Appendix 2).(98) Over the period covered by this study these diagnostic criteria were revised to reflect emergent disease (vCJD) and diagnostic advancement (CSF 14-3-3 protein, *PRNP* mutation testing and tonsil biopsy). Additional criteria are used by the NCJDSU to further characterise suspect prion disease cases that do not meet the diagnostic criteria, outlined in full below:

0.0 Unclassified: There is insufficient clinico-pathological information to classify the suspect prion disease case.

1.0 Definite case as defined in the diagnostic criteria.

2.0 Probable case as defined in the diagnostic criteria.

3.0 Possible case as defined in the diagnostic criteria.

4.1 Diagnosis unclear: Suspect prion disease cases that do not meet the diagnostic criteria as a definite, probable or possible case but for whom an alternate diagnosis has not emerged. Prion disease remains a differential diagnosis.

4.2 Prion disease unlikely: Prion disease is considered unlikely because of clinical features and/or results from investigations which do not support a diagnosis of prion disease. This group includes individuals in whom an alternate clinical diagnosis has been reached and those that have recovered clinically without an alternate diagnosis being reached.

4.3 Definitely not CJD: an alternate neuropathological diagnosis is available.

As a minimum suspect prion disease cases are assigned a case classification:

- at the time of referral to the NCJDSU.
- following visit by a NCJDSU neurologist.
- when the NCJDSU review is complete (when it becomes apparent that no further information regarding the suspect case will be forthcoming).
- on completion of the NCJDSU review a highest classification in life, based on clinical *not* neuropathological information is assigned

Case classification may be revised at any stage following initial classification if relevant information regarding the suspect case emerges.

Legal authority for collection of data

Prion diseases are not, nor have ever been, notifiable in the UK. There is no legal requirement for patients, their relatives or health care professionals to participate in disease surveillance.

Interface with other organisations

The NCJDSU is based in the Western General Hospital in Edinburgh, Scotland and affiliated with the University of Edinburgh. The NCJDSU is a WHO Collaborative Centre for Reference and Research on the Surveillance and Epidemiology of Human Transmissible Spongiform Encephalopathies (Figure 47). The NCJDSU collects and collates European CJD surveillance data for the European Centre for Disease Control (ECDC), co-ordinating the EUROCJD network. The NCJDSU is also directly involved in a number of other collaborative international networks including NEUROCJD and NEUROPRION. Of note NEUROCJD no longer exists but was in operation over the study period. Within the UK, the NCJDSU collaborates with the Institute of Child Health and the NPC (both located in London) in the surveillance of iCJD and genetic prion diseases respectively. Other organisations such as the General Registers Office (GRO) for England and Wales (and equivalent bodies in Scotland and Northern Ireland), the British Paediatric Surveillance Unit and the UKBTS work directly with the NCJDSU to provide information essential for disease surveillance. Reporting pathways will be outlined in the sections that follow. NCJDSU staff are directly involved in a number of Committees that inform public health policy both nationally and internationally, for example SEAC in the UK and internationally the European Medicines Agency.

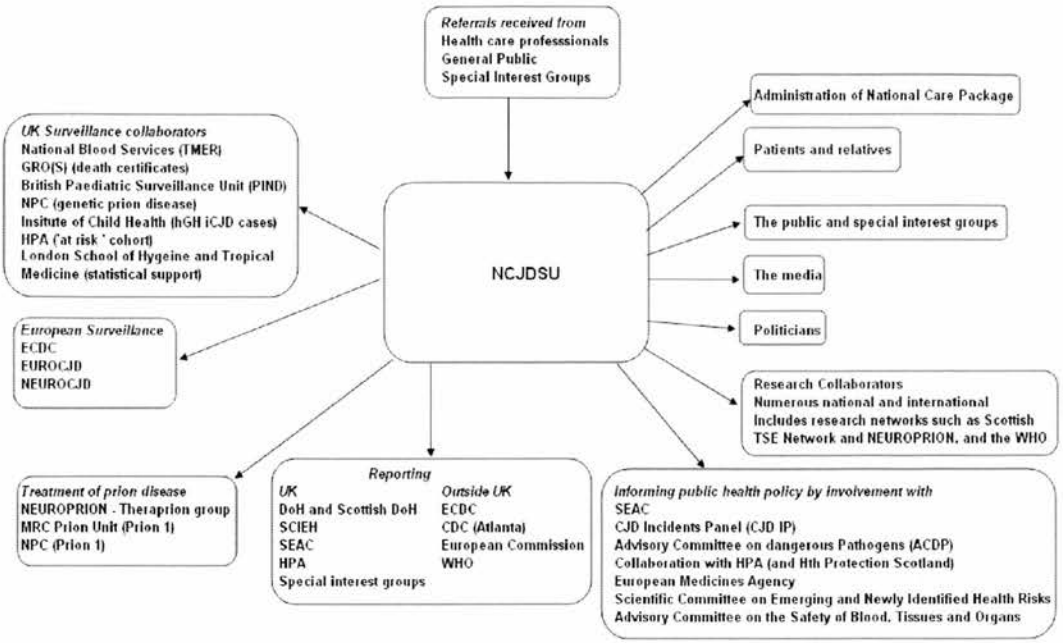

Figure 47 Diagram illustrating the organisations that the NCJDSU interfaces with in the UK and internationally

Data sources

Multiple and overlapping sources of data are used to maximise ascertainment of suspect cases. Suspect prion disease cases are ascertained through passive surveillance by direct notification from health care professionals, family members, the general public or specialist interest groups, and the review of death certificates coded under the specific rubric of CJD which are forwarded to the NCJDSU by the GRO for England and Wales (and equivalent bodies in Scotland and Northern Ireland). Certain health care professionals, including neurologists, neuropathologists and neurophysiologists receive a bi-annual reminder to refer any suspect prion disease case to the NCJDSU. Additional cases may be ascertained via the National CSF 14-3-3 protein service, neuropathology and molecular genetics laboratories, which are based in the NCJDSU. Referrals to these services are unsolicited and can be made without formal referral to the NCJDSU. For example, in 2006, the CSF 14-3-3 protein service processed 245 samples from patients in the UK of whom just 58 (23.7%) were formally referred to the NCJDSU as a suspect prion disease case. The annual numbers of referrals to the neuropathology and molecular genetics laboratories more closely mirror formal referrals to the NCJDSU. Enhanced active

surveillance has been carried out by the NCJDSU in a number of on-going and time limited, prospective and retrospective cohort studies, carried out in association with partners. Several of these studies were described in detail in chapter 1, including the PIND and the TMER studies.

Information collected

On notification of a suspect prion disease case, a designated neurologist from the NCJDSU contacts the referring source by telephone to obtain relevant case-related information including sociodemographic, clinical and diagnostic information, and details of any known risk factors for iatrogenic disease. A unique identifier, the NCJDSU number, is assigned to the suspect case and a paper-based case note generated. Case classification, as previous described, is assigned to the suspect case. A minimum dataset on all iCJD and genetic prion disease cases are held at the NCJDSU for surveillance purposes. These cases are not routinely followed up by the NCJDSU beyond this point. For suspect sCJD and vCJD cases verbal consent for the NCJDSU to contact the suspect cases' relative(s) to arrange a visit is obtained from the referrer at the point of referral where appropriate. A NCJDSU neurologist visits the patient and/or relatives, typically in hospital, hospice or at home. Where possible the neurologist is accompanied by a research nurse. During this visit relative(s) provide written informed consent for the NCJDSU to access the suspect cases' medical records. The NCJDSU neurologist confirms the clinical history with the relative(s). A detailed neurological examination of the suspect case is carried out. The medical case notes and all available investigations (for example EEGs and MRI scans) are reviewed by the NCJDSU neurologist. A proforma, the Patient Review and Examination form, is completed in writing by the NCJDSU neurologist at the time of this visit. The research nurse (or NCJDSU neurologist) completes a structured risk factor questionnaire in writing, which includes residential, occupational, dietary and medical histories with relative(s). During the visit the NCJDSU neurologist will request that the referrer inform the local CCDC using a standard reporting form, if the suspect case meets the diagnostic criteria as probable or definite case of prion disease. Following the visit, the NCJDSU neurologist requests copies of EEGs and MRI scans for central review at the NCJDSU. The former are reviewed by one of two designated neurologists (RK, RGW), the latter by

one of two designated neuroradiologists (DS, DC). Subsequent to the visit a letter is written to the referrer briefly outlining the clinical impression of the NCJDSU neurologist and stating the case classification based on the information available at that time. The responsibility for on-going case management remains with the referring clinical team, not the NCJDSU, although the NCJDSU will often advise on further investigations and clinical management, including infection control issues. Since 2000 a National Care Package has been available for CJD patients and their families. The aim of this is to ensure that the care and social needs of CJD patients and their families are adequately met. This National Care Package is administered by a National Care Co-ordinator based at the NCJDSU. Where the patient's family have granted permission, the NCJDSU neurologist will inform the National Care Co-ordinator of the case.

Where the patient is deceased at the time of notification or dies prior to a NCJDSU visit, the NCJDSU attempt to obtain as much clinical and diagnostic information as possible. Suspect cases that were not referred to the NCJDSU in life are typically referred by neuropathologists or ascertained by death certificate review. In the first instance the clinician responsible for the suspect cases' care is contacted and clinical and neuropathological information regarding suspect case, if available, is requested. For suspect cases ascertained through death certificate review in which there was no clinical suspicion of prion disease in life and no documentation supporting a diagnosis of prion disease, the suspect case will be classified accordingly and the NCJDSU record will be closed. Suspect cases that meet the diagnostic criteria as a definite, probable or possible case of sCJD or vCJD, based on the information available, are followed up. Permission is sought from the clinician that was responsible for the patients care to contact the cases relative(s). If consent is provided, a NCJDSU neurologist and research nurse visit the relative(s) to collect information regarding the suspect cases' clinical illness and complete the risk factor questionnaire. Where information is not available from the clinician responsible for the suspect cases' care, despite repeated attempts to contact the clinician and/or local health authority, the record will be closed.

When a suspect case dies, medical records from primary and secondary care are requested for review. These are reviewed by a NCJDSU neurologist and relevant information extracted and entered onto a proforma (The Final Review Form). Data from primary care are triangulated with data collected at the time of the NCJDSU visit and hospital medical records to obtain a complete clinical history.

The death certificates of all suspect cases referred to the NCJDSU are requested from the GRO (or equivalent bodies) so that the date and the underlying cause of death, as recorded on the death certificate, can be ascertained. Where post mortem examination has been carried out, the NCJDSU will endeavour to review any available neuropathological material. If this is unavailable, a copy of the post mortem report is requested.

NCJDSU case notes are periodically reviewed. The NCJDSU neurologist may contact the referrer to request an update on the suspect cases condition following the visit. Often an update on the clinical condition of a suspect case is received directly from the cases relatives; this may be through on-going contact with the National Care Co-ordinator. When no further information regarding the case is likely to become available the review is closed.

Data storage and issues of privacy

The NCJDSU retain a paper-based case note for each formal referral. This contains a hard copy of all proformas completed by NCJDSU staff, photocopies of suspect cases medical records where these have been accessed, EEG tracings and MRI images (where these have been provided), and all correspondence relating to the case. Case notes are held in secure fire-proof filing cabinets in a locked room. A minimal dataset on each suspect case referred to the NCJDSU is entered onto a minimum monitoring database maintained using Visual FoxPro by the Study Co-ordinator.(98) Any errors in data entry are corrected on an informal basis when new information becomes available or a change in cases classification occurs. Data from

the risk factor questionnaire are double entered onto a separate database also maintained using Visual FoxPro. A paper-based record of all miscellaneous contacts with the NCJDSU (contacts with the NCJDSU that have not resulted in a formal referral being made) is maintained for reference. Historically these were recorded on an adhoc basis by a NCJDSU neurologist, and when passed to the Study Co-ordinator, stored in lever arch files. Latterly an effort has been made to systematically record such contacts on an Excel spread sheet. Designated personnel are responsible for maintenance of electronic data held by the NCJDSU, including arrangements from back up. All data are held in accordance with the 1998 Data Protection Act (UK). Multi-centre research ethics committee (M-REC) approval was granted for data collected in relation to the case control study subsequent to passage of the 1998 Data Protection Act.

Data analysis and reporting

Routinely published surveillance data are analysed by the Study Co-ordinator. A designated statistician is employed by the NCJDSU and external statistical support is provided by the London School of Hygiene and Tropical Medicine. Pathways for data reporting have been established. Standardised reports are produced and disseminated according to agreed protocols. Each month the Study Co-ordinator updates the NCJDSU website with the number of suspect prion disease cases referred to the NCJDSU and the annual number of deaths from definite or probable prion disease according to aetiological subtype. Monthly figures are emailed by the NCJDSU to the Department of Health (DH) and Scottish Centre for Infection and Environmental Health (SCIEH). Each month the DH issues a press release and updates their website with these data; SCIEH publish these figures in their weekly report. A quarterly report summarising the number of definite or probable vCJD cases to date according to vital status is published on the NCJDSU website. Tables of definite or probable cases of vCJD by residence are sent quarterly by the NCJDSU to each Regional Epidemiologist and UK Health Department to be cascaded to relevant Districts. The NCJDSU provide data on the annual number of deaths and mortality rate from sCJD, iCJD and genetic prion disease (definite or probable cases) and the annual number of definite or probable vCJD cases according to year, to the EUROCJD group for publication on the EUROCJD website.

For each incident definite or probable vCJD case a number of agencies are informed. The DH is informed by email of the gender, age, case classification and vital status. In turn, the DH informs local government departments and the SEAC secretariat. The HPA are notified of the cases' NCJDSU number, gender, date of onset, date of referral to the NCJDSU, date of birth, date of death and date that the case was first classified as a definite or probable vCJD case. Finally, colleagues in the European Union CJD Surveillance System, WHO Headquarters, CDC Atlanta, European Commission, Alzheimer's Disease Society, Human BSE Foundation, BSE Enquiry and other interested parties are sent the gender, age and case classification of the incident case in addition to tables of the total number of definite or probable vCJD cases according to vital status.

As part of the TMER study, the Medical Director of the relevant UKBTS (determined by residential history) is notified immediately of an incident vCJD case (definite or probable). The Medical Director is provided with the case's name (including maiden name), gender, date of birth, residential history, donation history (dates and places of donation), case classification and country of residence at time of referral to NCJDSU. In addition an anonymised copy, stripped of patient identifiable data, is sent to the appropriate DH. The UKBTS are informed bi-annually of sCJD and genetic prion disease cases (definite or probable) that were identified as blood donors or were reported as being the recipient of labile blood components. The case's name (including maiden name) and gender are provided. For blood donors, the year of donation(s), home address at the time of donation(s) and location at which the donation(s) were made are supplied. For the recipients of blood product(s), the year of the transfusion(s), home address at the time of transfusion(s), hospital where the transfusion(s) occurred and the indication for the transfusion(s) are supplied.

Annually the NCJDSU produce a report which summarises the clinico-pathological epidemiology of human prion disease in the UK in the preceding year and outlines surveillance activities. This report is published on the NCJDSU website. Surveillance data are also disseminated to the scientific community through publications in peer-

reviewed journals and presentations at scientific meetings, nationally and internationally.

Resources required to operate the surveillance system annually

At inception the NCJDSU had an annual operational budget of £79,905 and employed one neurologist and one Study Co-ordinator. By 2006, the NCJDSU employed 34 staff (of whom 15 were primarily involved in clinical disease surveillance) and had an annual operational budget of £1.8 million (Table 28). Core funding is provided by DH (90%) and the Scottish Government Department of Health (10%). Additional funding is provided through research grants won in open competition; many of the staff employed by the NCJDSU are funded through such grants, for example the European Study Co-ordinator.

Table 28Annual resources available to operate the NCJDSU in 1990 and 2006

1990	2006
1 Consultant Neurologist	*Clinical disease surveillance*
1 Study Co-ordinator	2 Consultant Neurologists
	1 Senior Clinical Scientist (CSF)
	1 Senior Biomedical Scientist (CSF)
	1 Molecular Geneticist
	1 Laboratory technician (Genetics)
	2 Clinical Research Fellows
	2 Nurse Practitioners
	1 Study Co-ordinator
	1 European Study Co-ordinator
	1 Database Manager
	2 Administrative staff
	Other activities
	1 Consultant Epidemiologist
	2 Consultant Neuropathologists
	1 Chief Biomedical Scientist (Histopathology)
	3 Senior Biomedical Scientist (Histopathology)
	1 Senior Research Fellow (Biochemistry)
	3 Research Fellows (Biochemistry)
	1 Laboratory technician (Biochemistry)
	1 Research Assistant (Biochemistry)
	1 Statistician (Epidemiology)
	1 Business Manager
	4 Administrative staff
Annual Budget: £79,905	**Annual Budget: £ 1,811,696**

Performance of the surveillance system

The CDC criteria highlight eight key areas in evaluation of a surveillance system. Each will be addressed in turn. The metrics selected to examine one system attribute may also be applicable to another. Where this is the case, rather than presenting duplicate data, the reader has been signposted as appropriate.

Simplicity

> *"The simplicity of a surveillance system refers to both its structure and ease of operation. Surveillance systems should be as simple as possible while still meeting their objectives."*(222)

From the point of referral to the NCJDSU, detailed clinical, diagnostic and epidemiological data are collected from multiple data sources (Figure 48). The assessment and interpretation of clinical and diagnostic information is required for case classification; epidemiological data, including detailed sociodemographic, family, medical, residential, occupational, travel, behavioural, lifestyle and dietary histories data are collected to investigate putative risk factors for prion disease and explore possible routes of secondary transmission. Much of these data are collected when the NCJDSU neurologist visits the suspect case. This visit typically lasts for 3 hours, although it can take much longer. Owing to the distance travelled to meet each suspect case in person it is exceptionally rare for more than one visit to take place in a day, indeed each visit, including travel time, can take upward of 12 hours. Subsequent to the visit, time is spent entering electronic information on each suspect case onto the minimum reporting dataset and the case control study database (risk factor questionnaire). Data collected at interview are validated through the examination of medical records from primary and secondary care. Available diagnostic information including EEGs, MRI scans and neuropathological material are requested for review by designated staff at the NCJDSU. The UKBTS are informed immediately of all definite or probable vCJD cases (bi-annually for sCJD and genetic prion disease cases) to begin a process of tracing blood donations and recipients. Individuals meeting the diagnostic criteria as a definite or probable case of prion disease are able to access the National Care Package. Often the National Care Co-ordinator will remain in direct contact with the case's relatives, facilitating follow

up. The follow up of suspect cases that do not meet the diagnostic criteria is more challenging. Typically this would occur through NCJDSU staff contacting the referrer at intervals to request an update on the patient's progress. In addition the death certificates of all suspect cases referred to the NCJDSU are routinely requested and reviewed.

Whilst the NCJDSU provide data to a large number of agencies this process has been streamlined through the use of standard reporting formats, electronic data transfer and web publishing. A close working relationship with DH (and Scottish equivalent) and HPA ensures rapid communication where issues of national public health importance arise.

Interpretation: The surveillance system, whilst simple in design is operationally complex.

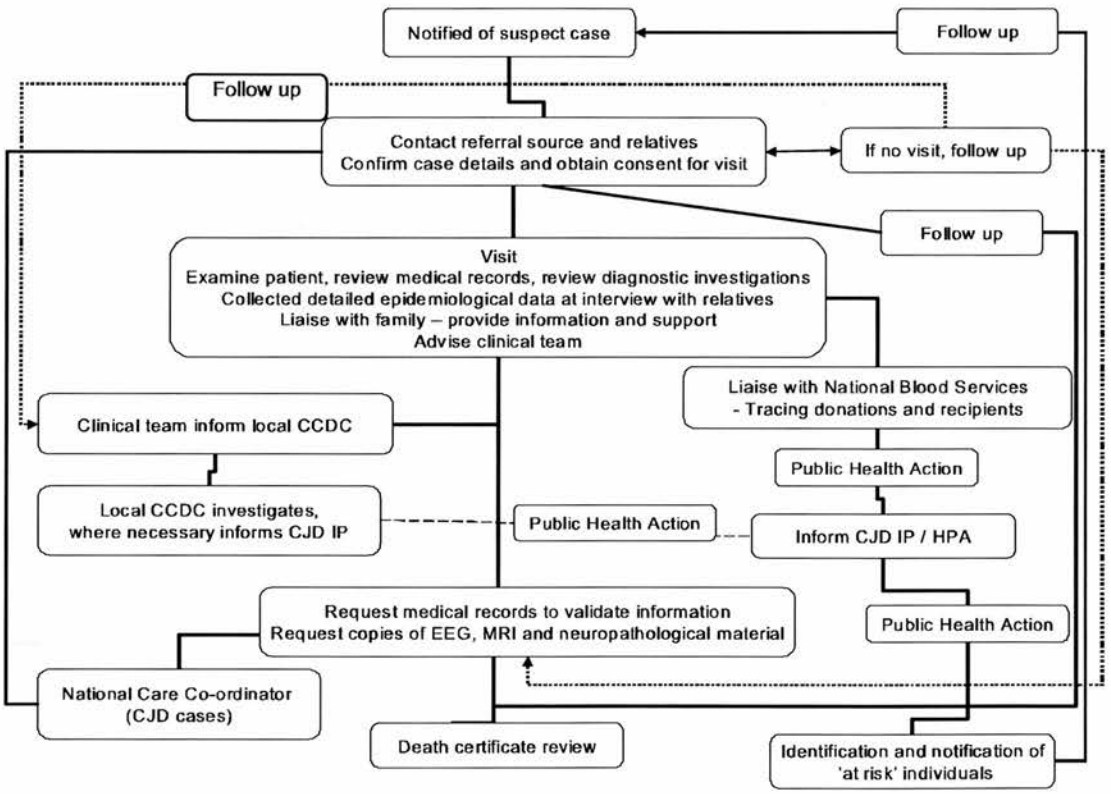

Figure 48 The surveillance pathway in the UK

Flexibility

> *"A flexible public health surveillance system can adapt to changing information needs or operating conditions with little additional time, personnel, or allocated funds. Flexible systems can accommodate, for example, new health-related events, changes in case definitions or technology, and variations in funding or reporting sources."*(222)

Over the study period the flexibility of the surveillance system has been challenged by emergent disease (vCJD), the introduction of novel diagnostic technologies (MRI, CSF 14-3-3 protein, *PRNP* Codon 129 genotyping and molecular subtyping) and changing case definitions (the incorporation of CSF 14-3-3 protein into diagnostic criteria for sCJD). The flexibility of the surveillance system can be evaluated by exploring how the system responded to these new demands.

The impact of vCJD on referral patterns and NCJDSU surveillance activities

The impact of vCJD on the operational performance and public health function of the surveillance system over this period can be explored by examining patterns of referral of suspect prion disease cases to the NCJDSU. The annual number of referrals received by the NCJDSU from 1990 through 2006 is shown in Figure 49. Between 1990 and 2006 the NCJDSU received a total of 2,154 referrals, of which 1,653 (76.7%) were suspect sCJD cases, 322 (15.0%) suspect vCJD cases, 121 (5.6%) suspect genetic prion disease cases and 58 (2.7%) suspect iCJD cases. The annual number of referrals increased from 53 per year in 1990 to peak at 179 per year in 2001 before falling to 109 per year in 2006. Over time there was a significant change in the distribution of referrals received by the NCJDSU according to disease subtype ($P<0.001$) (Figure 50). The annual number of suspect sCJD referrals received by the NCJDSU increased from 50 in 1990 to peak at 133 in 2001 before falling to 84 in 2006; for vCJD, from one in 1994 to a peak of 51 in 2000, before falling to 10 in 2006. The number of suspect iCJD and genetic prion disease cases referred was low, between zero and seven, and between zero and 13 referrals per

annum respectively, with year to year variation, but no obvious temporal trend.

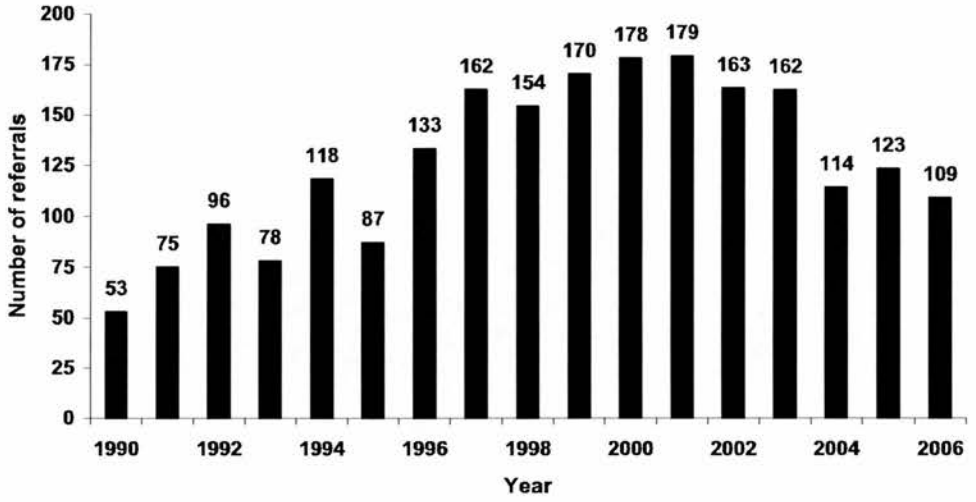

Figure 49 Annual number of referrals received by the NCJDSU, 1990 – 2006

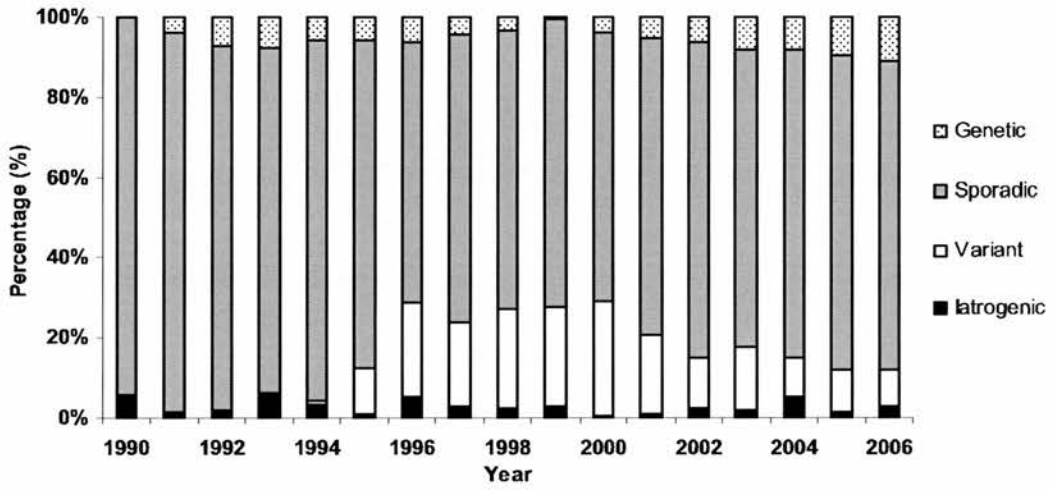

Figure 50 Distribution of referral received by the NCJDSU according to disease subtype, 1990 - 2006

Age standardised referral rates were examined to determine whether the observed increase in the number of referrals received by the NCJDSU was simply as a result of an increase in the size of the population under surveillance and population ageing over time. The age standardised referral rate increased from 0.94 (0.69 – 1.19) per million in 1990 to peak at 3.03 (2.58 – 3.47) per million in 2001, before falling to

1.78 (1.44 – 2.11) per million in 2006 (Figure 51). The annual percentage change (APC) in referral rate increased by 10.88% (7.33 – 14.55) from 1990 through 2000, then decreased by -10.05% (-15.37 - -4.38) from 2000 through 2006.

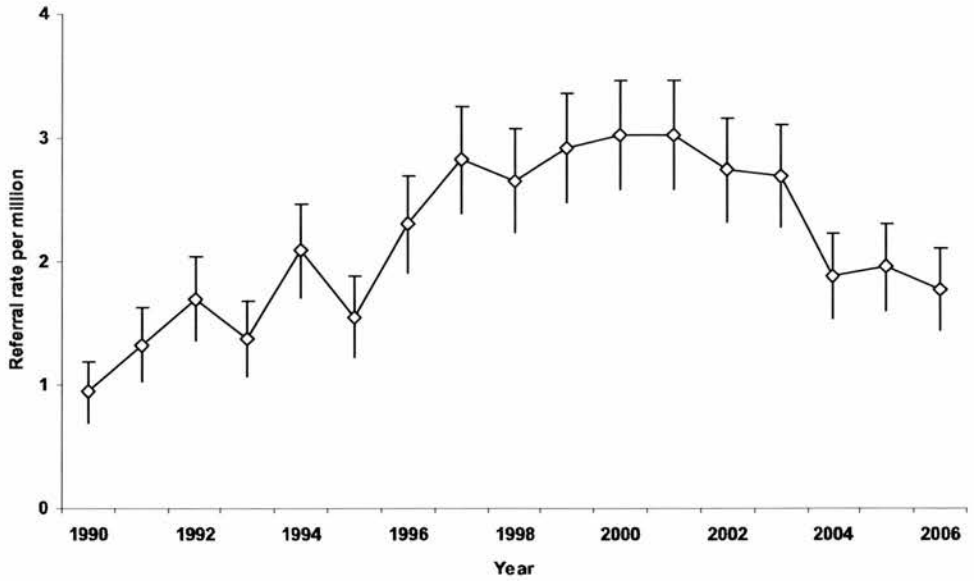

Figure 51 Age standardised rates of referral to the NCJDSU, 1990 – 2006

Age standardised referral rates according to disease subtype are shown in Figure 52. The referral rate for suspect sCJD increased from 0.89 (0.65 – 1.14) per million in 1990 to peak at 2.25 (1.87 – 2.63) per million in 2001 before falling to 1.37 (1.08 – 1.66) per million in 2006. From 1990 through 2001 there was a statistically significant increase in the referral rate for suspect sCJD, with an APC of 6.55% (3.33 – 9.87). A significant reduction in APC of -10.24% (-18.19 – -1.52) from 2001 through 2006 followed. The referral rate for suspect vCJD, increased from 0.02 (0.00 – 0.07) per million in 1994, to peak at 1.08 (0.78 – 1.38) per million in 2000, before falling to 0.20 (0.08 – 0.34) per million in 2006. Regression modelling fitted three joinpoints when modelling referral rates for vCJD over time. First, a non-significant increase in APC of 266.31% (-74.91 – 5248.67) from 1994 through 1996. Then, a non-significant increase in the APC of 10.09% (-17.98 – 47.76) from 1996 through 2000. Finally, a statistically significant decline in APC of -25.22% (-35.50 – -13.30) from 2000 through 2006. The overall age standardised referral rate for suspect iCJD was 0.06 (0.00 – 0.12) with no significant change over time (APC for 1990 through

2006: 0.83% (-4.89 – 6.89)). There was a gradual increase in the referral rate for suspect genetic prion disease over the study period from 0 per million in 1990 to 0.38 (0.16 – 0.59) per million in 2006, equating to an APC of 4.62% (1.17 – 8.19).

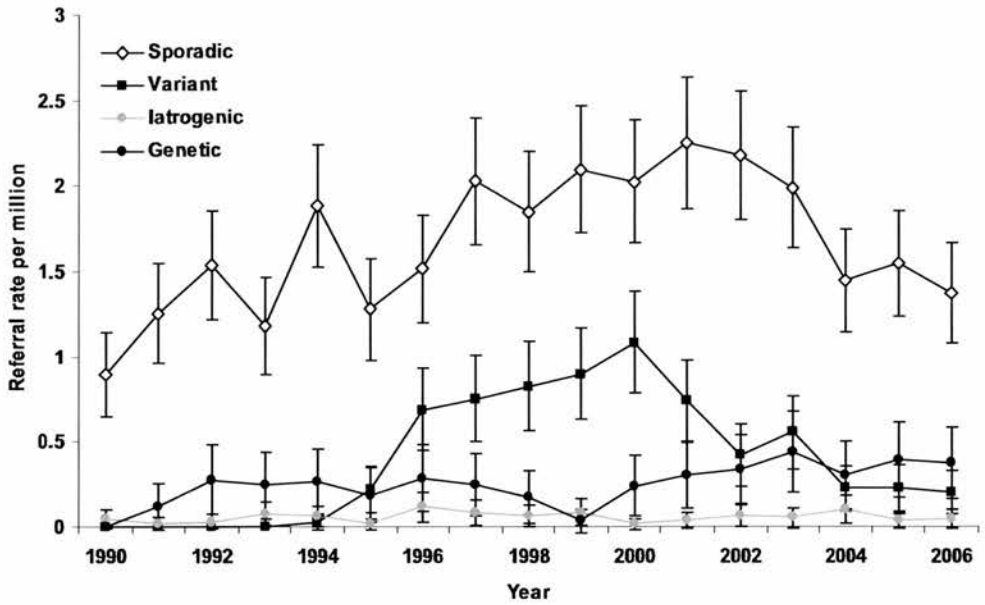

Figure 52 Age standardised referral rates according to disease subtype, 1990 - 2006

The response of the NCJDSU to an increase in the annual absolute number and rate of referral was assessed by examining the proportion of referrals that were visited by a NCJDSU neurologist across selected study years and the time from referral to neurologist visit. Over these years 801 referrals were received by the NCJDSU; 613 suspect sCJD cases, 122 suspect vCJD cases, 18 suspect iCJD cases and 48 suspect genetic prion disease cases. This section will focus on suspect sCJD and suspect vCJD cases only as the NCJDSU surveillance remit in relation to iCJD and genetic prion disease is limited.

A neurologist from the NCJDSU visited 379 (61.8%) suspect sCJD cases referred to the NCJDSU over the selected time period; 85.8% of all definite or probable sCJD cases, 52.8% of all possible sCJD cases and 29.7% of all non-sCJD cases (Table 29). In 1997 fewer than expected definite or probable sCJD cases were visited by a

neurologist from the NCJDSU (P<0.001), and in 1997 and 2000 fewer than expected non-sCJD cases were visited by a neurologist from the NCJDSU (P<0.001).

Table 29 Number of suspect sCJD cases visited by a NCJDSU neurologist each year according to case classification

Year of referral	Number (%) of suspect sCJD referrals visited by NCJDSU neurologist			
	All suspect sCJD cases	sCJD cases	Possible sCJD	Not sCJD
1991	48 (67.6)	29 (85.3)	1 (25.0)	18 (54.5)
1994	67 (63.2)	51 (92.7)	5 (45.5)	11 (28.2)
1997	59 (51.8)	48 (76.2)	4 (42.9)	7 (16.3)
2000	58 (48.7)	41 (83.7)	4 (50.0)	13 (21.7)
2003	82 (68.3)	71 (93.4)	1 (50.0)	10 (25.0)
2006	65 (78.3)	51 (82.3)	4 (100.0)	10 (58.8)
Total	**379 (61.8)**	**291 (85.8)**	**19 (52.8)**	**69 (29.7)**

CJD cases include classification 1.0 or 2.0; non-cases include classification 4.1, 4.2 and 4.3

The median number of working days from referral of a suspect sCJD case to visit by a NCJDSU neurologist according to year of referral and vital status at the time of visit is shown in Table 30. These data were not reliably recorded in 1991 – the date of neurologist visit was frequently recorded as the date of referral – hence this year has been excluded from analyses. Overall the median number of working days from referral to visit by a NCJDSU neurologist was 5 (3 – 9) days for suspect sCJD cases alive at the time of visit and 210 (110 – 286) days for suspect sCJD cases that were deceased at the time of visit. There was a statistically significant yearly variation in the median number of working days from referral to visit by a NCJDSU neurologist for both those alive (P<0.001) and deceased (P=0.012) at the time of visit, although the 95% confidence intervals for all years overlapped. The time from referral to visit was significantly longer when the suspect case was deceased at the time of visit compared to when the suspect case was alive at the time of visit. This is likely to reflecting the time taken for the NCJDSU to collect detailed clinico-pathological information on the suspect sCJD cases prior to visit to ensure that such individuals met the diagnostic criteria. In addition, given there are no public health implications of a diagnosis of sCJD it is often considered appropriate to delay approaching grieving relatives to obtain consent for a visit.

Table 30 Time from referral to visit by a NCJDSU neurologist, according to year of referral and vital status at time of visit

Year of referral	Median number of working days from referral of suspect sCJD case to visit (IQR)			
	Number	Alive at time of visit	Number	Deceased at time of visit
1994	49	4 (3 – 7)	18	239 (135 – 286)
1997	42	6 (3 - 11)	17	214 (141 – 286)
2000	43	7 (5 - 16)	15	142 (58 – 210)
2003	67	5 (3 - 9)	15	264 (213 – 270)
2006	57	4 (3 - 5)	8	60 (8 – 131)
All	**257**	**5 (3 - 9)**	**88**	**210 (110 – 286)**

A neurologist from the NCJDSU visited 77 (63.1%) of all suspect vCJD cases referred to the NCJDSU over selected years; 98.4% of all definite or probable vCJD case, all possible vCJD cases and 21.8% of non-vCJD (Table 31). There was no significant change over time in the proportion of suspect vCJD cases referred to the NCJDSU that were visited by a NCJDSU neurologist (P=0.568). The median number of working days from referral to the NCJDSU to visit by a NCJDSU neurologist was 7 (4 – 13) days in suspect vCJD cases alive at the time of NCJDSU visit; this was invariant over time (P=0.694). Just three suspect vCJD cases, all definite vCJD cases, were deceased at the time of visit. For these cases the number of working days from referral to visit was 4, 18 and 139 days, respectively. In the latter two cases referral came from a neuropathologist and vCJD had not been suspected in life therefore there was a delay in the neuropathologist informing the clinical team and in turn the clinical team informing the relatives of the diagnosis before the NCJDSU could attempt to obtain consent for a visit from the relatives.

Table 31 Number of suspect vCJD referrals visited by a NCJDSU neurologist each year according to case classification

Year of referral	Number (%) of suspect vCJD referrals visited by NCJDSU neurologist			
	All suspect vCJD cases	vCJD cases	Possible vCJD	Not vCJD
1994	**0 (0)**	0	0	0 (0)
1997	**22 (64.7)**	13 (100)	0	9 (42.9)
2000	**32 (62.8)**	28 (96.6)	1 (100)	3 (14.3)
2003	**17 (65.4)**	16 (100)	0	1 (10.0)
2006	**6 (60.0)**	5 (100)	0	1 (20.0)
Total	**77 (63.1)**	**62 (98.4)**	**1 (100)**	**14 (24.1)**

CJD cases include classification 1.0 or 2.0; non-cases include classification 4.1, 4.2 and 4.3

New diagnostic technologies

A flexible system would demonstrate increasing use of emergent technologies to support a diagnosis of prion disease in the suspect prion disease cases referred to the NCJDSU. To explore this, the use of investigations to support a diagnosis of sCJD or vCJD in suspect sCJD or vCJD cases referred to the NCJDSU over selected years was examined.

Over time a statistically significant increase in the proportion of suspect sCJD cases referred to the NCJDSU that underwent at least one EEG examination (P=0.015) during the course of their clinical illness was observed (Figure 53). Similar trends were seen in the use of MRI scanning (P<0.001) and CSF 14-3-3 protein examination (P<0.001). In 1991 for example 83.1% of suspect sCJD cases referred to the NCJDSU underwent one or more EEG examination, increasing to 91.6% in 2006. Corresponding figures for MRI scanning were 7% in 1991 increasing to 85.5% in 2006 and for CSF 14-3-3 protein examination 34.9% in 1997 increasing to 88.7% in 2006. Less than half of all suspect sCJD cases underwent *PRNP* Codon 129 genotyping or full sequencing of *PRNP* to test for mutations. There was no significant change in the proportion of suspect sCJD cases undergoing either investigation over time (P=0.302 and P=0.140 respectively). There was a significant fall in the proportion of suspect sCJD cases undergoing post mortem examination on expiration, from 62.7% in 1991 to 49.3% in 2006 (P=0.004). However among

suspect sCJD cases for whom neuropathological material was available, either from brain biopsy or post mortem, the use of molecular subtyping did increase (P<0.001).

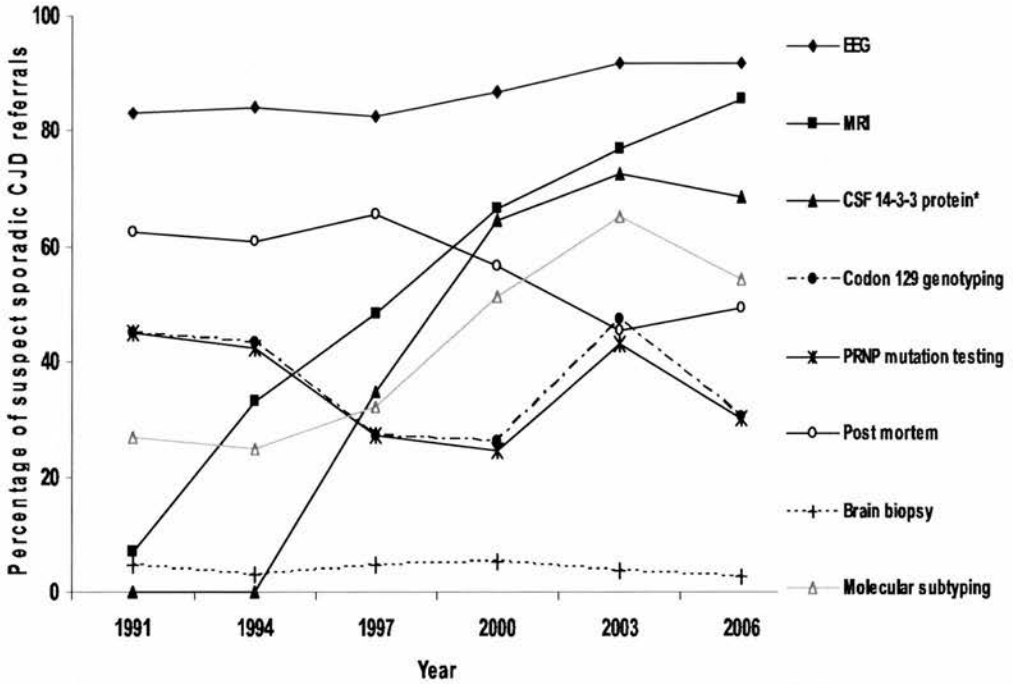

Figure 53 Proportion of suspect sCJD cases referred to the NCJDSU over selected years that underwent investigations that might support a diagnosis of sCJD

There was no significant change over time in the proportion of suspect vCJD cases undergoing MRI scanning (P=0.747) or CSF 14-3-3 protein examination (P=0.309) during the course of their clinical illness (Figure 54). However, over 90% of suspect vCJD cases underwent one or more MRI scan during the course of their clinical illness, and it should be noted that CSF 14-3-3 protein is of limited value in the investigation of suspect vCJD. Approximately half of all suspect vCJD cases referred to the NCJDSU underwent *PRNP* Codon 129 genotyping and full sequencing for *PRNP* mutations. This was invariant over time (P=0.802 and P=0.693 respectively). There was a significant fall in the proportion of suspect vCJD cases undergoing post mortem examination on expiration, from 81.8% in 1997 to 50.0% in 2006 (P=0.012). This was mirrored by a non-significant rise in the use of tonsil biopsy (P=0.071).

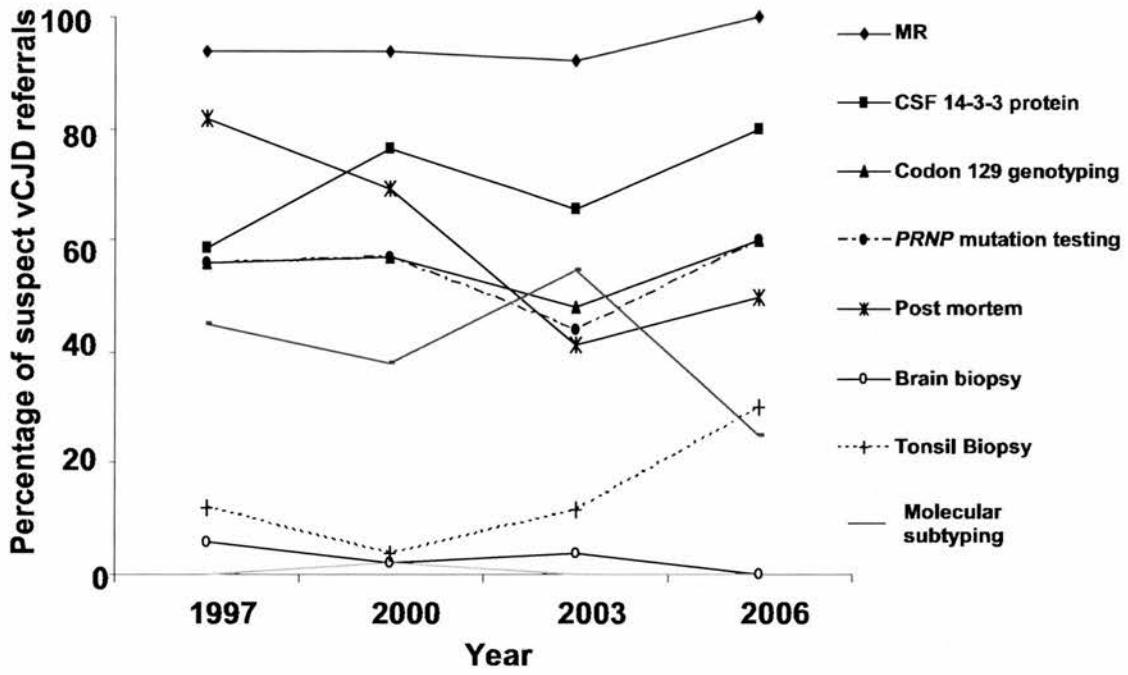

Figure 54 Proportion of suspect vCJD cases referred to the NCJDSU over selected years that underwent investigations that might support a diagnosis of vCJD

Changing diagnostic criteria

CSF 14-3-3 protein examination was incorporated into WHO diagnostic criteria for sCJD in 2000. Increasing use of CSF 14-3-3 protein in the investigation of suspect sCJD cases in the UK was described in the preceding section. An examination of the number of probable sCJD cases that met the diagnostic criteria based on EEG and clinical features or CSF 14-3-3 protein and clinical features gives some insight into the application of these adapted criteria by the NCJDSU as a measure of the flexibility of the system. Over 60% of all probable sCJD cases from 2000, 2003 and 2006 met the WHO diagnostic criteria based on CSF 14-3-3 protein and clinical features, compared to just approximately 15% based on EEG and clinical features (Table 32). This indicates that CSF 14-3-3 protein has made a substantial contribution to disease surveillance.

Table 32 Suspect sCJD cases meeting WHO diagnostic criteria as a probable case of sCJD based on EEG findings and clinical features or CSF 14-3-3 protein and clinical features

	Number (%) of probable sCJD cases	
Year	**EEG findings and clinical features**	**CSF 14-3-3 protein and clinical features**
2000	1 (12.5)	5 (62.5)
2003	6 (18.2)	19 (57.6)
2006	3 (11.1)	18 (66.7)
Total	**10 (14.7)**	**42 (61.8)**

Over these years only two thirds of individual meeting the diagnostic criteria as a definite, probable or possible sCJD cases underwent CSF 14-3-3 protein examination; 95% underwent EEG examination. To determine the potential under-ascertainment of sCJD cases through the under-utilisation of CSF 14-3-3 protein examination, the use of CSF 14-3-3 protein in the investigation of individuals classified at data censoring as a possible sCJD case was examined (Table 33). The maximum number of sCJD cases that may have been missed through under-utilisation of CSF 14-3-3 protein examination over this period was 11.

Table 33 Assessment of the potential degree of under-ascertainment of sCJD cases

Year	**Number of possible sCJD cases**	**Underwent EEG, n (%)**	**Underwent CSF 14-3-3 protein, n (%)**	**Possible under-ascertainment, n (%)**
2000	8	8 (100)	2 (25.0)	6 (75.0)
2003	2	2 (100)	0 (0)	2 (100)
2006	4	4 (100)	1 (25.0)	3 (75.0)
Total	**14**	**14 (100)**	**3 (21.4)**	**11 (78.6)**

Interpretation: The flexibility of the surveillance system has been challenged in the UK by emergent disease, new diagnostic technologies and changing diagnostic criteria. Following the emergence of vCJD in the UK a significant increase in the absolute number and age standardised rates of referral to the NCJDSU occurred, peaking between 2000 and 2001. This was driven by an increase in referrals of suspect sCJD and genetic prion disease in addition to suspect vCJD cases. The NCJDSU was able to respond to this increasing demand, visiting a consistently high

proportion of suspect vCJD cases in life. However, fewer than expected suspect sCJD cases were visited in 1997 and 2000 and an increase in the median number of working days from referral to visit was observed in these years. Given the limited implications of a diagnosis of sCJD, it could be argued that the system responded appropriately. Over time an increasing proportion of suspect sCJD cases underwent EEG, MRI scanning and CSF 14-3-3 protein examination during the course of their clinical illness. However there was evidence of potential under-ascertainment of sCJD cases attributable to the sub-optimal use of CFS 14-3-3 protein. Fewer than half of suspect sCJD cases underwent *PRNP* Codon 129 genotyping and full sequencing of *PRNP* for mutations, and post mortem rates in suspect sCJD and suspect vCJD cases fell across the study period. The former is crucial in excluding genetic prion disease which has a broad clinical phenotype and may be clinically indistinguishable from sCJD, and the latter in case confirmation. Overall, these data suggest that the NCJDSU is flexible and has responded appropriately to changing demands. However the possible under-ascertainment of sCJD cases and limited use of key diagnostic technologies is cause for concern. This latter issue will be addressed in greater detail in the section that follows on data quality.

Data Quality

> *"Data quality reflects the completeness and validity of the data recorded in the public health surveillance system".*(222)

The following metrics were considered:

1. The completeness of investigations to support a diagnosis of sCJD or vCJD in suspect sCJD and vCJD cases referred to the NCJDSU
2. Review by the NCJDSU of EEG, MRI and neuropathological studies from suspect sCJD cases and suspect vCJD cases referred to the NCJDSU
3. The extent to which multi-source clinical and diagnostic information on sCJD and vCJD cases (definite or probable) has been reviewed by the NCJDSU
4. The degree of missing data from key variables in the minimum monitoring dataset for all suspect prion disease cases referred to the NCJDSU

5. The degree of missing data as measured by unknown or blank responses on the risk factor questionnaire for any suspect sCJD case or suspect vCJD case for whom the risk factor questionnaire was completed
6. The completeness of follow up of all suspect sCJD cases and suspect vCJD cases that met the WHO diagnostic criteria at *any* stage in their clinical illness as a possible sCJD or vCJD case.

The quality of a diagnosis of sCJD or vCJD

In the preceding section I described the changing use of investigations which support a diagnosis of prion disease over selected years in suspect sCJD and suspect vCJD cases. In Tables 34 and 35, the completeness of investigations that support a diagnosis of sCJD or vCJD in suspect sCJD and suspect vCJD cases, according to case classification at data censoring, over selected years are described. In suspect sCJD cases there was evidence of differential use of EEG, MRI, CSF 14-3-3 protein, *PRNP* Codon 129 genotyping and *PRNP* mutation testing according to case classification. sCJD cases (classification 1.0 or 2.0) were more likely than non-sCJD cases (classification 4.1, 4.2 or 4.3) to undergo EEG examination ($P<0.001$), MRI scanning ($P<0.001$), *PRNP* Codon 129 genotyping ($P<0.001$) and *PRNP* mutation testing ($P<0.001$) and possible sCJD cases (classification 3.0), were less likely that other groups to undergo CSF 14-3-3 protein examination ($P<0.001$).

In suspect vCJD cases, there was no significant difference in the use of MRI scanning ($P=0.328$), CSF 14-3-3 protein ($P=0.500$) or tonsil biopsy ($P=0.204$) according to case classification, although the number of suspect vCJD cases (classification 1.0 or 2.0) undergoing tonsil biopsy was small. However non-cases (classification 4.1, 4.2 or 4.3) were less likely that vCJD cases to undergo *PRNP* Codon 129 genotyping ($P<0.001$) or *PRNP* mutation testing ($P<0.001$).

Table 34 Use of investigation that support a diagnosis of sCJD in suspect sCJD cases referred to the NCJDSU according to case classification (selected years)

		EEG	MRI	CSF 14-3-3 protein*	*PRNP* Codon 129 genotyping	*PRNP* mutation testing	Post mortem†	Brain biopsy	PrP typing**
Case classification at data censoring, N (%)	1.0	229 (89.5)	160 (62.5)	92 (51.7)	123 (48.4)	114 (44.5)	241 (94.9)	18 (7.0)	107 (41.8)
	2.0	82 (98.8)	63 (75.9)	59 (81.9)	36 (43.4)	37 (44.6)	0 (0)	0 (0)	0 (0)
	3.0	36 (100)	20 (55.6)	5 (23.8)	11 (30.6)	11 (30.6)	0 (0)	0 (0)	0 (0)
	4.1	35 (71.4)	27 (55.1)	30 (73.2)	9 (18.4)	9 (18.4)	0 (0)	9 (0)	0 (0)
	4.2	83 (76.9)	41 (38.0)	50 (68.5)	19 (17.6)	19 (17.6)	1 (1.5)	1 (0.9)	0 (0)
	4.3	64 (85.3)	26 (34.7)	21 (45.7)	24 (32.0)	23 (30.7)	71 (95.9)	6 (8.0)	0 (0)

*CSF 14-3-3 protein from 1997 onwards; †Denominator deceased referrals; **Limited to neuropathologically confirmed sCJD cases

Table 35 Use of investigation that support a diagnosis of vCJD in suspect vCJD cases referred to the NCJDSU according to case classification censoring (selected years)

		MRI	CSF 14-3-3 protein	*PRNP* Codon 129 genotyping	*PRNP* mutation testing	Post mortem†	Brain biopsy	Tonsil Biopsy	PrP typing**
Case Classification at data censoring, N (%)	1.0	45 (97.8)	31 (67.4)	42 (91.3)	41 (89.1)	43 (93.5)	3 (6.5)	6 (13.0)	24 (52.2)
	2.0	17 (100)	14 (82.4)	11 (68.8)	11 (68.8)	0 (0)	0	4 (23.5)	2 (50)
	3.0	1 (100)	1 (100)	1 (100)	1 (100)	0 (0)	0	0 (0)	0 (0)
	4.1	14 (87.5)	11 (68.8)	1 (6.3)	1 (6.3)	0 (0)	0	1 (6.3)	0 (0)
	4.2	25 (89.3)	20 (71.4)	5 (17.9)	5 (17.9)	0 (0)	0	1 (3.6)	0 (0)
	4.3	12 (85.2)	7 (50.0)	6 (42.9)	6 (42.9)	13 (92.9)	1 (7.1)	0 (0)	0 (0)

†Denominator deceased referrals; **Limited to CJD cases (definite or probable) with tissue (neuropathological or tonsil)

Review of investigations that support a diagnosis of sCJD and vCJD

It is crucial that the NCJDSU review investigations that support a diagnosis of sCJD or vCJD in suspect cases. The National CSF 14-3-3 protein laboratory is based at the NCJDSU. Quality assurance of this investigation can therefore be monitored. The same cannot be said of EEGs, MRIs and pathological studies which are carried out and reported throughout the UK. The proportion of suspect sCJD cases for whom the NCJDSU reviewed EEG, MRI and neuropathological studies, and the proportion of suspect vCJD cases for whom the NCJDSU reviewed EEG, MRI, neuropathological and pathological (tonsil biopsy) studies, can be considered as a measure of data quality. The denominator for these analyses is the number of suspect sCJD or vCJD cases known to have undergone these investigations over selected years.

Suspect sCJD cases

In just over half of all suspect sCJD cases that underwent MRI scanning, images were reviewed by the NCJDSU neuroradiologist (Table 36). There was an increase in the proportion of suspect sCJD cases in which MRI imaging was reviewed over time from 20.0% in 1991 to 71.8% in 2006 ($P<0.001$). EEGs were reviewed by the NCJDSU in 58.6% of all suspect sCJD cases that underwent EEG examination with year to year variation in this proportion but no discernible temporal trend ($P=0.243$). The NCJDSU reviewed neuropathological material obtained from brain biopsy in life or post mortem following death in 48.0% and 75.1% respectively of suspect sCJD cases for whom tissue was available with no significant change over time in either ($P=0.362$ and $P=0.112$ respectively).

Suspect vCJD cases

The corresponding data for suspect vCJD cases are presented in Table 36. MRI imaging was reviewed in a high proportion of suspect vCJD cases that underwent this investigation; EEG in a smaller proportion possibly reflecting the limited value of EEG in supporting a diagnosis of vCJD. Neuropathological tissue where available, was reviewed for all brain biopsies undertaken and over 80% of post mortem examinations. However tissue from tonsil biopsy was reviewed in only 58.3% of suspect vCJD cases undergoing this investigation.

Table 36 Review by NCJDSU of investigations that support a diagnosis of sCJD/vCJD in all suspect sCJD/vCJD cases referred to the NCJDSU over selected years

Year	All suspect sCJD cases, n (%)				All suspect vCJD cases, n (%)				
	MRI	EEG	Tissue from		MRI	EEG	Tissue from		
			Post mortem	Brain biopsy			Post mortem	Brain biopsy	Tonsil biopsy
1991	1 (20.0)	39 (66.1)	28 (66.7)	2 (66.6)	-	-	-	-	-
1994	7 (20.0)	55 (63.2)	44 (72.1)	0 (0.0)	-	-	-	-	-
1997	11 (25.0)	41 (44.1)	54 (77.1)	3 (60.0)	19 (59.4)	10 (33.3)	13 (72.2)	2 (100)	3 (42.9)
2000	50 (63.3)	58 (57.4)	45 (81.8)	4 (66.7)	34 (70.8)	28 (65.1)	25 (92.6)	1 (100)	1 (50.0)
2003	50 (56.5)	60 (58.3)	39 (81.3)	3 (75.0)	19 (79.2)	7 (33.3)	7 (100)	1 (100)	2 (66.7)
2006	51 (71.8)	51 (67.1)	25 (67.6)	0 (0.0)	8 (80.0)	1 (12.5)	0 (0.0)	0 (100)	1 (25.0)
Total	**172 (51.0)**	**304 (58.6)**	**235 (75.1)**	**12 (48.0)**	**80 (70.2)**	**46 (45.1)**	**46 (82.1)**	**4 (100)**	**7 (58.3)**

Multi-source information for sCJD and vCJD cases (definite or probable)

Multi-source information is collected by the NCJDSU for each sCJD and vCJD case: clinical examination of the case by a NCJDSU neurologist, interview with relatives, review of hospital medical records and review of medical records from primary care. Over selected years the completeness of multi-source information was assessed for all sCJD and vCJD cases (Table 37). Overall, a neurologist from the NCJDSU examined 90% of sCJD cases (alive at time of referral), with a decline in this percentage in 1997 and 2000. Relatives of sCJD cases were interviewed in similar proportions. Hospital records were reviewed in over 80% of sCJD cases, with a decline in this percentage in 2006. An increase over time was noted in the percentage of medical records from primary care that were reviewed for sCJD cases, although overall medical records from primary care were reviewed in only 15.6% of all sCJD cases. In all vCJD cases the NCJDSU neurologist examined the case and interviewed the relatives. In the majority of vCJD cases hospital records were reviewed and records from primary care accessed.

Table 37 Information available from various sources on sCJD cases and vCJD cases (definite and probable) referred to the NCJDSU according to year of referral

Year	Clinical Exam *		Interviewed Relatives		Reviewed Hospital records		Reviewed primary Care records†	
	sCJD	vCJD	sCJD	vCJD	sCJD	vCJD	sCJD	vCJD
1991	13(92.9)	-	27(90.0)	-	26(86.7)	-	0	-
1994	30(96.8)	-	43(89.6)	-	44(91.2)	-	2(4.2)	-
1997	35(89.7)	12(100)	46(78.0)	12(100)	49(83.1)	11(91.7)	2(3.4)	9(75.0)
2000	30(83.3)	23(100)	33(80.5)	24(100)	35(85.4)	24(100)	9(22.0)	20(83.3)
2003	31(91.2)	6(100)	40(93.0)	7(100)	37(86.0)	7(100)	13(30.2)	7(100)
2006	24(92.3)	3(100)	29(82.9)	3(100)	24(68.6)	1(50.0)	9(27.3)	1(50.0)
Total	**163(90.6)**	**44(100)**	**218(85.2)**	**46(100)**	**215(84.0)**	**43 (95.6)**	**(15.6)**	**37(82.2)**

*denominator alive at time of referral; †denominator deceased patients

Information was available from all of the above sources for 107 (31.6%) sCJD cases and 50 (81.0%) vCJD cases. Information was unavailable from any of the above sources in 25 sCJD cases (of which 21 were neuropathologically confirmed sCJD

cases); all 25 were deceased at the time of referral to the NCJDSU. In contrast, information was available from at least one source in all vCJD cases.

Missing data – key variables from minimum monitoring dataset

Case records of all suspect prion disease cases referred to the NCJDSU over selected years were examined to determine whether key variables from the minimum monitoring dataset identified by the WHO were missing (Table 38). As previously noted, date of referral was often recorded as either date of NCJDSU neurologist visit or date of death in 1991. Whilst there were no missing data for this variable it was not possible to validate this information. In suspect sCJD cases, where demise is rapid, month and year of neurologist visit or death are a reasonable proxy for month and year of referral; the same cannot be said for other disease subtypes, for example suspect vCJD cases or suspect genetic prion disease, which are associated with longer illness durations.

Table 38 Episodes of missing data from key variables in minimum monitoring dataset (all suspect prion disease referrals received in selected years), according to year

	Number of episodes where data were missing (%)					
Variable	**1991**	**1994**	**1997**	**2000**	**2003**	**2006**
Referral source	0	0	1 (0.6)	29 (16.3)	10 (6.2)	6 (5.6)
Date of referral	0	0	0	0	0	0
Sex	0	0	0	0	0	0
Date of birth	1 (1.3)	2 (1.8)	2 (1.2)	1 (0.6)	0	0
Date of onset	12 (16.0)	16 (13.7)	23 (14.3)	15 (8.4)	9 (5.6)	6 (5.6)
Residence at onset	5 (6.7)	7 (6.0)	9 (5.6)	7 (3.9)	4 (2.5)	2 (1.9)
Date of death	0	0	0	0	0	0

*dates required MM/YY

Referral source was well recorded in the early years of surveillance but missing in one in every six referrals received in 2000, and in over 5% of referrals received by the NCJDSU in 2003 and 2006. Date of symptom onset was missing in almost one in every six suspect prion disease cases referred to the NCJDSU in 1991, although there was evidence that recording of this variable improved over time. Residence at onset was missing in 6.7% of all suspect prion disease cases referred to the NJCDSU in 1991, falling to 1.9% in 2006. Of note over 70% of suspect prion disease cases

referred to the NCJDSU for whom key variables from the minimum monitoring dataset were missing were classified as non-cases (classification 4.1, 4.2 or 4.3). The majority of these were suspect sCJD cases, reflecting the large proportion of suspect sCJD referrals received by the NCJDSU relative to other disease subtypes.

As previously noted, multi-source data was available from clinical examination of the case, interview with relatives, review of medical records in primary care and review of medical records from secondary care in 107 sCJD and 50 vCJD cases. In this group multi-source information was used to verify the data recorded for the key variables of the minimum monitoring dataset described in Table 38 above. There were no inaccuracies in data entry. In a minority of the case notes reviewed (<1%) there was evidence that the Study Co-ordinator had triangulated data and corrected inaccuracies in the recording of variables from the minimum monitoring dataset.

Blank responses in risk factor questionnaire

Selected questions on the risk factor questionnaire (residential history, occupational history, medical including surgical history, family history and a history of blood donation / transfusion) were examined for suspect sCJD and vCJD cases for which this had been completed. With the exception of an occasional isolated omission, data recording was complete (98 – 100%).

The follow up of suspect sCJD and suspect vCJD cases

This section reviews the follow up of suspect sCJD and suspect vCJD cases referred to the NCJDSU over selected years that met the WHO diagnostic criteria as a possible, probable or definite case of sCJD or vCJD at any stage in the course of their clinical illness.

Suspect sCJD cases

In total 418 suspect sCJD cases referred to the NCJDSU over selected years met the diagnostic criteria as a possible (or greater) sCJD case at any stage in their clinical illness. At the time of data censoring a neuropathological diagnosis had been reached in 273 (65.3%), of whom 256 (93.8%) had sCJD. Pathologically confirmed non-sCJD cases (n=17) were most commonly Alzheimer's Disease or Lewy Body Dementia (Table 39). At data censoring, 36 cases remain classified as possible

sCJD; all were deceased with no prospect of further clinical information becoming available. Five suspect sCJD cases were classified as 4.1, diagnosis unclear; no further clinical information was available for these suspect cases despite repeated documented attempts at follow up. A further 23 individuals were, at the time of data censoring, classified as 4.2, indicating that sCJD was clinically unlikely. In this group an alternate clinical diagnosis had been reached for 17 (Table 40), a further two had improved clinically although a diagnosis had not been reached and in four an alternate clinical diagnosis was unavailable although the referring clinician confirmed that sCJD was no longer suspected.

Table 39 Cause of death as determined by a neuropathologist in suspect sCJD cases that met the diagnostic criteria as a possible (or greater) sCJD case during the course of their clinical illness but had an alternate neuropathologically confirmed diagnosis

Cause of death	Number
Alzheimer's Disease	6*
Lewy Body Dementia	4
Cerebrovascular Disease	2*
Amyloid Angiopathy	1
Angiotrophic Lymphoma	1
Cerebellar Encephalitis	1
Cerebral Lymphoma	1
Inflamed Leptomeninges	1
Multifocal calcifying leucoencephalopathy	1

*Dual pathology in one patient

Table 40 Alternate clinical diagnoses in individuals meeting the diagnostic criteria as a possible (or greater) case of sCJD at any stage during their clinical illness

Clinical Diagnosis	Number
Encephalitis ? cause	3
Lewy Body Dementia	3
Iatrogenic effects of drugs	2
Steroid responsive encephalopathy	1
Central Pontine Myelinolysis	1
Depression	1
Granulomatous disease	1
Multi-system atrophy	1
Myeloma	1
Normal Pressure Hydrocephalus	1
Paraneoplastic syndrome	1*
Seizures post head injury	1

*Brain not examined, primary lung tumour

A clinical or neuropathological diagnosis had not been reached at the time of data censoring for 41 suspect sCJD cases that met the diagnostic criteria as a possible (or greater) case of sCJD at any stage in their clinical illness, 36 individuals classified as possible sCJD cases and five classified as 4.1, diagnosis unclear. Details of the investigations undertaken to support a diagnosis of sCJD in this group during the course of their clinical illness are shown in Table 41. EEG was commonly undertaken but less than a third underwent CSF 14-3-3 protein examination whilst over half underwent MRI scanning. In four possible sCJD cases features on MRI scanning supported a diagnosis of sCJD. Approximately a third of suspect sCJD cases in this group underwent *PRNP* Codon 129 genotyping. The distribution of Codon 129 genotypes was 42.9% (6) methionine homozygote, 28.6% (4) methionine heterozygote and 28.6% (4) valine homozygote. Full sequencing for *PRNP* mutations was not carried out in any suspect sCJD case in this group, and none had undergone brain biopsy in life or autopsy following death.

Table 41 Investigations undertaken in suspect sCJD cases that met the diagnostic criteria as a possible (or greater) case of sCJD during the course of their clinical illness in whom a clinical or neuropathological diagnosis had not been reached at data censoring

Investigation	Number (%)
EEG	40 (97.6)
CSF 14-3-3 protein*	8 (32.0)
MRI	23 (56.1)
PRNP Codon 129 genotyping	14 (34.2)
PRNP Mutation testing	0 (0)
Brain Biopsy	0 (0)
Post mortem	0 (0)

*denominator limited to 1997 onward

A neurologist from the NCJDSU visited 335 (80.1%) suspect sCJD cases and/or their relatives that met the diagnostic criteria as a possible (or greater) sCJD case at any stage in their clinical illness. The reason why a visit was not undertaken, where known, is listed in Table 42.

Table 42 Reason why the NCJDSU did not visit suspect sCJD case that met the diagnostic criteria as a possible (or greater) sCJD case at any stage in their clinical illness

Reason for no visit from the NCJDSU	Number (%)
No response by referring clinician to request for next of kin's details	19 (22.9)
No response from family to postal invitation to participate in surveillance	12 (14.5)
Family declined interview	3 (3.6)
Family not yet approached – awaiting further clinical / diagnostic information	3 (3.6)
Clinician advised against contacting family	2 (2.4)
Family felt 'too soon' for visit	2 (2.4)
Reason unknown	24 (28.9)

Of the 41 suspect sCJD cases in whom a clinical or neuropathological diagnosis had not been reached (36 possible and 5 diagnosis uncertain), 40 (97.6%) were known to have died at data censoring; death certificates were available for review at the NCJDSU for 39 (97.5%). Three quarters (26) of suspect cases classified as possible CJD at data censoring had CJD recorded on their death certificate. In the remaining

possible cases the underlying cause of death was recorded as dementia (5), alzheimer's disease (1), stroke (1), brain tumour (1) and cause of death unknown (1). Four of the five individuals classified as 4.1 (diagnosis uncertain) were known to have died with the following recorded as the unerlying cause of death: dementia (1), Alzheimer's disease (1), bronchopneumonia (1) and stroke (1). The vital status of the fifth individual (referred in 2003 with a one year history of cerebellar symtpoms) was unknown. In total then just 2 suspect sCJD cases that met the diagnostic criteria as a possible sCJD case at any stage in their clinical illness were considered to have been lost to follow up, that is a clinical or neuropathological diagnosis had not been reached at data censoring and follow up information from primary care, secondary care or a death certificate was unavailable.

Suspect vCJD cases

In total 69 suspect vCJD cases referred to the NCJDSU over this period met the diagnostic criteria as a possible (or greater) vCJD case at any stage in their clinical illness. A neuropathologically confirmed diagnosis was reached in 49 (71.1%). Of these a diagnosis of vCJD was confirmed in 46 (93.9%). Three cases had an alternate neuropathological diagnosis: one Alzheimer's disease, one Alzheimer's Disease and amyloid angiography dual pathology and one subacute sclerosis panencephalitis. At data censoring 17 suspect vCJD cases in this group were classified as probable vCJD and one as a possible vCJD case. The latter individual died without post mortem examination; tonsil biopsy was not performed ante-mortem and due to movement artefact MRI scanning, although carried out, did not contribute to the diagnostic process. Clinical diagnoses (viral encephalitis and a functional illness) had been reached in the final two suspect vCJD cases in this group who were, at the time of data censoring, classified as 4.2, vCJD clinically unlikely.

In all but two instances a neurologist from the NCJDSU had visited the suspect vCJD cases in this group and/or their relatives to collect further information. In the latter two suspect vCJD cases the family had refused a visit from the NCJDSU; these suspect vCJD cases were classified, at the time of data censoring, as a probable vCJD case and 4.2, vCJD clinically unlikely. The vital status, at data censoring, of all suspect cases was known. Three individuals were known to be alive (two probable

vCJD cases and one individual classified as 4.2 (vCJD clinically unlikely)). Death certificates had been received by the NCJDSU for all but one deceased suspect case. The latter individual, a neuropathologically confirmed vCJD case underwent brain biopsy during life (tissue reviewed by the NCJDSU) and was known to have died abroad.

Interpretation: These data confirm the findings from the preceding section that examinations to support a diagnosis of sCJD and vCJD are being under-utilized in the investigation of suspect sCJD and vCJD cases. This may compromise surveillance efforts. The validation of clinical and diagnostic information has generally improved over time, although there remains room for further improvement, particularly in the review of medical cases records from primary and secondary care and EEGs in the investigation of suspect sCJD cases. Overall data recording was excellent and evidence of improvement over time. The follow up of suspect sCJD and vCJD cases that met the diagnostic criteria as a possible sCJD or vCJD case at any stage in the course of their clinical illness was also very good; less than 1% (2) of suspect sCJD, and no suspect vCJD cases that met the diagnostic criteria as a possible case at any stage in their clinical illness were lost to follow up.

Acceptability

> *"Acceptability reflects the willingness of individuals and organizations to participate in the surveillance system."* (222)

Acceptability can be measured in a number of ways. Firstly rates and patterns of referrals of suspect sCJD and vCJD cases can be considered. Secondly, the willingness of suspect sCJD and vCJD cases and their relatives to participate in surveillance can be examined. Thirdly, completion rates for questions in the risk factor questionnaire can be assessed. These latter two metrics were addressed under the subheading 'Data Quality' and will not be revisited here. Finally, the willingness of public health policy makers to use data from the NCJDSU to support and inform decision making can be examined.

Referral rates and patterns

Referral rates to the NCJDSU from 1990 through 2006 were described under the subheading 'Flexibility' and will not be recounted in detail here. A number of additional points may be considered when examining referral rates and patterns. Firstly, the proportion of suspect sCJD and vCJD cases referred to the NCJDSU in whom a neuropathologically confirmed diagnosis of sCJD and vCJD is reached. These data are shown in Figure 55. The WHO recommends that the number of suspect CJD cases referred to a surveillance system should exceed the number of confirmed cases by a factor of two. As can be seen in Figure 56 there has been variation over time in the proportion of suspect sCJD and vCJD cases referred to the NCJDSU in whom a neuropathological diagnosis of sCJD or vCJD was confirmed. However this has always been maintained at a factor of two or more.

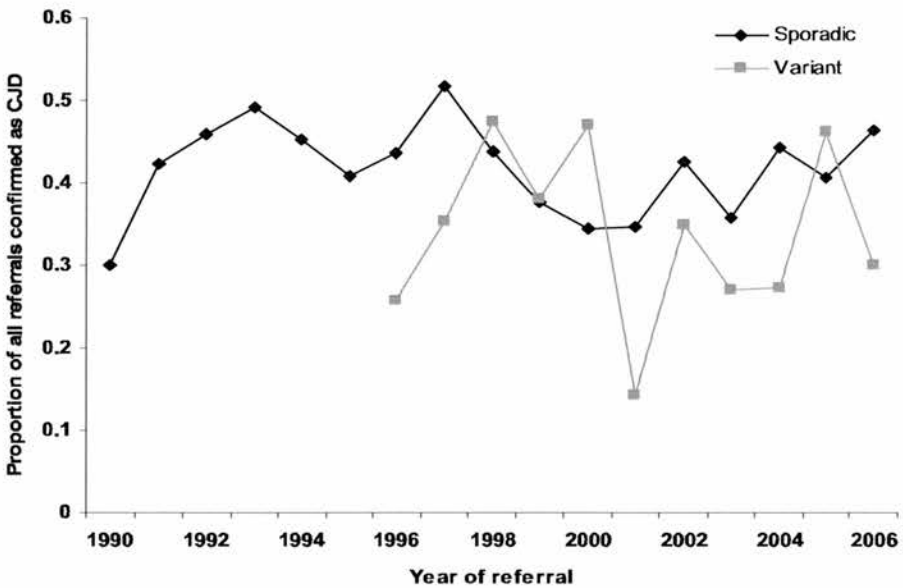

Figure 55 Proportion of all suspect sCJD and vCJD cases referred to the NCJDSU in whom a neuropathologically confirmed diagnosis of sCJD or vCJD was reached according to year of referral, 1990 – 2006

The second issue that should be considered is the source of referral to the NCJDSU. This metric was examined by analysing data from selected years of surveillance. A

declining reliance on death certificate data in the ascertainment of suspect prion disease cases could be considered a reflection of the acceptability of the surveillance system to health care professionals and the public. For example, in 1991, 18.3% (13) of all suspect sCJD cases referred to the NCJDSU were ascertained through death certificates review alone; in 2006 this figure was zero. A number of professional groups receive regular mailing asking them to refer suspect prion disease cases to the NCJDSU. These include neuropathologists, neurophysiologists and neurologists. Over selected study years 69.0% (49) of suspect sCJD cases and 71.1% (59) of suspect vCJD cases referred to the NCJDSU were referred by these groups. Unsolicited referrals are received from a number of other groups including general physicians, psychiatrists, other health care professionals and even the relatives of suspect cases. Over time the proportion of suspect sCJD referrals received by these groups increased, from 12.7% (9) in 1991 to 21.7% (18) in 2006 (P=0.002). For suspect vCJD cases from 23.1% (3) in 1997 to 50% (5) in 2006 (P=0.052).

It might be hypothesised that referrals from non-specialist groups are less desirable that those from specialists groups, as they are likely to have a lower sensitivity. As a measure of sensitivity, the proportion of all referrals received by each referral source that met the diagnostic criteria as a definite or probable case of sCJD and vCJD were examined. Overall the sensitivity was 39.0% (35.0 – 43.0) for suspect sCJD and 54.5% (45.2 – 63.7) for suspect vCJD (Table 43).

Table 43 Sensitivity of referral of suspect sCJD and vCJD cases from selected years, according to referrals source

	Sensitivity % (95% CI)	
	Suspect sCJD	**Suspect vCJD**
Neurologist	28.0 (23.3 – 32.8)	56.2 (45.9 – 66.5)
Neuropathologist	91.2 (83.9 – 98.6)	100
General Physician	50.8 (38.1 – 63.6)	75.0 (32.6 – 100)
Death Certificate	20.5 (7.8 – 33.1)	-
Psychiatrist	25.0 (6.0 – 44.0)	14.3 (0 – 40.2)
Neurophysiologist	50.0 (15.3 – 84.6)	-
Other	65.8 (50.7 – 80.9)	50 (19.0 – 81.0)
All	39.0 (35.0 – 43.0)	54.5 (45.2 – 63.7)

Predictably the sensitivity was greatest when the referral came from a neuropathologist for both suspect sCJD and suspect vCJD cases. Conversely, this was lowest when suspect cases were ascertained through death certificate review or referred by a psychiatrist. Interestingly the sensitivity was high when suspect cases were referred by the 'other' group, which includes other health care professionals and patients relatives. Sensitivity was greater for general physicians referring suspect vCJD cases than neurologist, although not significantly so. For suspect sCJD cases there was a significant difference. It should be considered however that whilst the referral to the NCJDSU may have come from a general physician many suspect sCJD and suspect vCJD cases are reviewed by a neurologist during the course of their clinical illness. For example, 89.9% (549) of all suspect sCJD and 86.9% (106) suspect vCJD cases referred to the NCJDSU were known to have been admitted to hospital during the course of their clinical illness. Suspect sCJD cases were most commonly admitted under a general physician, 33.1% (203), 24.5% (150) under the care of a neurologist and 13.5% (82) under the care of a geriatrician; suspect vCJD cases under a neurologist 40.2% (49) or mental health specialist, 24.6% (30), reflecting the prominent clinical features at onset. In total, 81.9% (502) of suspect sCJD and 95.1% (116) of all suspect vCJD cases referred to the NCJDSU were known to have been reviewed, as an in-patient or out-patient, by a neurologist.

A National Referral System was introduced by the Chief Medical Officer (CMO) in 2004. This system required a National Reporting Form to be faxed by the notifying clinician to NCJDSU, the NPC and the local CCDC. This system was intended to replace the less formal notification system that had been in operation which allowed referrers to contact the NCJDSU by telephone or in writing to informally discuss or formally refer suspect CJD cases. In 2006 the National Reporting Form was completed in just 14.8% (16) of all suspect prion disease cases referred to the NCJDSU. The low level of participation in completing the National Reporting Form indicates that this is not acceptable to referrers.

Use of surveillance data to inform public health policy

A final assessment of acceptability comes from the continued willingness of policy makers to utilise data produced by the NCJDSU to support and inform public health

decision making. For example, Prof. Will provided key testimony during the BSE Inquiry, other senior NCJDSU staff frequently respond to parliamentary questions regarding human prion disease and contribute to a number of committees providing expert independent scientific advice to government or directly developing public health policy, for example SEAC, the CJD IP and the ACDP.

Interpretation: High levels of participation of patients and relatives in disease surveillance, and an increasing proportion of suspect cases referred to the NCJDSU by a broadening range of referrers indicate that the system is acceptable. Limited use of the National Reporting Form suggests that this mechanism for referral is not acceptable and underscores the need for the NCJDSU to continue to accept referrals by a number of mechanisms.

Sensitivity

> *"The sensitivity of a surveillance system can be considered on two levels. First, at the level of case reporting, the proportion of cases of a disease or health conditions detected by the surveillance system can be evaluated. Second, the system can be evaluated for its ability to detect epidemics."* (222)

In the absence of a measure of the true occurrence of CJD in the UK a number of approaches can be adopted to assessing sensitivity. sCJD is not aetiologically linked to an exogenous exposure and geographically there is little variation in disease occurrence. The sensitivity of the NCJDSU could then be assessed by comparing the incidence of sCJD in the UK to the incidence of sCJD elsewhere. Such comparisons were drawn in chapter 1 and in the discussion of chapter 2 and will not be revisited here, beyond noting that temporal trends in sCJD incidence and mortality in the UK are comparable to temporal trends reported internationally. This is illustrated in Figure 56 which shows sCJD mortality rates reported by selected EUROCJD

collaborators from 1993 through 2006.(46)

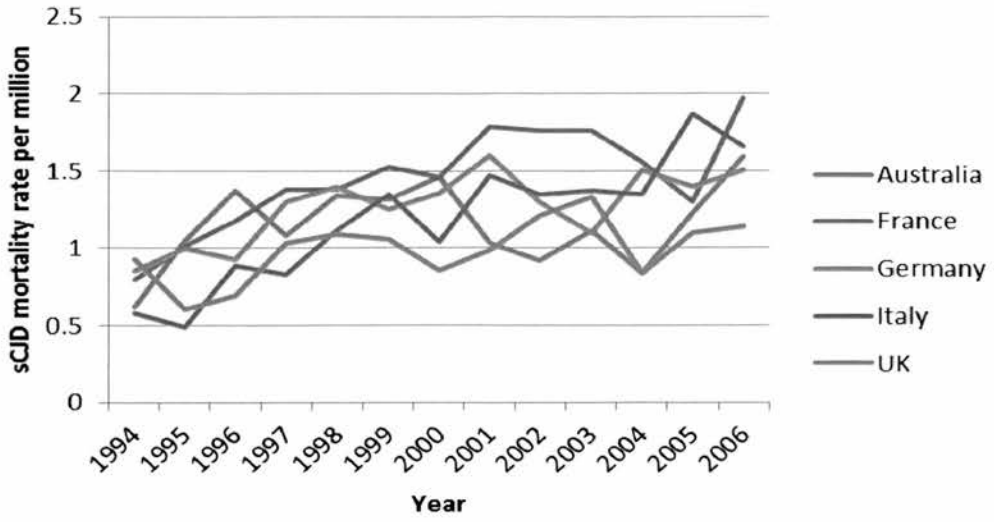

Figure 56 sCJD mortality rates (per million population) reported in selected EUROCJD countries, 1994 – 2006

Operationally, the NCJDSU aims to ascertain suspect cases sCJD and vCJD cases in life to facilitate rapid public health action where required. One approach to assessing sensitivity of the NCJDSU is to examine the proportion of all sCJD and vCJD cases ascertained by the NCJDSU that were referred to the NCJDSU in life. This is a measure of the sensitivity of the clinical surveillance system. In the section that follows the cohort of definite or probable sCJD and vCJD cases ascertained by the NCJDSU following death from 1990 through 2006 are described.

sCJD cases ascertained by the NCJDSU following death

From 1990 through 2006, 188 sCJD cases (177 definite and 11 probable) were referred to the NCJDSU following death, accounting for 21.1% of all sCJD cases ascertained by the NCJDSU over this period. The overall sensitivity of ascertaining sCJD cases in life was 78.5% (75.8 – 81.2), this increasing from 76.5% (62.2 – 90.7) in 1990 to 87.7% (79.7 – 93.7) in 2006 (Figure 57).

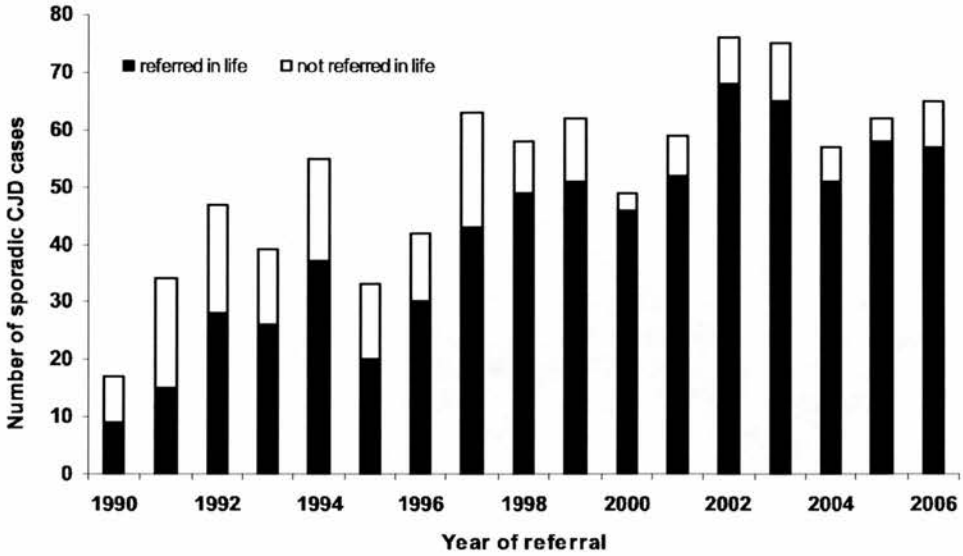

Figure 57 Annual number of sCJD cases ascertained by the NCJDSU according to vital status at time of referral, 1990 - 2006

The majority, 112 (59.6%), of these sCJD cases were referred to the NCJDSU by a neuropathologist (Figure 58). Of the six cases referred from 'other' sources, four were from family members, one from a CCDC and one from a virologist.

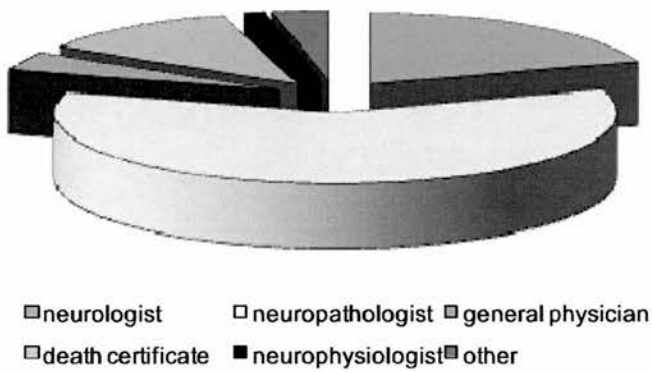

Figure 58 Referral source for sCJD cases referred to the NCJDSU after death, 1990 – 2006

sCJD case characteristics

In 22 sCJD cases no further information, beyond that provided in the time of referral, was available despite repeated (unsuccessful) attempts to obtain further information from primary and/or secondary care clinicians. This section will focus on the 166

sCJD cases (155 definite and 11 probable) for whom information from one or more sources (medical records from primary, medical records from secondary care or interview with the patients' relatives) was available. The characteristics of this group are described in Table 44. Of note, sCJD cases for whom further information from one or more of the sources described above was available did not differ significantly with respect to mean age (P=0.842) or sex (P=0.133) to those for whom no further information was available.

Table 44 Comparison of characteristics of sCJD cases referred to the NCJDSU according to vital status at the time of referral, 1990 - 2006

	Deceased at referral	**Referred in life**	**P value**
Number of sCJD cases (definite or probable)	166	705	
Men, n (%)	87 (52.4)	335 (47.5)	0.295
Mean age at death, Years (SD)	69.4 (9.7)	66.8 (9.7)	0.001
*Clinical presentation, n (%)**			
RPD	92 (55.4)	450 (63.9)	
Heidenhain Variant	7 (4.2)	40 (5.7)	
Psychiatric onset	14 (8.4)	28 (4.0)	
Slowly progressive dementia	12 (7.2)	36 (5.1)	
Cerebellar onset	15 (9.0)	84 (11.9)	0.006
Extra-pyramidal onset	3 (1.8)	0	
Stroke-like onset	5 (3.0)	17 (2.4)	
Sensory onset	7 (4.2)	12 (1.7)	
Other focal onset	9 (5.4)	31 (4.4)	
Investigations that might support a diagnosis of sCJD, n(%)			
Underwent one or more EEG examinations	120 (72.3)	685 (97.3)	<0.001
EEG typical	34 (28.3)	265 (38.7)	0.098
Underwent one or more CSF 14-3-3 protein examinations	0	430 (61.0)	
Underwent one or more MRI scans	69 (41.6)	525 (74.6)	<0.001
Features consistent with sCJD on MRI	15 (21.7)	186 (35.5)	0.154
Underwent *PRNP* Codon 129 genotyping	44 (26.5)	527 (74.8)	<0.001
Methionine homozygote	21 (47.7)	347 (65.8)	
Methionine heterozygote	9 (20.5)	92 (17.5)	0.002
Valine homozygote	14 (31.8)	88 (16.7)	
Underwent *PRNP* Mutation testing	8 (4.8)	423 (60.1)	<0.001
Underwent brain biopsy in life	2 (1.2)	32 (4.6)	<0.001
Brain biopsy diagnostic	1 (50.0)	24 (0.75)	-
Underwent post mortem examination following death	154 (92.8)	491 (69.7)	<0.001
Median illness duration, Months (IQR)	4.4 (2.5-8.8)	4.2 (2.7-7.7)	0.785

**Missing 2(1.2%) cases and 6(0.9%) cases*

Differential diagnoses considered in sCJD cases not referred to the NCJDSU in life

In three quarters of sCJD cases, 127 (76.5%), that were not referred to the NCJDSU in life, the clinical team managing the patient had considered a diagnosis of sCJD during the course of the clinical illness. sCJD was first considered as a differential diagnosis a median of 89 (53 – 185) days after onset and 25 (13 - 54) days prior to death in sCJD cases. Other differential diagnoses considered in life in this group are outlined in Table 45.

Table 45 Differential diagnoses considered by clinical team caring for sCJD cases that were not referred to the NCJDSU in life

Differential Diagnoses	Number
Paraneoplastic syndrome	20
Multi-infarct dementia	19
Alzheimer's Disease	11
Lewy Body Dementia	7
Cerebrovascular Disease	6
Unspecified Viral Encephalitis	4
Depression	4
Alcohol Related	4
Parkinson's Disease	3
Vasculitis	3
Multi-system Atrophy	3
Pick's disease	2
Corticobasilar Degeneration	2
Auto-immune Disease	2
Limbic Encephalitis	2
Motor Neurone disease	2
Dementia ? Cause	2
Cerebellar Degeneration ? Cause	1
Adult Reyes syndrome	1
Frontotemporal Dementia	1
Hypoxic Encephalopathy	1
Progressive supranuclear palsy	1

The features, signs, symptoms and supportive investigations that led clinicians to consider a diagnosis of sCJD are outline in Table 46 below.

Table 46 Key clinical features and investigations that led clinicians to consider a differential diagnosis of sCJD cases among sCJD cases not referred to the NCJDSU in life

	Features of illness that led sCJD to be considered	Number
Clinical signs / symptoms alone	RPD	20
	RPD + Cerebellar	22
	RPD + Myoclonus	28
	RPD + Visual	3
	RPD + Myoclonus + Cerebellar	5
	RPD + Myoclonus + Extra-pyramidal	1
	RPD + Cerebellar + Visual	1
	Cerebellar	2
	Cerebellar + Myoclonus	2
	Myoclonus	1
	Pyramidal	1
	Visual	1
	Dementia + Pyramidal + Extra-pyramidal	1
Clinical signs / symptoms and Supportive Investigations	RPD + EEG	11
	RPD + Myoclonus + EEG	10
	RPD + Cerebellar + EEG	2
	Cerebellar + EEG	1
	Visual + EEG	1
	RPD + MRI	1
Supportive Investigations alone	EEG	11
	MRI	2

Based on the clinical information available to the NCJDSU I determined the highest case classification that each sCJD case would have reached in life (Figure 59). There was insufficient clinical information to assign a case classification to one sCJD case (classification 0.0). Just under a fifth, 31 (18.9%) of sCJD cases met the WHO diagnostic criteria as a probable case of sCJD, 97 (58.4%) as a possible sCJD case and 37 (22.3%) did not meet the WHO diagnostic criteria (case classification 4.1).

A diagnosis of CJD had been considered in 30 of the 31 sCJD cases that met the WHO diagnostic criteria as a probable case of sCJD. In these patients sCJD was first considered as a differential diagnosis a median of 58 (37 – 78) days after onset and 18 (13 – 48) days prior to death. In one case a recommendation to refer to the NCJDSU was documented in the medical case notes 6 days prior to death. In four further sCJD cases (three that met the WHO diagnostic criteria as a possible sCJD case and one that did not meet the diagnostic criteria (case classification 4.1) in life), a recommendation to refer the patient to the NCJDSU was documented in the medical case notes prior to death. In these cases the median time from the recommendation to refer to the NCJDSU being documented in the medical case note to death was 15 (8 - 18) days.

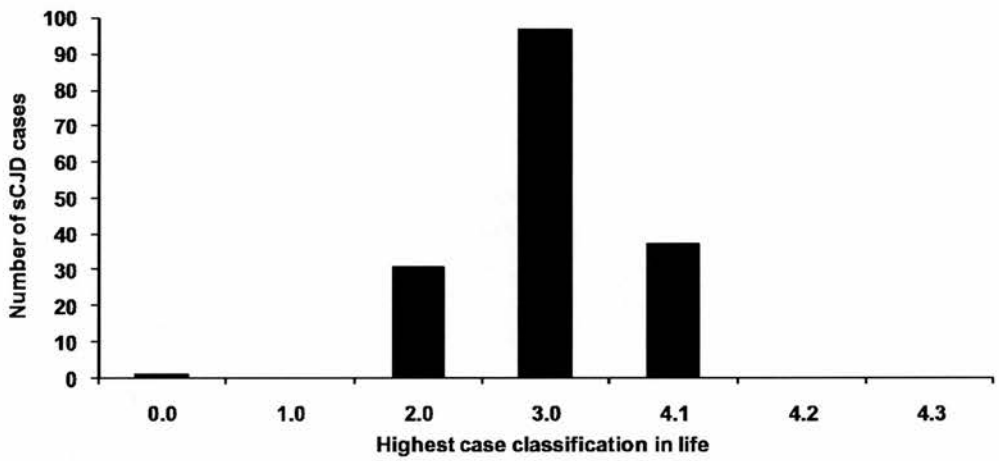

Figure 59 Highest case classification in life based on available clinical information for sCJD cases deceased at the time of referral to the NCJDSU, 1990 - 2006

vCJD cases ascertained by the NCJDSU following death

Over the entire study period four vCJD cases ascertained by the NCJDSU were deceased at the time of referral; all were neuropathologically confirmed cases in men aged 21.0 , 41.7, 69.9 and 74.9 years respectively. This group included one vCJD case attributable to the transfusion of labile blood products. Two cases presented with psychiatric onset and two rather unusually with RPD. In one case, a diagnosis of sCJD had been considered based on RPD and cerebellar signs; this case had evidence

of a 'pulvinar sign' on MRI but would not have met the WHO diagnostic criteria as a probable case of vCJD because of insufficient clinical features. One vCJD case met the WHO diagnostic criteria as a probable vCJD in life based on clinical features and the presence of the pulvinar sign on MRI. vCJD had not been considered in life in this individual who was one of the earliest vCJD cases (onset 1995) ascertained by the NCJDSU. The overall sensitivity of the clinical surveillance system at detecting cases in life was 97.6% (95.2 – 99.9) of vCJD cases.

The ability of the system to detect an epidemic

The sensitivity of the NCJDSU can also be assessed by examining the ability of the system to detect an epidemic. vCJD was first described in cases ascertained by the NCJDSU in 1996. Through the work of the NCJDSU the clinico-pathological epidemiology of vCJD has been described and the primary and secondary epidemics mapped. The clinical and epidemiological features of vCJD cases ascertained by the NCJDSU from 1990 through 2006 were described in Chapter 2 and will not be revised here. It is important to note that this metric captures the sensitivity of the whole system, clinical and pathological.

Interpretation: sCJD incidence/mortality rates in the UK have followed temporal trends which are consistent with international data, suggesting that the system is highly sensitive at detecting sCJD cases. The number of sCJD cases ascertained in life by the clinical surveillance system has increased over time. The clinical surveillance system is highly sensitive at detecting vCJD cases. The sensitivity of the NCJDSU was confirmed by its ability to detect and map both the primary and secondary epidemics of vCJD in the UK. This is a measure of the sensitivity of both the clinical and pathological aspects of the surveillance system.

Positive predictive value

> *"Predictive value positive (PVP) is the proportion of persons identified as having cases who actually do have the condition under surveillance."* (222)

Predictive value positive (PVP), more commonly referred to as the positive predictive value (PPV), can be considered as the proportion of suspect prion disease cases referred to the NCJDSU that actually had prion disease (definite or probable cases). Applying this definition the overall PPV of the NCJDSU from 1990 through 2006 was 57.5% (55.4 – 59.6). Over time this increased significantly from 37.7% (24.7 – 50.8) in 1990 to 76.1% (68.1 – 84.1) in 2006 (P<0.001). There was significant variation in PPV according to aetiological subtype. Overall the PPV for suspect sCJD cases was 54.5% (52.1 – 56.9). There was evidence of year to year variation in the PPV between 1990 and 2001, following which the temporal trend was toward an increase in PPV, from 45.0% (36.5 – 53.6) in 2001 to 76.2% (67.1 – 85.3) in 2006 (P<0.001). This increase in PPV was driven by an increase in the proportion of all sCJD referrals meeting the diagnostic criteria as a probable sCJD case, as illustrated in Figure 60, attributable to the contribution of CSF 14-3-3 protein examination.

Overall, the PPV for suspect vCJD was 51.9% (46.4 – 57.3), with year to year variation from, 29.0% (13.1 – 45.0) in 1996 to a high of 75.0% (56.0 – 94.0) in 2002 (P<0.001), although this did not follow a temporal trend. This is remarkable considering that vCJD was detected without a prior case definition. The overall PPV for iCJD and genetic prion disease were high at 94.7% (88.9 – 100) and 95.9% (92.3 – 99.4) respectively and invariant over time (P=0.618 and P=0.239 respectively).

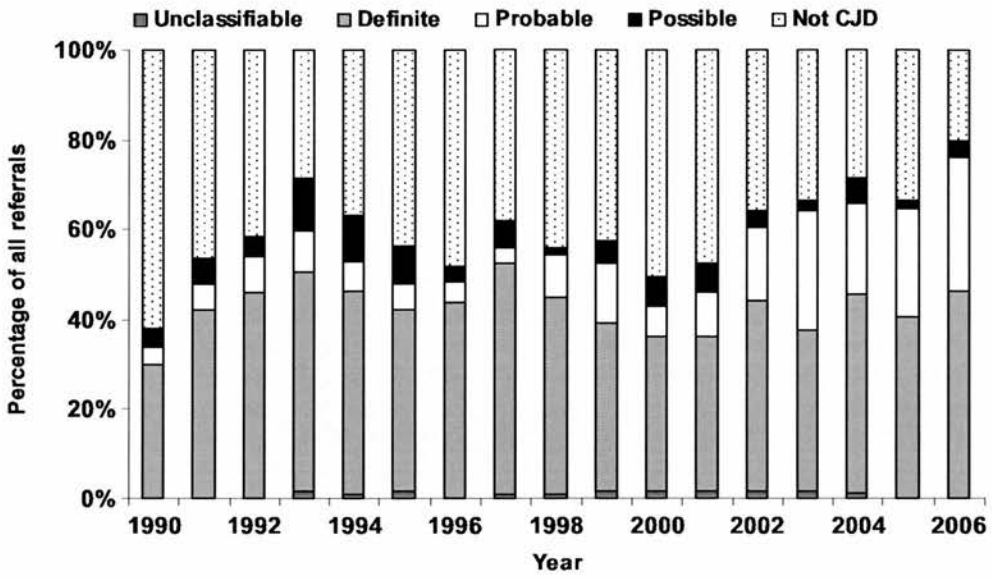

Figure 60 Case classification (at data censoring) of suspect sCJD cases referred to the NCJDSU according to year of referral, 1990 - 2006

Interpretation: The PPV of the NCJDSU was high and increased over time, overall and for suspect sCJD.

Usefulness

> *"A public health surveillance system is useful if it contributes to the prevention and control of adverse health-related events, including an improved understanding of the public health implications of such events."* (222)

The NCJDSU's objectives in 2006 were to provide accurate longitudinal data on the incidence and characteristics of all aetiological types of CJD and to study risk factors for the development of disease. Over the study period the NCJDSU has achieved many of its stated objectives. In chapter 2, using data from the NCJDSU, I described long-term trends in the epidemiology of prion diseases in humans in the UK according to disease subtype. During this period the NCJDSU identified and characterised a novel human prion disease, vCJD, and provided data from epidemiological and transmission studies in animals to support an aetiological link to

BSE in cattle. These data were rapidly dissemination to the scientific community, policy makers and public and, in turn, translated into public health practice and national, indeed international, policy. The NCJDSU has been uniquely placed through the on-going detection and characterisation of cases to develop and validate new diagnostic technologies in relation to vCJD (and other prion diseases) leading to the development and validation of diagnostic criteria for the purposes of disease surveillance.(186) In 2003 the NCJDSU identified secondary transmission of vCJD through a previously unrecognised route, the transfusion of labile blood components; it continues to investigate other potential routes of transmission including dentistry, surgery and maternal transmission through various studies including the on-going case control study and the PIND study. To date symptomatic vCJD has been identified in only *PRNP* Codon 129 methionine homozygotes. The identification of vCJD in a non-methionine homozygote would have significant public health implications. Experience from other human prion disease suggests that such individuals will have long incubation periods, lengthening the primary epidemic. However the major threat to public health is from potential secondary transmission of vCJD arising from an un-quantified population of asymptomatic, but potentially infectious individuals, who may be undergoing invasive medical procedures and donating blood and/or tissue. The NCJDSU have been working closely with the UKBTS to develop a blood test that can be used to identify abnormal prion protein. If successful this could be applied to screening donations of blood and/or tissue thereby limiting the potential for a secondary epidemic of vCJD.

Senior members of the NCJDSU are directly involved in influencing public health practice and policy in relation to the prevention and control of prion diseases in humans through involvement in numerous committees including SEAC, the ACDP, the CJD IP and the Advisory Committee on the Safety of Blood, Tissues and Organs (SaBTO). In turn on-going disease surveillance is crucial to providing epidemiological evidence to evaluate the success of control measures. Beyond a role in monitoring disease trends, identifying transmission routes and advising on prevention and control measures, the NCJDSU has had a role in therapeutic drug trials for those affected by human prion diseases. In addition, the NCJDSU

administer the National Care Package for CJD patients and their families and therefore work closely with health care professionals, special interest groups, patients and their families. A key role for the NCJDSU is to provide on-going advice and support to these groups during the patient's clinical illness and beyond. Senior member of the NCJDSU have held positions on the management committees of a number of charitable support organisations including the Human BSE Foundation, the UK CJD Support Network and other international support groups. In addition, the NCJDSU also responds to enquiries from the general public and media. For example between 2003 and 2006 the NCJDSU website received an average of 100,000 hits per annum.

Scientific research and surveillance activities are closely linked and interdependent. Fewer than half of the staff employed by the NCJDSU are primarily involved in disease surveillance; the majority are primarily involved in scientific research. The range of research is diverse, from transmission studies in animals in collaboration with the Institute for Animal Health at Edinburgh University, to evaluation of biomarkers for prion disease in humans with international collaborators (Anteprion and Prionscreen), to an evaluation of the clinical diagnostic criteria for CJD in association with international collaborators from the EUROCJD network. The NCJDSU were founding members of the Scottish TSE Network and participate in a range of other international surveillance and research networks including NEUROPRION and NEUROCJD (the NCJDSU co-ordinated the latter which is no longer in operation). Much of the research produced by the NCJDSU is externally funded and disseminated through peer review; between 1990 and 2006 an average of 30 peer-reviewed manuscripts per annum were published by NCJDSU staff, in addition to reports, book chapters conference presentations and abstracts, and other non-peer reviewed publications.

Finally, the on-going willingness of the DH and the Scottish Executive Health Department to provide core funding to the NCJDSU further underscores the usefulness of the activities or the surveillance system and its output.

Interpretation: The activities and output from the NCJDSU are useful, contributing to providing support and care for patients and their families, advancing scientific knowledge, sharing expertise of this rare disease, increasing surveillance activities worldwide including the development of international surveillance networks, and the development of public health policy in the UK and beyond.

Representativeness

> *"A surveillance system that is representative accurately describes the occurrence of a health event over time and its distribution by place and person."* (222)

A measure of representativeness ideally requires information on the true occurrence of disease in the population. This is not available. An assessment of representativeness can however be made by examining the geographical distribution of sCJD cases adjusted for the age and sex structure of the population. As previously noted sCJD is not aetiologically linked to an exogenous exposure and the incidence of sCJD shows very little variation according to geographical location. The age adjusted incidence rate of sCJD (definite or probable cases) in men and women was calculated for each country within the United Kingdom, England, Wales, Northern Ireland and Scotland over the entire study period (1990 – 2006). A Standardised Incidence Ratio (SIR) was calculated to compare the incidence of sCJD cases in each country relative to the incidence in England, which was taken as the reference population. Given the true occurrence of sCJD is not known these analyses were also carried out for *all* suspect sCJD cases referred to the NCJDSU to assess whether there was any significant difference in referral rates of suspect sCJD cases between countries. These data are shown in Table 47.

In men and women the greatest absolute number of suspect sCJD referrals came from England, however the rate of referral per million population was highest in Scotland at 2.03 (1.58 – 2.48) in men and 2.18 (1.75 – 2.62) in women. The SIR was significantly higher in men (144.50 (114.21 – 180.36)) and women (131.11 (106.44 – 159.78)) in Scotland compared to the national average, but no different from the

national average in Wales or Northern Ireland. Examining only sCJD cases ascertained by the NCJDSU, incidence rates per million population varied very little in men (0.80 – 0.90 per million population) according to country. In women the rate was significantly lower in Northern Ireland (0.41 (0.08 – 0.74) per million population) compared to other countries. However when examining SIR in both men and women, in all countries, these were not significantly different from 100. These data indicate that whilst rates of referral of suspect sCJD were higher in Scotland, there was no difference in the incidence of sCJD according to country in the UK.

Interpretation: These data confirm that the data collected by the surveillance system are representative.

Table 47 Age standardised reporting rates of sCJD according to country, 1990 – 2006

	Country	Men			Women		
		Total Number	Rate per million	Standardised incidence ratio (95% CI)	Total Number	Rate per million	Standardised incidence ratio (95% CI)
All suspect sCJD referrals	England	664	1.64 (1.51 – 1.76)	100	691	1.62 (1.50 – 1.74)	100
	Wales	42	1.75 (1.22 – 2.29)	99.77 (71.87 – 134.89)	49	1.92 (1.39 – 2.47)	111.75 (82.65 – 147.78)
	Northern Ireland	12	0.96 (0.42 – 1.50)	79.70 (41.00 – 139.45)	13	0.90 (0.41 – 1.38)	60.76 (32.22 – 104.05)
	Scotland	78	2.03 (1.58 – 2.48)	144.50 (114.21 – 180.36)	98	2.18 (1.75 – 2.62)	131.11 (106.44 – 159.78)
sCJD cases (definite or probable)	England	366	0.90 (0.81 – 1.00)	100	380	0.89 (0.80 – 0.98)	100
	Wales	21	0.88 (0.50 – 1.25)	90.43 (55.89 – 138.33)	25	0.98 (0.60 – 1.37)	103.52 (66.93 – 152.90)
	Northern Ireland	10	0.80 (0.30 – 1.29)	123.55 (58.85 – 227.69)	6	0.41 (0.08 – 0.74)	51.10 (18.40 – 111.65)
	Scotland	34	0.89 (0.59 – 1.18)	110.54 (76.50 – 154.52)	49	1.09 (0.79 – 1.40)	118.94 (87.97 – 157.28)

*country missing for n=6, excluded from these analyses

Timeliness

> *"Timeliness reflects the speed or delay between steps in a surveillance system."*(222)

Timeliness attempts to quantify the time that each step of surveillance takes. For example, the time from symptom onset to a diagnosis of prion disease being considered, the time from a diagnosis of prion disease being considered to the suspect case being notified to the NCJDSU and the time from this notification to public health action being initiated. For vCJD, where prevention and control measures to minimise the risk of onward iatrogenic transmission are initiated following the identification of a case by the surveillance system, timeliness is an important system attribute. This is less important in sCJD. Nevertheless there is merit in examining timeliness in relation to sCJD, as this is a process measure of the performance of the surveillance system.

Typically an assessment of timeliness would quantify the time from symptom onset to public health action. This is problematic. A number of intermediate steps exist between these events, some of which were outlined in the preceding paragraph, and many of which are outside the control of the NCJDSU. For example, it is the responsibility of the referring clinical team, not the NCJDSU, to inform the local CCDC who, in turn, collects and verifies information on the suspect cases past medical history, including invasive medical procedures, blood, organ and other tissue donation. Where an invasive medical procedure has been undertaken in an individual symptomatic of prion disease or prior to a diagnosis of prion disease being made, the local CCDC will send detailed information to the CJD IP who assess the potential for onward transmission of prion disease and determine what, if any, public health action is required on a case by case basis. In practice public health action may be taken prior to this occurring, for example surgical instruments may be removed from circulation and quarantined until a decision from the CJD IP is available. Each of these steps is beyond the control of the NCJDSU and data on the time interval between steps is unavailable. In practice at the time of referral the NCJDSU collect information on potential iatrogenic routes of transmission which facilitates prompt

public health action if required. Through the National Reporting Form the local CCDC should be informed at the same time as the NCJDSU of a suspect case and initiate public health action. In reality the National Reporting Form is rarely completed and the NCJDSU neurologist provides the referring clinical team with a form to complete and forward to the local CCDC at the time of visit. It is the responsibility of the NCJDSU to inform the UKBTS of definite or probable cases of vCJD. This occurs as soon as an individual meets the diagnostic criteria, typically on the date of notification or visit. Although there are limitations to this approach, the time from symptom onset to notification and the time from symptom onset to visit by a NCJDSU neurologist can be considered a measure of timeliness. In the analyses that follow timeliness using these metrics was assessed for suspect cases that were, at the time of data censoring, classified as definite or probable sCJD and vCJD cases (denoted sCJD cases and vCJD cases). Data from selected years was examined. The results from these analyses are shown in Table 48.

Table 48 Time intervals at various steps in disease surveillance for all sCJD cases (definite or probable) referred to the NCJDSU over selected years according to vital status at the time of referral

	sCJD cases (alive at referral)			sCJD cases (deceased at referral)		
Step in surveillance	**Number**	**Median Time (IQR), days**	**P for trend**	**Number**	**Median Time (IQR), days**	**P for trend**
Onset to first seeking medical attention	189	31 (17 – 75)	0.866	54	31 (15 – 122)	0.328
Medical attention to admission	207	33 (14 – 61)	0.162	47	32 (13 – 94)	0.598
Medical attention to review by neurologist	203	40 (22 – 66)	0.119	47	38 (19 – 111)	0.382
Admission to NCJDSU referral	238	25 (13 – 44)	0.980	69	48 (30 – 95)	0.342
Neurology review to NCJDSU referral	222	21 (8 – 43)	0.111	61	45 (25 – 115)	0.017
NCJDSU referral to NCJDSU visit	224	7 (5 – 14)	0.205	58	344 (199 – 449)	0.002
Overall intervals						
Onset to NCJDSU referral	256	97 (61 – 161)	0.816	79	183 (83 – 317)	0.751
Onset to NCJDSU visit	235	117 (71 – 217)	0.847	56	509 (425 – 720)	0.138

Table 49 Time intervals for various steps in disease surveillance for vCJD cases (definite or probable) referred to the NCJDSU over selected years

	vCJD cases (definite or probable)		
Step in surveillance	**Number**	**Median Time (IQR), days**	**P for trend**
Onset to first seeking medical attention	55	109 (56 – 189)	0.732
Medical attention to admission	54	115 (53 – 192)	0.252
Medical attention to review by neurologist	59	87 (42 – 173)	0.839
Admission to NCJDSU referral	55	21 (8 – 57)	0.330
Neurology review to NCJDSU referral	60	37 (16 – 78)	0.874
NCJDSU referral to NCJDSU visit	62	11 (6 – 18)	0.143
Overall intervals			
Onset to NCJDSU referral	63	257 (211 – 332)	0.449
Onset to NCJDSU visit	62	274 (233 – 352)	0.439

sCJD cases

For sCJD cases alive at the time of referral the overall median time from symptom onset to NCJDSU referral was 97 (61 – 161) days (Table 48). Approximately a third of this time was attributable to a delay in first seeking medical attention, a third from first seeking medical attention to hospital admission/neurologist review (which ever occurred first), and a third from hospital admission to NCJDSU referral. The median number of days from NCJDSU referral to visit by a NCJDSU neurologist was 7 (5 – 14) days. This latter step accounted for approximately 6% of the reporting delay (time from symptom onset to NCJDSU visit). The time taken for each of these steps did not change over the study period.

When sCJD cases deceased at the time of NCJDSU referral were considered the overall median time from symptom onset to NCJDSU referral was 183 (83 – 317) days. The time intervals from symptom onset to first seeking medical attention and first seeking medical attention to hospital admission/neurology review were comparable to those observed in sCJD cases alive at the time of NCJDSU referral. However the interval from hospital admission/neurology review to NCJDSU referral was longer, by an average of 23/24 days respectively. The overall median time from symptom onset to NCJDSU visit was 509 (425 – 720) days with the time from NCJDSU referral to NCJDSU visit accounting for approximately 65% of the reporting delay.

vCJD cases

vCJD analyses were not stratified according to vital status at referral. The number of vCJD cases deceased at the time of referral was small and sensitivity analyses revealed that including these cases with the overall cohort did not significantly change the estimates produced. Overall the median time from symptom onset to NCJDSU visit was 257 (211 – 332) days (Table 49). Approximately 40% of this time was attributable to a delay in first seeking medical attention, 45% from first seeking medical attention to hospital admission, approximately 10% from hospital admission/neurology review to NCJDSU referral and less than 5% from NCJDSU referral to visit by a NCJDSU neurologist. There was no significant change in the

time taken for any of these steps over the study period. Of note the time intervals were longer for vCJD, reflecting the longer illness duration and less specific clinical picture at onset.

An alternate measure of timeliness can be considered as the time taken to identify a change in disease occurrence. Within two years of symptom onset in the first vCJD case in the UK and within 10 years of peak exposure of the population to the BSE agent, a further 9 cases of vCJD had been identified, without a prior case definition of vCJD, in a population of 62 million people and these data published in the peer reviewed medical press. The early detection and characterisation of vCJD and in turn the identification through the TMER study of the secondary transmission of vCJD through the transfusion labile blood components, followed by the rapid dissemination of these data to health care professionals, the public and politicians facilitated prompt public health action both nationally and internationally. The 2001 Philips report, the official enquiry into BSE and vCJD in the UK,

> "*commend[ed] the sterling work of the CJDSU team, who so promptly detected the emergence of variant CJD and so efficiently established the clinical and pathological characteristics of the disease*".(24)

Established pathways for data reporting, the content of which is predefined, and most of which involve electronic data transfer or web publishing, ensures timely dissemination of surveillance data. Senior members of the NCJDSU contribute to key committees including SEAC, SaBTO and the ACDP which have national scientific advisory and public health policy and practice remits in relation to prion disease in humans. Involvement in such committees also facilitates rapid dissemination of important or novel findings from surveillance data. Finally, the NCJDSU enjoys an excellent working relationship with the DH (core funders) and Health Protection Agencies, facilitating open lines of communication with senior decision makers where issues of national public health importance arise.

Interpretation: Despite improving diagnostic technology and increased awareness of human prion diseases among the public and health care professionals there is no evidence of a temporal reduction in the time from symptom onset to the NCJDSU

being notified of a sCJD or vCJD cases. Less than 5% of the reporting delay for vCJD cases was attributable to the NCJDSU (time from NCJDSU referral to NCJDSU visit). Corresponding figures for sCJD cases referred in life and following death were approximately 6% and 65% respectively. The latter is likely to reflect a desire for the NCJDSU to obtain other information, for example post mortem reports or medical case notes to verify a clinical diagnosis before approaching grieving relatives to request a visit to collect detailed epidemiological and clinical data. It should also be considered that there are no significant public health implications of a diagnosis of sCJD, therefore such a delay is of limited significance. In identifying the primary and secondary vCJD epidemics the NCJDSU was found to have responded in a timely fashion to facilitate public health action. Moreover routine and exception reporting of data and communication with decision makers has been found to be timely.

Stability

> *"Stability refers to the reliability (i.e., the ability to collect, manage, and provide data properly without failure) and availability (the ability to be operational when it is needed) of the public health surveillance system."* (222)

The stability of the system can be considered as the systems continued ability to capture, analyse and disseminate surveillance data despite adverse events. The ability to capture data is dependent upon willingness of patients, their families and health care professionals to participate in surveillance and the ability of the surveillance system to respond to referrals. The former was discussed under the sub-heading 'Acceptability', the latter under the sub-heading 'Flexibility'. There are multiple methods by which a referral to the NCJDSU may be made, for example fax, telephone, email and in person, therefore if one method is unavailable due to unforeseen circumstances multiple other methods are accessible. The NCJDSU endeavour to visit suspect sCJD and vCJD cases in life. In 1990 this was carried out by a sole neurologist (RGW). By 2006 this was typically carried out by one of two clinical research fellows accompanied by a research nurse. In the event that these clinicians were unavailable, the senior neurologists (RK,RGW) fulfilled this role.

Where the research nurse was unable, the research fellow and senior neurologists had sufficient training to fulfil her role. Thus over time, through expansion, the stability of the system in this respect has improved. Diagnostic services such as the CSF 14-3-3 protein laboratory and the molecular genetics service have been established over the period studies. For these services adequate staffing to ensure continuity of the service, for example in the event of an extended absence from work, is available. Surveillance data are collected in writing and held in paper based files in the NCJDSU archives. Limited data, a minimum monitoring dataset and data relating to the case-control study, are entered onto an electronic database for analysis. Over time the volume of data held by the NCJDSU has increased, both numbers of cases and the amount of information collected per case; the NCJDSU investigation protocol ensures the collection of detailed clinical data extending far beyond the minimum monitoring dataset. To date there have been no instances of data loss from paper records, for example through flooding, however the potential exists for large volumes of data to be lost if such an event occurs. In the absence of a computerised backup there exists the potential for a catastrophic loss of data. In addition, the limited amount of clinical data held electronically limits the NCJDSU's ability to interrogate and analyse this rich data source beyond predetermined data reporting. The establishment of reporting pathways, as previously outlined, many of which utilize electronic data transfer, and the publishing of monthly figures on the NCJDSU website, ensures rapid dissemination of information to the public, media, health care professionals and policy makers. The NCJDSU actively engage in international surveillance and research networks, fostering opportunities to share expertise.

Interpretation: There is evidence of the stability of the NCJDSU increasing over time despite increasing demands. A lack of computerized data archiving is a potential threat to the stability of the system which should be addressed.

Summary of key findings

- The PHS system in the UK, though simple in design, is operationally complex.
- Overall the PHS system was found to be flexible in responding to a number of changing demands and stable over time.
- Overall, the quality of the data collected by the NCJDSU was good, with improvement in the validation of reported clinical, epidemiological and diagnostic data over time, and low rates of loss to follow up.
- The activities and outputs from the system were found to be acceptable and useful to patients, families, health care professionals and public health decision makers, and timely.
- Whilst the system was found to have a high sensitivity, improving PPV and collect representative data, this was undermined by evidence of sub-optimal, and differential, use of diagnostic investigations.
- Coupled with an increasing reliance of clinical diagnostic criteria due to falling autopsy rates this latter finding is of concern.
- An increasing PPV of the system in the face of falling referral rates of suspect cases may also be of concern as this may compromise the NCJDSU's ability to detect atypical disease phenotypes or novel prion diseases.

Discussion

In this chapter I reported the findings from the first ever evaluation of the National CJD Surveillance system in the UK. Here I discuss the key findings from this evaluation.

The quality of a diagnosis of sCJD or vCJD

Overall the PHS system was found to have performed well, meeting many of its stated objectives. However, the under-utilisation of diagnostic technology, particularly CSF 14-3-3 protein and EEG which are integral to the clinical diagnostic criteria in suspect sCJD, is cause for concern. The use of these investigations was sub-optimal in the assessment of all suspect sCJD cases, but more so in non-sCJD cases than cases. Resultantly, some non-sCJD cases will have failed to meet the diagnostic criteria because they did not undergo appropriate investigation. Combined with falling rates of post mortem examination, this may have resulted in under-ascertainment of sCJD cases.

An examination of the sensitivity of the surveillance system should provide some insight into whether this was the case. Unfortunately a direct measure of the sensitivity of the system was not available. Instead an indirect measure, sCJD incidence rates reported in the UK compared to those reported internationally, was selected. The incidence of sCJD has increased steadily over time in the UK and is in keeping with internationally reported rates. As post mortem rates have fallen the increasing sCJD incidence has been driven by a rising proportion of probable cases that have met the diagnostic criteria based on CSF 14-3-3 protein examination. It should however be considered that each ante-mortem diagnostic investigation has a false positive rate. For CSF 14-3-3 protein this has been quoted as up to 16%.(231) Reliance on clinical diagnostic criteria rather than case confirmation at autopsy will lead to misclassification of a small number of cases. The overall effect on annual sCJD incidence rates might be minimal, with the inclusion of non-cases compensating for the exclusion of cases.

In suspect vCJD there was evidence of under-utilisation of MRI in non-cases which may have a similar effect, although this was much less significant than for EEG and CSF 14-3-3 protein in sCJD, and by the end of the study period virtually all suspect vCJD cases underwent MRI scanning.

The proportion of suspect sCJD and vCJD cases undergoing *PRNP* Codon 129 genotyping and mutation testing was low with no evidence of improvement over time and again with evidence of differential use of these investigations according to case classification. This may have resulted in under-ascertainment of genetic prion disease cases which can be phenotypically indistinguishable from other aetiological prion diseases.

Falling post mortem rates also have implications for the PPV of the system. A low PPV may suggest that the system is not performing adequately. This is not necessarily the case. The WHO recommend that referrals to the system should exceed confirmed cases by a factor of two, to facilitate the detection of cases with unusual disease phenotypes that do not have sufficient clinical features to meet the clinical diagnostic criteria. In the UK referral criteria are broad to ensure this occurs. The significant increase in the PPV observed, in the face of falling referral rates at first glance may indicate increasing efficiency of the system at detecting cases. However this may also be cause for concern, if in achieving a high PPV, the sensitivity of the system has been compromised.

It has not been possible to formally assess the impact of increasing PPV on the sensitivity of the surveillance system with a direct measure. The ratio of neuropathologically confirmed to suspected CJD cases referred to the NCJDSU has been maintained at or below the level recommended by the WHO, despite falling referral rates in the latter period of the study. This has occurred as a result of falling post mortem rates leading to a reduction in the proportion of all referrals accounted for by neuropathologically confirmed cases, whilst the PPV of the system has increased as a result of the increasing ascertainment of probable sCJD cases based on CSF 14-3-3 protein examination. As outlined in the preceding passages, careful

consideration should be given as to whether this is appropriate, particularly in the face of falling referral and case confirmation rates.

A tension exists between the desires of the surveillance system and the clinical realities. Ideally all suspect CJD cases would undergo all investigations that might support a diagnosis of CJD, not solely to exclude other potentially treatable conditions and obtain a definitive clinical diagnosis, but also to inform surveillance practice. For example, to provide information about the validity of diagnostic tests such as CSF 14-3-3 protein and the diagnostic criteria into which they are incorporated. However, the NCJDSU does not directly manage patients with suspect CJD. Clinical decision making and patient management is retained by the referring clinical team. The decision to pursue a single or series of diagnostic investigations will be made by the local clinical team in consultation with the patients' significant others. Many factors will influence these decisions. For example a patient's clinical condition may negate the use of certain investigations. It may not be possible to obtain MRI images due to patient agitation. Invasive investigations such as lumbar puncture to obtain CSF, tonsil or brain biopsy in life may be declined by the patient's relatives on compassionate grounds, or these invasive investigations may prove technically difficult. Locally, there may be limited access to certain diagnostic technologies such as EEG or MRI. Accessing such investigations might require patient transfer to another facility which may be deemed inappropriate in the terminal phases of illness. CSF 14-3-3 protein, *PRNP* Codon 129 genotyping and mutation testing are only available from the NCJDSU or the NPC; centralization of these services ensures the validity of the investigations. These services are freely available and accessible remotely from throughout the UK therefore accessibility should not be a major issue. However other issues may determine the use of these services. For example a relative may refuse *PRNP* testing for mutations as a positive result may have implications for that individual and their family. It should also be considered that a clinician may decide not to pursue further investigations such as CSF 14-3-3 protein in a patient with a classic clinical course and a typical EEG if they feel that these features are sufficient to make a firm clinical diagnosis, or in a moribund patient in whom consent for post mortem examination on expiration has

been provided. Additional factors that may have contributed to falling post mortem rates in the UK were reviewed in the discussion in the preceding chapter. Improving the quality of diagnosis in suspect CJD therefore requires the engagement and agreement of patients, their relatives and health care professionals. It should be considered that aggressive pursuit of diagnostic investigations may have the unintended consequence of compromising the acceptability of the NCJDSU to referrers.

Other aspects of data quality

Several other aspects of data quality were assessed. Overall data quality was found to be very good, with low levels of missing data, low rates of loss to follow up and increasing levels of validation of clinical, diagnostic and epidemiological data. However areas for improvement were identified. For example, EEGs were reviewed in only two thirds of sCJD cases with evidence of review in a declining proportion of sCJD cases since 2000. Whilst EEGs are requested on suspect cases referred to the NCJDSU, referrers are under no obligation to provide these investigations. It is possible that referrers may not fully understand the importance of central review of EEGs for surveillance purposes and therefore be reluctant to send copies of EEGs that do not contribute to the diagnostic classification of the patient. For example in the case of an individual that has already met the diagnostic criteria based on clinical features and CSF 14-3-3 protein. Further issues relating to evaluation of EEG in the surveillance of sCJD will be discussed in greater detail in the Chapter that follows. The proportion of sCJD cases for which medical case notes from primary care were reviewed was also particularly low, although there was evidence of improvement over time. Accessing medical records can be challenging without specific legal authority to do so. Records may have been damaged or destroyed, or local health authorities may be reluctant to release them, unaware of the importance of the NCJDSU reviewing these. Further qualitative work may help elucidate the issues that have contributed to the poor performance in these two areas.

Stability

The NCJDSU collects a wealth of clinical, diagnostic and epidemiological data on CJD cases and suspect cases. However only a minority of these data are

electronically archived, the majority are paper based records. In many respects this is inefficient as it limits the NCJDSU's ability to quickly interrogate the data. In addition lack of computerized data archiving poses a potential threat to the stability of the system which should be addressed.

Flexibility

Overall the system was found to be flexible to a number of changing demands, including most significantly the emergence of vCJD. A fall in the proportion of sCJD cases reviewed by a NCJDSU neurologist in 1997 and non-sCJD cases in 1997 and 2000 was observed. This may indicate that the system was struggling to respond to a significant increase in demand. However reassuringly this did not impact on the NCJDSU's ability to review suspect vCJD cases or compromise timeliness in this respect. Given there are few public health implications of a diagnosis of sCJD, it could be argued that the system responded appropriately. The system was further able to respond to increasing demands for new diagnostic technologies through rapid expansion, including the introduction of diagnostic laboratories such as the national CSF 14-3-3 protein service, molecular genetics and protein biochemistry services. These expansions have successfully occurred without compromising stability.

Timeliness

The relative importance of timeliness depends upon the condition under surveillance. There are few public health implications of a diagnosis of sCJD. There are a small number of recognised routes of iatrogenic transmission of sCJD, most of which are now of historical importance only. This is not the case for vCJD. Iatrogenic transmission of vCJD via the transfusion of labile blood components has been documented. Due to the pathogenesis of vCJD and the inherent difficulties in decontaminating surgical instruments, instruments that have been used on vCJD cases, in both the symptomatic and asymptomatic phases of illness, may present a risk to others. In such cases prompt public health action is required to minimise the number of individuals exposed to potentially contaminated labile blood components or medical instruments and to inform those who have been exposed that they may be 'at risk' of vCJD such that they can, in turn, take appropriate precautions. An assessment of the timeliness of the NCJDSU is challenging because many of the

steps that occur prior to reporting are beyond the control of the NCJDSU; following reporting many of the steps that lead to public health action are also beyond the control of the NCJDSU. In this study a measure of the reporting delay was taken as the time from symptom onset to NCJDSU referral. Despite improving diagnostic technology and increased awareness of human prion diseases there was no temporal reduction in the time from symptom onset to NCJDSU referral. Over a third of the delay in reporting was due to delay in seeking medical attention with a similar period due to a delay in moving from primary to secondary care. This reflects the fact that symptom onset can be insidious and non-specific in nature. The remainder of the delay occurred between neurology review or hospital admission and NCJDSU referral, reflecting the time taken to reach a diagnosis. It is disappointing that this has not improved over time. As therapeutic treatment options begin to emerge the early identification of cases will become increasingly important.

Acceptability

The NCJDSU performed well on an overall assessment of acceptability. However it should be noted that referrers engaged poorly with the National Reporting Form. In theory, this form should stream-line the referral process, allowing referrers to inform the Unit, local CCDC and NPC simultaneously. It may be that referring clinical teams prefer to speak directly to a clinical colleague in the NCJDSU to discuss the differential diagnosis in a complex neurological case, rather than completing a form which has to be faxed to various external agencies. Having done so, they may feel that completion of the form, whilst requested, is unnecessary duplication of effort and does not appear directly relevant to the care of their patient. Others may find the prospect of having to visit a website to download the form and in turn fax it to 3 different agencies a time-consuming and low priority task in a busy schedule. There may be additional issues in accessing computing and facsimile facilities. Limited use of the National Reporting Form has not had a significant impact on referrals to the NCJDSU as a result of the continued willingness of the NCJDSU to accept referrals in other formats. Indeed increasing use of the National Referral Form may have the unintended consequence of reducing timeliness for the reasons highlighted above.

An examination of sCJD cases deceased at the time of referral to the NCJDSU revealed that the majority would have met the clinical diagnostic criteria as a possible or probable case of sCJD in life. In over 95% of those who would have met the clinical diagnostic criteria in life a diagnosis of sCJD had been considered in life; in a further group, referral to the NCJDSU had been recommended but was not followed through by the clinical team. There remain a small number of sCJD cases in whom a diagnosis of sCJD is suspected in life that are not referred to the NCJDSU, although this number is diminishing. Qualitative research may assist in determining the barriers to referral to the NCJDSU in this group.

Usefulness

Perhaps one of the most important system attributes examined was usefulness. In its activities and outputs the NCJDSU was found to have furthered scientific knowledge through national surveillance, closely linked to research activity, and facilitated the rapid transfer of knowledge to public health practice and policy, in addition to making substantial contributions to international disease surveillance and research. In many respects this is illustrated by central governments continued commitment to providing core funding for the NCJDSU.

Evaluation design

In this chapter I applied a widely recognised framework for the evaluation of public health surveillance systems.(222) PHS systems are diverse in their methodologies and objectives. The system attributes most relevant one system may not be relevant to another. In this evaluation the attributes I considered highest priority were data quality, sensitivity, flexibility, acceptability, timeliness and usefulness. It should be noted that this approach requires a subjective assessment of system attributes. A formal grading is not applied to the assessment of each system attribute to aid interpretation of evaluation findings. Rather the evaluator makes a subjective overall judgment as to whether the system is performing adequately and fit for purpose.

Many system attributes are interdependent. For example, if the quality of diagnosis is poor (data quality) then this will affect the ability of the system to detect disease, reducing sensitivity. If the system is unacceptable cases will not be referred which

will have a negative impact on sensitivity, PPV and representativeness. Improvement in one system attribute may compromise another. It is important to consider this in interpreting evaluation findings and making recommendations. For example, in the UK surveillance is both labour and time intensive. The lack of a single sensitive and specific ante-mortem diagnostic test, the diverse disease phenotype and specific requirement to identify novel clinico-pathological expressions of prion disease, necessitate broad referral criteria, the review of suspect cases in life and the collection and validation of extensive clinical and epidemiological data in order to maximise the sensitivity, PPV and representativeness of surveillance data. However this has resulted in an intrinsically simple system becoming operationally complex.

Where possible this evaluation assessed multiple metrics for each system attribute in an attempt to capture different facets of each attribute. For example, an assessment of data quality included a measure of missing data which is a simple assessment of the completeness of data recording that says little about the validity of the data collected. To assess the latter, the review of clinical, epidemiological and diagnostic information to validate data collected by the surveillance unit was also examined. The quality of the diagnosis of CJD is dependant, in part, upon the completeness of diagnostic investigation. This is an indirect measure of data quality but is nonetheless crucial to assess because it will influence the sensitivity and PPV of the system.

To evaluate some system attributes it was necessary to use indirect measures. For example, assessment of sensitivity and representativeness requires knowledge of the true number of prion disease cases occurring in the population. This is not known. In the UK the surveillance system utilises a range of data sources to maximise case ascertainment. Resultantly an external measure of the incidence of prion disease against which the NCJDSU could be assessed was not readily available. A number of retrospective studies have attempted to quantify whether there has been any systematic under-ascertainment of CJD cases in the UK. For example Majeed *et al* reviewed the medical case notes of patients identified from death certificates as dying from selected neurological conditions in England and Wales between 1979 and 1996.(248) The authors determined that no additional cases of sCJD or vCJD were

missed by the surveillance system in this sample. Hillier *et al* went further in re-examining neuropathological material, where available, from individuals aged 15 – 45 years old who had died in Wales between 1985 and 1995.(249) No cases of vCJD prior to the first cases being ascertained by the NCJDSU were identified. From 1998 to 2007, Piccardo *et al* undertook a retrospective review of neuropathological material from sCJD cases and atypical dementias dating back to the 1970s to determine whether vCJD cases had been missed.(250) This national study did not identify missed cases of vCJD but did identify a number of sCJD cases that had been misdiagnosed as atypical dementias. These studies are both labour and time intensive and not without limitation. The limitations of death certificate data in the absence of case confirmation will be discussed in detail in Chapter 5; retrospective examination of neuropathological material determines the sensitivity of a neuropathological diagnosis but does not determine the true incidence of disease, particularly in light of falling autopsy rates. I used readily available surveillance data to assess sensitivity. For example, a key objective of the surveillance system was the detection of vCJD. The system has therefore demonstrated that it is sensitive, having identified and characterised vCJD prior to a case definition being available. Two further measures of sensitivity were considered: the comparability of incidence/mortality rates of sCJD relative to internationally described rates and an assessment of the sensitivity of the clinical surveillance system at detecting sCJD and vCJD cases in life. The validity of these measures is not known.

Comparison with the literature

Two evaluations of CJD surveillance systems have utilised the CDC evaluation framework; both used indirect or proxy measures for the assessment of some system attributes. Robotin assessed the sensitivity of the Australian surveillance system by comparing rates of sCJD ascertained by the system to international rates, using a similar approach to the present study to assessing sensitivity.(233) In an examination of the EUROCJD network Pedro-Cuesta, developed a number of proxy indicators against which to assess sensitivity, including referral rates in those under 50 years of age and the genotypic profile of sCJD cases under 50 years of age.(231) These metrics reflected the overall aims of the surveillance network which were to identify vCJD and any change in the phenotype of sCJD in the young. This evaluation was

aided by an external data source, a central shared data repository to which collaborators reported data, and against which the contribution of individual countries could be assessed. In addition, in some analyses the UK was used as a reference population for comparison. These approaches were not possible in the present study and the metrics selected in the present study may not be valid in other evaluations. For example, vCJD has not been described in Australia, therefore the ability to detect an epidemic of vCJD could not be used as a measure of sensitivity, or the ability of the system to respond to this epidemic as a measure of flexibility in the study by Robotin.(233)

Strengths and limitations

There are a number of other limitations to this evaluation that should be considered. The main aim of this evaluation was to assess the performance of the system against its stated objectives. Beyond NCJDSU staff, external stakeholders were not identified and engaged in the process. The evaluation used quantitative data collected by the system against which the performance of the system was evidenced. These data were selected because they were readily available and accessible within the time frame (and resource) of this research and it had been prospectively, systematically collected by valid and reliable methods. For many analyses data from selected years, at 3 yearly intervals, were examined rather than all 16 years over which systematic prospective surveillance has been carried out. This approach was adopted for pragmatic reasons owing to the volume of data collection required and the need for manual data extraction from paper-based case records. The years selected were strategically identified, for example 1991 was the first full year of systematic prospective surveillance in the UK, 1994 the year before the first case of vCJD was described, 1997 the year following characterisation of vCJD, 2000 the year that CSF 14-3-3 protein was incorporated into the WHO diagnostic criteria for sCJD and 2006 the year that I was employed as a Research Registrar in Neurology by the NCJDSU and personally visited suspect cases to collect surveillance data. As previously noted, for some attributes it would have been desirable to examine external data and it may also have been useful to supplement these quantitative data with qualitative data to provide greater insight into the quantitative findings. Only limited data were available on iCJD and genetic prion disease cases against which to assess the

performance of the system therefore minimal analyses were carried out using these data. Finally, there is a genuine paucity of published studies of this nature against which to compare and contrast the validity of the evidence collected in this study to examine key system attributes and the subjective interpretation of the study findings.

Recommendations

Based on the findings of this evaluation the following recommendations are made:

- The differential use of investigations that support a diagnosis of sCJD or vCJD in sCJD or vCJD cases and non-cases warrants further investigation to determine whether specific groups, for example the elderly, are being systematically under-investigated and whether this may have contributed to the systematic under-ascertainment of cases.
- Barriers to the use of investigation that support a diagnosis of sCJD or vCJD in suspect sCJD or vCJD cases, particularly post mortem examination, should be explored with key stakeholders including relatives, health care providers and special interest groups to inform the development of strategies to facilitate the use of such technologies.
- Consideration should be given to whether a rising PPV of the system is desirable in the context of falling rates of referral of suspect sCJD and vCJD cases to the NCJDSU and an increasing reliance on clinical diagnostic criteria.
- Similarly consideration should be given as to whether there is a need for further contemporary studies, for example a capture-recapture study, to more directly examine sensitivity in light of changing PPV and the falling data quality of diagnosis reported.
- All surveillance data should be electronically archived to protect against data loss and ensure the ability to rapidly interrogate and analyse the rich surveillance data available to the NCJDSU
- Qualitative research should be considered to explore barriers to referral to the NCJDSU in the minority of cases in whom CJD is considered in life but referral to the NCJDSU is not made.

Regular evaluation of the NCJDSU should follow to ensure that the system is continues to meet it stated objectives and as part of an on-going process of quality improvement. The engagement of key stakeholders in this process is crucial. The dearth of published literature in this area is alarming given the rapid international expansion of CJD PHS system. The NCJDSU should endeavour to publish the output of regular evaluations of the system to inform practice, and use its considerable influence to encourage international collaborators to undertake regular evaluations of the performance of their systems.

Conclusions

In this Chapter, I presented the findings of the first ever evaluation of the NCJDSU in the UK. Over 16 years of prospective systematic surveillance the NCJDSU has performed well in meeting many of it stated objectives. However, falling post mortem rates and sub-optimal, and differential, use of investigations that support a diagnosis of sCJD or vCJD in suspect sCJD and vCJD cases are cause for concern. Falling rates of referral of suspect prion disease cases in the context of an increasing PPV of the system may compromise the NCJDSU's ability to detect atypical or novel prion diseases.

Chapter 4. Prospective validation of NCJDSU operational criteria for the assessment of electroencephalography (EEG) in suspect sCJD

Introduction

Reliable diagnosis of suspect CJD in life is important. This allows clinicians to exclude potentially treatable differential diagnoses, inform relatives of the likely prognosis and facilitates prompt public health action where necessary. However brain biopsy carries risk and rates of post mortem among suspect CJD cases in the UK are falling. In the absence of neuropathological confirmation, presumptive diagnosis of sCJD relies on a classical clinical history and characteristic findings on EEG (PSWC) and/or the detection of CSF 14-3-3 protein.(98) Whilst the validity of a CSF 14-3-3 protein test reported by a centralised national service can be assured, the same cannot necessarily be said for EEGs which are carried out and reported in medical centres across the UK. Given the importance of the EEG in diagnostic classification of suspect sCJD central review of EEGs by the PHS system is considered essential in the UK. Quantitative criteria for the assessment of EEG in suspect sCJD have been adopted by the WHO for surveillance purposes (Table 6). As previously noted there are practical difficulties in obtaining and accessing digitalised EEG recordings in the UK which have limited the use of these criteria in disease surveillance. Operational criteria are used by the NCJDSU in the assessment of EEG in case classification of suspect sCJD; these never been prospectively validated.

Aim

The aim of this study was to prospectively validate the NCJDSU operational criteria for the assessment of EEGs in case classification of suspect sCJD.

Methods

The surveillance protocol

The surveillance protocol has been described in detail in the preceding chapters of this thesis. Further details here are limited to those aspects directly relevant to the evaluation of EEGs. In the UK, representative samples from all EEGs carried out in the investigation of suspect CJD cases referred to the NCJDSU are requested for review. These are evaluated by one of two experienced clinicians (RGW, RK). In the early phase of disease surveillance a system of EEG classification was developed based on defined descriptive criteria which were employed by the same two individuals who have reviewed the EEGs in this study. This classification comprises of 5 broad categories (Table 50).

Table 50 Subjective criteria employed by the NCJDSU for EEG classification

Normal	Normal EEG
Non-specifically abnormal	Deterioration of normal background rhythms but essentially non-specifically abnormal.
Suggestive	Deterioration of normal background rhythms with the emergence of bi- or tri-phasic discharges, at times with some periodicity but only for short periods and not truly generalised
Highly suggestive	Marked deterioration, or absence of, of normal background rhythms. Periodic bi- or tri-phasic discharges, for longish segments, but not continuous throughout record and/or not always truly generalised.
Typical	Absence of normal background rhythms. Continuous, generalised, periodic bi- or tri-phasic discharges.

Within this scheme there is potential for overlap between categories, especially at the suggestive/highly suggestive boundary, and to a lesser extent at the highly

suggestive/typical boundary. This classification of EEGs was carried on into the more recent phase of surveillance but it did not necessarily map directly onto case classification as the case diagnostic criteria developed and changed. In general, normal, non-specific and suggestive EEGs were not considered to support elevation of a suspect sCJD case from possible to probable sCJD; typical EEGs were. The area of uncertainty concerned highly suggestive EEGs. Some were so highly suggestive, even if not absolutely typical, that they were used to support the diagnosis of probable sCJD; others were felt not to be suggestive enough. Whilst this does introduce a loosely defined and subjective element to the assessment of EEG in case classification, it was based very firmly on the extensive experience of RGW and RK, who had developed their judgments based on the outcome of previous case assessments. In this study, the decisions concerning EEG classification were based on the original 5 broad categories. Decisions concerning the use of the EEG in case classification were based on the principles outlined in the paragraph above.

Evaluation of EEGs

This study included all consecutive suspect sCJD cases referred to the NCJDSU between 1st January 2005 and 31st December 2006. All suspect sCJD cases were followed for a minimum of 2 years. Data were censored at 31st December 2008. Final case classifications at data censoring were used in these analyses. All sample EEGs received by the NCJDSU were anonymised and blindly reviewed by two independent clinicians (RGW, RK). The clinicians were not provided with any clinical data and EEGs were reviewed in a random order. The aim of this study was to prospectively validate the operational criteria used for the assessment of EEG for case classification in suspect sCJD. The clinicians were asked (Yes/No) whether would they use the EEG in case classification, meaning would they use the EEG to change case classification from possible to probable sCJD. In addition, the clinicians were asked to classify the EEG using the categories described in Table 50. The purpose of this was three fold (1) to assess the degree of inter-observer variation in the classification of EEGs using these criteria (2) to explore how well the criteria used in the EEG classification mapped to case classification in practice (3) to aid interpretation of any disagreement between reviewers in case classification. One clinician re-reviewed all EEGs to allow examination of intra-observer variance of

assessment of EEG for case classification. This reviewer was blinded to his previous grading and received the EEGs in a different order a minimum of one month after his initial review. At the time of this second review the clinician was asked whether he would use the EEG in case classification only.

Definitions

Studies of this nature typically consider the investigation as the unit of analysis, rather than the individual. Suspect sCJD cases often undergo multiple EEG examinations therefore disagreement between reviewers on the evaluation of one EEG may not in practice have an impact on overall case classification. In this study analyses were first carried out using EEGs as the unit of analysis. Multiple EEGs were considered per suspect case. Each EEG was considered as an independent observation. Analyses then considered the individual as the unit of analysis. If a suspect case was considered by a reviewer to have had an EEG at any stage in their clinical illness that could be used in case classification this individual was counted once as having an EEG used in case classification. Conversely, if the individual, despite serial investigations, did not have an EEG at any stage in their clinical illness that could be used in case classification, this individual was counted once as not having an EEG that could be used in case classification.

An assessment of the diagnostic value of a test (sensitivity, specificity, PPV and NPV) requires comparison of the performance of the test against a gold standard. Sensitivity was defined as the proportion of sCJD cases that had one or more EEG(s) which could be used for case classification. Specificity was defined as the proportion of non-cases that did not have any EEGs that could be used for case classification. PPV was defined as the proportion of suspect cases that had one or more EEGs that could be used for case classification who were sCJD cases and NPV the proportion of individuals that did not have any EEGs that could be used in case classification that were non-cases.

Definite diagnosis of sCJD requires neuropathological examination of tissue obtained from brain biopsy in life, or more commonly post mortem following death. In practice however a significant proportion of suspect sCJD cases do not undergo post

mortem examination or brain biopsy and clinical diagnostic criteria, based on clinical features and supportive investigations (EEG or CSF 14-3-3 protein), are applied that allow classification of suspect sCJD cases with an extremely high degree of diagnostic certainty. The use of clinical diagnostic criteria as a gold standard against which to assess the diagnostic utility of EEG is flawed because EEG are included in the clinical diagnostic criteria. For the purposes of this study two definitions of sCJD were considered. A narrow definition considered only individuals with a neuropathologically confirmed diagnosis of sCJD and neuropathologically confirmed non-cases (classifications 1.0 and 4.3 respectively). A broad clinical definition considered sCJD cases as meeting the WHO diagnostic criteria as a definite or probable case and non-cases as individuals that did not meet the WHO diagnostic criteria as a definite, probable or possible case (classifications 1.0, 2.0 or 3.0 and classifications 4.1, 4.2 or 4.3 respectively).

Statistical analysis

The median age at symptom onset and sex distribution of all suspect sCJD cases referred to the NCJDSU were described. The Wilcoxon Ranksum test was used to examine differences in median age at symptom onset between suspect cases for which EEG examination(s) were, and were not, available for review; Chi^2 test (Fisher's exact where assumptions were violated) were used to compare the sex distribution. Where a suspect sCJD case had undergone EEG examination and this was available to the NCJDSU for review, age at symptoms onset, median illness duration, the median number of EEGs per suspect case, the time to first EEG from symptom onset and time from last EEG to death were described. The Wilcoxon Ranksum test was used to compare these variables according to case classification and among suspect sCJD cases according to whether EEG was used in case classification at any stage in the clinical illness by either reviewer; Chi^2 test (Fisher's exact where assumptions were violated) were used to examine the sex distribution according to case classification. Further univariate analyses using these parametric and non-parametric tests as appropriate were carried out to determine whether the baseline characteristics and clinical features of sCJD cases (both broadly and narrowly defined) differed according to whether the EEG was or was not used in case classification.

Intra and inter-observer variance was assessed using the overall percentage agreement. A Kappa statistic was used to determine the intra and inter-observer agreement beyond that which would have arisen due to chance alone. A Kappa statistic based on exact agreement only acknowledges agreement or disagreement, without commenting on the extent of agreement or disagreement. Whilst this was appropriate to examine intra and inter-observer agreement in case classification (a dichotomous variable), it was not appropriate to examine intra and inter-observer agreement in the descriptive EEG classification which used ordered categorical data. If Reviewer 1 classified an EEG as typical whilst Reviewer 2 classified the same EEG as normal the extent of disagreement would be greater than if Reviewer 1 had classified the EEG as typical whilst Reviewer 2 classified the EEG as highly suggestive according to this criteria. To take this into account the Kappa statistic was weighted when analysing data on the classification of EEGs. Weights were pre-specified based on the following calculation: $1- |i-j| / (k-1)$, where i and j index the rows and columns of the ratings and k is the maximum number of ratings. Thus where there was complete agreement in EEG classification a weight of 1.0000 was applied and where there was complete disagreement (i.e. one Reviewer classified an EEG as typical and another as normal) a weight of 0.0000 was applied. Where there was disagreement between Reviewers in adjacent categories (i.e. typical and highly suggestive) a weight of 0.7500 was applied, where there was disagreement between Reviewers of two categories (i.e. typical and suggestive) a weight of 0.5000 and where there was disagreement between Reviewers of three categories (i.e. typical and non-specific) a weight of 0.2500 was applied. The Kappa statistic can take any value between + 1.0 (indicating complete agreement) and -1 (indicating complete disagreement). To aid interpretation I adopted the Landis and Koch classification (251) as follows: A Kappa statistic of <0.00 indicates no agreement, 0.00 - 0.20 slight agreement, 0.21 - 0.40 fair agreement, 0.41 - 0.60 moderate agreement, 0.61 - 0.80, substantial agreement and ≥ 0.81 almost perfect agreement. For each reviewer corresponding values for sensitivity, specificity, PPV and NPV were calculated. All statistical analyses were carried out using STATA (Version 11, Stata Corp.). A significance level of 0.05 was used throughout.

Results

Study population

From 1st January 2005 through 31st December 2006, 180 suspect sCJD were referred to the NCJDSU; 141 (78.3%) were known to have undergone an EEG examination during the course of their clinical illness. EEG was available for review at the NCJDSU for 108 (76.6%) suspect cases. Suspect cases in which EEG was unavailable for review comprised of 13 neuropathologically confirmed sCJD cases, 3 probable sCJD cases (based CSF 14-3-3 protein) and 17 non-cases (2 neuropathologically confirmed, the remainder had insufficient clinical features to meet the diagnostic criteria). This group did not differ from those suspect cases for whom an EEG was available for review with respect to age at onset (P=0.33) or sex (P=0.09). The baseline characteristics of suspect sCJD cases for which an EEG was available to the NCJDSU for review are outlined in Table 51.

In total, the two clinicians independently evaluated 166 EEGs from 108 suspect sCJD cases. This final sample included 87 sCJD cases (52 definite and 35 probable), 3 possible sCJD cases and 18 non- cases (10 pathologically confirmed and 8 with an alternate clinical diagnosis). In six probable sCJD cases, case classification was based on EEG alone. Overall the median number of EEGs per suspect sCJD case was 1 (range 1 – 4). Overall the time from symptom onset to first EEG was 96 days (57 – 172) and from last EEG to death was 31 days (17 – 50).

Table 51 Characteristics of suspect sCJD cases referred to the NCJDSU between 2005 and 2006 according to case classification

		Narrowly defined			Broadly defined		
	All suspect cases	**sCJD**	**Non-case**	**P value**	**sCJD**	**Non-case**	**P value**
Number	108	52	10		87	18	
Age at symptom onset,	69.0	70.5	65.0	0.287	70.0	65.5	0.514
Median years (IQR)	(61.0 – 77.0)	(61.0 – 78.0)	(62.0 – 66.0)		(61.0 – 77.0)	(62.0 – 70.00	
Male, n (%)	56 (51.9)	44 (58.7)	13 (59.1)	0.586	47 (54.0)	8 (44.4)	0.459
Illness duration,	134	122	376	<0.001	128	414	<0.001
Median days (IQR)	(95 – 231)	(91 – 188)	(219 – 1132)		(88 – 183)	(225 – 937)	
Number of EEG,	1 (1 – 4)	1 (1 – 4)	2 (1 – 4)	0.016	1 (1– 4)	1 (1 – 4)	0.274
Median (range)							
Time onset to first EEG,	96	92	299	0.020	86	229	0.002
Median days (IQR)	(57 – 172)	(52 - 177)	(124 – 615)		(46 – 150)	(119 – 615)	
Time last EEG to death,	31	30	86	0.024	31	91	0.034
Median days (IQR)	(17 – 50)	(15 – 53)	(20 – 298)		(16 – 48)	(30 – 299)	

Narrowly defined: sCJD case classification 1.0, non-case classification 4.3; broadly defined sCJD case classification 1.0 or 2.0, non-case classification 4.1, 4.2 or 4.3

Inter-observer variance in the classification of EEGs

Overall the percentage agreement between reviewers was 91.9%, with a weighted Kappa statistics of 0.681, indicating substantial agreement between reviewers in classifying EEGs using the descriptive criteria (Table 52).

Table 52 Agreement between reviewers examining all EEGs from all suspect sCJD cases using descriptive criteria for EEG classification

		Reviewer 2				
		Typical	**Highly suggestive**	**Suggestive**	**Non-specific**	**Normal**
Reviewer 1	**Typical**	5	7	0	0	0
	Highly suggestive	2	14	8	2	0
	Suggestive	0	10	31	8	0
	Non-specific	0	0	7	63	7
	Normal	0	0	0	1	1

Inter-observer variance in the assessment of EEG for case classification

The overall percentage agreement between clinicians in the evaluation of EEG for case classification was 89.2% when EEGs were considered the unit of analysis (Table 53). The Kappa statistic was 0.675, indicating substantial agreement. The clinicians disagreed on whether the EEG should be used for case classification in 18 EEGs (18 individuals). This group was comprised of ten neuropathologically confirmed sCJD cases, one neuropathologically confirmed non-case and seven probable sCJD cases; in the latter case classification was based on EEG alone in three cases. Limiting analyses to only EEGs from sCJD cases and non-cases (narrowly and broadly defined) made little difference to the degree of inter-observer variation or Kappa statistic.

When individuals rather than EEGs were considered the unit of analysis, the percentage agreement between reviewers and the Kappa statistics were slightly lower. This was of significance only when analyses were limited to individuals with

a neuropathological diagnosis. For this group the percentage agreement fell from 88.7% (EEG) to 82.3% (individual) with a corresponding fall in the Kappa statistics from 0.617 (EEG) indicating substantial agreement to 0.546 (individual) indicating moderate agreement. The clinicians disagreed on whether the EEG could be used for case classification in 16 individuals; ten neuropathologically confirmed sCJD cases, one neuropathologically confirmed non-case and five probable sCJD cases. In the latter, case classification was based on EEG alone in two cases.

Table 53 Inter-observer variance in the evaluation of EEG for case classification

		Unit of analysis	
		EEG	**Individual**
All suspect sCJD cases	**Agreement, n**	148	92
	Disagreement, n	18	16
	Percentage agreement, %	89.2	86.7
	Kappa Statistic	0.675	0.681
	P Value	<0.001	<0.001
Narrowly defined sCJD cases and non-cases	**Agreement, n**	86	51
	Disagreement, n	11	11
	Percentage agreement, %	88.7	82.3
	Kappa Statistic	0.617	0.546
	P Value	<0.001	<0.001
Broadly defined sCJD cases and non-cases	**Agreement, n**	143	88
	Disagreement, n	18	16
	Percentage agreement, %	88.8	84.6
	Kappa Statistic	0.672	0.640
	P Value	<0.001	<0.001

Mapping of EEG classification to case classification

Table 54 maps EEG classification to case classification based on EEG for reviewers 1 and 2. For both reviewers EEGs categorised as normal, non-specific or suggestive were not used in case classification. EEGs categorised as typical were always used in case classification and there was some variability in the use of highly suggestive EEGs in case classification.

Table 54 Mapping of EEG classification to case classification

EEG classification	Reviewer 1		Reviewer 2	
	Can the EEG be used in case classification			
	Yes	No	Yes	No
Normal	0	2	0	8
Non-specific	0	77	0	74
Suggestive	0	49	0	46
Highly Suggestive	12	6	31	0
Typical	20	0	7	0

Using EEG classification to aid interpretation of episodes of inter-observer variation in case classification

To aid interpretation of inter-observer disagreements (n=18), the EEG classification was examined. In 17 instances one reviewer interpreted the EEG as 'highly suggestive' whilst another 'suggestive'; in one instance the disagreement was 'highly suggestive' to 'non-specifically abnormal'.

Intra-observer variance

The overall percentage agreement between reviews in the evaluation of EEG for case classification was 92.8% when EEGs were considered the unit of analysis (Table 55). The Kappa statistic was 0.750, indicating substantial agreement between reviews. Altering the study population to examine EEGs from only sCJD cases and non-cases (narrowly or broadly defined) made little difference to the degree of intra-observer variation or Kappa statistic. When individuals rather than EEGs were considered the unit of analysis the percentage agreement, irrespective of the population studied, was 100% with a Kappa statistic of 1.000 indicating complete agreement.

Between reviews there was disagreement in the use of EEG for case classification in 12 EEGs (from 11 individuals). This group consisted of six neuropathologically confirmed sCJD cases and five probable sCJD cases. Of the probable sCJD cases, case classification based upon EEG findings alone in two cases.

Table 55 Intra-observer variance in the evaluation of EEG for case classification

		Unit of analysis	
		EEG	**Individual**
All suspect sCJD cases	**Agreement, n**	154	108
	Disagreement, n	12	0
	Percentage agreement, %	92.8	100
	Kappa Statistic	0.750	1.000
	P Value	<0.001	<0.001
Narrowly defined sCJD cases and non-cases	**Agreement, n**	90	62
	Disagreement, n	7	0
	Percentage agreement, %	92.8	100
	Kappa Statistic	0.732	1.000
	P Value	<0.001	<0.001
Broadly defined sCJD cases and non-cases	**Agreement, n**	149	104
	Disagreement, n	12	0
	Percentage agreement, %	92.6	100
	Kappa Statistic	0.748	1.000
	P Value	<0.001	<0.001

In five of the EEGs for which there was intra-observer disagreement, there was also inter-observer disagreement. This latter group included one probable sCJD case classified on EEG findings alone.

Sensitivity, specificity, positive and negative predictive values

The sensitivity, specificity, PPV and NPV of EEGs for each reviewer are outlined in Tables 56 (EEG unit of analysis) and 57 (individual unit of analysis) respectively. The sensitivity of EEG was low however the specificity and PPV were extremely high. For example when EEG was considered the unit of analysis estimates of sensitivity ranged from 10.6% to 33.8%. Corresponding values for specificity and positive predictive value were 77% to 100% and 72.7% to 100% respectively. Sensitivity increased, although not significantly so, when individuals rather than EEGs were considered the unit of analysis, at the expense of specificity and positive predictive value. Estimates of sensitivity ranged from 14.0% to 51.3% whilst estimates of specificity ranged from 55.5% to 100% and PPV from 71.3% to 100%.

Table 56 Sensitivity, specificity, positive and negative predictive value of EEG according to reviewer (unit of analysis EEG)

	Estimate % (95%CI)	Reviewer 1	Reviewer 2	Reviewer 1 (second review)
Narrow	Prevalence	77.0% (68.0 – 85.2)	77.0% (68.0 – 85.2)	77.0% (68.0 – 85.2)
Definition	Sensitivity	22.7% (13.8 – 33.8)	22.7% (13.8 – 33.8)	18.7% (10.6 – 29.3)
sCJD n=75	Specificity	100% (84.6 – 100)	95.5% (77.2 – 99.9)	100% (84.6 – 100)
Non-cases	PPV	100% (80.5 – 100)	94.4% (72.7 – 99.9)	100% (76.8 – 100)
n=22	NPV	27.5% (18.1 – 38.6)	26.6% (17.3 37.7)	26.5% (17.4 – 37.7)
Broad	Prevalence	80.0% (73.0 – 86.0)	80.0% (73.0 – 86.0)	80.0% (73.0 – 86.0)
definition	Sensitivity	24.8% (17.6 – 33.2)	28.7% (21.1 – 37.3)	20.2% (13.6 – 28.1)
sCJD n=129	Specificity	100% (89.1 – 100)	96.9% (83.8 – 99.9)	100% (89.1 – 100)
Non-case	PPV	100% (89.1 – 100)	97.4% (86.2 – 99.9)	100% (86.8 – 100)
n=32	NPV	24.8 (17.6 – 33.2)	25.2% (17.8 – 33.8)	23.7% (16.8 – 31.8)

Table 57 Sensitivity, specificity, positive and negative predictive value of EEG according to reviewer (unit of analysis individual)

	Estimate % (95%CI)	Reviewer 1	Reviewer 2	Reviewer 1 (second review)
Narrow	Prevalence	84.0% (72.0 – 92.0)	84.0% (72.0 – 92.0)	84.0% (72.0 – 92.0)
Definition	Sensitivity	30.8% (18.7 – 45.1)	30.8% (18.7 – 45.1)	25.0% (14.0 – 38.9)
sCJD n=52	Specificity	100% (69.2 – 100)	90% (55.5 – 99.7)	100% (69.2 – 100)
Non-cases	PPV	100% (79.4 – 100)	94.1% (71.3 – 99.9)	100% (75.3 – 100)
n=10	NPV	21.7% (10.9 – 36.4)	20% (9.6 – 34.6)	20.4% (10.2 – 34.3)
Broad	Prevalence	83.0% (74.0 – 89.5)	83.0% (74.0 – 89.5)	83% (74.0 – 89.5)
definition	Sensitivity	34.5% (24.6 – 45.4)	40.2% (29.9 – 51.3)	27.6% (18.5 – 38.2)
sCJD n=87	Specificity	100% (81.5 – 100)	94.4% (72.7 – 99.9)	100% (81.5 – 100)
Non-cases	PPV	100% (88.4 – 100)	97.2% (85.5 – 99.9)	100% (85.8 – 100)
n=18	NPV	24.0% (14.9 – 35.3)	24.5% (15.1 – 36.5)	22.2% (13.7 – 32.8)

There was no statistically significant difference in these estimates according to the definition of sCJD applied (narrow or broad). Nor was there a statistically significant difference in sensitivity, specificity, PPV or NPV between reviewers or reviews.

Sensitivity, specificity, positive and negative predictive values for CSF 14-3-3 protein

For comparison the sensitivity, specificity, positive and negative predictive values for CSF 14-3-3 protein in this population were calculated. Values for narrowly defined sCJD were 82.5% (67.2 – 92.7), 75.0% (34.9 – 96.8), 94.3% (80.8 – 99.3) and 46.2% (19.2 – 74.9) respectively; for broadly defined sCJD 87.3% (77.3 – 94.0), 42.9% (17.7 – 71.1), 88.6% (78.7 – 94.9) and 40.0% (16.3 – 67.7) respectively. At the time of writing MRI features were not included in the diagnostic criteria therefore these data have not been presented.

Timing of EEGs that were used in case classification

The first EEG that could be used for case classification was recorded a median of 65 days (41 – 97) after symptom onset and 22 days (15 – 34) before death. The timing of the first and last EEG that could be used for case classification is shown in Figure 61. Of note following the recording of an EEG that could be used for case classification six suspect sCJD cases underwent further EEG examinations.

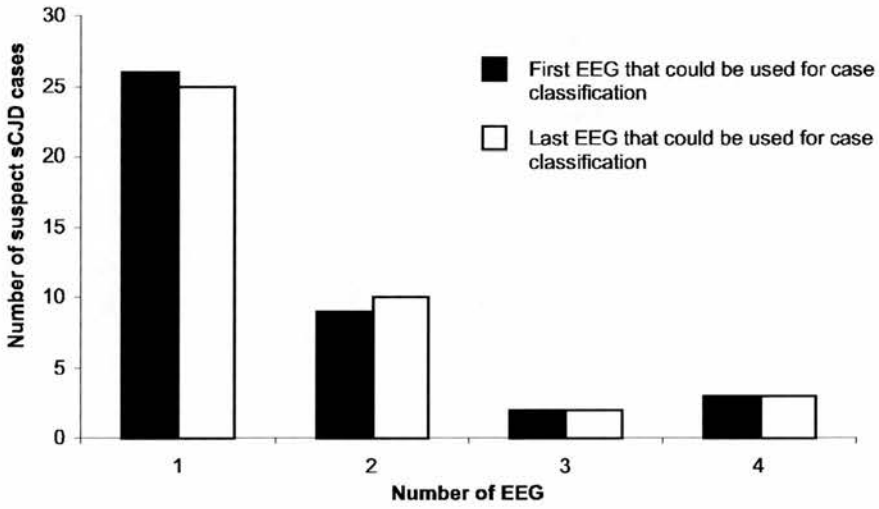

Figure 61 The timing of EEGs used for case classification

Characteristic EEG in a non-case

The EEG was considered characteristic of sCJD and would have been used in case classification by one reviewer in a neuropathologically confirmed non-case. Interestingly both reviewers considered the EEG in this individual to be highly

suggestive. This individual was a 53 year old male, heterozygote (Codon 129 MV) with an illness duration of 14 months, negative CSF 14-3-3 protein and MRI examinations. Whilst a definitive diagnosis was not reached, there was no evidence of abnormal prion protein on examination of tissue obtained from brain biopsy in this case.

sCJD cases that did not have characteristic EEGs

In 26 neuropathologically confirmed sCJD cases and 12 probable sCJD cases neither reviewer considered that features on EEG supported a diagnosis of sCJD. Table 58 compares the baseline characteristics of sCJD cases, narrowly and broadly defined, according to whether EEG was, or was not, considered by either reviewer to support a diagnosis of sCJD at any stage in the clinical illness.

Table 58 Comparison of baseline characteristic of sCJD cases (narrowly and broadly defined) that did not have EEG features supporting a diagnosis of sCJD at any stage in their clinical illness, compared to those that did

	Narrowly defined sCJD			Broadly defined sCJD		
	EEG supportive	**EEG not supportive**	**P value**	**EEG supportive**	**EEG not supportive**	**P value**
Number	26	26		38	49	
Age at symptom onset, Median years (IQR)	75 (61 – 78)	68 (60 – 74)	0.244	73 (61 – 78)	67 (61 – 74)	0.092
Male, n (%)	14 (53.8)	15 (57.7)	0.078	22 (57.9)	25 (51.0)	0.407
Illness duration, Median days (IQR)	106 (69 – 176)	145 (100 – 247)	0.072	109 (81 – 164)	145 (100 – 231)	0.047
Number of EEG, Median (range)	1 (1 – 4)	1 (1 – 4)	0.911	1 (1 – 4)	1 (1 - 4)	0.991
Time onset to first EEG, Median days (IQR)	69 (42 – 138)	131 (79 – 214)	0.023	71 (44 – 103)	107 (74 – 165)	0.016
Time last EEG to death, Median days (IQR)	18 (15 – 26)	33 (23 – 50)	0.051	21 (16 – 29)	34 (28 – 55)	0.007

Summary of key findings

- There was substantial agreement between reviewers in the classification of EEG using a 5 point descriptive scheme
- This scheme mapped well to case classification. Neither reviewer used normal, non-specific or suggestive EEGs in case classification and both used all typical EEGs. There was some variability in the use of highly suggestive EEGs in case classification.
- There was substantial agreement between reviewers in assessment of EEG for case classification.
- Reviewers disagreed on the classification of 10% of EEGs (accounting for 15% of all suspect sCJD referrals), although in less than 2% of EEGs2% of suspect sCJD cases) this disagreement was of clinical significance.
- Most cases of disagreement between reviewers arose as a result of close disagreement between adjacent categories, one reviewer classifying the EEG as highly suggestive whilst the other classified the EEG as suggestive.
- Intra-observer variance in the assessment of EEG for case classification was minimal.
- Disagreement between reviews was of clinical significance in just 2 EEGs from 2 individuals representing just 1.9% of the study population and 1.2% of all EEGs reviewed; when individuals were considered rather than EEGs there was complete agreement between reviews.
- The sensitivity of EEG was low however the specificity and PPV were high.
- Over the same period the sensitivity of CSF 14-3-3 protein was much higher than that of EEG with a marginally lower sensitivity and PPV.
- In one pathologically confirmed non-case EEG was considered characteristic and in the appropriate clinical context would have been used in case classification. Both reviewers considered this EEG to be highly suggestive, although only one indicated that they would use to EEG in case classification. A definitive diagnosis was not reached in this case although there was no evidence of PrP^{Sc} on tissue obtained from brain biopsy.

Discussion

In this chapter the operational criteria for the evaluation of EEG in case classification of suspect sCJD were prospectively validated. The main findings from this study will be discussed in the section that follows.

Interpretation of EEG in suspect sCJD

The interpretation of the EEG in suspect sCJD is challenging for a number of reasons.(103) Not all patients with sCJD develop PSWC on EEG. In the early, and less commonly in the late, stages of illness the EEGs may show non-specific slow wave abnormalities. Often the EEG progresses over the course of the clinical illness necessitating serial examinations that may be increasingly suggestive and broadly compatible with, but not entirely typical of, the PSWC classically associated with sCJD.

Nevertheless the assessment of EEG is required in case classification of sCJD. Whilst quantitative criteria for the assessment of digitalised EEG recordings have been developed these have not been prospectively validated in a large scale study and there are practical difficulties in applying such criteria in the UK.(98) In the absence of published criteria for the assessment EEG that can be practically applied in the UK, the NCJDSU developed operational criteria, albeit loosely defined, that have been employed by the same two clinicians since the inception of systematic prospective disease surveillance in the UK but never prospectively validated.

In the present study two experienced CJD clinicians reviewed EEGs using an internally developed descriptive scheme to classify EEGs and independently decided whether the EEG should be used for case classification in sCJD. The overall percentage agreement between reviewers was high with Kappa statistics indicating moderate to substantial agreement in both EEG classification and the use of EEG in case classification. The descriptive scheme mapped well to case classification. Where there was disagreement between reviewers interpretation of this was aided by an examination of the descriptive criteria. In most instances this was due to one reviewer considering the EEG suggestive whilst another considered the EEG highly

suggestive, in turn there was some variation between reviewers in the use of highly suggestive EEGs for case classification; one reviewer indicated that he would use all highly suggestive EEGs for case classification whilst another indicated that he would use just two thirds. These data highlight that interpretation of EEG, even in the hands of experienced clinicians is challenging.

Disagreement between reviewers was of clinical significance, that is case classification had been based on EEG alone, in just 3 cases. Nevertheless, given the rarity of sCJD misclassification of even a single case may distort national trend data with implications for national and indeed international public health policy. These cases were reviewed in detail by both clinicians following this study; neither was reclassified based on the findings from this study. This does however identify a need for periodic quality assurance of EEG assessment by the NCJDSU. Independent review of a random sample of EEGs from suspect sCJD cases by both clinicians could be undertaken with case by case review of any instances in which disagreement occurred to determine if case classification should be revised.

The remarkably consistent assessment of EEGs for case classification by one reviewer suggests that a single individual should examine all EEG for disease surveillance purposes. However such an approach would be neither stable nor sustainable in the UK. There is a further issue regarding stability and sustainability in the context of using un-validated criteria in the assessment of EEG where this assessment is in part subjective, based on the vast experience of the two clinicians that participated in this study. This study has made some progress toward addressing the issue of the validity of the NCJDSU operational criteria for the assessment of EEG in disease surveillance. However further characterisation of the more subjective elements relating to case classification are required. It would be important for the NCJDSU to ensure succession planning such that any change in personnel will not impact on this aspect of disease surveillance.

Central review of EEGs by the NCJDSU

The EEG was not available for review in almost a quarter (33) of suspect sCJD cases that were known to have undergone EEG examination. While the NCJDSU

endeavour to review all relevant investigation in suspect sCJD cases there is no statutory requirement for clinicians to provide these. The suspect cases for which EEG was unavailable did not differ in age or sex distribution from suspect sCJD cases for which EEG was available and EEG classification was not integral to case classification in any of these cases. However of these suspect cases the EEG was reported by the local neurophysiologist to have been consistent with a diagnosis of sCJD in almost a fifth (6). The criteria applied by local neurophysiologists in the assessment of EEGs in suspect sCJD are not known. It would be desirable to examine the inter-observer variance in EEG reporting between the NCJDSU and referring centres. Estimating this would go some way towards determining whether the central review of EEG is necessary for surveillance purposes. However referrals to the NCJDSU are made from hospitals across the UK. In this study the EEGs reviewed were recorded and interpreted in 69 different centres. Logistically a prospective study of this nature would be challenging. The reporting of EEGs in the UK is not standardised in such a way as to facilitate a study of this nature retrospectively.

It is unlikely that the assessment of EEGs in different centres, nationally and internationally is reproducible given the lack of practical objective criteria for this purpose. The approach taken by international PHS systems to assessing EEGs in case classification is unclear, although anecdotally there is said to be considerable variation (personal communication R.Knight). Transparency in this area is required to aid the interpretation of international surveillance data, particularly given the trend toward the study of pooled data from multiple international collaborators, most commonly in the EUROCJD group.

Comparison with the literature

Steinhoff et *al* in a small study of 29 suspect cases (15 sCJD and 14 non-cases based on clinical criteria) reported a Kappa statistic of 0.95 for intra-observer reliability in evaluation of EEG applying objective quantitative criteria indicating almost perfect agreement between reviewers.(105) The criteria used for assessment of EEG in this as previously noted, cannot be applied in the UK for practical reasons (Table 6). Moreover this study included sCJD cases and non-cases based on clinical diagnostic

criteria which include assessment of EEG. In the present study I was able to overcome this by carrying out additional analyses which limited the study population to only sCJD cases and non-cases with a neuropathologically confirmed diagnosis. The present study is the only published study to examine intra-observer variation in the assessment of EEG for case classification in the surveillance of sCJD.

Diagnostic value of the EEG in suspect sCJD

Until recently estimates of the sensitivity of EEG in sCJD have ranged from 58% – 66%.(96;97;104;105) In many countries including the UK, as illustrated in previous chapters of this thesis, the importance of EEG in the diagnostic evaluation of suspect sCJD cases has diminished since the incorporation of CSF 14-3-3 protein into the WHO diagnostic criteria.(107) A reduction in the sensitivity of EEG post-1997, the year that CSF 14-3-3 protein became widely available, has been reported by most EUROCJD collaborators.(107) In Germany for example the sensitivity and specificity of EEG in sCJD (definite or probable) was reported to be 32% and 94% respectively between 2001 and 2003,(106) compared to 64% and 91% between 1996 and 2000.(104) This has been attributed to the introduction of CSF 14-3-3 protein, which the authors speculate has led to suspect sCJD cases being referred to the PHS system at an earlier stage, prior to the onset of PSCW on EEG.(106) Using more contemporary data collected by 12 international collaborators between 1998 and 2007, Zerr *et al* examined the diagnostic utility of EEG, CSF 14-3-3 protein and MRI in 436 sCJD cases (definite or probable) and 141 non-cases (40% with a neuropathological diagnosis).(99) The reported sensitivity of EEG in this study was 44%, with a specificity of 92%; corresponding values for CSF 14-3-3 protein 86% and 68%; values not significantly different from those reported in the present study.

In the UK the sensitivity of EEG in the diagnosis of sCJD has been falling for some time. In the second chapter of this thesis I reported a fall in the sensitivity of EEG in sCJD cases, from 50.0% in 1990 to 33.0% in 2006 ($P<0.001$). In the third chapter of this thesis I demonstrated that there was no evidence to suggest a temporal reduction in time from symptom onset to suspect sCJD cases being referred to the NCJDSU that could be attributed to the introduction of CSF 14-3-3 protein. However the median time from symptom onset to positive investigation is shorter for CSF 14-3-3

protein (87 days (57 – 156)) than EEG (95 days (57 – 168)) and the sensitivity of CSF 14-3-3 protein is significantly higher. If diagnosis of probable sCJD can be reached using CSF 14-3-3 prior to EEG then an associated fall in the median number of EEG undertaken would be expected. A non-significant fall in the median number of EEG undertaken over the course of the clinical illness in sCJD cases was noted in the second chapter of this thesis. Combined these data suggest that CSF 14-3-3 protein has contributed to the fall in sensitivity of EEG through the earlier detection of probable sCJD cases. This may not be apparent in an examination of temporal trends in time to referral because the time between first positive CSF 14-3-3 protein examination and first positive EEG is short.

A fall in sensitivity of EEG in sCJD in the UK was first noted in 1995 when CSF 14-3-3 protein was an experimental assay and not widely used. The sensitivity of EEG continued to fall until 2000 and has remained at approximately 25-30% since. A number of factors may have contributed to this stabilisation. The use of CSF 14-3-3 protein in the investigation of suspect sCJD in the UK is less common that in other countries. EEG remains the most commonly used investigation to support a diagnosis of sCJD in suspect sCJD cases in the UK. In addition, in certain molecular subtypes of sCJD (MM1 and MV1), EEG is an exceptionally useful investigation. Given these sCJD cases often have a classical clinical illness, sCJD may be suspected in life and sequential EEGs may be preferentially used to investigate suspect cases over the more invasive investigations such as CSF 14-3-3 protein.

The specificity of EEG was exceptionally high in this study and comparable to published reports. In this study just one non-case was considered by a single reviewer to have an EEG that could in the appropriate clinical context be used for case classification. A definitive diagnosis was not reached in this case despite the individual undergoing brain biopsy in life. Heinemann *et al* reported that brain biopsy was non-diagnostic in 42% of suspect CJD cases that underwent this investigation in Germany between 1993 and 2005 (n=26). Only a quarter of suspect cases without a definite diagnosis following brain biopsy underwent post mortem examination therefore it is not known whether CJD cases were missed at brain

biopsy.(135) Although post mortem examination was not carried out on the suspect case identified in this study, neuropathological tissue from brain biopsy was evaluated by an experienced neuropathologist from the NCJDSU (JI) and in the absence of histological changes or PrP^{Sc} sCJD was excluded as a differential diagnosis.

For most diagnostic investigations the relationship between sensitivity and specificity tends to be reciprocal such that as one increases the other is compromised. In many respects for a condition such as sCJD it is preferable that a non-invasive investigation such as EEG has a high specificity. Whilst the specificity indicates that EEG is identifying individuals that do not have sCJD with a high degree of certainty, the low sensitivity of this investigation means that up to 75% of sCJD cases will be missed. In the investigation of suspect sCJD cases EEG should therefore be used conjunction with other diagnostic tools such as CSF 14-3-3 protein and MRI.

Conclusions

This study has confirmed the validity of the NCJDSU operational criteria for the assessment of EEG in case classification of suspect sCJD when applied by two experienced CJD clinicians. It is not clear whether these results would be reproducible in the hands of less experienced clinicians. Whilst the sensitivity of EEG in sCJD is low, the specificity is high. As a non-invasive investigation EEG remains a useful tool in the assessment of suspect sCJD cases if used in conjunction with other diagnostic technology.

Chapter 5. Death certificates in the surveillance of prion disease in the UK

Introduction

In the USA the analysis of death certificates is considered to be the most "*systematic and cost effective method of [CJD] surveillance.*"(227) The examination of death certificates is commonly used as an adjunct to other activities in the prion disease surveillance (Table 18). There are however remarkably few contemporary studies describing the diagnostic value of death certificate in prion disease surveillance, and the approach taken to identifying prion disease on death certificates varies between studies, some examining only the underlying cause of death as ICD coded,(51) others examining multiple cause of death both ICD coded and recorded in the literal text of the certificate.(50) Data describing the accuracy of ICD coding of prion diseases on death certificates in the UK and elsewhere, are lacking. A consensus as to the most valid approach to adopt when using death certificates in surveillance has not been reached. In this chapter I will address these critical gaps in the literature.

Aims and objectives

The overall aim of this chapter was to evaluate the use of death certificates in the surveillance of prion disease in the UK from 1990 through 2006.

Specific objectives were as follows:

1. To describe the use of death certificates in the ascertainment of suspect prion disease cases in the UK from 1990 through 2006.
2. To determine which approach to examining death certificates is optimal in ascertaining suspect prion disease cases.
3. To examine the diagnostic value of death certificates (sensitivity, specificity, PPV and NPV) in the surveillance of all human prion disease, sCJD cases and vCJD cases in the UK from 1990 through 2006.

4. To examine the accuracy of ICD coding of prion diseases on death certificates in the UK from 1990 through 2006
5. To compare age standardised mortality rates of human prion diseases produced by the surveillance methods adopted by the NCJDSU to age standardised mortality rates produced ascertaining prion disease cases using death certificates alone

Methods

Death certification in the UK

In the UK death certificates are completed by a physician. The certificate consists of two parts. In Part I, a sequence of up to three conditions that led to death are recorded. The condition that led directly to death is recorded in the first position. The underlying condition, to which all preceding conditions are attributable, is recorded in the last position. In Part II co-morbid conditions that may have contributed to, but did not directly cause death, are recorded. All death certificates are returned to the Office of National Statistics (ONS) (England and Wales) or General Register Office's (GRO) (Scotland and Northern Ireland) where they are coded according to the World Health Organisations International Classification of Diseases (ICD9 pre-1996 and ICD10 post-1996). The underlying cause of death is determined by the ONS/GRO and ICD coded accordingly. The ONS/GRO may ICD code multiple additional causes of death or co-morbid conditions at their own discretion. These need not directly correspond to the sequence entered by the certifying physician.

The surveillance protocol

Quarterly the ONS/GRO send all death certificates from the UK coded under the rubrics 046.1 'Jakob-Creutzfeldt Disease' (ICD9), 331.9 'Cerebral degeneration, unspecified' (ICD9), A81.0 'Creutzfeldt-Jakob Disease' (ICD10) or F02.1 'Dementia in Creutzfeldt-Jakob Disease' (ICD10) to the NCJDSU as part of routine surveillance practice. On expiration, death certificates are requested for all suspect prion disease cases referred to the NCJDSU. Of note 331.9 is not a CJD specific code.

Data collection

All suspect prion disease cases referred to the NCJDSU between 1st May 1990 and 31st December 2006 were followed for two years until 31st December 2008. A further six months was given to ensure the death certificates of individuals deceased as of 31st December 2008 had been received by the NCJDSU. The following information was extracted from death certificates and entered onto a password protected database: name, date of birth, sex, date of death, place of death, occupation of individual certifying death, causes of death as recorded at each position on the death certificate (literal text), underlying cause of death (ICD coded), all other ICD coded causes of death or co-morbidities contributing to death. These data were linked to the following information extracted from the NCJDSU case record: case classification, disease subtype (sporadic, variant, genetic or iatrogenic), date of referral to the NCJDSU and referral source. Data were then anonymised.

Cleaning and coding of death certificate data

The following conditions recorded in the literal text of a death certificate were considered to be indicative of a diagnosis of CJD or genetic prion disease: Creutzfeldt-Jakob Disease or Creutzfeldt-Jakob syndrome or Creutzfeldt-Jakob dementia (various spellings), Jakob-Creutzfeldt Disease or Jakob-Creutzfeldt syndrome or Jakob-Creutzfeldt dementia (various spellings), CJD, prion disease or prion dementia, spongiform encephalopathy or Gerstman-Straussler-Scheinker Syndrome (various spellings). The position in which this diagnosis was recorded on the death certificate noted. This was categorised as follows:

- Recorded in the literal text as the immediate cause of death (Part Ia)
- Recorded in the literal text as the underlying cause of death (The last position on Part I of the death certificate)
- Recorded in the literal text in any position (Part I or II)

Throughout this chapter I will refer to CJD recorded on the death certificate denoting that either CJD or genetic prion disease were recorded in the literal text of the death certificate.

CJD or genetic prion disease was considered to have been ICD coded on a death certificate if the following ICD9 (ICD10) codes were found: 046.1 (A81.0, F02.1). The position of coding was considered in the following categories:

- ICD coded as the underlying cause of death as determined by the ONS/GRO
- ICD coded anywhere on the death certificate

Throughout this chapter I will refer to CJD having been ICD coded on the death certificate if the aforementioned ICD codes appeared on the death certificate.

Age at death was treated as a continuous variable unless otherwise stated. The following age categories were used to examine the diagnostic utility of death certificates:

- vCJD: ≤ 30 years, 31–49 years, ≥ 50 years
- sCJD: ≤ 50 years, 50–59 years, 60–69 years, 70-79 years and ≥ 80 years

In the calculation of age-specific mortality rates, age was examined in 5 year bands with a lower age limit of <20 years and an upper age limit of ≥ 85 years.

Year group was considered in the following categories:

- sCJD: 1990–1995, 1996–2000 and 2001–2006
- vCJD: 1996–2000 and 2000–2006

In general two approaches to examining death certificates in this field have been adopted. One is to limit analyses to individuals for whom a neuropathologically confirmed diagnosis is available (classifications 1.0 and 4.3). An alternative approach, which more accurately reflects disease reporting practices, is to consider a case as a definite or probable prion disease case (classification 1.0 or 2.0) and non-case as a suspect case that failed to meet the diagnostic criteria (classification 4.1, 4.2 or 4.3). To reflect these differing approaches and ensure comparability with the existing literature analyses examining the diagnostic utility of death certificates applied two definitions as follows:

- A narrow (neuropathological) definition considered only neuropathologically confirmed cases (classification 1.0) and non-cases (classification 4.3).
- A broad (clinical) definition which considered cases as individuals meeting the WHO criteria as a definite or probable case (classification 1.0 or 2.0) and non-cases as individuals classified as 4.1, 4.2 or 4.3.

Statistical analyses

The baseline characteristics of all suspect prion disease cases referred to the NCJDSU between 1st May 1990 and 31st December 2006 according to disease subtype were described. The proportion of deceased cases for which a death certificate was available to the NCJDSU was quantified according to disease subtype and case classification. Non-parametric tests including the Wilcoxon-Ranksum test and Fishers exact test were used to compare the baseline characteristics of those suspect cases (deceased) for whom a death certificate was unavailable to the NCJDSU to those for whom a death certificate was not available to the NCJDSU.

The annual number of suspect prion disease cases ascertained by the NCJDSU through death certificate review alone was determined. Chi^2 test for trend was used to assess whether the proportion of all suspect prion disease cases ascertained by the NCJDSU through this route had changed significantly over time.

The proportion of suspect prion disease cases that had CJD recorded in the literal text and/or ICD coded on their death certificate, according to disease subtype and case classification, were described. The position that CJD was recorded in the literal text or ICD coded on the death certificate was considered.

To assess the diagnostic utility of death certificates the sensitivity, specificity, PPV and NPV of CJD ICD coded in any position on the death certificate was examined as this reflects current surveillance practice. Sensitivity was defined as the proportion of cases correctly identified by death certificates. Specificity defined as the proportion of non-cases correctly identified by death certificates. PPV was defined as the

proportion of those with prion disease on their death certificate that actually had prion disease, and NPV the proportion of non- cases that did not have prion disease on their death certificate. The latter measures, PPV and NPV, are dependent on the prevalence of disease in the population therefore disease prevalence estimates were provided. All prion disease cases were considered first, then sCJD and vCJD. The diagnostic utility according to age group and then year group was examined. Finally a linear regression model was fitted to assess whether, following adjustment for age group, the sensitivity of death certificates had increased over time. These analyses were carried out for both narrowly defined (pathological) and broadly defined (clinical) prion disease. These analyses were then repeated examining CJD recorded in the literal text or ICD coded in any position on the death certificate as it was determined in the course of this thesis that this approach to examining the fields on a death certificate produced the greatest yield for disease surveillance purposes.

The accuracy of ICD coding of death certificates was assessed by examining the number of suspect cases that had CJD recorded (literal text any position) but not ICD coded (any position) on their death certificates. In turn the number of suspect cases that had CJD ICD coded (any position) but not recorded (literal text any position). The changing proportions of these groups over time was assessed using Chi2 tests from trend to determine whether there was any temporal change in the accuracy of ICD coding.

The number of deaths from prion disease annually (definite or probable) as ascertained by the NCJDSU using all surveillance methodologies was quantified. The number of deaths from prion disease annually as determined by review of death certificates (CJD recorded in the literal text or ICD coded in any position) was quantified. Age-specific mortality rates in men and women were calculated using denominator data from mid-year population estimates in the UK for both. Directly age standardised prion disease mortality rates were calculated for both using denominator data from the 2001 Census. All analyses were carried out in STATA Version 10. A level of statistical significance of 0.05 is used throughout.

Results

Study population

In total 2,154 suspect prion disease cases were referred to the NCJDSU from 1st May 1990 through 31st December 2006 (Table 59). Three referrals were subsequently considered international cases due to their location at time of symptom onset. These individuals died and were certified in the UK and were therefore retained in analyses. As of 31st December 2008, 1894 (87.9%) suspect cases were deceased. Death certificates were available for 1879 (99.5%); 1504 suspect sCJD cases, 221 suspect vCJD cases, 53 suspect iCJD cases and 99 suspect genetic prion disease cases. A death certificate was unavailable for 15 suspect cases, a third (5) of whom were known to have died overseas. Suspect cases for whom a death certificate was not available did not differ significantly from suspect cases for whom a death certificate was available with respect to sex (P=0.507), case classification (P=0.229) or aetiological subtype (P=0.109) but were approximately 11 years younger at death, (55.2 (35.5 – 63.2) years *vs.* 66.4 (55.7 – 74.8) years, P=0.029).

Death certificates in the ascertainment of prion disease

Over the entire study period, 115 (5.3%) suspect prion disease cases were ascertained by the NCJDSU through death certificate review alone; 108 (93.9%) were suspect sCJD cases and 7 (6.1%) suspect genetic prion disease cases. Over time there was a statistically significant reduction in the proportion of all suspect prion disease cases ascertained by the NCJDSU through death certificate review alone, from 20.8% (11) in 1990 to 0% (0) in 2006 (P<0.001) (Figure 62). Over the entire study period, 30 prion disease cases (definite or probable) were ascertained by review of death certificates alone, representing 2.4% of all prion disease cases (definite or probable) ascertained by the NCJDSU. This figure fell from 10.0% (2) in 1990 to 0.0% (0) in 2006.

Table 59 Characteristics suspect prion disease cases referred to the NCJDSU according to aetiological subtype, 1990 – 2006

	sCJD	vCJD	iCJD	Genetic Prion Disease	All
Number (%)	1651 (76.7)	322 (14.9)	58 (2.7)	121 (5.6)	2154 (100)
Male (%)	797 (48.3)	171 (53.1)	34 (58.6)	56 (44.8)	1058 (49.1)
Median Age at death (IQR)	69.1 (62.3 – 76.4)	30.9 (24.7 – 41.3)	31.8 (28.5 – 37.3)	55.0 (45.4 – 61.8)	66.4 (55.6 – 74.8)
Case Classification, n (%)					
Pathologically CJD [1.0]	688 (41.7)	117 (36.3)	47 (81.0)	84 (69.4)	936 (43.5)
Definite or probable CJD [1.0 or 2.0]	892 (54.0)	167 (51.8)	54 (93.1)	116 (95.8)	1229 (57.1)
Possible CJD [3.0]	88 (5.3)	3 (0.9)	0	0	91 (4.2)
Clinically or pathologically not CJD [4.1, 4.2 or 4.3]	655 (39.6)	152 (47.2)	3 (5.2)	5 (4.1)	817 (37.9)
Pathologically not CJD [4.3]	226 (13.7)	36 (11.2)	0	1 (0.8)	263 (12.2)
Unclassifiable [0.0]	16 (1.0)	0	1 (1.7)	0	17 (1.0)
Number Dead (%)	1514 (91.6)	224 (69.6%)	54 (93.1)	102 (84.3)	1894 (87.9)
Death Certificate Available if Deceased (%)	1504 (99.7)	221 (98.7)	53 (100.0)	99 (99.0)	1879 (99.5)
Ascertained death certificate review alone, n (%)	108 (6.5)	0	0	7 (5.8)	115 (5.3)

[ ] indicate case classification

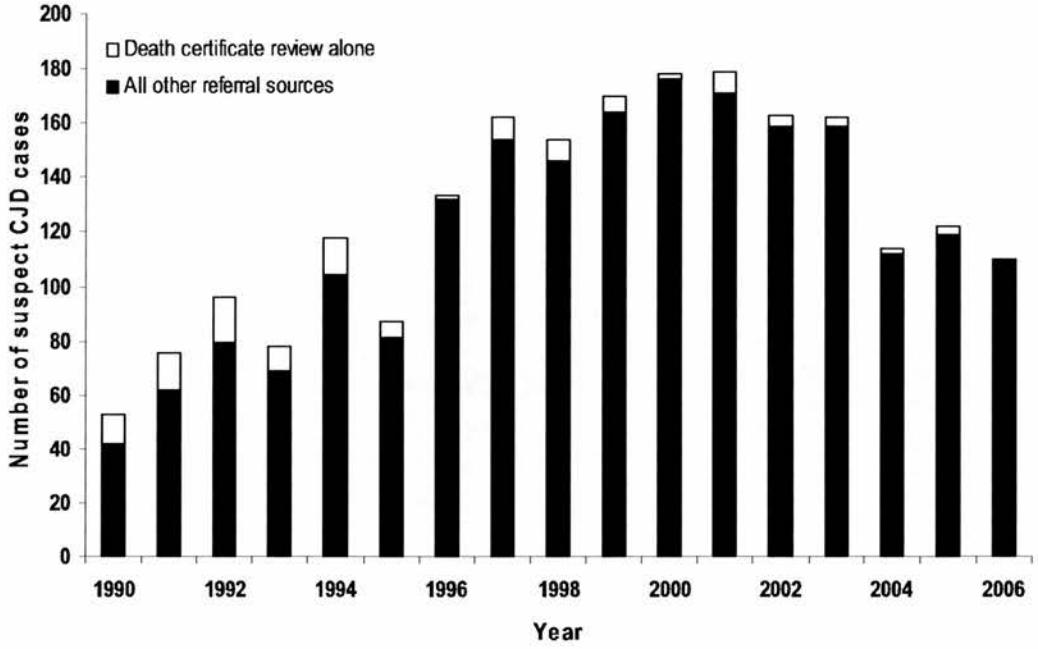

Figure 62 Number of suspect prion disease cases ascertained by the NCJDSU through review of death certificate alone

CJD recorded on death certificates

Overall, 800 (42.6%) suspect cases referred to the NCJDSU had CJD recorded in the literal text as the immediate cause of death (Part 1a) on their death certificate (Table 60a). This rose to 1108 (59.3%), when CJD recorded in the literal text as the underlying cause of death was considered. Rising further to 1191 (63.4%) when CJD recorded in the literal text in any position (Part I or II) on the death certificate was considered. Reassuringly, a high proportion of individuals that met the WHO diagnostic criteria as a definite, probable or possible case had CJD recorded in the literal text of their death certificate. This yield increased as multiple causes of death were considered. Examining ICD codes rather than literal text produced similar results although the yield was lower (Table 60b). For example, CJD was ICD coded in any position on the death certificate in 1128 (60.0%) suspect cases, but recorded in the literal text in any position on the death certificate in 1191 (63.4%) suspect cases. An examination of ICD coding and literal text in any position on the death certificate led to greatest yield, identifying 1227 (65.3%) suspect cases. Supplemental analyses

describing the underlying cause of death in prion disease cases (definite or probable) according to disease subtype can be found in Appendix 5.

As noted in the methods section the NCJDSU routinely request death certificates ICD 9 coded 046.1 or 331.9 and ICD 10 coded A81.0 or F02.1. The ICD 9 code 331.9, 'Cerebral degeneration, unspecified' is not specific to prion disease and was therefore was not included in these analyses. This code was recorded (any position) in just 12 death certificates received by the NCJDSU across the study period. None of these suspect cases were ascertained by death certificate alone. In just one out of these 12 suspect cases, CJD was recorded on the text of the death certificate. This was a pathologically confirmed case of sCJD that had been referred to the NCJDSU by a neuropathologist. Of note, no death certificates reviewed by the NCJDSU were ICD coded (any position) under the rubrics F02.1 across the study period.

The degree of misclassification of death certificates in cases (narrowly and broadly defined) varied according to disease subtype. For example, 16.6% of definite or probable sCJD cases did not have CJD recorded or ICD coded in any position on their death certificate compared to 17.9% of genetic prion disease cases, 6.3% of vCJD cases and 3.4% of iCJD cases ($P<0.001$). Further analyses, where possible, were therefore stratified according to disease subtype.

Supplemental analyses were carried out to determine whether any routinely available information recorded on the death certificate could be used to distinguish cases from non-cases in those certified as CJD, and whether cases that did and did not have CJD recorded on their death certificate differed significantly. These analyses have been presented in Appendix 6.

Table 60a Recording of CJD in literal text on death certificates of all suspect cases referred to NCJDSU by disease subtype and case classification

	sCJD	vCJD	iCJD	Genetic Prion Disease	All
CJD recorded as immediate cause of death (literal text), n (%)	621 (41.3)	110 (49.8)	22 (41.5)	47 (47.5)	800 (42.6)
Pathologically CJD [1.0]	350 (51.2)	69 (61.6)	21 (44.7)	35 (48.6)	461 (52.1)
Definite or probable CJD [1.0 or 2.0]	505 (56.9)	105 (66.0)	22 (41.5)	47 (49.5)	679 (56.8)
Possible CJD [3.0]	49 (57.0)	2 (66.7)	0	0	51 (57.3)
Clinically or pathologically not CJD [4.1, 4.2 or 4.3]	61 (11.8)	3 (5.0)	0	0	64 (11.0)
Pathologically not CJD [4.3]	23 (10.6)	3 (8.8)	0	0	26 (10.2)
Unclassifiable [0.0]	6 (46.2)	0	0	0	6 (46.2)
CJD recorded as underlying cause of death (literal text), n (%)	844 (56.4)	150 (67.9)	46 (88.5)	68 (69.4)	1108 (59.3)
Pathologically CJD [1.0]	491 (72.0)	98 (87.5)	42 (91.3)	54 (71.1)	685 (74.8)
Definite or probable CJD [1.0 or 2.0]	676 (76.1)	142 (89.9)	46 (88.5)	68 (72.6)	932 (78.4)
Possible CJD [3.0]	63 (73.4)	3 (100.0)	0	0	66 (74.2)
Clinically or pathologically not CJD [4.1, 4.2 or 4.3]	98 (19.1)	5 (8.5)	0	0	103 (17.8)
Pathologically not CJD [4.3]	37 (17.1)	4 (11.8)	0	0	41 (16.1)
Unclassifiable [0.0]	7 (53.9)	0	0	0	7 (53.9)
CJD recorded in any position (literal text), n (%)	910 (60.5)	155 (70.1)	50 (94.3)	76 (76.8)	1191 (63.4)
Pathologically CJD [1.0]	518 (75.7)	102 (91.1)	45 (95.7)	60 (77.9)	725 (78.8)
Definite or probable CJD [1.0 or 2.0]	714 (80.5)	147 (92.5)	50 (94.3)	76 (80.0)	987 (82.7)
Possible CJD [3.0]	66 (76.7)	3 (100.0)	0	0	69 (77.5)
Clinically or pathologically not CJD [4.1, 4.2 or 4.3]	119 (22.9)	5 (8.5)	0	0	124 (21.3)
Pathologically not CJD [4.3]	47 (21.7)	4 (11.8)	0	0	51 (20.1)
Unclassifiable [0.0]	11 (84.6)	0	0	0	11 (84.6)

[ ] indicates case classification

Table 60b Recording of CJD in ICD codes on death certificates of all suspect cases referred to NCJDSU by disease subtype and case classification

	sCJD	vCJD	iCJD	Genetic Prion Disease	All
CJD ICD coded as underlying cause of death, n (%)	857 (57.0)	151 (68.3)	45 (84.9)	60 (60.6)	1113 (59.3)
Pathologically CJD [1.0]	505 (73.8)	99 (88.4)	42 (89.4)	46 (59.7)	692 (75.2)
Definite or probable CJD [1.0 or 2.0]	686 (77.3)	144 (90.6)	45 (84.9)	60 (63.2)	935 (78.3)
Possible CJD [3.0]	63 (73.3)	2 (66.7)	0	0	65 (73.0)
Clinically or pathologically not CJD [4.1, 4.2 or 4.3]	98 (18.9)	5 (8.5)	0	0	103 (17.7)
Pathologically not CJD [4.3]	35 (16.1)	4 (11.8)	0	0	39 (15.4)
Unclassifiable [0.0]	10 (76.9)	0	0	0	10 (76.9)
CJD ICD coded in any position, n (%)	868 (57.8)	151 (68.3)	48 (90.6)	61 (61.2)	1128 (60.0)
Pathologically CJD [1.0]	510 (74.6)	99 (88.4)	43 (91.5)	46 (59.7)	698 (75.9)
Definite or probable CJD [1.0 or 2.0]	692 (78.0)	144 (90.6)	48 (90.6)	61 (64.2)	945 (79.2)
Possible CJD [3.0]	63 (73.3)	2 (66.7)	0	0	65 (73.0)
Clinically or pathologically not CJD [4.1, 4.2 or 4.3]	102 (19.7)	5 (8.5)	0	0	107 (18.4)
Pathologically not CJD [4.3]	38 (17.5)	4 (11.8)	0	0	42 (16.5)
Unclassifiable [0.0]	11 (84.6)	0	0	0	11 (84.6)
CJD recorded (literal text) or ICD coded in any position, n (%)	941 (62.6)	157 (71.0)	51 (96.3)	78 (78.8)	1227 (65.3)
Pathologically CJD [1.0]	544 (79.5)	104 (92.9)	46 (97.9)	62 (80.5)	756 (82.2)
Definite or probable CJD [1.0 or 2.0]	740 (83.5)	149 (93.7)	51 (96.3)	78 (82.1)	1018 (85.3)
Possible CJD [3.0]	66 (76.7)	3 (100)	0	0	69 (77.5)
Clinically or pathologically not CJD [4.1, 4.2 or 4.3]	123 (23.8)	5 (8.5)	0	0	128 (22.0)
Pathologically not CJD [4.3]	49 (22.6)	4 (11.8)	0	0	53 (20.9)
Unclassifiable [0.0]	12 (92.3)	0	0	0	12 (92.3)

[ ] indicate case classification

The diagnostic utility of death certificates

The diagnostic utility of death certificates in the surveillance of prion disease in the UK was assessed by examining the sensitivity, specificity, PPV and NPV of a death certificate diagnosis of CJD. These values were first assessed using CJD ICD coded in any position on the death certificate as this reflects current practice.

The sensitivity, specificity, PPV and NPV of CJD ICD coded on death certificates is shown in Tables 61 and 62 (narrowly defined). Overall the sensitivity (all prion disease) was 75.9% (75.0 – 78.6) with a specificity of 84.0% (78.9 – 88.3). PPV and NPV were 94.6% (92.7 – 96.1) and 48.6 (43.8 – 53.4) respectively. Thus death certificates correctly identified three out of every four prion disease cases and correctly identified five out of every six non-cases. The PPV was extremely high indicating that individuals with a death certificate diagnosis of prion disease had a high (95%) probability of having prion disease. However the NPV was low, half of those that did not have CJD ICD coded on their death certificate had prion disease. The sensitivity was highest in the youngest age group, although there was no discernable trend across age groups in these values. Data for sCJD very much followed the overall trend for all prion disease. For vCJD the sensitivity and NPV were higher than associated values for all prion disease and sCJD, with comparable levels of specificity and PPV. There was no discernable pattern according to age group. Across year groups there was an apparent increase in sensitivity between 1990 – 1995 and 2001 – 2006 for all prion disease and sCJD with no obvious improvement in the sensitivity for vCJD. Similar trends were observed when a broad case definition was applied (Tables 63 and 64).

Following adjustment for age group there was a statistically significant increase in sensitivity across year groups for all prion disease and sCJD (both narrowly and broadly defined) but not vCJD (Tables 65). For example, for each increase in year group these was a 7.5% (2.9 – 12.1) increase in the sensitivity of a death certificate diagnosis of all prion disease (narrowly defined). The corresponding value for sCJD was 8.4% (4.7 – 12.1).

Table 61 Sensitivity, specificity, positive and negative predictive value of CJD ICD coded in any position on a death certificate, according to disease subtype and age group (narrowly defined)

Disease subtype	Age group	Prevalence	Sensitivity	Specificity	PPV	NPV
All prion disease	All ages	79.0 (76.0 – 80.9)	75.9 (73.0 – 78.6)	84.0 (78.9 – 88.3)	94.6 (92.7 – 96.1)	48.6 (43.8 – 53.4)
	<50 years	86.0 (81.0 – 89.7)	83.2 (77.5 – 87.9)	86.1 (70.5 – 95.3)	97.3 (93.7 – 99.1)	46.3 (34.0 – 58.9)
	50 – 59 years	80.0 (74.0 – 85.7)	70.3 (62.5 – 77.4)	94.7 (82.3 – 99.4)	98.2 (93.6 – 99.8)	43.9 (33.0 – 55.3)
	60 – 69 years	82.0 (77.0 – 86.0)	77.4 (72.0 – 82.2)	83.3 (71.5 – 91.7)	95.5 (91.9 – 97.8)	44.6 (35.2 – 54.3)
	70 – 79 years	72.0 (67.0 – 77.3)	70.0 (63.5 – 76.1)	86.7 (77.5 – 93.2)	93.3 (88.2 – 96.6)	52.6 (43.9 – 61.1)
	≥80 years	65.0 (54.0 – 74.2)	78.3 (65.8 – 87.9)	63.6 (45.1 – 79.6)	79.7 (67.2 – 89.0)	61.8 (43.6 – 77.8)
sCJD	All ages	76.0 (73.0 – 79.0)	74.6 (71.1 – 77.8)	83.1 (77.4 – 87.9)	93.4 (91.0 – 95.3)	50.4 (45.1 – 55.8)
	<50 years	82.0 (68.0 – 92.0)	75.7 (58.8 – 88.2)	87.5 (47.3 – 99.7)	96.6 (82.2 – 99.9)	43.8 (19.8 – 70.1)
	50 – 59 years	80.0 (73.0 – 86.3)	73.0 (64.2 – 80.6)	93.3 (77.9 – 99.2)	97.8 (92.3 – 99.7)	45.9 (33.1 – 59.2)
	60 – 69 years	81.0 (76.0 – 85.3)	77.1 (71.4 – 82.1)	83.1 (71.0 – 91.6)	95.1 (91.2 – 97.6)	45.8 (36.1 – 55.7)
	70 – 79 years	72.0 (66.0 – 76.9)	71.2 (64.6 – 77.2)	86.7 (77.5 – 93.2)	93.2 (88.2 – 96.6)	54.1 (45.3 – 62.8)
	≥80 years	65.0 (54.0 – 74.2)	78.3 (65.8 – 87.9)	63.6 (45.1 – 79.6)	79.7 (67.2 – 89.0)	61.8 (43.6 – 77.8)
vCJD	All ages	77.0 (69.0 – 83.3)	88.4 (81.0 – 93.7)	88.2 (72.5 – 96.7)	96.1 (90.4 – 98.9)	69.8 (53.9 – 82.8)
	<30 years	91.0 (81.0 – 96.5)	88.1 (77.1 – 95.1)	100 (54.1 – 100)	100 (93.2 – 100)	46.2 (19.2 – 74.9)
	30 – 49 years	72.0 (59.0 – 82.5)	90.7 (77.9 – 97.4)	76.5 (50.1 – 93.2)	90.7 (77.9 – 97.4)	76.5 (50.1 – 93.2)
	≥50 years	48.0 (26.0 – 70.2)	80.0 (44.4 – 97.5)	100 (71.5 – 100)	100 (63.1 – 100)	84.6 (54.6 – 98.1)

Table 62 Sensitivity, specificity, positive and negative predictive value of CJD ICD coded in any position on a death certificate, according to disease subtype and year group (narrowly defined)

Disease subtype	Year Group	Prevalence	Sensitivity	Specificity	PPV	NPV
All prion disease	1990 – 1995	76.0 (71.0 – 80.4)	69.0 (62.8 – 74.7)	78.2 (67.4 – 86.8)	90.9 (85.8 – 94.6)	44.5 (36.0 – 53.3)
	1996 – 2000	75.0 (71.0 – 78.9)	74.6 (69.6 – 79.1)	87.0 (79.4 – 92.5)	94.5 (91.1 – 96.9)	53.2 (45.8 – 60.5)
	2001 - 2006	85.0 (81.0 – 88.6)	82.4 (77.8 – 93.7)	86.0 (74.2 – 93.7)	97.1 (94.4 – 98.8)	45.8 (36.1 – 55.7)
sCJD	1990 – 1995	72.0 (66.0 – 77.2)	69.2 (62.2 – 75.6)	77.6 (66.6 – 86.4)	88.8 (82.7 – 93.3)	49.6 (40.3 – 58.9)
	1996 – 2000	73.0 (68.0 – 77.8)	68.7 (62.3 – 74.6)	87.1 (78.0 – 93.4)	93.5 (88.7 – 96.7)	50.7 (42.3 – 59.0)
	2001 – 2006	83.0 (79.0 – 87.3)	83.8 (78.7 – 88.1)	84.6 (71.9 – 93.1)	96.4 (93.1 – 98.5)	51.2 (40.1 – 62.1)
vCJD	1996 – 2000	74.0 (64.0 – 82.0)	89.5 (80.3 – 95.3)	85.2 (66.3 – 95.8)	94.4 (86.4 – 98.5)	74.2 (55.4 – 88.1)
	2001 – 2006	85.0 (68.0 – 94.9)	92.9 (76.5 – 99.1)	100 (47.8 – 100)	100 (86.8 – 100)	71.4 (29.0 – 96.3)

Table 63 Sensitivity, specificity, positive and negative predictive value of CJD ICD coded in any position on a death certificate, according to disease subtype and age group (broadly defined)

Disease Subtype	Age group	Prevalence	Sensitivity	Specificity	PPV	NPV
All prion disease	All ages	67.0 (65.0 – 69.4)	79.1 (76.7 – 81.4)	81.6 (78.3 – 84.7)	89.8 (87.8 – 91.6)	65.7 (62.1 – 69.1)
	<50 years	81.0 (76.0 – 84.6)	85.9 (81.3 – 89.8)	88.1 (77.8 – 94.7)	96.7 (93.7 – 98.6)	60.2 (49.8 – 70.0)
	50 – 59 years	69.0 (63.0 – 74.5)	74.2 (67.4 – 80.3)	89.4 (80.8 – 95.0)	94.0 (88.9 – 97.2)	60.8 (51.7 – 69.4)
	60 – 69 years	70.0 (66.0 – 73.7)	80.5 (76.1 – 84.5)	79.1 (71.9 – 85.2)	89.9 (86.1 – 93.0)	63.8 (56.6 – 70.5)
	70 – 79 years	60.0 (56.0 – 64.8)	73.9 (68.4 – 78.9)	83.0 (76.8 – 88.1)	86.9 (82.0 – 90.9)	67.5 (61.1 – 73.5)
	≥80 years	47.0 (39.0 – 54.9)	80.0 (69.2 – 88.4)	70.6 (59.7 – 80.0)	70.6 (59.7 – 80.0)	80.0 (69.2 – 884)
sCJD	All ages	63.0 (61.0 – 65.7)	78.0 (75.1 – 80.7)	80.3 (76.6 – 83.6)	87.2 (84.6 – 89.4)	68.1 (64.2 – 71.8)
	<50 years	66.0 (53.0 – 77.4)	79.1 (64.0 – 90.0)	86.4 (65.1 – 97.1)	91.9 (78.1 – 98.3)	67.9 (47.6 – 84.1)
	50 – 59 years	68.0 (62.0 – 74.5)	77.0 (69.5 – 83.4)	87.1 (77.0 – 93.9)	92.9 (86.9 – 96.7)	63.5 (53.1 – 73.1)
	60 – 69 years	68.0 (64.0 – 72.6)	80.4 (75.8 – 84.5)	78.7 (71.4 – 84.5)	89.1 (85.1 – 92.4)	64.9 (57.6 – 71.7)
	70 – 79 years	60.0 (55.0 – 64.6)	75.0 (69.5 – 80.0)	82.8 (76.6 – 87.9)	86.8 (81.8 – 90.8)	68.8 (62.2 – 74.8)
	≥80 years	47.0 (39.0 – 54.9)	80.0 (69.2 – 88.4)	70.6 (59.7 – 80.0)	70.6 (59.7 – 80.0)	80.0 (69.2 – 88.4)
vCJD	All ages	73.0 (67.0 – 78.7)	90.6 (84.9 – 94.6)	(91.5 (81.3 – 97.2)	96.6 (92.3 – 98.9)	78.3 (66.7 – 87.3)
	<30 years	90.0 (83.0 – 95.5)	90.6 (82.3 – 95.8)	88.9 (51.8 – 99.7)	98.7 (93.1 – 100)	50.0 (24.7 – 75.3)
	30 – 49 years	69.0 (58.0 – 78.2)	91.9 (82.2 – 97.3)	85.7 (67.3 – 96.0)	93.4 (84.1 – 98.2)	82.8 (64.2 – 94.2)
	≥50 years	35.0 (20.0 – 53.5)	83.3 (51.6 – 97.9)	100 (84.6 – 100)	100 (69.2 – 100)	91.7 (73.0 – 99.0)

Table 64 Sensitivity, specificity, positive and negative predictive value of CJD ICD coded in any position on a death certificate, according to disease subtype and year group (broadly defined)

Disease Subtype	Year group	Prevalence	Sensitivity	Specificity	PPV	NPV
All prion disease	1990 – 1995	62.0 (57.0 – 66.1)	71.8 (66.1 – 771.)	69.9 (62.5 – 76.7)	79.3 (73.7 – 84.1)	60.8 (53.7 – 67.6)
	1996 – 2000	62.0 (58.0 – 65.8)	76.3 (71.8 – 80.3)	92.2 (88.2 – 95.3)	94.1 (91.0 – 96.4)	70.4 (65.1 – 75.3)
	2001 - 2006	76.0 (72.0 – 79.0)	85.3 (81.9 – 88.2)	78.2 (71.1 – 84.2)	92.5 (89.7 – 94.7)	62.9 (55.9 – 69.6)
sCJD	1990 – 1995	57.0 (52.0 – 61.8)	72.4 (66.1 – 78.2)	69.6 (62.1 – 76.4)	75.8 (69.5 – 81.4)	65.7 (58.3 – 72.6)
	1996 – 2000	58.0 (54.0 – 62.7)	71.2 (65.4 – 76.5)	92.3 (87.6 – 95.6)	92.8 (88.4 – 95.9)	69.8 (63.8 – 75.3)
	2001 – 2006	72.0 (68.0 – 75.7)	85.9 (82.1 – 89.2)	77.0 (69.5 – 83.4)	90.6 (87.1 – 93.3)	68.0 (60.5 – 74.9)
vCJD	1996 – 2000	65.0 (57.0 – 73.5)	89.4 (80.8 – 95.0)	91.1 (78.8 – 97.5)	95.0 (87.7 – 98.6)	82.0 (68.6 – 91.4)
	2001 – 2006	85.0 (75.0 – 91.8)	95.5 (87.3 – 99.1)	91.7 (61.5 – 99.8)	98.4 (91.6 – 100)	78.6 (49.2 – 95.3)

Table 65 Regression Co-efficients for changing sensitivity of death certificate diagnosis of CJD over time

Definition	Disease subtype	Year Group		Age group	
		Regression co-efficient (95% CI)	P value	Regression co-efficient (95% CI)	P value
Narrow	All prion disease	7.5 (2.9 – 12.1)	0.004	-1.5 (-4.1 – 1.1)	0.240
	sCJD	8.4 (4.7 – 12.1)	<0.001	-0.8 (-2.9 - 1.3)	0.435
	vCJD	12.8 (-31.0 – 56.6)	0.422	-3.2 (-30.0 – 23.6)	0.731
Broad	All prion disease	7.5 (3.2 – 11.7)	0.002	-1.7 (-4.2 – 0.7)	0.151
	sCJD	7.9 (4.4 – 11.4)	<0.001	-1.1 (-3.1 – 0.9)	0.242
	vCJD	14.0 (-15.6 – 43.7)	0.229	-3.9 (-22.1 – 14.2)	0.538

Further analyses assessed these values when CJD was recorded in the literal text or ICD coded in any position on the death certificate as it had been demonstrated in earlier analyses that this approach led to the greatest yield of suspect prion disease cases. These results are reported in Appendix 7. In brief, whilst the overall trends were similar, identifying CJD on a death certificate using both literal text and ICD codes resulted in a higher sensitivity without compromising specificity significantly.

Accuracy of ICD coding of death certificates

There was evidence of ICD coding inaccuracies on 6.3% (135) of all death certificates reviewed. Three quarters of inaccuracies occurred when CJD was recorded in the literal text on the death certificate but was not ICD coded.

In 99 suspect cases, CJD was recorded in the literal text of the death certificate (any position) but not ICD coded on the certificate (any position). The classification and disease subtypes in this group are outlined in Table 66.

Table 66 Classification and disease subtype of suspect cases for whom CJD was recorded in the literal text of their death certificate but not ICD coded

Case classification	Disease subtype			
	sCJD	vCJD	iCJD	Genetic prion disease
Definite or probable [1.0 or 2.0]	48 (34)	5	3 (3)	17 (16)
Possible [3.0]	3	0	0	0
Not CJD [4.1, 4.2 or 4.3]	21 (11)	1	0	0
Unclassifiable [0.0]	1	0	0	0

() indicates neuropathological diagnosis; [ ] indicates case classification

The cause(s) of death that were ICD coded on the death certificate of these suspect cases are described in Appendix 8. In one suspect prion disease cases, 'Other atypical viruses of the central nervous system' was ICD coded (A81.8). In the ICD coding manual a footnote indicates that this refers to Kuru. In a further eight suspect cases 'Atypical virus infection of central nervous system, unspecified', with a footnote indicating that this refers to 'Prion disease of central nervous system not otherwise specified' was ICD coded (A81.9). This group included five genetic prion disease cases, two sCJD and one iCJD case (all pathologically confirmed cases) and one

probable sCJD. Current algorithms for identifying prion disease on death certificates in the UK do not consider these codes.

In a further 36 suspect cases CJD was ICD coded, when CJD had not been mentioned in the literal text in any position on the death certificate. This group comprised largely of neuropathologically confirmed cases (Table 67). The causes of death as stated on the death certificate in these cases, according to position, are shown in Appendix 8.

Table 67 Case classification and disease subtype of suspect cases for which CJD was ICD coded but not recorded in the literal text of the death certificate

Case classification	Disease subtype			
	sCJD	vCJD	iCJD	Genetic prion disease
Definite or probable [1.0 or 2.0]	26 (26)	2 (2)	1 (1)	2 (2)
Possible [3.0]	0	0	0	0
Not CJD [4.1, 4.2 or 4.3]	4 (2)	1	0	0
Unclassifiable [0.0]	1	0	0	0

() indicates pathological diagnosis; [ ] indicates case classification

Over time there was a statistically significant reduction in the proportion of death certificates on which CJD was ICD coded when CJD had not been recorded in the literal text of the death certificate (P<0.001). As can be seen from Figure 63 this type of error has not occurred since 2003 and peaked between 1996 and 1998, a period over which ICD classification changed from the 9th to 10th revision. However there was no change in proportion of death certificates on which CJD was not ICD coded when CJD had been recorded in the literal text of the death certificate (P=0.687). This latter group accounted for the greatest number of ICD coding inaccuracies on death certificates. The effect persisted when individuals that received an ICD code of A81.8 and A81.9 were considered as having been correctly identified as having CJD (P=0.678).

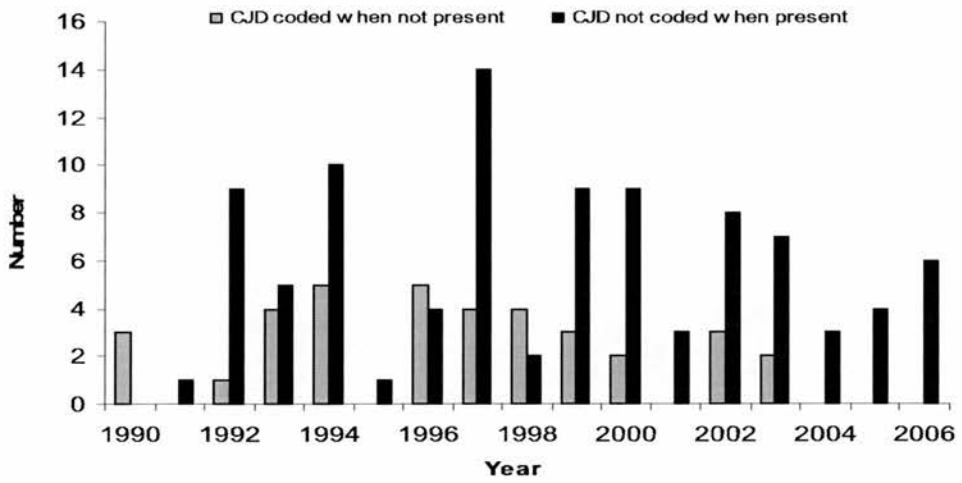

Figure 63 Number of inaccuracies in coding of death certificates in suspect prion disease cases according to year

Prion disease mortality rates

Figure 64 shows age-adjusted prion disease mortality rates according the method of case ascertainment. There was no significant difference in the mortality rates produced when all methods of case ascertainment employed by the NCJDSU were compared (definite or probable cases) to those produced by an examination of death certificates alone (all suspect cases identified from death certificate review).

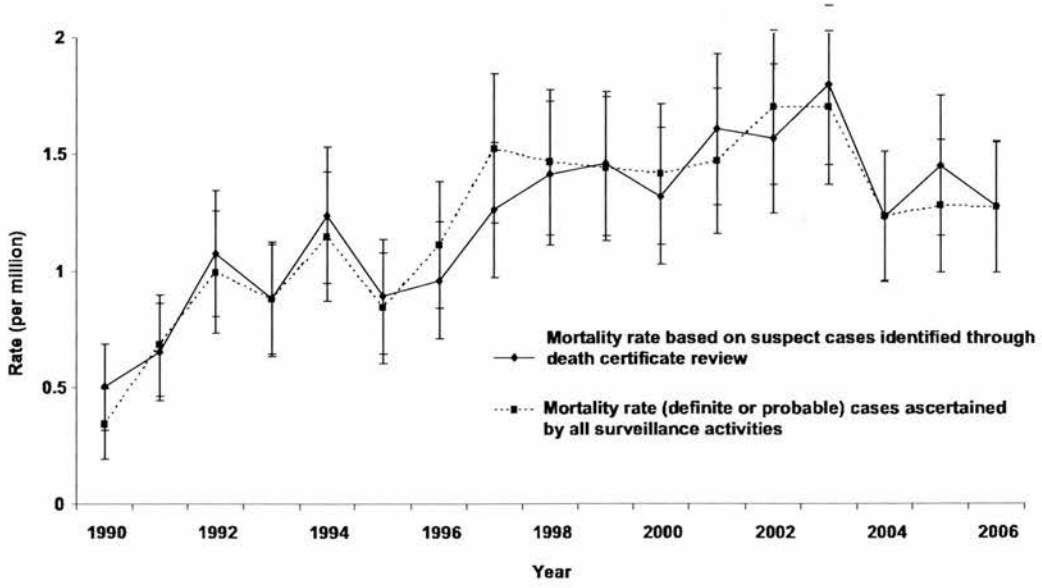

Figure 64 Age standardised prion disease mortality rate (per million population) according to method of case ascertainment

Sex-specific and age-specific mortality rates produced by examination of death certificates alone and all other surveillance methods are shown in Table 68. As can be seen there is no significant difference in either sex-specific mortality rates or age-specific mortality rates produced using these approaches to surveillance.

Table 68 Number and rate of deaths from prion disease according to sex and age group as ascertained by all surveillance methods (definite or probable cases) and death certificate review alone (all suspect cases identified on death certificates)

		All surveillance methods*		Death certificates only†	
		Number of deaths	Rate per million	Number of deaths	Rate per million
Sex	Male	610	1.26 (1.16 - 1.36)	605	1.25 (1.15 - 1.34)
	Female	619	1.21 (1.11 - 1.31)	622	1.22 (1.12 - 1.31)
Age group (years)	<20	32	0.13 (0.08 - 0.17)	30	0.12 (0.08 - 0.16)
	20-24	37	0.56 (0.38 - 0.74)	34	0.52 (0.34 - 0.69)
	25-29	54	0.75 (0.55 - 0.95)	51	0.71 (0.52 - 0.90)
	30-34	55	0.73 (0.54 - 0.92)	49	0.65 (0.47 - 0.83)
	35-39	38	0.52 (0.35 - 0.68)	30	0.41 (0.26 - 0.56)
	40-44	40	0.57 (0.40 - 0.75)	39	0.56 (0.38 - 0.73)
	45-49	45	0.69 (0.49 - 0.89)	42	0.64 (0.45 - 0.83)
	50-54	77	1.27 (0.98 - 1.55)	66	1.09 (0.82 - 1.35)
	55-59	119	2.13 (1.75 - 2.52)	109	1.95 (1.59 - 2.32)
	60-64	173	3.50 (2.98 - 4.02)	172	3.48 (2.96 - 4.00)
	65-69	194	4.26 (3.66 - 4.86)	209	4.59 (3.97 - 5.22)
	70-74	146	3.63 (3.04 - 4.22)	146	3.63 (3.04 - 4.22)
	75-79	142	4.41 (3.69 - 5.14)	147	4.57 (3.83 - 5.31)
	80-84	59	2.62 (1.96 - 3.29)	78	3.47 (2.70 - 4.24)
	≥85	15	0.84 (0.42 - 1.27)	24	1.35 (0.81 - 1.89)

*Definite and probable cases of CJD; † CJD recorded in literal text or ICD coded in any position

From these data the use of death certificates as the sole method of case ascertainment in the surveillance of prion diseases in the UK produces comparable mortality rates to the current approach to disease surveillance. However this is as a result of the inclusion of non-cases in the mortality figures (Figure 65). Relative to all other surveillance approaches the use of death certificates alone under-ascertains prion

disease cases in those under 60 years of age and over-estimates prion disease cases in those over 60 years of age (Figures 66 and 67).

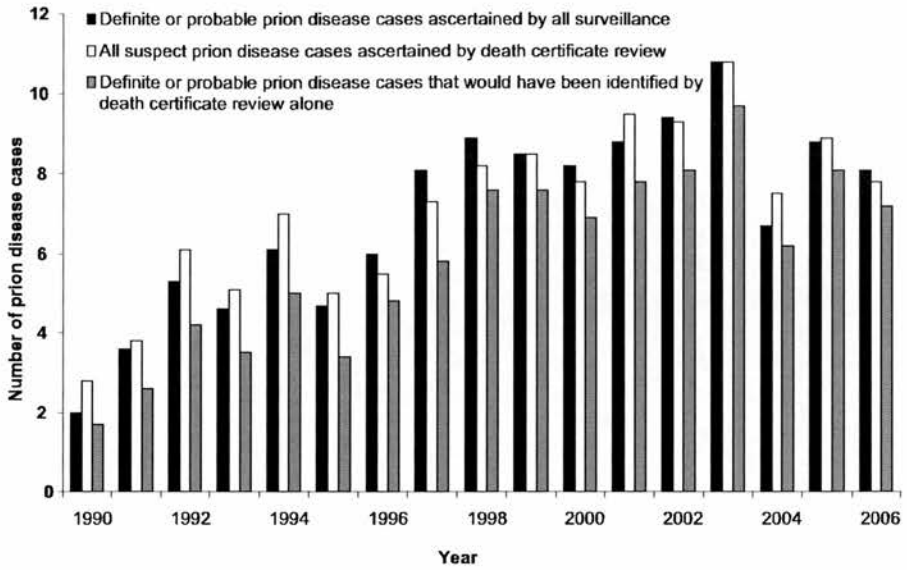

Figure 65 Number of prion disease cases as ascertained by all surveillance methods and by death certificate review alone, 1990 – 2006

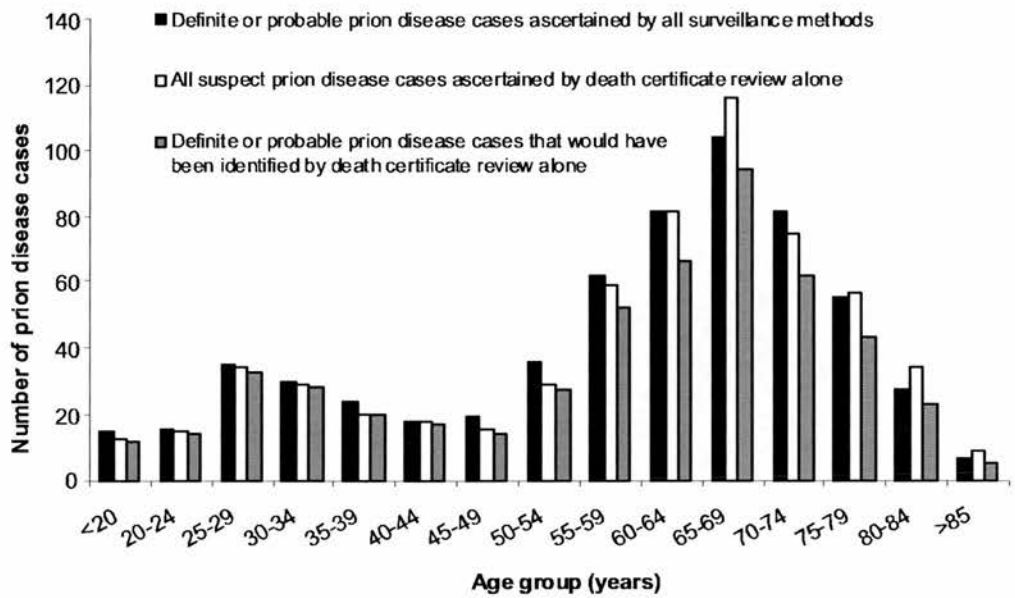

Figure 66 Number of prion disease cases ascertained by all surveillance methods and by death certificate review alone according to age group (men), 1990 – 2006

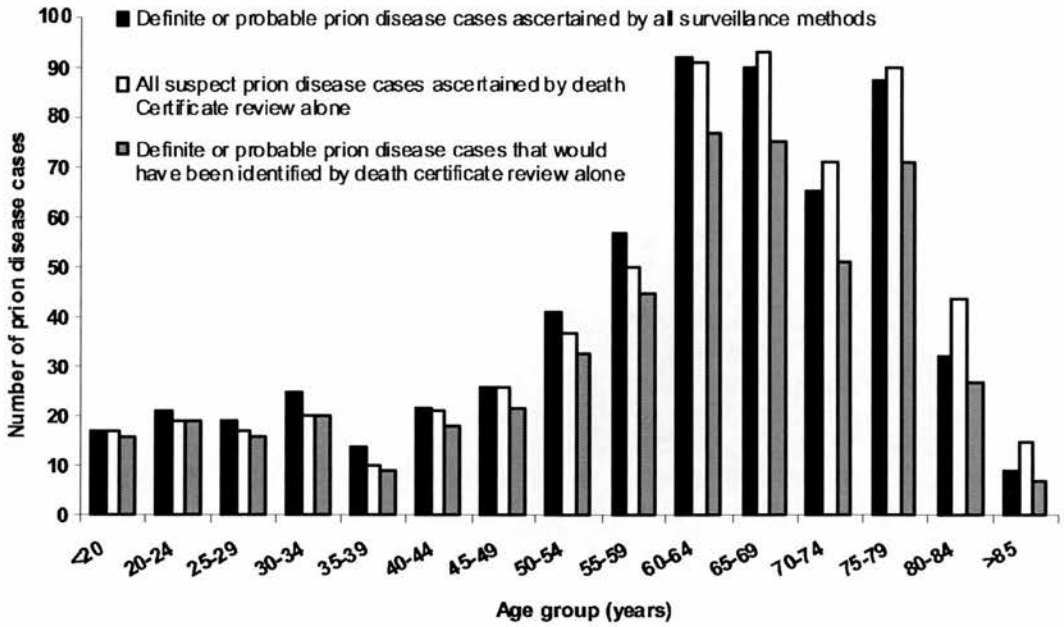

Figure 67 Number of prion disease cases ascertained by all surveillance methods and by death certificate review alone according to age group (women), 1990 – 2006

Summary of key findings

- In the UK the reliance on death certificates alone as a method of case ascertainment in the surveillance of prion disease has diminished with the establishment of systematic prospective surveillance.
- The yield from reviewing death certificates is maximal when both the literal text recorded on the death certificate and ICD codes ascribed to this text are reviewed, and when multiple causes of death are considered.
- The sensitivity and specificity of a death certificate diagnosis of prion disease in the UK are high. This is greatest in those aged under 50 years of age and for vCJD.
- The sensitivity of a death certificate diagnosis of prion disease and sCJD has increased over time following adjustment for age.
- Use of death certificates alone produces similar sex-specific, age-specific and age-adjusted prion disease mortality rates to the combined surveillance approaches currently adopted in the UK. However, this is as a result of the inclusion of non-cases and the exclusion of cases.

Discussion

In this chapter I examined the role of death certificates in the surveillance of prion disease in the UK. This is the first longitudinal study to examine the diagnostic utility of death certificates in the surveillance of prion disease over time, adjusting for age and disease subtype. Using data collected prospectively over 16 years from 2,154 suspect prion disease cases, including over 1200 pathologically confirmed cases, this is the largest study of its kind. To the best of my knowledge this is the only contemporary study that has explored the optimal approach to using death certificates in the surveillance of prion disease. The key findings from this study will be discussed here.

The use of death certificates in the ascertainment of suspect prion disease

As systematic prospective surveillance has gained momentum in the UK the reliance on death certificates in the ascertainment of suspect prion disease cases has fallen. Will noted that 42% of definite or probable CJD cases in the UK were ascertained by death certificate review alone in the 1970s, falling to 13% in the period 1980 – 1984 and falling further to 6% in 1990 – 1992 (the first years of prospective surveillance).(72) In the present study just 30 definite or probable prion disease cases were ascertained by death certificate review alone from 1990 through 2006, representing 2.4% of all definite or probable cases ascertained by the NCJDSU over this period; in 2006 no cases were ascertained by death certificate review alone.

Surprisingly few studies are available for comparison. Most studies reporting the clinico-pathological epidemiology of prion disease as ascertained by surveillance systems do not describe the source of referrals in sufficient detail to determine the relative contribution that death certificates have made to case ascertainment. In Australia, Collins *et al* report that a quarter of definite or probable prion disease cases ascertained by the surveillance system from 1970 through 1999 were identified by death certificate review alone. Prospective surveillance was not initiated in Australia until 1993, prior to this surveillance was retrospective which might explain the high proportion of cases ascertained by this route.(238) Perhaps the most widely

cited study is by Davanipour *et al* who examined 69 pathologically confirmed sCJD cases and 5 non-cases ascertained from 11 states in the USA between 1986 and 1988.(227) This study took place prior to systematic prospective disease surveillance in the USA, at a time when there was limited understanding of the clinico-pathological heterogeneity of sCJD and limited diagnostic technology. The authors contacted neuropathologists and hospitals requesting information on sCJD and reviewed routinely collated mortality data from death certificates. In the USA each episode of care within a hospital stays is ICD coded with multiple diagnoses at the time of discharge for billing purposes. The authors requested information on all hospital stays ICD coded 046.1 (under the rubrics CJD). The same approach to identifying sCJD on death certificates was adopted. It is unclear from the manuscript whether multiple causes of death were considered. The response rate to the study was extremely low. Just 29% of neuropathologists and 36% of hospitals contacted by the authors responded to a request for information about suspect sCJD cases over the study period. As noted in previous chapters of this thesis, given the nature of sCJD it is likely that a clinical case would seek medical attention and be admitted to hospital during the course of their clinical illness. Moreover in typical cases that death will occur after a short illness, most likely in hospital. It is very likely then that the high proportion of sCJD cases ascertained by death certificate review alone in this study has arisen as a result of the poor response rate in other groups. For example, had the response rate from hospitals been higher the authors may have found that a greater proportion of cases were identified through review of hospital case records in addition to death certificates rather than death certificates alone. Whilst a reported 42% of all pathologically confirmed sCJD cases were ascertained by review of death certificates alone, just 27% of the suspect sCJD cases initially identified by the authors were included in the study. Just one out of every six suspect sCJD case identified by death certificates alone was included in the final sample as the authors were unable to obtain clinical or neuropathological data to verify the diagnosis in other suspect cases. These data should then be interpreted with caution as they are likely to be subject to bias.

More recently, Conti *et al* conducted a data linkage study in Italy, using routinely collected mortality data from death certificates to determine the completeness of active CJD surveillance from 1993 through 1999.(226) It is noteworthy that the surveillance protocol in Italy does not include routine review of death certificates. Overall 64 suspect CJD cases were identified by death certificate review that had not been identified by the surveillance unit. Annually this figure fell from 12 suspect cases in 1993, representing 22% of all suspect CJD deaths, to 5 suspect cases in 1999, representing 4% of all suspect CJD deaths. The major limitation of this study is that the final diagnosis in death certificate only cases is unclear. The authors did not attempted to validate the diagnosis through the review of medical case records instead rather dubiously stating that it is

> *"reasonable to assume that more that 90% of the 64 deaths recorded only by ISTAT [death certificates] are really cases of TSE who were not reported to the CJD register".*

The optimal approach to using death certificates in the surveillance of prion disease

A number of approaches to examining death certificates in the surveillance of prion disease have been adopted, although few studies justify the approach taken. The vast majority of studies examine only the underlying cause of death as ICD coded on the death certificate. The ICD codes selected, and the periods of transition from ICD 9 to ICD 10, vary internationally. For example in the study by Conti *et al* temporal trends in CJD mortality from 1993 through 1999 were reported using the ICD codes 046.1 and 331.5 recorded as the underlying cause of death.(226) Information regarding the completion of death certification in Italy and the process for ICD coding these data was not provided. Doi *et al* reported temporal trends in CJD mortality in Japan from 1979 through 2004 using the underlying cause of death ICD coded as 046.1 (ICD 9 until 1994) and A81.0 (ICD 10 from 1994).(47) Whilst the authors report that a clinician is responsible for completing the death certificate it is not clear who is responsible for ICD coding these data. In Canada, ElSaadany *et al* produced a similar study examining the underlying cause of death ICD coded under the rubrics 046.1 when describing mortality trends from 1979 through 1997.(51) The authors justified the use of the underlying cause of death by stating that

"as CJD is considered a disease that contributes to death directly, our study used the underlying cause of death code to identify all cases of CJD".

This assumes that those certifying CJD cases correctly complete death certificates and in turn that those ICD coding death certificate data correctly identify the underlying cause of death from the certificate. The examples provided above are from three countries in different continents. Despite using death certificate data to describe temporal trends in CJD mortality, none of the studies described the process of completion of death certificates and coding of death certificate data in their respective countries. Nor do they provide any information on the accuracy of ICD coding.

The most comprehensive and contemporary study of prion disease mortality using death certificate data is from Holman and co-workers in the USA.(50) The authors examine multiple causes of deaths ICD coded under the rubrics of 046.1 (ICD 9 1979 – 1998) and A81.0 (ICD 10 1999 – 2006). Following a change in ICD coding in 1999 (from ICD 9 to ICD 10), a coding related under-ascertainment of prion disease cases was noted. At this time software was introduced to allow literal text search for CJD and CJD related diagnoses on death certificates. The introduction of this software was however staggered such that in 1993 less than half (18) of all States were using this software. It was not until 2003 that all states were using the software. This may have introduced information bias. Whilst the authors used additional surveillance efforts to validate the diagnosis of CJD in selected groups, for example in those under 55 years of age, not all diagnoses were validated by neuropathological examination or review of medical records, again possibly introducing bias. The use of software to search the literal text of death certificates allowed the authors to exclude those death certificates on which CJD was ICD coded but not recorded and conversely include those certificates on which CJD was recorded but not ICD coded. However, the authors do not provide data to quantify the degree of coding inaccuracies which is a major limitation of the study. To the best of my knowledge there are no contemporary published studies reporting the coding accuracy of death certificates in prion disease.

In the UK death certificates are reviewed as a 'safety net' as part of routine surveillance practice. Death certificates ICD coded under the rubrics of 046.1 or 331.9 (ICD 9) and A81.0 or F02.1 (ICD 10) are requested from the GROS / ONS. The ICD 9 code 331.9 is not a CJD specific code but certificates coded under this rubric were requested in an attempt to identify additional cases. The death certificates of all suspect prion disease cases referred to the NJCDSU are also reviewed. In this chapter I have shown that examining the literal text recorded on death certificates in addition to ICD codes results in a higher yield of suspect cases. The greatest yield comes from examining multiple causes of death rather than the underlying cause of death. For example examining CJD ICD coded (046.1, A81.0 or F02.1) as the underlying cause of death identified 78.3% of definite or probable prion disease cases. Interestingly the figure for CJD recorded as the underlying cause of death was virtually identical suggesting that in the UK coders are correctly identifying and coding the underlying cause of death on death certificates. Examining the literal text and aforementioned ICD codes in any position on the death certificate identified 85.3% of all definite or probable prion disease cases, equating to an additional 83 cases. This also resulted in the identification of an additional 25 non-cases (14 of which had a neuropathologically confirmed diagnosis). The latter point highlights the need for review of medical and neuropathological records in the verification of data from death certificates. The 2010 study by Holman *et al* reported that CJD was recorded as the underlying cause of death in 83% of deaths in individuals identified from disease surveillance activities and death certificate review as having prion disease, in the USA between 1979 and 2006.(50) However as previously noted the authors did not verify the diagnosis in all suspect cases. Whilst the diagnosis was verified in some groups, the proportion of those with a verified clinical or pathological diagnosis of prion disease, that had CJD recorded as the underlying cause of death was not reported.

ICD coding inaccuracies

An additional advantage of examining the literal text on death certificates was that it allowed an assessment of coding inaccuracies. Approximately 6% (135) of death certificates reviewed in this study had evidence of inaccuracies in ICD coding. Three quarters of these inaccuracies related to CJD not being ICD coded when it was

recorded on the death certificate. There is no evidence of any improvement in this area over time. It is possible that suspect prion disease cases in the UK have been missed as a result of coding inaccuracies. However all of the 73 definite or probable cases that had CJD recorded in the text of their death certificate but not ICD coded had been ascertained by the NCJDSU prior to death certificate review, therefore it is likely that the number of missed cases as a result of coding inaccuracies would be small.

Over the entire study period 53 pathologically confirmed non-cases had CJD either recorded in the literal text or ICD coded on their death certificate. In the UK death certificates may be issued prior to the completion of neuropathological examination. For example a death certificate in a suspect CJD case may be issued following autopsy but prior to immunohistochemical results from brain tissue being available. The latter process can take several weeks and given that a death certificate is required prior to burial or cremation this would be an unacceptable delay in proceedings for the patient's relatives. As further information becomes available the death certificate should be updated to accurately reflect the final diagnosis. Updated death certificates have not been received by the NCJDSU in these cases. It is unclear whether this is due to updated certificates not being issued, or the GROS/ONS not forwarding these certificates to the NCJDSU. However, this highlights the need to verify the information obtained from death certificates. Through routine surveillance activities the NCJDSU has obtained neuropathological reports and in almost half (n=23) reviewed neuropathological material, from these cases to verified that final diagnosis.

Practical issues in relation to reviewing death certificates

An examination of the literal text on death certificates is challenging. In the process of this study I manually reviewed all death certificates received by the NCJDSU in the 16 years since the inception of prospective surveillance. This was a labour and time intensive task. Moreover this study was only feasible because the GROS/ ONS historically sent a copy of the death certificate to the NCJDSU. A recent change in the GROS/ ONS protocols for data sharing mean that they now no longer send death certificates to the NCJDSU, instead providing ICD coded death certificate data only.

This study has highlighted the importance of reviewing both ICD codes and literal text on death certificates. Future research should focus on the development of software, appropriate to the UK which could be applied to the literal text of death certificate data collated nationally. In addition to increasing the yield of suspect cases identified by death certificates this approach will allow a regular and systematic assessment of coding inaccuracies and identification of the most useful ICD codes for the purposes of surveillance. This would also allow an assessment of whether prion disease cases, not referred to the NCJDSU by other sources and not identified by an examination of ICD codes on review of death certificates, are being missed in the UK. There would of course be cost implications to software development and potential difficulties in obtaining agreement from the GROS / ONS in Scotland, England, Northern Ireland and Wales to run such software. Nevertheless these issues are likely to be surmountable as Holman and colleagues have demonstrated in the USA.

Data from this study suggest that the ICD codes currently utilised by the NCJDSU in the UK, whilst consistent with the approach adopted internationally, may not be optimal. For example the ICD 10 code F02.1 was not recorded in any position on the death certificate of any suspect prion disease case. However, nine prion disease cases were coded under the rubrics A81.8 and A81.9, CJD specific codes and are not currently examined by the NCJDSU. The latter cases would have been missed had surveillance been reliant on death certificate review alone. The ICD codes that are selected for use in the review of death certificates for disease surveillance should be regularly reviewed to ensure that these are optimal.

The diagnostic value of death certificates in the surveillance

There is a paucity of studies examining the diagnostic value of death certificates in the surveillance of prion disease. The most widely referenced study is by Davanipour *et al.*(227) The limitations of this study have been outlined and should be considered in interpreting the findings of this study. The sensitivity, specificity, PPV and NPV of death certificates in this population was 79.9% (68.3 – 88.4), 0% (0 – 52.2%), 91.7% (81.6 – 97.2) and 0% (0 – 23.2). These data indicate that in this highly selected population death certificates correctly identified four out of every five sCJD

case. However, death certificates were very poor at excluding individuals without sCJD. Despite the inherent limitations of this study, it is considered by some to have demonstrated that death certificates are able to identify over 80% of sCJD cases in the USA. Few citing authors acknowledge that Davanipour *et al* conclude their manuscript by noting that whilst death certificates are a readily available and low cost means of ascertaining CJD cases,

> *"review of medical records and pathology reports and verification of diagnosis* ***must*** *follow the identification of potential cases by death certificates."(227)*

In Italy, Conti *et al* linked death certificate data to the CJD surveillance register.(226) Death certificates identified just 46.6% of all definite or probable CJD cases ascertained by the surveillance system between 1993 and 1999. This fell from 47.2% in 1993 to 42.0% in 1999. Under ascertainment of CJD cases based on death certificate review was greatest in those aged 60 years and over. The reason for the relatively low sensitivity of death certificates in this population is unclear. The sensitivity of death certificates fell over the study period despite increasing case ascertainment by the surveillance unit. The authors attribute this in part to poor communication between the surveillance unit and certifying clinicians. In this study 20% of CJD cases that did not have CJD ICD coded on their death certificate as the underlying cause of death, did not have a neurological disorder ICD coded as the underlying cause of death either. In the present study just over half of all definite or probable prion disease cases that did not have CJD recorded as their underlying cause of death had a neurological diagnosis recorded as the underlying cause of death. Following a careful examination of underlying causes of death I considered it likely that the immediate cause of death was incorrectly recorded as the underlying cause of death in 7.6% of definite or probable prion disease cases overall. In all other cases was plausible that the underlying cause of death as recorded on the death certificate was truly the underlying cause of death, even in the presence of a clinically apparent neurological condition. Improvements in the quality of death certification may be achieved through additional training of clinicians and a greater appreciation within the clinical community of the value of routinely collected mortality data in epidemiological research and disease surveillance. An examination

of multiple causes of death would circumvent these issues in the UK. However, it is unclear from the data presented in the study by Conti *et al* whether an examination of multiple causes of death would have led to an increase the sensitivity of death certificates in the Italian population.(226)

In the present study the overall sensitivity, specificity, PPV and NPV of CJD ICD coded in any position on death certificates was 75.9% (73.0 – 78.6), 84.0% (78.9 – 88.3), 94.6% (92.7 – 96.1) and 48.6% (43.8 – 53.4) when pathologically confirmed cases were considered and 79.1% (76.7 – 81.4), 81.6% (78.3 – 84.7), 89.8% (87.8 – 91.6) and 65.7% (62.1 – 69.1) when definite or probable cases were considered. The sensitivity of death certificates was highest in those under 50 years of age. There was no discernable trend in sensitivity across age groups when those over 50 years of age where examined. Values for sCJD were slightly lower that for all prion disease but followed similar trends. The sensitivity, specificity, PPV and NPV for death certificates in vCJD was exceptionally high at 88.4% (81.0 – 93.7), 88.2% (72.5 – 96.7), 96.1% (90.4 – 98.9) and 69.8 (53.9 – 82.8) respectively. This is perhaps unsurprising given vCJD typically affects a younger age group, less than 50 years old, and overall the sensitivity of death certificates is greatest in this age group. Moreover in the UK vCJD has had an exceptionally high profile and there are significant implications for patients, families and health care providers of a diagnosis of vCJD, not least a complex compensation system that requires a diagnosis of vCJD to be reached on the balance of probability. It should be noted however that, particularly in the early years of the vCJD primary epidemic, there was considerable stigma associated with a diagnosis of vCJD such that families may have requested that vCJD was not recorded on a death certificate (personal communication R. Knight).

Over time there was a statistically significant increase in the ability of death certificates to identify all prion disease cases and sCJD cases (pathologically confirmed and definite or probable cases). Will noted an increase in the sensitivity of death certificates (definite or probable CJD cases) over time in the UK, from 39% in the 1960s to 67% in the early 1990s.(72) The present study has shown that the

sensitivity has continued to increase over time, from 71.8% (66.1 – 77.1) in the period 1990 – 1995 to 85.3% (81.9 – 88.2) in the period 2001 – 2005. Following adjustment for age this increase in sensitivity over time persisted. It is likely that this increase sensitivity of death certificates has arisen as a result of systematic prospective surveillance in the UK. Increasing awareness of all forms of prion disease as a result of the primary vCJD epidemic may also have contributed to this trend. A trend toward an increase in the sensitivity of death certificates in vCJD was observed however this was not statistically significant. It may be that analyses in this group were underpowered due to the smaller sample size (146 cases with pathologically confirmed diagnoses and 218 cases with a clinical or pathological diagnosis).

In this study it was possible to compare the values of sensitivity, specificity, PPV and NPV obtained from death certificates that identified CJD based on ICD coding, as is the current approach in the surveillance of CJD in the UK, to CJD identified by examination of literal text and ICD codes from multiple causes of death. Reassuringly, the sensitivity of death certificates increased when multiple causes of death and both text and ICD coding were considered without a substantial fall in specificity. This suggests that adopting the latter approach to surveillance in the UK would increase the yield of prion disease cases identified without resulting in a significant increase in the number of non-cases identified.

Prion disease mortality rates

There was no significant difference in sex-specific, age-specific or age-adjusted prion disease mortality rates produced by examination of death certificates alone and produced by data obtained from definite or probable prion disease cases ascertained by combined surveillance activities. Reassuringly the mortality rates produced in this study by both methods were consistent with international reports of prion disease mortality and followed recognised age-specific trends. The age-specific mortality rates produced by death certificates alone led to a slight under-estimation of mortality rates in those under 50 years of age and a marginal over estimation in mortality rates in those over 60 years of age. However the 95% confidence intervals overlapped indicating that these differences were not statistically significant. This

equilibrium has arisen because whilst death certificates identify over 80% of prion disease cases, they also incorrectly identify approximately 20% of non-cases. In the UK death certificate review alone would then produce a reasonable estimate of prion disease mortality rates.

Strengths and limitations

This study examined death certificate data on all suspect prion disease cases referred to the NCJDSU from 1990 through 2006. Death certificates were unavailable for a small number of suspect cases. It is unlikely that the exclusion of these individuals introduced significant bias to this study. A direct visual inspection of the death certificate allowed examination of material that might otherwise have been available had an automated computerised search retrieved records. This allowed subjective assessment of the content of the certificate, particularly in relation to the literal text. Whilst this approach was useful for the purposes of this research it is not a practical approach that could be easily transferred to routine surveillance practice. Estimates of the sensitivity and specificity of a death certificate diagnosis of CJD produced in this study are based on an examination of the death certificates from suspect prion disease cases referred to the NCJDSU in life or following death, for whom it was possible to obtain further information (clinical and pathological) to verify diagnoses. This study does not however consider the hundreds of thousands of deaths in the UK annually that are not referred to the NCJDSU in life and that do not have reference to prion disease on their death certificate. Assuming that the NCJDSU achieves high levels of case ascertainment, it is likely that the present study provides a reasonable estimate of the sensitivity of a death certificate diagnosis of CJD but a biased estimate (underestimating) of the true specificity of a death certificate diagnosis of CJD.

Conclusions

In the UK the use of death certificates in the ascertainment of suspect prion disease cases has diminished over time with the establishment of systematic prospective surveillance. Death certificate review is a sensitive and specific way to identifying prion disease. However this is likely to have arisen as a direct consequence of systematic prospective surveillance. The yield from examining death certificates is

maximal when both ICD codes and literal text in multiple causes of death are considered. Death certificates in the UK produce a valid estimate of prion disease mortality for monitoring trends over time. However, this is as a result of the inclusion of a small number of non-prion disease cases at the expense of the exclusion of a small number of prion disease cases. These findings highlight the importance of the verification of data from death certificates through an examination of clinical and pathological data.

Chapter 6: General Discussion and Conclusions

The thesis evaluated various aspects of the surveillance of CJD in the UK, from 1990 through 2006. In Chapter 2, the epidemiology and diagnostic features of prion disease in the UK were described using data collected by the NCJDSU over 16 years of systematic prospective surveillance. In Chapter 3, using an established framework, the performance of NCJDSU was formally evaluated. In Chapter 4, a study to validate the NCJDSU operational criteria for the assessment of EEG in case classification of sCJD was carried out. Finally, in Chapter 5, the use of death certificates in the surveillance of prion disease in the UK was examined. The key findings from this thesis are summarised below. A discussion of these findings, placing them in context of the future challenges of prion disease PHS in the UK follows.

Summary of key findings

- Systematic prospective surveillance of CJD was initiated in the UK in 1990, the aim of which was to detect any change in the clinico-pathological phenotype of CJD that could be attributable to BSE in cattle; in 1996 this was realised.
- The primary vCJD epidemic in the UK has been smaller than predicted and in decline since 2000.
- Secondary transmission of vCJD via the transfusion of labile blood components has been described. The risk of iatrogenic transmission of vCJD via other health care associated procedures is unquantified.
- The pathogenesis of vCJD is poorly understood. The number of asymptomatic but potentially infectious individuals in the population is unknown.
- Uncertainties remain around the susceptibility of non-methionine homozygote genotypes to vCJD and the phenotypic expression of disease in such individuals; long incubation periods are likely.

- The public health implications and the unanswered questions about the epidemiology and pathogenesis of the vCJD, provide an imperative to continue the PHS of all forms of prion disease for the foreseeable future.
- The NCJDSU performed well between 1990 and 2006 and would be in a strong position to continue to undertake PHS of prion disease in the UK.
- However, falling post mortem rates have led to an increasing reliance on clinical diagnostic criteria in the UK and there is credible evidence that the use of investigations to support a diagnosis in suspect sCJD and suspect vCJD cases is sub-optimal and differential.
- In addition, the PPV of the system rose over the study period which in the face of falling referral rates may compromise the ability of the NCJDSU to detect atypical disease phenotypes or entirely novel prion diseases.
- It was not possible to determine directly whether systematic under-ascertainment of prion disease have occurred as a result. There was however evidence to suggest possible under-ascertainment of genetic prion disease, which may be indistinguishable from sCJD, in the UK.
- NCJDSU operational criteria for the assessment of EEG in case classification of sCJD were validated; given the subjective elements to these criteria it is unclear whether these findings would be reproducibility in the hands of less experienced clinicians.
- Clarification of the more subjective elements of the NCJDSU operational criteria for the assessment of EEG in case classification are required to ensure stability and sustainability.
- The sensitivity of EEG in sCJD is low but specificity high; EEG remains a useful, non-invasive test in the investigation of suspect sCJD cases, if used in conjunction with other diagnostic tools.
- The use of death certificates in the ascertainment of suspect prion disease cases has diminished with the establishment of systematic prospective surveillance.
- Death certificate review is a sensitive and specific way to monitor prion disease mortality in the UK. However the high diagnostic value of death certificates in the UK is likely to have arisen as a direct consequence of systematic prospective

surveillance. The impact that a change in the surveillance protocol would have on the value of death certificates in this context is unclear.

Future challenges in PHS of human prion diseases

A primary aim of systematic prospective PHS of CJD in the UK was the identification of a change in the clinico-pathological phenotype of CJD that could be attributable to BSE in humans. Following the identification of vCJD, a key driver for on-going surveillance in the UK was to understand the public health implications of this disease and thus facilitate prompt public health action where required. The large epidemic of vCJD that was feared by some has not occurred and the primary epidemic of vCJD has been in declined in the UK since 2000. Our knowledge of the epidemiology of vCJD, and other human prion diseases, has been expanded dramatically through systematic prospective surveillance in the UK. Although uncertainties exist around the potential for a secondary epidemic of vCJD models predict that this too will be small.(172) Currently the only recognised route of secondary transmission of vCJD is via the transfusion of labile blood components. Public health control measures have been put in place to minimise the risk of health care associated iatrogenic transmission of vCJD. Progress is being made toward the development of a blood test that could be used to screen blood and organ donations for abnormal prion protein which would potentially further reduce the size of any secondary epidemic. In this context and in the current financial climate systematic prospective surveillance in the UK, in its current form, may no longer be considered viable and alternate models of disease surveillance may be sought.

Referral to the NCJDSU

Unlike many other prion disease PHS systems, the UK system aims to identify and review suspect cases in life. The enables the collection of detailed clinical, diagnostic and epidemiological data, used to investigate putative risk factors for disease and evaluated diagnostic technology, and also facilitate prompt public health action where required. The ability of the NCJDSU to detect phenotypically diverse or novel prion disease in life will be in part determined by the referral and review of atypical cases. The willingness of individuals to refer suspect prion disease cases to the

NCJDSU and of patients and their relatives to participate in surveillance was demonstrated in Chapter 3. A high proportion of all suspect cases referred to the NCJDSU were visited and assessed by a NCJDSU neurologist. However rates of referral to the NCJDSU have fallen in recent years whilst the PPV of the system has increased. This may compromise the NCJDSU's ability to detect atypical disease phenotypes or entirely novel human prion diseases. The NCJDSU should consider whether the current ratio of referrals to cases, definite or probable, not simply definite, is appropriate. Any effort to increase referral rates through enhanced contact should also consider the broadening range of health care professionals referring to the NCJDSU.

Should prion disease be notifiable in the UK?

Should CJD become a notifiable disease in the UK to aid referral? Compulsory notifiable disease reporting appears to have had mixed effects on referral rates elsewhere. In Slovakia referrals to the surveillance system fell following the introduction of compulsory notification as referrer's awaited case confirmation before contacting the PHS system.(24) Other countries report no significant change in referral rates.(230) A case definition is required for compulsory notification. In the UK the identification of novel human prion diseases is a key objective of the surveillance system. It is extremely unlikely that such cases would meet established diagnostic criteria. Broad criteria for referral to the NCJDSU are therefore essential. In addition, part of the success of the NCJDSU appears to be the value that referring clinicians place on the ability to discuss a complex clinical case with a colleague who may be able to offer advice and support in the investigation and management of that case. Compulsory reporting may prohibit or inhibit such a dialogue. In this context I would suggest that compulsory notification might act as a barrier rather than facilitator, to the notification of atypical or unusual clinical or pathological cases.

Diagnostic technology

The diagnosis of CJD in life requires the application of diagnostic criteria based on clinical features and supportive investigations. If the clinical phenotype of disease changes these diagnostic criteria may be of limited value. This has occurred to some extent in sCJD. An increasingly diverse clinical phenotype has been described in

sCJD which varies according to *PRNP* Codon 129 genotype and prion protein type. In turn, diagnostic investigations vary in sensitivity across the spectrum of molecular subtypes. A recent manuscript by Zerr *et al* noted that the sensitivity of the diagnostic criteria for sCJD could be improved by the addition of MRI findings which would be particularly useful in ascertaining rarer molecular subtypes of the disease. The NCJDSU has been shown to be flexible to responding to changing demand, such as changing diagnostic criteria, in the past. However it should be considered that an unforeseen impact of increasingly sensitive and specific clinical diagnostic criteria in life may be a further reduction in post mortem rates following death.

Rather uniquely the NCJDSU identified vCJD prior to an established case definition being available for the disease. The first few cases of vCJD were identified through autopsy examination; young age at symptom onset may have increased the likelihood of consent being granted for post mortem examination. Further cases were identified by direct referral to the NCJDSU from families, in part as a result of the intense media interest. It is worth reflecting on whether vCJD would have been so quickly ascertained and characterised had it emerged in the elderly population; there is already evidence to suggest under-ascertainment of sCJD in this population despite surveillance efforts. The development of diagnostic criteria for vCJD was aided considerably by the remarkably consistent clinico-pathological phenotype. Despite this due to the small number of cases it was almost 15 years before the first study validating the diagnostic criteria was published. The assessment of many investigations including EEG and MRI is subjective. Validation of diagnostic criteria for vCJD was only possible because cases and diagnostic investigations had been reviewed centrally by individuals with considerable experience of human prion diseases and a sizable number of cases and non-cases had undergone post mortem examination to obtain a neuropathologically confirmed diagnosis. Falling post mortem rates will not only limit our ability to detect novel or phenotypically diverse form of prion disease in humans, but also our ability to develop and validate diagnostic criteria to facilitate the on-going identification of the cases in life and death.

Alternate models of PHS

The model of systematic prospective CJD surveillance in the UK is both labour and time intensive, requiring considerable clinical expertise and associated with significant financial cost. Whilst the NCJDSU has performed well, in the current financial climate, as the primary BSE and vCJD epidemics wanes and novel threats to human health emerge, the political will and public health imperative to continue to fund systematic prospective CJD surveillance in its current model may diminish. Alternate financially sustainable models of PHS surveillance may be sought. Much of the cost associated with prospective systematic surveillance in the UK relates to the cost of visits during which the NCJDSU has direct contact with cases and their families. Many surveillance systems, for example France, Australia and USA do not have direct contact with cases and yet are able to produce broadly comparable surveillance data, including in the examples of France and USA ascertaining cases of vCJD.

Ascertaining cases through laboratory results: CSF 14-3-3 protein

The PHS model in France relies primarily upon review of requests for CSF 14-3-3 protein. Adoption of this model in the UK would prove problematic for a number of reasons. The UK CSF 14-3-3 protein laboratory received 330 requests in 2008 (the end of follow up for this study), twice as many as formal referrals to the NCJDSU. However, only 1 in every 5 samples was from an individual that met the diagnostic criteria as a definite or probable prion disease case, indicating that the PPV of referrals to this service low. Combined these data provide some evidence that CSF 14-3-3 protein is being used, inappropriately, as a screening test by some clinicians. A negative investigation does not mean that a suspect case does not have prion disease. This is an important point. CSF 14-3-3 protein is a test that has been validated for sCJD in a specific reference population; outside this reference population measures of sensitivity and specificity are invalid. CSF 14-3-3 protein is not of value in vCJD for example and may not then be pursued. In sCJD, false positive rates of up to 16% have been quoted for CSF 14-3-3 protein.(231) These individuals would require careful review and follow up, as would individuals with negative CSF 14-3-3 protein to capture false negatives and ensure that cases, particularly those with atypical phenotypes in whom CSF 14-3-3 protein is of limited

value, are not missed. Given the number of requests received by the CSF 14-3-3 protein service in the UK, follow up of all requests to the service would perhaps be as time and labour intensive as the current arrangement.

There are further issues to be considered. The use of this service to identify suspect cases is only of value if the investigation is widely pursued in all suspect cases. There is clear evidence of sub-optimal use of this investigation in suspect sCJD in the UK. It is entirely possible that the forthcoming addition of MRI to the diagnostic criteria for sCJD will result in preferentially used of this investigation rather than CSF 14-3-3 protein. In individuals with a supportive MRI scan there would be no additional clinical value in undertaking lumbar puncture examination for CSF 14-3-3 protein if a treatable differential diagnosis has been excluded. In this context, I would predict that there will be a reduction in requests for CSF 14-3-3- protein over time in the UK following this amendment to the diagnostic criteria.

Ascertaining cases through death certificate review

An alternative model that could be considered is that adopted in the USA. Routine analysis of mortality data obtained from death certificates are supplemented by the review of medical records in all suspect vCJD cases notified to public health authorities and the Centre for Disease Control (CDC), and all other prion disease cases aged less than 55 years old that have been identified from death certificate review. This system prioritises the collection and validation of detailed information in suspect cases for which there are potential public health implications. Mortality data are routinely collected, readily available and low cost. This may therefore be a sustainable option for disease surveillance. There is an issue relating to timelineness, particularly in suspect vCJD, where public health action may be required. This is addressed by encouraging the referral in life of suspect vCJD cases, rather than all suspect prion disease cases to the public health authorities or CDC. The ability of the system to detect vCJD has not been significantly challenged however. Just three cases of vCJD have been described in the USA; two cases were diagnosed in the UK and the third was diagnosed clinically, undergoing both brain and tonsil biopsies in life. It is not known therefore whether this system would be sensitive enough to detect vCJD including novel clinical phenotypes if employed in the UK. The USA is

the only country with a mature PHS system that has not reported an increasing in sCJD mortality in recent years, and the only country to report racial differences in sCJD mortality. These findings may be entirely novel, or may be as a result of systematic under-ascertainment of sCJD cases. This should also be considered.

Data from this thesis confirmed that death certificates in the UK have a high sensitivity for identifying prion disease, particularly vCJD. It is likely however that the activities of the NCJDSU have made a significant contribution to this. It is not known whether a change in surveillance activities in the UK would result in a fall in the sensitivity of a death certificate diagnosis of prion disease. It should also be considered that the interpretation of death certificate data requires the review of medical records and neuropathological material. These steps are both labour and time intensive. They require on-going cooperation of clinicians and local health authorities in providing medical case records, an area that the NCJDSU is currently under-performing in. There may also be issues relating to the reliability and completeness of information relating to clinical history, clinical signs, date of onset and so on, obtained from a retrospective review of medical case notes alone. Perhaps more importantly, it is also unclear whether such a system would detect a further change in clinical phenotype of CJD or indeed a novel human prion disease given the narrow referral criteria.

Optional appraisal

The current model of disease surveillance in the UK is expensive to deliver. In the present financial climate the long term sustainability of this model is uncertain. Two alternate models for delivering a sustainable prion disease PHS system in the UK have been explored, each with inherent limitations. Any change to the model of delivery of surveillance should first and foremost consider the objectives of the surveillance system and the resource available to achieve these. Indeed in the context of scarce resources it may be desirable and necessary to revise the objectives of the system to focus more directly on vCJD given the limited public health implications of other forms of human prion disease.

Were I to speculate on which model the PHS system in the UK might adopt in the future I would suggest a system in which the routine review of mortality data are supplemented by direct referral of suspect cases with the central review of medical case records and diagnostic data to ensure diagnostic verification in all suspect cases where possible, but without a neurologist visiting cases. Prioritisation would be given to those cases in which there are public health implications of a diagnosis. This system would be complemented by established surveillance activities such as the on-going TMER and PIND studies and enhanced surveillance in selected groups such as the 'at risk' cohort. The NCJDSU has been found to be both stable and flexible over time and will be well placed to adapt to any change in the model of surveillance. However any change to the model of PHS should be made following a full option appraisal to identify the most sustainable and viable model for meeting the systems objectives with the resources available.

Screening blood and organs for PrP^{Sc}

The discovery of a single, safe, sensitive and specific ante-mortem blood test for prion disease will be the most significant forthcoming development to shape PHS. The impact that such a test will have on PHS is unclear. This will in part be determined by the properties of the test (the sensitivity and specificity) and the prevalence of asymptomatic infection in the population. There are major ethical considerations to introducing a blood test to screening blood and organ donations for PrP^{Sc}, not least that the pathogenesis of vCJD is poorly understood therefore the significance of a positive result would, based on current scientific knowledge, be unknown. Nevertheless, any screening test, if introduced, is likely to result in the identification of an increasing number of individuals that will be designated 'at risk' for public health purposes in whom there will be a requirement for enhanced surveillance. This group will consist of those genuinely 'at risk' and false positives from the screening test; given that the prevalence of asymptomatic vCJD infection in the population is currently thought to be low, the PPV of any screening test is likely to be low, resulting in a large number of false positives who will undoubtedly be subject to public health protection measures. There is also the requirement for a confirmatory test for those individuals screening positive. Such a test does not currently exist. Expertise and consistency in the interpretation of the results of both

screening and diagnostic tests are crucial and pathways for patient care must be developed to minimise the potential harm experienced by patients through screening. Of course even with a screening program in place clinical prion disease cases will continue to emerge that will require public health action. The NCJDSU are likely to have a major role in these forthcoming developments.

Future research

There is a clear imperative for on-going prion disease PHS. Evaluation should be a key component of the PHS, conducted regularly to ensure that the system continues to meet its objectives. Comparative international data are crucial to interpret surveillance data from the UK. It is therefore important that international PHS systems also undertake regular evaluations. Given the increasing reliance on diagnostic criteria transparency about the surveillance protocols used by international collaborators, for example the approach to assessment of EEGs or MRI scans, is increasingly important. In addition, I would call for international collaborators to publish details of rates of case confirmation to aid the interpretation of routinely published incidence and mortality rates. Multi-site international studies examining the reproducibility and validity of the approaches taken to assess of EEG and MRI in disease surveillance would be welcomed. The NCJDSU may wish to further explore the key areas of concern that were identified in the evaluation in Chapter 3. In addition, acknowledging that the current model of disease surveillance in the UK may not be sustainable in the long-term, an option appraisal exploring other possible models of delivering PHS surveillance should be considered.

Conclusions

Prospective systematic CJD surveillance in the UK has successfully identified and characterised a novel prion disease, vCJD, in humans. Secondary transmission of vCJD via a previously unrecognised route has provided the public health imperative to continue disease surveillance for the foreseeable future. The NCJDSU in the UK is well placed to achieve this.

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Appendix 1

Additional web-based resources accessed in search of grey literature

UK Government Departments or Affiliated Agencies
Department of Health
Department of Environment, Food and Rural Affairs
Food Standards Agency
The National Archives (access to BSE Inquiry)
Spongiform Encephalopathy Advisory Committee (SEAC)
CJD Incident Panel
Health Protection Agency
NIBSC CJD Resource Centre
Advisory Committee on Dangerous Pathogens
vCJD Trust

Research / Surveillance Networks
Scottish TSE network
EUROCJD
NEUROCJD
NEUROPRION
TMER Study
Medical Research Council: TSE Research List
National CJD Surveillance Unit
National Prion Clinic

European Resources
European Commission
European Food Safety Authority (EFSA)
European Centre for Disease Prevention and Control

Worldwide resources
Centre for Disease Control and Prevention
World Health Organization
National Prion Disease Pathology Surveillance Center (USA)
Alberta Prion Research Institute (Canada)

Patient Groups
CJD Support Network (UK)
CJD Support Network (USA)
Human BSE Foundation
CJD Advice Network
CJD Alliance
Brain and Spine Foundation

Appendix 2

WHO Diagnostic criteria for human prion diseases (98)

SPORADIC CJD

Definite
Neuropathological/immunocytochemical confirmation is required for a diagnosis of definite sCJD

Probable
Rapidly progressive dementia, **and at least two** of the following four symptoms:

a. myoclonus
b. visual or cerebellar problems
c. pyramidal or extra-pyramidal features
d. akinetic mutism

plus typical electroencephalogram (EEG) with generalised triphasic periodic complexes at approximately 1 per second

or

clinical criteria for *possible s*CJD **and** a positive assay for CSF 14-3-3 protein

Possible
Rapidly progressive dementia, **two** of the symptoms listed in above (a-d) **and** an illness duration of less than 2 years.

VARIANT CJD

Definite
A progressive neuropsychiatric disorder **and** neuropathological confirmation of the disease, showing spongiform change and extensive PrPSc deposition with florid plaques throughout the cerebrum and cellebellum.

Probable
A progressive neuropsychiatric disorder of a duration greater than 6 months, where routine investigations do not suggest an alternative diagnosis **and at least four** of the following five symptoms:

a. early psychiatric symptoms (depression, anxiety, apathy withdrawal, delusions)
b. persistent painful sensory symptoms (including both frank pain and/or unpleasant dysaesthesia)
c. ataxia
d. myoclonus or chorea or dystonia
e. dementia

An EEG will not show the typical appearances of sporadic CJD, **or** no EEG has been performed and there is a symmetrical high signal in the posterior thalamus on a MRI brain scan. The patient would have had no history of potential iatrogenic exposure and no evidence of a familial form of TSE.

or

A progressive neuropsychiatric disorder for a period of longer than six months, where routine investigations do not support an alternative diagnosis, and where there is no history of potential of

iatrogenic exposure or evidence of a genetic form of prion disease, **plus** a tonsil biopsy which is positive for PrPSc.

Possible
A progressive neuropsychiatric disorder of a duration greater than 6 months, where routine investigations do not suggest an alternative diagnosis, and there is no history of potential iatrogenic exposure or evidence of a genetic prion disease, **and at least four** out of five of the symptoms listed above (a-e) **and** an EEG that does not show the typical appearance of sCJD **or** no EEG has been performed.

IATROGENIC CJD

Definite
A neuropathological diagnosis of CJD in a patient with a recognised risk factor for iatrogenic CJD

Probable
A progressive predominantly cerebellar syndrome in a human pituitary growth hormone recipient, or a clinical diagnosis of probable sCJD (see above) in a patient with a recognised risk factor for iatrogenic CJD

Relevant exposure risks for iatrogenic CJD
The relevance of any exposure to disease causation must take into account the timing of the exposure in relation to disease onset

- Treatment with human pituitary growth hormone, human pituitary gonadotrophin or human dura mater graft
- Corneal graft in which the corneal donor has been classified as definite or probable human prion disease
- Exposure to neurosurgical instruments previously used in a case of definite or probable human prion disease
- Transfusion of blood from a donor subsequently diagnosed with vCJD*

This list is provisional as previously unrecognised mechanisms of human prion disease may occur

*note cases of acquired vCJD as a result for transfusion of blood from a donor subsequently diagnosed with vCJD are designated vCJD cases for disease surveillance purposes

GENETIC PRION DISEASE

Definite
A neuropathological confirmation of prion disease, **plus either** *definite* genetic prion disease in a first degree relative (i.e. a parent, child or sibling), **or** a pathogenic *PRNP* mutation

Probable
A progressive neuropsychiatric disorder **plus** either *definite* or *probable* genetic prion disease in a first degree relative, **or** a pathogenic *PRNP* mutation

Pathogenic *PRNP* Mutations

- *PRNP* Mutations associated with GSS neuropathological phenotype
  P102L, P105L, A117V, G131V, F198S, D202N, Q212P, Q217R, M232T, 192 bpi
- *PRNP* Mutations associated with CJD neuropathological phenotype
  D178N-129V, V180I, V180I+M232R, T183A, T188A, E196K, E200K, V203I, R208H, V210I, E211Q, M232R, 96 bpi, 120 bpi, 144 bpi, 168 bpi, 48 bp*del*
- *PRNP* Mutations associated with FFI neuropathological phenotype
  D178N-129M
- *PRNP* Mutation associated with vascular PrP amyloid

Y145s

- *PRNP* Mutations associated with proven but unclassified prion disease
  H187R, 216 bpi
- *PRNP* Mutations associated with neuro-psychiatric disorder, but not proven prion disease
  I138M, G142S, Q160S, T188K, M232R, 24 bpi, 48 bpi, 48 bpi + nucleotide substitution in other octapeptides
- *PRNP* Mutations without clinical and neuropathological data:
  T188R, P238S
- *PRNP* Polymorphisms with established influence on phenotype:
  M129V
- *PRNP* polymorphisms with suggested influence on phenotype:
  N171S, E219K, 24 bp deletion
- *PRNP* polymorphisms without established influence on phenotype:
  P68P, A117A, G124G, V161V, N173N, H177H, T188T, D202D, Q212Q, R228R, S230S

Appendix 3

Clinical presentation sCJD (230)

It may not be possible to classify a particular case. If the case has good data but does not clearly fit into one of the specified categories then the code 'other' should be used. If there are insufficient data to categorise the cases then 'not specified' should be used.

Rapidly progressive dementia (RPD)

The majority of cases will probably be in this category. Precise presenting symptoms will vary from case to case. The picture is one of an encephalopathic illness with dementia and diverse other neurological features, progressing rapidly over weeks to a few months with no individual cognitive or physical deficit being present alone for more than two weeks

Heidenhain Variant

These cases present with impairment of visual acuity and/or field, progressing on to clinical blindness without other significant clinical deficit for the first two weeks of illness. Visual symptoms might include visual loss, visual inattention, visual illusions and hallucinations. It is essential that the symptoms progress to cortical blindness. Cases with other onsets that progress to include cortical blindness are not included in this category.

Pure psychiatric onset

These cases present with psychiatric symptoms such as depression, anxiety, paranoia and delusions, without the presence of other features for a period of at least four weeks. Non-specific malaise or apathy do not count unless accompanied by some of the above symptoms. Visual or auditory hallucinations alone do not count but may accompany the above fates. It may be difficult to distinguish between the early features of dementia and a more specifically psychiatric onset. Behavioural change straightforwardly due to a developing dementia is not included in this category. The essential characteristics of this presentation is that the patients present with a disturbance that suggests a psychiatric disturbance rather than an obvious dementia and specifically neurological features are absent.

Slowly progressive dementia

These cases present with a slowly progressive dementia, developing over months to years without any other significant neurological features for the first six months.

Pure cerebellar onset

Presentation is with a progressive cerebellar syndrome without other significant features.

Extra-pyramidal onset

Presentation is with an extra-pyramidal syndrome involving Parkinsonian features with or without chorea, athetosis or dystonia but without other significant features for at least two weeks.

Stroke-like onset

Presentation is abrupt enough for a diagnosis of stroke to be entertained in the initial stages.

Sensory symptoms at onset

Presentation with somato-sensory symptoms alone for at least two weeks. Such symptoms might include parasthesia, dysaesthesis, numbness, specifically neurogenic pain etc but would not include vague, non-specific aches and pains. This category does not include presentation with special sensory symptoms (i.e. visual, auditory, olfactory, gustatory). Sensory symptoms may be present along with other symptoms (for example as part of a RPD) but this category is for essentially 'pure' sensory presentation.

Other

None of the presentations describe above is applicable

Not specified

There is no clear clinical information available or the information does not allow a definite classification according to the above criteria.

Appendix 4

Clinical presentation vCJD (185)

Psychiatric features
Anxiety
Irritability
Insomnia
Social withdrawal
Loss of interest
Dysphoria
Aggression
Tearfulness
Agitation
Weight loss
Psychomotor retardation
Behavioural change
Anergia
Poor performance
Hypersomnia
Hallucinations
Paranoid delusions
Inappropriate affect
Obsessive features
Suicidal ideation
Panic attacks
Diurnal mood variation
Loss of confidence
Bizarre behaviour
Paranoid ideation
Lack of emotion
Change in eating preferences

Neurological features
Gait disturbance
Impairment of language
Pyramidal features
Impaired coordination
Impaired concentration
Poor memory
Myoclonus
Dementia
Abnormality of ocular motility
Hypoaesthesia
Tremor
Paraesthesia
Dystonia
Chorea
Other involuntary movements
Pain
Visual symptoms
Primitive reflexes
Swallowing impairment
Incontinence
Headache
Dizziness
Dysdiadochokinesia
Extra-pyramidal features
Seizures
Facial weakness
Taste disturbance
Hyperacusis

Appendix 5

Supplemental analyses describing the underlying cause of death recorded in the literal text of death certificates in prion disease cases (definite or probable) are reported here, according to disease subtype (Table 69).

CJD was recorded as the underlying cause of death in 676 (76.1%) sCJD cases. In 264 (39.1%) sCJD was identified as the underlying cause of death; in two vCJD. Of the sCJD cases (n=208) that did not have CJD recorded as the underlying cause of death, a neurological disorder was recorded as the underlying cause of death in 118 (56.7%). In 67 (32.2%), representing 7.6% of all sCJD cases, the diagnosis recorded as underlying cause of death was most likely the immediate cause of death, for example, cardiac arrest or pneumonia.

The underlying cause of death was recorded as CJD in 142 (89.9%) vCJD cases, of which the majority, 110 (77.5%) were identified as vCJD cases on their death certificate; two were identified as iCJD cases. Of the vCJD cases (n=17) that did not have CJD recorded as the underlying cause of death, 8 (47.1%) had a neurological diagnosis recorded as the underlying cause of death. The remaining 9 (52.9%) vCJD cases, representing 5.4% of all vCJD cases, the immediate cause of death was most likely recorded as the underlying cause of death, for example pneumonia.

The underlying cause of death was recorded as CJD in 46 (88.5%) iCJD cases. Of these 21 were identified as being aetiologically iCJD with cadaveric-derived hGH being identified as the route of exposure in ten and cadaveric-derived dura mater grafting in one. Of the remaining iCJD cases, four had a neurological diagnosis recorded as the underlying cause of death. For two, representing 3.7% of all iCJD cases, the immediate cause of death was most likely recorded as the underlying cause of death (both pneumonia).

Finally, the underlying cause of death was recorded as CJD in 72.6% (68) of definite or probable genetic prion disease cases; 32 (47.1%) of these were identified as being of a genetic aetiology and 6 (8.9%), sporadic. Of the remaining 26 cases, half (13) had a neurological diagnosis recorded as the underlying cause of death and the other half most likely had their immediate cause of death incorrectly recorded as the underlying cause of death.

Table 69 Underlying cause of death as recorded in the literal text of death certificate in definite and probable prion disease case according to disease subtype

Underlying cause of death	Number (%)
sCJD	**887 (100)**
CJD	676 (76.2)
Neurological Disease	118 (13.3)
Cardiovascular Disease	14 (1.6)
Gastrointestinal Disease	4 (<0.1)
Malignancy	3 (<0.1)
Other	2 (<0.1)
Most likely immediate cause of death (pneumonia, pulmonary embolism, sepsis, cardiac arrest)	67 (7.6)
vCJD	**159 (100)**
CJD	142 (89.3)
Neurological Disease	8 (5.0)
Most likely immediate cause of death (pneumonia, immobility, hypostasis)	9 (5.7)
Genetic Prion Disease	**52 (100)**
CJD	46 (88.5)
Neurological Disease	4 (7.7)
Most likely immediate cause of death (pneumonia)	2 (3.8)
iCJD	**94 (100)**
CJD	68 (72.3)
Neurological Disease	13 (13.8)
Most likely immediate cause of death (pneumonia, sepsis)	13 (13.8)

Appendix 6

These analyses consider two issues. Firstly, whether any routinely available information recorded on the death certificate could be used to distinguish CJD cases from non-cases in those certified as having CJD. Secondly, whether there was any significant difference between cases of CJD that did and did not have CJD recorded on their death certificate. In the analyses that follow only CJD stated or coded in any diagnostic position was considered as assessment of death certificates using this criteria had produced the greatest yield. Analyses were first carried out on the entire cohort and then stratified according to disease subtype given that the degree of misclassification of death certificates was shown to vary according to subtype. There were insufficient non-cases to analyse data from iCJD or genetic prion disease cases. Stratified analyses were therefore limited to sCJD and vCJD

Further methodological issues and definitions

In addition to cause of death, the date and place of death and name of the individual certifying death is recorded on a death certificate. Where a physician is unable to certify a death for medico-legal reasons, the case is referred to the coroner (in Scotland, the Procurator Fiscal). This individual determines whether a death requires further investigation. He/she may be satisfied that the death can be certified, or may request a post mortem examination to determine the cause of death. Where the cause of death cannot be determined following post mortem examination or death is deemed to have occurred due to violent or unnatural causes an inquest will be held. This is a publically held, legal investigation into the circumstances of a death. Following an inquest the coroner may issue a death certificate. Where an inquest into a death has been held the verdict of this inquest is recorded. The place of death was determined from each death certificate and categorised as follows: Hospital, hospice, an individuals own home or home of next of kin, nursing or residential home and other. The individual certifying death was categorised as follows based upon information available on each death certificate:

- Hospital doctor: Cases where death occurred in hospital and death was not certified by a neuropathologist or coroner/procurator fiscal
- Hospice doctor: Cases where death occurred in a hospice and death was not certified by a neuropathologist or coroner/procurator fiscal
- General practitioner: Cases where death occurred at home or in nursing or residential care and death was not certified by a neuropathologist or coroner/procurator fiscal
- Deaths certified by a coroner or procurator fiscal
- Deaths certified by a neuropathologist or pathologist

Univariate analyses using the Chi^2 test (and non-parametric equivalents where assumptions violated) and Wilcoxon Ranksum test were carried out.

Results

There was no difference in the sex distribution of cases and non-cases certified as CJD (Table 70). Cases were younger than non-cases and more likely to die in a hospice or at home than non-cases, although the greatest proportion of both cases and non-cases died in hospital. Reflecting this cases and non-cases were most frequently certified by a hospital physician although the distribution of certification between cases and non-cases was significantly different, such that cases were more likely to be certified by a coroner than non-cases.

When analyses were stratified by disease subtype age at death remained significantly lower in sCJD cases compared to non-cases but few other associations remained statistically significant (Table 71). For vCJD there was no significant difference between cases and non-cases on statistical testing although the statistical power to detect an effect was limited by the small number of non-cases (Table 72).

Table 70 Comparison of death certificate data from suspect CJD cases referred to the NCJDSU that had CJD recorded in the literal text or ICD coded (any position) on their death certificate according to case classification (all disease subtypes)

		Narrowly defined*			Broadly defined†		
		CJD case	Non-case	P value	CJD case	Non-case	P value
Total number (%)		756	50		1018	128	
Male, n (%)		384 (50.8)	27 (54.0)	0.660	511 (50.2)	66 (51.6)	0.708
Median Age at Death, Years (IQR)		62.8 (49.3 – 71.0)	71.4 (64.1 – 80.6)	<0.001	63.5 (49.7 – 71.8)	71.1 (64.2 – 79.7)	<0.001
Place of Death	**Hospital**	476 (63.0)	43 (86.0)	0.050	621 (61.0)	95 (74.2)	0.007
	Hospice	92 (12.3)	3 (6.0)		137 (13.5)	10 (7.8)	
	Usual home	117 (15.5)	3 (6.0)		172 (16.9)	8 (6.3)	
	Nursing or residential home	68 (9.0)	1 (2.0)		85 (8.4)	15 (11.7)	
	Other	1 (0.1)	0		1 (0.1)	0	
	Unknown	2 (0.3)	0		2 (0.2)	0	
Certifier	**Hospital doctor**	361 (47.8)	37 (74.0)	0.039	500 (49.1)	85 (66.4)	<0.001
	Hospice doctor	64 (8.5)	2 (4.0)		106 (10.4)	9 (7.0)	
	General Practitioner	120 (15.9)	5 (10.0)		188 (18.5)	27 (21.1)	
	Coroner	203 (26.9)	6 (12.0)		211 (20.7)	7 (5.5)	
	Neuropathologist	1 (0.1)	0		1 (0.1)	0	
	Unknown	7 (0.7)	0		12 (1.1)	0	

*CJD case classified as 1.0 and non-case as 4.3; † CJD case classified as 1.0 or 2.0 and non-case as 4.1, 4.2 or 4.3

Table 71 Comparison of death certificate data from suspect sCJD cases referred to the NCJDSU that had CJD recorded in the literal text or ICD coded (any position) on their death certificate according to case classification

		Narrowly defined*			Broadly defined†		
		CJD case	Non-case	P value	CJD case	Non-case	P value
Total number (%)		544	46		740	123	
Male, n (%)		269 (49.5)	25 (54.4)	0.314	360 (48.7)	59 (48.0)	0.889
Median age at death, Years (IQR)		67.3 (60.8 – 74.8)	73.8 (67.6 – 80.7)	<0.001	67.6 (61.2 – 74.7)	71.6 (65.2 – 79.8)	<0.001
Place of Death	**Hospital**	398 (73.2)	40 (87.0)	0.290	528 (71.4)	91 (74.0)	0.086
	Hospice	58 (10.7)	3 (6.5)		93 (12.6)	10 (8.1)	
	Usual home	46 (8.5)	2 (4.4)		67 (9.1)	7 (5.7)	
	Nursing or residential home	42 (7.7)	1 (2.2)		52 (7.0)	15 (12.2)	
	Other	0	0		0		
	Unknown						
Certifier	**Hospital doctor**	312 (57.4)	35 (76.1)	0.322	438 (59.2)	82 (66.7)	0.002
	Hospice doctor	45 (8.3)	2 (4.4)		80 (10.8)	9 (7.3)	
	General Practitioner	61 (11.2)	4 (8.7)		91 (12.3)	26 (21.1)	
	Coroner	120 (22.1)	5 (10.9)		120 (16.2)	6 (4.9)	
	Neuropathologist	1 (0.2)	0		1 (0.1)	0	
	Unknown	5 (0.9)	0		10 (1.4)	0	

*CJD case classified as 1.0 and non-case as 4.3; † CJD case classified as 1.0 or 2.0 and non-case as 4.1, 4.2 or 4.3

Table 72 Comparison of death certificate data from suspect vCJD cases referred to the NCJDSU that had CJD recorded in the literal text or ICD coded (any position) on their death certificate according to case classification

		Narrowly defined*			Broadly defined†		
		vCJD cases	Non-cases	P value	vCJD cases	Non-cases	P value
Total number (%)		104	4		149	5	
Male, n (%)		62 (59.6)	2 (50.0)	0.701	83 (55.7)	3 (60.0)	0.849
Median Age at Death, Years (IQR)		28.8 (24.3 – 34.9)	39.4 (31.2 – 46.1)	0.071	28.4 (22.3 – 34.5)	33.8 (30.0 – 44.8)	0.089
Place of Death	**Hospital**	31 (29.8)	3 (75.0)	0.488	37 (24.8)	4 (80.0)	0.135
	Hospice	22 (21.2)	0		30 (20.1)	0	
	Usual home	42 (40.4)	1 (25.0)		69 (46.3)	1 (20.0)	
	Nursing or residential home	8 (7.7)	0		12 (8.1)	0	
	Other	0	0		0	0	
	Unknown	0	0		0	0	
Certifier	**Hospital doctor**	19 (18.3)	2 (50.0)	0.611	25 (16.8)	3 (60.0)	0.180
	Hospice doctor	11 (10.6)	0		16 (10.7)	0	
	General Practitioner	34 (32.7)	1 (25.0)		63 (42.3)	1 (20.0)	
	Coroner	39 (37.5)	1 (25.0)		44 (29.5)	1 (20.0)	
	Neuropathologist	0	0		0	0	
	Unknown	1 (1.0)	0		1 (0.7)	0	

*CJD case classified as 1.0 and non-case as 4.3; † CJD case classified as 1.0 or 2.0 and non-case as 4.1, 4.2 or 4.3

Comparison was next drawn between cases (narrowly and broadly defined) that did, and did not, have CJD recorded in the literal text or ICD coded in any position on their death certificate. Table 73 includes all disease subtypes, whilst Table 73 and Table 75 examine only sCJD and vCJD cases respectively. Overall there was no difference in the sex distribution of cases according to whether CJD was on the death certificate or not. Cases that had CJD on their death certificate were younger (narrowly defined CJD: median of 62.8 years old (45.3 – 71.0) *vs.* 66.4 years old (58.2 – 74.6), P<0.001). There was no difference in median duration of illness between groups. A greater than expected proportion of cases that had CJD recorded or coded on their death certificate died in a hospice or at home, and accordingly a greater proportion were certified by a general physician or hospice doctor when compared to those who did not have CJD on their death certificate. In the latter group a greater than expected proportion of cases died in hospital or in nursing home care and more than expected were certified by a coroner.

When sCJD cases only were examined there was no longer a significant difference in age between groups, however the median illness duration in the group that had CJD on their death certificate was shorter than for the group that did not. The trends in place of death and the individual responsible for certifying death for sCJD cases were no different from the overall cohort.

Analyses of vCJD cases should be interpreted with caution due to a lack of statistical power as a result of the small sample size. There was no significant difference between either group with respect to sex, age, median illness duration or certifier. However vCJD cases that had CJD on their death certificate were more likely to die at home than those who did not, the latter comparison group being more likely to die in hospital. This finding, also observed for all subtypes and in the analysis of sCJD cases only, may reflect diagnostic certainty in that patients in whom a clinical or pathological diagnosis has been reached may be more likely to be discharged home to receive end of life care. This is less likely to occur if the diagnosis remains unclear, for example if a reversible cause for the illness has not yet been excluded.

Table 73 Comparison of characteristics of cases that did and did not have CJD recorded in the literal text or ICD coded (any position) on their death certificate (all disease subtypes)

		Narrowly defined CJD*			Broadly defined CJD†		
		Certified CJD	Not Certified CJD	P value	Certified CJD	Not Certified CJD	P value
Total number		756	164		1018	176	
Male, n (%)		384 (50.8)	81 (49.4)	0.745	511 (50.2)	88 (50.0)	0.962
Age at Death, Median Years (IQR)		62.8 (49.3 – 71.0)	66.4 (58.5 – 74.6)	<0.001	63.5 (49.7 – 71.8)	66.7 (58.5 – 74.4)	<0.001
Illness Duration, Median Months (IQR)		6.0 (3.0 – 12.4)	6.1 (2.8 – 12.9)	0.804	5.7 (3.0 – 11.4)	6.1 (2.8 – 12.9)	0.629
Place of Death	**Hospital**	476 (63.0)	121 (73.8)	<0.001	621 (61.0)	131 (74.4)	<0.001
	Hospice	92 (12.2)	7 (4.3)		137 (13.5)	7 (4.0)	
	Usual home	117 (15.5)	15 (9.2)		172 (16.9)	17 (9.7)	
	Nursing or residential home	68 (9.0)	19 (11.6)		85 (8.4)	19 (10.8)	
	Other	1 (0.1)	0		1 (<0.1)	0	
	Unknown	2 (0.3)	2 (1.2)		2 (0.2)	2 (1.4)	
Certifier	**Hospital doctor**	361 (47.8)	78 (47.6)	0.003	500 (49.1)	87 (49.4)	<0.001
	Hospice doctor	64 (8.5)	4 (2.4)		106 (10.4)	4 (2.3)	
	General Practitioner	120 (15.9)	20 (12.2)		188 (18.5)	22 (12.5)	
	Coroner	203 (26.9)	58 (35.4)		211 (20.7)	58 (33.0)	
	Neuropathologist	1 (0.1)	0		1 (0.1)	0	
	Unknown	7 (0.9)	4 (2.4)		12 (1.2)	5 (2.8)	

*CJD case classified as 1.0 and non-case as 4.3; † CJD case classified as 1.0 or 2.0 and non-case as 4.1, 4.2 or 4.3

Table 74 Comparison of characteristics of sCJD cases that did and did not have CJD recorded in the literal text or ICD coded (any position) on their death certificate

		Narrowly defined sCJD*			Broadly defined sCJD†		
		Certified CJD	Not Certified CJD	P value	Certified CJD	Not Certified CJD	P value
Total number		544	140		740	147	
Male, n (%)		269 (49.5)	66 (47.1)	0.626	360 (48.7)	70 (47.6)	0.820
Age at Death, Median Years (IQR)		67.3 (60.8 – 74.8)	68.3 (61.1 – 75.2)	0.453	67.6 (61.2 – 74.7)	68.6 (61.1 – 74.9)	0.507
Illness Duration, Median Months (IQR)		4.2 (2.6 – 7.9)	5.7 (2.8 – 12.2)	0.021	4.1 (2.6 – 7.5)	5.3 (2.8 – 11.6)	0.009
Place of Death	**Hospital**	398 (73.2)	107 (76.4)	0.004	528 (71.4)	112 (76.2)	<0.001
	Hospice	58 (10.7)	5 (3.6)		93 (12.6)	5 (3.4)	
	Usual home	46 (8.5)	10 (7.1)		67 (9.1)	12 (8.2)	
	Nursing or residential home	42 (7.7)	16 (11.4)		52 (7.0)	16 (10.9)	
	Other	0	0		0	0	
	Unknown	0	2 (1.4)		0	2 (1.4)	
Certifier	**Hospital doctor**	312 (57.4)	69 (49.3)	<0.001	438 (59.2)	73 (49.7)	<0.001
	Hospice doctor	45 (8.3)	2 (1.4)		80 (10.8)	2 (1.4)	
	General Practitioner	61 (11.2)	14 (10.0)		91 (12.3)	16 (10.8)	
	Coroner	120 (22.1)	52 (37.1)		120 (16.2)	52 (35.4)	
	Neuropathologist	1 (0.2)	0		1 (0.1)	0	
	Unknown	5 (1.0)	3 (2.1)		10 (1.4)	4 (2.7)	

*CJD case classified as 1.0 and non-case as 4.3; † CJD case classified as 1.0 or 2.0 and non-case as 4.1, 4.2 or 4.3

Table 75 Comparison of characteristics of vCJD cases that did and did not have CJD recorded in the literal text or ICD coded (any position) on their death certificate

		Narrowly defined vCJD*			Broadly defined vCJD†		
		Certified CJD	Not Certified CJD	P value	Certified CJD	Not Certified CJD	P value
Total number		104	8		149	10	
Male, n (%)		62 (59.6)	4 (50.0)	0.594	83 (55.7)	6 (60.0)	0.791
Age at Death, Median Years (IQR)		26.3 (22.4 – 41.5)	28.8 (24.3 – 34.9)	0.786	28.5 (22.3 – 34.6)	27.7 (22.9 – 39.6)	0.865
Illness Duration, Median Months (IQR)		13.4 (10.9 – 17.1)	11.5 (10.4 – 13.6)	0.299	13.6 (11.0 – 17.2)	12.4 (11.2 – 16.6)	0.510
Place of Death	**Hospital**	31 (29.8)	5 (62.5)	0.266	37 (24.8)	7 (70.0)	0.033
	Hospice	22 (21.2)	2 (25.0)		30 (20.2)	2 (20.0)	
	Usual home	42 (40.4)	1 (12.5)		69 (46.3)	1 (10.0)	
	Nursing or residential home	8 (7.7)	0		12 (8.1)	0	
	Other	0	0		0	0	
	Unknown	1 (1.0)	0		1 (0.7)	0	
Certifier	**Hospital doctor**	19 (18.3)	2 (25.0)	0.453	25 (16.8)	4 (40.0)	0.111
	Hospice doctor	11 (10.6)	2 (25.0)		16 (10.7)	2 (20.0)	
	General Practitioner	34 (32.7)	1 (12.5)		63 (42.3)	1 (10.0)	
	Coroner	39 (37.5)	3 (37.5)		44 (29.5)	3 (30.0)	
	Neuropathologist	0	0		0	0	
	Unknown	1 (1.0)	0		1 (0.7)	0	

*CJD case classified as 1.0 and non-case as 4.3; † CJD case classified as 1.0 or 2.0 and non-case as 4.1, 4.2 or 4.3

Appendix 7

The sensitivity, specificity, PPV and NPV of a death certificate diagnosis of CJD (recorded in the literal text or ICD coded in any position) are outlined in Tables 76-79. The overall sensitivity for all prion disease was higher using this approach than simply examining ICD coding alone, with no significant difference in other measures. For narrowly defined prion disease the overall sensitivity was 82.2% (79.5 – 84.6), with a specificity of 80.0% (74.5 – 84.8), PPV of 93.8 (91.9 – 95.4) and NPV of 54.9 (49.7 – 60.1). The sensitivity was highest in the youngest age group but there was no discernable pattern across age groups. Again values for sCJD followed the overall trend for all prion disease whilst values for vCJD were significantly higher. The overall trends were the same irrespective of whether a narrow or broad definition of prion disease was applied, although as for previous analyses values were slightly higher when a broad definition of prion disease was applied. Once again following adjustment for age there was a statistically significant increase in sensitivity over time when all prion disease and sCJD were examined but not vCJD (Table 80).

Table 76 Sensitivity, specificity, positive and negative predictive value of CJD recorded in the literal text or ICD coded in any position on a death certificate, according to disease subtype and age group (narrowly defined)

Disease subtype	Age group	Prevalence	Sensitivity	Specificity	PPV	NPV
All prion disease	All ages	79.0 (76.0 – 80.9)	82.2 (79.5 – 84.6)	80.0 (74.5 – 84.8)	93.8 (91.9 – 95.4)	54.9 (49.7 – 60.1)
	<50 years	86.0 (81.0 – 89.7)	92.1 (87.6 – 95.3)	86.1 (70.5 – 95.3)	97.5 (94.3 – 99.2)	64.6 (49.5 – 77.8)
	50 – 59 years	80.0 (74.0 – 85.7)	80.0 (72.8 – 86.0)	94.7 (82.3 – 99.4)	98.4 (94.4 – 99.8)	53.7 (41.1 – 66.0)
	60 – 69 years	82.0 (77.0 – 86.0)	82.5 (77.5 – 86.8)	78.3 (65.8 – 87.9)	94.6 (90.9 – 97.1)	49.5 (39.1 – 59.9)
	70 – 79 years	72.0 (67.0 – 77.3)	73.7 (67.3 – 79.5)	79.5 (69.2 – 87.6)	90.4 (85.1 – 94.3)	53.7 (44.4 – 62.7)
	≥80 years	65.0 (54.0 – 74.2)	81.7 (69.6 – 90.5)	60.6 (42.1 – 77.1)	79.0 (66.8 – 88.3)	64.5 (45.5 – 80.8)
sCJD	All ages	76.0 (73.0 – 79.0)	79.5 (76.3 – 82.5)	78.4 (72.3 – 83.7)	92.2 (89.7 – 94.2)	54.4 (48.6 – 60.1)
	<50 years	82.0 (68.0 – 92.0)	83.8 (68.0 – 93.8)	87.5 (47.3 – 99.7)	96.9 (83.8 – 99.9)	53.8 (25.1 – 80.8)
	50 – 59 years	80.0 (73.0 – 86.3)	79.5 (71.3 – 86.3)	93.3 (77.9 – 99.2)	98.0 (92.9 – 99.8)	52.8 (38.6 – 66.7)
	60 – 69 years	81.0 (76.0 – 85.3)	82.2 (76.9 – 86.7)	78.0 (65.3 – 87.7)	94.1 (90.2 – 96.8)	50.5 (39.9 – 61.2)
	70 – 79 years	72.0 (66.0 – 76.9)	75.0 (68.6 – 80.7)	79.5 (69.2 – 87.6)	90.3 (85.0 – 94.3)	55.5 (46.1 – 64.6)
	≥80 years	65.0 (54.0 – 74.2)	81.7 (69.6 – 90.5)	60.6 (42.1 – 77.1)	79.0 (66.8 – 88.3)	64.5 (45.4 – 80.8)
vCJD	All ages	77.0 (69.0 – 83.3)	92.9 (86.4 – 96.9)	88.2 (72.5 – 96.7)	96.3 (90.8 – 99.0)	78.9 (62.7 – 90.4)
	<30 years	91.5 (81.3 – 97.2)	100 (54.1 – 100)	100 (54.1 – 100)	100 (93.4 – 100)	54.5 (23.4 – 83.3)
	30 – 49 years	72.0 (59.0 – 82.5)	97.7 (87.7 – 99.9)	76.5 (50.1 – 93.2)	91.3 (79.2 – 97.6)	92.9 (66.1 – 99.8)
	≥50 years	48.0 (26.0 – 70.2)	80.0 (44.4 – 97.5)	100 (71.5 – 100)	100 (63.1 – 100)	84.6 (54.6 – 98.1)

Table 77 Sensitivity, specificity, positive and negative predictive value of CJD recorded in the literal text or ICD coded in any position on a death certificate, according to disease subtype and year group (narrowly defined)

Disease subtype	Year group	Prevalence	Sensitivity	Specificity	PPV	NPV
All prion disease	1990 – 1995	76.0 (71.0 – 80.4)	75.1 (69.2 – 80.4)	69.2 (57.8 – 79.2)	88.5 (83.3 – 92.5)	47.0 (37.6 – 56.5)
	1996 – 2000	75.0 (71.0 – 78.9)	82.1 (77.6 – 86.0)	84.3 (76.4 – 90.5)	94.0 (90.7 – 96.4)	61.0 (53.0 – 68.6)
	2001 - 2006	85.0 (81.0 – 88.6)	87.5 (83.5 – 90.9)	86.0 (74.2 – 93.7)	97.3 (94.7 – 98.8)	54.4 (43.6 – 65.0)
sCJD	1990 – 1995	72.0 (66.0 – 77.2)	73.8 (67.1 – 79.9)	68.4 (56.7 – 78.6)	85.7 (79.5 – 90.6)	50.5 (40.5 – 60.5)
	1996 – 2000	73.0 (68.0 – 77.8)	77.0 (71.0 – 82.2)	83.5 (73.9 – 90.7)	92.7 (88.0 – 95.9)	57.3 (48.1 – 66.1)
	2001 – 2006	83.0 (79.0 – 87.3)	86.1 (81.3 – 90.1)	84.6 (71.9 – 93.1)	96.5 (93.3 – 98.5)	55.0 (43.5 – 66.2)
vCJD	1996 – 2000	74.0 (64.0 – 82.0)	93.4 (85.3 – 97.8)	85.2 (66.3 – 95.8)	94.7 (86.9 – 98.5)	82.1 (63.1 – 93.9)
	2001 – 2006	85.0 (68.0 – 94.9)	96.4 (81.7 – 99.9)	100 (81.7 – 99.9)	100 (87.2 – 100)	83.3 (35.9 – 99.6)

Table 78 Sensitivity, specificity, positive and negative predictive value of CJD recorded in the literal text or ICD coded in any position on a death certificate, according to disease subtype and age group (broadly defined)

Disease subtype	Age group	Prevalence	Sensitivity	Specificity	PPV	NPV
All prion disease	All ages	67.0 (65.0 – 69.4)	85.3 (83.1 – 87.2)	78.0 (74.5 – 81.3)	88.8 (86.9 – 90.6)	72.1 (68.4 – 75.6)
	<50 years	81.0 (76.0 – 84.6)	93.1 (89.5 – 95.8)	88.1 (77.8 – 94.7)	97.0 (94.2 – 98.7)	75.6 (64.6 – 84.7)
	50 – 59 years	69.0 (63.0 – 74.5)	82.6 (76.5 – 87.7)	89.4 (80.8 – 95.0)	94.6 (90.0 – 97.5)	69.7 (60.2 – 78.2)
	60 – 69 years	70.0 (66.0 – 73.7)	85.8 (81.7 – 89.2)	75.9 (68.5 – 82.4)	89.2 (85.4 – 92.2)	69.8 (62.3 – 76.5)
	70 – 79 years	60.0 (56.0 – 64.8)	79.1 (73.9 – 83.7)	77.1 (70.5 – 82.9)	84.1 (79.2 – 88.2)	70.7 (64.0 – 76.9)
	≥80 years	47.0 (39.0 – 54.9)	84.0 (73.7 – 91.4)	64.7 (53.6 – 74.8)	67.7 (57.3 – 77.1)	82.1 (70.8 – 90.4)
sCJD	All ages	63.0 (61.0 – 65.7)	83.4 (80.8 – 85.8)	76.3 (72.3 – 79.9)	85.7 (83.2 – 88.0)	72.9 (68.9 – 76.6)
	<50 years	66.0 (53.0 – 77.4)	86.0 (72.1 – 94.7)	86.4 (65.1 – 97.1)	92.5 (79.6 – 98.4)	76.0 (54.9 – 90.6)
	50 – 59 years	68.0 (62.0 – 74.5)	82.9 (76.0 – 88.5)	87.1 (77.0 – 93.9)	93.3 (87.7 – 96.9)	70.1 (59.4 - 79.5)
	60 – 69 years	68.0 (64.0 – 72.6)	85.8 (81.6 – 89.3)	75.5 (67.9 – 82.0)	88.4 (84.4 – 91.6)	70.9 (63.3 – 77.7)
	70 – 79 years	60.0 (55.0 – 64.6)	80.4 (75.2 – 84.8)	76.9 (70.2 – 82.7)	84.0 (79.0 – 88.1)	72.2 (65.4 – 78.3)
	≥80 years	47.0 (39.0 – 54.9)	84.0 (73.7 – 91.4)	64.7 (73.7 – 91.4)	67.7 (57.3 – 77.1)	82.1 (70.8 – 90.4)
vCJD	All ages	73.0 (67.0 – 78.7)	93.7 (88.7 – 96.9)	91.5 (81.3 – 97.2)	96.8 (92.6 – 98.9)	84.4 (73.1 – 92.2)
	<30 years	90.0 (83.0 – 95.5)	88.9 (85.3 – 97.4)	88.9 (51.8 – 99.7)	98.8 (93.2 – 100)	57.1 (28.9 – 82.3)
	30 – 49 years	69.0 (58.0 – 78.2)	96.8 (88.8 – 99.6)	85.7 (67.3 – 96.0)	93.8 (84.8 – 98.3)	92.3 (74.9 – 99.1)
	≥50 years	35.0 (20.0 – 53.5)	83.3 (51.6 – 97.9)	100 (84.6 – 100)	100 (69.2 – 100)	91.7 (73.0 – 99.0)

Table 79 Sensitivity, specificity, positive and negative predictive value of CJD recorded in the literal text or ICD coded in any position on a death certificate, according to disease subtype and year group (broadly defined)

Disease subtype	Year group	Prevalence	Sensitivity	Specificity	PPV	NPV
All prion disease	1990 – 1995	62.0 (57.0 – 66.1)	78.0 (72.6 – 82.7)	63.0 (55.3 – 70.2)	77.1 (71.8 – 81.9)	64.1 (56.4 – 71.3)
	1996 – 2000	62.0 (58.0 – 65.8)	83.8 (79.8 – 87.2)	89.8 (85.3 – 93.3)	93.1 (89.9 – 95.5)	77.2 (71.9 – 81.9)
	2001 - 2006	76.0 (72.0 – 79.0)	90.3 (87.4 – 92.7)	76.4 (69.1 – 82.6)	92.3 (89.6 – 94.5)	71.6 (64.3 – 78.1)
sCJD	1990 – 1995	57.0 (52.0 – 61.8)	77.3 (71.3 – 82.6)	62.6 (54.9 – 69.8)	73.1 (67.0 – 78.6)	67.7 (59.8 – 74.9)
	1996 – 2000	58.0 (54.0 – 62.7)	79.7 (74.4 – 84.3)	89.2 (84.0 – 93.2)	91.1 (86.8 – 94.4)	76.0 (69.9 – 81.4)
	2001 – 2006	72.0 (68.0 – 75.7)	89.5 (86.0 – 92.4)	75.0 (67.3 – 81.7)	90.2 (86.8 – 93.0)	73.5 (65.9 – 80.3)
vCJD	1996 – 2000	65.0 (57.0 – 73.5)	92.9 (85.3 – 97.4)	91.1 (78.8 – 97.5)	95.2 (88.1 – 98.7)	87.2 (74.3 – 95.2)
	2001 – 2006	85.0 (75.0 – 91.8)	97.0 (89.5 – 99.6)	91.7 (61.5 – 99.8)	98.5 (91.7 - 100)	84.6 (54.6 – 98.1)

Table 80 Regression Co-efficients for changing sensitivity of death certificate diagnosis of CJD over time

Definition	Aetiological Subtype	Year Group Regression co-efficient (95% CI)	P value	Age group Regression co-efficient (95% CI)	P value
Narrow	All prion disease	6.8 (2.4 – 11.3)	0.006	-3.2 (-5.8 - -6.3)	0.019
	sCJD	7.1 (3.1 – 11.1)	0.002	-1.0 (-3.3 – 1.3)	0.372
	vCJD	12.6 (-30.5 – 55.7)	0.422	-6.0 (-32.4 – 20.4)	0.520
Broad	All prion disease	6.5 (2.2 – 10.8)	0.006	-2.4 (-4.8 – 0.1)	0.061
	sCJD	6.9 (2.5 – 11.4)	0.006	-0.8 (-3.3 – 1.7)	0.486
	vCJD	12.2 (-23.7 – 48.1)	0.360	0.7 (-21.3 – 22.7)	0.926

Appendix 8

The underlying causes of death as ICD coded on death certificates of suspect cases that had CJD recorded in the literal text of their death certificate but not ICD coded are shown in Table 81. In Table 82 the causes of death as recorded in the literal text of death certificates of suspect cases that had CJD ICD coded without mention of CJD in the literal text are shown.

Table 81 Causes of death as ICD coded in suspect prion disease cases with CJD recorded in the literal text of the death certificate (any position) but not ICD coded (any position)

ICD 9	Diagnosis corresponding to ICD code	ICD 10	Diagnosis corresponding to ICD code
1629	Malignant neoplasm of bronchus/ lung	A818 *	Other atypical virus infections of CNS
2041*	Chronic lymphoid leukaemia	A819 (8)*	Atypical virus infection of CNS
2533*	Pituitary dwarfism	A872*	Lymphocytic choriomeningitis
2901 (20)*	Presenile dementia	C541	Malignant neoplasm of endometruim
2950*	Schizophrenic Disorders	E852 (2)*	Heredofamilial amyloidosis
2989*	Unspecified psychosis	F812*	Specific disorders of arithmetical skills
3239*	Unspecified cause of encephalitis, myelitis, and encephalomyelitis	G122	Motor Neuron Disease
3319*	Cerebral degeneration, unspecified	G319 (2)*	Degenerative disease of CNS
3498 (2)*	Other specified disorders of CNS	G98*	Other disorders of CNS
410	Acute myocardial infarction	I219*	Acute myocardial infarction
4151	Pulmonary embolism and infarction	I259	Chronic ischaemic heart disease
436 (2)*	Stroke, subtype not specified	I269*	Pulmonary embolism without cor pulmonale
4660	Acute bronchitis	I64	Stroke, subtype not specified
485 (15)*	Bronchopneumonia, organism unspecified	J181*	Lobar pneumonia, unspecified
5789	Haemorrhage of gastrointestinal tract	J411*	Mucopurulent chronic bronchitis
8769*	Open wounds of back	W061*	Fall involving bed
887	Traumatic amputation of arm and hand		

*indicate definite or probable cases, 14 pre-senile dementia, 10 bronchopneumonia

Table 82 Causes of death according to position for individuals that had a CJD related ICD code on their death certificate without mention of CJD in the literal text of their death certificate

Position	Cause of death	Number
Part 1a	Acute confusional state	1
	Aspiration pneumonia	1
	Bronchopneumonia	15 (3)
	Cardiac Arrest / Cardiorespiratory arrest	2 (1)
	Cerebrovascular Accident	3
	Dementia	3 (1)
	Diabetes Mellitus	1
	Encephalitis	2
	Encephalopathy (unknown cause)	1
	Multi-infarct dementia	1
	Neurodegenerative condition	1
	Pulmonary embolism	3
	Rapidly progressive dementia	1
	Septicaemia secondary to aspiration	1
Part 1b	Cerebral Arterial Atherosclerosis	1
	Deep Vein Thrombosis	2
	Dementia	1
	Encephalitis	1
	Encephalopathy (unknown cause)	4
	Nevin Jones Syndrome	1
	Presenile dementia	1
	Progressive neurodegenerative Disease	3
	Spongiform myelin encephalopathy	1 (1)
Part 1c	Alzheimer's Disease	1
	Encephalopathy	1
	Immobilisation due to Parkinsonism	1
Part II	Dementia	4 (3)
	Iron Deficiency Anaemia	1
	Neurological Disorder	1
	Osteoarthritis	1
	Prostate Cancer	1

( ) indicate definite or probable prion disease case