Contact with pigs and cats associated with high prevalence of *Toxoplasma* antibodies among farmers

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Abstract

Antibodies to *Toxoplasma* were measured by enzyme linked immunosorbent assay (ELISA) in sera from 159 abattoir workers, 142 pig farmers, and 106 grain or berry farmers. Farmers occupationally exposed to pigs had antibodies in 53 (37%) cases, abattoir workers in 40 (25%) cases, and farmers not exposed to pigs in 24 (23%) cases. In each group antibodies were more prevalent among those who had a cat or cats in the household. Controlling for age and cat contacts changed the prevalences less than one percent. The results indicate that pig farmers might have an occupational risk of toxoplasmosis. As the prevalence of antibodies among abattoir workers was about the same as among the referent farmers, it seems unlikely that infection from *Toxoplasma* could be acquired by mere handling of raw meat.

The *Toxoplasma* parasite infects many animal species. In humans, the infection is usually subclinical and in symptomatic cases the diagnosis is often missed.¹ After the primary infection, viable *Toxoplasma* cysts probably persist throughout life.² *Toxoplasma* has a worldwide distribution. The prevalence of infection among adults varies greatly in different populations; up to 90% are infected in Paris and up to 50% in the United States.³

Humans are known to be infected by at least three routes: transplacental infection in congenital transmission,⁴ infection by the cyst through ingestion of raw meat,^{5,6} and infection by the oocyst from contact with infected cats or exposure to contaminated soil.^{1,7,8} Routes of infection may vary, depending on hygienic and social factors, on humidity, and on occupations, manners, and customs of inhabitants.^{9,10}

Although cats are considered to play a central part in the spreading of toxoplasmosis,^{1,7,8,11} several epi-

demiological studies,^{9,12-17} including some in Scandinavia,¹⁸⁻²⁰ have failed to show increased prevalence of antibodies among those in contact with cats.

Toxoplasma infection is highly prevalent in swine in many countries,²¹ but the prevalence of antibodies to *Toxoplasma* in slaughtered pigs in Finland is only about 2.5% as measured by ELISA.²²

We studied the prevalence of antibodies to *Toxoplasma* among pig farmers, grain or berry raising farmers, and abattoir workers to find possible associations with cat and pig contacts.

Materials and methods

Workers from the killing floor of six pig processing abattoirs situated in central and northern Finland were asked to participate. In today's Finnish abattoirs these workers use bare hands in contact with the flesh of the carcasses. For the sake of hygiene the use of gloves, customary for other jobs in meat processing plants, is forbidden in work on the killing floor. A total of 177 workers were asked to fill in a questionnaire and to give a blood sample for antibody testing; 159 (90%) blood samples were obtained from the abattoir workers.

Similar questions were sent to 270 pig farmers and 310 farmers whose first or second line of production was either grain or berry raising. The grain or berry farmers were not involved in meat production and lived in the Kuopio district in eastern Finland. The source of the names and addresses was different for the two farmer groups. The data for the pig farmers were from the register of the local cooperative meat processing company, which covers virtually all the swine producers in the area. The grain or berry farmers were identified from the register of the local agricultural advisory association.

The questionnaire was returned by 248 (92%) pig farmers and by 266 (86%) control farmers. The farmers who were over the age of 65 or were no longer active in farming were excluded, as were the control farmers who had cattle or pigs. The register used to obtain the grain or berry farmers contained much outdated information as 26 farmers were no longer active in farming and 52 were over 65 years of age; a further 20 kept cows. Another three replies had no

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identifying name and three were received too late. Thus only 162 grain or berry farmers were asked to submit a blood sample. For the pig farmers, 35 were excluded because they lived outside the area included in the study, 10 kept no pigs, and one kept cows. This reduced the number of pig farmers to 224. According to a power estimation about 200 subjects would be needed and, consequently, a random sample of 206 from the 224 pig farmers was asked to participate in the blood sampling.

Blood samples for the *Toxoplasma* antibody testing were obtained from 142 (69%) pig farmers and from 106 (65%) grain or berry farmers. All blood samples were collected from the cubical vein in spring 1989.

The age of the subjects varied between 20 and 65 years. The mean age of the pig farmers who were asked to participate in the blood sampling was 42 and for those whose blood samples were obtained it was 43. The equivalent mean ages for the grain or berry farmers were 46 and 47 and for abattoir workers 39 and 39 years respectively. The proportion of women varied among the study groups; 20% of the pig farmers who were asked for blood samples were women. The percentage for those actually sampled was 23%. The equivalent percentages for the control farmers were 15 and 13, and 10 for both groups among the abattoir workers. Twenty four per cent of the pig farmers, 17% of the control farmers, and 48% of the abattoir workers were current smokers.

As eating of raw or undercooked meat is extremely rare in Finland, information about this practice was not sought. A habit of tasting raw minced meat while preparing meals was reported by 13% of the pig farmers, by 10% of the control farmers, and by none of the abattoir workers.

The samples of blood were allowed to clot and sera were separated by centrifugation and stored at -20°C until analysis. *Toxoplasma* specific IgG antibodies were primarily determined by a semiquantitative enzyme linked immunosorbent assay (ELISA) using a commercial kit (ToxEnz G-96, Northumbria Biologicals Limited, Northumber-

land, UK) with peroxidase labelled antihuman IgG as conjugate and 3,3', 5,5' tetramethyl benzene as chromogenic substrate. As in our previous larger study⁴² borderline sera with ambiguous results were re-examined by a quantitative *Toxoplasma gondii* IgG ELISA (Labsystems, Helsinki, Finland) with alkaline phosphatase labelled antihuman IgG as conjugate and p-nitrophenylphosphate as substrate.

According to the instruction leaflet of ToxEnz G-96, the sera that gave an absorbance higher than that obtained with the low titre positive control were considered antibody positive, the sera with absorbance values of between 90 and 95% of that obtained with the low titre positive control were considered borderline, and the sera with absorbance less than 90% of that obtained with the low titre control serum were interpreted as antibody negative. The instruction leaflet of ToxEnz G-96 does not state the antibody concentration of the low titre positive control serum in international units (IUs). We examined the low titre control serum against toxoplasma reference serum (500 IU/ml, Biotrol, Paris, France) and found that the positive limit value in the ToxEnz G-96 kit was 15 IU/ml, which is generally considered to be the cut off value between negative and positive titres, for instance in the Labsystems' *Toxoplasma gondii* IgG ELISA. Only 15 (4%) borderline sera were found in this study.

The prevalence of antibodies was compared in the three worker groups between those who had cats and those who had no cats in their households. Risk differences (RDs) with their 95% confidence intervals (95% CIs) were calculated.²³

Results

Among all subjects 117 (29%) were positive for *Toxoplasma*, 53 (37%) among the pig farmers, 24 (23%) among the grain or berry farmers, and 40 (25%) among the abattoir workers (table 1). The RD for the pig farmers compared with the control farmers was 11% (95% CI 3 to 26%). For the

Table 1 Numbers of *Toxoplasma* positive subjects among the pig farmers, the abattoir workers, and the grain or berry farmers according to age, sex, and having a cat or cats in their households

	Pig farmers No (%)	Abattoir workers No (%)	Grain or berry farmers No (%)	Total No (%)
All	53 (37)	40 (25)	24 (23)	117 (29)
Age (y):				
30 or under	10 (50)	4 (13)	1 (17)	15 (27)
31-40	13 (29)	18 (30)	7 (24)	38 (29)
41-50	13 (32)	13 (26)	5 (18)	31 (26)
Over 50	17 (45)	5 (26)	11 (26)	33 (33)
Sex:				
Men	41 (38)	36 (25)	18 (20)	95 (28)
Women	12 (36)	4 (25)	6 (43)	22 (35)
Having cats:				
Yes	35 (41)	9 (35)	12 (29)	56 (37)
No	18 (32)	31 (23)	12 (18)	61 (24)

abattoir workers, compared with the control farmers, the RD was only 3% (95% CI -8 to 13%). More pig farmers than controls were positive in all age groups.

Among the 64 women in the whole study 22 (34%) were positive compared with 96 (28%) among the 348 men ($p > 0.10$, 95% CI 0-13%). This difference was almost all due to the higher prevalence of positive subjects among the grain or berry farmers.

At the time of blood sampling 86 (60%) of the 142 pig farmers, 41 (39%) of the 106 grain or berry farmers, and 26 (16%) of the 159 abattoir workers had a cat or cats in their households.

Having a cat or cats in the household was associated with increased positivity to *Toxoplasma*. In the whole material 37% of those having a cat or cats were positive, compared with 24% among those with no cat (RD 13%, 95% CI 4 to 22%). The RD and the 95% CI among the pig farmers, controls, and abattoir workers were 9% (-8 to 26%), 11% (-6 to 28%), and 11% (-8 to 32%) respectively. The prevalences of positive subjects after standardisation for age and ownership of cat or cats were 36% for the pig farmers, 25% for the abattoir workers, and 23% for the control farmers.

The current smokers were less often positive (25%) than the subjects not smoking (30%). The 95% CI of this 5% risk difference included zero.

Tasting of raw minced meat while preparing food was reported by 41 farmers. Among them 11 (37%) were positive for *Toxoplasma* compared with 65 (31%) positives among the 210 farmers who reported not having that habit (RD 6%, 95% CI -10 to 22%).

Discussion

In their comprehensive review article Dutkiewicz *et al* mention toxoplasmosis as an occupational biohazard among both agricultural workers breeding animals and workers in facilities processing animal products.²⁴ We found significantly more antibodies among pig farmers compared with controls, but not among the abattoir workers.

The possible work related nature of toxoplasmosis was first considered in 1961 in two studies. In a Japanese study 61 (68%) of 90 skin tested abattoir workers were positive compared with only 30% among three control groups (pluck handlers, ham making workers, and farmers).²⁵ At the Georgia training school for mental defectives, USA, 703 patients were serologically tested and 51% of the positive patients were found among the inhabitants of two cottages compared with only 15% positive patients among the other 10 cottages. The authors state that "the only related identifiable factor which differentiated these two cottages was that they housed the patients who assisted in the farm work and in the care of the farm animals."²⁶

In 1974 Rhodesian slaughtermen were reported to have twice as much antibody to *Toxoplasma* as European or African blood donors.²⁷ A year later, the prevalence of antibodies against *Toxoplasma* measured by indirect haemagglutination among Brazilian slaughterhouse workers (72%) was found to be twice as high as that reported for persons associated with a veterinary school (46%) in the same area.²⁸ For the killing floor workers the prevalence of antibodies *Toxoplasma* was 65%.²⁸ In a Slovak study 39% of 1179 subjects employed in animal production and the meat industry were found to have antibodies against *Toxoplasma* but the study had no reference group; nor was any reference prevalence given.²⁹ In another Slovak study antibodies against *Toxoplasma* were measured by three different methods (complement fixation, indirect immunofluorescence assay, and ELISA), again with no reference group. Among the employees in the meat industry, however, the slaughterhouse workers had the highest rate of antibodies and the highest geometric means of antibody titres for *Toxoplasma* by all three methods.³⁰ Women employed in agriculture, flower production, and the food industry were found to be at risk of toxoplasmosis in a Swiss case-control study considering several potential risks of *Toxoplasma* during pregnancy. Odds ratios were 5.6 for those in farm work and flower production and 2.7 for those in the food industry. The number of subjects in both groups was small, however, six in the first and 24 in the second.³¹

In Poland antibodies to *Toxoplasma* were studied among health service workers, workers in poultry processing plants, inseminators, farmers, army workers, and control persons. Higher titres and more frequent positive reactions with *Toxoplasma* antigen in relation to the controls were obtained for persons occupationally exposed to *Toxoplasma gondii*.³² On the other hand, no differences in the prevalence of antibodies to *Toxoplasma* were found among persons in different occupations in a study made in La Guadeloupe, French West Indies. The occupational groups considered were white collar workers, blue collar workers, agricultural workers, traders, and craftsmen. No distinction between those exposed to pigs and others was made for the agricultural workers.³³ In India livestock or pet owners had about 1.3 times more prevalent antibodies than campus residents with no apparent animal contacts.³⁴

Studies concerning the risk of veterinarians and veterinary students have produced contradictory results. In the first two studies the risks of toxoplasmosis were compared between veterinary and medical students.^{35 36} The veterinary students were more often skin test positive, but if their more prevalent rural residence was considered, the difference in positivity between the groups disappeared. Small animal practitioners in San Francisco³⁷ and

Canada³⁸ had no more antibodies than the population at large³⁷ or a selected control group.³⁸ Furthermore, no differences were found between veterinarians, other workers with an occupational cat and dog contact, and a non-veterinarian control group with no occupational animal contacts.³⁹ When students and personnel in veterinary colleges in California and Brazil were compared, the Brazilians were twice as often positive but no particular species of animals could be incriminated.¹⁴ A study with the aim of relating animal contact and other epidemiological factors to toxoplasmosis among personnel and students of Iowa Veterinary College concluded that "the toxoplasmosis problem is complex and probably involves a multiplicity of factors, some of which may be still unrecognised."⁴⁰ Exposure to cats in a research setting has been found to carry little or no risk of infection from *Toxoplasma*.¹⁶

To our knowledge, no previous Scandinavian or Finnish studies focus on the occupational nature of infection from *Toxoplasma*. No relation between antibodies to *Toxoplasma* and abattoir work was found in this study, which seems to contradict at least some earlier reports.^{25 28 30} The level of hygiene in today's Finnish abattoir is different from that in the early 1960s in Japan or later in eastern Europe. Most of the Finnish abattoir workers work bare handed. It should be noted, however, that the abattoir workers in Finland are put on sick leave immediately they receive hand wounds, including minor scratches. Consequently, handling of raw meat with hands that have the skin intact seems to carry no risk of infection from *Toxoplasma*.

The comparison of control farmers with the abattoir workers carries a possible bias; these groups are not comparable with respect to their living environments. The abattoir workers have a more urban way of living than the grain or berry farmers. Although differences in positivity between urban, town, or rural environments were not found in a large nationwide study made in the United States,⁴¹ opposite results were obtained when environmental backgrounds of college students were considered.³⁶ Living in rural districts appeared to represent a predisposing factor for the acquisition of infection from *Toxoplasma* at younger ages in Norway.²⁰

To the best of our knowledge, this is the first study to compare farmers exposed and not exposed to swine. After standardising for age and the ownership of a cat or cats the risk difference was about 11% indicating that pig farmers have a somewhat increased risk of infection from *Toxoplasma* because of occupation. The relation between antibodies to *Toxoplasma* and pigs is not necessarily a causal one. If the soil and environment of the farm, including the swine confinement buildings, are contaminated with oocysts the farmer is exposed to oocysts in the dust during work in the piggery and in a similar manner pigs are infected by the feed.

In a Finnish study that investigated the presence of antibodies to *Toxoplasma* among pregnant women in the Helsinki area (mainly urban population), the positivity among the 16 733 women studied was 20% with a small increase with age. In the age group 35 to 39 the prevalence among pregnant women was 26%.⁴² These prevalences are close to those of our present study for grain or berry farmers (23%) and for abattoir workers (25%).

Epidemiological studies that have investigated the association of *Toxoplasma* positivity with exposure to household cats have been contradictory. Ganley and Comstock listed 12 studies concerning the association of cats and tests positive for *Toxoplasma gondii* published before 1978. In seven of them no association with cats was noticed.⁴³ Only in three studies was the association obvious. In one study no association with cats alone was noticed⁴⁴ and in a Finnish study an association was found with pedigree cats but not with house cats.⁴⁵

By contrast with two Scandinavian studies,^{19 20} we found a clear relation between a cat or cats in the household and antibodies to *Toxoplasma* in all the three groups studied, even though the 95% CI included zero due to the limited number of subjects. Stray-Pedersen and Lorentzen-Styr²⁰ asked about contacts with cats and Hultdt *et al*¹⁹ considered families living with these animals.¹⁹ Our question focused on cats in the household, not on physical contact with them. For the spreading of *Toxoplasma*, the environment contaminated by the cats' oocysts is important,⁴⁶ not the physical contact itself.¹⁵ This property is measured only indirectly by the three studies, ours included. Furthermore, it can be presumed that the cat contact of the rural population is more stable than that in urban areas. Thus the present cat contact of our study of farmers would be a better estimate of the life long contact with cats, which really matters as toxoplasma antibodies are known to last for decades.⁴⁷

None of the previously cited studies concerning the association of animal contact and antibodies to *Toxoplasma* have been follow up studies. Ours is also a cross sectional one. Antibodies to *Toxoplasma* are known to persist for decades.⁴⁷ Subjects considered to have no animal contact in cross sectional studies might have been exposed earlier in their lives. Thus the association between antibodies to *Toxoplasma* and animal contact is weakened and the cross sectional design tends to underestimate the true association.

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