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INFECTION OF FOXES BY *ECHINOCOCCOCUS MULTILOCULARIS* IN URBAN AND SUBURBAN AREAS OF NANCY, FRANCE: INFLUENCE OF FEEDING HABITS AND ENVIRONMENT

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Summary:

This study evaluated the impact of biological and environmental factors on the infection of red foxes *(Vulpes vulpes)* by *Echinococcus multilocularis* in an endemic area of north-east France. From January 2004 to April 2006, 127 foxes were examined for *E. multilocularis* and their stomach contents analysed. The effect of year, season, age, sex and urbanisation level on *E. multilocularis* presence was estimated using a General Linear Model (GLM) with logit link, (*i.e.* logistic regression). Urbanisation level was the only influencing factor, with a decreasing gradient from rural [54 %, CI 95 % (40-68)] to peri-urban [31 %, CI 95 % (15-52)] and urban area [4 %, CI 95 % (0.7-15)]. The consumption of *Arvicola terrestris* and *Microtus* sp., grassland species, the main presumed intermediate hosts of *E. multilocularis*, was studied by the same approach. The two species were consumed less in the urban area and more in autumn than in spring. Anthropogenic food consumption was linked to urbanisation and to age. The frequency of anthropogenic food consumption decreased in the rural area. A global model explaining the presence of *E. multilocularis* and including urbanisation level and diet was then elaborated. Independently of urbanisation, there was a suggestion of less *E. multilocularis* infection with anthropogenic food consumption. Red foxes consuming *Microtus* sp. and *A. terrestris* had higher worm burden than those that did not. The results suggest that the decreasing gradient observed from rural to urban area is linked to behaviour and feeding habits.

KEY WORDS : *Echinococcus multilocularis*, *Vulpes vulpes*, urbanization, rodents, anthropogenic food.

Résumé : INFECTION DES RENARDS PAR *ECHINOCOCCUS MULTILOCULARIS* DANS LES ZONES URBAINES ET PÉRI-URBAINES DE NANCY, FRANCE : INFLUENCE DU RÉGIME ALIMENTAIRE ET DE L'ENVIRONNEMENT

*Cette étude évalue l'impact des facteurs biologiques et environnementaux sur l'infection du renard roux (*Vulpes vulpes*) par* Echinococcus multilocularis *dans une région endémique du nord-est de la France. De janvier 2004 à avril 2006, 127 renards ont été examinés pour le diagnostic d'*E. multilocularis *et une analyse du régime alimentaire a été réalisée à partir des contenus stomacaux. L'effet de l'année, de la saison, de l'âge, du sexe et du niveau d'urbanisation sur la présence d'*E. multilocularis *a été estimé en utilisant un modèle linéaire général (GLM) à lien logit (c.-à-d. régression logistique). Le niveau d'urbanisation était le seul facteur influençant la présence du parasite avec un gradient décroissant du secteur rural [54 %, IC 95 % (40-68)] à peri-urbain [31 %, IC 95 % (15-52)] et urbain [4 %, IC 95 % (0.7-15)]. La consommation d'*Arvicola terrestris *et de* Microtus arvalis, *espèces prairiales considérées comme hôtes intermédiaires principaux, a été analysée en utilisant le même type de modèle. Les deux espèces étaient moins consommées dans le secteur urbain et davantage consommées en automne qu'au printemps. La consommation de nourriture anthropogène était liée au niveau d'urbanisation et à l'âge. La fréquence de la consommation de nourriture anthropogène diminuait dans le secteur rural. Un modèle global expliquant la présence d'*E. multilocularis *comprenant le niveau d'urbanisation et le régime alimentaire a alors été ajusté. Indépendamment de la zone, les résultats suggèrent une diminution de l'infestation liée à la consommation de nourriture anthropogène. Les renards roux consommant les espèces* Microtus *sp. et* A. terrestris *indiquaient une charge parasitaire plus élevée. Ces résultats suggèrent que le gradient décroissant observé du secteur rural à urbain est lié aux changements comportementaux et alimentaires.*

MOTS CLÉS : Echinococcus multilocularis, Vulpes vulpes, *urbanisation, rongeurs, nourriture anthropogène.*

INTRODUCTION

The colonisation of cities by red foxes was initially a British phenomenon reported in the 1940s (Harris, 1977; Macdonald & Newdick, 1982). Since the beginning of the 1990s, an increased fox population has been observed in France and other European countries (Artois, 1997; Ruette & Stahl, 2003) leading to a colonisation of urban areas by foxes (Gloor *et al.*, 2001; Deplazes *et al.*, 2004). The changes in population dynamics were assumed to be, in part, the result of successful oral vaccination programs against rabies (Chautan *et al.*, 2000; Gloor *et al.*, 2001). Therefore, more red foxes are observed in urban areas and villages and the proximity of wild fauna to human populations may be a public health risk, particularly with regard to alveolar echinococcosis.

This is a lethal helminthosis of the northern hemisphere, due to infection of the liver by the larval stages of *Echinococcus multilocularis* (Schantz *et al.*, 1995).

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Transmission to human occurs through the accidental ingestion of eggs excreted by the final host. From 1982 to 2000, 42 % of the registered European human cases were diagnosed in France (Kern *et al.*, 2003). Although rare, this disease is a real public health concern due to high pathogenicity, late diagnosis and stringent treatment (Eckert & Deplazes, 2004). The parasite life cycle involves red foxes as definitive hosts and several small mammals as intermediate hosts. In Europe *Microtus arvalis* and *Arvicola terrestris,* two grassland species, are considered the main intermediate hosts (Giraudoux *et al.*, 2002). Domestic carnivores, such as dogs and cats, are potential hosts. Even though cats' behaviour could favour their infection, among rodents predation, their physiology presents a lower susceptibility to infection and eggs excretion (Kapel *et al.*, 2006). Contribution of domestic carnivores to human exposure in Europe is still unclear and their proximity to man may pose a major zoonotic risk (Petavy *et al.*, 2000; Romig *et al.*, 2006b). In Europe, the red fox is the main source of environmental contamination (Deplazes & Eckert, 2001). Recent studies have shown the presence of *E. multilocularis* in red fox in various cities: Copenhagen (Kapel & Saeed, 2000), Geneva (Fischer *et al.*, 2005), Prague (Martinek *et al.*, 2000), Stuttgart (Romig *et al.*, 1999), Zürich (Hofer *et al.*, 2000) in Europe, Sapporo (Tsukada *et al.*, 2000) and Otaru (Yimam *et al.*, 2002) in Japan. The prevalence among urban foxes was lower than among suburban or rural foxes. However it occurs a considerable infection risk to human due to the high fox population in cities (Romig *et al.*, 2006a). Definitive host contamination is due to eating rodents, and epidemiological studies have shown the impact of their availability on rural red fox infection (Delattre *et al.*, 1988; Giraudoux *et al.*, 2002; Raoul *et al.*, 2003) and more recently in urban red fox infection (Hegglin *et al.*, 2007). Diet analysis of urban red foxes has revealed changes in feeding habits, compared to the more traditional biotopes, with a larger consumption of anthropogenic food (Baker *et al.*, 2000; Contesse *et al.*, 2004; Doncaster *et al.*, 1990). Deplazes *et al.* (2004) point out that these changes may affect the life cycle of *E. multilocularis* in urban foxes.

The aim of this study was to investigate the effect of environmental and dietary factors on the infection of urban and suburban red foxes in eastern France. The results will be discussed in the light of earlier studies in other countries.

MATERIAL AND METHODS

Study site

The study site was the conurbation of Nancy, in the Lorraine region of north-east France, an endemic area for *E. multilocularis* (Aubert *et al.*, 1987). The study area (230 km^2) was divided into urban, peri-urban and rural zones (62, 75 and 93 km^2, respectively) (Fig. 1). The urban zone, with 255,632 inha-

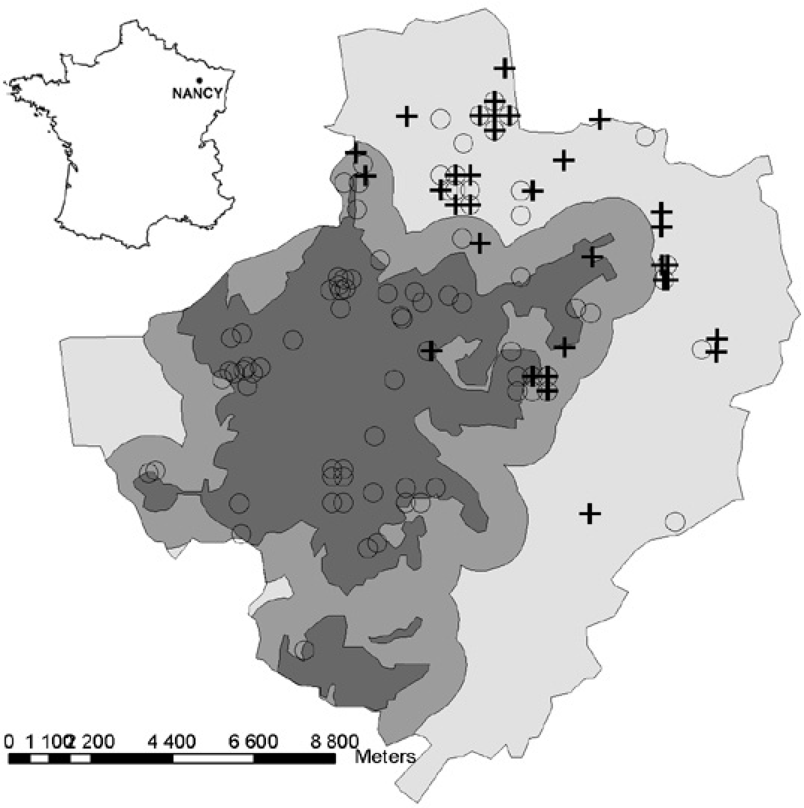

Fig. 1. – Location of the Nancy conurbation and the urban (dark gray), peri-urban (gray), rural (light gray) areas, with points of origin of foxes analysed for *Echinococcus multilocularis*. Location of positive cases: cross; location of negative cases: circle.

bitants (INSEE census, 1999), comprised the urban center and its suburbs. The urban zone was a built-up area including some recreational areas like parks, family gardens and small fields. The peri-urban zone was defined as a one km large buffer zone around the urban unit. The peri-urban landscape comprised a mix of recreational areas and agricultural areas, with pastures, fields, meadows and woodland. The rural zone was principally farmland.

Fox sampling

From January 2004 to April 2006, data from 127 red foxes were collected. Samples were taken from foxes caught by authorised trappers, foxes freshly killed on the road, or shot during hunting season, except in urban area where this is prohibited. It was assumed that the cause of the death did not influence the parasitic status of the animal. The geographical location, year, season of the death, sex and age were recorded for each animal. Age was evaluated by counting annual cementum layers in transversal sections of the lower canines (Artois & Salmon, 1981).

Parasitological examination

After safe collection, intestines were deep-frozen at – 80° C during 10 days for decontamination and stored at – 30° C. They were analysed according to the reference technique of sedimentation and counting (SCT) (Eckert *et al.*, 2001) with modifications. The whole intestine was incised longitudinally and cut into four pieces. All pieces were transferred into a 1 litre plastic jar with water. After shaking for a few seconds, pieces were extracted one after the other and intestinal mucous was scraped twice between fingers. After 30 minutes, the sediment was washed with water through a 1 mm sieve. After one hour, the sediment was re-suspended and a few ml were examined under stereoscopic microscope (50 × magnification) on a rectangular plastic dish with a counting grid. When *E. multilocularis* were not seen, all the sediment was examined. Where worms were detected, the total burden was estimated from the count of 20 % of the re-suspended sediment. *E. multilocularis* identification was based on morphology (Dorchies *et al.*, 2002).

Stomach analysis

Stomachs were collected at necropsy and stored at – 30° C. Their contents were removed and washed through a 0.5 mm sieve. The presence of *A. terrestris*, *Microtus* sp. and anthropogenic food was recorded. Rodents were identified by their teeth according to the method of Chaline *et al.* (1974). When rodent teeth were not found, identification of prey species was assessed by examining 10 µm cross sections of hairs, according to the methods of Day (1966) and Debrot *et al.* (1982). Anthropogenic food was presumed from the presence of pet food, plastic, metallic or paper packaging, and scavenged human food.

Statistical analysis

Logistic regression was used to analyse the association of urbanisation level, year, season, age class or sex with the probability of infection by *E. multilocularis*. Model was built considering urbanisation level as the most promising independent variable followed by season, age class, sex and possible interactions. Similar analyses were carried out on the impact of urbanisation level, year, season, age class or sex on the consumption of *Microtus* sp. and *A. terrestris* and anthropogenic food. Consumption was expressed as the proportion of stomachs containing the index food item among all stomachs, including the empty ones. Furthermore, logistic models combining the effects of environmental factors and dietary changes on fox infection were elaborated. Models were compared using the information theoretic approach as outlined by Burnham & Anderson (2002) corrected for small sample sizes (AICc) and presented according to the method of Anderson *et al.* (2001). The model considered the most appropriate for the data set was the model with lowest AICc value. When two AICc values differed by less than two, we selected the most parsimonious model, *i.e.*, with less parameters. Odds ratios (OR) were computed from the model parameters. Confidence intervals (CI) of predictive values were generated via bootstrapping of 1,000 replicates (Davison & Hinkley, 1997). The ability of the selected models to fit the data was assessed with the Pearson goodness-of-fit test. The discriminatory performance for final logistic equation was tested with a Receiver Operating Characteristic (ROC) approach. The area under the ROC curve (AUC) defined by the false positive rates and true positive rates was used as a measure of accuracy. The nearer to one, the more accurately the model describes the data (Fielding & Bell, 1997). The distribution of worm burden among zones and among food items, being far from normal, with few extremely high values, comparisons were performed using a Kruskal-Wallis test. Analyses were performed using R 2.4.1 (R Development Core Team, 2004).

RESULTS

127 foxes were collected, including 49 from urban area (39 %), 26 from peri-urban area (20 %) and 52 from rural area (41 %). 56 were collected in winter, 36 in spring, 18 in summer and 17 in autumn. The sex ratio of 1.27 revealed higher proportion of males. The age range was divided into four classes: 0-1 year

Model	LL	K	n/K	AICc	Δ_i	w_i
Zone	– 60.3	3	42.3	126.8	0.0	0.94
Zone + Season	– 59.8	6	21.2	132.3	5.5	0.06
Zone + Season + Age	– 59.0	10	12.7	139.9	13.1	0
Zone + Season + Age + Sex	– 56.6	13	9.8	142.4	15.6	0
Zone + Season + Age + Sex + Year	– 56.3	15	8.5	147.0	20.2	0
Null model	– 77.5	1	127.0	157.0	30.2	0

LL = maximised log-likelihood; K = number of estimated parameters; n/K = number of observations/K; AICc = second-order Akaike index criterion; Δ_i = difference between AICc and minimum AICc; w_i = Akaike weights.

Table I. – Model selection to explain *E. multilocularis* infection with environmental and biological factors as descriptive variables.

(53 foxes), 1-2 years (22), 2-3 years (22) and more than 3 years (21). The age of nine specimens could not be determined since their jaws were crushed.

E. MULTILOCULARIS INFECTION

38 foxes (30 %) were infected. Two of 49 urban foxes were positive. They were juvenile males found close to the peri-urban zone (Fig. 1). The distribution of the eight positive cases in peri-urban area revealed that six were collected close to the urban area, with three in the same place. Table I shows that second-order Akaike index criterion (AICc) values were lower in the model with only a zone effect. The model failed to detect any effect of year, season, age and sex. The prevalence in urban foxes was lower than in rural foxes (OR = 0.04; CI = 0.01-0.14) with a small difference between peri-urban and rural foxes (OR = 0.38; CI = 0.14-1.01). Prevalence in rural (54 %, CI 40-68 %), in peri-urban (31 %, CI 15-52 %) and in urban area (4 %, CI 0.7-15 %) shows a decreasing gradient of infection from rural to urban area (Fig. 2). Pearson goodness-of-fit test (p = 0.41) and AUC of 0,78 indicated a good fit for the model.

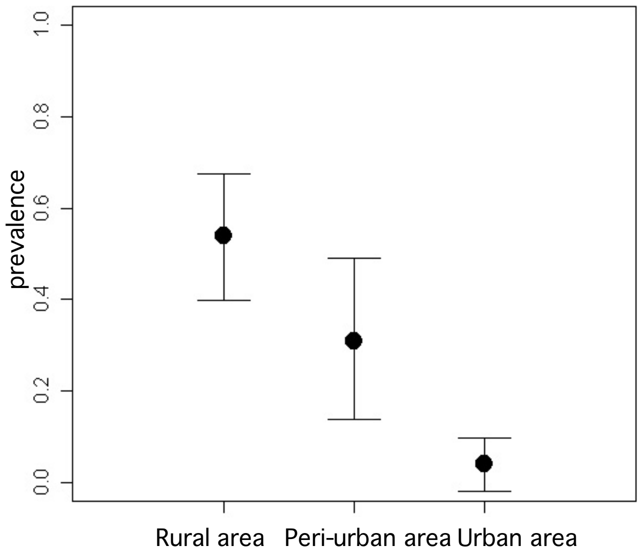

Fig. 2. – Prevalence of *Echinococcus multilocularis* in red foxes according to urbanisation level. Lines represent 95 % confidence intervals.

E. MULTILOCULARIS BURDEN

The infection intensity ranged from two to 135,565 adult worms, with a total of 163,070 (Table II). A Kruskal-Wallis test failed to detect differences in worm burden between zones (p = 0.169). The total biomass of worms in the urban area (84 adult worms) represented 0.05 % of the total biomass in the study area, while rural red foxes harboured 13 % and peri-urban foxes 87 % of the total biomass. 37 % of the infected foxes harboured less than 100 worms and 26 % more than 1,000, with the highest score of 135,565 in one juvenile peri-urban fox representing 83 % of the total worm biomass.

ARVICOLA TERRESTRIS AND *MICROTUS* SP. CONSUMPTION

33 stomachs were empty (35 %) and 94 had contents. 18 % (CI 11-28 %) of non-empty stomachs contained *A. terrestris* and/or *Microtus* sp , *i.e.* 13 % (CI 8-21 %) of all stomachs. Comparison of models revealed that AICc (Table III) were lower in the model including zone and seasonal effects (Pearson goodness-of-fit test: p = 0.67, AUC = 0.75). The predictions of the selected model

	No. of analysed foxes	No. of infected foxes	E. m biomass (%)	Median	Minimum	Maximum	Total worm burden
Rural area	52	28	13	190	2	7220	21,100
Peri-urban area	26	8	87	840	6	135,565	141,886
Urban area	49	2	0.05	42	4	80	84
Total	127	38	100	190	2	135,565	163,070

Table II. – Distribution of *E. multilocularis* worm burden in red foxes along the gradient of urbanisation.

are presented in figure 3. Consumption frequency was lower in the urban area than in the rural area (OR = 0.08, CI = 0.01-0.44) whereas no clear influence of peri-urban areas was observed (OR = 0.80, CI = 0.20-2.79). A seasonal variation revealed a higher consumption in autumn than in spring (OR = 13.12, CI = 2.16-115.27). This phenomenon was observed mainly for rural and peri-urban foxes (Fig. 3). No clear difference was obser-

a) *Microtus* sp. and *Arvicola terrestris* consumption

Model	LL	K	n/K	AICc	Δ_i	w_i
Zone + Season	– 42.2	6	21.2	97.1	0.0	0.7
Zone	– 46.5	3	42.3	99.3	2.2	0.23
Null model	– 50	1	127	102	4.9	0.06
Zone + Season + Age	– 41.7	10	12.7	105.3	8.2	0.01
Zone + Season + Age + Sex	– 41.3	13	9.8	111.8	14.7	0
Zone + Season + Age + Sex + Year	– 40.8	15	8.5	115.8	18.7	0

b) Anthropogzenic food consumption

Model	LL	K	n/K	AICc	Δ_i	w_i
Zone + Age	– 44.8	7	18.1	104.5	0	0.78
Zone	– 50.9	3	42.3	107.9	3.4	0.14
Zone + Age + Season	– 43.6	10	12.7	109.1	4.6	0.08
Zone + Age + Season + Sex	– 43.0	13	9.8	115.3	10.8	0
Null model	– 57.0	1	127.0	115.9	11.4	0
Zone + Age + Season + Sex + Year	– 42.7	15	8.5	119.7	15.2	0

LL = maximised log-likelihood; K = number of estimated parameters; n/K = number of observations/K; AICc = second-order Akaike index criterion; Δ_i = difference between AICc and minimum AICc; w_i = Akaike weights.

Table III. – a) Model selection to explain *Microtus* sp. and *Arvicola terrestris* consumption by red foxes; b) Model selection to explain anthropogenic food consumption by red foxes.

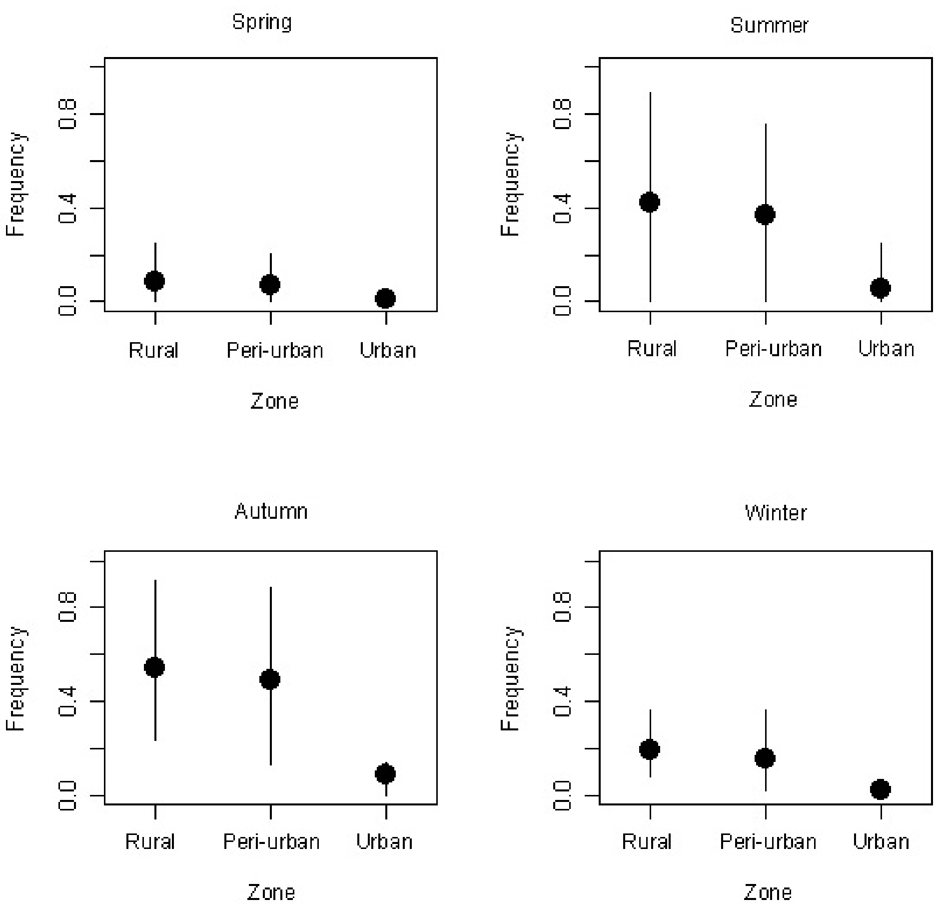

Fig. 3. – Occurrence frequency and 95 % confidence interval of *Arvicola terrestris* and/or *Microtus* sp. in fox stomachs from rural, peri-urban and urban areas during consecutive seasons.

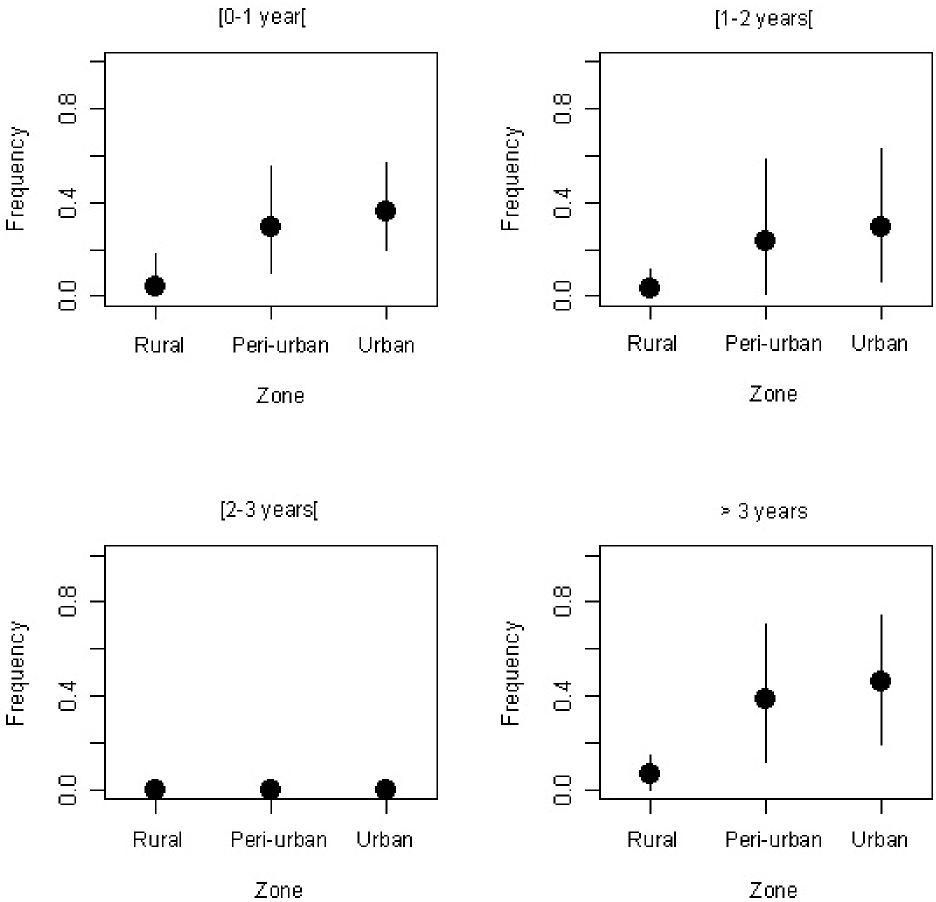

Fig. 4. – Occurrence frequency and 95 % confidence interval of anthropogenic food in fox stomachs from rural, peri-urban and urban area along consecutive age classes.

ved between winter and spring (OR = 2.60, CI = 0.59-18.28). The large confidence interval seen in summer (OR = 7.93, CI = 0.72-97.44) does not allow an estimate of seasonal influence.

Anthropogenic food consumption

Anthropogenic food consumption was seen in 22 % (CI 15-32 %) of the stomach with contents, *i.e.* 13 % (CI 11-24 %) of all stomachs. AICc (Table III) were lower in the model including zone and age (Pearson goodness-of-fit test: p = 0.79, AUC = 0.80). Compared to the rural area data, frequency of anthropogenic food consumption increased in peri-urban (OR = 8.92, CI = 1.72-69.77) and more significantly in urban areas (OR = 12.67, CI = 2.94-90.80). Predictive values and confidence intervals revealed no differences between peri-urban and urban areas for each age class (Fig. 4). For 2-3 years old foxes, no consumption of anthropogenic food was found.

Impact of diet on *E. multilocularis* infection

The smallest AICc were observed for the model including the effect of zone and anthropogenic consumption. However, the small observed differences between AICc values (< 2) did not allow discrimination between "zone + anthro" and "zone" and "zone + anthro + mat" (Table IV). Retaining the most parsimonious model, prevalence was mainly explained by the zone (Pearson goodness-of-fit test: p = 0.41, AUC = 0.78). Foxes from the urban area had the lowest prevalence (OR = 0.04, CI = 0.007-0.17) with a likely decrease of infection prevalence of foxes with anthropogenic food consumption (OR = 0.17, CI = 0.009-1) independent of urbanisation. Among infected foxes, a difference in worm burden was detected between those animals having eaten *A. terrestris* and/or *Microtus* sp. and those that did not; Kruskal-Wallis test (p = 0.018).

DISCUSSION

Gradient of infection and urbanisation level

This study records the presence of the parasite in the urban area of a French city with a decreasing gradient from rural to urban area. The same trend was observed in Switzerland: Zürich (Hegglin *et al.*, 2007; Hofer *et al.*, 2000) and Geneva (Fischer *et al.*, 2005). The first report of an urban cycle for *E. multilocularis* concerned the city of Zürich. Diagnosis in red foxes in this city revealed a high preva-

Model	LL	K	n/K	AICc	Δ_i	w_i
Zone + Anthro	– 58.4	4	31.8	125.1	0.0	0.50
Zone	– 60.3	3	42.3	126.8	1.7	0.21
Zone + Anthro + Mat	– 58.2	5	25.4	126.9	1.8	0.20
Zone + Mat	– 60.1	4	31.8	128.6	3.5	0.09
Anthro	– 72.6	2	63.5	149.3	24.2	0.00
Mat	– 76.2	2	63.5	156.5	31.4	0.00

Anthro = anthropogenic food consumption; Mat = *Microtus* sp. and *Arvicola terrestris* consumption; LL = maximised log-likelihood; K = number of estimated parameters; n/K = number of observations/K; AICc = second-order Akaike index criterion; Δ_i = difference between AICc and minimum AICc; w_i = Akaike weights.

Table IV. – Model selection to explain *E. multilocularis* infection.

lence of *E. multilocularis* between 16.5 % in the urban area, 39.5 % in adjacent areas and 62.9 % in peri-urban area (Hegglin *et al.*, 2007). In the present study, the gradient of infection reached 4 % in the urban area, 31 % in the peri-urban area and 54 % in the rural area. This may indicate a higher disturbance of transmission dynamics in the urban area of Nancy compared to peri-urban and rural areas. The analysis of the effect of urbanisation level, year, season, age class and sex have shown that urbanisation only explained fox infection. Other studies have revealed a seasonal prevalence in an urban area with higher rate in winter for young males (Hofer *et al.*, 2000) and more pronounced variation along the urbanisation gradient in juveniles foxes than in adult foxes (Hegglin *et al.*, 2007). In our study urban positive cases were juveniles males. Though the real origin of those juveniles could not be established, this finding may be due to the dispersion of juveniles from the peri-urban area. It revels that more attention must be deployed on the sex and age of foxes for the understanding of the urban dynamics transmission.

DIETARY CHANGES IN RED FOXES

The studies in rural areas showed that, for each stage of the parasite and on various scales (individual, population, biotope, etc.), several environmental and behavioural variables influenced distribution and parasite dynamics (Deplazes *et al.*, 2004; Giraudoux *et al.*, 2003; Miterpakova *et al.*, 2006). In Europe, at the regional level, it has been shown that transmission to the definitive host requires focal rodent species like *M. arvalis* and *A. terrestris* (Delattre *et al.*, 1988). Hegglin *et al.* (2007) have observed that urbanisation lead to a decrease of vole species consumption by foxes and to a decrease of *E. multilocularis* prevalence in this definitive host. Same trend was observed in Nancy. The present study provides evidence of fewer *A. terrestris* and *Microtus* sp. in the urban fox diet than in the peri-urban. No clear difference was observed between peri-urban and rural consumption of the intermediate hosts. This may be due to the decreased availability of voles because of a higher urbanisation, and/or to changes in the foxes' feeding habits. Giraudoux *et al.* (2001), Pleydell *et al.* (2004) and Raoul *et al.* (1999) have emphasised the impact of grassland on transmission dynamics, and Stieger *et al.* (2002) have found evidence of infected *A. terrestris* in the lawns of urban parks within the city of Zürich. A comparison of the features of green areas and parks in different cities may prove an interesting way to understand how urban planning may impact transmission. In our study, there was less anthropogenic food consumption in the rural zone than in the peri-urban zone. Such findings are consistent with those from other European cities (Baker *et al.*, 2000; Contesse *et al.*, 2004; Doncaster *et al.*, 1990). However, Contesse *et al.* (2004) described a zone-related variation of the anthropogenic food category, with a higher consumption in the urban area than in the peri-urban area. In our study, no clear zone-related distinction in anthropogenic food consumption was observed between the peri-urban and urban areas. Our results also indicated that, for each urbanisation level, anthropogenic food consumption affected negatively the proportion of infected foxes, and worm burdens were heavier where foxes ate *A. terrestris* and *Microtus* sp. An understanding of the dynamics of *E. multilocularis* infection at the interface of an endemic area and a low infection zone might be useful for proposing new strategies to control the parasite.

INFECTION PRESSURE IN URBAN AND RECREATIONAL AREA

Our findings, and those of earlier studies, suggest that more attention should be paid to peri-urban red foxes. These animals have feeding habits closer to those of rural foxes. In our study, the peri-urban prevalence of *E. multilocularis* (31 %) was close to the rural prevalence (54 %). Moreover, one peri-urban juvenile fox harboured 83 % of the total worm biomass sampled. The peri-urban area of the city of Nancy has many recreational areas and private gardens. Therefore, fewer peri-urban red foxes could contaminate this highly used environment. Furthermore, the parasite population seemed to be highly aggregated spatially, which could lead to micro-foci of infection, hard to detect, rather than to a uniform distribution of infective material. In Europe, infected people are mainly farmers, dog owners, people in contact with foxes (Kern *et al.*, 2004), and people living in areas of high vole density (Viel *et al.*, 1999). Since the year 2000, the incidence of human cases in Switzerland has increased. This emerging epidemic situation is in accordance with the increase of red fox population and the urbanisation of *E. multilocularis* cycle that occurred 10-15 years earlier and corresponds to the time delay of clinical manifestation in humans (Schweiger *et al.*, 2007). In regards to the increased risk of infection for man, control programs have already been carried out. Treatment with praziquantel baits has been initiated in Zürich (Hegglin *et al.*, 2003) and in progress in Sapporo (Nonaka *et al.*, 2005) and we suggest that the study of environmental contamination on a detailed field scale may be helpful for such planning control measures.

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REFERENCES

ANDERSON D.R., LINK W.A., JOHNSON D.H. & BURNHAM K.P. Suggestions for presenting the resuls of data analyses. *Journal of Wildlife Management*, 2001, *65*, 373-378.

ARTOIS M., LANGLAIS M. & SUPPO C. Simulation of rabies control within an increasing fox population. *Ecological Modelling*, 1997, *97*, 23-34.

ARTOIS M. & SALMON D. Détermination expérimentale du sexe et de l'âge chez le renard roux (*Vulpes vulpes*): Validité et reproductibilité des techniques choisies. *Mammalia*, 1981, *45*, 375-378.

AUBERT M., JACQUIER P., ARTOIS M., BARRAT M.J. & BASILE A.M. Le portage d'*Echinococcus multilocularis* par le renard (*Vulpes vulpes*) en Lorraine. Conséquences sur la contamination humaine. *Recherche Médicale Vétérinaire*, 1987, *163* (10), 839-843.

BAKER P.J., FUNK S.M., HARRIS S. & WHITE P.C.L. Flexible spatial organization of urban foxes, *Vulpes vulpes*, before and during an outbreak of sarcoptic mange. *Animal Behaviour*, 2000, *59*, 127-146.

BURNHAM K.P. & ANDERSON D.R. Model selection and multimodel inference: a practical information-theoretic approach, 2nd ed. Springer-Verlag, New York, 2002.

CHALINE J., BAUDVIN H., JAMMOT D. & SAINT GIRONS M.C. Les proies des rapaces - petits mammifères et leur environnement. Doin, Paris, 1974.

CHAUTAN M., PONTIER D. & ARTOIS M. Role of rabies in recent demographic changes in red fox (*Vulpes vulpes*) population in Europe. *Mammalia*, 2000, *64*, 391-410.

CONTESSE P., HEGGLIN D., GLOOR S., BONTADINA F. & DEPLAZES P. The diet of urban foxes (*Vulpes vulpes*) and the availability of anthropogenic food in the city of Zurich, Switzerland. *Mammalian Biology - Zeitschrift fur Saugetierkunde*, 2004, *69*, 81-95.

DAVISON A.C. & HINKLEY D.V. Bootstrap methods and their application. Cambridge University Press, 1997.

DAY M.G. Identification of hair and feather remains in the guts and faeces of stoats and waesels. *Journal of Zoology*, 1966, *148*, 201-217.

DEBROT S., FIVAS G., MERMOD C. & WEBER J.M. Atlas des poils de mammifères d'Europe, Institut de Zoologie de l'Université de Neuchâtel. Edition Neuchâtel, 1982.

DELATTRE P., PASCAL M., GIRAUDOUX P. & DAMANGE J.P. Caractéristiques écologiques et épidemiologiques de l'*Echinococcus multilocularis* au cours d'un cycle complet des populations d'un hôte intermédiaire (*Microtus arvalis*). *Canadian Journal of Zoology*, 1988, *66*, 2740-2750.

DEPLAZES P. & ECKERT J., Veterinary aspects of alveolar echinococcosis, a zoonosis of public health significance. *Veterinary Parasitology*, 2001, *98*, 65-87.

DEPLAZES P., HEGGLIN D., GLOOR S. & ROMIG T. Wilderness in the city: the urbanization of *Echinococcus multilocularis*. *Trends in Parasitology*, 2004, *20*, 77-84.

DONCASTER C.P., DICKMAN C.R. *et al.* Feeding ecology of red foxes (*Vulpes vulpes*) in the city of Oxford, England. *Journal of Mammalogy*, 1990, *71* (2), 188-194.

DORCHIES P., KILANI M. & MAGNAVAL J.F. *Echinococcus granulosus* et *Echinococcus multilocularis*. Les animaux et l'homme exposés aux mêmes dangers. *Bulletin de la société Vétérinaire Pratique de France*, 2002, *86* (2), 74-90.

ECKERT J. & DEPLAZES P. Biological, epidemiological, and clinical aspects of echinococcosis, a zoonosis of increasing concern. *Clinical Microbiology Review*, 2004, *17*, 107-135.

ECKERT J., DEPLAZES P., CRAIG P.S., GEMMEL M.A., GOTTSTEIN B., HEATH D., JENKINS D.J., KAMIYA M. & LIGHTOWLERS M. Echinococcosis in animals: clinical aspects, diagnosis and treatment, *in*: WHO/OIE manual on echinococcosis in humans and animals: a public health problem of global concern. Eckert J.G., M.A Meslin, F.X., Pawlowski, Z.S. (eds), WHO/OIE, 2001, 72-99.

FIELDING A.H. & BELL J.F. A review of methods for the presence/absence models. *Environmental Conservation*, 1997, *24*, 38-49.

FISCHER C., REPERANT L.A., WEBER J.M., HEGGLIN D. & DEPLAZES P. *Echinococcus multlocularis* infections of rural, residential and urban foxes (*Vulpes vulpes*) in the canton of Geneva, Switzerland. *Parasite*, 2005, *12*, 339-346.

GIRAUDOUX P., CRAIG P.S., DELATTRE P., BAO G., BARTHOLOMOT B., HARRAGA S., QUERE J.P., RAOUL F., WANG Y., SHI D. & VUITTON D.A. Interactions between landscape changes and host communities can regulate *Echinococcus multilocularis* transmission. *Parasitology*, 2003, *127* (Suppl,), S121-S131.

GIRAUDOUX P., DELATTRE P., TAKAHASHI K., RAOUL F., QUERE J.P., CRAIG P.S. *et al.* Transmission ecology of *Echinococcus multilocularis* in wildlife: what can be learned from comparative studies and multi-scale approaches? *In*: Echinococcoses: a global problem of emerging disease. Craig P. (ed.), NATO Sciences series, London, 2002, 251-266.

Giraudoux P., Raoul F., Bardonnet K., Vuillaume P., Tourneux F., Cliquet F., Delattre P. & Vuitton D.A. Alveolar echinococcosis: characteristics of a possible emergence and new perspectives in epidemiosurveillance. *Médecine et Maladies Infectieuses*, 2001, *31*, 247-256.

Gloor S., Bontadina F., Hegglin D., Deplazes P. *et al.* The rise of urban fox populations in Switzerland. *Mammalian. Biology*, 2001, *66*, 155-164.

Harris S. Distribution, habitat utilization and age structure of a suburban fox (*Vulpes vulpes)* population. *Mammal Review*, 1977, *7*, 25-39.

Hegglin D., Bontadina F., Contesse P., Gloor S. & Deplazes P. Plasticity of predation behaviour as a putative driving force for parasite life-cycle dynamics: the case of urban foxes and *Echinococcus multilocularis* tapeworm. *Functional Ecology*, 2007, *21*, 552-560.

Hegglin D., Ward P.I. & Deplazes P. Anthelmintic baiting of foxes against urban contamination with *Echinococcus multilocularis. Emerging Infectioud Diseases*, 2003, *9*, 1266-1272.

Hofer S., Gloor S., Muller U., Mathis A., Hegglin D. & Deplazes P. High prevalence of *Echinococcus multilocularis* in urban red foxes (*Vulpes vulpes*) and voles (*Arvicola terrestris*) in the city of Zurich, Switzerland. *Parasitology*, 2000, *120* (Pt 2), 135-142.

Kapel C.M.O. & Saeed I. *Echinococcus multilocularis* – en ny zoonotisk parasit I Denmark. *Dansk Veterinaer*, 2000, *83*, 14-16.

Kapel C.M.O., Torgerson P.R., Thompson R.C.A. & Deplazes P. Reproductive potential of *Echinococcus multilocularis* in experimentally infected foxes, dogs, raccoon dogs and cats. *International Journal for Parasitology*, 2006, *36*, 79-86.

Kern P., Ammon A., Kron M., Sinn G., Sander S., Petersen L.R., Gaus W. & Kern P. Risk factors for alveolar echinococcosis in humans. *Emerging Infectious Diseases*, 2004, *10* (12), 2088-2093.

Kern P., Bardonnet K., Renner E., Auer H., Pawlowski Z., Ammann R.W. & Vuitton D.A. European echinococcosis registry: human alveolar echinococcosis, Europe, 1982-2000. *Emerging Infectious Diseases*, 2003, *9*, 343-349.

Macdonald D.W. & Newdick M.T. The distribution and ecology of foxes, *Vulpes vulpes*, in urban areas. Proceedings of 2nd european ecological symp. *Urban ecology*, 1982, 123-135.

Martinek K., Kolãrovã L. & Cerveny J. Echinococcus multilocularis in carnivores from the Klatovy distrtrict of the Czech Republic. *Public Health Nutrition*, 2000, *75* (1), 61-66.

Miterpakova M., Dubinsky P., Reiterova K. & Stanko M. Climate and environmental factors influencing *Echinococcus multilocularis* occurrence in the Slovak Republic. *Annals of Agricultural and Environmental Medicine*, 2006, *13*, 235-242.

Nonaka N., Kamiya M. & Oku Y. Towards the control of *Echinococcus multilocularis* in the definitive host in Japan. *Parasitology International*, 2005, *55*, S263-S266.

Petavy A.F., Tenora F., Deblock S. & Sergent V. *Echinococcus multilocularis* in domestic cats in France. A potential risk factor for alveolar hydatid disease contamination in humans. *Veterinary Parasitology*, 2000, *87*, 151-156.

Pleydell D.R.J., Raoul F., Tourneux, F. Danson F.M., Graham A.J., Craig P.S. & Giraudoux P. Modelling the spatial distribution of *Echinococcus multilocularis* infection in foxes. *Acta Tropica*, 2004, *91*, 253-265.

R Development Core Team. R: a language and environment for statistical computing. Computing R.F.f.S., ed., Vienna, Austria, 2004.

Raoul F., Depierre V., Scheifler R., Mass R., Petavy A.F., Vuitton D. & Giraudoux P. Landscape effect on *Echinococcus multilocularis* winter prevalence in foxes in the Jura massif, France. *Helminthologia*, 1999, *36* (Suppl.), 48.

Raoul F., Michelat D., Ordinaire M., Decote Y., Aubert M., Delattre P., Deplazes P. & Giraudoux P. *Echinococcus multilocularis*: secondary poisoning of fox population during a vole outbreak reduces environmental contamination in a high endemicity area. *International Journal for Parasitology*, 2003, *33*, 945-954.

Romig T., Bilger B., Dinkel A., Merli M. & Mackenstedt U. *Echinococcus multilocularis* in animal hosts: new data from western Europe. *Helminthologia*, 1999, *36* (3), 185-191.

Romig T., Dinkel A. & Mackenstedt U. The present situation of echinococcosis in Europe. Taeniasis/cysticercosis and echinococcosis with focus on Asia and the Pacific. *Parasitology International*, 2006a, 55, S187-S191.

Romig T., Thoma D. & Weible A.K. Echinococcus multilocularis: a zoonosis of anthropogenic environments? *Journal of Helminthology*, 2006b, *80*, 207-212.

Ruette S. & Stahl P. Comparaison entre les comptages nocturnes de renards réalisés en dix ans d'intervalles dans neuf régions françaises. *Faune sauvage*, 2003, *258*, 47-49.

Schantz P., Chai J., Craig P.S., Eckert J., Jenkins D.J. & Macpherson C.N.L. Epidemiology and control of hydatid disease, *in*: *Echinococcus* and hydatid disease. Thompson R.C.A., Lymbery A.J. (eds). CAB International, Wallingford Oxon, 1995, 233-331.

Schweiger A., Ammann R.W., Candinas D., Clavien P.A., Eckert J., Gottstein B., Halkic N., Muellhaupt B., Prinz B.M., Reichen J., Tarr P.E., Torgerson P.R. & Deplazes P. Human alveolar echinococcosis after fox population increase, Switzerland. *Emerging Infectious Diseases*, 2007, *13*, 878-882.

Stieger C., Hegglin D., Schwarzenbach G., Mathis A. & Deplazes P. Spatial and temporal aspects of urban transmission of *Echinococcus multilocularis. Parasitology*, 2002, *124*, 631-640.

Tsukada H., Morishima Y., Nonaka N., Oku Y. & Kamiya M. Preliminary study of the role of red foxes in *Echinococcus multilocularis* transmission in the urban area of Sapporo, Japan. *Parasitology*, 2000, *120* (Pt 4), 423-428.

Viel J.F., Giraudoux P., Abrial V. & Bresson-Hadni S. Water vole (*Arvicola terrestris* Scherman) density as risk factor for human alveolar echinococcosis. *American Journal of Tropical Medecine and Hygiene*, 1999, *61*, 559-565.

Yimam A.E., Nonaka N., Oku Y. & Kamiya M. Prevalence and intensity of *Echinococcus multilocularis* in red foxes (*Vulpes vulpes* Schrencki) and raccoon dogs (*Nyctereutes procyonoides albus*) in Otaru City, Hokkaido, Japan. *Japan Journal of Veterinary Research*, 2002, *49*, 287-296.

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