Animal-friendly production systems may cause re-emergence of Toxoplasma gondii

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Abstract

Toxoplasmosis is still one of the most common parasitic infections in the world, although in Europe improvements in hygiene and the introduction of 'total' indoor farming in livestock production have rapidly diminished the problem during the past decades. As a result of public dislike, however, introduction of alternative and more acceptable animal-friendly livestock production systems including outdoor access are gaining ground. Potentially these systems can lead to increased prevalence of certain zoonotic diseases, including Toxoplasmosis. To retain prevalence of this disease in humans at current levels, emphasis should be on disease control at farm-level. This article provides an analysis of various risk factors for farm animals to get infected with *Toxoplasma gondii*. Access of cats to the farm premises, the use of compost and goat whey, and rodent control were identified as possible risk factors that should be addressed. Consumers should be aware of the fact that *Toxoplasma* infection, besides through meat, can also be caused by the uptake of contaminated water, soil, fruit and vegetables.

Additional keywords: organic production, risk factors, rodents, parasitic infections

Introduction

Toxoplasmosis, caused by *Toxoplasma gondii*, is currently the most prevalent parasitic zoonotic disease throughout the world (Tenter *et al.*, 2000). It is an important cause of abortion in humans and livestock (sheep) and was recently shown to be the third cause of death following food-borne illnesses (Mead *et al.*, 1999). In humans it is further known to cause mental retardation, encephalitis and blindness. Although not fatal, ocular Toxoplasmosis is probably the most frequently occurring complication of this

disease (Holland, 2003) whereby eventually 24% of the affected persons become blind (Bosch-Driessen *et al.*, 2002). Over the years a large number of anti-parasitic drugs have been developed to treat patients with Toxoplasmosis, with good results under certain conditions. However, the drugs developed so far were not always effective in the treatment of ocular Toxoplasmosis (Stanford *et al.*, 2003). So at present, prevention of *Toxoplasma* infection is the only strategy to combat the blinding complications of Toxoplasmosis. Since meat consumption is one of the main risk factors, prevention should also be aimed at livestock production.

Over the past decades animal farming in Western Europe has drastically changed. Some of these changes are not in favour of animal welfare and have led to alternative production systems such as organic animal husbandry. At this moment, the impact of these animal-friendly production systems on certain zoonotic diseases such as Toxoplasmosis is not clear. In this short review we shall discuss risk factors for an animal to become infected with *Toxoplasma* and present a risk analysis with a number of control points to limit the on-farm risk.

Parasitology

Toxoplasma gondii is a ubiquitous protozoan parasite capable of infecting virtually all warm-blooded vertebrates in the world. It is an obligate intracellular organism belonging to the Coccidian family. Three strains of *Toxoplasma gondii* have been defined, (type I, type II and type III), of which type I is extremely virulent for mice and type II has been associated with the majority of Toxoplasmosis cases in AIDS patients (Boothroyd & Grigg, 2002; Klaren & Kijlstra, 2002). Type III is present in animals and has been detected in AIDS patients, but does not seem to be associated with ocular Toxoplasmosis (Boothroyd & Grigg, 2002).

The parasite has a complex life cycle whereby Felidae (cats) function as the definitive host, i.e., the sexual part of the life cycle takes place in these animals. Fusion of gametocytes and zygotes takes place in the gut of catlike animals leading to the formation of eggs (oocysts). Cats have been shown to shed millions of oocysts via their faeces into the environment during a period of a few weeks (Dubey, 2001). Oocysts must mature (sporulate) for I–5 days to become infective for other hosts. Sporulated oocysts can remain in a moist environment for some years (Dubey & Beattie, 1988) since they are resistant to a large number of threats such as heat and cold. Further prolongation of the oocyst's lifetime may be due to the uptake by other organisms in the soil or water. Some West European countries (e.g. the Netherlands, Belgium, United Kingdom, Germany and Denmark) have optimum climatic conditions for a parasite like *Toxoplasma gondii* to thrive, because of their moist summers and generally temperate winters. As they also have a large population of pet cats, it is not remarkable that Toxoplasmosis is now recognized as a serious health problem in these countries.

When an intermediate host ingests a sporulated *Toxoplasma* oocyst the parasite transforms into a stage called the tachyzoite. Tachyzoites can infect virtually any nucleated cell type, although a tropism for certain cell types (for instance retinal vascular endothelial cells) has been reported (Smith *et al.*, 2004). After invading a cell, the

tachyzoites can rapidly divide and after death of the host cell they will invade adjacent cells or – after traveling through the blood stream – attach to cells elsewhere in the body. The tachyzoite stage can transform into a slowly dividing bradyzoite. This stage of the parasite is able to form a cyst wall around a large family of dividing parasites, thereby protecting the parasite against the mounting immune response of the host. The stage differentiation from tachyzoite to bradyzoite is thought to be triggered by certain cytokines of the cellular host immune response (Klaren & Kijlstra, 2002). The parasite can remain dormant in its encysted stage whereby release of parasites from the cyst is associated with a weaning cellular immune response (Nath & Sinai, 2001). The cysts have a predilection for certain sites in the host such as muscle, brain or retinal tissues. Ingesting tissues of an intermediate host containing tissue cysts infects carnivorous and omnivorous animals. This is also the route whereby cats (and other Felidae) become infected, thereby closing the life cycle of the parasite.

Congenital transmission is a unique method that this parasite has developed to maintain an infectious reservoir in certain intermediate hosts. During pregnancy the tachyzoites can transfer the placenta and infect the developing offspring. In some species this can occur during successive pregnancies (Owen & Trees, 1998; Webster, 2001; Marshall *et al.*, 2004). In humans, congenital transmission is thought to be a one-time event, occurring when a pregnant woman becomes infected with the parasite for the first time in her life. For sheep and rodents, evidence has been reported indicating that this may occur during successive pregnancies. Although congenital transmission has been reported in pigs, it is not known whether transmission can occur during multiple pregnancies. In rodents, the congenitally infected offspring may transfer the disease to their offspring leading to a long-lasting reservoir of *Toxoplasma gondii* even in the absence of Felidae as definitive host (Webster, 1994). In immuno-competent humans a primary infection is followed by a lifelong immunity causing the parasite to remain in its encysted stage during lifetime, preventing transfer of infection to the fetus in women who have encountered the infection prior to pregnancy.

Epidemiology

Due to the ubiquitous presence of the parasite, infection in humans is quite common. In some populations up to 100% of the individuals have been shown to be seropositive for *Toxoplasma* (Tenter, 2000). Prevalence rates differ, depending amongst other things on the environmental conditions of oocyst survival. Prevalence of *Toxoplasma* is high in humid tropical areas and low in hot and dry areas. Prevalence in cold areas is also low. The prevalence of infection in a number of European countries (e.g. United Kingdom, Sweden and the Netherlands) shows a rapid decline over the past years (Walker *et al.*, 1992; Nokes *et al.*, 1993). A study performed in the Netherlands in 1987 showed that 50% of the population aged 30–34 years had experienced a previous *Toxoplasma* infection, whereas a study performed in 1996 showed that 37% of this age group was seropositive (Figure 1; L.M. Kortbeek, unpublished results). It is not clear why prevalence has dropped over the past years. It may be due to a change in consumption patterns, food handling and outdoor activities. On the other hand, the



Figure 1. *Toxoplasma* seroprevalence in the Dutch population by age groups in the years 1987 (n = 28,000) and 1996 (n = 7521).



Figure 2. Toxoplasma infection in Dutch slaughter pigs in the period 1969–2001.

decreased prevalence in the human population parallels a decrease in *Toxoplasma* infection rates observed in pigs (Figure 2). The change to intensive farm management practices whereby animals are confined within buildings may have contributed to a decrease in *Toxoplasma* seroprevalence. This in turn may have led to fewer consumers becoming infected.

Risk factors

Risk factors for both humans and livestock include the ingestion of fruit, vegetables, soil or water contaminated with sporulated oocysts shed into the environment by cats during a few weeks following infection (Tenter, 2000). It is assumed that the latter only occurs once in the lifetime of a cat. Prevalence studies in European cats have shown that approximately 50% of the cats have experienced a *Toxoplasma* infection, indicating that these pets are the cause of a serious environmental burden of parasites (Webster, 2001). Disposal of cat litter boxes via the toilet or into compost garbage collection systems may pose as yet unknown environmental problems. Infection of marine mammals along the west coast of the United States is thought to be due to the outlet into the Pacific Ocean of sewage systems containing cat box litter disposed via home toilets (Dubey *et al.*, 2003b). Compost production systems may not reach temperatures high enough to kill sporulated oocysts. Compost, which is used by a few pig farms to improve iron uptake (I.A.J.M. Eijck, personal communication), may be involved in the transfer of Toxoplasmosis to pigs.

The main risk factor for humans to contract Toxoplasmosis is the consumption of raw or undercooked meat from animals that have been previously infected with the parasite (Cook et al., 2000). It is not known how many tissue cysts result in the infection of human beings, but ingestion of one cyst (containing hundreds of bradyzoites) is sufficient for a cat to become infected. Pigs, goats, sheep and poultry are the major meat sources of human infection (Tenter, 2000; Aspinall et al., 2002;). One pig may be consumed by 200–400 different individuals so that there is a tremendous amplification of the risk to become infected (Fehlhaber, 2001). During the production of various meat products, meat of many animals is mixed, which also amplifies the risk in cases where only a few animals would be infected (Aspinall et al., 2002). Of interest is the fact that beef is not an important source of human infection (Dubey & Beattie, 1988). Although cattle can become infected with Toxoplasma gondii, this does not result in the appearance of infectious tissue cysts in their meat. Epidemiological studies in Europe have indicated that meat consumption could account for almost 60% of the Toxoplasma infections, whereas contact with soil (gardening) may be held responsible for approximately 20% of the cases (Cook et al., 2000). Kitchen hygiene with respect to the handling of meat may also be a risk factor for humans to contract Toxoplasmosis. Examples include the use of the same knife to cut raw meat and subsequently cut fresh salad or the tasting of raw minced meat during flavouring with salt and pepper (Kapperud et al., 1996; Tenter et al., 2000).

Toxoplasma gondii can also be transmitted via the milk of infected goats (Riemann *et al.*, 1975; Tenter *et al.*, 2000). It is not known whether goat cheese prepared from animals shedding the parasite is still infectious. Byproducts of goat cheese processing (whey) are sometimes fed to pigs, which may potentially lead to the transfer of *Toxoplasma* to these animals.

Toxoplasma gondii is effectively killed by heating at temperatures above 67 °C for a few minutes (Dubey, 2000). Overnight freezing at –12 °C also kills the majority of tissue cysts (Kotula *et al.*, 1991), whereas the curing of meat with salt does not seem to

affect the parasite immediately (Dubey, 2000). Irradiation with low dosages of Cesium 137 can also be a suitable method to destroy the parasite (Dubey *et al.*, 1998).

Animal-friendly production systems

Indoor housing of farm animals is not regarded as being in favour of animal welfare, and due to social pressure the bioindustry in Western Europe is urged to re-introduce outdoor housing. In the Netherlands different animal-friendly pig-production systems have therefore been introduced. On modern intensive farms following a high hygiene protocol, pigs are housed indoors mostly on concrete-slatted floors and are fed regular pelleted pig feed. So-called free-range pigs are allowed outdoor access, are given straw bedding and are also fed regular pelleted pig feed. Pigs from organic farms are kept according to regulations set up by the European Union (EU regulation nr 2092/91), which includes outdoor access, straw bedding and 'organic' pig feed. Organic pig feed often contains the same (plant) ingredients as regular pig feed, but is grown on farms that do not use inorganic fertilizers or pesticides. Since the BSE crisis, in the European Union it is not allowed to feed pigs with products of 'animal' origin. So pigs can be considered as vegetarians, although they are in fact omnivorous.

Whether animal-friendly production systems lead to a re-emergence of Toxoplasma infections is not yet known. This question was therefore the subject of research performed in the past few years by the Animal Sciences Group of Wageningen University and Research Centre. Results show that indoor housed pigs were free from Toxoplasma infection whereas almost 3% of the animals raised in animal-friendly production systems had previously been infected with Toxoplasma (Kijlstra et al., 2004). On 70% of the organic farms tested, sows were seropositive whereby per farm on average 15% of the sows showed evidence of a previous *Toxoplasma* infection (I.A.J.M. Eijck; personal communication). The source of infection is not exactly known yet, but may include ingestion of sporulated oocysts deposited in the environment via cats. Pigs are known to catch rats and mice and earlier research concerning risk factors for Toxoplas*ma* infection on pig farms has already shown that poor rodent control is involved (Weigel et al., 1995). Our own field studies confirm these observations. All conventional pig farmers use chemical rodenticides, whereas up to 30% of the farmers on animal-friendly farms do not, but rather rely on the use of cats for rodent control on their premises. An analysis of risk factors and possible means of controlling Toxoplasma infection on organic pig farms based on HACCP methodology is presented in the Appendix.

Since cats have been recognized as an important risk factor (Dubey *et al.*, 1995a), the effect was studied of *Toxoplasma* vaccination of farm cats on *Toxoplasma* seroprevalence in pigs. Some time after the introduction of the cat vaccine, the seroprevalence showed a small but statistically significant decrease in the seroprevalence of pigs where cats had been vaccinated (Mateus-Pinilla *et al.*, 1999). The small change could point to other more important risk factors, such as transfer via the ingestion of rodents. As will be mentioned later, rodents can be an important reservoir of the parasite even when cats had been absent for a long period of time (Webster, 2001).

Chickens are also a potential source of *Toxoplasma* infection in humans (Tenter *et al.*, 2000). Most chickens used for consumption are raised indoors and probably do not have access to a source of *Toxoplasma* infection during their short lifetime. Organic chickens raised for meat production are allowed outdoor access and are slaughtered at an older age. Due to their longer life and due to the fact that they have access to various *Toxoplasma* sources, organic chickens could potentially become infected with *Toxoplasma*. So far this has not been studied but recent data show that a large percentage of free-ranging chickens have been infected with the parasite (Dubey *et al.*, 2003a). At present there are no epidemiological data supporting the hypothesis that 'animal friendly' farming will have an impact on the incidence of human Toxoplasmosis. But this is also because at present the market share of organic meat is only a few percent.

Toxoplasma infection in rodents: prevalences and routes of transmission

Many studies have focused on the prevalence of *Toxoplasma* infection in wildlife animals. In this section we shall confine ourselves to rodents that may play a role in the transmission of infection to farm animals like pigs and poultry.

From field studies conducted on pig farms in Illinois (USA) during 1992 and 1993 it was learned that 2.1% of the house mice (*Mus musculus*) were seropositive for *Toxoplasma* (Dubey *et al.*, 1995b). Infectious parasites were recovered from heart or brain tissue in 0.5% of the mice investigated. Sera from Missouri and Kansas (USA) collected in the period December 1974 – December 1987 and analysed for the presence of antibodies to *Toxoplasma gondii* showed a low prevalence (3%) of antibodies in mice (*Mus musculus* and *Peromyscus* spp.) and rats (*Rattus norvegicus* and *Sigmodon hispidus*), while medium-sized herbivores, like squirrels (*Sciurus* spp.), rabbits (*Sylvilagus floridanus*) and muskrats (*Ondatra zibethicus*) had prevalences of about 18% (Smith & Frenkel, 1995). Webster (1994) studied the prevalence of *Toxoplasma gondii* within 6 UK farmstead wild rat populations (*Rattus norvegicus*) and reported a mean prevalence of 35%. No statistically significant age, sex or site differences were observed in prevalence between or within populations irrespective of habitat type or presence of cats.

The prevalence of *Toxoplasma gondii* in the Czech Republic was < 1% in insectivores (n = 578), 12% in carnivores (n = 112), 1% in rodents except muskrats (*Ondatra zibethicus*) (n = 5163), 24% in muskrats (n = 437), 5% in lagomorphs (n = 293), 0% in ruminants (n = 456) and 2% in wild boars (*Sus scrofa*) (n = 136) (Hejlicek *et al.*, 1997). Another study from the Czech Republic showed *Toxoplasma* infection in 47% of the muskrats from a site with water heavily polluted with municipal wastes and 9% in muskrats from 3 sites with water slightly polluted with wastes, stressing the role of waste water as a source of *Toxoplasma* infection (Nezval & Literak, 1994).

A study from Ontario (Canada) showed that 11% of the mice (*Mus musculus*), 5% of the deer mice (*Peromyscus*), 3% of the rats (*Rattus norvegicus*) and less than 2% of the sparrows (*Passer domesticus*) investigated were seropositive. All samples from short-tailed field mice (*Microtus pennsylvanicus*), squirrels (*Sciurus carolinensis*), chipmunks (*Tamias striatus*), meadow jumping mice (*Zapus hudsonius*) and starlings (*Sturnus*)

vulgaris) were seronegative (Tizard et al., 1978).

How wild rodent populations become infected with *Toxoplasma gondii* is not exactly known, but congenital transmission may perpetuate the infection over successive generations. Webster (2001) concluded that this mode of transmission is the predominant route for *Rattus norvegicus*. Marshall *et al.* (2004) presented evidence that 75% of the transmission in the house mouse (*Mus musculus*) also occurs via the congenital route (Owen & Trees, 1998). Also studies in sheep favour this mode of transmission (Duncanson *et al.*, 2001).

Toxoplasma gondii has been shown to affect the behaviour of its intermediate host, the rat: its chance of being predated by cats is increased. Rats, which normally avoid areas with cat scent, become attracted to cat urine odour following infection with *Toxoplasma gondii* (Berdoy *et al.*, 2000).

Conclusions

Toxoplasmosis is an important parasitic disease worldwide causing substantial health problems in humans as well as in farm animals. Currently no vaccine is available and anti-parasitic drugs are either not effective (ocular Toxoplasmosis) or are associated with serious adverse side-effects. This means that prevention is currently the best method to manage the disease. Since the majority of human infections are due to the consumption of meat, emphasis should be on control of the disease at farm level or on implementing measures to test animals before the meat reaches the consumer. Indoor housing of farm animals as practised in intensive animal husbandry has resulted in the abrogation of the *Toxoplasma* problem in the pig industry. However, animal-friendly livestock-production systems are associated with a potentially higher prevalence of *Toxoplasma* infections and extra attention is needed to control transfer of infection on these farms. The access of cats to the farm premises, the use of compost and goat whey, and rodent control have been identified as risk factors that should be addressed on these farms.

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Risk analysis of Toxoplasma gondii infection on organic pig-production facilities.

Part	Hazard	Chance ¹	Severity ²	Motivation	Control measure
Environment	Introduction of <i>Toxoplasma</i> due to pigs rooting in the earth	н	0	Oocysts can survive in soil for prolonged periods of time, after cat has shed infected faeces on the pasture. Not all pigs will become infected.	 Limit the number of cats on the farm No kittens Vaccination of the cats Sterilization of the cats Keep cats away from pasture Take male cat to defend farm area
	Introduction of <i>Toxoplasma</i> by pig due to uptake of dead or live worms or flies	г	ц	Uptake by a pig of worms and flies infected with <i>Toxoplasma</i> is a seldom event. Only an individual pig may become infected.	against cats from neighbours - Fly control
	The pig may become infected with <i>Toxoplasma</i> by eating an infected dead mouse	7	н	The chance of a pig eating a dead mouse can occur a few times per year. It is not known how many mice are infected with <i>Toxoplasma</i> (estimated a few percent). Only an individual may become infected.	 Rodent control Use of live traps to prevent dead animals in the outdoor area or in the pigsty.
	Introduction of <i>Toxoplasma</i> on the farm via a live mouse or a dead or live bird.	I	ц	The chance of a pig eating a live mouse or a dead or live bird is small. If it does occur only an individual animal may become infected.	- Rodent control - Improvement of farm hygiene
	Introduction of <i>Toxoplasma</i> by pig via cat faeces	0	0	In the Netherlands there are always cats in the local farm environment. The defaecation of cats on the outdoor area may be limited. On the other hand, at least 50% of the cats will have shed oocysts in their lifetime. Shed oocysts can persist for years. Many pigs can take up these oocysts, indicating that several pigs on the farm may become infected.	 Limit the number of cats on the farm No kittens Vaccination of cats Sterilization of cats Sterilization of cats Tate male cat to defend farm area avainst cats from neichbours
Management	Infection of pigs with <i>Toxoplasma</i> through application of compost (iron supplementation)	7	7	Compost may contain litter or faeces from cat-boxes, compost may contain litter or faeces from cat-boxes, possibly containing oocysts. Not all parts of compost reach temperatures high enough to inactivate oocysts. Since piglets are sometimes given compost, they are at risk of becoming infected with <i>Toxoplasma</i> via this route.	According to Dutch farming regulations, pig farmers are forbidden to use compost during pig production

Part	Hazard	Chance	Severity	Motivation	Control measure
Water	Pigs can become infected with <i>Toxoplasma</i> by drinking water contaminated with oocysts	0	ñ	If pigs drink water from ponds, ditches or canals there is a fair chance of them becoming infected with <i>Toxoplasma</i> . Cats can drop faeces in the neighbour- hood of ponds canals and ditches, which may drain into the water following rainfall. Whether this can occur in the Netherlands is not known. All pigs having access to this water source may become infected.	- Do not allow access to water from ponds, canals or ditches
	Pigs can become infected by drinking water from an infected well Spreading of <i>Toxoplasma</i> due	г	~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~	The chance of infection of a well is small if the well is deep. If the well does become infected then nearly all pigs on the farm will become infected. The chance of an infected mouse drowning in a water	- Test well water - Close the water reservoir
	to infected insects or rodents getting into a local intermediate water reservoir			reservoir is low. The chance of insects drowning in a water reservoir is higher. The number of oocysts in insects is not exactly known and probably very small. Due to dilution in the water the overall chance of a pig becoming infected via this route is small. The severity may be large because the water is supplied to various pens and many animals may become infected.	
	Pigs become infected due to a dead bird in a local intermedite water reservoir Dice become infected with	ц	н с	The chance of an infected bird drowning in a water reservoir is low. The cysts will probably not leave the dead bird and thus will not infect the pigs.	- Close the water reservoir Eased and users should another
Leeu	Toxoplasma because the feed is contaminated with cat faeces or dead infected rodents	-	Ś	reneted pig recurb thate at high chiper attracts and under high pressure. This limits the chance of oocysts present in the original ingredients to survive. If temperatures are too low and feed ingredients are contaminated, infectious oocysts may remain in the feed leading to several pigs becoming infected.	 recu producers should guarance temperatures during feed processing of at least 65 °C during 5 minutes
	Pigs become infected with <i>Toxoplasma</i> because cat faeces or dead rodents enter feeding system	7	<i>c</i>	If the faeces are from an infected cat or if the rodent is infected, several pigs may become infected	 No access of cats to feeding system No access of rodents to feeding system Farm hygiene

Appendix (cont'd)

Feed	Pigs become infected with	Ι	3	If infected cats can drop faeces onto stored feed	- No access of cats or rodents to farm
	Toxoplasma because non-			products or if infected rodents have access to these	storage sites of by-products or other
	processed feed (by-products			products (and can die there), this can lead to	feed ingredients
	and individual ingredients,			contamination of the feed products with both oocysts	- Feed producers should guarantee a
	including hay and straw) are			and (rodent tissue) cysts. Several animals can become	cat- and rodent-free storage system
	contaminated with cat faeces			infected if fed with these products.	(including control system)
	or dead rodents				
	Pigs become infected with	I	3	Some pig farmers feed organic goat-whey to their pigs.	- Forbid feeding goat-whey
	Toxoplasma because they are			If whey is obtained from Toxoplasma-infected goats,	
	fed (infected) goat-whey			the whey can contain Toxoplasma tachyzoites. This	
				may lead to infection of all animals on the farm.	
Piglets	External supply of piglets	7	2	Piglets can become congenitally infected during	- The supply of piglets should come
	may introduce <i>Toxoplasma</i>			pregnancy or during the lactation period. Transport	from Toxoplasma-free farms
	on the farm			of such pigs to a finishing farm will lead to the	
				presence of infected slaughter pigs. Not all pigs will	
				become infected.	
	Piglets become infected due	7	I	There is a chance of Toxoplasma infection being	- Prevent cannibalism by introducing
	to cannibalism			transmitted through cannibalism. Cannibalism of ears	distracting elements (playing material)
				or tails containing infectious Toxoplasma cysts may	
				lead to the infection of other piglets. A few piglets	
				will become infected via this route.	
Instruments	Pigs can become infected	2	Ι	The chance of instruments or tools on the farm	- The use of clean instruments and
and tools	with Toxoplasma due to the			being infected with oocysts (cat faeces or dead rodents)	tools
	presence of oocysts or cysts			is very small. Carry-over of cysts via instruments	
	on instruments and tools			(castration or vaccination) is low too. Only an	
				individual animal may become infected.	
Visitor and	Pigs can become infected	I	Ι	The chance of shoes or clothes from visitors	- The use by visitors of clean shoes and
animal care-	with Toxoplasma through			becoming infected with cat faeces is low.	clothes before entering the farm area
taker	visitors and animal caretakers				
	via oocysts on their shoes or				
	clothes				

Part	Hazard	Chance	Severity	Motivation	Control measure
Dung	Pig can become infected	Ι	Ι	Removal of dung from the farm may lead to	- Only clean wagons and trucks have
	with <i>Toxoplasma</i> via dung transport			introduction of pathogens on the farm if the trucks or other machines used for transport are not clean prior	access to the farm
				to entering the farm. Presence of <i>Toxoplasma</i> oocysts on these trucks via cat faeces is hypothetical. If this is	
				the case only an individual pig may become infected.	
Finishing pigs	Finishing pigs can become	I	61	Regulations oblige transporters to clean their trucks	- Always use clean transport
	infected with Toxoplasma			before loading new animals. The chance of trucks	
	due to the presence of cat			containing cat faeces or dead rodents is small. If trucks	
	faeces or dead rodents in the			are not clean this may lead to several animals becoming	
	trucks used for transport			infected. The time between loading and slaughter is	
				such that this will not lead to gross infection of the	
				animals.	
Insemination	Pigs become infected with	I	I	Transfer of Toxoplasma via sperm has not yet been	
and boars	T <i>oxoplasma</i> via sperm			reported.	
Cadavers	Pigs become infected with	I	2	The chance of transferring Toxoplasma infection via	- Remove cadavers immediately
	Toxoplasma through			the pig cadavers is small. Pigs are not infectious on the	
	cannibalism of cadavers or			outside and transfer only occurs through cannibalism	
	due to uptake of cadaver			or uptake by rodents or cats, which in turn can serve	
	material by cats or rodents			as a risk factor. Several pigs may become infected via	
				this route	

¹ Chance estimates whether the described risk occurs on the farm. It is ranked from 1 to 3; with 1 = chance of occasion is low, it occurs rarely or is theoretical; 2= chance of occasion is m ediocre, it can occur or occurs several times a year; 3= chance of occasion is high, it occurs frequently.

influence on a single pig to all pigs in the pen; 2 = the occasion has an influence on a part of all the pigs on-farm (pig unit); 3 = the occasion has influence on (almost) all the pigs on- \cdot Severity gives an estimate of the number of animals possibly affected by *Toxoplasma* when the risk becomes manifest. It is also ranked from 1 to 3; with 1 = the occasion has an farm.

Appendix (cont'd)