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Some lifestyle-related factors and risk of chronic renal failure

- a population-based approach

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SUMMARY

Some renal diseases, i.e. rapidly progressive glomerulonephritis, are sufficient causes of a rapid, permanent total loss of renal function. However, the majority of renal diseases progress slowly over decades, initially often without symptoms, sometimes making it difficult to define the aetiologies. There is growing evidence that a multitude of lifestyle-related and environmental factors influence the risk and the progression rate of chronic renal failure (CRF), although genetic factors also appear to be of importance. Prevention is important since the prognosis of end-stage renal disease treated with dialysis is poor, but mortality is substantially increased also in patients with mild CRF.

To identify risk factors for CRF, we performed a population-based nation-wide case-control study. The study base was the entire Swedish population born in Sweden and aged 18-74 years. Eligible as cases were subjects who had a serum creatinine that for the first time and permanently exceeded 300 μmol/l (men) or 250 μmol/l (women) during the two-year study period, 1996-1998. The final study population included 926 cases and 998 randomly selected controls from the study base. A face-to-face interview and a self-administered questionnaire provided information about various exposures.

Despite an overall non-significant association, high daily smoking dose, long duration of the smoking habit, and a high cumulative dose were associated with a significant excess risk of CRF. In smokers with a cumulative dose of >30 pack-years, the risk was increased by 52 % compared to non-smokers. A more than two-fold increased risk among heavy smokers was observed for CRF classified as nephrosclerosis, but significant positive associations were also noted with glomerulonephritis, and among women – also with diabetic nephropathy. Other tobacco use than smoking was unrelated to risk of CRF.

A high protein intake was strongly and positively related to an increased risk of diabetic nephropathy. We cannot rule out that this association might be the result of reverse causality and recall bias, however, in an analysis confined to diabetic cases and controls, an almost 3-fold risk gradient with protein intake remained, albeit imprecise due to small numbers. Protein intake was not associated with other types of renal disease. We could not confirm our hypothesis that a high intake of antioxidants reduces risk of CRF, with the possible exception that a high intake of vitamin E was linked to a low risk in individuals with hypertension.

Being overweight in early adulthood and obese at anytime in life was associated with an increased risk of CRF. A body mass index exceeding 30 kg/m^2 in men and 35 kg/m^2 in women anytime during lifetime was linked to 3-to 4-fold increases in risk. Although much of the excess risk was driven by the higher prevalence of hypertension and diabetes among obese, an additional pathway may exist. Birth weight was unrelated to risk of CRF, while a short stature was associated with CRF, at least in men.

Isolated regular use of either paracetamol or aspirin was associated with a 2.5-fold increased risk of CRF overall, with a positive dose-response. The elevations in risk were observed for most types of underlying renal diseases, albeit not always statistically significant. To avoid bias due to analgesic use triggered by CRF symptoms, we disregarded more recent use, but the risks became only slightly attenuated. Our results are consistent with an exacerbating effect of paracetamol and aspirin, but, we cannot exclude that predisposing conditions of CRF may have prompted analgesic use.

To my family

LIST OF PAPERS

I. Ejerblad E, Fored CM, Lindblad P, Fryzek JP, Dickman PW, Elinder CG, McLaughlin JK, Nyrén O.
Association between smoking and chronic renal failure in a nationwide population-based case-control study.
Journal of American Society of Nephrology, 2004 Aug; 15 (8): 2178-85

II. Ejerblad E, Ye W, Fored CM, Lindblad P, Wolk A, McLaughlin JK, Bälter K, Nyrén O.
Risk of chronic renal failure in relation to protein and antioxidant intake; a nationwide population-based study in Sweden.
In manuscript

III. Ejerblad E, Fored CM, Lindblad P, Fryzek JP, McLaughlin JK, Nyrén O.
Obesity and risk of chronic renal failure.
Submitted for publication

IV. Fored CM, Ejerblad E, Lindblad P, Fryzek JP, Dickman PW, Signorello LB, Lipworth L, Elinder CG, Blot WJ, McLaughlin JK, Zack MM, Nyrén O.
Acetaminophen, Aspirin, and chronic renal failure.
New England Journal of Medicine, 2001 Dec; 345 (25): 1801-8

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LIST OF ABBREVIATIONS

AAN	Analgesic-associated nephropathy
ACE	Angiotensin-converting enzyme
BMI	Body mass index
CI	Confidence interval
CKD	Chronic kidney disease
CRF	Chronic renal failure
ESRD	End-stage renal disease
GFR	Glomerular filtration rate
HN	Hypertensive nephropathy
LBW	Low birth weight
LPD	Low protein diet
NSAID	Non-steroid anti-inflammatory drug
OR	Odds ratio
OS	Oxidative stress
OTC	Over-the-counter
pmp	Per million people
PY	Pack-years
ROS	Reactive oxygen species
RRT	Renal replacement therapy
SRAU	The Swedish register of renal replacement therapy
WHO	World Health Organisation

INTRODUCTION

The incidence of end-stage renal disease (ESRD) is rising rapidly in many countries [1, 2], although the increase has been less marked in Sweden [3]. Major driving forces behind the global growth of the prevalence pool of treated ESRD patients appear to be broader acceptance of older and sicker patients into renal replacement therapy, improved survival from concurrent or underlying diseases owing to better management of e.g., cardiovascular diseases and diabetes, and increased occurrence of renal disease [4]. Recently, there have been several reports from quite a few countries demonstrating that early signs of kidney disease are common; up to 10-15 % of the general population either has proteinuria, haematuria or a reduced glomerular filtration rate [5, 6]. Both proteinuria and a small reduction in glomerular filtration rate are strong predictors of future risk of established renal failure [7, 8].

The survival among patients treated with dialysis, particularly among the elderly, is no better than that documented in many cancers [3], but even earlier stages of renal failure are associated with 2-to 3-fold increased risks of death [9]. Patients with renal failure have a high morbidity related to the kidney disease and the prevalence of cardiovascular diseases is high. In addition, the financial burden posed by ESRD is heavy [1]. Against this background, prevention of chronic renal failure must be of high importance. There is growing evidence that a multitude of lifestyle-related factors or environmental factors, many of them preventable, might influence the occurrence or progression of chronic renal failure (CRF) [10, 11].

The aim of this work was to evaluate if, and to what extent, some lifestyle-related factors influence the risk of CRF in Sweden.

BACKGROUND

DEFINITIONS

Chronic renal failure (CRF) is a pathophysiologic process with multiple aetiologies, resulting in a progressive loss of function and number of functioning units (nephrons) in the kidney. Due to the initially asymptomatic nature of renal disease, the condition is often undetected until late in the course. The renal failure can lead to end-stage renal disease (ESRD), which is defined as a condition where the irreversible loss of renal function is of a sufficient degree to render the patient permanently dependent upon renal replacement therapy (dialysis or transplantation) in order to survive [12].

Measurement of serum creatinine is the most commonly used screening test for renal failure. However, it is an insensitive measure, as much as 50 % of the nephron mass may be lost before creatinine concentration increases, and levels are influenced by several factors such as sex, age, body mass, muscle mass and diet [13]. Estimated glomerular filtration rate (GFR) is the best measure of the level of kidney function, and in clinical practice measurements of serum creatinine typically form the basis for such estimates. The widely used Cockcroft-Gault equation, which includes age, weight, sex, and serum creatinine, provides a rather valid assessment of GFR [14]. In young adults the GFR is approximately 125 ml/min, but after the age of 30 it declines by on average 1 ml/min annually [15]. If GFR falls below 5 to 10 % of normal, continued survival without renal replacement therapy (RRT) becomes impossible.

The National Kidney Foundation has suggested the following definition of chronic kidney disease (CKD): established kidney damage with structural or functional abnormalities or a glomerular filtration rate <60 ml/min/1.73 m^2 for three months or more [14]. The classification of stages of CKD is based on the level of kidney function measured by GFR, where stage 1 represents kidney damage with normal or elevated GFR and stage 5 represents a GFR of less than 15 ml/min or treatment with dialysis. However, this classification has not been generally adopted, and a new international modified version is currently being developed [16].

DESCRIPTIVE EPIDEMIOLOGY

Registers of patients on renal replacement therapy (RRT) have been established in several developed countries. The Swedish register of renal replacement therapy (SRAU) started in 1991. Thus, the incidence and prevalence of patients with ESRD accepted for RRT are known in countries with registers, although the absolute numbers of ESRD are uncertain. In less developed countries economic factors and health-care resources strongly affect the RRT rate [2]. Typically, the occurrence of kidney disease overall in the general population is unknown, with exceptions in countries or areas where incidence or prevalence studies have been performed.

Occurrence of end-stage renal disease

By the end of year 2002, 6,761 patients were on renal replacement treatment in Sweden (36 % with haemodialysis, 12 % with peritoneal dialysis and 52% had a well-functioning transplant), corresponding to a prevalence of 756 per million people (pmp). Every year more than 1100 new patients enter the Swedish register, corresponding to an incidence of 125 pmp per year [3].

Internationally, the incidence rates and prevalence differ substantially between countries. Japan has the highest prevalence worldwide (1730 pmp in 2001) followed by Taiwan and USA [2]. The incidence rate in USA reached 333 pmp per year in 2002 [17], and varied in Europe between 76 (Iceland) and 170 pmp per year (Dutch-speaking Belgium) in 2002 [18]. A high incidence is also reported from Japan (252 pmp/year) [19], whereas it is lower in Australia and New Zealand (92 and 107 pmp/year, respectively) [20].

The need for RRT increases steeply with age; more than half of the patients treated with dialysis worldwide are over 65 years [21]. In Sweden, the average age of the prevalent population treated with dialysis or renal transplantations in 2002 was 56.8 years, an increase of 4.7 years since the start of the register in 1991 [3]. The mean age of new patients in 2002 was 63.4 years.

Occurrence of kidney disease

Early-stage kidney disease is commonly encountered in the population. In the US it is estimated that 11 % of the adult population (about 19 million individuals) have some sign of chronic kidney disease (proteinuria and/or reduced GFR). Approximately 5 % have an estimated GFR of less than 60 mL/min [6]. In Australia, 16 % of the adult population were reported to have at least one indicator of kidney damage (proteinuria, hematuria, or reduced GFR) and about 11 % had a GFR that was lower than 60 ml/min [5]. In a study from United Kingdom 0.56 % of the general population had a serum creatinine of ≥180 micromol/L (men) and ≥135 micromol/L (women) [22]. From another UK study the incidence rate of CKD (defined as persistently increased serum creatinine of ≥150 micromol/L) was 1,701 pmp per year [23].

Trends

End-stage renal disease is an escalating public health problem throughout the world. In 2001, approximately 1.5 million people were undergoing treatment for ESRD, an increase by 7 % compared with previous year. It is expected that the number of patients will reach 2.5 million by 2010 [2]. A rapid rise is anticipated in developing countries. In Japan the RRT rate has increased by 11 % annually during the last decade [24], in Europe by 4.8 % [25] and in the US by 5.3 % in the early 1990s, albeit the increase has fallen during the last years to below 2 % [32]. In Sweden, the incidence rate increased during the 1990s, but has remained relatively stable during the last 5-year period [3]. Since 1991 all types of RRT in Sweden have increased in prevalence, the annual average increment has been 5.1%.

Sex differences

There is a striking gender difference in the incidence of patients with ESRD accepted for RRT. In 2002, the female to male ratio was 1: 1.7 in Sweden and 1: 1.4 in the United States [3, 32]. It appears that men have a more rapid progression of renal insufficiency compared with women, at least in non-diabetic renal diseases [27, 28]. This finding is supported by animal studies [29]. Sex hormones are suggested to mediate the difference in progression rate, but it remains unclear whether the presence of testosterone or the absence of estrogens, or both factors combined, might explain the greater susceptibility of the male kidney [30].

Racial/Ethnic differences

There are dramatic ethnic/racial discrepancies in ESRD occurrence. A remarkable, up to eightfold increased rate of RRT is seen among indigenous groups in Australia and New Zealand compared with the non-indigenous population [31]. In US, the incidence of ESRD is four times higher among blacks than in whites [32]. The reasons for these differences are not fully understood but it appears that both genetic, environmental, lifestyle habits and possibly health care-related factors are of importance [33]. A high prevalence of hypertension and diabetes in some populations contributes to the variation in risk, in addition, it appears that individuals with these conditions in some ethnic/racial groups have a higher susceptibility to kidney damage and a more rapid progress towards ESRD. However, also other types of renal diseases are more common in some populations, e.g., glomerulonephritis in Afro-Americans and in indigenous groups in Australia/New Zealand [32, 34]. Since low birth weight (LBW) has been tentatively linked to ESRD, and LBW is common in some high-risk populations, this factor has been proposed to be one contributory reason for the disparate incidence rates [35]. An alternative interpretation, however, is that ethnicity might explain the association between birth weight and risk of ESRD observed in some studies.

Prognostic aspects

According to the Swedish register, the mortality is approximately 28 per 100 person-years among patients in dialysis and 3 per 100 person-years among patients with kidney transplants. The crude five-year survival among patients in dialysis is approximately 23 %, and it varies between 70 % in the youngest patient group to 14 % among patients older than 65 years. The five-year survival among patients with kidney transplants is approximately 85 % [3]. Also moderate CRF is associated with a substantially increased mortality; an estimated GFR of 30-44 mL/min almost doubles the risk of death compared with the corresponding risk in the general population, and a more than tripled mortality is seen among patients with a GFR of 15-30 ml/min [9].

ETIOLOGY AND RISK FACTORS

The "multi-hit" theory of renal disease

Some renal diseases, i.e. rapidly progressive glomerulonephritis, are sufficient causes for a rapid permanent total loss of renal function. However, the majority of renal diseases progress slowly over decades, initially often without symptoms, sometimes making it difficult to define the aetiologies. There are strong indications that environmental and lifestyle factors influence kidney function over time, although genetic factors also appear to be of importance [36]. First, renal function declines generally with aging also in healthy subjects, however, the decline is not uniform but varies substantially between individuals [37]. Secondly, the incidence of renal injury varies considerably among individuals who are at-risk for chronic kidney disease (e.g. hypertension, diabetes mellitus). Finally, the rate of loss of renal function is highly variable among individuals with the same underlying cause of renal injury [11]. This individual variability probably reflects the multifactorial nature of the etiologic and biologic mechanisms involved in a substantial part of chronic renal failure.

In the "multi-hit" hypothesis of chronic renal failure it is suggested that a "single hit" in many cases is insufficient for causing overt renal disease, but in combination with several hits, overtly progressive renal disease may develop [38]. It is known from autopsy studies that the number of nephrons varies substantially in humans, from 300,000 to 1,200,000 per kidney [39]. A congenitally small number of nephrons might constitute a "first hit" which predisposes for overt renal disease later in life. Although no strong candidate genes have yet been identified, inheritance of hypothetical genotypes associated with particular susceptibility may similarly constitute a first hit. Acquired loss of functioning nephrons due to renal disease, ageing, obesity, and a multitude of harmful exposures throughout life, would constitute "second hits". Several risk factors for the occurrence or progression of CRF are now identified or suspected (Table 1).

Table 1. Established or suspected factors associated with the occurrence or progression of chronic renal failure.

Specific kidney diseases	Race and ethnicity
Hypertension	Familial aggregation /hereditary factors
Diabetes	Low birth weight
Insulin resistance /Hyperinsulinemia	Short stature
Hyperlipidemia	Obesity
Chronic anaemia	Cigarette smoking
Proteinuria	Illicit drug use
Oxidative stress	Analgesics /other nephrotoxic drugs
Older age	High intake of proteins
Male gender	Lead, Cadmium and other heavy metals
Low socio-economic status	Organic solvent

Progression of renal disease

Although certain renal diseases and renal damaging conditions or exposures might have their own specific pathophysiologic mechanisms that initially lead to nephropathy, experimental animal data suggests that a wide variety of kidney diseases have a common pathway for progression to CRF [40]. It appears that when glomerular filtration rate falls to about half of normal, further loss of function often ensues, regardless of the initial causes of kidney damage [10]. The common pathway of progression includes systemic hypertension, glomerular hypertension, increased glomerular permeability and proteinuria, ultimately leading to inflammation, scarring, and renal failure. It appears that the excessive protein filtration, caused by the glomerular hypertension, might *per se* have toxic effects on the kidneys and increases the rate of progression [40, 41].

Based on results from studies in rats, Brenner *et al* suggested, already in the 1980s, that hyperfiltration and glomerular hypertension may play important roles [42]. Hyperfiltration is observed in diabetes and obesity, but also in any condition associated with a reduced number of nephrons [43]. To compensate nephron loss, the glomerular plasma flow rate and glomerular capillary hydrostatic pressure increase in the surviving nephrons, thus raising the single-nephron glomerular filtration rate. These changes are adaptive initially, by maintaining the overall GFR. However, the glomerular hypertension has negative long-term effects and causes progressive renal sclerosis in a self-perpetuating vicious cycle, whereby nephron loss due to sclerosis further increases flow and pressure in the remaining glomeruli leading to a gradual progress of CRF [42]. Angiotensin II seems to be a central mediator of the observed glomerular hemodynamic changes, but it also controls other factors that might be of importance in the progression of kidney disease, such as the production of oxygen species, the regulation of cytokines and profibrotic growth factors, among others. Inappropriate activation of other systems, such as the sympathetic system, the endothelin system and of aldosterone, has also been implicated in the progression of CRF [44].

Renal disease

Renal diseases are of course the strongest risk factor for CRF and ESRD. Glomerulo-nephritis used to be the leading cause of ESRD, but during the last decades there has been a worldwide shift in the diagnosis of ESRD leading to RRT; diabetic nephropathy and hypertensive nephropathy are now the most common diagnoses [3, 25, 32]. The incidence of ESRD of uncertain origin has increased in many countries during the last decades. In Europe, this group doubled in a 10-year-period, and in 1999 22 % of the new patients entering the European register had not a clear diagnosis [25]. In Sweden, 9.2 % of the incident patients had an unknown cause of the ERSD in 2002 [3].

Hypertension

There is compelling evidence from the epidemiological literature that hypertension causes a decline in renal function [8, 45, 46] and increases risk of ESRD [47, 48]. However, some investigators have questioned whether non-malignant hypertension (in contrast to malignant) is an important initiator of kidney disease [49, 50]. Although the evidence that

hypertension accelerates the progression of already existing renal failure is overwhelming, there is a lack of conclusive data from clinical trials that aggressive treatment of hypertension reduces risk of kidney disease *onset*. A meta-analysis, including 10 prospective controlled trials investigating the relation between hypertension and subsequent loss of renal function, could not confirm that optimised medical treatment of non-malignant hypertension prevented kidney disease [49]. It is conceivable, though, that the duration of the clinical trials was too short to clearly demonstrate the beneficial effect of good blood pressure control.

The diagnosis of hypertensive nephropathy (HN) now accounts for 18 % of incident cases of ESRD in Sweden, and 30 % of new patients in United States [3, 32]. However, the entity "hypertensive nephropathy", i.e., CRF as a consequence of hypertension, deserves some extra comment. Since hypertension accompanies virtually all renal diseases it might be difficult to disentangle whether hypertension caused the renal failure or was a consequence of it. A clear diagnosis of HN requires that a renal biopsy is performed, which is rarely done. Hypertensive nephropathy is usually diagnosed on clinical grounds alone and is sometimes a diagnosis of exclusion, or made when the cause of renal failure is unknown [51]. In a Japanese study of ESRD patients only 2.6 % of those diagnosed with HN had a biopsy performed [52]. Biopsy studies of patients clinically diagnosed with HN have confirmed that misclassification, indeed, exists [53, 54]. In one study, only half of the HN patients, in fact, had genuine HN, while the rest had "atheromatous vascular disease" [54], and in a case report, renal biopsies from two patients showed signs of focal segmental sclerosis [53]. It appears that the clinical entity of HN is composed of a mixture of CRF due to hypertension, renovascular disease, small-vessel arteriosclerotic disease within the kidney, but it might also include unidentified primary renal parenchymal diseases [55].

Diabetes

Diabetes contributes importantly to the increasing burden of ESRD [56, 57], and the rapidly rising trend in type II diabetes prevalence throughout the world is of major concern [58]. There is considerable variation in incidence of type II diabetes-linked ESRD between populations and countries, and it appears that this marked difference cannot solely be explained by variations in the prevalence of type 2 diabetes [59]. There are indications that genetic susceptibility for nephropathy development may be in operation in both type I and type II diabetes, although gene hunting studies have been unable to identify any particular mutations which could explain the development of diabetic nephropathy in the majority of diabetic patients [60]. Beside genetic factors, changing environmental or behavioural factors appear to be of importance for the development of diabetic nephropathy. In Pima Indians, where both type II diabetes and diabetic nephropathy are highly prevalent, the incidence rate of proteinuria among type 2 diabetics has increased nearly two-fold during the last 40 years, despite improvements in plasma glucose and blood pressure [61].

Tobacco

Historical remarks

Tobacco denotes a stimulant of leaves from any of numerous Nicotiana species. It has been used for centuries, initially by indigenous populations in America for medicinal and ceremonial purposes. It was brought to Europe by Columbus, primarily as a medicinal plant as it was thought to act as a panacea for a diversity of medical conditions. Later, tobacco started to be used as a stimulant, and early in the 20th century the habit of using tobacco products was widespread [62].

Prevalence of tobacco users

It is estimated that the total number of cigarette smokers worldwide is one billion [63]. Generally, the tobacco smoking prevalence is decreasing in developed countries and increasing in developing countries. Cigarette smoking has decreased steadily during the past decades in Sweden; the prevalence is now the lowest in Europe. Between 1980 and 2003 the proportion smokers fell from 36% to 17% among men and from 29% to 18% among women [64]. In contrast, the use of smokeless tobacco has increased in Sweden, particularly among male adolescents and athletes. In 1999, 19 % of men and 1% of women were daily users of the Swedish product snus [65].

General health effects of tobacco

Tobacco smoking is considered to be the most common identifiable cause of adult death in developed countries, with the exception of hypertension. The World Health Organization estimated that 4.9 million premature deaths from smoking occurred in year 2000, and the yearly number is expected to be 10 millions in 2020 if current smoking patterns continue [63]. The effect of smokeless tobacco on human health is more uncertain. Swedish studies about health effects of snus have shown no clear link between such use and various types of cancer [66-68]. A Swedish cohort study showed an increased risk of death from cardiovascular disease among users of smokeless tobacco [69], but this finding was not confirmed in a US study [70].

Tobacco and the kidney

In recent decades a growing body of literature has emerged, supporting the idea that smoking is associated with adverse effects on the kidneys. Strongest evidence exists for a detrimental effect of smoking on the kidneys in diabetes, but most data point to a similar smoking-related negative impact on kidneys in individuals with hypertension or a pre-existing renal disease. Moreover, experimental studies and population-based epidemiological studies indicate that smoking may cause renal damage also in healthy individuals, independent of other factors.

In diabetes, smoking has been linked to increased risks of microalbuminuria development, accelerated progression from microalbuminuria to manifest proteinuria, and accelerated progression of manifest renal failure [71]. Most data support a two- to three-fold increased risk of nephropathy among smoking diabetics compared with non-smokers [71] although the WHO Multinational Study of Vascular Disease in Diabetes did not find any association between smoking and CRF after, on average, 8.4 years of follow-up [72]. Initially, the increased renal risk from smoking was only reported in type I diabetes [73, 74], but it is now recognised also in patients with type 2 diabetes [75, 76]. It

appears that some diabetics are more sensitive to renal effects of smoking than others, and a genetic susceptibility to develop adverse renal effects from smoking among diabetics has been hypothesised [77].

In two cross-sectional studies, smoking was a strong predictor of both micro-albuminuria and macroalbuminuria in hypertensive individuals [78, 79], and a prospective study showed that smoking was the most powerful predictor of a decline in renal function among individuals with severe hypertension [80]. In contrast, a large cohort study of almost 12.000 hypertensive men could not confirm a statistically significant relation between smoking and risk of ESRD after a minimum of 14-years' follow-up [47].

The role of smoking in primary renal disease is less investigated. Two case-control studies could not verify the hypothesis that smoking increases risk for *appearance* of glomerulonephritis [81, 82]. In contrast, studies have yielded positive associations between smoking and progression rate among men with glomerulonephritis or autosomal dominant polycystic kidney disease [83, 84]. Studies investigating the role of smoking on progression in lupus nephritis have reported disparate results [85, 86].

A number of cross-sectional studies from the general population in Europe, USA and Australia have demonstrated a higher prevalence of albuminuria/proteinuria among smokers compared with non-smokers, independent of diabetes, hypertension and other factors [87-90]. A case-control study among elderly reported that an increased serum creatinine level was independently related to smoking [91]. A cohort study with individuals free of renal disease at entry, found a 42 % increased risk of renal impairment (the fifth or lower percentile) among smokers, after a median follow up of 18.5 years [8]. Another prospective study found a significant association between cigarette smoking and risk of ESRD [92].

To my knowledge there are no previous studies that have investigated whether smokeless tobacco might be a risk factor for kidney disease.

Protein intake

Already more than 50 years ago Addis suggested that a low protein diet (LPD) could preserve renal function in patients with CRF [93]. He hypothesised that a LPD would reduce the workload of surviving nephrons in diseased kidneys and thus minimise further loss of renal function. Brenner et al extended this view and postulated the "hyperfiltration theory", based on animal studies. He suggested that sustained excesses of dietary protein cause increases in renal blood flow and glomerular filtration rates that lead to "intrarenal hypertension", ultimately resulting in progressive glomerular sclerosis and deterioration of renal function [42].

Protein intake and progression rate of renal failure

Numerous studies have now investigated the effect of a LPD on progression rate in various renal diseases, showing somewhat disparate results [94-99]. Although the largest trial was inconclusive [96], post hoc analyses indicated that a LPD retarded the deterioration of renal function [100]. Several meta-analyses report that dietary protein

restriction slows the progression of moderate and severe, both diabetic and non-diabetic, renal diseases [101-103], but the effect appears to be modest. These results, together with the fact that a LPD relieves symptoms related to uraemia, has led to a recommendation of a protein intake of about 0.6 g/kg/day in persons with moderate or advanced CRF [104]. A limitation of protein intake in kidney transplant recipients may also be beneficial; this strategy appears to stabilise renal function [105].

Protein intake and effect on kidneys without pre-existing disease

Whether an excessive protein intake can be detrimental in subjects without kidney disease is less evaluated. *Acute* effects of a protein load (increases in renal blood flow and glomerular filtration rate) have repeatedly been demonstrated, not only in animals but also in physiological experiments among humans without pre-existing renal disease [15]. Data on long-term effects of a *chronic* high protein intake on healthy kidneys is scant. In the Nurses Health Study there was no association between protein intake, estimated from a food frequency questionnaire, and decline in glomerular filtration rate among women with an initially normal renal function, after 11 years of follow-up [106]. In contrast, in women with a mild renal failure at baseline (GFR between 55 and 80 mL/min), high protein intake was linked to a more rapid loss of renal function. One cross-sectional study reported that a 0.1 g/kg/day increment of protein intake was independently associated with an increased risk of microalbuminuria [107], while another study found a positive relation between a high protein intake and microalbuminuria only in persons with both diabetes and hypertension [108]. In type I diabetics, a high protein intake was linked to proteinuria [109].

Oxidative stress and antioxidant intake

What is oxidative stress?

Reactive oxygen species (ROS) are not only toxic by-products of metabolism but act also as essential biomolecules in cell signalling and regulation. ROS are tentatively involved in various physiological processes such as cell proliferation and differentiation, apoptosis, immunity, and defence against microorganisms [110]. Oxidative stress (OS) occurs when there is an imbalance between the production of ROS and the capacity of the antioxidant defence system. The defence is constituted by enzyme systems that remove ROS and non-enzymatic endogenous scavengers, but dietary antioxidants, such as vitamin C, vitamin E, beta-carotene, and other micronutrients (i.e. caretonoids, polyphenols and selenium) play a major role in maintaining the oxidative balance [111]. The main sources of vitamin E are certain vegetable oils, whole grains, nuts and green leafy vegetables. Fruit and vegetables are the major sources of many other dietary antioxidants – not only vitamin C and beta-carotene. Oxidative stress may lead to genetic alterations, impaired cell function, and plain tissue damage, and is implicated in the pathogenesis of cancers, cardiovascular disease, hypertension and other chronic diseases [111].

Oxidative stress and the kidney

It is evident that oxidative stress is present in ESRD [112]. It has been attributed mainly to the uremic state *per se* and to the effect of the haemodialysis process [113]. However, there are several lines of evidence suggesting that OS has a role in the pathogenesis of CRF. In experimental studies, OS causes pathophysiological changes typically seen in renal scaring, such as cell proliferation, apoptosis, inflammation and vascular injury, and pathological changes in the kidneys similar to those seen in chronic kidney disease [114]. In addition, excessive amounts of ROS have been recorded in animal models of kidney diseases for glomerulonephritis, interstitial fibrosis and hypertensive nehprangio-sclerosis, among others [114]. In humans, oxidative stress is not only present in uraemia but also in patients with mild CRF [115], and there is no close relationship between the degree of OS and the stage of renal failure [116], facts suggesting that OS may have a pathogenetic significance. Furthermore, both in animal and in humans, treatment with antioxidants appears to ameliorate kidney damage [114].

Several observational studies have found that a high intake of antioxidants, at least vitamin E, reduces risk of cardiovascular disease [117]. There are limited data from the general population whether or not a large intake of antioxidants protects against kidney disease. In an American cross-sectional study, serum vitamin E levels were not significantly associated with serum creatinine elevations, whereas vitamin C was inversely related to hypercreatininemia in two out of three studied races/ethnic groups [118]. In the latter study a high serum vitamin A appeared to increase the probability of serum creatinine elevation. Another cross-sectional study found no association between serum alpha-tocopherol and elevated urinary albumin, while concentrations of certain diet-derived carotenoids were inversely associated with albuminuria [119].

The seemingly protective effect of antioxidants against cardiovascular diseases found in observational studies has not been confirmed in controlled prospective studies [120, 121]. There are no such prospective intervention studies aimed at preventing CRF in the general population, but several studies have investigated the impact of antioxidant supplements, mostly vitamin E, on the progression rate in already established kidney diseases. Vitamin E administration resulted in a normalisation of elevated baseline creatinine clearance in type 1 diabetes [122], decreased proteinuria in children with focal segmental glomerulosclerosis [123] and (in combination with vitamin C) in type 2 diabetes [124]. Treatment with vitamin C reduced albumin excretion rate among diabetics [125].

Obesity

Prevalence of obesity

Obesity has become a major problem around the world. Although this phenomenon used to be a result of altered dietary patterns and a sedentary lifestyle among people in affluent developed countries, obesity is now a rapidly emerging problem also in developing countries. Obesity rates have risen 3-fold or more since 1980 in some areas of North America, the United Kingdom, Eastern Europe, the Middle East, the Pacific Islands, Australia and in China. According to estimates by the World Health Organisation (WHO), one billion adults are overweight (body mass index [BMI] ≥25

kg/m^2), and of these, at least 300 million are obese (BMI ≥30 kg/m^2) [126]. There are, further, great concerns about the rising prevalence of overweight and obesity among school children and adolescents. According to WHO's definition of overweight and obesity [127], 64 % of the adult population in United States are overweight (BMI ≥25 kg/m^2), almost one third are obese (BMI ≥30), and close to 5 % of the population are considered to be extremely obese (BMI ≥40) [128]. Although Sweden has one of the lowest rates of overweight and obesity in Europe, rates are increasing [129]. The prevalence of obesity (BMI ≥30) in 1996/1997 was reported to be 11.9 % and 10 %, among Swedish women and men, respectively. Obesity contributes significantly to the burden of chronic diseases, numerous diseases are caused or made worse by obesity, such as cardiovascular diseases, cancers, type 2 diabetes, and hypertension, among others [130].

Epidemiological studies linking obesity to kidney disease

The alarming increment of obesity worldwide has been paralleled by a steadily increasing incidence of ESRD due to hypertension and type 2 diabetes [3, 32]. Unquestionably, much of the excess risk for CRF observed among obese people [131, 132] is driven by the increased prevalence of hypertension and/or type II diabetes [133, 134]. However, it also appears that obese individuals diagnosed with hypertension and diabetes are at higher risk of developing nephropathy, compared with leaner subjects with these conditions, independent of blood pressure, glucose concentrations and other factors. In epidemiological studies, a high BMI was independently linked to proteinuria among diabetics [135, 136], and hypertensive subjects [137].

Obesity may also aggravate pre-existing nephropathies. Excessive body weight was reported to be an independent risk factor for progression in IgA nephritis [138]. Results from a cross-sectional study investigating the effects of excess weight on renal function after unilateral nephrectomy strongly indicate that obesity has an adverse effect on remaining nephrons [139]. In this study 92 % of obese subjects had developed protienuria and/or renal insufficiency 20 years after the nephrectomy, whereas these complications were only found in 12 % of non-obese subjects. In multivariate analyses, BMI at time of the nephrectomy was the only statistically significant factor related to development of renal disease, independent of hypertension, diabetes and other factors. In another study obesity was reported to increase risk of graft failure after renal transplantation [140].

There are indications that obesity may also affect the kidneys in otherwise healthy subjects. Experiments in obese Zucker rats, a genetic model of obesity, have revealed various renal disturbances, such as hyperfiltration and glomerulosclerosis [141, 142]. Renal function studies in humans suggest that obese subjects have an elevated GFR and/or renal blood flow [143, 144], and it appears that even moderate overweight is associated with unfavourable renal hemodynamic patterns [145]. Epidemiological studies from the general population have found a link between BMI and proteinuria [146, 147] and with a reduction in GFR [8], independent of other factors. Further evidence for a link between obesity and kidney damage is provided by the fact that weight loss in obese reduces proteinuria and hyperfiltration [148, 149].

Suggested mechanisms of obesity-induced kidney damage

The mechanisms behind the adverse effect of obesity on kidneys are not completely known, but there is an intriguing similarity between the hemodynamic renal effects of obesity and those of diabetes. In both conditions an increased renal blood flow and glomerular filtration rate is observed, mediated by vasodilatation of the afferent arterioles [131, 150, 151]. Animal studies suggest that this hyperfiltration and increased glomerular hydrostatic pressure causes progressive glomerulosclerosis [42, 150].

The hormone Leptin, mainly produced in adipocytes, has been implicated in the pathogenesis of obesity-related kidney damage [131, 150, 151]. In animals leptin stimulates cellular proliferation and increases the expression of prosclerotic cytokines that may lead to glomerulosclerosis. A low-grade systemic inflammation, commonly seen among obese, may also be of importance [131, 150, 151]. It is known that adipocytes produce cytokines and that obese subjects often have elevated levels of CRP, and higher levels of CRP have been linked to a reduced GFR. Other suggested mechanisms include an up-regulated activity of the renin angiotensin system, effects of obesity-related hyperlipidemia and increased oxidative stress [131, 150, 151].

There are suggestions from human studies that the *fat distribution* may be of importance for the development of kidney disease. These studies showed that central adiposity is a greater risk factor for kidney disease than peripheral adiposity [146, 152]. Central adiposity is closely linked to the metabolic syndrome [153], and evidence relating this syndrome to renal disease has emerged [154]. Several conditions of the metabolic syndrome, such as diabetes, hypertension and possibly dyslipidemia, are established risk factors for the initiation or progression of CRF. However, insulin resistance and concomitant hyperinsulinemia, other features of the metabolic syndrome, may also be related to CRF risk, even in the absence of clinically manifest diabetes. Cross-sectional studies suggest that insulin resistance/hyperinsulinemia is associated with microalbuminuria [155], and with CRF [156], independent of diabetes and other factors. Suggested adverse renal effects from hyperinsulinemia are preglomerular vasodilatation and subsequent glomerular hypertension, stimulation of the expression of inflammatory collagen and promotion of lipogenesis and subsequent renal lipotoxicity [154].

Birth weight

Perinatal programming

The hypothesis that the fetal environment may have consequences for adult health was first proposed by David Barker. He reported that low birth weight (LBW) was associated with an increased risk of dying from ischemic heart disease [157]. The elevated risk appeared to be confined to those who were small at birth as a result of growth retardation, rather than to those born prematurely. A considerable number of reports are now published relating intrauterine malnutrition to a number of diseases in adult life, such as cardiovascular disease, hypertension, diabetes and renal disease [158, 159]. This concept is called perinatal programming, or the fetal origins of adult disease. However, more recent data indicate that growth patterns in childhood may also be of importance for future risk of disease [159].

Experimental studies suggest that not only energy *restricted* maternal nutrition, but also *altered* nutrition, or prenatal exposure to certain substances, may increase adult risk of chronic disease in the offspring [159]. The mechanisms by which perturbations in the fetal environment increases the susceptibility for chronic diseases in adult life are not established, but disturbances in several pathways and systems are suggested, such as the hypothalamic-pituitary-adrenal axis, the sympathetic nervous system and the renin-angiotensinogen system [160].

The kidneys appear to be particularly sensitive to an unfavourable prenatal environment [160, 161]. Studies of human fetuses and new-borns showed an association between intrauterine growth retardation and nephron number or reduced renal volumes. Animal studies have revealed that maternal restriction of energy or protein results in a reduced number of nephrons, but also restriction of Vitamin A or iron, as well as intrauterine exposure to certain antibiotics, hyperglycaemia and high levels of glucocorticoids, may reduce the nephron number. It is suggested that a small number of nephrons, "oligonephropathy", may result in hyperfiltration and glomerular hypertension, which might lead to increased future risks of glomerulosclerosis, hypertension and renal failure [162].

Previous epidemiological studies of birth weight and kidney disease

Two previous case-control studies from the United States found that a birth weight below 2.5 kg was independently linked to an increased risk of ESRD among whites [164] and blacks [164, 165]. In a cross-sectional study in an Australian Aborigine community (where both kidney disease and low birth weight is common), a 2.5-fold increased risk of albuminuria was observed among those with a low birth weight, independently of the presence of hypertension or diabetes [35]. In contrast, neither a cohort study, nor a case-control study of type I diabetics, found any relation between birth weight and nephropathy [166, 167]. A study among Pima Indians revealed a u-shaped relation between birth weight and albuminuria in diabetics [168].

Height and kidney disease

Beside low birth weight, short stature has also been suggested to be a possible indicator of an unfavourable intrauterine environment. In cross-sectional studies, short stature was linked to the degree of albuminuria in both diabetics and non-diabetics [169-171]. A small cohort study of type I diabetics showed that a short stature was associated with an increased progression rate of nephropathy [172].

Analgesics

Analgesics are widely used in the general population, both as self-medication sold over-the-counter (OTC) and as prescribed drugs by physicians. In Sweden in 1996, approximately 30 % and 20 % of women and men, respectively, reported use of OTC analgesics during a two-week recall period, while 12 % of the women and 7 % of the men had used analgesics with prescription [173]. Currently, the most commonly prescribed analgesic drug is aspirin, while paracetamol (acetaminophen) is the number one OTC analgesic, followed by ibuprofen [174].

Analgesics and kidney damage-the early reports

Fifty years ago, Spuhler and Zollinger observed an association between chronic interstitial nephritis and excessive consumption of combination analgesics containing phenacetin [175]. Soon thereafter, reports of an increased occurrence of renal papillary necrosis among heavy users of phenacetin started appearing [176, 177]. A prospective study among women in Switerland found a small but significant increased risk of proteinuria, heamaturia and elevated serum creatinine after heavy use of phenacetin-containing products [178].

In the Swedish town of Huskvarna it was observed that the number of deaths from uraemia had increased dramatically during the 50s. The most striking findings from an examination of uraemia-related deaths during 1952-61 was that 80 % of those who had died from uraemia had been employed at the factory of the Huskvarna Company and among these, 65 % used to be regular consumers of analgesics with phenacetin. The origin of this abuse could be traced back to the Spanish influenza pandemic of 1918-1919 when a physician named Hjorton was working in Huskvarna. He prescribed a compound consisting of caffeine, phenazone and phenacetin – "Hjortons powder", against fever and muscle pain. Soon it was believed that this powder was strengthening also in healthy individuals, and the phenacetin use became a social habit in Huskvarna, and thereafter the powder spread to the rest of Sweden as an over-the-counter drug. The abuse was particularly pronounced at the factory of Huskvarna Company, the employees were encouraged to take a few "powders" regularly throughout the working day to be able to keep up the productivity and to reduce strain. A study among the employees at the factory was set up, and it revealed that among users of phenacetin 34 % had renal damage of varying degree, whereas only 2.4 % of non-users had such injury [179].

"Classical" analgesic-associated nephropathy (AAN)

The nephropathy associated with analgesic use was initially called "phenacetin nephropathy", since phenacetin appeared to be the common denominator. However, soon other analgesics also came under suspicion and the name was replaced by "analgesic-associated nephropathy" (AAN). This condition has been characterised as a slowly progressing renal disease, more commonly seen in women then in men, that relentlessly advances to ESRD unless the analgesic use is discontinued. Related clinico-pathological features described are relatively non-specific; anaemia, hypertension, peptic ulcers, weight loss, sterile pyuria, hematuria, defect urinary concentration and acidification, among others [180]. The histological changes typically seen in AAN are renal papillary necrosis, capillarosclerosis and chronic interstitial nephritis [181], although not pathognomonic for AAN. Also the "hallmark" of AAN – renal papillary necrosis – is seen in other conditions such as diabetes mellitus, urinary tract obstruction or infection and sickle cell disease [182].

AAN emerged as a public health problem in many countries such as Switzerland, Sweden, Denmark and Australia [180]. Australia reported the highest incidence, in some geographic areas almost 30 % of patients with ESRD were diagnosed with AAN in 1979 [180]. After legislative measures against phenacetin-containing analgesics, which coincided with a ban of combination analgesics in some countries, the incidence started to fall in Sweden and in other countries. Yet, still in 1989, 16 % and 21 % of ESRD in

Belgium and Switzerland, respectively, was attributed to AAN [183]. In 2002, the European dialysis register reported that the incidence of ESRD due to AAN was low (on average <2 pmp/year) and stable since 1980 [18]. In some countries, such as Sweden, Finland, Norway and Greece, the entity of AAN has virtually disappeared among patients entered into the register, whereas the incidence is higher in Switzerland and Belgium. In US, only 0.1 % of incident ESRD patients in 1997 were diagnosed with AAN [184].

There are no universally accepted diagnostic criteria for AAN. The diagnosis is usually made on the basis of exclusion of other renal diseases and on information of analgesic exposure. Some years ago it was suggested that non-contrast CT scan would be the gold standard for diagnosing AAN; certain specific morphological criteria such as papillary calcification, bumpy contours of the kidneys and decreased renal volume, were claimed to have a high specificity and sensitivity for AAN [185]. However, this method has not been generally adopted [186]. There has been a striking geographical variation in incidence of AAN. Ecological studies indicated that countries or areas with high incidence also had a large consumption of analgesics [180, 187]. An inverse relationship between the proportion of patients reaching ESRD due to an unknown cause and the proportion with a diagnosis of AAN was noted [188]. This could indicate an underestimation of AAN in some geographic areas. Since there are no clear diagnostic criteria for AAN the geographic variation may reflect differences in the threshold for making this diagnosis.

Which analgesic(s) is nephrotoxic?

Although phenacetin was almost invariably sold as a combination drug, phenacetin was quickly accused of being the culprit. In addition, phenacetin had other unacceptable adverse effects, such as anaemia, cyanosis and methaemoglobinia [180]. This led to successive legislative measures; in Sweden phenacetin became a prescription drug in 1961 and thereafter it was gradually removed from the market. The availability was restricted in several other countries during the following decade [189]. Soon, several objections against "the phenacetin nephropathy concept" were raised; numerous animal studies with phenacetin failed to produce renal papillary necrosis whereas other analgesics did, the removal of phenacetin in some countries did not reduce the incidence of AAN, and the prognosis for AAN was better if all analgesic were discontinued and not only phenacetin [180].

Paracetamol (acetaminophen), rapidly became a popular drug after the removal of phenacetin from the market [190]. Already early on, its widespread use generated considerable concern since paracetamol is the major metabolite of phenacetin. However, many other types of analgesics, such as salicylates, other NSAIDS and phenazones also came under suspicion [180]. Animal studies indicated that several analgesics, particularly salicylates, were capable of causing nephrotoxicity, including renal papillary necrosis [191]. In the 1970s a new hypothesis emerged, namely that it was the *combination* of analgesics that was responsible for the renal damage [180]. In 1996 an Ad Hoc committee of the National Kidney Foundation stated that "AAN is caused by use of analgesic mixtures containing at least two anti-pyretic agents and caffeine, or codeine, on a daily basis, for at least 3 years" [192]. This statement was strongly influenced by the widely spread view that an isolated ban of phenacetin failed to reduce

the incidence of AAN in Australia and Belgium. Not until combined mixtures were banned a couple of years later in Australia, there was a decline in the incidence. No such decline was observed in Belgium, where mixed analgesics were still available. However, this conclusion has recently been re-evaluated and claimed to be incorrect [193]. In 1999, a peer-review committee of international scientists reviewing the more recent research on analgesics and nephropathy stated that whereas the association for phenacetin is established there is insufficient evidence for a relation between non-phenacetin combined analgesics and nephropathy [194].

Cyclooxygenase inhibitors have well-documented nephrotoxicity. They are known to cause acute renal failure in susceptible patients (i.e., those with volume depletion, CRF, diabetes, congestive heart failure, or old age). In addition, they may cause hypertension, and thus increase the risk of kidney damage. Cases of interstitial nephritis, AAN and renal papillary necrosis have been reported after cycloxygenase inhibitor intake, but there is insufficient data to implicate these drugs as important contributors to *chronic* renal disease [195].

May analgesics cause CRF other than "classical AAN" and/or exacerbate renal failure?

During the last two decades, results from epidemiological studies have suggested that analgesics do not only cause "classical AAN" but may also increase risk of CRF or ESRD in general. Moreover, there are indications that analgesic use may exacerbate pre-existing CRF. Most case-control studies have reported an increased risk of CRF with the use of phenacetin [196, 197], and several other have found positive associations also with other types of analgesics, but often with lower risk estimates than for phenacetin. There are reports of positive associations between use of paracetamol and occurrence of ESRD [198] or newly diagnosed CRF [196]. Other studies found a positive relationship with aspirin and other NSAIDs [197], as well as with combination drugs [199]. In contrast, Murray *et al* found no link between any analgesic drug and ESRD in another case-control study [200]. Likewise, in a study from the "Physicians health cohort", where analgesic use was related to changes in serum creatinine from samples undertaken in 1982 and in 1996, it was concluded that moderate intake of paracetamol, aspirin or other NSAIDS among initially healthy men was *not* associated with risk for impaired renal function [201]. However, in this study creatinine measurements were available for no more than 4,517 subjects out of 22,071, and the precision of the relative risk estimates was fairly low. Moreover, subjects with clinically manifest CRF, either at entry or at follow-up, as well as those dying during the follow-up (14 with CRF) were not included in the computations.

Suggested mechanisms of nephrotoxicity

The mechanisms involved in analgesic-induced renal injury remains unclear, but direct cell injury, free radical formation, prostaglandin inhibition, reduced medullary blood flow and possibly an immunological mechanism are suggested modes of actions [202, 203]. Paracetamol accumulates in the renal medulla, and it is proposed that metabolization of paracetamol results in reactive oxygen radicals, which may cause cell necrosis. It is claimed that aspirin may exacerbate this toxicity by depleting stores of reduced glutathione, which normally prevents bindings of reactive intermediates [202], but this theory has been challenged because it is based largely on unverified extrapolations to

humans from artificial in vitro systems [194]. Prostaglandin inhibition by aspirin and other NSAIDs causes redistribution of renal blood flow from the renal medulla to the renal cortex potentially resulting in medullary ischemia and eventual necrosis of the renal papillae [204].

Other risk factors

Socio-economic status

It is evident that socio-economic status is linked to the development of ESRD, both low income and low educational level elevates risk [205-207]. In the United States, risk varied up to 7-fold with educational level [207]. Some of the variations across educational levels appeared to depend on unequal access to medical treatment, but this did not solely explain the large risk difference. In a previously published paper with data originating from our case-control study, we reported that a low socio-economic status (based on occupations), and low educational level, were risk indicators for chronic renal failure [208]. Socio-economic status *per se* could reasonably not increase risk of kidney disease, but is rather a marker for not yet identified risk factors. The elevated risk among individuals with lower socio-economic status remained after adjusting for potential confounders, such as smoking, alcohol, body mass index and analgesic use. In addition, the prevalence of type 2 diabetes and hypertension were evenly distributed across socio-economic strata, and could not explain the difference in CRF risk. It is unlikely that unequal availability of medical treatment contributes importantly to the uneven distribution of risk since Sweden has a public health care system with equal access for all.

Occupational exposures

Several occupational exposures have long been accused of impairing renal function and causing CRF [209]. Exposure to organic solvents has predominantly been linked to the appearance and exacerbation of glomerulonephritis [210]. Previous literature has suggested an adverse renal effect from silica and several heavy metals, such as cadmium, chromium and lead [211]. The most persuasive evidence exists for cadmium; it causes proteinuria [212] and has been linked to increased risk of ESRD [213]. With data from our nation-wide case-control study, the association between occupation and occupational exposures with risk of CRF has previously been investigated [214, 215]. Although there was substantial variation in risk of CRF across occupational groups, this variation was not explained by exposure to lead, mercury, chromium or grain dust. In contrast, the results suggested a small increased risk from exposure of cadmium and silica. Organic solvents were not linked to risk. These results may partly reflect a limited range of exposure owing to strictly enforced Swedish legislation regarding workers' protection.

Dyslipidemia

Renal failure, in early as well as advanced stages, is associated with abnormalities in lipoprotein metabolism. Dyslipidemia appears to be independently associated with increased progression rate of CRF in patients with kidney disease [216, 217], and with increased risk of graft loss after renal transplantation [218]. Moreover, there are indications that dyslipidemia might initiate kidney disease. Two cohort studies in the

general population reported links between elevated plasma triglycerids, high total serum cholesterol and low high-density-lipoprotein cholesterol on the one hand and increases in serum creatinine at follow-up on the other [219, 220].

Genetic susceptibility

There are indications that a generalised genetic susceptibility contributes to the development of ESRD [36, 221, 222]. The observation that there is a clear familial aggregation of ESRD due to diabetes, hypertension and glomerulonephritis, initiated the search for specific "candidate genes" that might be involved in renal diseases. It is suggested from various types of genetic association studies that genes of the renin-angiotensinogen system and genes coding for cytokines and growth factors might be of interest, among others [221]. Some specific mutations are suggested to increase the susceptibility for glomerular damage, and are implicated in the aetiology of focal segmental nefrosclerosis [223], at least in familial variants – their applicability in sporadic focal segmental nephrosclerosis is more uncertain. However, in the search for specific candidate genes for renal genetic susceptibility, independent evaluations have frequently provided contradictory results. This might be due to the limited extent to which each specific gene contributes; the susceptibility is more likely a result of influences from many genes [36]. Further, the studies have been carried out in different populations with different methodological approaches, possibly contributing to the disparate results. In addition, many studies were hampered by poor statistical power [221]. Studies using the genome scan approach, which has the potential for a more comprehensive evaluation of inheritance throughout the genome and to locate previously unknown genes related to diseases, have recently found evidence of susceptibility loci for diabetic nephropathy [224].

AIMS

The overall aim of this study was to identify risk factors for chronic renal failure. The specific aims included in this thesis are;

To estimate the strength of the association, if any, of cigarette smoking and other use of tobacco with risk of chronic renal failure.

To investigate whether variations in intake of protein and antioxidant vitamins, such as vitamin C, beta-carotene and vitamin E, are associated with corresponding variations in subsequent risk of chronic renal failure.

To explore possible associations between obesity and risk of CRF development, and to study if height and/or birth weight are related to this risk.

To evaluate whether or not analgesic intake is linked to risk of chronic renal failure.

SUBJECTS AND METHODS

SETTING

We conducted a nation-wide population-based case-control study during the period May 20, 1996 through May 31, 1998. The study base was defined as the person-time generated by all native Swedes, aged 18-74 years, who were living in Sweden during the study period. The Swedish Population Register includes information on date of birth, citizenship and country of birth on all residents in Sweden, and provided a well-defined source population including 5.3 million persons, whose eligibility and vital status could be followed throughout the case and control ascertainment period. Before start of the study, all regional ethics committees and the Swedish Data Inspection Board approved the study protocol.

CASES

Men and women, belonging to the study base, with newly diagnosed CRF (i.e., a serum creatinine found to exceed 300 μmol/L [3.4 mg/dL] in men and 250 μmol/L [2.8 mg/dL] in women, for the first time and permanently, during the study period) due to renal causes were eligible as cases. To verify the chronicity of the renal failure, a second creatinine test was obtained three months after the first measurement. The threshold of this second creatinine test was set lower (250 μmol/L for men and 200 μmol/L for women), to allow for day-to day variation. If the creatinine level had fallen below these values, the patient was considered non-eligible. Only patients with a disease "within" the kidney were eligible. All patients with prerenal causes (e.g., severe heart failure) or postrenal causes (i.e. obstruction of the urinary tract) were non-eligible. Eligible patients who had received kidney transplants before start of the study period were excluded.

To ensure that eligible cases throughout the country were identified, we set up a comprehensive organisation that included 68 medical laboratories covering practically all inpatient and outpatient care in Sweden. These laboratories provided monthly lists with all patients who had undergone serum creatinine measurements. We also initiated collaboration with physicians working at all hospitals (n=60) where patients with renal diseases are treated. These physicians reviewed medical records of all patients who were found to have serum creatinine levels corresponding to our inclusion criteria, and decided about final eligibility. They also asked the patients about participation in the study. The diagnosis of the underlying condition that caused the renal failure was determined by the treating physician, and was based on routine clinical work-up.

Of the 1,189 eligible cases identified during the study period, 926 could be enrolled (78 % participation rate). In another 35 patients the eligibility could not be determined. The reasons for non-participation were early death in 69 cases, very poor clinical condition that precluded participation in 83, and patient refusal in 111.

POPULATION-BASED CONTROLS

On three occasions during the study period, controls were randomly selected by Statistic Sweden from 10-year age and gender strata in the Swedish Population register. The number of selected controls in each stratum was chosen to mimic the age and sex distribution among cases (i.e., frequency matching on age and gender). All controls provided informed consent before enrolment. Of the 1,130 control subjects who were contacted, 998 participated (75 % participation rate). Of the remaining, 221 individuals refused to participate, 56 could not be located, and 55 were too ill to participate.

DATA COLLECTION

All cases and controls first received a mailed self-administered questionnaire. It encompassed a wide range of questions about marital status, education, birth weight, anthropometric measures, dietary habits, tobacco use and alcohol consumption. Thereafter, professional interviewers from Statistics Sweden conducted a computer-aided face-to-face interview, using laptop computers. This interview covered questions on medical history, occupational history and work-related exposures, and detailed information on use of non-narcotic analgesics (using a picture guide of relevant packages). This interview lasted, on average, somewhat longer for cases (80 minutes), compared with controls (70 minutes). This difference was largely explained by the more complicated medical history among cases. The interviewers also scrutinised the mailed questionnaire and assisted study participants in completing missing answers. We were not able to blind the interviewers to the case/control status of the interviewees, but they were unaware of the study hypotheses and were specially trained to conduct the interview of all subjects in a strictly standardised manner.

Tobacco (paper I)

Information about tobacco habits was obtained through our questionnaire. The study participants reported whether they had used any type of tobacco (cigarettes, cigars, pipe smoking or snuff), and if the use was ever regular. Our definition of regular tobacco use was smoking either at least one cigarette per day or at least one cigar or pipe per week, or using snuff at least once a week, for a period of 6 months or more. If any tobacco use was reported as regular, we inquired about starting and stopping dates, amounts used, as well as changes in use over time.

Diet (paper II)

We focused on diet 10 years prior to interview. Dietary habits were assessed by a food-frequency questionnaire, including 106 common foods and beverages. Most items had 9 predetermined response categories of consumption ranging from “never/seldom” to “three or more times per day”. The participants also estimated the portion size of most items (“small, “medium” or “large”), although some of the items had a predetermined portion size. We also asked if the study participants had changed their diet during a period lasting for at least for 2 years, what kind of diet they had switched to (e.g., a diet

with reduced protein, vegetarian diet, diabetic diet), and during which years the diet was changed. In addition, the questionnaire also inquired about vitamin supplements.

Body mass, birth weight and height (paper III)

The self-administered questionnaire encompassed several questions on various aspects of body weight, such as current weight, highest lifetime weight, weight at age 20, 40, 60, weight gains and weight losses. The interviewees also reported their height. The questionnaire inquired about birth weight in 6 predefined categories (<2500 g, 2500-2999 g, 3000-3499 g, 3500-3999 g, ≥ 4000 g, or "do not know"). The exact birth weight could also be reported, if it was known.

Analgesics (paper IV)

Before the computer-aided interview started, the study participants reviewed a booklet with colour pictures of the packaging of all analgesics containing paracetamol (acetaminophen) or phenacetin marketed in Sweden between 1960-1996, together with 78 of the most sold brands (out of 174) of other non-narcotic analgesics during that period. Moreover, in order to check that the self-reports were reasonable, the interviewers had detailed lists of dates of introduction on (and withdrawal from) the Swedish market for all types, brands and preparations of analgesics. If the interviewees reported regular use of any brand (e.g., according to our definition; at least one tablet twice a week for two months or more), detailed questions followed on dosage and duration for each period of regular use, and age of the study participant when each period of use started. If subjects had taken more than 20 tablets of an analgesic but reported no regular use (sporadic use), the lifetime cumulative number of tablets of that analgesic was estimated. Nonusers of an analgesic were defined as those whose lifetime cumulative intake did not exceed 20 tablets. The interview also covered questions on changes in the patterns of use and reasons for these changes, as well as indications for analgesic use.

Other covariates (paper I-IV)

Educational level was based on the number of years of education and categorized into ≤ 9 years, 9-12 years and ≥13 years. During the face-to-face interview information was obtained on every occupation lasting for more than one year. The individual's socio-economic status (SES), based on reported occupations, was derived from the official Swedish socio-economic classification scheme [225]. We could, further, assess the "household SES" since we also inquired about occupations among spouses and parents. Alcohol intake was assessed from the mailed questionnaire with separate questions about beer, wine and liquor consumption 10 years before the interview. The average frequency and amount of alcohol was converted to total consumption of pure alcohol in grams per week, and categorized into quartiles, according to the distribution among controls.

STATISTICAL METHODS

To estimate the relationship between risk of chronic renal failure and possible risk factors we used unconditional logistic regression models. We computed odds ratios (OR) and 95 % confidence intervals (CI), as measures of relative risk. First, we estimated ORs in simple age-and sex-adjusted models, next we explored whether confounding occurred. The frequency matching factors (sex and age in 10-year groups) were always kept in the model, other covariates were retained if they were of statistical importance, assessed using the likelihood ratio test [226], or of *a priori* biological importance. Hypertension was not added in the final models since it may not be the cause, but instead the consequence of CRF at this stage. However preliminary analyses including hypertension did not change any risk estimates in this thesis. To test whether effect modification occurred, we conducted both stratified analyses and tested statistically for interactions in the final models through likelihood ratio tests. When trend test were performed, we constructed semi-continuous variables by assigning each category of exposure a score equal to the mean value. Then we fitted the resulting scores into the model and tested for linear trend. Analyses were carried out for CRF overall and for major groups of disease-specific CRF.

Tobacco (paper I)

Never regular smokers of any tobacco type constituted the reference group. In analyses comparing never regular users to former and current regular users, the classification of tobacco use was based on tobacco status 5 years prior to interview. The purpose was to reduce the possibility that symptoms of early CRF influenced tobacco us. To assess dose-effect relations we analysed lifetime average intensity, duration and cumulative quantity. For cigarette smoking we used average number of cigarettes per day, years and pack-years, respectively. The full multivariate model included gender, age, education, alcohol consumption, use of salicylates and paracetamol. Mutual adjustments for cigarette smoking, cigar smoking, pipe smoking, and snuff use were made, when appropriate. No interaction terms needed to be included in the model. We conducted stratified analyses according to gender, underlying renal disease, diabetes status, and ever versus never use of angiotensin-converting enzyme (ACE) inhibitors. In some tables results from men and women are presented separately because the risk estimates differed somewhat in these analyses, however, there were no statistical significant differences in results between genders.

Diet (paper II)

To compute average daily intake of calories and nutrients, frequency data of food items were multiplied by the nutrient composition of foods and beverages based on the National Food Administration handbook [227] and by coefficients corresponding to reported portion sizes (“small, “medium”, or “large”). The latter coefficients, specific for each gender, were derived from a previous validation study using 4-weeks’ food record method. Nutrients were energy-adjusted according to the residual method [228]. The reported mean energy intake among study participants was 2150 kcal/day. Intake derived from supplements was added to the energy-adjusted nutrients. Quintiles of

intake were created based on the distribution of controls. Univariate and multivariate analyses were performed to estimate the risk of CRF in different strata of protein and antioxidant intake. Analyses on protein intake were conducted in all subjects whereas we restricted our analyses of vitamin, vegetables and fruit intake to the non-diabetic stratum. The rationale for this decision was expected reverse causation among diabetics regarding vegetable and fruit intake. In all analyses, the category representing the lowest intake was used as reference. In the final multivariate model we included sex, age, education, highest BMI in lifetime, smoking, alcohol consumption, use of salicylates and paracetamol. Birth-weight and socio-economic status (instead of education) were not included since they did not change the risk estimates, neither was occupational exposures since it was unrelated to CRF risk. Presence of diabetes was not used as a covariate in the regression model, instead we stratified according to diabetes status. The results did not differ between men and women, hence, data for both genders was combined in the final analyses.

Obesity, birth weight and height (paper III)

In the final main analyses, we decided to stratify data by gender, since the ORs differed somewhat between men and women, however, formal test of interaction was not significant. In subanalyses, data from men and women was combined due to otherwise small numbers. Body mass index (BMI=body weight divided by height raised to the second power, kg/m^2) was divided into quartiles according to the distribution among controls, and the lowest quartile of BMI served at the reference in analyses. In addition, BMI was categorised according to WHO´s definition of overweight and obesity (overweight; BMI $\geq$25 kg/m^2, obesity; $\geq$30, severe obesity $\geq$35) [127], where subjects without overweight constituted the reference group. Few subjects at age 20 had a BMI exceeding 30 kg/m^2; subsequently BMI at that age was dichotomised into <25 and $\geq$ 25. Height in centimetres was categorised into quartiles. In addition, to increase the resolution of risk assessments among the shortest study participants, the quartile including the shortest subjects was further divided into tertiles. The quartile with the tallest study participants served as the reference group. Birth weight was kept in the predefined categories, the category including those with a birth weight between 3000 and 3499 grams was the referent. The final multivariate analyses included age, education, smoking, alcohol consumption, and use of paracetamol and aspirin. Mutual adjustments for BMI, birth weight and height did not alter the risk estimates, and were therefore omitted. There was no significant interaction between any of the covariates.

Analgesics (paper IV)

In the final analyses on analgesic use and CRF risk, we included in the model sex, age, smoking, level of education, regular use of other analgesics, and finally an interaction term between paracetamol and aspirin (when applicable). The reference category in analyses of a given analgesic was nonusers of that analgesic. For each analgesic we analysed the independent effect of that analgesic on CRF risk for sporadic use and regular use. We further estimated risks associated with duration, average dose consumed during regular periods of use, and cumulative lifetime dose of each

analgesic. There was no heterogeneity in risks between gender so data for men and women were combined. In an attempt to diminish the risk that symptoms related to renal failure prompted use of analgesics, we conducted lagged analyses disregarding the use of paracetamol and aspirin during the latest 5 and 10 years before interview.

RESULTS

GENERAL RESULTS (PAPERS I-IV)

The female to male ratio for the 926 cases enrolled in the study was 1: 1.8 (Table 2). The median serum creatinine at inclusion was 336 μmol/L for men, and 281 μmol/L for women, while the median predicted creatinine clearance was 22 and 19 mL/min, respectively. A majority of the cases were in the pre-uremic stage; 80 % had a creatinine level below 400 μmol/L, only 6 % had a predicted clearance lower than 10 mL/min. Approximately one third of female and male cases were diagnosed with diabetic nephropathy, somewhat more frequently as a consequence of type II than of type I diabetes (Table 2). The second largest group was patients with glomerulonephritis (28 % of men and 16% of women), followed by renal vascular disease (17 and 12 % of men and women, respectively). The renal diagnoses were based on biopsies in 30 %.

Table 2. Participating case patients with chronic renal failure. Measures of renal function and underlying diagnosis.

	Men (n=597)		Women (n=329)	
	Median	Range	Median	Range
Serum creatinine at inclusion (μmol/L)[1]	336	300-2,475	281	250-1,680
Creatinine clearance (mL/min)[2]	22	2-53	19	3-35
	No of cases	(%)	No of cases	(%)
Diagnosis group				
Diabetic nephropathy	180	(30)	106	(32)
Type I diabetes	75		46	
Type II diabetes	97		54	
Unknown	8		6	
Glomerulonephritis	168	(28)	54	(16)
IgA nephropathy	55		8	
No renal biopsy	40		14	
Unclassified on biopsy	27		15	
Proliferative	18		8	
Focal segmental sclerosis	13		3	
Crescentic glomerulonephritis	8		4	
Other	7		2	
Renal vascular disease	100	(17)	39	(12)
Benign hypertension	91		34	
Malignant hypertension	4		4	
Other	5		1	
Other diagnosis	149	(25)	130	(40)
Hereditary disease	58		40	
Systemic disease or vasculitis	40		42	
Other diagnosis	23		32	
Unknown renal disease	28		16	

[1] Conversion factor for conventional Unit (mg/dL) is 88.4. [2] Predicted creatinine clearance (Cockroft-Gault formula).

Thirteen percent reported that they had not received a diagnosis of renal disease before inclusion in this study. The median duration of known kidney disease among the other cases was 4 years, and the mean duration was 10 years.

The mean age was 58 years for men and 57 years for women both among cases and controls (Table 3). Approximately 40 % of cases and controls were in the highest age group (65-74 years). Compared with controls, both female and male cases lived more often alone and were on average less well educated. Both individual (not shown) and household socioeconomic status, based on reported occupations, was, on average, lower among cases than among controls. The prevalence of self-reported hypertension was high among cases: 87 % of men and 85 % of women, compared with approximately 25 % of male and female controls. Diabetes, present in slightly more than one third of the case patients, was reported by 7 % of the control subjects (both sexes). The proportion of alcohol users was lower among the cases, but the mean consumption was higher among cases (data not shown).

Table 3. Selected characteristics of cases and controls.

	Men		Women	
	Cases n=597 %	Controls n=653 %	Cases n=329 %	Controls n=345 %
Age at interview, years				
18-24	1	2	2	2
25-34	6	5	9	8
35-44	10	9	11	10
45-54	22	18	19	20
55-64	21	21	19	20
65-74	41	45	41	41
Household SES[1]				
professionals	17	23	12	18
intermediate non-manual	20	21	16	23
assistant non-manual	16	16	26	21
skilled manual	28	23	19	15
unskilled manual	11	9	22	13
self-employed	7	8	6	10
Education, years				
≤ 9 years	59	54	57	49
10-12 years	22	23	24	28
>12 years	18	22	18	23
Married or living together	76	81	83	88
Diabetes	35	7	37	7
Hypertension	87	25	85	26

[1] The highest occupational SES group within the subjects family

TOBACCO USE (PAPER I)

Smoking and overall risk of chronic renal failure

The prevalence of "current" smokers 5 years before interview was 31 % of cases and 29 % of controls, whereas 27 % of cases and controls were former regular users at that point (Table 4). Overall, current regular cigarette smoking was not significantly associated with risk of CRF (OR 1.14, 95 % CI 0.89-1.47), neither was former cigarette smoking (OR 1.18, 95 % CI 0.90-1.55). However, the risk increased with average number of cigarettes smoked per day, number of years smoked and number of pack-years. In relation to never regular smokers, smoking more than 20 daily cigarettes was associated with a statistically significant 51 % excess risk of CRF, and a cumulative dose of >30 pack-yeas yielded a 52 % increased risk. The OR for those who had smoked >40 years, in relation no non-smokers, was 1.45 (95 % CI 1.00-2.09).

Table 4. Odds ratios (OR) and 95 percent confidence intervals (95 % CI) for chronic renal failure among smokers [1]

	Cases		Controls		OR [2]	(95 % CI)
	N	(%)	N	(%)		
Ever regular use of cigarettes , cigars or pipe						
No [3]	348	(38)	420	(42)	1.00	(referent)
Yes	567	(61)	574	(58)	1.17	(0.95-1.44)
Regular use of cigarettes						
Former	252	(27)	268	(27)	1.18	(0.90-1.55)
Current	291	(31)	285	(29)	1.14	(0.89-1.47)
Average number of cigarettes/day						
1-10	128	(14)	171	(17)	0.89	(0.66-2.11)
11-20	298	(32)	286	(29)	1.24	(0.96-1.60)
>20	117	(13)	96	(10)	1.51	(1.06-2.15)
Duration of cigarette smoking , years						
1-20	180	(19)	225	(23)	0.99	(0.74-1.31)
21-40	259	(28)	238	(24)	1.23	(0.94-1.60)
>40	104	(11)	90	(9)	1.45	(1.00-2.09)
Number of pack-years, cigarettes						
1-15	202	(22)	263	(26)	0.95	(0.73-1.24)
16-30	207	(22)	184	(18)	1.32	(1.00-1.75)
>30	134	(15)	106	(11)	1.52	(1.08-2.14)

[1] Smoking status 5 years prior to interview.

[2] Adjusted for age, sex, education, alcohol, use of paracetamol and salicylates. Analyses of cigarette smoking also adjusted for pipe smoking, cigar smoking and snuff use.

[3] Never regular smokers of any tobacco type was the reference group for all comparisons.

In analyses stratified by diabetes status we found a 30 % increased risk of borderline statistical significance linked to heavy smoking among non-diabetics (OR=1.3, 95 % CI 1.0-1.9), whereas no similar excess in risk was seen among heavy smoking diabetics

(data not shown). However, the statistical precision was low; only 68 controls had diabetes. Among never users of ACE inhibitors, smoking >20 pack-years was associated with a statistically significant 70 % increased risk of CRF, in relation to non-smokers. This risk increase was not found in the stratum of ever users of ACE inhibitors (OR 0.7, 95 % CI 0.3-2.0), but the statistical power was low. This apparent interaction was not statistically significant. Adjustment for hypertension did not change the risk estimates.

Separated analyses of risk for disease-specific CRF

Among current regular smokers, the strongest overall association was found for CRF classified as nephrosclerosis (OR=1.9, 95 % CI 1.1-3.2, both genders combined), the association being slightly stronger for women (data not shown). This risk increased in a dose-response manner (Table 5), and women who had smoked more than 20 pack-years had an almost 5-fold increased risk of nephrosclerosis. There was a weaker and statistically non-significant association between regular smoking and glomerulo-nephritis, and the association was somewhat stronger and close to significant among former smokers (OR 1.5, 95 % CI 1.0-2.4). A dose-response trend was observed for both men and women (Table 5), but most results were statistically insignificant, except the risk increase associated with a high daily dose (OR for >15 cigarettes/day=1.7, 95 % 1.1-2.6).

Table 5. Odds ratios and 95 percent confidence intervals for chronic renal failure by type of renal disease among cigarette smokers, dose-effect analyses.

		Diabetic nephropathy (n=286)		Glomerulonephritis (n=222)		Nephrosclerosis (n=139)	
		Odds ratio [1,2] (95 % confidence interval)					
Average number/day							
All	1-15	0.8	(0.6-1.2)	1.2	(0.8-1.8)	1.8	(1.1-3.0)
	>15	1.4	(1.0-2.1)	1.7	(1.1-2.6)	1.4	(0.8-2.6)
Men	1-15	0.7	(0.4-1.2)	1.2	(0.7-2.0)	1.6	(0.8-3.2)
	>15	1.1	(0.7-1.9)	1.7	(1.0-2.8)	1.4	(0.7-2.8)
Women	1-15	1.1	(0.6-2.0)	1.1	(0.5-2.4)	2.3	(1.0-5.2)
	>15	2.0	(1.0-4.1)	1.9	(0.8-4.7)	1.5	(0.4-5.5)
Number of pack-years							
All	1-20	0.9	(0.6-1.3)	1.3	(0.9-2.0)	1.3	(0.8-2.3)
	>20	1.4	(0.9-2.1)	1.6	(1.0-2.5)	2.2	(1.3-3.8)
Men	1-20	0.7	(0.4-1.2)	1.3	(0.8-2.1)	1.2	(0.6-2.5)
	>20	1.1	(0.6-1.8)	1.6	(0.9-2.7)	1.8	(0.9-3.5)
Women	1-20	1.1	(1.0-4.5)	1.2	(0.6-2.6)	1.4	(0.5-3.4)
	>20	2.1	(1.0-4.5)	1.7	(0.6-4.5)	4.9	(1.8-13.5)

[1] Never regular smokers of any tobacco type was the reference group for all comparisons.

[2] Adjusted for sex (when appropriate), age, education, alcohol, use of paracetamol and salicylates, pipe smoking, cigar smoking and snuff use.

Overall, there was no statistical link between regular current or former smoking and diabetic nephropathy, however a nearly significant risk increase was observed among currently smoking women (OR=1.6, 95 % CI 0.9-3.0), but not among men (OR =0.8, 95 % CI 0.4-1.3). Heavy smoking among women (>20 pack-years), was associated with a more than doubled risk of diabetic nephropathy, in relation to non-smokers (Table 5). Hereditary disease was unrelated to smoking whereas there was a significant *inverse* relation with CRF caused by systemic disease/vasculitis, but this latter association did not exhibit any consistent dose-effect trend (Table 6).

Table 6. Odds ratios and 95 percent confidence intervals for chronic renal failure by type of renal disease among cigarette smokers, dose-effect analyses.

		Hereditary disease (n=98)		Systemic disease/ vasculitis (n=82)		Other (n=99)	
		Odds ratio [1,2] (95 % confidence interval)					
Average number/day							
All	1-15	1.3	(0.8-2.3)	0.5	(0.2-0.9)	1.2	(0.7-2.0)
	>15	1.0	(0.5-1.9)	0.5	(0.2-1.0)	0.8	(0.4-1.5)
Men	1-15	1.3	(0.6-2.7)	0.6	(0.2-1.7)	0.4	(0.2-1.0)
	>15	1.3	(0.6-3.0)	0.4	(0.1-1.2)	0.4	(0.2-1.1)
Women	1-15	1.4	(0.6-3.1)	0.3	(0.1-0.9)	2.4	(1.2-5.0)
	>15	0.6	(0.2-2.5)	0.6	(0.2-1.8)	0.8	(0.2-2.6)
Number of pack-years							
All	1-20	1.4	(0.8-2.3)	0.3	(0.2-0.7)	1.1	(0.6-1.9)
	>20	0.9	(0.4-1.8)	0.7	(0.4-1.5)	0.9	(0.5-1.7)
Men	1-20	1.3	(0.6-2.7)	0.5	(0.2-1.3)	0.4	(0.2-1.0)
	>20	1.3	(0.5-3.0)	0.6	(0.2-1.7)	0.4	(0.2-1.1)
Women	1-20	1.5	(0.7-3.3)	0.1	(0.0-0-5)	2.2	(1.0-4.8)
	>20	0.5	(0.1-2.2)	1.2	(0.4-3.5)	1.2	(0.4-3.3)

[1] Never regular smokers of any tobacco type was the reference group for all comparisons.

[2] Adjusted for sex (when appropriate), age, education, alcohol, use of paracetamol and salicylates, pipe smoking, cigar smoking and snuff use.

Other type of tobacco use and risk of chronic renal failure

Ever regular pipe smoking was reported by 21 % of male cases and 20 % of male controls. Snuff was used by 17 % and 18 % of male cases and controls, respectively. Very few women reported any regular pipe or cigar smoking, or use of snuff. After adjustment for other tobacco products, no significant associations were found between CRF (overall or by subtype) and regular pipe smoking, cigar smoking, or use of oral snuff. The OR among ever regular pipe smokers relative to never-smokers of any tobacco was 1.0 (95 % CI 0.5-2.1), among cigar smokers 0.7 (95 % CI 0.2-2.4), and among regular users of snuff 0.8 (95 % CI 0.6-1.1). These findings pertained to all measures of dose-effect relations (data not shown).

PROTEIN INTAKE (PAPER II)

We found a weak, statistically non-significant positive association between intake of protein 10 years prior to interview and CRF overall (Table 7). The adjusted OR was 1.4 (95 % CI 1.0-1.9) among those in the highest quintile of protein intake compared with those in the lowest. In stratified analyses according to underlying major subtypes of CRF, the elevated risk only pertained to diabetic nephropathy, where a strong dose-dependent association was seen. Subjects in the highest quintile of intake had a six-fold increased risk of CRF compared with those in the lowest quintile. Few cases had been recommended a protein-reduced diet due to CRF, and excluding those from analyses did not change any estimates. When we restricted the analysis to cases and controls with diabetes, the increased risk with a high protein intake remained, although the OR was lower and did not quite attain statistical significance (OR=2.9, 95 % CI 1.0-8.2).

Table 7. Odds ratios and 95 % confidence interval for chronic renal failure, overall and by subtype, associated with energy-adjusted dietary protein intake 10 years prior to interview.

	All	Diabetic nephropathy	Renal vascular disease	Glomerulo-nephritis	Other
	Odds ratio[1] (95 % confidence interval)				
Protein intake (g/day)					
<73.2	1.0 (referent)	1.0 (referent)	1.0 (referent)	1.0 (referent)	1.0 (referent)
73.2-80.8	1.2 (0.9-1.6)	2.4 (1.3-4.5)	1.4 (0.8-2.5)	1.6 (1.0-2.6)	0.7 (0.4-1.0)
80.9-86.6	1.0 (0.7-1.4)	2.7 (1.4-5.1)	1.2 (0.6-2.2)	0.9 (0.5-1.5)	0.7 (0.4-1.0)
86.7-93.4	1.2 (0.9-1.6)	4.4 (2.3-7.5)	1.1 (0.6-2.0)	1.0 (0.6-1.6)	0.8 (0.5-1.2)
>93.4	1.4 (1.0-1.9)	6.0 (3.4-10.7)	0.7 (0.4-1.4)	1.5 (0.9-2.4)	0.7 (0.5-1.1)

[1]Adjusted for age, sex, education, smoking, alcohol, highest BMI in lifetime, use of paracetamol and salicylates, and energy intake

ANTIOXIDANT INTAKE (PAPER II)

There were no important differences in risk estimates for any of the disease-specific types of CRF (renal vascular disease, glomerulonephritis and "other renal diseases"), therefore results are presented combined. Diabetics were excluded from our analyses. A high weekly intake of vegetables was not statistically significantly associated with risk of CRF, the OR was 0.7 (95% CI 0.5-1.1) among those in the highest quintile of intake in relation to those in the lowest. Unexpectedly, a high intake of fruits was positively associated with CRF risk (OR =1.5 for those in the highest quintile compared with the lowest), but the association was statistically non-significant and showed no clear dose-effect trend. We did not find any clear associations between intake of vitamin C, folic acid, vitamin A or betacarotene and risk of CRF (data not shown). There was a tendency of an inverse association between intake of vitamin E and CRF risk, the OR was 0.7 (95 % CI 0.5-1.0) for subjects with a vitamin E intake corresponding to the fourth and fifth highest quintiles compared with those in the lowest quintile.

We conducted analyses with stratification for various factors that, according to the previous literature, are allegedly associated with an increased oxidative stress, such as smoking, obesity and hypertension. Among hypertensive subjects, an intake of vitamin E corresponding to the fourth or fifth highest quintile in relation to the lowest, at least halved the CRF risk, although no clear dose-response trend emerged (Table 8). No such reduction in risk was seen among individuals without hypertension. The p-value for a formal test of interaction was 0.05. Our data did not reveal other significant effect modification of any factor on the association between antioxidant intake and risk of CRF (data not shown).

Table 8. Odds ratios (OR) and 95 % confidence intervals (CI) among non-diabetics for risk of chronic renal failure associated with intake of vitamin E from diet and supplements 10 years prior to interview. Analyses stratified by hypertension status.

	Hypertension	No hypertension
	OR [1] (95 % CI)	OR [1] (95 % CI)
Vitamin E (mg/day)		
<4.54	1.0 (referent)	1.0 (referent)
4.54-5.16	0.6 (0.4-1.1)	1.2 (0.6-2.5)
5.17-5.82	1.0 (0.6-1.8)	1.3 (0.6-2.8)
5.83-6.79	0.4 (0.2-0.8)	1.3 (0.6-3.0)
>6.79	0.5 (0.4-0.9)	1.3 (0.6-3.0)

[1] Adjusted for age, sex, education, smoking, alcohol, highest BMI in lifetime, use of paracetamol and salicylates, and energy intake.

BODY MASS, HEIGHT AND BIRTH WEIGHT (PAPER III)

Obesity and overall risk of chronic renal failure

We observed a positive association between BMI based on highest reported weight in lifetime and overall risk of CRF. When study participants were categorized using WHO´s definitions of obesity and overweight, we found relatively strong dose-dependent associations across the range of BMI categories in both sexes (Table 9). Women and men with a BMI ≥35 kg/m^2 had 3 and 4-fold increases in risks, respectively, compared with those without overweight. When using quartiles as cut points, significant risk elevations were only found among men, those in the highest quartile of BMI had a 2.3-fold increased risk of CRF overall, compared with those in the lowest (data not shown). Men and women having a BMI of 25 kg/m^2 or more at age 20, had statistically significant 3-fold increased risks of CRF compared to subjects with a BMI below 25 at that age (Table 9). Weaker positive associations were observed between BMI at time of interview and CRF risk, at least for men (OR 1.9), but these associations did not reach statistical significance (data not shown).

Table 9. Odds ratios (OR) for chronic renal failure associated with body mass index (BMI). Cutpoints in accordance with WHO's definition of overweight and obesity.

	Men		Women	
	No of cases/controls	OR[1] (95% CI)	No of cases/controls	OR[1] (95% CI)
Highest BMI in lifetime, (kg/m^2)				
<25.00	129 / 213	1.0 (referent)	96 / 136	1.0 (referent)
25.00-29.9	265 / 323	1.4 (1.0-1.9)	115 / 133	1.2 (0.8-1.8)
30.0-34.9	130 / 79	2.7 (1.9-4.0)	49 / 46	1.4 (0.8-2.4)
≥35.00	56 / 18	4.4 (2.4-8.2)	48 / 17	3.1 (1.6-6.1)
BMI at age 20 (kg/m^2)				
<25.0	377 / 506	1.0 (referent)	211 / 274	1.0 (referent)
≥25.0	92 / 40	3.1 (2.1-4.8)	30 / 13	3.0 (1.4-6.1)

[1] Adjusted for age, education, smoking, alcohol, and use of paracetamol and salicylates

Stratified analyses by hypertension and diabetes status

In analyses restricted to non-diabetics and/or non-hypertensives, we found that subjects with a highest lifetime BMI of 35 kg/m^2 or more had higher risk of CRF even though they did not have clinically overt diabetes (OR 2.2, 95 % CI 1.3-3.8), or hypertension (OR 2.8, 95 % 1.0-8.1) (Table 10). In analyses confined to subjects with neither diabetes, nor hypertension, the point estimate was similar, but due to small numbers no statistical significance was obtained. Subjects without diabetes and hypertension who reported overweight at age 20 had a statistically significant 3-fold increased risk, compared with those with normal weight at that age.

Table 10. Odds ratios (OR) for chronic renal failure associated with body mass index (BMI). Cutpoints in accordance with WHO's definition of overweight and obesity. Analyses restricted to subjects without self-reported diabetes and/or hypertension.

	No diabetes		No hypertension		No diabetes or hypertension	
	OR[1]	(95% CI)	OR[1]	(95% CI)	OR[1]	(95% CI)
Highest BMI in lifetime (kg/m^2)						
<25	1.0	(referent)	1.0	(referent)	1.0	(referent
25-29.9	1.3	(1.0-1.7)	1.3	(0.8-2.0)	1.1	(0.6-1.8)
30-34.9	2.0	(1.4-2.8)	1.8	(1.0-3.5)	1.2	(0.5-2.6)
≥35.0	2.2	(1.3-3.8)	2.8	(1.0-8.1)	2.1	(0.6-7.6)
BMI at age 20 (kg/m^2)						
<25.00	1.0	(referent)	1.0	(referent)	1.0	(referent)
≥25.00	2.4	(1.6-3.6)	3.6	(1.8-7.1)	3.0	(1.4-6.4)

[1] Adjusted for age, sex, education, smoking, alcohol, use of paracetamol and salicylates.

Separated analyses of risk for disease-specific CRF

In analyses of risks for disease-specific CRF, the highest risk was found for diabetic nephropathy. OR was 2.8 (95 % CI 1.8-4.4) for those with a BMI ≥30 kg/m^2 compared with BMI<25. The risk increase pertained only to diabetic nephropathy associated with type II diabetes. Those with severe obesity (BMI>35) had a more than 17-fold excess risk of type II diabetes-related nephropathy (data not shown). However, elevations in risk were also seen for all other types of nephropathies. In relation to subjects with a normal weight, those with a BMI of 30 or more had a statistically significant 2.4-fold increased risk of CRF due to nephrosclerosis, and the risk of CRF due to glomerulonephritis was doubled. Likewise, elevated BMI at age 20 yielded increases in risk for all major types of CRF (data not show).

Height and birth weight

We found a 90 % increased risk of CRF overall among the shortest men (<169 cm) compared with the tallest (Table 11). A corresponding 50 % excess in risk among the shortest women (<158 cm) was not statistically significant. The association was strongest for glomerulonephritis (OR=2.8, 95% CI 1.6-5.9 for men and women combined), and diabetic nephropathy (OR=2.0, 95 % CI 1.1-3.4), while the 60 % excess in risk for nephrosclerosis was statistically non-significant.

Table 11. Odds ratios (OR) for chronic renal failure associated with height and birthweight.

	Men OR[1] (95% CI)	Women OR[1] (95% CI)
Height, (cm)		
Sex-specific quartiles[2, 3]		
Q4 (highest quartile)	1.0 (referent)	1.0 (referent)
Q3	1.1 (0.8-1.5)	0.8 (0.5-1.4)
Q2	1.1 (0.8-1.5)	1.0 (0.6-1.6)
Q1 a (lowest quartile, highest subtertile)	1.2 (0.7-2.0)	0.7 (0.4-1.4)
b	1.2 (0.7-1.8)	1.3 (0.7-2.6)
c (lowest quartile, lowest subtertile)	1.9 (1.2-3.0)	1.5 (0.8-2.7)
Birth weight, (gram)		
<2500	0.9 (0.4-2.0)	0.8 (0.3-2.1)
2500-2999	1.4 (0.9-2.1)	1.1 (0.6-1.9)
3000-3499	1.0 (referent	1.0 (referent)
3500-3999	1.1 (0.8-1.5.)	1.0 (0.6-1.7)
≥4000	1.4 (0.9-2.2)	0.8 (0.4-1.4)

[1] Adjusted for age, sex, education, smoking, alcohol, and use of paracetamol and salicylates.

[2] Q4: men 183-204 ; women: 168-186; Q3: men: 178-182; women: 165-167; Q2: men: 173-177; women 161-164; Q1a: men: 172; women: 160; Q1b: men: 169-171; women: 158-159; Q1c: men: 154-168; women: 144-157

[3] Quartile number one divided in sub-tertiles.

A relatively large number of study participants did not recall their birth weight (236 cases and 244 controls). Few study participants reported a low birth weight, only 30 cases and 33 controls had a birth weight below 2500 kg. After adjustment for potential

confounders the analyses did not reveal any significant associations between self-reported birth weight and CRF risk (Table 11). The OR for women with a birth weight of less than 2500 g compared with those with a birth weight of 3000-3499 g, was 0.8 (95 % 0.3-2.1), the corresponding OR for men was 0.9 (95% CI 0.4-2.0). In further analyses, we tested whether LBW could effect modify the association between obesity and CRF, but this could not be confirmed (data not show).

ANALGESIC USE (PAPER IV)

Aspirin was the most commonly used non-narcotic analgesic; 37% of the cases and 19% of the controls reported periods of regular use. Paracetamol was used regularly by 25% of cases and by 12% of controls. Regular use of other analgesics, such as dextropropoxyphene, NSAIDs and pyrazolones was also more frequent in cases than in controls. Only 17 cases and 17 controls reported ever use of phenacetin, and 4 cases and 6 controls had used this analgesic substance regularly.

Isolated effects of paracetamol and aspirin

Among subjects who did not use aspirin regularly, the regular use of paracetamol was associated with a 2.5-fold increased risk of CRF (Table 12). A clear dose-effect trend with lifetime cumulative dose emerged (p for trend<0.001). A high average daily dose of paracetamol was strongly linked to risk of CRF. OR among subjects who took ≥ 1.4 g/day during periods of regular use was 5.3 (95 % 1.8-15.1), relative to non-users.

Table 12. Odds ratios for chronic renal failure associated with the lifetime use of either paracetamol or aspirin among subjects who did not use the other analgesic regularly.[1]

	Paracetamol use OR (95% CI)	Aspirin use OR (95% CI)
Never used	1.0	1.0
Ever used	1.3 (1.0-1.6)	1.5 (1.2-1.8)
Use or used regularly	2.5 (1.7-3.6)	2.5 (1.9-3.3)
Cumulative dose		
1-99 g	1.2 (0.9-1.5)	1.4 (1.1-1.7)
100-499 g	1.3 (0.9-1.8)	1.6 (1.2-2.1)
≥500 g	3.3 (2.0-5.5)	1.9 (1.3-2.9)

[1] The odds ratios are adjusted for age, sex, level of education, smoking status, use of other analgesics, and the interaction between aspirin use and paracetamol use. P<0.001 for the trend toward greater risk with increasing cumulative doses of paracetamol; P=0.01 for the trend toward greater risk with increasing doses of aspirin. Regular use was defined as the use of at least two tablets per week for a period of two months or longer.

Likewise, among subjects who did not use paracetamol regularly, the regular use of aspirin was independently and significantly linked to CRF risk. The cumulative dose showed a dose-dependent pattern of association, although the trend was slightly weaker than that for paracetamol use (Table 12). In addition, those who had taken ≥ 1.4 g/day on average during periods of regular use had a more than 3-fold increased risk (OR 3.3, 95 % CI 1.4-8.0). The duration of paracetamol or aspirin use was not related to CRF risk (data not shown).

Paracetamol, aspirin and disease-specific CRF risks

The odds ratios for disease-specific types of CRF associated with isolated regular use of paracetamol varied between 1.6 and 3.6, but statistical significance was only attained for renal failure classified as diabetic nephropathy and CRF associated with systemic disease/vasculitis (Table 13). However, except for CRF due to renal vascular disease and glomerulonephritis, all dose-related trends in risk were statistically significant. Regular use of aspirin in the absence of regular use of paracetamol was associated with statistically significant 2-fold or higher increases in risk for all types of CRF, except for renal disease caused by systemic disease/vasculitis. In contrast, dose-risk trends were only significant for glomerulonephritis.

Table 13. Odds ratios for chronic renal failure associated with isolated regular use of paracetamol or aspirin according to the type of underlying renal disease [1]

	Paracetamol use			Aspirin use		
	n	OR[2]	(95% CI)	n	OR[3]	(95% CI)
Diabetic nephropathy	42	3.6	(2.1-6.0)	68	2.9	(1.9-4.5)
Glomerulonephritis	17	1.6	(0.9-3.0)	57	2.6	(1.4-4.8)
Nephrosclerosis	12	1.7	(0.8-3.7)	39	2.1	(1.3-3.5)
Hereditary renal disease	6	2.2	(0.8-5.9)	19	3.1	(1.6-6.0)
Systemic disease / Vasculitis	15	2.8	(1.2-6.5)	8	1.1	(0.4-2.8)
Other renal disease	13	2.1	(0.9-4.6)	22	3.7	(1.8-7.7)

[1] Adjustments were made for age, sex, level of education, smoking status, use of other analgesics, and the interaction between aspirin use and paracetamol use. Regular use was defined as the use of at least two tablets per week for a period of two months or longer.

[2] The reference group was nonusers of paracetamol without regular aspirin use.

[3] The reference group was nonusers of aspirin without regular paracetamol use.

Combined effects of paracetamol and aspirin

In analyses comparing regular users of both paracetamol and aspirin with regular users of *only aspirin*, we found an increased CRF risk among users of both analgesics (OR=2.2, 95 % CI 1.4-3.5). The risk increased significantly with increasing cumulative lifetime dose of paracetamol (p for trend=0.03). The effect of combined use of paracetamol and aspirin, relative to regular use of *only paracetamol*, did not reach statistical significance (OR 1.6, 95 % CI 0.9-2.7). However, the trend toward greater risk with increasing cumulative lifetime dose of aspirin was significant (p=0.02).

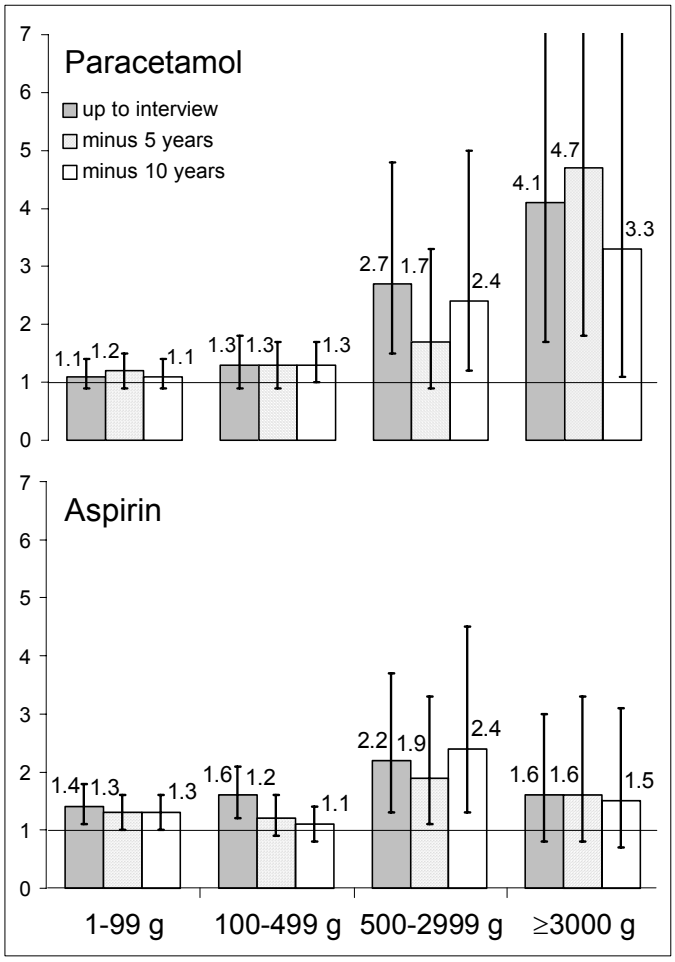

Figure 1. Odds ratios and 95% CI for chronic renal failure according to the cumulative lifetime dose of paracetamol or aspirin in analyses based on different periods of exposure. The odds ratios are for comparison with nonusers of both classes of analgesics

Isolated effects of paracetamol and aspirin, disregarding recent exposure

A larger proportion of cases than controls started to use analgesics regularly within five years prior to interview, 35 % versus 25 % for paracetamol, and 37 % versus 28 % for aspirin. In an attempt to avoid reverse causation we disregarded the 5 and 10 most recent years of exposure in analyses, but the odds ratios were only slightly attenuated (Figure 1).

Other types of analgesics

No elevated risks for CRF were seen in association with use of other types of analgesics than paracetamol and aspirin (data not shown).

DISCUSSION

METHODOLOGICAL CONSIDERATIONS

Choice of study design

Intervention studies, or clinical trials, where the exposure or the treatment is randomly allocated to the participants by the investigators, preferably in a double-blind fashion, yield the most valid effect estimates owing to the inherent control of known and unsuspected confounding. However, due to issues of cost, feasibility and ethics, other types of studies than intervention studies are needed in many circumstances. In the case of disease aetiology epidemiology, where the effects of supposedly hazardous risk factors are evaluated, intervention studies are typically not an option. If cohort and case-control studies are properly designed and well conducted, and if data are correctly analysed and interpreted, such observational studies may contribute significantly to our understanding of the aetiology of many diseases.

Case-control studies are better suited for studying rare diseases than are cohort studies. Notwithstanding the increasing trend, ESRD is still a relatively rare condition. The incidence of RRT is 125 per million person-years in Sweden [3]. The incidence of CRF, as defined in our study, was not exactly known during our planning phase. However, we did not expect the incidence to be very high and concluded that the most efficient way to study risk factors for moderate CRF would be a case-control approach.

The accuracy of epidemiological studies depends on the validity and precision. Lack of validity (systemic errors) may follow from selection bias, misclassification of exposure and/or outcome, and confounding. The main determinants of precision are sample size, exposure prevalence, and outcome frequency. Lack of precision causes random errors.

Validity

Selection bias

Selection bias might occur if case and control subjects do not represent the very same study base. Fortunately, the prerequisites in Sweden, with a geographically well defined population, the ubiquitous use of National Registration Numbers that are assigned to every resident, a very efficient population administration that holds a complete and continuously updated computerized population register, combined with an entirely public and homogenous health care, make case ascertainment and truly random sampling of controls from a well-defined study base unusually simple. Selection bias may also occur whenever the inclusion of cases or controls into the study depends in any other way on the exposure of interest. The population-based design and the fortunate circumstances cited above seem to insure that the identified cases and selected controls adequately represent the study base, without any biased selection linked to the studied exposures.

In an attempt to identify all incident cases within the study base we set up a comprehensive study organisation involving all nephrology departments and other medical departments treating patients with kidney diseases, as well as essentially all medical laboratories in Sweden. Since the ascertainment of cases was based on serum creatinine tests in routine clinical practice, undertaken because of various symptoms and not as screening, it is important that all symptomatic individuals in the study base, regardless of their exposure to the studied risk factors and confounding factors, had the same chance of having a serum creatinine test performed. Although it is conceivable that that people with co-morbidities were more likely to be tested than were those without, the creatinine cutpoint in our case definition was deliberately placed at a level where symptoms were expected to almost invariably prompt medical consultations. And since Sweden has a public health care system with equal access for everybody, and the costs are virtually completely covered by a mandatory social insurance, we do not believe that social or other lifestyle-related factors have importantly affected the probability of creatinine testing. During our study period we identified 1, 189 eligible cases, which corresponds to an incidence of 115 patients per million person-years. This is somewhat lower than the incidence rate of patients starting RRT in Sweden. This might indicate that we did not manage to identify all eligible cases. However, it must be noted that we did not include patients over 74 years and those who not were born in Sweden. In addition, patients with pre-renal or post-renal disease were not eligible.

Our case definition was carefully chosen. If the cut point of serum creatinine would have been placed to far into the course of CRF, then it might have led to losses of cases caused by rapid deterioration or death, potentially related to the exposure under investigation. On the other hand, if the cut point was placed too early, when the disease had not yet manifested clinically, there is a risk of ascertainment (detection) bias. With our case definition, we do not believe that there is a large pool of unidentified eligible cases in the population. This reasoning will, in the next section, be exemplified by the importance of an appropriate case definition in paper I (tobacco exposure), but the reasoning is valid also for the other papers. It is known that cigarette smoking increases risk of death among patients with advanced CRF [229], thus, a higher cut-point of CRF would conceivably result in a selective loss of CRF patients who were smokers, which would result in an underestimation of the true association. Contrary, an overestimation of the link between smoking and CRF would presumably be the case if the creatinine level was set to low, then smoking-related morbidity would probably increase the likelihood of serum creatinine testing and hence CRF detection. Hence, our choice reflects our balancing to avoid the above-mentioned risks.

Non-participation may be another potential source of selection bias if it is linked to status of exposure under investigation. We cannot exclude the possibility that subjects who refused to take part in the study or whose poor condition prohibited participation, may have had other lifestyle habits than those who participated. This might have biased our results in any direction. However, we managed to obtain a reasonably high participation rate, and the difference in this rate between cases and controls was relatively small (75 % of cases and 78 % of controls).

Misclassification of exposure

The major threat to the validity of case-control studies is recall bias, i.e., differential misclassification of exposure due to disparate reporting by cases and controls. Recall bias might distort the true association in any direction, but it is likely to arise only when the disease in itself increases the likelihood of over- or underestimation of the exposure, or when there are preconceptions – among interviewees or interviewers – about the association between the exposure and the disease. A generally poor recollection of the exposure, similar in cases and controls, results in non-differential misclassification, which tends to bias any association towards the null.

The fact that our study hypotheses were unknown to the public and that the interview covered a multitude of exposures, not only the exposures of particular interest, probably reduced the risk of recall bias. In addition, self-reported information on tobacco habits and on anthropometric measures are previously reported to be relatively accurate [230-232]. However, there is a systemic tendency for overweight subjects to underestimate their body size, and conversely, an over-estimation among very lean subjects [233]. This might have biased our results in paper III. If this misclassification was non-differential between cases and controls, it would bias the results towards null. Recall of remote diet is inevitably associated with some misclassification, however it appears that diet as far back as 10 years may be recalled with an acceptable level of misclassification [228]. In the study of CRF and analgesic use, we used photographs of products and their packaging, which is known to improve the accuracy of the recall of drug intake [234]. However, like in all case-control studies, despite our efforts to avoid misclassification of exposure, we cannot rule out that non-differential or possibly differential recall of exposure in cases and controls may have had some impact on the results.

Another source of information bias that may lead to misclassification of exposures comes from erroneous collection or interpretation of responses by interviewers. Although we not were able to blind the interviewers to the case/control status of the subjects, they were trained to follow the questionnaire carefully in a standardised manner, and they were unaware of the study hypotheses. Therefore, this type of bias is not likely in our study

Misclassification of outcome

We did not estimate serum creatinine levels among control subjects, hence, we cannot completely exclude the presence of control individuals with a moderate renal insufficiency. If this potential misclassification of case/control status is unrelated to the studied exposures, then the observed exposure-CRF associations may have been biased towards the null. However, because asymptomatic pre-uraemic CRF of the magnitude required for being classified as a case is likely to be exceedingly rare (see above), it is not likely that an appreciable number of controls were, in fact, misclassified cases.

Confounding

In non-experimental and non-randomised studies confounding may threaten the validity. Confounding is a mixing of the effect of a main exposure on the disease with that of another factor. This factor must be associated with the main exposure and, independently of this exposure, be a risk factor of the disease. Further, the confounding factor must not be a link in the causal chain between the studied exposure and the outcome disease. To avoid confounding effects from sex and age we frequency-matched cases and controls with regard to sex and age (in 10-year strata). We obtained

detailed information on known or suspected risk factors for CRF and in multivariate analyses we adjusted for these factors. However, if the confounding factors are measured too crudely, there may still be room for some residual confounding. Moreover, potentially there are not yet identified risk factors for CRF that could be related to exposures under investigation in this work, thus, we cannot entirely rule out some impact of confounding.

In all observational studies when assessing effects of drug intake on disease outcome, the potential impact of confounding by indication must be considered. This is a bias that arises when the indication for a drug is related to the risk of a future health outcome of interest. This type of confounding was of major concern in paper IV. Many of our cases had underlying diseases that are known to increase risk of CRF; some of these diseases might also prompt the use of analgesics. However, the associations between the use of analgesics and the risk of CRF were not consistently stronger among subjects with predisposing diseases typically causing frequent aches and pains than among subjects without such conditions (e.g. glomerulonephritis).

Our retrospective design may also include difficulties in establishing the temporal relationship between exposure and outcome. Early signs of CRF might have introduced use of analgesics, thus, the positive association observed would in that case be explained by reverse causality. In an attempt to disentangle whether analgesic use increased risk of CRF or if symptoms related to CRF triggered analgesic use, we performed lagged time analyses. In these analyses we disregarded the use of analgesic during the 5 and 10 years before the interview. This resulted only in minor reduction of the risk estimates, which indicates that the use of analgesics was unlikely to be commonly prompted by symptoms from CRF. It is unlikely that many cases had symptoms from their CRF 10 years prior to interview, but in contrast, the underlying conditions might have been present for decades, and they may have been associated with aches and pain that triggered analgesic use. Thus, in the lagged analyses we could not satisfactorily evaluate the potential impact of confounding by indication, and this type of bias cannot completely be ruled out in paper IV.

The influence of reverse causation must be considered in all three other papers. We cannot exclude the possibility that cases had been counselled to stop smoking or that they had quitted due to symptoms from the renal failure (paper I). Indeed, we noticed that more cases than controls had stopped smoking during the last 5 years prior to interview. To diminish the possible impact of reverse causation we did not consider smoking data at time of interview but analysed data based on smoking status 5 years earlier. In paper II, to reduce the risk that symptoms from CRF and/or professional advice had led to changes in dietary habits, we chose to inquire about dietary habits 10 years prior to interview. Excluding subjects who reported that they had changed their diet at any point (i.e. to protein reduced, diabetic or vegetarian diet) did not alter the relative risk estimates. In the analyses of CRF and antioxidant intake we decided to restrict our analyses to non-diabetic subjects since diabetic patients with advanced nephropathy are likely to have had diabetes for many years, quite often more than 10 years. A switch to a diabetic diet presumably results in a higher intake of fruits and vegetables, thus increasing the probability of reversed causation. Whereas there was a positive association between highest BMI in lifetime and CRF (paper III), current BMI

was not significantly related to CRF. This suggests that the renal failure had led to some weight loss.

Precision

Chance must always be considered as an alternative explanation of observed associations, or of lack of associations. In our study we used 95 % confidence intervals to provide information about the precision of our results. The relatively large number of cases yielded generally good statistical power to detect moderately strong associations in the main analyses. Given the observed dose-response, chance alone is an unlikely explanation of the positive associations seen between smoking, obesity or analgesic intake and CRF (paper I, III, IV). However, it must be kept in mind that a large-scale case-control study that investigates many exposures in relation to several outcome diseases (disease-specific types of CRF) simultaneously gives rise to multiple significance testing, which inflates the risk of type I error. Moreover, we had too low statistical power to detect a weak- to moderately strong association between low birth weight and CRF, hence the risk of type II error was not negligible. We performed numerous subgroup analyses, which might increase the risk for type I error, while simultaneously being associated with considerable risks for type II errors due to small numbers.

FINDINGS AND IMPLICATIONS

Tobacco (paper I)

We observed a moderately increased, dose-dependent risk for CRF among cigarette smokers, not explained by age, gender, body mass, hypertension, education, or use of analgesics or alcohol. Increases in risk seemed to be limited to nephrosclerosis and glomerulonephritis in both genders, and to diabetic nephropathy in women. However, due to small numbers in the subgroups analyses, there was a lack of precision in analyses on smoking and other types of CRF. Our finding confirms the general impression of smoking as a renal risk factor [71].

Our results suggest that heavy cigarette smoking increases the CRF risk at least as much for women as for men. Some previous studies have reported that the risk of renal damage from smoking appears to be restricted to men [83, 84, 90, 147] which has prompted speculations about a biological background for the gender difference [235]. This difference is not implausible, because men are generally more likely to progress to ESRD [28]. However, most of these studies were limited by small samples of women, and in some of the studies the smoking exposure was very low among women. In line with our results, a recent study pointed to the same renal risk from smoking among women as among men [236].

Perhaps not surprising, we obtained the strongest association between smoking and nephrosclerosis. Previous studies have suggested that smoking might increase risk of

nephropathy among hypertensive subjects [78, 79]. Of even more importance is the fact that the diagnosis group "nephrosclerosis" most likely includes individuals with atherosclerotic disease in the small and large vessels of the kidneys, and not only subjects with hypertensive nephrosclerosis [51], as previously discussed in this thesis. As smoking is a well-known risk factor for atherosclerosis related to cardiovascular disease, it is not surprising that it also increases risk for atherosclerotic changes in the renal arteries [237]. However, autopsy studies have revealed that smoking also appears to be associated with *intrarenal* damage of small vessels, both in individuals with [238] and without pre-existing kidney disease [239, 240]. Thus, it is likely that smoking-induced atherosclerotic processes related to the kidney may both initiate kidney damage and contribute to the progression rate in all types of renal diseases. Several mechanisms may be involved in inducing damage of renal microvasculature, cigarette smoking causes direct endothelial injury [241] and increases smooth muscle cell proliferation [242]. Other suggested mechanisms for smoking-induced renal injuries include adverse effects from elevated oxidative stress and increased release of cytokines [243], and negative impact from an increased activity of the sympathetic nerve system [244].

Our findings are not consistent with an adverse effect on the kidneys from use of oral snuff, pipe smoking or cigar smoking. These results must be interpreted cautiously because our power to detect any dose-response increases in risk was limited. There is no obvious explanation for the discrepancy in effects of cigarette smoking on the one hand and pipe and cigar smoking on the other hand, the most likely explanation is chance. Compared to cigarette smoking, use of smokeless tobacco causes more prolonged sustained increases of nicotine levels [245]. Although such use has been associated with an adverse cardiovascular risk profile (i.e. negative effects on lipid profiles, increased oxidative stress and increased insulin resistance), it appears that the cardiovascular risk is lower for users of smokeless tobacco than for cigarette smokers [245] if elevated at all [70, 246]. The mechanisms behind this discrepancy in cardiovascular risk are not completely understood. By analogy, it is reasonable to assume that the atherosclerotic changes observed in kidneys of smokers might be less pronounced among users of snuff.

Protein intake (paper II)

Recently, a cohort study was unable to find any link between a high protein intake and reduction of GFR in women without renal failure, whereas such a reduction was observed at follow-up in women with a mild renal failure at baseline [106]. In our study we could not disentangle whether a high protein intake would initiate kidney disease or increase the rate of progression. However, we can conclude that a high protein intake, within the range observed in our Swedish population, was not a risk factor for development of incident moderately severe CRF of non-diabetic type.

In contrast, we found a positive strong association between protein intake and diabetic nephropathy. This finding is in line with results from other studies. In a cross-sectional study of type I diabetics, a protein intake exceeding 20% of total food energy intake was associated with an increased risk of proteinuria [109]. Another study yielded a positive relation between a high protein intake and microalbuminuria, but only among

those who also had hypertension [108]. However, our positive link between a high intake of protein and diabetic nephropathy might, at least partly, be the result of recall bias and particularly reverse causation. We observed that the median protein intake was higher not only among diabetic cases, but also among diabetic controls, compared with non-diabetics. This might have been the result of diet modifications due to diabetes. Although Swedish diabetics without nephropathy are recommended a similar protein intake as individuals without diabetes (10-15 % of total energy intake), a too zealous reduction of fat and carbohydrate consumption might have led to a proportionate increase in protein intake. However, in an analysis confined to diabetic cases and controls, an almost 3-fold risk gradient with protein intake remained, albeit imprecise due to a small number of diabetic controls. Some reservation must be raised for the possibility that case patients may have had diabetes longer than the diabetic controls, and that some of the latter may not have been on diabetes diet 10 years ago. It is biological plausible that a high protein intake might be more harmful in diabetics than in non-diabetics. Both type I and type II diabetes are conditions known to exhibit an increased glomerular filtration rate, caused by increases in plasma flow and glomerular capillary pressure [247]. Subsequently, the extra burden of hyperfiltration related to excessive protein intake [15] might be particularly deleterious in diabetes.

The protein intake in our population was modest, and it cannot be excluded from our study that a very high intake of protein might have adverse long-term consequences on the kidneys. In the era of an alarming increase of obesity, high-protein weight-loss diets have a growing popularity in several countries. To my knowledge there are no long-term studies performed that can evaluate the effect of this diet on the kidneys.

Antioxidant intake (paper II)

We hypothesized that a diet rich in antioxidants would protect against CRF. However, our results contradict any strong link between intake of antioxidant vitamins and risk of CRF overall or various renal diseases. There is limited data from previous studies, and the results are inconsistent. A low serum level of vitamin C has been associated with creatinine elevations [118], another study found that low levels of certain diet-derived carotenoids were related to an increased risk of albuminuria [119].

Beside dietary patterns, several other factors seem to influence the antioxidant status in individuals, i.e. hypertension [248], smoking habits, alcohol consumption, psychological stress and ultraviolet light exposure [249]. In an attempt to elucidate whether subjects with a presumed increased state of oxidative stress benefit more from a high antioxidant intake, we conducted analyses stratified by smoking, alcohol consumption, and hypertension status. Our data lend some support for a moderately strong inverse association with high vitamin E intake among individuals with hypertension. Our finding is biologically plausible since oxidative stress appears to be an important factor in hypertension, presumably both as a cause and a consequence [248]. We only observed this risk reduction from a high intake of vitamin E, and not from any other antioxidant vitamin. This is in line with observational studies investigating the association between antioxidants and cardiovascular diseases, where vitamin E was more strongly linked to risk reduction than vitamin C or beta-carotene [117]. However, our observed association

lacked a dose-effect trend, and there were no indications of positive effects in any other stratum, including that of heavy smokers, where oxidative stress is equally implicated. Moreover, we made multiple analyses, and we cannot exclude chance as an alternative explanation. Our finding requires confirmation in other studies.

Since other compounds in fruits and vegetables than those specifically investigated might be of importance, we also analysed the association of crude intake of fruits and vegetables with CRF risk. Although there was a weak inverse association for vegetable intake and a weak positive association for fruit intake, no statistical significant results were obtained, and dose-effect trends were absent. Thus, our data did not support any overall protective effect from fruits and vegetables.

Deficiency of folic acid causes hyperhomocysteinemia, which may cause oxidative injury to the vascular walls [250], possibly including intrarenal capillaries. Hyperhomocysteinemia is commonly seen in ESRD, but has also been associated with microalbuminuria (an early indicator of kidney damage) [107], although this finding was not repeated in another cross-sectional study [119]. Our results do not indicate that a diet low in folic acid would increase risk of CRF.

Body mass, height and birth weight (paper III)

We found that both overweight in young ages, and obesity any time later in life, significantly increased risk of CRF overall. Our results confirm an accumulating body of clinical and experimental data implicating obesity as an important etiologic factor in renal disease [151, 251].

It is well known that obesity strongly increases risk of diabetes and hypertension [133, 134] and that both these conditions are important contributors to ESRD [48, 57]. Not surprisingly, we found the strongest positive association between a high BMI and risk of diabetic nephropathy (related to type II diabetes), and the second highest risk within the group nephrosclerosis (almost all patients were reported having hypertension as underlying cause of this diagnosis). Nevertheless, risk elevations were also observed among those diagnosed with glomerulonepritis and "other renal diseases". However, many of the cases with these renal diagnoses also had hypertension, and some were diabetics. It is clear that hypertension accompanies virtually all types of renal disease, not only as a cause, but also frequently as a consequence of the renal failure. We could not disentangle whether the CRF had led to hypertension, or if patients with glomerulonephritis and "other renal disease" also had a diagnosis of hypertension preceding the kidney disease, potentially related to obesity. To further elucidate the effect of BMI on CRF risk, independently of hypertension and diabetes, we conducted analyses stratified on diabetes and hypertension status. These results indicate that risk elevations remained in subjects without diabetes and in those without hypertension. Analyses confined to subjects who reported neither of these conditions produced a point estimate of similar magnitude, although there was a lack of precision due to small numbers. Notwithstanding that the adverse renal effects from obesity appear to be driven mainly by hypertension and diabetes, our findings give a suggestion that obesity might also damage the kidneys through other mechanisms. However, we cannot

exclude the possibility that undiagnosed hypertension and diabetes might have played a pathophysiological role.

In renal biopsies from severely obese patients with proteinuria, focal segmental glomerulosclerosis (FSGS) and/or glomerulomegaly are common findings [252-254]. The occurrence of this condition seems to have increased; in a clinicopathologic study from New York, the proportion of all renal biopsies exhibiting obesity-related FSGS or glomerulomegaly increased ten-fold from 1986 to 2000 [253]. In our study this diagnosis was not common, although a low rate of biopsies may have entailed underascertainment. Only 16 of our cases were diagnosed with FSGS, and of these only one had a lifetime highest BMI that exceeded 35 kg/m^2. Since obesity-induced FSGS has been mainly associated with morbid obesity, this entity might be a smaller problem in the comparably lean Swedish population, than in other populations with higher prevalence of severe obesity.

In contrast to other studies [164, 165] we did not find that low birth weight increases risk of CRF. However, our results must be interpreted cautiously. The analyses were hampered by a relatively large proportion of missing data on birth weight, and the statistical power was poor since few subjects reported low birth weight. In addition, we do not know whether the study participants reporting low birth weight were small for gestational age or born prematurely. It appears that intrauterine growth retardation rather than premature birth is of importance in this context [157]. A short stature could conceivably not be a risk factor per se, but is suggested to be linked with reduced number of nephrons [255]. Contrary to our result of birth weight, a short stature was related to an increased CRF risk, at least among men. This finding provides some support for the hypothesis that "oligonephropathy" increases risk of CRF.

Analgesic use (paper IV)

Both intake of paracetamol and aspirin were positively linked with increased risk of CRF, and the associations showed dose-response patterns. These associations are consistent with the findings of several previous case-control studies [196-198]. However, earlier cases-control studies aimed at investigating the relation between analgesics and CRF have been claimed to suffer from methodological flaws such as selection, information and confounding biases [194, 256]. One major concern is that several studies did not adequately adjust for phenacetin use. We do not think our results are biased from previous phenacetin use. We adjusted for phenacetin, moreover, since very few individuals had used phenacetin it is unlikely that such use would have importantly influenced CRF risk. Underreporting of phenacetin is unlikely, since the sales of this drug were restricted in 1961, and phenacetin shortly after disappeared from the Swedish market. Several of the previous studies were not population-based, and it was sometimes questionable whether the cases were drawn from the same population as the controls. We had a population-based setting, and controls were a true random sample from the study base. Other weaknesses in previous studies include lack of adequate adjustment for other potential confounders, such as race, sex, proxy response status and socio-economic status – weaknesses that we could avoid. Unlike all but one of the earlier studies, we used a visual aid, which is known to improve recall of drug intake

[234], and we obtained detailed information about lifetime analgesic use in face-to-face interviews. To diminish the risk that symptoms related to CRF prompted use of analgesics, our case patients had earlier-stage CRF than in most of the previous studies where the case definition was ESRD. In an attempt to elucidate a potential role of reverse causation, we performed analyses disregarding the latest 5 and 10 years of analgesic intake, but the risk estimates were only slightly attenuated.

In conclusion, I believe that our study was carefully planned and we managed to avoid most shortcomings. However, it must be kept in mind that not only symptoms from CRF, but also from underlying diseases causing CRF may lead to increased analgesic consumption (as discussed in "general discussion", page 50), and like in earlier studies, we were unable to confidently rule out "confounding by indication" as a contributing explanation of our results. The interpretation of the association between aspirin use and CRF is further complicated by the fact that individuals with cardiovascular diseases are treated with aspirin. Since it is well known that chronic renal failure and cardiovascular diseases are closely interrelated, it can be expected that CRF patients have a higher consumption of aspirin due to this indication.

It can be concluded that there is previous strong evidence for a causal link between phenacetin and AAN [194], but this could not be evaluated in our study since very few subjects had used phenacetin. Although confounding by indication cannot be completely ruled out, our results are consistent with an exacerbating effect of paracetamol, and – with less certainty – of aspirin, on CRF. The results of a newly published study support an exacerbating effect of analgesics not including phenacetin. This study included 78 subjects with AAN, i.e., CRF in the absence of other renal diagnoses and a positive history of excessive analgesic use. Among subjects who continued to take analgesics, renal function declined 3.53 ml/min/year faster than among the patients who stopped taking analgesics. After 64-70 months of follow-up, continued analgesic use conferred a 6-fold increase in risk of death or progression to ESRD [257]. Another recent study, from the physician's Health Study cohort, could not confirm any accelerated decline in renal function among analgesic-using individuals without pre-existing renal disease, but the investigators dichotomized the trend over time, and the cutpoint used for deterioration may have been set on a level that was consistent with normal, age-related loss of function [201]. The results from these two cohort studies, together with our findings, might, however, indicate that non-phenacetin analgesics do not constitute strong renal risk factors among healthy individuals but may increase progression rate among individuals with pre-existing kidney disease.

Socioeconomic status, gender and risk factors

As previously reported, there was a markedly increased risk of CRF among patients with a lower educational level and lower socio economic status (based on occupations) in our study [208]. We also found that men have a higher risk of CRF than females. The female to male ratio of cases in our study was 1: 1.8. This gender distribution is similar to the distribution observed in the Swedish register of patients on renal replacement therapy. The exposures that were associated with risk of CRF in this thesis could not explain the effects of socio-economic status or gender. None of the exposures were

stronger risk factors in those with lower socio-economic status or in men. Thus, these variations in risk observed in our study (as well as in many other previous studies) depend on yet unidentified factors.

CONCLUSIONS

- Heavy cigarette smoking is a risk factor for the development of chronic renal failure. The increase in relative risk is at least as strong in women as in men. Chronic renal failure classified as nephrosclerosis was the subtype that was most strongly linked to smoking, but significant positive associations were also noted with glomerulonephritis, and among women, with diabetic nephropathy.

- Other tobacco use, such as cigar smoking, pipe smoking and use of oral snuff, was not related to risk of chronic renal failure.

- A high intake of antioxidants from diet and supplements do not appear to protect against chronic renal failure, with the possible exception that a high intake of vitamin E might be protective in individuals with hypertension.

- A diet rich in proteins may increase risk of diabetic nephropathy, however, reverse causation cannot be excluded as an alternative explanation of the findings. A high protein intake, within the range observed in our Swedish population, does not increase risk of non-diabetic moderate chronic renal failure.

- Being overweight in early adulthood or obese anytime in life is linked to an increased risk of chronic renal failure. Although much of the excess risk is driven by the higher prevalence of hypertension and diabetes among obese, an additional pathway may exist.

- A low birth weight appears not to increase risk of chronic renal failure in the Swedish population. However, some reservations must be made due to low statistical power and potential misclassification. A short stature, another possible marker of a low number of nephrons, is associated with increased risk of chronic renal failure, at least among men.

- Regular use of paracetamol and aspirin might exacerbate chronic renal failure. However, we cannot rule out confounding by indication as an alternative explanation, i.e. that predisposing conditions for chronic renal failure prompted analgesic consumption.

FUTURE STUDIES

Population-based study of early signs of renal disease

Signs of early kidney damage are common in other countries [5, 6]. It would be of interest to conduct a cross-sectional study with a large random sample from the Swedish population to estimate the prevalence of any signs of kidney damage (hematuria, proteinuria and reduced glomerular filtration rate) and to explore their relation to suspected risk factors. With sufficient funding it would be exciting to form a cohort from this cross-sectional study, and to follow the study participants with medical check-ups and new questionnaires every fifth year, i.e., to investigate into the long-term development of renal function, the incidence of CRF, and possible risk factors, in a stratum of subjects without evidence of impaired renal function (with or without predisposing diseases), and in strata with various signs of early kidney damage (again, with or without predisposing diseases).

The study would include;

-A careful medical check-up to evaluate to what extent specific renal diseases, hypertension, and diabetes might explain signs of early kidney damage. In addition detailed information on cardiovascular diseases, other medical conditions, and drug treatments.

-Blood pressure, weight, measurements of fat distribution, and height.

-Blood and urine samples for testing of proteinuria, hematuria, serum/blood concentrations of creatinine, glucose, haemoglobin, urate and lipids, testing of insulin resistance/hyperinsulinemia, inflammatory markers and HbA1C in diabetics. Extra blood samples saved to have the potential to do genetic testing of renal susceptibility genes and assess serum levels of antioxidant vitamins.

-A questionnaire encompassing questions on education, socio-economic factors, birth weight, family history of renal disease and lifestyle factors such as tobacco, alcohol and analgesic use.

Cohort studies with incident cases of individuals with diabetes and hypertension

There are of course many factors of interest to study in these groups of patients, both for initiation of nephropathy and progression. Of particular interest might be to perform genetic association studies of signs of nephropathy in relation to angiotensinogen converting enzyme gene and angiotensin II receptor gene haplotypes. In addition it could be evaluated whether specific haplotypes benefit more from therapy with angiotensinogen converting enzyme inhibitors or angiontensin II receptor blockers. Among diabetics, initially free from nephropathy, it would be valuable to prospectively follow whether a high protein intake increase risk of kidney damage.

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REFERENCES

1. Lysaght MJ. Maintenance dialysis population dynamics: current trends and long-term implications. J Am Soc Nephrol 2002; 13:S37-40.
2. Moeller S, Gioberge S, Brown G. ESRD patients in 2001: global overview of patients, treatment modalities and development trends. Nephrol Dial Transplant 2002; 17:2071-6.
3. SRAU. (2003). Renal Replacement Therapy in Sweden 1991-2002 (in Swedish). Svenskt Register för Aktiv Uremivård.
4. Port FK. End-stage renal disease: magnitude of the problem, prognosis of future trends and possible solutions. Kidney Int Suppl 1995; 50:S3-6.
5. Chadban SJ, Briganti EM, Kerr PG, et al. Prevalence of kidney damage in Australian adults: The AusDiab kidney study. J Am Soc Nephrol 2003; 14:S131-8.
6. Coresh J, Astor BC, Greene T, Eknoyan G, Levey AS. Prevalence of chronic kidney disease and decreased kidney function in the adult US population: Third National Health and Nutrition Examination Survey. Am J Kidney Dis 2003; 41:1-12.
7. Iseki K, Ikemiya Y, Iseki C, Takishita S. Proteinuria and the risk of developing end-stage renal disease. Kidney Int 2003; 63:1468-74.
8. Fox CS, Larson MG, Leip EP, Culleton B, Wilson PW, Levy D. Predictors of new-onset kidney disease in a community-based population. Jama 2004; 291:844-50.
9. Go AS, Chertow GM, Fan D, McCulloch CE, Hsu CY. Chronic kidney disease and the risks of death, cardiovascular events, and hospitalization. N Engl J Med 2004; 351:1296-305.
10. Brenner BM. Retarding the progression of renal disease. Kidney Int 2003; 64:370-8.
11. McClellan WM, Flanders WD. Risk factors for progressive chronic kidney disease. J Am Soc Nephrol 2003; 14:S65-70.
12. Dennis L. Kasper EB, Anthony S. Fauci, Stephen L. Hauser, Dan L. Longo, J. Larry Jameson, and Kurt J. Isselbacher. (2003) Harrison's Principles of Internal Medicine, 16th Edition. Part 11; Disorders of the kidney and urinary tract. McGrawHill.
13. Parmar MS. Chronic renal disease. Bmj 2002; 325:85-90.
14. K/DOQI clinical practice guidelines for chronic kidney disease: evaluation, classification, and stratification. Am J Kidney Dis 2002; 39:S1-266.
15. King AJ, Levey AS. Dietary protein and renal function. J Am Soc Nephrol 1993; 3:1723-37.
16. Clase CM, Garg AX, Kiberd BA. Classifying kidney problems: can we avoid framing risks as diseases? Bmj 2004; 329:912-5.
17. USRDS. The United States Renal Data system, Am J Kidney Dis; 42:S15:1-230.
18. ERA-EDTA. European Renal Association-European Dialysis and Transplant Association registry website; Annual report 2002. (accessed Jan. 25, 2005, at http://www.era-edta-reg.org/index.jsp).

19. Iseki K, Tozawa M, Iseki C, Takishita S, Ogawa Y. Demographic trends in the Okinawa Dialysis Study (OKIDS) registry (1971-2000). Kidney Int 2002; 61:668-75.
20. McDonald SP, Russ GR, Kerr PG, Collins JF. ESRD in Australia and New Zealand at the end of the millennium: a report from the ANZDATA registry. Am J Kidney Dis 2002; 40:1122-31.
21. Sims RJ, Cassidy MJ, Masud T. The increasing number of older patients with renal disease. Bmj 2003; 327:463-4.
22. John R, Webb M, Young A, Stevens PE. Unreferred chronic kidney disease: a longitudinal study. Am J Kidney Dis 2004; 43:825-35.
23. Drey N, Roderick P, Mullee M, Rogerson M. A population-based study of the incidence and outcomes of diagnosed chronic kidney disease. Am J Kidney Dis 2003; 42:677-84.
24. Usami T, Koyama K, Takeuchi O, Morozumi K, Kimura G. Regional variations in the incidence of end-stage renal failure in Japan. Jama 2000; 284:2622-4.
25. Stengel B, Billon S, Van Dijk PC, et al. Trends in the incidence of renal replacement therapy for end-stage renal disease in Europe, 1990-1999. Nephrol Dial Transplant 2003; 18:1824-33.
26. USRDS. The United States Renal Data System. Am J Kidney Dis 2003; 42:S15:1-230.
27. Neugarten J, Acharya A, Silbiger SR. Effect of gender on the progression of nondiabetic renal disease: a meta-analysis. J Am Soc Nephrol 2000; 11:319-29.
28. Seliger SL, Davis C, Stehman-Breen C. Gender and the progression of renal disease. Curr Opin Nephrol Hypertens 2001; 10:219-25.
29. Silbiger SR, Neugarten J. The role of gender in the progression of renal disease. Adv Ren Replace Ther 2003; 10:3-14.
30. Neugarten J. Gender and the progression of renal disease. J Am Soc Nephrol 2002; 13:2807-9.
31. McDonald SP, Russ GR. Current incidence, treatment patterns and outcome of end-stage renal disease among indigenous groups in Australia and New Zealand. Nephrology (Carlton) 2003; 8:42-8.
32. URSD (2004). United States Renal Data System web-site; Annual data report 2004. (accesssed Jan. 25, 2005. at http://www.usrds.org/adr.htm).
33. Lopes AA. Relationships of race and ethnicity to progression of kidney dysfunction and clinical outcomes in patients with chronic kidney failure. Adv Ren Replace Ther 2004; 11:14-23.
34. Stewart JH, McCredie MR, McDonald SP. The incidence of treated end-stage renal disease in New Zealand Maori and Pacific Island people and in Indigenous Australians. Nephrol Dial Transplant 2004; 19:678-85.
35. Hoy WE, Rees M, Kile E, Mathews JD, Wang Z. A new dimension to the Barker hypothesis: low birthweight and susceptibility to renal disease. Kidney Int 1999; 56:1072-7.
36. Bowden DW. Genetics of kidney disease. Kidney Int Suppl 2003:S8-12.
37. Lindeman RD, Tobin J, Shock NW. Longitudinal studies on the rate of decline in renal function with age. J Am Geriatr Soc 1985; 33:278-85.
38. Nenov VD, Taal MW, Sakharova OV, Brenner BM. Multi-hit nature of chronic renal disease. Curr Opin Nephrol Hypertens 2000; 9:85-97.

39. Nyengaard JR, Bendtsen TF. Glomerular number and size in relation to age, kidney weight, and body surface in normal man. Anat Rec 1992; 232:194-201.
40. Remuzzi G, Bertani T. Pathophysiology of progressive nephropathies. N Engl J Med 1998; 339:1448-56.
41. Tryggvason K, Pettersson E. Causes and consequences of proteinuria: the kidney filtration barrier and progressive renal failure. J Intern Med 2003; 254:216-24.
42. Brenner BM, Meyer TW, Hostetter TH. Dietary protein intake and the progressive nature of kidney disease: the role of hemodynamically mediated glomerular injury in the pathogenesis of progressive glomerular sclerosis in aging, renal ablation, and intrinsic renal disease. N Engl J Med 1982; 307:652-9.
43. Brenner BM, Lawler EV, Mackenzie HS. The hyperfiltration theory: a paradigm shift in nephrology. Kidney Int 1996; 49:1774-7.
44. Gross ML, Amann K. Progression of renal disease: new insights into risk factors and pathomechanisms. Curr Opin Nephrol Hypertens 2004; 13:307-12.
45. Young JH, Klag MJ, Muntner P, Whyte JL, Pahor M, Coresh J. Blood pressure and decline in kidney function: findings from the Systolic Hypertension in the Elderly Program (SHEP). J Am Soc Nephrol 2002; 13:2776-82.
46. Ishida K, Ishida H, Narita M, et al. Factors affecting renal function in 119 985 adults over three years. Qjm 2001; 94:541-50.
47. Perry HM, Jr., Miller JP, Fornoff JR, et al. Early predictors of 15-year end-stage renal disease in hypertensive patients. Hypertension 1995; 25:587-94.
48. Klag MJ, Whelton PK, Randall BL, et al. Blood pressure and end-stage renal disease in men. N Engl J Med 1996; 334:13-8.
49. Hsu CY. Does non-malignant hypertension cause renal insufficiency? Evidence-based perspective. Curr Opin Nephrol Hypertens 2002; 11:267-72.
50. Kincaid-Smith P. Hypothesis: obesity and the insulin resistance syndrome play a major role in end-stage renal failure attributed to hypertension and labelled 'hypertensive nephrosclerosis'. J Hypertens 2004; 22:1051-5.
51. Bleyer AJ, Chen R, D'Agostino RB, Jr., Appel RG. Clinical correlates of hypertensive end-stage renal disease. Am J Kidney Dis 1998; 31:28-34.
52. Iseki K, Miyasato F, Uehara H, et al. Outcome study of renal biopsy patients in Okinawa, Japan. Kidney Int 2004; 66:914-9.
53. Freedman BI, Iskander SS, Buckalew VM, Jr., Burkart JM, Appel RG. Renal biopsy findings in presumed hypertensive nephrosclerosis. Am J Nephrol 1994; 14:90-4.
54. Zucchelli P, Zuccala A. Can we accurately diagnose nephrosclerosis? Nephrol Dial Transplant 1995; 10:2-5.
55. Bleyer AJ, Appel RG. Risk factors associated with hypertensive nephrosclerosis. Nephron 1999; 82:193-8.
56. Ritz E, Rychlik I, Locatelli F, Halimi S. End-stage renal failure in type 2 diabetes: A medical catastrophe of worldwide dimensions. Am J Kidney Dis 1999; 34:795-808.
57. Brancati FL, Whelton PK, Randall BL, Neaton JD, Stamler J, Klag MJ. Risk of end-stage renal disease in diabetes mellitus: a prospective cohort study of men screened for MRFIT. Multiple Risk Factor Intervention Trial. Jama 1997; 278:2069-74.

58. Zimmet P. The burden of type 2 diabetes: are we doing enough? Diabetes Metab 2003; 29:6S9-18.
59. Bergrem H, Leivestad T. Diabetic nephropathy and end-stage renal failure: the Norwegian story. Adv Ren Replace Ther 2001; 8:4-12.
60. Berger M, Monks D, Wanner C, Lindner TH. Diabetic nephropathy: an inherited disease or just a diabetic complication? Kidney Blood Press Res 2003; 26:143-54.
61. Nelson RG, Morgenstern H, Bennett PH. An epidemic of proteinuria in Pima Indians with type 2 diabetes mellitus. Kidney Int 1998; 54:2081-8.
62. Routh HB, Bhowmik KR, Parish JL, Parish LC. Historical aspects of tobacco use and smoking. Clin Dermatol 1998; 16:539-44.
63. WHO. (2004) World Healths organization web-site; Tobacco Free Initiative (TFI). (accessed Jan 03. 2005, at http://www.who.int/tobacco/health_priority/en).
64. SCB. Statistiska centralbyrån website; Statistik rökvanor. (acessed Jan. 25, 2005 at http://www.scb.se/statistik/LE/LE0101/1980I01/HA1203.xls.)
65. Fagerstrom KO, Schildt EB. Should the European Union lift the ban on snus? Evidence from the Swedish experience. Addiction 2003; 98:1191-5.
66. Lagergren J, Bergstrom R, Lindgren A, Nyren O. The role of tobacco, snuff and alcohol use in the aetiology of cancer of the oesophagus and gastric cardia. Int J Cancer 2000; 85:340-6.
67. Lewin F, Norell SE, Johansson H, et al. Smoking tobacco, oral snuff, and alcohol in the etiology of squamous cell carcinoma of the head and neck: a population-based case-referent study in Sweden. Cancer 1998; 82:1367-75.
68. Schildt EB, Eriksson M, Hardell L, Magnuson A. Oral snuff, smoking habits and alcohol consumption in relation to oral cancer in a Swedish case-control study. Int J Cancer 1998; 77:341-6.
69. Bolinder G, Alfredsson L, Englund A, de Faire U. Smokeless tobacco use and increased cardiovascular mortality among Swedish construction workers. Am J Public Health 1994; 84:399-404.
70. Accortt NA, Waterbor JW, Beall C, Howard G. Chronic disease mortality in a cohort of smokeless tobacco users. Am J Epidemiol 2002; 156:730-7.
71. Orth SR. Smoking--a renal risk factor. Nephron 2000; 86:12-26.
72. Colhoun HM, Lee ET, Bennett PH, et al. Risk factors for renal failure: the WHO Multinational Study of Vascular Disease in Diabetes. Diabetologia 2001; 44:S46-53.
73. Christiansen JS. Cigarette smoking and prevalence of microangiopathy in juvenile-onset insulin-dependent diabetes mellitus. Diabetes Care 1978; 1:146-9.
74. Sawicki PT, Didjurgeit U, Muhlhauser I, Bender R, Heinemann L, Berger M. Smoking is associated with progression of diabetic nephropathy. Diabetes Care 1994; 17:126-31.
75. Biesenbach G, Grafinger P, Janko O, Zazgornik J. Influence of cigarette-smoking on the progression of clinical diabetic nephropathy in type 2 diabetic patients. Clin Nephrol 1997; 48:146-50.
76. Rossing K, Christensen PK, Hovind P, Tarnow L, Rossing P, Parving HH. Progression of nephropathy in type 2 diabetic patients. Kidney Int 2004; 66:1596-605.

77. Orth SR. Effects of smoking on systemic and intrarenal hemodynamics: influence on renal function. J Am Soc Nephrol 2004; 15:S58-63.
78. Mimran A, Ribstein J, DuCailar G, Halimi JM. Albuminuria in normals and essential hypertension. J Diabetes Complications 1994; 8:150-6.
79. Horner D, Fliser D, Klimm HP, Ritz E. Albuminuria in normotensive and hypertensive individuals attending offices of general practitioners. J Hypertens 1996; 14:655-60.
80. Regalado M, Yang S, Wesson DE. Cigarette smoking is associated with augmented progression of renal insufficiency in severe essential hypertension. Am J Kidney Dis 2000; 35:687-94.
81. Yaqoob M, Bell GM, Percy DF, Finn R. Primary glomerulonephritis and hydrocarbon exposure: a case-control study and literature review. Q J Med 1992; 83:409-18.
82. Wakai K, Nakai S, Matsuo S, et al. Risk factors for IgA nephropathy: a case-control study with incident cases in Japan. Nephron 2002; 90:16-23.
83. Orth SR, Stockmann A, Conradt C, et al. Smoking as a risk factor for end-stage renal failure in men with primary renal disease. Kidney Int 1998; 54:926-31.
84. Stengel B, Couchoud C, Cenee S, Hemon D. Age, blood pressure and smoking effects on chronic renal failure in primary glomerular nephropathies. Kidney Int 2000; 57:2519-26.
85. Ward MM, Studenski S. Clinical prognostic factors in lupus nephritis. The importance of hypertension and smoking. Arch Intern Med 1992; 152:2082-8.
86. Font J, Ramos-Casals M, Cervera R, et al. Cardiovascular risk factors and the long-term outcome of lupus nephritis. Qjm 2001; 94:19-26.
87. Pinto-Sietsma SJ, Mulder J, Janssen WM, Hillege HL, de Zeeuw D, de Jong PE. Smoking is related to albuminuria and abnormal renal function in nondiabetic persons. Ann Intern Med 2000; 133:585-91.
88. Halimi JM, Giraudeau B, Vol S, et al. Effects of current smoking and smoking discontinuation on renal function and proteinuria in the general population. Kidney Int 2000; 58:1285-92.
89. Hillege HL, Janssen WM, Bak AA, et al. Microalbuminuria is common, also in a nondiabetic, nonhypertensive population, and an independent indicator of cardiovascular risk factors and cardiovascular morbidity. J Intern Med 2001; 249:519-26.
90. Briganti EM, Branley P, Chadban SJ, et al. Smoking is associated with renal impairment and proteinuria in the normal population: the AusDiab kidney study. Australian Diabetes, Obesity and Lifestyle Study. Am J Kidney Dis 2002; 40:704-12.
91. Bleyer AJ, Shemanski LR, Burke GL, Hansen KJ, Appel RG. Tobacco, hypertension, and vascular disease: risk factors for renal functional decline in an older population. Kidney Int 2000; 57:2072-9.
92. Whelton P, Randall B, Neaton J, Stamler J, Brancati F, Klag M. Cigarette smoking and ESRD incidence in men screened for the MRFIT. (abstract). J Am Soc Nephrol 1995; 6:408a.
93. Addis T. Glomerular nephritis: diagnosis and treatment. New York: Macmilian 1948.

94. D'Amico G, Gentile MG, Fellin G, Manna G, Cofano F. Effect of dietary protein restriction on the progression of renal failure: a prospective randomized trial. Nephrol Dial Transplant 1994; 9:1590-4.
95. Ihle BU, Becker GJ, Whitworth JA, Charlwood RA, Kincaid-Smith PS. The effect of protein restriction on the progression of renal insufficiency. N Engl J Med 1989; 321:1773-7.
96. Klahr S, Levey AS, Beck GJ, et al. The effects of dietary protein restriction and blood-pressure control on the progression of chronic renal disease. Modification of Diet in Renal Disease Study Group. N Engl J Med 1994; 330:877-84.
97. Wingen AM, Fabian-Bach C, Schaefer F, Mehls O. Randomised multicentre study of a low-protein diet on the progression of chronic renal failure in children. European Study Group of Nutritional Treatment of Chronic Renal Failure in Childhood. Lancet 1997; 349:1117-23.
98. Remuzzi A, Perticucci E, Battaglia C, D'Amico G, Gentile MG, Remuzzi G. Low-protein diet and glomerular size-selective function in membranous glomerulopathy. Am J Kidney Dis 1991; 17:317-22.
99. Locatelli F, Alberti D, Graziani G, Buccianti G, Redaelli B, Giangrande A. Prospective, randomised, multicentre trial of effect of protein restriction on progression of chronic renal insufficiency. Northern Italian Cooperative Study Group. Lancet 1991; 337:1299-304.
100. Levey AS, Greene T, Beck GJ, et al. Dietary protein restriction and the progression of chronic renal disease: what have all of the results of the MDRD study shown? Modification of Diet in Renal Disease Study group. J Am Soc Nephrol 1999; 10:2426-39.
101. Fouque D, Wang P, Laville M, Boissel JP. Low protein diets delay end-stage renal disease in non diabetic adults with chronic renal failure. Cochrane Database Syst Rev 2000:CD001892.
102. Kasiske BL, Lakatua JD, Ma JZ, Louis TA. A meta-analysis of the effects of dietary protein restriction on the rate of decline in renal function. Am J Kidney Dis 1998; 31:954-61.
103. Pedrini MT, Levey AS, Lau J, Chalmers TC, Wang PH. The effect of dietary protein restriction on the progression of diabetic and nondiabetic renal diseases: a meta-analysis. Ann Intern Med 1996; 124:627-32.
104. Kopple JD. National kidney foundation K/DOQI clinical practice guidelines for nutrition in chronic renal failure. Am J Kidney Dis 2001; 37:S66-70.
105. Bernardi A, Biasia F, Piva M, et al. Dietary protein intake and nutritional status in patients with renal transplant. Clin Nephrol 2000; 53:3-5.
106. Knight EL, Stampfer MJ, Hankinson SE, Spiegelman D, Curhan GC. The impact of protein intake on renal function decline in women with normal renal function or mild renal insufficiency. Ann Intern Med 2003; 138:460-7.
107. Hoogeveen EK, Kostense PJ, Jager A, et al. Serum homocysteine level and protein intake are related to risk of microalbuminuria: the Hoorn Study. Kidney Int 1998; 54:203-9.
108. Wrone EM, Carnethon MR, Palaniappan L, Fortmann SP. Association of dietary protein intake and microalbuminuria in healthy adults: Third National Health and Nutrition Examination Survey. Am J Kidney Dis 2003; 41:580-7.

109. Toeller M, Buyken A, Heitkamp G, et al. Protein intake and urinary albumin excretion rates in the EURODIAB IDDM Complications Study. Diabetologia 1997; 40:1219-26.
110. Thannickal VJ, Fanburg BL. Reactive oxygen species in cell signaling. Am J Physiol Lung Cell Mol Physiol 2000; 279:L1005-28.
111. Willcox JK, Ash SL, Catignani GL. Antioxidants and prevention of chronic disease. Crit Rev Food Sci Nutr 2004; 44:275-95.
112. Himmelfarb J, McMonagle E. Manifestations of oxidant stress in uremia. Blood Purif 2001; 19:200-5.
113. Descamps-Latscha B, Drueke T, Witko-Sarsat V. Dialysis-induced oxidative stress: biological aspects, clinical consequences, and therapy. Semin Dial 2001; 14:193-9.
114. Tylicki L, Rutkowski B, Horl WH. Antioxidants: a possible role in kidney protection. Kidney Blood Press Res 2003; 26:303-14.
115. Gazdikova K, Gvozdjakova A, Kucharska J, Spustova V, Braunova Z, Dzurik R. Oxidative stress and plasma concentrations of coenzyme Q10, alpha-tocopherol, and beta-carotene in patients with a mild to moderate decrease of kidney function. Nephron 2001; 88:285.
116. Oberg BP, McMenamin E, Lucas FL, et al. Increased prevalence of oxidant stress and inflammation in patients with moderate to severe chronic kidney disease. Kidney Int 2004; 65:1009-16.
117. Rimm EB, Stampfer MJ. The role of antioxidants in preventive cardiology. Curr Opin Cardiol 1997; 12:188-94.
118. Chen J, He J, Ogden LG, Batuman V, Whelton PK. Relationship of serum antioxidant vitamins to serum creatinine in the US population. Am J Kidney Dis 2002; 39:460-8.
119. Rowley K, O'Dea K, Su Q, Jenkins AJ, Best JD. Low plasma concentrations of diet-derived antioxidants in association with microalbuminuria in Indigenous Australian populations. Clin Sci (Lond) 2003; 105:569-75.
120. Vivekananthan DP, Penn MS, Sapp SK, Hsu A, Topol EJ. Use of antioxidant vitamins for the prevention of cardiovascular disease: meta-analysis of randomised trials. Lancet 2003; 361:2017-23.
121. Meagher EA. Treatment of atherosclerosis in the new millennium: is there a role for vitamin E? Prev Cardiol 2003; 6:85-90.
122. Bursell SE, Clermont AC, Aiello LP, et al. High-dose vitamin E supplementation normalizes retinal blood flow and creatinine clearance in patients with type 1 diabetes. Diabetes Care 1999; 22:1245-51.
123. Tahzib M, Frank R, Gauthier B, Valderrama E, Trachtman H. Vitamin E treatment of focal segmental glomerulosclerosis: results of an open-label study. Pediatr Nephrol 1999; 13:649-52.
124. Gaede P, Poulsen HE, Parving HH, Pedersen O. Double-blind, randomised study of the effect of combined treatment with vitamin C and E on albuminuria in Type 2 diabetic patients. Diabet Med 2001; 18:756-60.
125. McAuliffe AV, Brooks BA, Fisher EJ, Molyneaux LM, Yue DK. Administration of ascorbic acid and an aldose reductase inhibitor (tolrestat) in diabetes: effect on urinary albumin excretion. Nephron 1998; 80:277-84.

126. WHO. World health Organization website; Obesity and overweight. (accessed Jan. 2., 2005. at http://www.who.int/dietphysicalactivity/publications/facts/obesity/en/.)
127. Physical status: the use and interpretation of anthropometry. Report of a WHO Expert Committee. World Health Organ Tech Rep Ser 1995; 854:1-452.
128. Flegal KM, Carroll MD, Ogden CL, Johnson CL. Prevalence and trends in obesity among US adults, 1999-2000. Jama 2002; 288:1723-7.
129. Lissner L, Johansson SE, Qvist J, Rossner S, Wolk A. Social mapping of the obesity epidemic in Sweden. Int J Obes Relat Metab Disord 2000; 24:801-5.
130. O'Brien PE, Dixon JB. The extent of the problem of obesity. Am J Surg 2002; 184:4S-8S.
131. Hall JE, Henegar JR, Dwyer TM, et al. Is obesity a major cause of chronic kidney disease? Adv Ren Replace Ther 2004; 11:41-54.
132. Iseki K, Ikemiya Y, Kinjo K, Inoue T, Iseki C, Takishita S. Body mass index and the risk of development of end-stage renal disease in a screened cohort. Kidney Int 2004; 65:1870-6.
133. Mokdad AH, Ford ES, Bowman BA, et al. Prevalence of obesity, diabetes, and obesity-related health risk factors, 2001. Jama 2003; 289:76-9.
134. El-Atat F, Aneja A, McFarlane S, Sowers J. Obesity and hypertension. Endocrinol Metab Clin North Am 2003; 32:823-54.
135. Savage S, Nagel NJ, Estacio RO, Lukken N, Schrier RW. Clinical factors associated with urinary albumin excretion in type II diabetes. Am J Kidney Dis 1995; 25:836-44.
136. Spangler JG, Konen JC. Hypertension, hyperlipidemia, and abdominal obesity and the development of microalbuminuria in patients with non-insulin-dependent diabetes mellitus. J Am Board Fam Pract 1996; 9:1-6.
137. Ribstein J, du Cailar G, Mimran A. Combined renal effects of overweight and hypertension. Hypertension 1995; 26:610-5.
138. Bonnet F, Deprele C, Sassolas A, et al. Excessive body weight as a new independent risk factor for clinical and pathological progression in primary IgA nephritis. Am J Kidney Dis 2001; 37:720-7.
139. Praga M, Hernandez E, Herrero JC, et al. Influence of obesity on the appearance of proteinuria and renal insufficiency after unilateral nephrectomy. Kidney Int 2000; 58:2111-8.
140. Meier-Kriesche HU, Arndorfer JA, Kaplan B. The impact of body mass index on renal transplant outcomes: a significant independent risk factor for graft failure and patient death. Transplantation 2002; 73:70-4.
141. Fiske WD, 3rd, Blouin RA, Mitchell B, McNamara PJ. Renal function in the obese Zucker rat. Int J Obes 1986; 10:175-83.
142. Maddox DA, Alavi FK, Santella RN, Zawada ET, Jr. Prevention of obesity-linked renal disease: age-dependent effects of dietary food restriction. Kidney Int 2002; 62:208-19.
143. Reisin E, Messerli FG, Ventura HO, Frohlich ED. Renal haemodynamic studies in obesity hypertension. J Hypertens 1987; 5:397-400.
144. Chagnac A, Weinstein T, Korzets A, Ramadan E, Hirsch J, Gafter U. Glomerular hemodynamics in severe obesity. Am J Physiol Renal Physiol 2000; 278:F817-22.

145. Bosma RJ, van der Heide JJ, Oosterop EJ, de Jong PE, Navis G. Body mass index is associated with altered renal hemodynamics in non-obese healthy subjects. Kidney Int 2004; 65:259-65.
146. Pinto-Sietsma SJ, Navis G, Janssen WM, de Zeeuw D, Gans RO, de Jong PE. A central body fat distribution is related to renal function impairment, even in lean subjects. Am J Kidney Dis 2003; 41:733-41.
147. Tozawa M, Iseki K, Iseki C, Oshiro S, Ikemiya Y, Takishita S. Influence of smoking and obesity on the development of proteinuria. Kidney Int 2002; 62:956-62.
148. Chagnac A, Weinstein T, Herman M, Hirsh J, Gafter U, Ori Y. The effects of weight loss on renal function in patients with severe obesity. J Am Soc Nephrol 2003; 14:1480-6.
149. Morales E, Valero MA, Leon M, Hernandez E, Praga M. Beneficial effects of weight loss in overweight patients with chronic proteinuric nephropathies. Am J Kidney Dis 2003; 41:319-27.
150. de Jong PE, Verhave JC, Pinto-Sietsma SJ, Hillege HL. Obesity and target organ damage: the kidney. Int J Obes Relat Metab Disord 2002; 26:S21-4.
151. Praga M. Obesity--a neglected culprit in renal disease. Nephrol Dial Transplant 2002; 17:1157-9.
152. Scaglione R, Ganguzza A, Corrao S, et al. Central obesity and hypertension: pathophysiologic role of renal haemodynamics and function. Int J Obes Relat Metab Disord 1995; 19:403-9.
153. Vega GL. Obesity and the metabolic syndrome. Minerva Endocrinol 2004; 29:47-54.
154. Bagby SP. Obesity-initiated metabolic syndrome and the kidney: a recipe for chronic kidney disease? J Am Soc Nephrol 2004; 15:2775-91.
155. Mykkanen L, Zaccaro DJ, Wagenknecht LE, Robbins DC, Gabriel M, Haffner SM. Microalbuminuria is associated with insulin resistance in nondiabetic subjects: the insulin resistance atherosclerosis study. Diabetes 1998; 47:793-800.
156. Chen J, Muntner P, Hamm LL, et al. Insulin resistance and risk of chronic kidney disease in nondiabetic US adults. J Am Soc Nephrol 2003; 14:469-77.
157. Barker DJ. Fetal origins of coronary heart disease. Br Heart J 1993; 69:195-6.
158. Godfrey KM, Barker DJ. Fetal nutrition and adult disease. Am J Clin Nutr 2000; 71:1344S-52S.
159. Ingelfinger JR, Woods LL. Perinatal programming, renal development, and adult renal function. Am J Hypertens 2002; 15:46S-49S.
160. Marchand MC, Langley-Evans SC. Intrauterine programming of nephron number: the fetal flaw revisited. J Nephrol 2001; 14:327-31.
161. Ingelfinger JR. Pathogenesis of perinatal programming. Curr Opin Nephrol Hypertens 2004; 13:459-64.
162. Brenner BM. The etiology of adult hypertension and progressive renal injury: an hypothesis. Bull Mem Acad R Med Belg 1994; 149:121-5; discussion 125-7.
163. Mackenzie HS, Lawler EV, Brenner BM. Congenital oligonephropathy: The fetal flaw in essential hypertension? Kidney Int Suppl 1996; 55:S30-4.
164. Lackland DT, Egan BM, Fan ZJ, Syddall HE. Low birth weight contributes to the excess prevalence of end-stage renal disease in African Americans. J Clin Hypertens (Greenwich) 2001; 3:29-31.

165. Fan Z, Lipsitz S, Egan B, Lackland D. The impact of birth weight on the racial disparity of end-stage renal disease. Ann Epidemiol 2000; 10:459.
166. Eshoj O, Vaag A, Borch-Johnsen K, Feldt-Rasmussen B, Beck-Nielsen H. Is low birth weight a risk factor for the development of diabetic nephropathy in patients with type 1 diabetes? A population-based case-control study. J Intern Med 2002; 252:524-8.
167. Jacobsen P, Rossing P, Tarnow L, Hovind P, Parving HH. Birth weight - a risk factor for progression in diabetic nephropathy? J Intern Med 2003; 253:343-50.
168. Nelson RG, Morgenstern H, Bennett PH. Birth weight and renal disease in Pima Indians with type 2 diabetes mellitus. Am J Epidemiol 1998; 148:650-6.
169. Gould MM, Mohamed-Ali V, Goubet SA, Yudkin JS, Haines AP. Microalbuminuria: associations with height and sex in non-diabetic subjects. Bmj 1993; 306:240-2.
170. Olivarius Nde F, Vestbo E, Andreasen AH, Mogensen CE. Renal involvement is related to body height in newly diagnosed diabetic women aged 40 years or over. Diabetes Metab 2001; 27:14-8.
171. Rossing P, Tarnow L, Nielsen FS, Boelskifte S, Brenner BM, Parving HH. Short stature and diabetic nephropathy. Bmj 1995; 310:296-7.
172. Jacobsen P, Rossing K, Tarnow L, et al. Progression of diabetic nephropathy in normotensive type 1 diabetic patients. Kidney Int Suppl 1999; 71:S101-5.
173. Antonov KI, Isacson DG. Prescription and nonprescription analgesic use in Sweden. Ann Pharmacother 1998; 32:485-94.
174. SOS. (2004) Socialstyrelsen website; statistik över läkemedelsförsäljningen 2003 (accessed Jan. 2., 2005. at http://www.socialstyrelsen.se/NR/rdonlyres/E3E02806-7A12-4495-B9CF-FEED4C2FCE58/2888/2004462.pdf).
175. Spuhler O, Zollinger HO. Die chronish-interstitielle Nephritis. Zeit Klin Med, 1953:1-50.
176. Burry AF. A profile of renal disease in Queensland: results of an autopsy survey. Med J Aust 1966; 1:826-34.
177. Hultengren N. Renal papillary necrosis. A clinical study of 103 cases. Acta Chir Scand 1961; Suppl:1-84.
178. Dubach UC, Levy PS, Minder F. Epidemiological study of analgesic intake and its relationship to urinary tract disorders in Switzerland. Helv Med Acta 1968; 34:297-312.
179. Grimlund K. Phenacetin and Renal Damage at a Swedish Factory. Acta Med Scand 1963; 174:SUPPL405:1-26.
180. Prescott LF. Analgesic nephropathy: a reassessment of the role of phenacetin and other analgesics. Drugs 1982; 23:75-149.
181. Kincaid-Smith P. Pathogenesis of the renal lesion associated with the abuse of analgesics. Lancet 1967; 1:859-62.
182. Griffin MD, Bergstralhn EJ, Larson TS. Renal papillary necrosis--a sixteen-year clinical experience. J Am Soc Nephrol 1995; 6:248-56.
183. Wing AJ, Brunner FP, Geerlings W, et al. Contribution of toxic nephropathies to end-stage renal failure in Europe: a report from the EDTA-ERA registry. Toxicol Lett 1989; 46:281-92.
184. USRDS, System USRD, Bethesda M, US Department of Health and Human services. National Institutes of Health. Annual Data report. 1997:p 30.

185. Elseviers MM, De Schepper A, Corthouts R, et al. High diagnostic performance of CT scan for analgesic nephropathy in patients with incipient to severe renal failure. Kidney Int 1995; 48:1316-23.
186. Schnuelle P, Van Der Woude FJ. Analgesics and renal disease in the postphenacetin era. Am J Kidney Dis 2003; 42:385-7.
187. Elseviers MM, De Broe ME. Combination analgesic involvement in the pathogenesis of analgesic nephropathy: the European perspective. Am J Kidney Dis 1996; 28:S48-55.
188. Kincaid-Smith P. Effects of non-narcotic analgesics on the kidney. Drugs 1986; 32:109-28.
189. Buckalew VM, Jr. Habitual use of acetaminophen as a risk factor for chronic renal failure: a comparison with phenacetin. Am J Kidney Dis 1996; 28:S7-13.
190. Matzke GR. Clinical consequences of nonnarcotic analgesic use. Ann Pharmacother 1997; 31:245-8.
191. Porter GA. Acetaminophen/aspirin mixtures: experimental data. Am J Kidney Dis 1996; 28:S30-3.
192. Henrich WL, Agodoa LE, Barrett B, et al. Analgesics and the kidney: summary and recommendations to the Scientific Advisory Board of the National Kidney Foundation from an Ad Hoc Committee of the National Kidney Foundation. Am J Kidney Dis 1996; 27:162-5.
193. Michielsen P, de Schepper P. Trends of analgesic nephropathy in two high-endemic regions with different legislation. J Am Soc Nephrol 2001; 12:550-6.
194. Feinstein AR, Heinemann LA, Curhan GC, et al. Relationship between nonphenacetin combined analgesics and nephropathy: a review. Ad Hoc Committee of the International Study Group on Analgesics and Nephropathy. Kidney Int 2000; 58:2259-64.
195. Gambaro G, Perazella MA. Adverse renal effects of anti-inflammatory agents: evaluation of selective and nonselective cyclooxygenase inhibitors. J Intern Med 2003; 253:643-52.
196. Sandler DP, Smith JC, Weinberg CR, et al. Analgesic use and chronic renal disease. N Engl J Med 1989; 320:1238-43.
197. Morlans M, Laporte JR, Vidal X, Cabeza D, Stolley PD. End-stage renal disease and non-narcotic analgesics: a case-control study. Br J Clin Pharmacol 1990; 30:717-23.
198. Perneger TV, Whelton PK, Klag MJ. Risk of kidney failure associated with the use of acetaminophen, aspirin, and nonsteroidal antiinflammatory drugs. N Engl J Med 1994; 331:1675-9.
199. Pommer W, Bronder E, Greiser E, et al. Regular analgesic intake and the risk of end-stage renal failure. Am J Nephrol 1989; 9:403-12.
200. Murray TG, Stolley PD, Anthony JC, Schinnar R, Hepler-Smith E, Jeffreys JL. Epidemiologic study of regular analgesic use and end-stage renal disease. Arch Intern Med 1983; 143:1687-93.
201. Kurth T, Glynn RJ, Walker AM, et al. Analgesic use and change in kidney function in apparently healthy men. Am J Kidney Dis 2003; 42:234-44.
202. Duggin GG. Combination analgesic-induced kidney disease: the Australian experience. Am J Kidney Dis 1996; 28:S39-47.
203. Gault MH, Barrett BJ. Analgesic nephropathy. Am J Kidney Dis 1998; 32:351-60.

204. Sabatini S. Pathophysiologic mechanisms in analgesic-induced papillary necrosis. Am J Kidney Dis 1996; 28:S34-8.
205. Byrne C, Nedelman J, Luke RG. Race, socioeconomic status, and the development of end-stage renal disease. Am J Kidney Dis 1994; 23:16-22.
206. Young EW, Mauger EA, Jiang KH, Port FK, Wolfe RA. Socioeconomic status and end-stage renal disease in the United States. Kidney Int 1994; 45:907-11.
207. Perneger TV, Whelton PK, Klag MJ. Race and end-stage renal disease. Socioeconomic status and access to health care as mediating factors. Arch Intern Med 1995; 155:1201-8.
208. Fored CM, Ejerblad E, Fryzek JP, et al. Socio-economic status and chronic renal failure: a population-based case-control study in Sweden. Nephrol Dial Transplant 2003; 18:82-8.
209. de Broe ME, D'Haese PC, Nuyts GD, Elseviers MM. Occupational renal diseases. Curr Opin Nephrol Hypertens 1996; 5:114-21.
210. Ravnskov U. Hydrocarbon exposure may cause glomerulonephritis and worsen renal function: evidence based on Hill's criteria for causality. Qjm 2000; 93:551-6.
211. Wedeen RP. Occupational and environmental renal disease. Semin Nephrol 1997; 17:46-53.
212. Jarup L. Cadmium overload and toxicity. Nephrol Dial Transplant 2002; 17:35-9.
213. Hellstrom L, Elinder CG, Dahlberg B, et al. Cadmium exposure and end-stage renal disease. Am J Kidney Dis 2001; 38:1001-8.
214. Fored CM, Nise G, Ejerblad E, et al. Absence of association between organic solvent exposure and risk of chronic renal failure: a nationwide population-based case-control study. J Am Soc Nephrol 2004; 15:180-6.
215. Fored CM. Dissertation; Risk factors for the development of chronic renal failure. Epidemiological studies on the role of analgesic use, occupational exposures and socioeconomic background. Department of Medical Epidemiology. Stockholm: Karolinska Institutet, 2003.
216. Samuelsson O, Mulec H, Knight-Gibson C, et al. Lipoprotein abnormalities are associated with increased rate of progression of human chronic renal insufficiency. Nephrol Dial Transplant 1997; 12:1908-15.
217. Cusick M, Chew EY, Hoogwerf B, et al. Risk factors for renal replacement therapy in the Early Treatment Diabetic Retinopathy Study (ETDRS), Early Treatment Diabetic Retinopathy Study Report No. 26. Kidney Int 2004; 66:1173-9.
218. Castello IB. Hyperlipidemia: a risk factor for chronic allograft dysfunction. Kidney Int Suppl 2002:73-7.
219. Muntner P, Coresh J, Smith JC, Eckfeldt J, Klag MJ. Plasma lipids and risk of developing renal dysfunction: the atherosclerosis risk in communities study. Kidney Int 2000; 58:293-301.
220. Schaeffner ES, Kurth T, Curhan GC, et al. Cholesterol and the risk of renal dysfunction in apparently healthy men. J Am Soc Nephrol 2003; 14:2084-91.
221. Buraczynska M, Ksiazek A. Searching for a genetic risk profile in end-stage renal disease. Med Sci Monit 2001; 7:1376-80.
222. Freedman BI. Susceptibility genes for hypertension and renal failure. J Am Soc Nephrol 2003; 14:S192-4.

223. Winn MP. Approach to the evaluation of heritable diseases and update on familial focal segmental glomerulosclerosis. Nephrol Dial Transplant 2003; 18:vi14-20.
224. Bowden DW, Colicigno CJ, Langefeld CD, et al. A genome scan for diabetic nephropathy in African Americans. Kidney Int 2004; 66:1517-26.
225. SEI. Swedish socioeconomic classification (English summary). Reports on Statistical Co-ordination 1982:4. Statistics Sweden: Stockholm, 1982.
226. Breslow NE, Day NE. The analyses of case-control studies. Statistical methods in cancer research. Vol 1. Lyon: International Agency for Research on Cancer, 1980:192-246.
227. National Food Administration U. Vikttabeller för livsmedel och maträtter,. 1988.
228. Willett W. Nutritional epidemiology, 2nd. ed. Oxford University Press, New York 1998.
229. Foley RN, Herzog CA, Collins AJ. Smoking and cardiovascular outcomes in dialysis patients: the United States Renal Data System Wave 2 study. Kidney Int 2003; 63:1462-7.
230. Patrick DL, Cheadle A, Thompson DC, Diehr P, Koepsell T, Kinne S. The validity of self-reported smoking: a review and meta-analysis. Am J Public Health 1994; 84:1086-93.
231. Troy LM, Michels KB, Hunter DJ, et al. Self-reported birthweight and history of having been breastfed among younger women: an assessment of validity. Int J Epidemiol 1996; 25:122-7.
232. Tamakoshi K, Yatsuya H, Kondo T, et al. The accuracy of long-term recall of past body weight in Japanese adult men. Int J Obes Relat Metab Disord 2003; 27:247-52.
233. Kuskowska-Wolk A, Karlsson P, Stolt M, Rossner S. The predictive validity of body mass index based on self-reported weight and height. Int J Obes 1989; 13:441-53.
234. West S, Strom B. Validity of pharmacoepidemiology drug and diagnosis data, in Pharmacoepidemiology, edited by Strom BL, Chichester, john Wiley and Sons Ltd., 1994, pp 549-580, 1994.
235. Orth SR, Ritz E. Adverse effect of smoking on renal function in the general population: are men at higher risk? Am J Kidney Dis 2002; 40:864-6.
236. Haroun MK, Jaar BG, Hoffman SC, Comstock GW, Klag MJ, Coresh J. Risk factors for chronic kidney disease: a prospective study of 23,534 men and women in Washington County, Maryland. J Am Soc Nephrol 2003; 14:2934-41.
237. Appel RG, Bleyer AJ, Reavis S, Hansen KJ. Renovascular disease in older patients beginning renal replacement therapy. Kidney Int 1995; 48:171-6.
238. Lhotta K, Rumpelt HJ, Konig P, Mayer G, Kronenberg F. Cigarette smoking and vascular pathology in renal biopsies. Kidney Int 2002; 61:648-54.
239. Black HR, Zeevi GR, Silten RM, Walker Smith GJ. Effect of heavy cigarette smoking on renal and myocardial arterioles. Nephron 1983; 34:173-9.
240. Oberai B, Adams CW, High OB. Myocardial and renal arteriolar thickening in cigarette smokers. Atherosclerosis 1984; 52:185-90.
241. Blann AD, McCollum CN. Adverse influence of cigarette smoking on the endothelium. Thromb Haemost 1993; 70:707-11.

242. Cucina A, Sapienza P, Corvino V, et al. Nicotine-induced smooth muscle cell proliferation is mediated through bFGF and TGF-beta 1. Surgery 2000; 127:316-22.
243. Wesson DE. The relationship of cigarette smoking to end-stage renal disease. Semin Nephrol 2003; 23:317-22.
244. Grassi G, Seravalle G, Calhoun DA, et al. Mechanisms responsible for sympathetic activation by cigarette smoking in humans. Circulation 1994; 90:248-53.
245. Gupta R, Gurm H, Bartholomew JR. Smokeless tobacco and cardiovascular risk. Arch Intern Med 2004; 164:1845-9.
246. Hergens MP, Ahlbom A, Andersson T, Pershagen G. Swedish moist snuff and myocardial infarction among men. Epidemiology 2005; 16:12-6.
247. O'Bryan GT, Hostetter TH. The renal hemodynamic basis of diabetic nephropathy. Semin Nephrol 1997; 17:93-100.
248. Vaziri ND. Roles of oxidative stress and antioxidant therapy in chronic kidney disease and hypertension. Curr Opin Nephrol Hypertens 2004; 13:93-9.
249. Moller P, Wallin H, Knudsen LE. Oxidative stress associated with exercise, psychological stress and life-style factors. Chem Biol Interact 1996; 102:17-36.
250. Loscalzo J. The oxidant stress of hyperhomocyst(e)inemia. J Clin Invest 1996; 98:5-7.
251. Hall JE, Jones DW, Kuo JJ, da Silva A, Tallam LS, Liu J. Impact of the obesity epidemic on hypertension and renal disease. Curr Hypertens Rep 2003; 5:386-92.
252. Kasiske BL, Crosson JT. Renal disease in patients with massive obesity. Arch Intern Med 1986; 146:1105-9.
253. Kambham N, Markowitz GS, Valeri AM, Lin J, D'Agati VD. Obesity-related glomerulopathy: an emerging epidemic. Kidney Int 2001; 59:1498-509.
254. Praga M, Morales E, Herrero JC, et al. Absence of hypoalbuminemia despite massive proteinuria in focal segmental glomerulosclerosis secondary to hyperfiltration. Am J Kidney Dis 1999; 33:52-8.
255. Yudkin JS, Phillips DI, Stanner S. Proteinuria and progressive renal disease: birth weight and microalbuminuria. Nephrol Dial Transplant 1997; 12:10-3.
256. McLaughlin JK, Lipworth L, Chow WH, Blot WJ. Analgesic use and chronic renal failure: a critical review of the epidemiologic literature. Kidney Int 1998; 54:679-86.
257. Mackinnon B, Boulton-Jones M, McLaughlin K. Analgesic-associated nephropathy in the West of Scotland: a 12-year observational study. Nephrol Dial Transplant 2003; 18:1800-5.