

Supporting information

Vector competence of *Ixodes ricinus* instars for the transmission of *Borrelia burgdorferi* sensu lato on different small mammalian hosts

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Table S1: An overview of samples sizes of captured small mammals with live and lethal traps per season between 2018 and 2022 in Son, Viken county, Norway.

		2018		2019		2020		2021		2022	
	Sum	Spring	Fall	Spring	Fall	Spring	Fall	Spring	Fall	Spring	Fall
% Live traps		100	80	80	80	48	48	48	48	0	0
<i>A. sylvaticus</i>	156	2	9	20	16	27	30	23	11	11	7
<i>M. agrestis</i>	14	1	0	0	3	0	9	0	0	0	1
<i>M. glareolus</i>	258	3	16	4	12	10	143	10	27	10	23
<i>S. araneus</i>	106	2	27	1	21	1	36	1	14	0	3
<i>S. minutus</i>	23	0	3	0	5	3	11	0	1	0	0
Sum	557	8	55	25	57	41	229	34	53	21	34

Table S2: Sequences and adjusted primer set and probe concentrations in respective multiplex real-time quantitative PCR assays for detecting A) *Borrelia burgdorferi* s.l. and *Anaplasma phagocytophilum* and B) *I. ricinus* and *I. trianguliceps*, with respective target genes, product size (bp), sequences, and concentration (μM). Infection prevalence of *A. phagocytophilum* was not considered in the present study.

A) Infection prevalence	Target	bp	Sequence	μM
<u><i>A. phagocytophilum</i></u>	16S rRNA	75		
F primer			ATGGAAGGTAGTGTGGTTATGGTATT	0.900
R primer			TTGGTCTTGAAGCGCTCGTA	0.900
Probe			[HEX]TGGTGCCAGGGTTGAGCTTGAGATTG[TAMRA]	0.125
<u><i>B. burgdorferi</i> s.l.</u>	23S rRNA	77		
F primer			CGAGTCTTAAAAGGGCGATTTAGT	0.700
R primer			GCTTCAGCCTGGCCATAAATAG	0.700
Probe			[6FAM]AGATGTGGTAGACCCGAAGCCGAGTG[TAMRA]	0.175
B) Tick species				
<i>I. ricinus</i>	<i>ITS2</i>	72		
F primer			TTGACTGTGTCGGATCGTGG	0.600
R primer			TCGCTCCGAGAACGTGAAAA	0.600
Probe			[5TexRd-XN]ACGCATGTTTATGCTTTCTTGCGTTGCG[Iowa black]	0.200
<i>I. trianguliceps</i>	<i>ITS2</i>	128		
F primer			CGCGAACGTTGGAATCGTAC	0.600
R primer			GGCTCCAGTATTCATCGGGG	0.600
Probe			[5HEX]CCGTGCGTCGTAGCCATCCGT[3IAbRQSp]	0.200

Table S3: Population level mean and median and interquartile range (IQR) of larval *Ixodes ricinus* tick burden on individual hosts, proportion of hosts infected with *B. burgdorferi* s.l., mean proportion of fed *I. ricinus* larvae, and mean proportion of fed and *B. burgdorferi* s.l. infected *I. ricinus* larvae on captured small mammals between 2018 and 2022 in Son, Viken county, Norway. SE denotes the standard error of the sample proportion for host infection, and model (logit) scale standard errors for larval feeding and larval infection, respectively.

	Larval burden			Host infection		Larval feeding		Larval infection	
	Mean	Median	IQR	%	SE	Mean	SE	Mean	SE
<i>Apodemus sylvaticus</i>	31	19	11-39	0.30	0.04	0.36	0.17	0.04	0.53
<i>Myodes glareolus</i>	24	16	9-31	0.40	0.03	0.31	0.17	0.11	0.43
<i>Sorex araneus</i>	20	13	6-20	0.51	0.05	0.46	0.22	0.07	0.42

Table S4: Estimates of parameters in generalized linear mixed models on successful feeding in *I. ricinus* larvae and nymphs, and *B. burgdorferi* s.l. infection in *I. ricinus* larvae, on small mammals captured in South-East Norway (2018-2022) as a function of season and whether the host was found dead or alive. Significant *P*-values are italicized. Random intercepts of “trap station”, “host ID” and “year” were included. The intra-class correlation (ICC) of the random intercepts were calculated by the R-package performance version 0.10.4.

Parameter	Estimate	Std. error	<i>z</i>	<i>P</i>
<u>Larval feeding</u> [2]	(logit-link)			
Intercept [1]	-1.197	0.206	-5.800	<0.001
Season = Spring	-0.087	0.127	-0.684	0.494
Status = Live	0.666	0.118	5.650	<0.001
<u>Nymphal feeding</u> [3]	(logit-link)			
Intercept [1]	-0.449	0.392	-1.146	0.252
Season = Spring	0.323	0.342	0.945	0.345
Status = Live	0.772	0.373	2.068	0.039
<u>Larval infection</u> [4]	(logit-link)			
Intercept [1]	-3.144	0.379	-8.292	<0.001
Season = Spring	-0.336	0.574	-0.585	0.559
Status = Live	0.771	0.530	1.455	0.146

[1]: The intercept corresponds to a host found dead in fall.

[2]: The ICC of the random intercept “host ID” = 0.202 and “year” = 0.040. The random intercept variance for “trap station” was close to zero.

[3]: The ICC of the random intercept “host ID” = 0.156, “year” = 0.016, and “trap station” = 0.007.

[4]: The ICC of the random intercept “trap station” = 0.274. The random intercept variance for “host ID” and “year” were close to zero.