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***Toxocara*-infestations in Austria: a study on the risk of infection of farmers, slaughterhouse staff, hunters and veterinarians**

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Abstract A total of 585 persons from several occupational groups (farmers, slaughterhouse staff, hunters, veterinarians) exposed to *Toxocara* infestations and 50 persons of a control group were tested for the presence of specific antibodies to the *Toxocara canis* antigen using an enzyme-linked immunosorbent assay and a western blot. Farmers showed the highest seroprevalence (44%), followed by veterinarians (27%), slaughterhouse staff (25%) and hunters (17%), whereas only 2% of the individuals of the control group were seropositive. Thus, the risk to *Toxocara* infestation is 39, 18, 16 and 9 times higher for farmers, veterinarians, slaughterhouse staff (some workers were part-time farmers) and for hunters, respectively, when compared to the control group. The main source of infection in rural areas seems to be (roaming) farm cats and dogs that have not been dewormed. The results are discussed with a view to potential risk factors and preventive measures, in terms of veterinary and human medicine.

Introduction

While *Toxocara canis* ('dog roundworm') and *Toxocara cati* ('cat roundworm') have been known as parasites of dogs, foxes and cats for more than 200 years, it was not until the early 1950s that they were also recognised as human pathogens (Wilder 1950; Beaver et al. 1952). Today, one can distinguish between several forms of toxocarosis (apart from the clinically inconspicuous *Toxocara* infestation), such as the larva migrans visceralis (LMV) syndrome (Beaver et al. 1952), the ocular larva migrans syndrome (OLM) (Wilder 1950; Dietrich et al. 1998), inapparent (or covered) toxocarosis (Bass et al. 1987; Taylor et al. 1987), the common toxocarosis (Auer et al. 1990; Magnaval et al. 1994) and cerebral toxocarosis (Magnaval et al. 2001; Auer and Aspöck 2004). Other clinical pictures (i.e. bronchial asthma, epilepsy, rheumatic arthritis) have also been assumed and are discussed as being the result of *Toxocara* infestations (Gillespie 1993; Glickman 1993).

Humans acquire the infection by the oral ingestion of the eggs from *Toxocara* infected dog, fox or cat faeces (smear infection) or by the consumption of raw or not adequately cooked paratenic hosts (i.e. chicken, rabbits, snails) (Nagakura et al. 1989; Romeu et al. 1989; Dubinsky et al. 1995). The L₃ larvae leave the eggs in the small intestine and reach the liver by haematogenous or lymphogenous routes or through active migration. They can, subsequently, be transported to all human organs via the heart and systemic circulation.

Both *Toxocara* species are spread worldwide and can, thus, also be found in Central Europe. Young dogs (and foxes) and cats, in particular, are the carriers of *T. canis* and *T. cati*, respectively, and may shed more than 50,000 eggs/g of faeces daily. The percentage of natural hosts patently infected with *Toxocara* sp. decreases with increasing age, as a result of immunity (Coggin 1998; Rommel et al. 2000).

In Austria, infection rates of up to 18% have been reported for dogs (Hinaidy 1971, 1976; Supperer and

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Hinaidy 1986) and of more than 60% for cats (Kasiecka 1982). Red foxes (*Vulpes vulpes*) are also often infected with *T. canis*, with prevalences of up to 47% in Austria (Suchentrunk and Sattmann 1994; Lassning 1996) and even up to 70% in Germany (Steinbach et al. 1994). In addition, epidemiological studies on environmental contamination carried out in Vienna, the provinces of Lower Austria, Salzburg, the Tyrol and Styria, have shown that up to 14% of faeces, soil and sand samples from public parks were contaminated with *Toxocara* eggs (Auer and Aspöck 1995, 2004; Kutzer et al. 1995; Auer 2003).

Two seroepidemiological screenings among a normal human population, comprising of more than 6,000 pregnant women from all over Austria, on one hand, and more than 7,500 18-year-old men (military recruits) in the provinces Tyrol, Vorarlberg and Salzburg, on the other hand, revealed seroprevalences of 1.4 and 3.7%, respectively (Walder 1987; Walder and Aspöck 1988; Auer and Aspöck 1995, 1998). However, a survey among Styrian veterinarians yielded a seroprevalence of 27% (Deutz et al. 1996; Nowotny et al. 1997).

Within the recent seroepidemiological study, the sera of farmers, slaughterhouse staff and hunters, on the one hand, and a control group, on the other, were examined for specific anti-*Toxocara* antibodies, a questionnaire analysed and the risks of infection due to age, sex, occupation and husbandry evaluated for the different groups; for direct comparison data of the already tested Styrian veterinarians (Deutz et al. 1996, 2002, 2003; Nowotny et al. 1997) were included into the study.

Material and methods

Volunteers

In 1999, a total of 152 farmers and 147 slaughterhouse workers from Styria, 149 hunters from Styria and Burgenland and 50 persons of a control group, residents of Graz (the capital city of Styria), who did not belong to the occupational groups described above, were examined. Table 1 shows the age and sex distribution of these volunteers.

Table 1 Age and sex distribution of volunteers

Occupational group	Number of volunteers tested						
	Total	Male			Female		
		No.	Age Ø	Min. – max.	No.	Age Ø	Min. – max.
Farmers	152	82	42	22–70	70	42	24–64
Slaughterhouse workers	147	109	34	15–58	38	32	20–59
Hunters	149	146	50	22–76	3	41	38–43
Veterinarians (Deutz et al. 1996; Nowotny et al. 1997)	137	107	41	25–72	30	39	24–60
Control group	50	22	44	28–58	28	38	19–54

Questionnaire

In order to collect personal and epidemiological data, questionnaires were issued to the volunteers. The following parameters were collected: sex, age, type of veterinarian practice, type of activity in slaughtering facilities and animal husbandry (dogs/cats, livestock, other animals). The questionnaires were subsequently evaluated in an anonymised form. Statistical analysis was carried out using the four field table and a χ^2 test.

Parasitological–serological tests

Blood samples were taken using the Vacutainer System. The blood samples were centrifuged, packaged, frozen and stored at -70 °C until serological tests were carried out.

All sera were tested by an enzyme immunoassay (ELISA) as the basic test (de Savigny 1975; de Savigny et al. 1979) and a western blot (WB) for confirmation; in both tests sera were examined for the presence of IgG antibodies, using the excretory–secretory (E/S) antigen of *T. canis*. Serum samples were considered serologically positive if they showed clearly positive reactions in the ELISA and displayed the typical reactivity pattern in the WB (Magnaval et al. 1994; Auer and Aspöck 1995, 1998).

Differential blood count

Differential blood counts were prepared from blood samples (EDTA blood) of 104 farmers, slaughterhouse workers, hunters and the control group. Examination was carried out under a microscope and the percentage of eosinophilic granulocytes was calculated according to standard methods; the standard eosinophil count was taken to be 0–4%.

Results

Results of parasitological–serological tests:

In total, antibodies to the *T. canis* E/S antigen were detected in 164 (25.8%) out of 635 sera, tested using

Table 2 Antibody prevalences among different occupational groups and the control group

Occupational group	N_{pos}	N_{neg}	$N_{\text{♂pos}}$	$N_{\text{♂neg}}$	$N_{\text{♀pos}}$	$N_{\text{♀neg}}$
Farmers	67/152 (44.1%)	85/152 (55.9%)	37/82 (45.1%)	45/82 (54.9%)	30/70 (42.9%)	40/70 (57.1%)
Slaughterhouse workers	37/147 (25.2%)	110/147 (74.8%)	35/109 (32.1%)	74/109 (67.9%)	2/38 (5.3%)	36/38 (94.7%)
Hunters	22/149 (14.8%)	127/149 (85.2%)	22/146 (15.1%)	124/146 (84.9%)	0/3	3/3
Veterinarians (Deutz et al. 1996; Nowotny et al. 1997)	37/137 (27.0%)	100/137 (73.0%)	29/107 (27.1%)	78/107 (72.9%)	8/30 (26.7%)	22/30 (73.3%)
Control group	1/50 (2.0%)	49/50 (98.0%)	0/22	22/22	1/28 (3.6%)	27/28 (96.4%)
Total	164/635 (25.8%)	471/635 (74.2%)	123/466 (26.4%)	343/466 (73.6%)	41/169 (24.3%)	128/169 (75.7%)

ELISA and western blot. In detail, 67 out of 152 (44.1%) farmers, 37 of 147 (25.2%) slaughterhouse workers, 37 of 137 (27.0%) veterinarians and 22 of 149 hunters (14.8%) were serologically positive, whereas only one serum of the control group (2.0%) was reactive in ELISA and WB (Table 2).

Antibody prevalence and age:

Seropositivity increased with increasing age in the group of veterinarians. No age dependence could be observed in the group of farmers, slaughterhouse workers and hunters; among these occupational groups, the highest seroprevalence was observed in the 41–50 age class.

Antibody prevalence and sex:

Table 2 shows the antibody prevalences among males and females in the different groups tested. In total, males showed higher seroprevalences than females; a significant correlation between antibody prevalence and sex could be observed only in the group of the slaughterhouse workers.

Antibody prevalence and occupation:

A statistically significant correlation was found between the exposure factor, occupation and the *Toxocara* antibody prevalence ($P < 0.05$). Compared to the control group, all other occupational groups showed a much higher risk of infection (odds ratio, $OR > 1$), as assessed by the four field table and a χ^2 test. The calculation showed a 38-fold increase in risk ($OR = 38.6$; 95% confidence interval: 5.198–286.991) for a *Toxocara* infestation among farmers as compared to the control group; veterinarians had a 18-fold [2.14–136.05], slaughterhouse workers a 16-fold [2.19–123.58] and hunters a 9-fold [1.30–74.91] increase than the un-exposed occupational group (i.e. control group).

Dogs and cats as risk factors:

Dogs and cats were kept by 27% of farmers, by 22% of slaughterhouse workers, by 73% of hunters, by 53% of veterinarians and by 42% of subjects of the control group.

No statistically significant correlation was detected between *Toxocara* infestation and the risk factors of a 'cat' or 'dog as pet'. A significant relation, however, was found between the seroprevalence of *Toxocara* among farmers and slaughterhouse workers, on the one hand, and the keeping of cats and dogs as pets ($P < 0.05$), on the other. 32 of 104 *Toxocara* positive farmers and slaughterhouse workers reported keeping dogs and cats as pets. The OR was 2.236, indicating that the risk of acquiring a *Toxocara* infestation increases by a factor of 2.236 if dogs and cats are kept as pets.

Antibody prevalence and eosinophilia:

22 of the total of 104 *Toxocara* positive volunteers (farmers, slaughterhouse workers) showed a percentage of eosinophils of more than 4% in the differential blood count. The values of 82 volunteers were in the normal range. Since more than half of the volunteers with eosinophilia were *Toxocara* negative, no statistically significant relation was established between eosinophilia and toxocarosis.

Discussion

Twenty years after the LMV syndrome had been described in the USA by Beaver et al. (1952), Styria was the first Austrian province, where human toxocarosis cases were diagnosed and treated (Wendler 1972). The first case of covert 'toxocarosis' in a 5-year-old boy was also observed in the province of Styria (Varga et al. 1998). In 1995, a comprehensive screening programme on zoonoses among Styrian veterinarians was carried out; it revealed an antibody prevalence of 27% to the *T. canis* E/S antigens (Deutz et al. 1996; Nowotny et al. 1997). This result induced a further screening study,

primarily among those persons who were in 'occupational contact' with the veterinarians (i.e. farmers, hunters). Thus, 152 Styrian farmers, 149 hunters from Styria and the southern Burgenland, and 147 Styrian slaughterhouse workers, as well as a control group deriving from the capital city of Graz, were examined for specific antibodies to the *T. canis* E/S antigens by 'classical' ELISA (de Savigny et al. 1979; Auer and Aspöck 1995, 1998) and WB (Magnaval et al. 1994; Auer and Aspöck 1995, 1998).

Based upon the *Toxocara* seroprevalence of the 'normal population' of <5% in Austria (Walder 1987; Walder and Aspöck 1988; Auer and Aspöck 1995, 1998), Germany (Kimmig et al. 1991), and Switzerland (Stürchler et al. 1996), our recent study obviously showed extremely high infection rates among farmers (44%) and rather high antibody prevalences among veterinarians (27%) (Deutz et al. 1996; Nowotny et al. 1997), slaughterhouse workers (25%; one-fourth of whom were also part-time-farmers), and hunters (15%); whereas the control group revealed only a 2% infection rate. The conclusion of this study is that the exposure to *Toxocara* spp. is essentially higher in rural than in urban areas, although dogs and also the cats (as pets) are densely populated in Central European countries (Prange et al. 2000). In our opinion this phenomenon is due to one main reason, the rather low standard of health care (i.e. deworming) for cats and dogs in rural areas. Our opinion is based on several facts:

1. In rural areas cats and dogs (since many farmers keep cats and dogs) roam freely on farms, some of them also extending to the neighbourhood (contamination of vegetable gardens, orchards),
2. A (more or less) permanent contamination of the environment of the farmers' houses, of grass and hay stored in barns (Stürchler et al. 1990),
3. The contamination of the fur of cats and dogs (Wolfe and Wright 2003),
4. The high tenacity of the eggs (*Toxocara* eggs may survive in a humid environment for several months up to 4 years. The eggs resist cold periods but are sensitive to desiccation and temperatures of more than 30–35 °C),
5. The ignorance of people on the parasites and their ways of transmission,
6. The relatively high costs of anthelmintics for deworming dogs and particularly of cats.

Thus, it is obvious that farmers and part-time farmers (among 25% of the slaughterhouse workers were part-time farmers), and also veterinarians who have occupational contact with farmers, and their environment are living in persisting risk of infection and reinfection in such rural areas. On the other hand, the *Toxocara* seroprevalence of hunters is 'only' 15% and much lower than that of farmers, slaughterhouse workers and veterinarians; this phenomenon might be due to the fact that hunting-dogs, usually of pure-blooded origin and of

high cost value, are kept at a high hygienic and healthy standard (i.e. regular visits to the veterinarians and regular deworming procedures).

Our study did not reveal other *Toxocara* risk factors (i.e. age, gender) – at least statistically confirmed ones – although the antibody prevalence was a little bit higher in older and in male probands.

The results obtained in this study represent the basis for the development and establishment of proper preventive measures, both in veterinary (deworming management, hygiene measures, instruction of animal owners) and human medicine (enhanced consideration of toxocarosis in differential diagnosis). Primarily, toxocarosis control must be directed at the high shedding rates of young dogs and cats and the possibility of lactogenic transmission. Pets should be dewormed for the first time, 2 weeks post-partum, in order to avoid or reduce the shedding of eggs (Rommel et al. 2000). Moreover, other preventive tools should be taken into consideration, e.g. regular deworming actions of young cats and dogs (preferably initiated by health authorities), the removal of dog and cat faeces, and, finally, the killing of *Toxocara* eggs by heating up to 70 °C. All occupational groups tested here in the present study, showed an elevated risk to *Toxocara* infestation in comparison to people living in urban areas; thus, such people, particularly farmers and part-time farmers, have to be aware of the consequences of the disease 'toxocarosis' and should, therefore, change their (personal) view concerning the hygienic status of their cats and dogs.

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