Introduced Siberian chipmunks are more heavily infested by ixodid ticks than are native bank voles in a suburban forest in France

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ABSTRACT

By serving as hosts for native vectors, introduced species can surpass native hosts in their role as major reservoirs of local pathogens. During a 4-year longitudinal study, we investigated factors that affected infestation by ixodid ticks on both introduced Siberian chipmunks *Tamias sibiricus barberi* and native bank voles *Myodes glareolus* in a suburban forest (Forêt de Sénart, Ile-de-France). Ticks were counted on adult bank voles and on adult and young chipmunks using regular monthly trapping sessions, and questing ticks were quantified by dragging. At the summer peak of questing *Ixodes ricinus* availability, the average tick load was 27–69 times greater on adult chipmunks than on adult voles, while average biomass per hectare of chipmunks and voles were similar. In adult chipmunks, individual effects significantly explained 31% and 24% of the total variance of tick larvae and nymph burdens, respectively. Male adult chipmunks harboured significantly more larvae and nymphs than adult females, and than juveniles born in spring and in summer. The higher tick loads, and more specifically the ratio of nymphs over larvae, observed in chipmunks may be caused by a higher predisposition – both in terms of susceptibility and exposure – to questing ticks. Tick burdens were also related to habitat and seasonal variation in age- and sex-related space use by both rodents. Introduced chipmunks may thus have an important role in the dynamics of local vector-borne pathogens compared with native reservoir hosts such as bank voles.

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1. Introduction

Many of the recorded cases of emerging infectious diseases in wildlife result from the introduction of allochtonous host species that become invasive (Daszak et al., 2000; Dobson and Foufoupoulos, 2001; Tompkins and Poulin, 2006). Introduced species modify the composition of a host community and create new relationships between hosts and parasites (Krakau et al., 2006; Thieltges et al., 2009). They may acquire native parasites, especially from phylogenetically related native host species or when the parasite has a large spectrum of compatible host species (Barton, 1997; Azakawa, 2005; Tompkins and Poulin, 2006; Pisanu et al., 2009). Introduced species can then surpass the role of native hosts as major reservoirs (Craine et al., 1995; Daszak et al., 2000; Lv et al., 2009). Theoretical works have shown that the composition of a host community influences the dynamics of diseases, especially those transmitted by generalist vectors (Dobson, 2004; Begon, 2008; Logiudice et al., 2008). Vector dynamics depend on its survival and on host infestation, which, in turns, depend on differences in resistance levels of host species, vector feeding preference and host–vector con-

tact (Dizij and Kurtenbach, 1995; Godsey et al., 1987; Shaw et al., 2003). In the field, very few studies have investigated the consequences of the introduction of a new host species on vector dynamics, whereas several studies have investigated different established host communities (LoGiudice et al., 2003; Krasnov et al., 2007; Swaddle and Calos, 2008; Welc-Falęciak et al., 2008). In addition, factors affecting the acquisition of parasites by introduced vertebrates have received little attention to date (Prenter et al., 2004; Tompkins and Poulin, 2006; Kelly et al., 2009).

Ixodid ticks are true generalists, which mean that they can exploit a large spectrum of different host species and are vectors for many microparasites (Pérez-Eid, 2007). Infestation by ixodids depends on factors such as the micro-geographical variability in the availability and activity of the developmental stages (Hubálek et al., 2003), tick population dynamics (Randolph et al., 2002), or on temporal availability of alternative hosts (Shaw et al., 2003; Ostfeld et al., 2006). Hard tick infestation also depends on the host's endocrine system (Hughes and Randolph, 2001) and related immunogenetic (Charbonnel et al., 2006) and behavioural status (Barnard and Behnke, 2006; Boyer et al., 2010). More specifically, individual variation in parasite loads can be attributed to sex, males being more heavily infected than females because of their distinct behaviours, altered immune functions or their larger body

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size (Poulin, 1996; Moore and Wilson, 2002; Wilson et al., 2002; Harrison et al., 2010).

In this paper, we examine the determinants of infestation by ixodid species on the Korean form of the Siberian chipmunk, *Tamias* (= *Eutamias*) *sibiricus barberi* Johnson and Jones, 1955 (see Obolenskaya et al., 2009), introduced into France (Chapuis, 2005). Imported from Korea to be sold in pet shops since the 1960s, Siberian chipmunks have been released into the wild since the 1970s in at least 11 woody areas in France (Chapuis, 2005; Marmet and Chapuis, 2007). In the population of the Forêt de Sénart, chipmunks were found to be much more heavily infested by larvae and nymphs of *Ixodes ricinus* than were native muroid rodents living in sympatry, and suspected to be an important reservoir for Lyme borreliosis (Vourc'h et al., 2007). Moreover, we found that variation in ixodid burdens on individual chipmunks was related to both differences in habitat characteristics and personality (Boyer et al., 2010). Here we report how age and sex affect variation in tick burden based on monthly monitoring of the Siberian chipmunk population in the Forêt de Sénart over 4 years by capture-mark-recapture in two distinct habitats (Marmet, 2008; Marmet et al., 2009), together with an estimation of the dynamics of ticks that are questing for hosts on the vegetation. These results are compared with monthly data collected over 2 years on one of the most abundant muroid species, the bank vole, *Myodes* (= *Clethrionomys*) *glareolus*, that cohabits with chipmunks on our study site (Vourc'h et al., 2007; Pisanu et al., 2008, 2009). We especially focussed on the difference in nymph and larvae burdens between these two rodents according to adult host biomass per hectare at the period of both maximum availability of questing ticks and the presence of feeding stages. Additionally, using mixed models, a special effort was made to quantify variability in tick load on individual chipmunks (Patterson and Lello, 2003).

2. Materials and methods

2.1. Study site and populations

The study site was the "Parc de la Faisanderie" (14 ha; 02°29′36″E, 48°39′24′N), located in the Forêt de Sénart, a 3200 ha suburban forest 22 km southeast from Paris, France. This area is occupied by an oak-hornbeam forest separated into two kinds of habitat differing in their arboreal and herbaceous cover: (i) an open area in the northern part (6 ha), and (ii) a closed one in the southern part (6.5 ha); the remaining 1.5 ha part of the study site consisted of a lawn and stonewall buildings (Marmet, 2008; Marmet et al., 2009). As part of a broader study on the ecology of introduced Siberian chipmunks (Marmet, 2008), the tick study was based on the capture of individually marked chipmunks using 104 Sherman© traps baited with peanut butter, and sited using a Global Positioning System Leitz® (accuracy: ±1 m) distributed throughout the study site (distance between two neighboring traps = 38 ± 9 m, n = 169; Marmet et al., 2009). Trapping was carried out from March to October, between 2005 and 2008. Between 2005 and 2007 we performed two monthly trapping sessions of three and five consecutive days, respectively. A single, 5-day long trapping session was undertaken every month in 2008. During a trapping session, chipmunks were weighed, sexed, examined and immediately released. Each newly trapped chipmunk was marked using a s.c. transponder chip. Chipmunks were classified in three age groups depending of their body mass and the date of their first capture: (i) adults born the previous year or before; (ii) spring cohort juveniles (J1), emerging from their natal burrow between April and June; and (iii) summer cohort juveniles (J2), emerging between August and September. During this study, 47 individuals out of the 734 different chipmunks trapped could not be precisely aged, and were excluded from analyses.

Bank voles were regularly caught on our trapping grid. A sample up to 30 voles were euthanized monthly by cervical dislocation in 2007 and 2008, immediately put into a plastic bag, and frozen for later analysis in the laboratory. Animals were weighed to the nearest gram and sexed. Overall, 346 adults weighing at least 18 g, were selected for analyses. All trapping and handling procedures were conducted in accordance with the French legislation.

2.2. Densities of hosts

Adult chipmunk densities were calculated in June 2007 and 2008 by using the DENSITY 4.4 software which offers a direct estimation of density by fitting a simple spatial model to animal trapping histories by using simulation and inverse prediction (Efford, 2004; Efford et al., 2004: http://www.otago.ac.nz/density/). For bank voles, population size was estimated using the 'Removal' algorithm in CAPTURE software (Otis et al., 1978; http://www.mbr-pwrc.usgs.gov/software.html) from marked-recaptured individuals caught during three consecutive days in 100 baited INRA© live-traps equally spaced among 200 m long lines and deployed in the habitats (Vourc'h et al., 2007; Pisanu et al., 2008). These abundances were then divided by an effective trapping area (Parmenter et al., 2003) deduced by adding a boundary strip of 25 m wide on each side of the line (Pisanu et al., 2008), which is close to the estimation of the half mean maximal distances moved by these small rodents (Brown, 1956; Kikkawa, 1964; Randolph, 1973).

2.3. Feeding ticks

During examination of chipmunks, we counted tick larvae, nymphs and adults only on the head (Craine et al., 1995; Schmidt et al., 1999), using eye lenses ($3\times$ magnification). A sub-sample of adult ticks was removed and identified using the morphological key of Pérez-Eid (2007). Overall, 45 adult ticks were observed from 2870 trapping events on 734 chipmunks between 2005 and 2008. Twenty-six of them were removed and identified to species. All were identified as *Ixodes acuminatus*. Because there were so few adult ticks, this stage was not considered in this study. We focused on earlier life-stages of ixodids of which >95% have been identified as *I. ricinus* in a previous study in the Forêt de Sénart (Vourc'h et al., 2007).

Ticks on bank voles were collected from the entire body for counting and identification to species level under an optical microscope (all done by M. Marsot). Both larvae and nymph of *I. ricinus* represented 93.1% of identified specimens (n = 786), the remaining belonging to *Dermacentor reticulatus* (2.9%), *Ixodes trianguliceps* (2.0%) and *I. acuminatus* (1.9%). *Ixodes ricinus* is the major vector for Lyme borreliosis in Europe. The agent has been isolated in the two other species of *Ixodes* but their role in transmission has not been demonstrated. The role of *D. reticulatus* is poorly known (Pérez-Eid, 2007). No adult ticks were recovered from bank voles. Because we included all ixodid species for chipmunks, we did the same for bank voles.

2.4. Questing I. ricinus

Questing stages of ticks were sampled monthly by dragging a 1 m² woollen blanket over the vegetation on two 0.5 ha sites of the study area, one in the open and another in the closed forest habitat. Within each site, 15 U of 10 m² were randomly dragged in 2005 and 2006, and 16 U in 2007 and 2008. Drags were conducted early in the afternoon, avoiding rainy days, and nymphs and larvae were counted exhaustively. A total of 1082 questing nymphs and 36 adults were collected, of which all were further identified under the microscope as *I. ricinus* except one adult of *D. reticulatus* (E. Ferquel, personal observation).

2.5. Data analyses

For chipmunks, due to the lack of J1s until April and of J2s until August each year during the study period, and the absence of J1s until July in 2006 (Marmet et al., 2009), we investigated the variation in larval and nymphal counts according to year, habitat, age and sex only in 2005, 2007 and 2008 without including a month effect. As adults were present throughout the study period, we also tested for the effect of habitat, year, month and sex on variation in their tick counts. We used mixed models (Pinheiro and Bates, 2000; Patterson and Lello, 2003; Bolker et al., 2009; Zuur et al., 2009) both to estimate individual consistency in tick load, also including autocorrelation structure due the temporal nature of data collection, and to test for the effects of year, habitat, month, age and sex (see Supplementary data S1).

Using Generalized Linear Models with negative binomial errors and a log link (Wilson and Grenfell, 1997; Venables and Ripley, 2002; Crawley, 2007), we investigated the effects of habitat, year, month and sex on tick counts on adult voles. For questing nymphs, we used data collected from April to October of each summer period as only a few nymphs were collected in March. We investigated the effects of habitat, year and month. Models were backward selected using likelihood ratio tests (noted LRT). Because we could not reach satisfactory fits with the model on questing larvae, only descriptive patterns of variation in their abundance are presented.

In all the models described above, sources of variation were investigated up to two-way interactions between factors. We used non-transformed mean numbers and their standard errors in all the figures and in the table. All the statistical analyses were done using R 2.8.1 (R Development Core Team, 2008, http://www.R-project.org).

3. Results

3.1. Feeding larvae

We did not detect any significant temporal autocorrelation in our model of larval loads on adult chipmunks (Supplementary Table S1). Chipmunk identity significantly explained 31.1% of the variation in repeated larval counts. We found a significant interaction between year and month. Each year, infestation by larvae on adult chipmunks increased from March until May, June or July, depending on the year of the study. Larvae then decreased until October (Fig. 1). A significant effect of year and habitat was detected (Supplementary Table S1), with chipmunks inhabiting the closed habitat being more heavily infested by larvae than those in the open habitat in 2006 and 2008, but not in 2005 and 2007 (Table 1). Finally, a significant interaction between year and sex was observed (Supplementary Table S1): between 2005 and 2007, males had higher loads of larvae than females, except in 2008 where both sexes had a low degree in larval infestation (Table 1). The three other two-way interactions were not significant (Supplementary Table S1).

Including juveniles, and controlling for significant temporal and identity effects (Supplementary Table S1), a significant interaction between age and sex was found (Supplementary Table S1). The maximum in mean larval counts for J1s were on males (Table 1), and larval infestation on J2s remained identical over the study period (Fig. 1; Table 1). Overall, adult males harboured significantly more larvae than adult females and than J1s and J2s (Table 1). We also found significant effects of year and habitat (Supplementary Table S1), both following the same pattern as for adult variation in larval infestation (Table 1).

On bank voles, the best minimal model that could be retained was a three-way interaction between year, month and sex (df = 7, LRT = 17.73, P = 0.01). Males had significantly higher loads of larvae than females (Table 1), with highest loads between July and September in 2007 (Fig. 2). Overall, both females and males had lower larval burdens in 2008 than in 2007.

3.2. Feeding nymphs

We did not find any temporal autocorrelation in the model of nymph infestation, and chipmunk identity significantly explained 23.9% of the variation in nymph counts with a strong fit to the data (Supplementary Table S2). Controlling for the identity effect, we found a significant interaction between year and month (Supplementary Table S2). Between 2005 and 2008, yearly counts of nymphs on adults began to increase in March, reaching a peak in May or in June, then decreasing in July or August, and remaining stable until October (Fig. 1). No other two-way interaction was found to be significant (Supplementary Table S2). Overall, adult males had heavier nymph burdens than females (Table 2).

Controlling for a significant identity effect, but not any temporal autocorrelation, the model including juveniles gave a satisfactory fit to the data (Supplementary Table S2). There was no effect of habitat, and no interaction between factors, but a single effect of age, sex or year (Supplementary Table S2). Separating by age and sex, adult females hosted significantly fewer nymphs than adult males, and adults of both sexes than both sexes of J1s and J2s (Table 2). Additionally, adult males had heavier loads of nymphs in 2005.

On bank voles, we found a significant effect of habitat (df = 1, LRT = 5.69, P = 0.02) and of sex (df = 1, LRT = 4.99, P = 0.03), but not of month or year taken as single effect, and all these factors tested in interaction. Voles in the closed habitat had a significantly higher mean nymph load (0.28 ± 0.05, n = 152) than in the open habitat (0.16 ± 0.03, n = 194). Males had a significantly higher nymph load (0.27 ± 0.04, n = 175) than females (0.15 ± 0.04, n = 171).

3.3. Questing ticks

For questing nymphs, we found significant effects for habitat and year, habitat and month, and year and month (Table 3). Overall, questing nymph abundance varied seasonally (Fig. 1), increasing from March until June or July depending on the year, and then decreasing until October. Nymphs were much less abundant on the open habitat than on the closed habitat, except for May and June 2008 (Fig. 1). Mean counts (Fig. 1) followed a regular seasonal pattern, reaching a peak between May and September, except in 2006 when very a high abundance of larvae occurred in the closed habitat in May.

Questing larvae were more irregularly distributed over time in the open habitat, with very low abundance in 2005 and 2006, and monthly variation in 2007 and 2008.

3.4. Biomass of hosts per hectare

At the peak of questing tick availability in June, the average biomass of chipmunks was 420 g/ha in 2007 and 309 g/ha in 2008 (based on the product of mean body mass and density: Table 4), whereas it was 363 g/ha in 2007 and 264 g/ha in 2008 for bank voles.

4. Discussion

Siberian chipmunks introduced in the Forêt of Sénart hosted approximately 27–69 times more ixodid ticks than native bank voles at the peak of questing stage availability in June (see Table 1), despite representing similar biomasses. Even if our biomass density for voles is biased due to uncertainty about the estimation of an
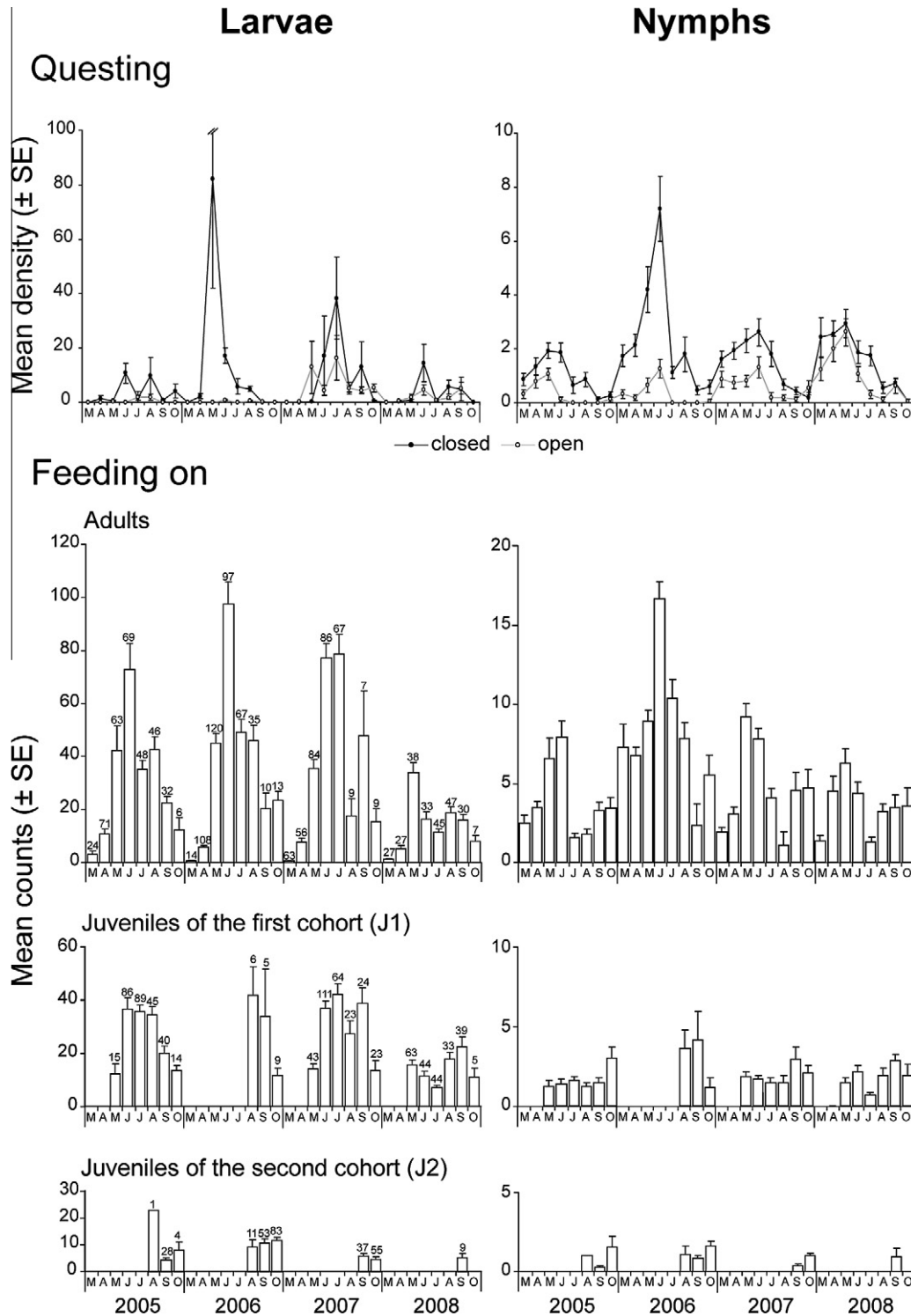
Fig. 1. Monthly variation in mean density of questing stages of *Ixodes ricinus* per 10 m² according to open and closed habitats, and mean counts of feeding stages of ixodids according to the ages of chipmunks between 2005 and 2008 on the Forêt de Sénart (France). Numbers of examined hosts are indicated above bars for larvae, the same applying for nymphs. An absence of sample size indicates no chipmunks were trapped. SE, standard error.

effective trapping area around the trapping lines (Parmenter et al., 2003; Efford, 2004), we are confident that the difference in tick infestation rates between the two species is strong enough to be reliable.

An adult chipmunk is four to five times larger than an adult vole (Table 4). Such a difference in body size allows these ground squirrels to contact questing ticks efficiently at more variable heights above ground level than smaller rodents such as voles. Indeed, lar-

vae quest lower than nymphs (Mejlon and Jaenson, 1997). This hypothesis is supported by the observation that chipmunks appear to feed larvae and nymphs in an approximately c.10:1 ratio, which is close to the one observed for both stages of questing ticks (see Fig. 1). This is in marked contrast to small rodents, which are generally so small that they run about beneath available questing ticks. Indeed, in voles trapped and analyzed between May and July

Table 1
Mean number of ixodid larvae, according to year, age, sex and habitat, on Siberian chipmunks in the Forêt de Sénart (France) between 2005 and 2008, and on adult bank voles in 2007 and 2008.

Year	Age	Sex	Closed habitat					Open habitat				
			Chipmunks			Voles		Chipmunks			Voles	
			N	n	Mean ± SE	N	Mean ± SE	N	n	Mean ± SE	N	Mean ± SE
2005	Ad.	♀	36	159	30 ± 4	–	–	23	108	30 ± 3	–	–
		♂	13	51	43 ± 7			8	41	67 ± 17		
	J1	♀	28	78	32 ± 4	–	–	13	60	31 ± 4	–	–
		♂	29	59	32 ± 4			27	91	26 ± 2		
	J2	♀	14	16	6 ± 2	–	–	3	4	3 ± 1	–	–
		♂	8	10	4 ± 1			3	3	5 ± 4		
2006	Ad.	♀	47	196	44 ± 3	–	–	24	93	33 ± 3	–	–
		♂	30	115	55 ± 7			19	60	47 ± 8		
	J1	♀	4	11	17 ± 6	–	–	1	2	5; 3	–	–
		♂	3	5	47 ± 13			1	2	25; 14		
	J2	♀	18	34	11 ± 2	–	–	22	31	9 ± 1	–	–
		♂	25	41	9 ± 1			30	41	11 ± 2		
2007	Ad.	♀	38	149	35 ± 3	41	2 ± 1	20	74	27 ± 4	56	1 ± 0.2
		♂	31	107	60 ± 6	45	6 ± 1	18	51	47 ± 7	57	5 ± 1
	J1	♀	30	88	28 ± 2	–	–	22	59	25 ± 3	–	–
		♂	37	80	33 ± 4			29	61	36 ± 4		
	J2	♀	23	33	5 ± 1	–	–	4	7	7 ± 4	–	–
		♂	27	33	4 ± 1			18	20	3 ± 1		
2008	Ad.	♀	42	99	19 ± 2	35	1 ± 0.2	18	47	9 ± 2	39	1 ± 0.2
		♂	32	75	17 ± 2	31	1 ± 0.2	22	33	12 ± 3	42	1 ± 0.3
	J1	♀	39	85	14 ± 1	–	–	25	48	9 ± 1	–	–
		♂	30	52	16 ± 2			29	45	10 ± 2		
	J2	♀	2	2	0; 5	–	–	1	1	5	–	–
		♂	5	5	5 ± 3			1	1	3		

Ad., adult; J1, J2, juveniles; N, number of chipmunks individuals examined; n, number of examinations of these chipmunks. Voles were sampled and examined once; SE, standard error.
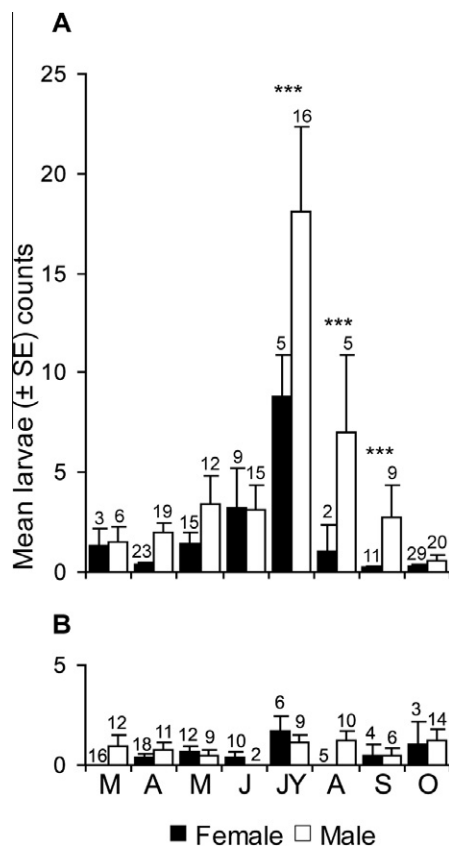
Fig. 2. Variation in monthly mean ixodid tick counts according to sex in adult bank voles at Forêt de Sénart (France) in (A) 2007 and (B) 2008. ***: $P < 0.001$; SE, standard error. Months March to October are indicated by initials.

2005 (Vourc'h et al., 2007), this ratio was approximately c.83-fold, ~46-fold in May–July 2007, and ~53-fold in May–July 2008, indicating that voles picked up far fewer nymphs than larvae. The infestation ratio between larvae and nymphs is of prime importance for the maintenance of pathogenic agents in reservoir hosts (Randolph and Storey, 1999). For the dynamics to be maintained in an ecosystem, reservoirs must host both stages because they become infected by nymphs, and transmit the infection to larvae, that subsequently become infected questing nymphs. Because chipmunks have such a high ratio of larvae to nymphs, and because Vourc'h et al. (2007) have shown that chipmunks are infected by Lyme disease, we can expect, if those are confirmed as reservoir hosts, that those will produce more infected ticks than the bank voles and thus may be major reservoir hosts where they have been introduced with vectors.

Some unmeasured characteristics of rodent hosts are responsible for individual variation in infestation by *Ixodes* spp. (Brunner and Ostfeld, 2008). Such predisposition to infestation (Hudson and Dobson, 1995) significantly explained why some Siberian chipmunks were consistently more heavily infested by tick larvae and nymphs than others, mostly *I. ricinus*, and confirms results of an earlier behavioural study based on a sub-sample of chipmunks at the same study site (Boyer et al., 2010). This result is also in accordance with patterns of infestation by *I. ricinus* on yellow-necked wood mice *Apodemus flavicollis* (Perkins et al., 2003). Adult Siberian chipmunks at the Forêt de Sénart show strong multi-year site fidelity in home range (Marmet et al., 2009). Therefore, any consistency in exposure to questing stages of ticks (Wilson et al., 1990, 2002) could partly explain individual host consistency in tick loads.

Tick burdens in adult rodents, both native and introduced, were related to sex, and to age in chipmunks. These results are in accordance with highest frequency of occurrence in *I. ricinus* found on larger male rodents (Randolph, 1975; Perkins et al., 2003; Harrison et al., 2010). Siberian chipmunks do not show sexually dimorphic

Table 2
Mean number of ixodid nymphs on male and female Siberian chipmunks of different ages and during different years, in the Forêt de Sénart (France).

Age	Sex	2005		2007		2008	
		n	Mean ± SE	n	Mean ± SE	n	Mean ± SE
Adults	♀	267	3.2 ± 0.3	223	1.7 ± 0.4	146	3.1 ± 0.3
	♂	92	5.2 ± 0.7	158	2.0 ± 0.4	108	3.7 ± 0.4
J1	♀	138	1.6 ± 0.2	147	1.7 ± 0.2	133	1.7 ± 0.2
	♂	150	1.6 ± 0.2	141	2.0 ± 0.2	97	1.2 ± 0.2
J2	♀	20	0.7 ± 0.2	40	0.9 ± 0.2	3	1.7 ± 2.0
	♂	13	0.4 ± 0.2	53	0.7 ± 0.1	6	0.5 ± 0.4

J1, J2, juveniles; n, number of examinations of chipmunks; SE, standard error.

Table 3
Backward selection of generalized linear models using likelihood ratio tests (LRTs) exploring the variation in questing *Ixodes ricinus* nymphs.

Sources of variation	df	LRT	P
Habitat:Year	8	25.60	<0.001
Habitat:Month	14	39.32	<0.001
Year:Month	14	33.08	<0.001

Table 4
Mean body mass (±standard error, in g) and density ([95% confidence interval], in number/ha) of adult of chipmunks *Tamias sibiricus* and bank voles *Myodes glareolus* in June 2007 and 2008 in Sénart (France).

Year	Chipmunks			Bank voles		
	n	Mass	Density	n	Mass	Density
2007	61	100 ± 1	4.2 [1–8]	24	25 ± 1	14.5 [13–21]
2008	37	103 ± 1	3.0 [1–4]	12	22 ± 1	12.0 [9–21]

body size (Obolenskaya et al., 2009). Thus, any sex differences in tick load may be caused by differences in genetics (Coltman et al., 2001), physiology (Perez-Orella and Schulte-Hostedde, 2005), immune status (Hughes and Randolph, 2001) or in activity, exploration and space use (Boyer et al., 2010). Moreover, the lower infestation by both tick stages on juveniles compared with adults can be mainly related to the limited space used by juveniles (Marmet et al., 2009, 2010), as already described for small rodents (Brown, 1956; Kikkawa, 1964; Randolph, 1973, 1975).

Yearly fluctuation in infestation by ticks on rodents can be partly attributed to spatial differences in survival or activity of questing stages (Randolph et al., 2002; Randolph, 2004). Habitat was found to influence larval load on chipmunks in the Forêt de Sénart (Boyer et al., 2010), with lower infestations on hosts inhabiting the open habitat than on the closed habitat of the study site. This is in accordance with the highest number of questing stages of *I. ricinus* observed on the closed habitat where soil-litter cover is more humid and deeper (Marmet, 2008), thus more suitable for questing *I. ricinus* (Pérez-Eid, 2007). Moreover, the closed habitat offers preferred resting sites for roe deer, *Capreolus capreolus*, which are important hosts for *I. ricinus* (Wilson et al., 1990; Perkins et al., 2006). Additionally, availability in alternative rodent hosts can modify the temporal abundance of ixodids on chipmunks (Schmidt et al., 1999; Brunner and Ostfeld, 2008). Wood mouse density on our study site increased by 2.3-fold in June between 2007 and 2008 (data not shown). The increased capture of infesting larvae by wood mice could partly explain the decrease in ixodids on both chipmunks and bank voles. Indeed, wood mice host twice as many ticks as bank voles (Vourc'h et al., 2007). This difference was related to lower immune resistance in mice than in voles (Kurtenback et al., 1994; Dizij and Kurtenbach, 1995). More data are needed to investigate the role of wood mice influencing the pattern of infestation by ixodids in our small rodent community.

This study confirms earlier findings that introduced chipmunks harbour many more *I. ricinus* compared with native rodent host species in a boreal European suburban forest (Vourc'h et al., 2007). Our study highlights the well-known roles of spatial, temporal and host individual characteristics – i.e. age and sex – in seasonal patterns of ixodid infestations in rodents (Perkins et al., 2003; Brunner and Ostfeld, 2008). It raises the question of the predisposition, either in susceptibility or exposure (Hudson and Dobson, 1995) for an introduced rodent to native parasites (Tompkins and Poulin, 2006; Kelly et al., 2009) with a short history of co-adaptation – i.e. less than 80 generations over ~40 years – and its potential role in the increased dynamics of the Lyme borreliosis by outweighing the role of its main native reservoir, the bank vole (Vourc'h et al., 2007), in French suburban forests where the vector is present.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.ijpara.2010.03.012.

References

Azakawa, M., 2005. Perspectives of host–parasite relationships between rodents and nematodes in Japan. Mammal Study 30, S95–S99.
Barnard, C.J., Behnke, J.M., 2006. Behavior, life history strategies and parasite infection in rodents. In: Morand, S., Krasnov, B., Poulin, R. (Eds.), Micromammals and Macroparasites: From Evolutionary Ecology to Management. Springer-Verlag, Tokyo, Japan, pp. 475–511.
Barton, D., 1997. Introduced animals and their parasites: the cane Toad, *Bufo marinus*, in Australia. Aust. J. Ecol. 22, 316–324.
Begon, M., 2008. Effect of host diversity on disease dynamics. In: Ostfeld, R.S., Keesing, F., Eviner, V.T. (Eds.), Effects of Ecosystems on Disease and of Disease on Ecosystems. Princeton University Press, Princeton, pp. 12–29.
Bolker, B.M., Brooks, M.E., Clark, C.J., Geange, S.W., Poulsen, J.R., Stevens, M.H.H., White, J.-S.S., 2009. Generalized linear mixed models: a practical guide for ecology and evolution. Trends Ecol. Evol. 24, 127–135.
Boyer, N., Réale, D., Marmet, J., Pisanu, B., Chapuis, J.-L., 2010. Personality, space use, and tick load in a population of Siberian chipmunk, *Tamias sibiricus*. J. Anim. Ecol. 79, 538–547.
Brown, L.E., 1956. Movements of some British small mammals. J. Anim. Ecol. 25, 54–71.
Brunner, J.L., Ostfeld, R.S., 2008. Multiple causes of variable tick burdens on small-mammal hosts. Ecology 89, 2259–2272.

Chapuis, J.-L., 2005. Répartition en France d'un animal de compagnie naturalisé, le Tamia de Sibérie (*Tamias sibiricus*). Rev. Ecol. (Terre Vie) 60, 239–253.

Charbonnel, N., Goüy de Bellocq, J., Morand, S., 2006. Immunogenetics of micromammal–macroparasites interactions. In: Morand, S., Krasnov, B., Poulin, R. (Eds.), Micromammals and Macroparasites: From Evolutionary Ecology to Management. Springer-Verlag, Tokyo, pp. 401–442.

Coltman, D.W., Pilkington, J., Bruuk, L.E.B., Wilson, K., Pemberton, J.M., 2001. Positive genetic correlation between parasite resistance and body size in a free-living ungulate population. Evolution 55, 2116–2125.

Craine, N.G., Randolph, S.E., Nuttall, P.A., 1995. Seasonal variation in the role of grey squirrels as hosts of *Ixodes ricinus*, the tick vector of the Lyme disease spirochaete, in a British woodland. Folia Parasitol. 42, 73–80.

Crawley, M.J., 2007. The R Book. John Wiley & Sons, Ltd., Chichester.

Daszak, P., Cunningham, A.A., Hyatt, A.D., 2000. Emerging infectious diseases of wildlife – threats to biodiversity and human health. Science 287, 443–449.

Dizij, A., Kurtenbach, K., 1995. *Clethrionomys glareolus*, but not *Apodemus flavicollis*, acquires resistance to *Ixodes ricinus* L., the main European vector of *Borrelia burgdorferi*. Parasitol. Immunol. 17, 177–183.

Dobson, A., 2004. Population dynamics of pathogens with multiple host species. Am. Nat. 164, S64–S78.

Dobson, A., Foufoupoulos, J., 2001. Emerging infectious pathogens of wildlife. Philos. Trans. R. Soc. Lond. B 356, 1001–1012.

Efford, M.G., 2004. Density estimation in live-trapping studies. Oikos 106, 598–610.

Efford, M.G., Dawson, D.K., Robbins, C.S., 2004. Density: software for analysing capture-recapture data from passive detector arrays. Anim. Biodiv. Conserv. 27, 217–228.

Godsey, M., Amundson, T., Burgess, E., Schell, W., Davis, J., Kaslow, R., Edelman, R., 1987. Lyme disease ecology in Wisconsin – distribution and host preferences of *Ixodes dammini*, and prevalence of antibody to *Borrelia burgdorferi* in small mammals. Am. J. Epidemiol. 37, 180–187.

Harrison, A., Scantlebury, M., Montgomery, W.I., 2010. Body mass and sex-biased parasitism in wood mice *Apodemus sylvaticus*. Oikos. doi:10.1111/j.1600-0706.2009.18072.x.

Hubálek, Z., Halouzka, J., Juøicová, Z., 2003. Host-seeking activity of ixodid ticks in relation to weather variables. J. Vector Ecol. 28, 159–165.

Hudson, P.J., Dobson, A.D., 1995. Macroparasites: observed patterns in naturally fluctuating animal populations. In: Grenfell, B.T., Dobson, A.P. (Eds.), Ecology of Infectious Diseases in Natural Populations. Cambridge University Press, pp. 144–176.

Hughes, V.L., Randolph, S.E., 2001. Testosterone depresses innate and acquired resistance to ticks in natural rodent hosts: a force for aggregated distribution of parasites. J. Parasitol. 87, 49–54.

Kelly, D.W., Paterson, R.A., Townsend, C.R., Poulin, R., Tompkins, D.M., 2009. Parasite spillback: a neglected concept in invasion ecology? Ecology 90, 2047–2056.

Kikkawa, J., 1964. Movement, activity and distribution of the small rodents *Clethrionomys glareolus* and *Apodemus sylvaticus* in woodland. J. Anim. Ecol. 33, 259–299.

Krakau, M., Thieltges, D.W., Reise, K., 2006. Native parasites adopt introduced bivalves of the North Sea. Biol. Invasions 8, 919–925.

Krasnov, B., Stanko, M., Morand, S., 2007. Host community structure and infestation by ixodid ticks: repeatability, dilution effect and ecological specialization. Oecologia 154, 185–194.

Kurtenbach, K., Dizij, A., Martin Seitz, H., Margos, G., Moter, S.E., Kramer, M.D., Wallich, R., Schaible, U.E., Simon, M.M., 1994. Differential immune response to *Borrelia burgdorferi* in European wild rodent species influence Spirochete transmission to *Ixodes ricinus* L. (Acari: Ixodidae). Infect. Immun. 62, 5344–5352.

LoGiudice, K., Ostfeld, R.S., Schmidt, K.A., Keesing, F., 2003. The ecology of infectious disease: effects of host diversity and community composition on Lyme disease risk. Proc. Natl. Acad. Sci. USA 100, 567–571.

LoGiudice, K., Duerr, S., Newhouse, M., Schmidt, K., Killilea, M., Ostfeld, R., 2008. Impact of host community composition on Lyme disease risk. Ecology 89, 2841–2849.

Lv, S., Zhang, Y., Liu, H.-X., Hu, L., Yang, K., Steinmann, P., Chen, Z., Wang, L.-Y., Utzinger, J., Zhou, X.-N., 2009. Invasive snails and an emerging infectious disease: results from the first national survey on *Angiostrongylus cantonensis* in China. PLoS Negl. Trop. Dis. 3, e368.

Marmet, J., 2008. Traits d'histoire de vie du Tamia de Sibérie *Tamias sibiricus*, espèce exotique naturalisée dans la Forêt de Sénart (Essonne): démographie, biologie de la reproduction, occupation de l'espace et dispersion. Ph.D. Thesis, Muséum National d'Histoire Naturelle, p. 172.

Marmet, J., Chapuis, J.-L., 2007. Répartition de l'écureuil de Corée (*Tamias sibiricus*), animal de compagnie exotique introduit en France: résultats de l'enquête nationale de 2005. In: Tillon, L. (Ed.), Les Mammifères forestiers, actes du XXVIIIème colloque francophone de Mammalogie de la SFEPM, Rambouillet, France. Collection dossiers forestiers, pp. 27–31.

Marmet, J., Pisanu, B., Chapuis, J.-L., 2009. Home range, range overlap, and site fidelity of introduced Siberian chipmunks in a suburban French forest. Eur. J. Wildl. Res. 55, 497–504.

Marmet, J., Pisanu, B., Chapuis, J.-L., 2010. Natal dispersal of introduced Siberian chipmunks, *Tamias sibiricus*, in a suburban forest. J. Ethol. doi:10.1007/s10164-010-0215-3.

Mejlon, H.A., Jaenson, T.G.T., 1997. Questing behaviour of *Ixodes ricinus* ticks (Acari: Ixodidae). Exp. Appl. Acarol. 21, 747–754.

Moore, S.L., Wilson, K., 2002. Parasites as a viability cost of sexual selection in natural populations of mammals. Science 297, 2015–2018.

Obolenskaya, E.V., Lee, M.Y., Dokuchaev, N.E., Oshida, T., Lee, M.S., Lee, H., Lissovsky, A.A., 2009. The diversity of Palaearctic chipmunks (*Tamias*, Sciuridae). Mammalia 73, 281–298.

Ostfeld, R.S., Canham, C.D., Oggenfuss, K., Winchcombe, R.J., Keesing, F., 2006. Climate, deer, rodents, and acorns as determinants of variation in Lyme-disease risk. PLoS Biol. 4, 1058–1068.

Otis, D.L., Burnham, K.P., White, G.C., Anderson, D.R., 1978. Statistical inference from capture data on closed animal populations. Wildl. Monogr. 62, 1–135.

Parmenter, R.R., Yates, L.Y., Anderson, D.R., Burnham, K.P., Dunnum, J.L., Franklin, A.B., Friggens, M.T., Lubow, B.C., Miller, M., Olson, G.S., Parmenter, C.A., Pollard, J., Rexstad, E., Shenk, T.M., Stanley, T.R., White, G.C., 2003. Small-mammal density estimation: a field comparison of grid-based vs. web-based density estimators. Ecol. Monogr. 73, 1–26.

Patterson, S., Lello, J., 2003. Mixed models: getting the best use of parasitological data. Trends Parasitol. 19, 370–375.

Pérez-Eid, C., 2007. Les tiques: identification, biologie, importance médicale et vétérinaire. Lavoisier, Paris.

Perez-Orella, C., Schulte-Hostedde, A.I., 2005. Effect of sex and body size on ectoparasite loads in the northern flying squirrel (*Glaucomys sabrinus*). Can. J. Zool. 83, 1381–1385.

Perkins, S.E., Cattadori, I.M., Tagliapietra, V., Rizzoli, A.P., Hudson, P.J., 2003. Empirical evidence for key hosts in persistence of a tick-borne disease. Int. J. Parasitol. 33, 909–917.

Perkins, S.E., Cattadori, I.M., Tagliapietra, V., Rizzoli, A.P., Hudson, P.J., 2006. Localized deer absence leads to tick amplification. Ecology 87, 1981–1986.

Pinheiro, J., Bates, D., 2000. Mixed-Effects Models in S and S-PLUS. Springer, New York.

Pisanu, B., Marmet, J., Beaucournu, J.-C., Chapuis, J.-L., 2008. Diversité du cortège en Siphonaptères chez le Tamia de Sibérie (*Tamias sibiricus* Laxmann) introduit en Forêt de Sénart. Parasite 15, 35–43.

Pisanu, B., Lebailleux, L., Chapuis, J.-L., 2009. Why do Siberian chipmunks *Tamias sibiricus* (Sciuridae) introduced in French forests acquired so few intestinal nematode species from native sympatric Murids? Parasitol. Res. 104, 709–714.

Poulin, R., 1996. Sexual inequalities in helminth infections: a cost of being a male? Am. Nat. 147, 287–295.

Prenter, J., MacNeil, C., Dick, J.T.A., Dunn, A.M., 2004. Roles of parasites in animal invasions. Trends Ecol. Evol. 19, 385–390.

Randolph, S.E., 1973. A tracking technique for comparing individual home ranges of small mammals. J. Zool. Lond. 170, 509–520.

Randolph, S.E., 1975. Patterns of distribution of the tick *Ixodes trianguliceps* Birula on its hosts. J. Anim. Ecol. 44, 451–474.

Randolph, S.E., 2004. Tick ecology: processes and patterns behind the epidemiological risk posed by Ixodid ticks as vectors. Parasitology 129, S37–S65.

Randolph, S.E., Storey, K., 1999. Impact of microclimate on immature tick-rodent host interactions (Acari: Ixodidae): implications for parasite transmission. J. Med. Entomol. 36, 741–748.

Randolph, S.E., Green, R.M., Hoodless, A.N., Peacey, M.F., 2002. An empirical quantitative framework for the seasonal population dynamics of the tick *Ixodes ricinus*. Int. J. Parasitol. 32, 979–989.

Schmidt, K.A., Ostfeld, R.S., Schauber, E.M., 1999. Infestation of *Peromyscus leucopus* and *Tamias striatus* by *Ixodes scapularis* (Acari: Ixodidae) in relation to the abundance of hosts and parasites. J. Med. Entomol. 36, 749–757.

Shaw, M.T., Keesing, F., McGrail, R., Ostfeld, R.S., 2003. Factors influencing the distribution of larval blacklegged ticks on rodent hosts. Am. J. Trop. Med. Hyg. 68, 447–452.

Swaddle, J.P., Calos, S.E., 2008. Increased avian diversity is associated with lower incidence of human West Nile infection: observation of the dilution effect. PLoS ONE 3, e2488.

Thieltges, D.W., Reise, K., Prinz, K., Jensen, K.T., 2009. Invaders interfere with native parasite–host interactions. Biol. Invasions 11, 1421–1429.

Tompkins, D.M., Poulin, R., 2006. Parasites and biological invasions. In: Allen, R.B., Lee, W.G. (Eds.), Biological Invasions in New Zealand. Ecological Studies, vol. 186. Springer-Verlag, Berlin, Heidelberg, pp. 67–84.

Venables, W.N., Ripley, B.D., 2002. Modern Applied Statistics with S, fourth ed. Springer, New York.

Vourc'h, G., Marmet, J., Chassagne, M., Bord, S., Chapuis, J.-L., 2007. *Borrelia burgdorferi* sensu lato in Siberian Chipmunks (*Tamias sibiricus*) introduced in suburban forests. Vector Borne Zoonotic Dis. 7, 637–642.

Welc-Falęciak, R., Bajer, A., Behnke, J.M., Siński, E., 2008. Effects of host diversity and the community composition of hard ticks (Ixodidae) on *Babesia microti* infection. Int. J. Med. Microbiol. 298, 235–242.

Wilson, K., Grenfell, B.T., 1997. Generalized linear modelling for parasitologists. Parasitol. Today 13, 33–38.

Wilson, K., Bjørnstad, O.N., Dobson, A.P., Merler, S., Poglayen, G., Randolph, S.E., Read, A.F., Skorping, A., 2002. Heterogeneities in macroparasite infections: patterns and processes. In: Hudson, P.J., Rizzoli, A., Grenfell, B.T., Heesterbeek, H., Dobson, A.P. (Eds.), The Ecology of Wildlife Diseases. Oxford University Press, Oxford, UK, pp. 6–44.

Wilson, M.L., Ducey, A.M., Litwin, T.S., Gavin, T.A., Spielman, A., 1990. Microgeographic distribution of immature *Ixodes dammini* ticks correlated with that of deer. Med. Vet. Entomol. 4, 151–159.

Zuur, A.F., Ieno, E.N., Walker, N.J., Saveliev, A.A., Smith, G.M., 2009. Mixed Effects Models and Extensions in Ecology with R. Springer, New York.