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Echinococcus multilocularis in Belgium: Prevalence in red foxes (*Vulpes vulpes*) and in different species of potential intermediate hosts

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Abstract

Echinococcus multilocularis causes a rare but potentially lethal zoonotic infection in humans. This tapeworm is known to be endemic in foxes in several countries of Western and Central Europe. In Western Europe, the common vole (*Microtus arvalis*) and the water vole (*Arvicola terrestris*) are considered to be the most important intermediate host species of this cestode whereas the red fox is by far the most important final host. The purpose of this study was to provide data on the prevalences in Wallonia (Southern part of Belgium) both in the red fox and in different potential intermediate hosts.

A total of 990 red foxes were examined between January 2003 and December 2004 for the presence of *E. multilocularis*. The average prevalence was 24.55% (22.38–27.87). Out of 1249 rodents or insectivores belonging to the species *Apodemus sylvaticus*, *Arvicola terrestris*, *Clethrionomys glareolus*, *Microtus arvalis*, *Microtus agrestris* and *Sorex araneus*, only one *M. arvalis* (out of 914–0.11% (0.003–0.61) and one *C. glareolus* (out of 23–4.3% (0.1–21.9) were found to be infected. However, the muskrat (*Ondatra zibethicus*) seems to be a good intermediate host as 11.18% (9.72–12.76) of the animals ($n = 1718$) were found to be infected. A positive correlation was found between the prevalences in foxes and in muskrats in each of the different geological regions. This study indicates that the muskrat is highly sensitive to this zoonotic tapeworm and could perhaps represent a good bioindicator when studying the epidemiology of this parasitic infection in Belgium and in other countries where the muskrat is present.

Keywords: *Echinococcus multilocularis*; Red fox; Muskrat; Rodents; Belgium; Epidemiology

1. Introduction

Echinococcus multilocularis is a small tapeworm belonging to the Taenidae family. This parasite is the causative agent of alveolar echinococcosis, a zoonotic parasitic disease which causes a severe hepatic disorder in humans.

The presence of the parasite is limited to the northern hemisphere. *E. multilocularis* is present in several countries of Western and Central Europe, in some of the northern American states (Leiby et al., 1970; Storandt et al., 2002) and in Northern

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and Central Eurasia (Ito et al., 2003; Vuitton et al., 2003).

In Europe, the typical life cycle is sylvatic with foxes (*Vulpes vulpes*) as the main definitive host and rodents, particularly those of the family Arvicolidae, as intermediate hosts. Less frequently other species of the families Canidae and Felidae are final hosts, including the dog and cat (Eckert et al., 2001).

Increasing fox densities were recorded in several European countries more particularly in suburban and urban areas. Recent studies investigating the potential risk in urban areas in Central Europe indicated that infected foxes seemed to represent an important reservoir for the parasite even in an urban environment (Hofer et al., 2000; Tsukada et al., 2000).

In several reports, the common vole (*M. arvalis*) and the water vole (*A. terrestris*) are considered as the most important intermediate host species of *E. multilocularis* in Europe. Their significance differs according to the various countries and regions. Few data are available about the parasite prevalences in rodents, but in general they are fairly low (<1–6%) as compared to those in foxes from the same area (20–60%) (Hofer et al., 2000). However, studies in France and Switzerland indicated that high-endemic foci of rodent *E. multilocularis* infections do exist and in these cases prevalences of *E. multilocularis* in the water vole can reach 39% (Eckert et al., 2001).

In Belgium, the presence of *E. multilocularis* was recorded for the first time in the South-Eastern of the country in 1991 (Brochier et al., 1992). An additional study in 1997 revealed that 51.0% (74/145) of the red foxes collected in the same area were positive (Losson et al., 1997). In 2002, the presence of the adult tapeworm was detected in other regions of Wallonia (Losson et al., 2003). However, no data are available about the intermediate hosts of *E. multilocularis* in Belgium.

Consequently the present study was conducted in order to evaluate in Wallonia (Southern Belgium) the prevalence of *E. multilocularis* infection both in its main final host, the red fox, and its potential intermediate hosts species.

2. Material and methods

2.1. Location

Wallonia is the southern administrative part of Belgium and has a total surface area of 16,844 km^2. It is divided into five distinct geological areas. Minimum and maximum altitudes are <100 and 692 m above sea level respectively.

2.2. Collection and post mortem examination of the red foxes

A total of 990 red foxes were collected between January 2003 and December 2004 and were examined for the presence of *E. multilocularis*.

Foxes were either shot or killed in road accidents. They were transported in sealed plastic bags and kept at −20 °C until further examinations. At necropsy, the animals were sexed and their age was evaluated (Eckert et al., 2001): the foxes were divided into three age categories. Cub refers to foxes of up to 6 months of age, sub-adult refers to foxes of 7–12 months of age and adult refers to foxes older than 12 months of age.

The term "juvenile" is used to describe foxes under one year of age and consequently the category includes both cubs and sub-adults (Harris, 1978). The age evaluation was done by morphological changes of teeth that can be used to distinguish foxes under and above 1 year of age.

The entire gut was collected and kept for at least 10 days at −80 °C in order to inactive the infective material. After thawing the gut was opened and 15 mucosal scrapings were collected from the proximal, middle and posterior parts of the small intestine. The collected material was examined under a dissecting microscope under a 24–40× magnification (Eckert et al., 2001).

The identification of the worms was performed according to the size and the appearance of the uterus of the last segment.

2.3. Collection and post mortem examination of the intermediate hosts

Different species of voles were trapped in an area highly endemic for alveolar echinococcosis located in the Luxemburg province at an altitude of 460–495 m above sea level. The trap used for the common voles was a Tomahawk's Humane Mouse Trap, Model 101 (Tomahawk live trap, Wisconsin) and for the water voles a Topcat trap (Topcat GmbH, Switzerland). The landscape was predominantly composed of pastures and crops. Muskrats were provided by the staff of the regional muskrat control programme. The animals are trapped twice a year in spring and autumn.

Voles and muskrats were tagged (date and place of capture) and stored at −20 °C until further use. At post mortem the animals were sexed and weighted. Muskrat population was divided into adult and sub-adult (Pankakoski, 1980).

At necropsy, liver and abdominal viscera were examined visually for the presence of cystic lesions. Identification on *E. multilocularis* infection was based on gross and microscopical appearance of lesions. However in the case of very small, atypical or calcified liver lesions, the method of choice for identifying *E. multilocularis* was a PCR technique (Gottstein et al., 2001). In the present study, the slightly modified inner primer EM H15-H17 was used (Dinkel et al., 1998).

2.4. Statistical analysis

The prevalence of infection was estimated with a 95% confidence interval (95% CI) assuming a binomial exact distribution. Trends of prevalence in foxes and muskrats were compared by using Spearman's rank correlation. The potential influence of sex, and season (only in foxes), and age on the carriage of the infection in red foxes and rodents was assessed. Statistical analysis of these data was done using a Chi square (Chi^2) test and the trend for a parameter toward risk factors was evaluated by mean of odds ratio with 95% confidence interval; a P value < 0.05 was considered as significant. The possible relationships between age and time of collection of foxes and between the levels of infection with *E. multilocularis* in muskrats foxes in the different geographical area were assessed by a Cochran-Mantel-Haenszel (CMH) method (ORMH). This test was distributed as a Chi square test with respectively two free degree (age and time of collected foxes) and one free degree (muskrat and foxes infections). The Breslow-Day test was used to evaluate the homogeneity of the odds ratios.

3. Results

3.1. Examination of foxes

The number of foxes collected from the different region varied considerably according to fox population but also and mainly according to the hunters' participation in the survey.

Nevertheless, the parasitological results will be presented according to the different geological areas of Wallonia.

In the 990 foxes examined during this study, the average rate of infection with *E. multilocularis* was 24.55% (22.38–27.87).

Differences were observed among the different natural regions (Fig. 1). In the southern high plateau, prevalences varied from 33.8% (26.1–42.2) (48/142) to 40.84% (34.2–47.8) (87/213) in the Belgian Lorraine and in the Ardennes respectively. Surprisingly, the highest prevalence was recorded in the Fagne-Famenne-Calestienne (61.8% (43.6–77.8) (21/34)). Furthermore, the lowest prevalences were recorded in the Condroz (24.86% (18.7–31.8) (45/181)) and in the Hesbaye area (10% (7.3–13.3) (42/420)) where altitude is under 100 m above the sea level.

A preliminary evaluation indicated that the gender of foxes had no effect on the prevalence (females: 43.81% ($n = 435$); males: 56.19% ($n = 559$); $Chi^2 = 3.21$, $P =$

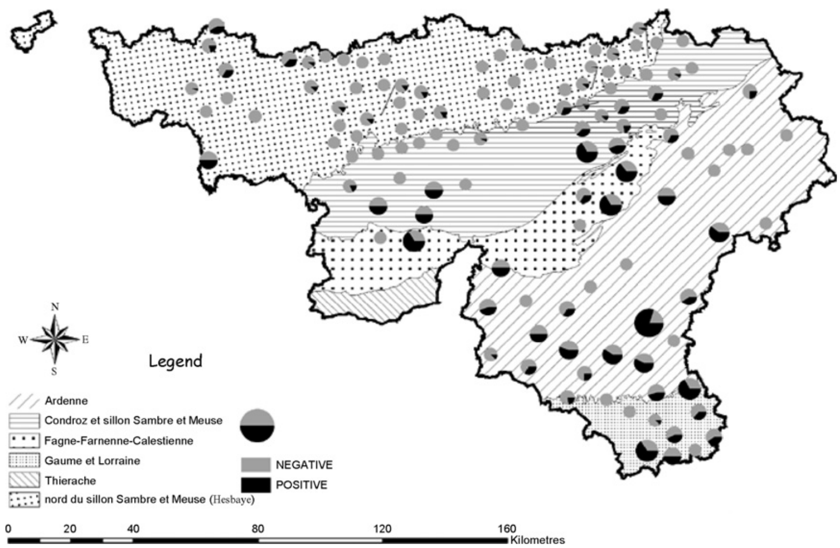

Fig. 1. Hanosset *Echinococcus multilocularis* in Belgium: prevalence in red foxes (*Vulpes vulpes*) and in different species of potential intermediate hosts. Prevalence of *E. multilocularis* carriage by the red fox (*Vulpes vulpes*) in the geological areas of Wallonia (2003–2004). The diameter of the circle is proportional to the number of examined foxes in a given place whereas the proportion of positive samples is indicated in black.

Table 1
Percentage of *E. multilocularis* infection in the different species of microtines trapped in the study area

Species	N_t/N_p [a]	Percentage of infection (95% CI) [b]
Apodemus sylvaticus	0/55	0 (0–5.3)
Arvicola terrestris	0/2	0 (0–78)
Clethrionomys glareolus	1/23	4.3 (0.1–21.9)
Microtus arvalis	1/914	0.11 (0.003–0.61)
Microtus agrestis	0/39	0 (0 7.4)
Sorex araneus	0/216	0 (0–1.38)
Total	2/1249	0.16 (0.02–0.58)

[a] N_t: Number of tested animals, N_p: number of positive animals.
[b] The 95% confidence interval (binomial exact).

0.074). In the summer and the autumn, foxes were significantly more often infected than in the winter and in the spring ($Chi^2 = 4.92$, $P = 0.03$) (OR: 1.4 (95% CI: 1.04–1.98)) and prevalence rates differed significantly in function of the age of foxes ($Chi^2 = 12.52$, $P = 0.002$). Sub-adults (OR*:* 5.12 (95% CI: 1.93–13.58)) and adults foxes (OR: 3.73 (95% CI: 1.59–8.72)) were significantly more often infected than cubs. The possible relationship between the age of foxes and the time of collection was made, using the CMH test. In fact the age of animals was found to be a confounding factor for the season because the crude OR was higher than the OR calculated for each age stratum: cubs (OR*:* 0.52 (95% CI: 0.06–4.76)), sub-adults (OR*:* 0.73 (95% CI: 0.16–3.39)), and adults (OR*:* 4.00 (95% CI: 1.00–1.96)).

3.2. Examination of rodents

Between March 2003 and September 2004, 1249 rodents were trapped in a highly endemic area (more or less 60% according to previous surveys carried out in the same area). The distribution of species and the numbers of *E. multilocularis* infected rodents are given in Table 1. Only one *C. glareolus* and one *M. arvalis* were found to be infected by the metacestode of *E. multilocularis* (0.16% (95% CI: 0.02–0.58)). The cysts measured between 5 and 20 mm in diameter. They contained numerous protoscolices.

In contrast, 11.18% (95% CI: 9.72–12.76) of the 1718 muskrats were carrying the metacestode of *E. multilocularis*. In agreement with the results in red foxes, differences were observed between the different geological areas of Wallonia (Table 2). The gender of the animal had no effect on the infection ($Chi^2 = 0.04$, $P = 0.84$). However the prevalence in the sub-adult muskrats was lower than in adult animals ($Chi^2 = 20.32$, $P = 6.56E^{-6}$).

The percentage of infected foxes and the percentage of infected muskrat are statistically highly correlated (Spearman's rank correlation coefficient = 1; $P < 0.0001$). Moreover all percentages observed in foxes are higher than those observed in muskrat whatever the natural area (Chi^2; $P < 0.0001$). In foxes, the percentage of infection observed in Hesbaye and in Fagne-Famenne natural areas are respectively significantly lower (Chi^2; $P < 0.001$) and higher (Chi^2; $P = 0.02$) than the others. In muskrat, the percentage of infection observed in Hesbaye and both in Fagne-Famenne and Ardenne natural areas are respectively significantly lower (Chi^2; $P = 0.001$) and higher (Chi^2; $P = 0.005$) than the others.

4. Discussion

As described in most European countries, a spectacular increase in the distribution and density of red foxes occurred in Belgium during the last two decades. This increase could be explained by a reduction in the mortality rate due to an intensive campaign of vaccination against rabies (oral baits), the

Table 2
Prevalence of *E. multilocularis* carriage in the red fox (*Vulpes vulpes*) and the muskrat (*Ondatra zibethicus*) in the different geological areas of Wallonia (2003–2004)

Natural area	Foxes N_t/N_p [a]	Percentage of infected foxes (95% CI) [b]	Muskrat N_t/N_p [a]	Percentage of infected muskrat (95% CI) [b]
Hesbaye	42/420	10 (7.3–13.3)	13/644	2.02 (1.08–3.43)
Condroz	45/181	24.86 (18.7–31.8)	15/227	6.61 (3.75–10.66)
Fagne-Famenne	21/34	61.8 (43.6–77.8)	80/347	23.06 (18.73–27.85)
Ardenne	87/213	40.84 (34.2–47.8)	65/320	20.31 (16.04–25.14)
Belgian Lorraine	48/142	33.8 (26.1–42.2)	19/180	10.56 (6.47–15.99)
Total	248/990	24.55 (22.38–27.87)	192/1718	11.18 (9.72–12.76)

[a] N_t: Number of tested animals, N_p: number of positive animals.
[b] The 95% confidence interval (binomial exact).

opportunist behavior of the red foxes and the nature conservation measures (Vervaeke et al., 2003). This work confirms a previous study (Losson et al., 2003) which indicated that *E. multilocularis* was widely distributed in Wallonia where environmental conditions are suitable for the establishment of the life cycle. The prevalences decrease from the South-Eastern part to the North-Western part of the region and this is probably related to the altitude (which decreases from the east to the west). However sporadically hyperendemic foci were recorded in the different investigated geological areas. Indeed heterogeneous spatial distribution patterns appear to be a particular epidemiological feature of the parasite (Tackmann et al., 1998) and this is due to the highly aggregated nature of the infection pressure (Morgan et al., 2004). This could, at least in part, explain the very high prevalence in Fagne-Famenne (61.8%). The present results indicate that adult foxes exhibit a lower prevalence as mentioned previously and this could be due to the induction of a specific immune response to the parasite or to variations in infection pressure (Torgerson, 2006). The present study when compared to another one dealing with the same parasite and performed in the same region of Belgium (Losson et al., 2003) seems to indicate a wider parasite distribution particularly in the low land. This phenomenon could be related to the migration of infected red foxes to new places potentially suitable to the establishment of the life cycle. This was recorded in Flanders (Northern part of Belgium) (Vervaeke et al., 2003). In Belgium the only available data were dealing with the carriage of *E. multilocularis* by the red fox (Losson et al., 2003; Vervaeke et al., 2003). In particular the role of microtine rodents and other potential intermediate such as the muskrat (Borgsteede et al., 2003) is unknown. In several studies highly endemic areas were usually associated with large population of different rodent species and specific climatic and environmental conditions (Giraudoux et al., 1997). Taking into account a high level of infection in red foxes (61.5%), high rainfalls (1176.15 $\pm$ 84.67 mm/year) and the presence of pastures favorable for the common voles (*M. arvalis*) and water vole (*A. terrestris*) (Giraudoux, 1991) which are considered as major intermediate host in Western and Central Europe (Eckert et al., 2001), it was surprising to find only 0.16% of small rodents to be infected with the metacestode. However only 2 animals out of 1249 belonged to the species *A. terrestris*. This was probably due to the type of traps used in the present study.

Nevertheless, the low level of infection in *M. arvalis* (1/914, i.e. 0.11%) would suggest that the species plays a minor role in the life cycle of *E. multilocularis* in the selected area. Alternatively the low prevalence rate in the species could be due to a high parasite-induced host mortality resulting in low observed prevalences in this host. In contrast a high level of infection (11.7%) was found in the muskrat with a good correlation (OR: 3.4495 (95% CI: 2.6947–4.4159)) between the prevalence in foxes and in muskrats in each of the different geological regions (Table 2). In fact, this species could be considered as a good bioindicator in the investigated tool territory. The survival of the eggs of *E. multilocularis* is dependent on the climatic conditions (Staubach et al., 2001) more particularly rainfalls and humidity. The humid soil conditions may be favorable for the persistence of the parasite and these suitable habitats are occupied by the muskrat. Additionally this species has a longer life span than the microtines and the percentage of infection was higher in the adults than in the juveniles. Different hypotheses can explain this observation. In *Echinococcus granulosus* infected intermediate hosts there is no evidence of parasite-induced immunity (Torgerson and Heath, 2003). Additionally, the growth of the metacestode of *E. multilocularis* in the rodent tissues requires a minimum of 2–4 months before the production of protoscoleces (Eckert et al., 2001). Under natural conditions the muskrat is probably not a common prey for the red fox. Other studies in France (Boussinesq et al., 1986), the Netherlands (Borgsteede et al., 2003) and Germany (Baumeister et al., 1997) reported the presence of *E. multilocularis* in the muskrat but at lower prevalences. In Belgium, the muskrat are trapped by professional team and the cadavers are left on river banks. In such conditions, freshly killed muskrats are available for the red foxes and then could play locally a major role in the life cycle maintenance. Studies are planned to verify this hypothesis. In conclusion this study confirms the wide distribution of *E. multilocularis* in Belgium although the carriage by the red fox varies widely from one place to another. The common vole (*M. arvalis*) did not seem to play a significant role as intermediate host. In contrast the muskrat is highly sensitive and could perhaps represent a good bioindicator when studying the epidemiology of this parasite infection in Belgium and elsewhere.

References

Baumeister, S., Pohlmeyer, K., Kuschfeldt, S., Stoye, M., 1997. Prevalence of *Echinococcus multilocularis* and other metacestodes and cestodes in the muskrat (*Ondatra zibethicus* Link 1795) in Lower Saxony. Deut. Tierärztl. Woch. 104, 448–452.

Boussinesq, M., Bresson, S., Liance, M., Houin, R., 1986. A new natural intermediate host of *Echinococcus multilocularis* in France: the muskrat (*Ondatra zibethicus* L). Ann. Parasitol. Hum. Comp. 61, 431–434.

Borgsteede, F., Tibben, J., Van der Giessen, J., 2003. The musk rat (*Ondatra zibethicus*) as intermediate host of cestodes in the Netherlands. Vet. Parasitol. 117, 29–36.

Brochier, B., Coppens, P., Losson, B., Aubert, M., Bauduin, B., Barrat, J., Costy, F., Peharpre, D., Pouplard, L., Pastoret, P.P., 1992. Enquête sur l'infestation du renard roux (*Vulpes vulpes*) par *Echinococcus multilocularis* en province du Luxembourg (Belgique). Ann. Med. Vet. 136, 497–501.

Dinkel, A., Von Nickisch-Rosenegk, M., Bilger, B., Merli, M., Lucius, R., Romig, T., 1998. Detection of *Echinococcus* multilocularis in the definitive host: coprodiagnosis by PCR as an alternative to necropsy. J. Clin. Microbiol. 36, 1871–1876.

Eckert, J., Gemmell, M., Meslin, F., Pawlowski, Z.S., 2001. WHO/OIE Manual on Echinococcosis in Humans and Animals: A Public Health Problem of Global Concern. World Health Organization, Paris.

Giraudoux, P., 1991. Utilisation de l'espace par les hôtes du ténias multiloculaire (*Echinococcus multilocularis*) conséquence épidémiologiques. Thèse Université de Bourgogne, 90 pp.

Giraudoux, P., Delattre, P., Habert, M., Quere, J.-P., Deblay, S., Defaut, R., Duhamel, R., Moissenet, M., Salvi, D., Truchetet, D., 1997. Population dynamics of fossorial water vole (*Arvicola terrestris* scherman): a land use and landscape perspective. Agr. Ecosyst. Environ. 66, 47–60.

Gottstein, B., Saucy, F., Deplazes, P., Reichen, J., Demierre, G., Busato, A., Zuercher, C., Pugin, P., 2001. Is high prevalence of *Echinococcus multilocularis* in wild and domestic animals associated with disease incidence in humans? Emerg. Infect. Dis. 7, 408–412.

Harris, S., 1978. Age determination in the red foxes (*Vulpes vulpes*): an evaluation of technique efficiency as applied to a sample of suburban foxes. J. Zool. 184, 91–117.

Hofer, S., Gloor, S., Muller, U., Mathis, A., Hegglin, D., Deplazes, P., 2000. High prevalence of *Echinococcus multilocularis* in urban red foxes (*Vulpes vulpes*) and voles (*Arvicola terrestris*) in the city of Zürich, Switzerland. Parasitology 120, 135–142.

Ito, A., Sako, Y., Yamasaki, H., Mamuti, W., Nakaya, K., Nakao, M., Ishikawa, Y., 2003. Development of EM 18-immunoblot and EM 18-Elisa for specific diagnosis of alveolar echinococcosis. Acta Trop. 00, 1–10.

Leiby, P., Carney, W., Woods, C.E., 1970. Studies on sylvatic echinococcosis. III: Host occurrence and geographic distribution of *Echinococcus multilocularis* in the north central United States. J. Parasitol. 56, 1141–1150.

Losson, B., Mignon, B., Brochier, B., Bauduin, B., Pastoret, P.P., 1997. Infestation du renard roux (*Vulpes vulpes*) par *Echinococcus multilocularis* dans la province du Luxembourg (Belgique): résultats de l'enquête effectuée entre 1993 et 1995. Ann. Med. Vet. 141, 149–153.

Losson, B., Kervyn, T., Detry, J., Pastoret, P.P., Mignon, B., Brochier, B., 2003. *Echinococcus multilocularis* in the red fox (*Vulpes vulpes*) in southern Belgium: a prevalence study. Vet. Parasitol. 117, 23–28.

Morgan, E.R., Milner-Gulland, E.J., Torgerson, P.R., Medley, G.F., 2004. Ruminating on complexity: macroparasites of wildlife and livestock. Trends Ecol. Evol. 19, 181–188.

Pankakoski, E., 1980. An improved method for age determination in the musk rat, *Ondatra zibethicus* (L). Ann. Zool. Fennici 17, 113–121.

Staubach, C., Thulke, H.H., Tackmann, K., Hugh-Jones, M., Conraths, F.J., 2001. Geographic information system-aided analysis of factors associated with spatial distribution of *Echinococcus multilocularis* infections of foxes. Am. J. Trop. Med. Hyg. 65, 943–948.

Storandt, S., Virchow, D., Dryden, M., Hygnstrom, S., Kazacos, K.R., 2002. Distribution and prevalence of *Echinococcus multilocularis* in wild predator in Nebraska, Kansas, and Wyoming. J. Parasitol. 88, 422–425.

Tackmann, K., Löschner, U., Mix, H., Staubach, C., Thulke, H.H., Conraths, F.J., 1998. Spatial distribution patterns of *Echinococcus multilocularis* (Leuckart 1863) (Cestoda: Cyclophyllidea: Taeniidae) among red foxes in an endemic focus in Brandenburg, Germany. Epidemiol. Infect. 120, 101–109.

Torgerson, P.R., 2006. Canid immunity to *Echinococcus* spp.: impact on transmission. Parasite Immunol. 28, 295–303.

Torgerson, P.R., Heath, D.D., 2003. Transmission dynamics and control options for *Echinococcus granulosus*. Parasitology 127, S143–S158.

Tsukada, H., Morishima, Y., Nonaka, N., Oku, Y., Kamiya, M., 2000. Preliminary study of the role of red foxes in *Echinococcus multilocularis* transmission in the urban area of Sapporo, Japan. Parasitology 120, 423–428.

Vervaeke, M., Dorny, P., Vercammen, F., Geerts, S., Brandt, J., Van den Berghe, K., Verhagen, R., 2003. *Echinococcus multilocularis* (Cestoda Taeniidae) in Red foxes (*Vulpes vulpes*) in northern Belgium. Vet. Parasitol. 115, 257–263.

Vuitton, D.A., Zhou, H., Bresson-Hadni, S., Wang, Q., Piarroux, M., Raoul, F., Giraudoux, P., 2003. Epidemiology of alveolar echinococcosis with particular reference to China and Europe. Parasitology 127, 87–107.