Parasitological survey of rats in rural regions of Croatia

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ABSTRACT: A parasitological survey of 255 rats, trapped in rural regions of Croatia, was carried out. The survey revealed infection with 7 helminth species (2 cestodes, 5 nematodes and eggs of 2 accidental parasites). Also, 8 ectoparasite species (2 fleas, 3 lice, and 3 mites) were found. Overall infection rate was 72.6% (185 infected out of 255 rats). Eighty two of 255 rats (32.2%) were infected with ectoparasites and 166 out of 255 (65.1%) with endoparasites. Among endoparasites the most frequent parasite was *Hymenolepis diminuta* (36.9%) and among ectoparasites *Polyplax spinulosa* (14.5%). Analysing association of parasite infection with sex, age and location (household and village) we found out that none of the found parasites was sex related. On the other hand, several parasite infections increased with age and depended on habitat. As accidental finding, study showed that rats with mites tended to be infected with fleas and rats with nematodes tended to be infected with cestodes.

Keywords: rat; endoparasites; ectoparasites; prevalence; risk factors

Synanthropic rodents, particularly those living in close association with man, play a significant role in human health, welfare and economy. It has to be stressed that their arthropod ectoparasites are important vectors of pathogenic microorganisms and they can also be important reservoirs for parasitic zoonoses like trichinellosis and capillariosis. No doubt, the increase in rodent populations can be followed by the increase in zoonotic diseases. Hepatic capillariosis is a zoonosis seldom described in people. Human infection occurs by the consumption of the food or water contaminated with embryonated eggs, previously released from rat liver through cannibalism, predation or decomposition of carcasses. Different authors have reported a wide range of parasites but most investigations were carried out in tropical areas. Nama and Parihar (1976) reported 8 helminth species, found in 149 trapped rats in India. Yen *et al.* (1996) found in China the overall infection rate of 29.6% in 199 rats. Namue and Wongsawad (1997) found 33 out of 38 trapped rats to be infected (86.8%)

in Thailand. In Egypt, Abd el-Wahed *et al.* (1999) reported overall infection rate of 54% in 172 rats. Studies of parasites in brown rats in Europe have been limited to just few reports. The most detailed investigations were carried out in order to determine the prevalence of *Capillaria hepatica*. Ceruti *et al.* (2001) screened 47 brown rat livers for the presence of *C. hepatica* infection and found 36% to be infected. In France Davoust *et al.* (1997) trapped 82 rats and found 44% to be infected with *C. hepatica* and 21% with fleas. The most striking were the results of Farhang-Azad (1977) who examined 845 brown rats and found 75% to be infected with *C. hepatica*. The aim of our study was to define the prevalence of parasite infections in brown rats in Croatia since similar investigations were not carried out in our country. Also, we wanted, to compare the prevalence data of parasites in two different rural regions and investigate the influence of habitat on parasitic infections. In addition, we wanted to establish relation of parasite infections with sex and age of rats.

MATERIAL AND METHODS

The investigation was carried out in two randomly selected rural regions of Croatia in period of February–June 2001. The rats were captured in 19 village households in 2 randomly selected villages. At the beginning of the study 20 households were randomly selected from each village. In the village of Otok (A) 17 (85%) and in the village of Domagovic (B) 2 (10%) households agreed to participate in the study.

In both villages, very low biosanitary conditions were observed (especially in village B), such as open garbage dumps (including live stock carcasses) and drainage system was poor or absent. The investigation was carried out in small private rural farms where pigs have been reared, exclusively. In most farms a variety of poultry, cats and dogs were present as well.

Animals

A total of 255 Norway rats (*Rattus norvegicus*) were trapped alive and euthanized individually with ether in a glass container. The carcasses and the paper with which the glass vessel was wiped were closed in marked plastic bags and sealed. The bags were transported to laboratory, and kept at 4°C until examination. The rats were examined in the period of 24 h after trapping. Among them, 210 rats originated from 17 households of the village of Otok (A) and 45 from 2 households of the village of Domagović (B). Depending on the total weight and length of body, rats were categorised in 3 groups, as juveniles, subadults and adults. There were 51 (20%) juveniles, defined by the weight of 0–100 g and the length of 0–15 cm, 83 (32.6%) were subadults, weighted 100–300 g and their length was 15–30 cm and 121 (47.5%) were defined as adults, by the weight of 300–500 g and the length of 30–45 cm. Gender was determined by visual inspection of external sexual organs. Among 255 rats 96 (37.7%) were males and 159 (62.4%) were females.

Parasitological examination

Before autopsy, the fur and the skin were macroscopically examined for the presence of fleas and lice. The skin scrapings and hair specimens were microscopically examined for the presence of mites and lice. All rats were eviscerated and intestines were slit lengthwise. The gut contents were examined by eye inspection followed by the light microscope examination. In addition, the presence of capillarid eggs was determined by screening of faecal speciments from ampula recti with sodium chloride flotation method. The livers were examined for the presence of metacestodes. In order to define the prevalence of eggs, adult or larval stages of *C. hepatica*, livers were inspected and when lesions were noted further examination of the tissue was performed. All other livers were screened by artificial digestion method (0.5% pepsin, 1% HCl).

Statistical analysis

The significance of individual factors (household location, village, sex and age) as determinants for parasite infection, was investigated by univariate and multivariate logistic regression model, using individual parasitological status (positive or negative) as a dependent variable. The null hypothesis was that none of the individual factors (indipendant variables) was associated with parasite infection. The critical probability was set at $P = 0.05$. Statistical analysis was performed using computer package STATA 6 (Stata Press, College station, Texas, USA).

RESULTS

Totally 255 rats were captured and examined. Parasite infections were found in 185 (72.6%) rats. Among rats from village A 69.1% were found infected and from village B 88.9% rats harboured parasites.

Ectoparasites (8 species) assigned to four orders (*Siphunculata*, *Mallophaga*, *Siphonaptera* and *Acarina*) were found in 82 rats (32.2%).

The sucking lice *Poliplax spinulosa* had the highest prevalence and was observed in 14.5% of rats (Table 1). Two chewing lice species, *Trimenopon jenningsi* and *Gyropus ovalis* were observed in 4.71% of rats (Table 1). The capture location (household) within village as a risk factor is associated with infection with *T. jenningsi* (OR = 1.406, 95% CI 1.041–1.900, $P = 0.026$).

The flea species *Leptopsylla segnis* and *Ceratophylus fasciatus* were found in 13.3% of rats. The infection with *C. fasciatus* significantly increased with age

Table 1. The prevalence of ectoparasites detected on rats of different gender, location and age groups

Category	Group	*P. spinulosa* (%)	*T. jenningsi* (%)	*G. ovalis* (%)	*L. segnis* (%)	*C. fasciatus* (%)	*M. musculi* (%)	*M. musculinus* (%)	*D. sanguineus* (%)
Gender	male	11.5	5.21	2.08	1.04	10.4	4.17	2.08	0
	female	16.4	1.89	1.26	1.26	13.2	6.29	0.63	1.26
Village	A	16.2	3.81	1.9	0.95	8.1	2.86	0	0
	B	6.67	0	0	2.22	31.1	17.8	6.67	4.44
Age	juveniles	5.88	0	0	0	1.96	1.96	1.96	0
	subadults	15.7	2.41	1.2	0	14.5	6.29	1.2	0
	adults	17.4	4.96	2.48	2.48	14.9	4.17	0.83	1.65
Total		14.5	3.14	1.57	1.18	12.1	5.49	1.18	0.78

(OR = 1.866, 95% CI 1.082–3.216, *P* = 0.025), and was found in significantly higher number in village B (31.1%) than in village A (8.10%) (OR = 5.127, 95% CI 2.298–11.440, *P* < 0.001) (Table 1). Three mite species, *Myobia musculi*, *Myocoptes musculinus* and *Dermanyssus sanguineus* were recoverd from 7.45% of rats with *M. musculi* being the most prevalent mite (5.49%) (Table 1). Although thoughfully checked infected rats did not show attributable skin lesions. Geographic position (village) as a risk factor was associated with *M. musculi* infection which was higher in village B (17.8%) than village A (2.86%) (OR = 7.351, 95% CI 2.410–22.416, *P* < 0.001) (Table 1).The rats with mites tended to be infected with fleas as well (OR = 7.59, 95% CI 2.818–20.470, *P* < 0.001). Statistical analysis showed that presence of observed ectoparasites was not related with sex.

Parasites (worms and eggs) from seven nematode genera (*Capillaria*, *Syphacia*, *Nippostrongylus*, *Heterakis*, *Strongyloides*, *Ascaris*, *Toxocara*) and two tapeworm genera (*Hymenolepis*, *Taenia*) were recovered from 166 (65.1%) rats.

The cestode *Hymenolepis diminuta* (36.9%) and nematodes *Heterakis spumosa* (25.9%) and *Capillaria* sp. (18%) were the most prevalent helminths (Table 2).

Infections of intestinal nematodes were significantly correlated with age (OR = 2.278, 95% CI 1.613–3.218, *P* < 0.001), especially for *H. spumosa* (OR = 2.787, 95% CI 1.758–4.418, *P* < 0.001), *Capillaria* sp. (OR = 2.556, 95%

CI 1.507–4.335, *P* < 0.001) *Syphacia muris* (OR = 6.927, 95% CI 1.001–47.937, *P* = 0.050) and *Nippostrongylus brasiliensis* (OR = 3.24, 95% CI 1.781–5.899, *P* < 0.001). The same association was found for cestode *H. diminuta*, whose prevalence of infection also increased with age (OR = 1.96, 95% CI 1.365–2.817, *P* < 0.001) as well as for *T. taeniaeformis* larvae (OR = 4.621, 95% CI 1.898–11.247, *P* < 0.001). The difference in intensity of parasite infections between two selected villages was observed for *Capillaria* sp. (OR = 2.512, 95% CI 1.205–5.238, *P* = 0.014) *S. muris* (OR = 19.7, 95% CI 3.833–95.753, *P* < 0.001) and *N. brasiliensis* (OR = 2.818, 95% CI 1.341–5.925, *P* = 0.006) and all were higher in village B (Table 2). Also, according to our studies rats from village A had relatively few parasites per household. The significant difference between households was observed in prevalence of *T. taeniaeformis* larvae (OR = 0.921, 95% CI 0.860–0.987, *P* = 0.002), *H. spumosa* (OR = 0.932, 95% CI 0.889–0.977, *P* = 0.003), *Capillaria* sp. (OR = 0.914, 95% CI 0.866–0.967, *P* = 0.002) and *N. brasiliensis* (OR = 0.920, 95% CI 0.870–0.974, *P* = 0.004). The rats with observed nematode infection tended to be infected with cestodes as well (OR = 1.135, 95% CI 1.072–1.202, *P* < 0.001). More than half (56.9%) of rats were infected with nematodes and cestodes at the same time.

Accidental parasite eggs of *A. suum* were found in 21 (8.24%) rats and *Toxocara* sp. in 5 (1.96%) rats. Statistical analysis showed that presence of these eggs was not related to any of risk factors.

Table 2. The prevalence of endoparasites detected on rats of different gender, location and age groups

Category	Group	*H. diminuta* (%)	*T. taeniaeformis* larvae (%)	*Capillaria* sp. (%)	*C. hepatica* (%)	*S. muris* (%)	*N. brasiliensis* (%)	*H. spumosa* (%)	*S. ratti* (%)	*A. suum* eggs (%)	*Toxocara* sp. eggs (%)
Gender	male	35.4	13.5	18.8	0	1.04	14.6	24.0	2.08	9.38	0
	female	37.7	8.81	17.6	0.63	5.03	18.2	27.0	0.63	7.55	3.14
Village	A	37.1	11.4	15.2	0	0.95	13.8	23.8	0	9.52	1.9
	B	35.6	6.67	31.1	2.22	15.6	31.1	35.6	6.67	2.22	2.22
Age	juveniles	13.7	0	1.96	0	0	1.96	1.96	0	11.8	0
	subadults	38.6	6.02	16.9	0	1.2	12.0	25.3	0	1.2	1.2
	adults	45.5	18.2	25.6	0.83	6.61	26.5	36.4	2.48	11.6	3.31
Total		36.9	10.6	18	0.39	3.53	16.9	25.9	1.18	8.24	1.96

DISCUSSION

In our study, we have found out that the parasite community of brown rats in Croatia is characterized by the small number of species of ecto and endoparasites, a total of only 15 species of parasites.

The finding of *P. spinulosa* with the highest prevalence among ectoparasites is in the correlation with findings of Soliman *et al.* (2001) who found *P. spinulosa* as a dominant louse species on brown rats. There was no significant association between lice infection and sex, age and geographical position (village). Our results are not in agreement with findings of Volf (1991) who has established that the degree of *P. spinulosa* infection depends on sex and age. The high prevalence of infection with *C. fasciatus* and *M. musculi* (especially in village B) can be attributed to very low biosanitary conditions in village B and is in correlation with Linardi *et al.* (1985) who found that area of rats capture is related to the degree of infection with fleas. The low prevalence of *C. fasciatus* in juveniles is probably biased due to low number of juveniles captured in village B. Our finding that mite infections were not related with sex can not be explained at the moment, and is not correlated with findings of Soliman *et al.* (2001) who claimed that infection with most ectoparasites were significantly higher on male rats, while Webster and Macdonald (1995) found significantly more female rats infected with mites and male rats infected with lice. From what we know, the relation of parasite infections and sex in rats has not been investigated, yet.

The high prevalence of *H. diminuta*, *H. spumosa* and *Capillaria* sp. in our results is in the correlation to the findings of Yen *et al.* (1996) and of Acinboade *et al.* (1981) who found that the incidence of *H. diminuta* was high among rats trapped in the villages. Studies in Germany (Nickel and Buchwald, 1979) have shown that *H. spumosa* was present in 58.3%, *H. diminuta* in 44.1% and *Taenia taeniaeformis* larvae in 3.3% of rats. Infections with intestinal helminths were significantly correlated with age what is in agreement with Webster and Macdonald (1995) (with the exception of *N. brasiliensis*) who also found an age-prevalence increase and Mafiana *et al.* (1997) who found that rats among the weight class over 100 g were more infected with helminths than those with the weight less than 100 g. The difference in intensity of parasite infections between two selected villages can be attributed to different levels of sanitation in those villages and the significant difference in prevalences between households indicate clustering of parasites among rats from the same household.

The findings of accidental parasite eggs, not related to any of risk factors suggest that all categories of rats are coprophagous. The similar prevalence of parasites in rats as incidental carriers was found by Sahin (1979) for *A. lumbricoides* eggs (2.8%) and Webster and Macdonald (1995) for *T. cati* eggs (15%).

It is interesting, having in mind low hygienic conditions that *C. hepatica* in the liver was found just in one animal. This is completely different from results of many authors world wide. Conlogue *et al.* (1979) found *C. hepatica* in 82% of 86 rats in Connecticut and Davoust *et al.* (1997) found *C. hepatica* in 44% of rats.

Interpretation of the significant difference in the prevalence of *C. hepatica* can be the result of differences in infection among urban and rural rat populations (Sinniah *et al.*, 1979). The authors reported significantly different prevalence among urban (synanthropic rats) and jungle rats (low prevalence) in comparison to field rats trapped from agricultural areas (high prevalence).

We can conclude, that our findings, regarding *C. hepatica* have no considerable public health implications at the moment, but the presence of a large population of infected synanthropic rats requires further deratisation campaigns and rats monitoring.

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