



Cox, R. and Groner, M. L. and Todd, C. D. and Gettinby, G. and Patanasatienkul, T. and Revie, C. W. (2017) Mate limitation in sea lice infesting wild salmon hosts : the influence of parasite sex ratio and aggregation. Ecosphere, 8 (12). ISSN 2150-8925 , <http://dx.doi.org/10.1002/ecs2.2040>

This version is available at <https://strathprints.strath.ac.uk/64427/>

Strathprints is designed to allow users to access the research output of the University of Strathclyde. Unless otherwise explicitly stated on the manuscript, Copyright © and Moral Rights for the papers on this site are retained by the individual authors and/or other copyright owners. Please check the manuscript for details of any other licences that may have been applied. You may not engage in further distribution of the material for any profitmaking activities or any commercial gain. You may freely distribute both the url (<https://strathprints.strath.ac.uk/>) and the content of this paper for research or private study, educational, or not-for-profit purposes without prior permission or charge.

Any correspondence concerning this service should be sent to the Strathprints administrator: strathprints@strath.ac.uk

Mate limitation in sea lice infesting wild salmon hosts: the influence of parasite sex ratio and aggregation

R. COX,^{1,†} M. L. GRONER,¹ C. D. TODD,² G. GETTINBY,³ T. PATANASATIENKUL,¹ AND C. W. REVIE¹

¹Department of Health Management, Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, Prince Edward Island, C1A 4P3 Canada

²Scottish Oceans Institute, University of St. Andrews, St. Andrews, KY16 8LB UK

³Department of Mathematics and Statistics, University of Strathclyde, Glasgow G1 1XH UK

Citation: Cox, R., M. L. Groner, C. D. Todd, G. Gettinby, T. Patanasatienkul, and C. W. Revie. 2017. Mate limitation in sea lice infesting wild salmon hosts: the influence of parasite sex ratio and aggregation. *Ecosphere* 8(12):e02040. 10.1002/ecs2.2040

Abstract. Mate limitation in dioecious parasite species has the potential to impact parasite population growth. Our focus of interest was the influence of parasite sex distribution among hosts on parasite reproduction and transmission dynamics for populations of ectoparasitic sea lice (*Lepeophtheirus salmonis* Krøyer) establishing on wild juvenile salmon hosts. The data included more than 139,000 out-migrating juvenile pink salmon (*Oncorhynchus gorbuscha* (Walbaum)) and chum salmon (*Oncorhynchus keta* (Walbaum)) in British Columbia, Canada, sampled over nine years. For almost all years, the sex ratio of the reproductive stages of the sea lice was female-biased. The probability of a female being able to mate (i.e., of being attached to a fish also carrying a male louse) increased with increasing parasite abundance and parasite aggregation. We compared, with expected modeling predictions, the observed prevalence of pairs of sea lice (i.e., one reproductive louse of each sex) on a given fish and the observed probability of a female being able to mate. These comparisons showed that male and female sea lice tend to be distributed together rather than separately on hosts. Distribution together means that sea lice are distributed randomly on hosts according to a common negative binomial distribution, whereas distribution separately means that males are distributed according to a negative binomial and females are distributed in their own negative binomial among hosts. Despite the tendency for distribution together we found that, in every year, at least 30% of reproductive female sea lice experience mate limitation. This Allee effect will result in submaximal rates of parasite reproduction at low parasite abundances and may limit parasite transmission. The work has important implications for salmon parasite management and the health both of captive farm salmon populations and migratory wild stocks. More broadly, these results demonstrate the potential impact of mate limitation as a constraint to the establishment and spread of wild ectoparasite populations.

Key words: dioecious parasite; host–parasite interactions; mate limitation; sea lice; sex ratio; wild salmon.

Received 3 July 2017; revised 24 August 2017; accepted 1 September 2017; final version received 29 October 2017. Corresponding Editor: Andrew W. Park.

Copyright: © 2017 Cox et al. This is an open access article under the terms of the Creative Commons Attribution License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.

† **E-mail:** rucox@upe.ca

INTRODUCTION

In dioecious parasite species, the distribution of males and females among individual hosts in a population influences reproductive potential. For most species, parasites tend to be aggregated

among hosts (Anderson and Gordon 1982), and the host population therefore represents a collection of patches that harbor parasites (Poulin 2008). This aggregated distribution can result in an uneven frequency of encounter between males and females (Morand et al. 1993, Poulin 2008).

Parasite reproduction will depend on the parasite sex ratio and also on whether male and female parasites are distributed together or separately among hosts (May 1977). Distribution together assumes that the sex of each parasite is random and that individuals are equally likely to be female or male (assuming an equal sex ratio). At the other extreme, distribution separately results in independent distributions of each sex among their hosts and increases the probability of hosts being infested with parasites of just one sex. The difference between these extremes can alter parasite reproduction and, as a result, influence transmission to new hosts (Bradley and May 1978). Furthermore, it indicates that individual hosts in a host–parasite association could potentially comprise either a source or a sink of infection for the parasite population (Chaves et al. 2007, Fengyang and Poulin 2011).

Many dioecious parasite species have detrimental impacts on public or animal health. Understanding the heterogeneity of reproduction of such species can inform control and prevention strategies (Grant 2002). For example, an inability of an individual parasite to encounter a mate at low density—an Allee effect—could result in parasite transmission breakpoints (or tipping points; Macdonald 1965, May 1977). Conversely, ignoring the importance of mating strategies could lead to misinterpretation of the reproductive potential of empirically sampled parasites (Dittmar et al. 2011). Despite the importance of mate pairing in parasite transmission, it has most often been assessed only by theoretical modeling (May 1977, May and Woolhouse 1993, Cornell et al. 2004) and very rarely through empirical work (Morand et al. 1993, Krkošek et al. 2012).

The present study focuses on the aquatic ectoparasite salmon louse, *Lepeophtheirus salmonis* Krøyer, which is a caligid copepod associated with numerous species of host salmonid fishes (Boxaspen 2006). As for other ectoparasites of marine fish, this species can compromise the health of both wild and farmed hosts, and concerns persist regarding its regulatory role on host populations (Costello 2009, Jansen et al. 2012, Krkošek et al. 2013). As a dioecious species, *L. salmonis* reproduces sexually, with adult males typically forming mate-guarding pre-copulatory pairs with pre-adult females. Although the

transfer of spermatophores and extrusion of fertilized oocytes occur only once the female has molted to the adult stage (Ritchie et al. 1996), the pre-adult females are considered reproductively active because they can engage adult males in mate guarding (Hull et al. 1998). Both genders are mobile over the body surface of the fish, and both have limited swimming abilities. Initial infestation of salmon arises from host encounter of the final planktonic larval stage (copepodid). The duration of the planktonic developmental stages lasts long enough that self-reinfestation of a given host is extremely unlikely. At present, there is a knowledge gap concerning the sex ratio and the extent of co-occurrence of the genders of *L. salmonis* on host populations in both the Atlantic and Pacific oceans. These distributional patterns among host fish undoubtedly will have marked consequences for *L. salmonis* reproduction and are of fundamental importance to understanding successful transmission to new hosts.

To date, some empirical data and theoretical models for wild fish (Krkošek et al. 2012) have suggested that mate limitation may constrain population growth when sea lice occur at densities below two adult females per host. Furthermore, although the population dynamics of the parasite on farmed and wild fish are quite different, empirical data (Stormoen et al. 2013) and models (Stormoen et al. 2013, Groner et al. 2014) also suggest constraints on mating at densities below two to three adult females per farmed Atlantic salmon. Available models do not, however, take account of the effect of male and female distribution among the hosts, which theory suggests can alter mating success (May 1977). These models also make assumptions about equal sex ratios and aggregation, which are not well quantified for sea lice. Here, we address the need for quantification of parasite sex aggregation and its predicted consequences on population demographics.

The reason that little is known about the sex distribution of *L. salmonis*, or indeed of any marine or terrestrial ectoparasite, among host individuals, is that neither scientific studies nor commercial monitoring typically reports parasite sex. For wild and farmed salmon species, data collection of sea lice infestations typically focuses on enumerating attached larval (chalmus) stages and mobiles (i.e., pre-adult and adult stages) on host fish; although adult females and gravid

females are commonly distinguished and recorded, the incidence of pre-adult and adult males is recorded only rarely (Peacock et al. 2016). Among the few studies that have recorded sea louse sex, there does not appear to be a consensus regarding the sex ratio of reproductive stages. Studies of wild Pacific or Atlantic salmon have indicated either a predominance of adult female sea lice (Todd et al. 2000, Gottesfeld et al. 2009) or of adult male sea lice (Todd et al. 2005, Connors et al. 2010). Studies on farmed Atlantic salmon (which, in some cases, have been repeatedly exposed to anti-parasitic treatments) have reported evidence for male dominance (Bron et al. 1993), female dominance (Todd et al. 2005), and either male or female dominance at different farm locations (Revie 2006). It is therefore likely that local and regional characteristics of the host population (e.g., fish size and age), the parasite population (e.g., parasite dispersal), and environmental conditions (e.g., season, temperature) may influence the parasite sex ratio.

Understanding the mechanisms that influence sea louse reproductive potential is of ecological and practical importance. Such knowledge will have applications for the management of both farmed and wild salmon, with specific implications regarding the method of louse monitoring (which usually distinguishes only female adults) on farmed fish, the reliability and applicability of host-parasite population models, and the efficacy of treating farmed fish in order to control salmon louse populations. To better understand the dynamics of parasite transmission among wild hosts of juvenile Pacific salmon, we used nine years of empirical data to determine the sex ratio of sea lice on individual hosts. We compared the results to theoretical models to determine how male and females are distributed on hosts. From these results, we determined the potential for female sea lice to mate and we assessed both the extent and likely consequences of mate limitation in these populations.

METHODS

Data description

We analyzed an existing database detailing counts of *Lepeophtheirus salmonis* on juvenile wild Pacific salmon in the Knight Inlet and Broughton Archipelago regions of British Columbia, Canada

(Broughton Archipelago Monitoring Program [BAMP] initiative). The data, which span the years 2004–2012, were compiled from three separate studies which used different sampling protocols. For one study, between 2004 and 2009, fish were caught by beach seine netting and were sampled live (Krkošek et al. 2005). In the second study, also between 2004 and 2009, fish were lethally sampled by beach or purse seine netting by the Department of Fisheries and Oceans Canada (Jones and Hargreaves 2007). Purse seine netting occurs from a ship and is necessarily undertaken in deeper water than beach seine netting. In 2010, a third study was initiated under BAMP that merged the data from these two earlier studies; it also adopted a standard protocol that comprised only beach seine netting and lethal sampling (Patanasatienkul et al. 2013). A subset of these data (live sampled fish caught by beach seine netting in 2006–2009) has been used in a previous publication about mate limitation (Krkošek et al. 2012). Another subset of the data (the lethally sampled fish caught by purse and beach seine netting in 2004 and 2005) has been used to record abundance and sex ratio of sea lice (Connors et al. 2010). However, neither study quantified the relationship between male and female parasite distribution among hosts.

Repeated sampling took place each year at either weekly, biweekly, or monthly intervals between March and July. In order to assess within-year temporal patterns of salmon louse infestations, we separated the annual data into three sampling units of approximately equal duration and host sample number, which we denote as early (days of the year 83–124), middle (days of the year 125–155), and late (days of the year 156–188). In total, there were 150 different sampling locations across all years, with the number of locations sampled within a given year varying between 78 (2012) and 118 (2007 and 2008).

Sampled fish were out-migrating juvenile pink (*Oncorhynchus gorbuscha* (Walbaum)) and chum salmon (*Oncorhynchus keta* (Walbaum)), which ranged in length from 21 to 174 mm (average 53 mm). After the initial description of the data in the *Results* section, the data for both species were combined for analyses. Although some differences in the level of aggregation of sea lice between the two-host species have been noted (Krkošek et al. 2012), there was no difference in

prevalence of sea lice between species, and infestation intensity was significantly different in fewer than half of the years studied (Patanasatiengkul et al. 2013). We decided, therefore, that we did not wish to introduce host species as another variable into our analysis. Moreover, combining the data for the two-host species is reasonable because these salmonids comprise mixed-species shoals within which parasites potentially can disperse among fish (Krkošek et al. 2012). It is important to note that these salmon were infested both with *L. salmonis* and another caligid copepod, *Caligus clemensi* (Parker and Margolis 1964), which was identified where possible to species, developmental stage, and sex. Our analysis focused on *L. salmonis* only. The following classifications were used for the parasites sampled from the body surface of each fish: chalimus, pre-adult male, pre-adult female, adult female (without egg strings), gravid female (with egg strings), and adult male. We use the following definitions throughout: mobile sea lice = pre-adults + adults; reproductive females = pre-adult females + adult females + gravid adult females; reproductive males = adult males.

We include both pre-adult and adult female sea lice as reproductive stages. Pre-adult females are included because adult males preferentially mate-pair with pre-adult females (Hull et al. 1998). A mate-guarding male thus cannot interact with any other female. Adult females were included because adult males may also mate-pair with them (Todd et al. 2005). Exclusion of adult females would underestimate the true mating probability (Krkošek et al. 2013). Although males mate preferentially with pre-adult females, we did not separate pre-adult and adult females into two different analyses because adult females are able to mate and be fertilized repeatedly by different males (Todd et al. 2005). Pre-adult males were excluded from the reproductive male category because they do not form reproductive pairs (Krkošek et al. 2012).

Data analysis

Preliminary descriptive data were derived for the prevalence of infestation (\pm Clopper–Pearson 95% confidence intervals [CI] for a binomial distribution), mean abundance, and mean intensity of male and female pre-adults and adults (\pm 95% bias-corrected and accelerated bootstrap CIs

estimated for 2000 iterations) using QPweb (Reiczigel et al. 2013). Abundance is defined as the number of parasites per sampled host, and intensity is defined as the number of parasites per infested host (Margolis et al. 1982).

Nonparametric Spearman rank correlation was used to compare the number of male sea lice vs. female sea lice on fish.

Aggregation of sea lice was calculated using the following two related measures. We include both measures to enable comparison with existing literature.

1. Variance-to-mean ratio (VMR) = (s^2/m) , where m = mean parasite load per host, s = standard deviation, and n = number of hosts. Variance-to-mean ratio ≥ 1 indicates a negative binomial distribution.
2. Aggregation coefficient k of the negative binomial, calculated by maximum-likelihood estimation using QPweb (Reiczigel et al. 2013). k is inversely proportional to the level of aggregation, in that $k = 5$ is considered aggregated whereas $k = 1$ is highly aggregated (Wilson et al. 2001). Note the relationship between VMR and k : $VMR = m/k + 1$. As k tends to infinity, the negative binomial distribution approaches the Poisson with $VMR = 1$.

Two measures were used to assess male and female sea louse infestation on hosts:

$P_{(o)}$: Observed fraction of the host population infested with at least one male plus one female louse.

$\Phi_{(op)}$: Observed pairing probability of female sea lice, that is, the fraction of female sea lice on hosts that have the opportunity to mate with a male. Although males and females tend to be spatially segregated on large adult host fish (Todd et al. 2000), for the present juvenile salmon we made the assumption that a female louse on a fish had the opportunity to mate if a male was present on the same host. Pairing probability was calculated for a promiscuous population, $\Phi_{(op)}$; that is, all females on a host are presumed to mate, provided that at least one male is present on the same host. Genetic paternity analyses show that *L. salmonis* are promiscuous on wild and farmed Atlantic salmon and multiple mating is not uncommon in this species (Todd et al. 2005). For comparison, we made the same calculations for a monogamous

population, $\Phi_{(om)}$; that is, one female mates with one male on the same host.

Observed values of P and Φ were compared to expected estimates using a chi-squared test. Due to the number of tests conducted ($n = 31$), a Bonferroni correction using $p < 0.0016$ was applied. Expected values were calculated using equations formulated by May (1977), which estimate male–female aggregation for two different and extreme cases: (1) males and females distributed together, or (2) males and females distributed separately among host individuals. In case 1, the genders are distributed randomly in a single negative binomial distribution with mean m and parameter k , each individual louse is equally likely to be female or male. Case 2 is the converse extreme, whereby female and male sea lice are distributed separately, with female sea lice distributed in a negative binomial (with mean $m/2$ and parameter k) and the males also distributed in their own negative binomial distribution (also with a mean $m/2$ and parameter k). For each equation: m = mean parasite load per host; k = aggregation coefficient of parasites in the host population.

Case 1: Males and females are distributed together:

$P_{(t)}$ is the inverse of the probability that there are no sea lice on fish or that both sea lice are of the same sex, and is calculated according to May (1977) as:

$$P_{(t)}(m, k) = 1 - 2(1 + m/(2k))^{-k} + (1 + m/k)^{-k}$$

$\Phi_{(t)}$ is the probability that a given female will be paired and for a promiscuous population is as follows:

$$\Phi_{(t)}(m, k) = 1 - (1 + m/(2k))^{-1-k}$$

Case 2: Males and females are distributed separately:

$P_{(s)}$ is the product of the probability that there is one or more female and the probability that there is one or more male:

$$P_{(s)}(m, k) = (1 - (1 + m/(2k))^{-k})^2$$

$\Phi_{(s)}$ is the probability that a given female will be paired and for a promiscuous population is as follows:

$$\Phi_{(s)}(m, k) = 1 - (1 + m/(2k))^{-k}$$

The models predict that $P_{(o)}$ and $\Phi_{(o)}$ will be greater in a population where genders are distributed together rather than separately. As k tends to infinity (i.e., parasites are distributed randomly according to the Poisson), cases 1 and 2 converge.

Although these models make the assumption—which is not adhered to in our data—that the sex ratio is unity, Bradley and May (1978) indicate that this assumption is not of qualitative importance. However, because our data show an unequal sex ratio, we also compared our results to theoretical populations that have a biased sex ratio (May and Woolhouse 1993), for a promiscuous population:

$$\Phi(q; m, k) = 1 - (1 + mp/k)^{-k-1}$$

where q = the fraction of all reproductive sea lice that are female, and $p = 1 - q$ the fraction of male sea lice. The assumption of this model is that male and female sea lice are aggregated together rather than separately (case 1 described above). We were unable to compare to a model that assumes male and females are aggregated separately (case 2) because May and Woolhouse (1993) did not formulate case 2.

RESULTS

Descriptive analysis

A total of 143,234 salmon were sampled between 2004 and 2012, of which 51% were pink ($n = 72,609$) and 49% were chum ($n = 70,625$). Of these fish, 2.3% (3297) were excluded from analysis (3221 of which were caught in 2004): 3261 fish carried sea lice that were not identified to species, 17 were infested with mobile sea lice that were not identified to sex, and 19 were infested with mobile sea lice that were not identified to adult or pre-adult. These fish had been caught primarily using beach seine nets (80 pink, 3206 chum) compared to ship-based purse seine netting (9 pink, 2 chum). Further analysis was conducted on the remaining 139,937 fish (52% pink; 48% chum).

The majority (88%) of fish were caught using beach seine (64,938 pink; 58,284 chum) and 12% using purse seine (7582 pink, 9133 chum). Previous analysis of the same dataset has shown that

there are significant associations between the gear type and fish size, with purse seine-caught fish being significantly longer than those captured with a beach seine at the same location and time (Patanasatiengkul et al. 2013). Because fish size may influence parasite load (Patanasatiengkul et al. 2013), separate analyses were conducted on fish that were caught using the two different methods.

Lepeophtheirus salmonis prevalence and intensity

The prevalence of mobile *L. salmonis* varied between years, being lowest in 2009 when the proportion of fish infested was 0.016 (0.014 [95% CI: 0.013, 0.016] beach-caught and 0.029 [95% CI: 0.023, 0.036] purse-caught), and highest in 2004, when the proportion of fish infested was 0.193 (0.121 [95% CI: 0.115, 0.127] beach-caught and 0.382 [95% CI: 0.367, 0.397] purse-caught; Appendix S3: Table S1). Co-occurrence of *Caligus clemensi* is shown in Appendix S4: Table S1. The predominant fish species differed among years (in particular, pink dominated from 2008 onwards, while chum dominated in 2004). For all years, a total of 6.6% (9302) of fish were infested with mobile sea lice, showing a mean intensity of 0.85 (95% CI: 0.843, 0.857) females and 1.06 (95% CI: 1.029, 1.631) males (Appendix S1: Fig. S1).

Sex distribution of L. salmonis

The proportion of mobile sea lice that were female in a given year varied between 0.32 and 0.53 (Fig. 1). This proportion only exceeded 0.5 (indicating a greater proportion of female than male sea lice) for purse seine-caught fish in 2006. On excluding the pre-adult males (which are not yet reproductively active), the sex ratio of reproductive sea lice was female-biased in all years, with the proportion of sea lice that were female ranging between 0.63 and 0.77 (beach seine) and between 0.61 and 0.76 (purse seine).

Distribution of male and female sea lice on individual fish

We now focus on the occurrence of male and female sea lice on individual fish. In total, 6506 fish were infested with at least one reproductive sea louse (Table 1). The majority carried sea lice of only one sex, while only 1364 (21%) fish carried reproductive sea lice of both sexes. The sex ratio of reproductive sea lice on hosts was female biased (Fig. 2). There were only 31 gravid females recorded, which infested 30 different fish. Such a low frequency of gravid female parasites is a reflection of the hosts being juvenile and having been in saltwater for only a few weeks.

For fish infested with reproductive sea lice of both sexes ($n = 1364$), 607 had an equal proportion

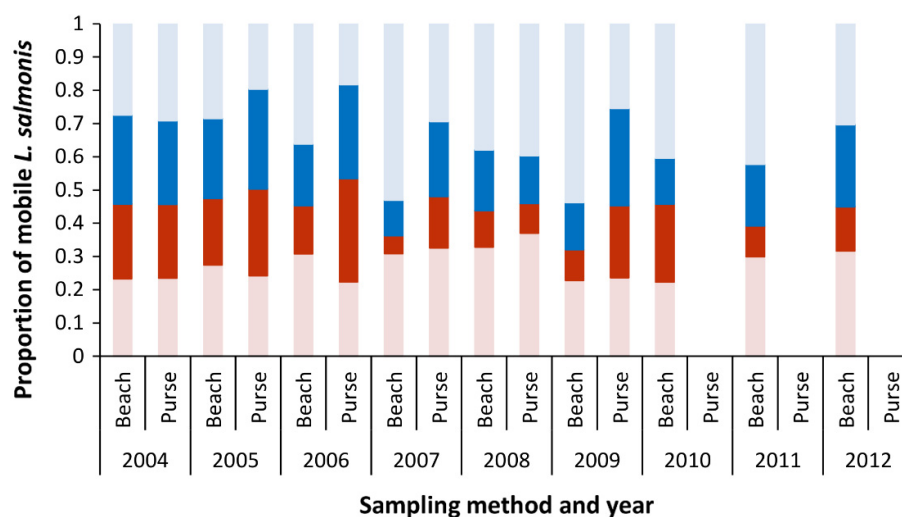


Fig. 1. Proportion of male and female life stages in the population of mobile *Lepeophtheirus salmonis*. Figure includes only fish infested with at least one mobile louse ($n = 9302$). Pre-adult female: light pink; adult female: dark red; adult male: dark blue; pre-adult male: light blue.

Table 1. Distribution of reproductive *Lepeophtheirus salmonis* on wild Pacific salmon sampled between 2004 and 2012 in the Broughton Archipelago Monitoring Program (BAMP) region.

Year and seine net	No. fish								Maximum no.	
	Sampled	Infested with reproductive sea lice	Infested with F only	Infested with M only	Infested with F and M	SR > 0.5 (F-biased)	SR < 0.5 (M-biased)	SR = 0.5	F per fish	M per fish
Beach										
2004	11,174	1163	480	207	476	239	71	166	22	12
2005	4022	413	235	105	73	25	1	47	6	4
2006	17,668	845	578	190	77	18	7	52	5	10
2007	23,933	662	501	123	38	5	7	26	4	4
2008	25,901	717	470	148	99	35	6	58	5	7
2009	21,419	142	98	43	1	0	0	1	2	1
2010	5975	199	150	37	12	1	1	10	2	2
2011	5922	52	34	15	3	0	0	3	2	1
2012	7208	294	182	91	21	3	2	16	3	4
Purse										
2004	4320	1363	632	254	477	227	76	174	30	21
2005	2176	232	133	66	33	7	6	20	6	4
2006	2264	161	98	42	21	5	1	15	3	4
2007	2006	71	47	15	9	2	2	5	3	2
2008	3094	127	88	18	21	7	2	12	3	3
2009	2855	65	41	21	3	0	1	2	3	4
2010	0	–	–	–	–	–	–	–	–	–
2011	0	–	–	–	–	–	–	–	–	–
2012	0	–	–	–	–	–	–	–	–	–
Total	139,937	6506	3767	1375	1364	574	183	607		

Notes: Beach, beach seine netting; Purse, purse seine netting; M, male; F, female (adult, pre-adult, and gravid); SR, sex ratio. Dashes indicate no fish were sampled.

of male and female sea lice, while the proportion of females was >0.5 (female-biased) on 574 fish and <0.5 (male-biased) for 183 fish (Table 1). There was a positive correlation between the number of male and female reproductive sea lice on these fish (Spearman rank correlation: Beach: $r = 0.526$, $p < 0.001$, $n = 800$; Purse: $r = 0.478$, $p < 0.001$, $n = 564$; Appendix S2: Fig. S1).

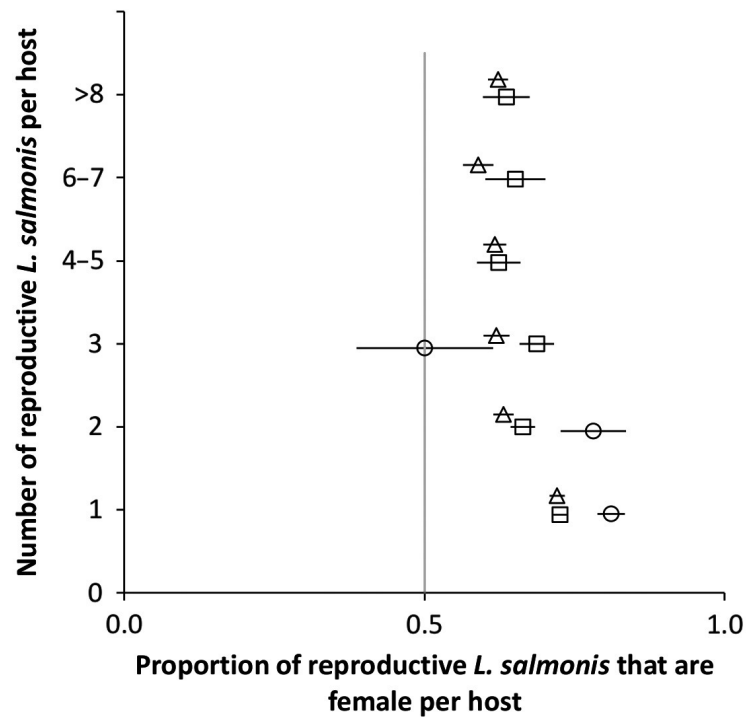
Probability of *L. salmonis* mate encounter

The indices of aggregation (VMR, k), the prevalence of reproductive sea lice, prevalence of pairs of reproductive sea lice ($P_{(o)}$), and pairing probability of female reproductive sea lice ($\Phi_{(o)}$) are all shown for early, middle, and late sampling units within years, and separated by capture method in Appendices S5 and S6. We excluded just ten sampling units when <10 sea lice were sampled. We also excluded three sampling units when the calculation of k returned a value of infinity. The latter occurred when the mean and variance were equal (and thus $VMR = 1$ and $k = m/(VMR - 1)$).

The mean prevalence of reproductive sea lice within a sampling unit ranged between 0 and 0.559 (95% CI for maximum: 0.532, 0.585) for beach seine-caught fish and between 0 and 0.323 (95% CI for maximum: 0.302, 0.346) for purse seine-caught fish (Appendix S6: Table S1). For all sampling units, the distribution of reproductive sea lice among the host population followed a negative binomial indicated by a VMR > 1 . For beach seine-sampled fish, VMR ranged from 1.03 to 5.04, whereas for purse seine-sampled fish, VMR ranged from 1.08 to 6.44 (Appendix S5: Table S1). The VMR of reproductive males exceeded that of reproductive females in 7 of 20 beach seine units and 5 of 11 purse seine units. With the inclusion of pre-adult male sea lice in the calculation of VMR, the VMR of mobile males exceeded that of mobile females in 14 of 20 beach seine units and for 10 of 11 purse seine units.

We also compared how louse abundance and VMR influenced $\Phi_{(op)}$. As expected, $\Phi_{(op)}$ increased with VMR and as the mean abundance increased (Fig. 3a, b, respectively). The assumption

a) Beach seine netting



b) Purse seine netting

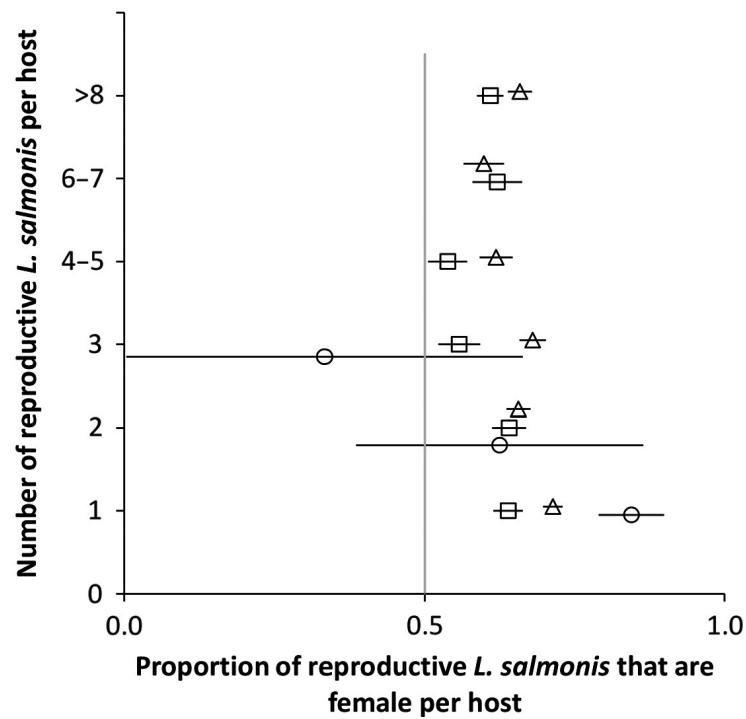


Fig. 2. Sex ratio of *Lepeophtheirus salmonis* as a function of infestation intensity on wild Pacific salmon. Data for

(Fig. 2. *Continued*)

pink and chum are combined, and data are pooled across years. Figures include only fish infested with at least one reproductive louse ($n = 6506$). Symbols represent sampling time units (\pm standard error). Circle: early; square: middle; triangle: late. The vertical gray line indicates a sex ratio of 0.5.

of promiscuous mating also resulted in higher mating probabilities than monogamous mating in most sampling units (12 of 20 beach seine units and 9 of 11 purse seine units). Values of $\Phi_{(op)}$ ranged from 0 to 0.71, whereas values of $\Phi_{(om)}$ were consistently lower and ranged from 0 to 0.45 (Appendix S6: Table S1). Fig. 3 highlights, under the assumption of promiscuous mating, that a maximum of 71% of the female louse population was able to find a mate on the same host at the time of sampling. Thus, at least 29% (and, in some years, up to 100%) of the female louse population were unable to find a mate. Thus, mate limitation occurred in all of our sampling units spanning abundances between 0 and 2 reproductive sea lice per host and for VMR ranging between 0 and 7.

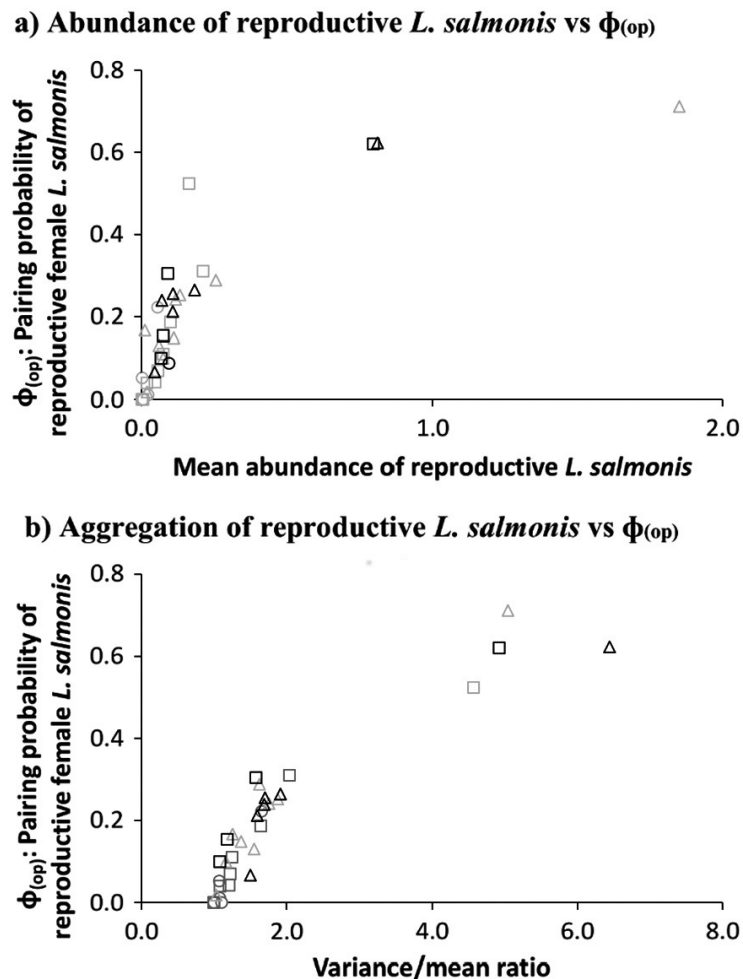


Fig. 3. Interaction between the abundance of *Lepeophtheirus salmonis* on fish, the level of *L. salmonis* aggregation, and the pairing probability of reproductive female *L. salmonis* ($\Phi_{(op)}$). Symbols represent sampling time units. Circle: early; square: middle; triangle: late. Light gray: beach seine netting; black: purse seine netting.

Mate limitation may well occur at abundances exceeding two reproductive sea lice per host, but data were not available beyond this range.

Distribution of sea lice together or separately

As noted previously, of the fish that were infested with reproductive sea lice, the majority (79%) were infested with sea lice (or a single louse) of only one sex. In order to formally test whether the genders tend to be distributed together or separately, we compared the observed and estimated values of P and Φ using a chi-squared test (Appendix S6: Table S1).

For beach-caught fish, $P_{(o)}$ (range: 0–0.265 [95% CI for maximum: 0.242, 0.290]) was not significantly different from $P_{(t)}$ in any of the 20 sampling units. $P_{(o)}$ was significantly different from $P_{(s)}$ in 13 of 20 sampling units. For purse seine-caught fish, $P_{(o)}$ (range: 0–0.11 [95% CI for maximum: 0.099, 0.129]) was not significantly different from $P_{(t)}$ for any of the 11 sampling units, whereas, by contrast, $P_{(o)}$ was significantly different from $P_{(s)}$ in 6 of 11 sampling units. Results suggest that male and female sea lice tend to be distributed together on hosts, because $P_{(o)}$ tended to be more similar to $P_{(t)}$ rather than to $P_{(s)}$ (Fig. 4a). If males

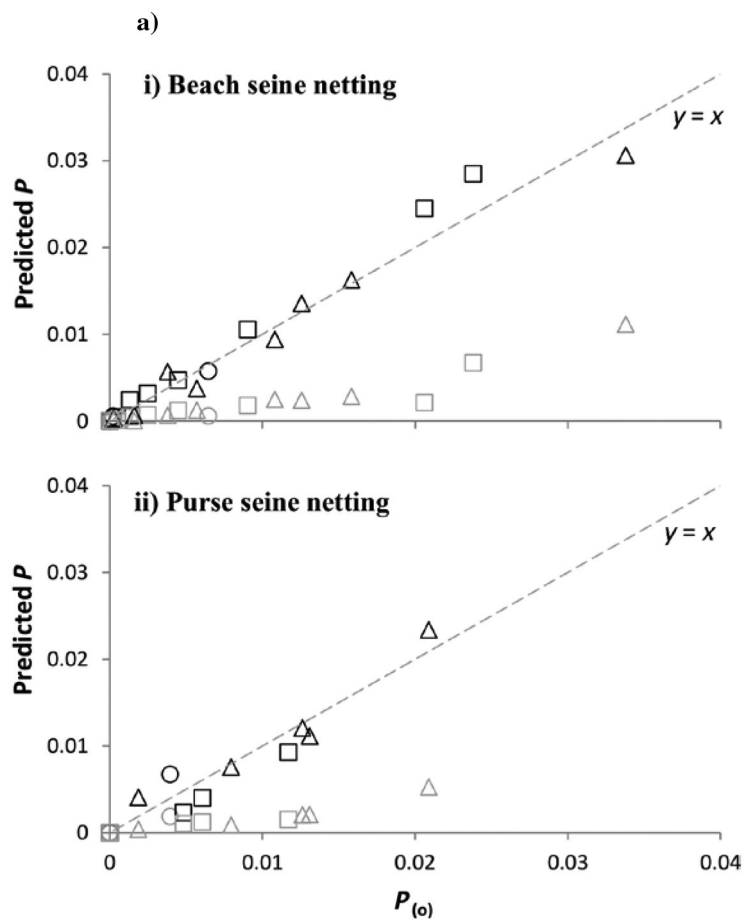
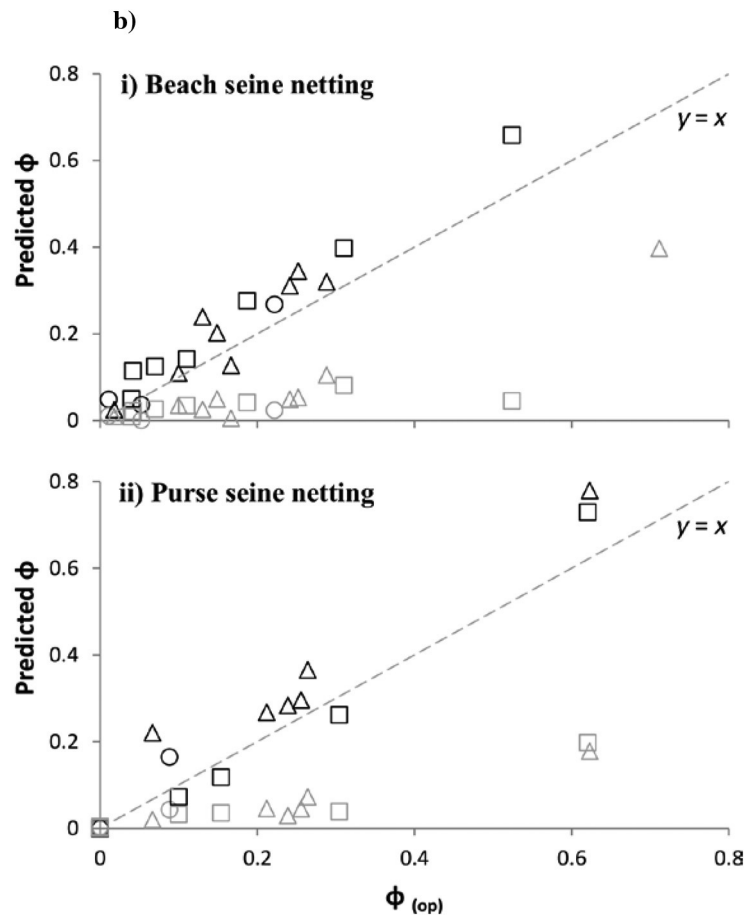


Fig. 4. Comparison of observed and estimated values of the prevalence of male–female pairs of *Lepeophtheirus salmonis* (P) and of the pairing probability of reproductive female *L. salmonis* on host fish (Φ). Symbols represent sampling time units. Circle: early; square: middle; triangle: late. The dashed line indicates equality of observed and predicted values of either P or Φ . (a) Comparison of $P_{(o)}$ (x -axis) with predicted $P_{(t)}$ (black) and $P_{(s)}$ (gray). (b) Comparison of $\Phi_{(op)}$ (x -axis) with predicted $\Phi_{(t)}$ (black) and $\Phi_{(s)}$ (gray). (c) Comparison of $\Phi_{(op)}$ (x -axis) with predicted $\Phi_{(t)}$ (black) and $\Phi_{(s)}$ (gray) assuming equal sex ratio (as in b). Additional red data points show $\Phi_{(t)}$ for a population with unequal sex ratio.



(Fig. 4. Continued)

and females were distributed separately, an even greater proportion of the host fish would be expected to be infested with sea lice of one sex (greater than the 79% recorded here) and a smaller proportion to be infested with both genders (i.e., a value closer to $P_{(s)}$).

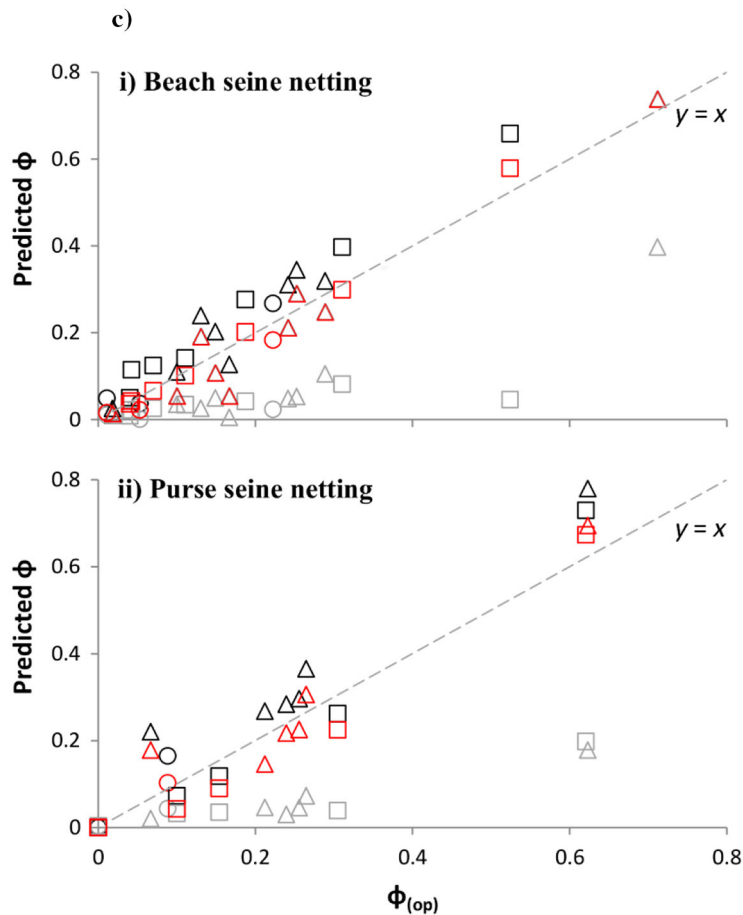
For beach-caught fish, $\Phi_{(op)}$ was significantly different from $\Phi_{(t)}$ in 12 of 20 sampling units, and from $\Phi_{(s)}$ in 18 of 20 sampling units. Fig. 4b shows that $\Phi_{(op)}$ was consistently more similar to $\Phi_{(t)}$ than to $\Phi_{(s)}$. The results for purse seine-caught fish were similar. Note that $\Phi_{(op)}$ was significantly different from $\Phi_{(t)}$ in 6 of 11 sampling units, and from $\Phi_{(s)}$ in 11 of 11 sampling units.

Using additional formulations provided by May and Woolhouse (1993), we showed that $\Phi_{(t)}$ for a biased sex ratio tended to be less than $\Phi_{(t)}$ for an unbiased sex ratio; however, values differed only by a mean of 0.06 ± 0.008 for beach-

and purse seine-caught fish (Fig. 4c). $\Phi_{(op)}$ was significantly different from $\Phi_{(t)}$ for a biased sex ratio in one sampling unit for beach-caught fish and in two sampling units for purse seine-caught fish. Although models to estimate $\Phi_{(s)}$ for unequal sex ratios were not available for comparison, our analysis supports the hypothesis that male and female sea lice tend to be distributed together.

DISCUSSION

The availability of mates will promote or limit the reproductive potential of individuals within any population. Such fundamental constraints could be exacerbated for aquatic ectoparasites because they tend to be isolated from potential mates infesting other host fish. We have shown that for wild out-migrating juvenile Pacific



(Fig. 4. Continued)

salmon, only a small proportion of fish (between 0.01% and 27% of fish per sampling unit per year) was infested with at least one male and one female reproductive *Lepeophtheirus salmonis* (i.e., $P_{(o)}$ ranged between 0.0001 and 0.265). Furthermore, at the time of sampling, only a small proportion of the reproductive female population was attached to hosts where they could encounter a reproductive male louse on the same host. In all years, except 2004, between 72% and 97% of reproductive females were attached to a host without a male and were therefore unable to mate. 2004 showed the highest abundance of sea lice, and yet in that year, there was still a considerable proportion of reproductive female sea lice (36%) that were unable to mate at the time of sampling. Such mate limitation in these early stages of parasite establishment indicates a

submaximal rate of reproduction which will likely reduce effective parasite transmission.

Lepeophtheirus salmonis aggregation

The aggregated distribution ($k < 1$) of reproductive sea lice that we recorded is typical among parasite populations (Anderson and Gordon 1982, Shaw and Dobson 1995, Shaw et al. 1998). The pattern likely results from multiple factors including heterogeneity in (1) individual host exposure to infection (Murray 2002) and host susceptibility (Lysne and Skorping 2002, Ugelvik et al. 2017), (2) sea louse choice of host, or (3) competition with other parasite species (Morand et al. 1999, Bottomley et al. 2005). As expected, we showed that increased aggregation tended to increase the probability of female pairing (May 1977, Harrison and Bennett 2012).

There did not appear to be a clear difference in the level of aggregation of male and female reproductive sea lice; however, among the mobile sea lice, the VMR of mobile males exceeded that of mobile females in the majority of sampling units (14/20 beach seine sampling units and 10/11 purse seine sampling units). The movement of male sea lice between hosts is a possible mate-searching behavior and has been observed in farmed fish held in high-density cages (Ritchie 1997), under experimental laboratory conditions in aquaria (Hull et al. 1998, Bandilla et al. 2008, Connors et al. 2008, 2011, Stephenson 2012) and for juvenile host fish in sentinel cages in the field (Pert et al. 2014). Perhaps more surprisingly, it has been inferred for free-ranging wild hosts in the natural environment (Connors et al. 2010). Such movement is likely to be a contributory factor to the higher level of male aggregation.

Lepeophtheirus salmonis sex ratio

Biased sex ratios are to be expected in any natural population (Poulin 2008, Dittmar et al. 2011) and can arise from differential selective pressures acting on either sex at any developmental stage (Trivers and Willard 1973). One mechanism that is likely to generate the biased ratios observed in the present data is the differential rate of development and survivorship of the sexes. The female bias among the reproductive sea lice (in part due to our inclusion of pre-adult females but exclusion of pre-adult males) may arise because females tend to live longer than males (Stien et al. 2005, Todd et al. 2006). Increased mortality as a result of risky mate-searching behavior by males could also increase this female bias. The male bias of the mobile sea lice (resulting from a dominance of pre-adult sea lice, which is characteristic of recent infestation) occurs because males develop to the pre-adult stage more quickly than females (Johnson and Albright 1991).

Sex ratios can depend also on parasite density and level of aggregation. The sex ratio of nematode species, for example, is less strongly female-biased at higher infestation intensities (Poulin 1997). Differences in infestation intensity therefore may be one contributory factor to the variation among reports regarding sea louse sex ratios on salmon species. The present study demonstrates a male bias for the mobile sea louse stages, which concurs with a prior study of Pacific salmon that

also reported male bias for adult sea lice on juvenile hosts (and which used a subset of the data; Connors et al. 2010). Furthermore, we showed female bias for the reproductive stages, which is commensurate with most studies of adult wild Pacific and Atlantic salmonids (Bristow et al. 1996, Jacobsen and Gaard 1997, Todd et al. 2000, Beamish et al. 2005, Butterworth et al. 2008, Gottesfeld et al. 2009). These latter reports are for hosts carrying a predominance of female sea lice because they had resided at sea for some time and likely had little opportunities for new infestation.

Compared to an equal sex ratio, a female bias would have a limiting effect on the pairing probability of individual females if the mating system were monogamous. Nonetheless, the pairing probability does increase when the mating system is promiscuous (May and Woolhouse 1993, Morand et al. 1993), as is the case for *L. salmonis* (Todd et al. 2005). As expected, therefore, promiscuous mating resulted in higher mating probabilities with estimated $\Phi_{(op)}$ being up to 1.8 times greater than $\Phi_{(om)}$.

Aggregation of genders together or separately

In 1978, Bradley and May stated that it would be valuable to have more evidence regarding when aggregation together and separately should be expected. However, more than 30 yr later we could find few data that describe the aggregation of male and female parasites within any host population. Our dataset provided a unique opportunity to assess the parasite sex distribution on >100,000 host individuals over a 9-yr period, to better understand the success of individual reproduction and its consequences for parasite transmission dynamics.

Although the presently sampled host fish were juvenile, out-migrating salmon in their first months of infestation, parasite abundance was sufficiently high for us to conclude that males and females tend to be distributed together on hosts. While a large proportion of infested fish (79%) carried sea lice of only one sex, we demonstrated a positive correlation between numbers of male and female sea lice on fish that were infested with at least one louse of each sex. This is important because a negative correlation might indicate differences between sexes in infestation rates or host preference, or some avoidance of the opposite sex. We then showed, with theoretical models for the individual host fish, that male and female sea lice

tend to be distributed together rather than separately, across most sampling units. Of course, the level of separation of the genders is, in reality, a continuum. Statistically, distribution separately is more likely when the average parasite load is low (because there are only a few parasites, which are unlikely to be attached to the same fish). Conversely, distribution together is more likely when the infestation load is high (with more fish likely to be infested with parasites of both genders; Bradley and May 1978). In order to compare distribution together vs. separately, we applied models that assumed an equal sex ratio because models for unequal sex ratio were not available. Two further analyses do, however, support our conjecture of aggregation together. First, when assuming an equal sex ratio, $\Phi_{(t)}$ was only slightly greater than $\Phi_{(t)}$ for a female-biased sex ratio; and second, $\Phi_{(op)}$ was similar to $\Phi_{(t)}$ for a female-biased sex ratio. It may therefore be reasonably assumed that values of $\Phi_{(s)}$ for a female-biased sex ratio would be considerably smaller.

Mate limitation

Although theoretical models have been used to show how mating probabilities under different mating systems may result in an Allee effect (May 1977, Cornell et al. 2004), the phenomenon has rarely been assessed empirically because comprehensive datasets comprising sex-specific parasite information are scarce. Our analyses provide empirical evidence in support of the assertion that mate limitation, or a component Allee effect, does indeed apply to these populations and that this may reduce the effectiveness of parasite transmission. From the present data, at the time of sampling, at least 36% (and in most years more than 70%) of the female louse population per year had not yet had the opportunity to mate in this establishing population in which the abundance of reproductive sea lice per host varied between 0 and 1.85, and the abundance of reproductive female sea lice per host varied between 0 and 1.16. Although it is not surprising that pairing was low at low sea louse abundance, it would be of considerable ecological and applied value to better understand how the level of pairing changes at abundances above the levels recorded in these data. Only Krkošek et al. (2012; using a subset of the 9 yr of data that we analyzed) and the present study have investigated whether an

Allee effect pertains to the population dynamics of sea lice colonizing wild juvenile Pacific salmon.

In support of our empirical data, Krkošek et al. (2012) previously showed theoretically that mate limitation could occur when parasite abundance fell below approximately two per host fish. Furthermore, their modeling indicated that an Allee effect is not necessarily expected at mean abundances below two sea lice per host. It is noteworthy that they did suggest that their work may have overestimated the potential for an Allee effect because the adult females were excluded from the analysis. Note that here we chose to include adult females in our analyses, as well as allowing for promiscuity, and yet we could show that mate limitation does indeed occur.

Clearly, our data represent only a series of snapshots in time. A female louse that is solitary on a host fish might have the opportunity to mate in the future (Ritchie et al. 1996, Hull et al. 1998), and once mating occurs, that female will be able to produce multiple egg strings. Furthermore, any behavioral or developmental constraint causing mate limitation will be dampened as the abundance of sea lice increases, as well as by mate-searching behavior (mobility between host fish) and promiscuity. Nevertheless, these caveats do not detract from the important result that females are limited in their opportunity to mate at low parasite intensity, and that this limitation could represent a critical phase in effective salmon louse transmission to other wild or captive farmed hosts.

The lower parasite abundance on small, wild juvenile hosts that we have described, compared to previously published reports for larger subadult or adult hosts, leads us to suggest that juvenile fish have the potential to act as a sink for the parasite population, whereas larger subadult or adult fish may well provide a parasite source. Such source-sink dynamics have been shown to impact the transmission of other parasite populations (Chaves et al. 2007, Fengyang and Poulin 2011) and have clear implications both for natural populations and for interactions between farmed and wild host species. Small juvenile salmonid hosts will be subject to a higher per capita mortality rate (from a multitude of sources; e.g., Lothian et al. 2017) compared to larger subadult or adult salmon. It is this high rate of mortality that will reduce the onward reproduction of the parasite, which means that the

parasites on these small juvenile hosts have only a limited time window within which to mate. Such density-independent mortality, together with biased sex ratios, can significantly lower the abilities of a population to survive due to the Allee effect (Schmickl and Karsai 2010). Related to this, it is interesting to note that although juvenile pink salmon can suffer mortality as a result of predation by other salmon species, sea lice have been recorded to escape predation on their host by trophic transmission (i.e., attaching to the predator species; Connors et al. 2011). By contrast, the subadult or adult hosts might act as sources in the transmission dynamics of the parasite. This is because they typically have greater infestations due to the accumulation of parasites over time and because their larger body size may be able to support more parasites. This difference may well be critical for parasite regulation, because populations can be regulated in a patchy environment only if the subdivided population exhibits source-sink dynamics (Pulliam 1988); that is, some patches (sources) can sustain exponential growth, whereas others (sinks) do not provide the necessary conditions for population growth.

Data assumptions

From these analyses, we were able to account for a number of factors that might influence observed parasite abundance, including time of year, sampling gear type, fish species, and fish length (Patanasatienkul et al. 2013). In relation to the efficacy of sampling, two factors that can result in loss of parasites are the capture method and exposure of sampled fish to brackish water (Todd et al. 2000). It is apparent, however, that capture method has a minimal effect on sex-biased retention of sea lice (Todd et al. 2000), and there is no evidence in the literature that loss of sea lice resulting from fish encountering reduced salinity would be sex biased.

Exclusion of hosts on which parasites had not been identified (in particular data from 2004) might influence results, and in particular, our calculation that 36% of female reproductive sea lice in 2004 were unable to mate because they were attached to a host without a male louse. Although the number of hosts that were excluded represented 17% of all fish sampled in that year, our data from other years also support our demonstration of the prevalence of mate limitation.

One additional possible factor that we could not assess in the present analysis was spatial clustering of sea louse infestations (Patanasatienkul et al. 2015). While assessment of parasite spatial distribution was beyond the scope of this analysis, it may be beneficial in future to investigate whether parasite aggregation varies according to location. The present data comprised a total of 150 sampling locations, but we were not able to disaggregate the data on a spatial level without compromising statistical power.

Implications

The present results have important implications in relation to our understanding of sea louse transmission for both wild and farmed salmonid hosts. These data, and specifically the identification of an Allee effect, will inform the structuring and interpretation of models describing these host-parasite associations. They also highlight the need to appraise the accuracy of parasite abundance estimations obtained during routine monitoring programs. Previous studies that have modeled sea louse population dynamics have often focused on female sea lice to the exclusion of male sea lice, or have assumed that the louse sex ratio is equal and that the genders are independently and randomly distributed among hosts (Revie et al. 2005, Groner et al. 2013). Our work shows how the sex ratio of sea lice on wild Pacific salmon could well influence sea louse reproduction. If the sex ratio is not taken into account, then it is possible to underestimate or overestimate the average parasite load (Dittmar et al. 2011) and thereby the reproductive potential of the population. Our work also shows how reproductive potential is influenced by the degree to which male and female sea lice are distributed together or separately among hosts. The distinction between the two cases is not trivial since, for the same values of m and k , there may be 75% pairing when distributed together compared to only 20% pairing when distributed separately (Bradley and May 1978). Introducing aggregation into models of parasite dynamics can mostly destroy the tipping point of transmission (Macdonald 1965) if the genders are distributed together. Thus, any model that fails to account for aggregation may not be robust (Bradley and May 1978).

While our analysis focused on wild fish, the findings should also be considered in a farm

setting, where data on sea louse sex distribution (before and after anti-parasite treatment) could aid predictions of parasite abundance and effective treatment (Robbins et al. 2010, Stormoen et al. 2013). Farms often apply louse treatments when the average intensity of female sea lice per fish exceeds a pre-defined threshold. Accordingly, additional information on the level of aggregation and the sex ratio of sea lice may be especially advantageous at critical times of the production cycle, particularly if counts could be used to estimate the likely tipping point of the louse population. Furthermore, if parasite treatment could be sex-specific then targeted control at a critical time may help to sustain mate limitation and thereby limit onward transmission. Reducing the number of female sea lice during the early stages of infestation may be more effective than interventions later in the fish stocking cycle, by which time males may have developed to sexual maturity and increased in prevalence. Stormoen et al. (2013) suggested that using cleaner wrasse for parasite control on farms may delay the release of parasite larvae early in the salmon production cycle because the wrasse tend to target adult female sea lice. This may be the reason that wrasse tend to be successful at controlling louse populations on Atlantic fish farms in the first year of production but are less successful later in the production cycle (Treasurer 2002).

In summary, we assessed two aspects of the parasite transmission dynamics that are rarely considered—the consequences of aggregation of parasite sexes between hosts and the numbers of pairs of reproducing sea lice. We emphasize that ignoring the details of sex distribution and the heterogeneity of reproduction in any host–parasite system could lead to errors in empirical sampling, in effective control measures and in predictive modeling.

ACKNOWLEDGMENTS

Research was undertaken thanks to funding from the SALMODIS project and the Canada Excellence Research Chairs Program. In particular, the CERC Aquatic Epidemiology Visiting Scientist initiative provided support for an extended visit by C.D.T. to work with colleagues at UPEI. We are also grateful for being given access to the relevant subsets of data collected in the Broughton Archipelago Monitoring Program

(BAMP) initiative (www.bamp.ca). Finally we thank two anonymous reviewers for their insightful and constructive comments.

LITERATURE CITED

- Anderson, R. M., and D. M. Gordon. 1982. Processes influencing the distribution of parasite numbers within host populations with special emphasis on parasite-induced host mortalities. *Parasitology* 85: 373–398.
- Bandilla, M., T. Hakalahti-Siren, and E. Teelervo Valtonen. 2008. Patterns of host switching in the fish ectoparasite *Argulus coregoni*. *Behaviour Ecology and Sociobiology* 62:975–982.
- Beamish, R., C. Neville, R. Sweeting, and N. Ambers. 2005. Sea lice on adult Pacific salmon in the coastal waters of Central British Columbia, Canada. *Fisheries Research* 76:198–208.
- Bottomley, C., V. Isham, and M. G. Basanez. 2005. Population biology of multispecies helminth infection: interspecific interactions and parasite distribution. *Parasitology* 131:417–433.
- Boxaspen, K. 2006. A review of the biology and genetics of sea lice. *ICES Journal of Marine Science* 63: 1304–1316.
- Bradley, D. J., and R. M. May. 1978. Consequences of helminth aggregation for the dynamics of schistosomiasis. *Transactions of the Royal Society of Tropical Medicine and Hygiene* 72:262–273.
- Bristow, G. A., T. Alvik, and H. Bohne. 1996. Some parasites of marine salmonids from Tanafjorden, Finnmark, Norway. *Bulletin of the Scandinavian Society for Parasitologists* 6:25–32.
- Bron, J. E., C. Sommerville, R. Wootten, and G. H. Rae. 1993. Influence of treatment with dichlorvos on the epidemiology of *Lepeophtheirus salmonis* (Kroyer, 1837) and *Caligus elongatus* Nordmann, 1832 on Scottish salmon farms. Pages 263–274 in G. A. Boxshall and D. DeFaye, editors. *Pathogens of wild and farmed fish: sea lice*. Taylor and Francis, London, UK.
- Butterworth, G. A., K. F. Cubbit, and R. S. McKinley. 2008. The prevalence, density and impact of *Lepeophtheirus salmonis* (Kroyer) infestation on juvenile pink salmon (*Oncorhynchus gorbuscha*) from the central coast of British Columbia, Canada. *Fisheries Research* 91:35–41.
- Chaves, L. F., M. J. Henandez, A. Dobson, and M. Pascual. 2007. Sources and sinks: revisiting the criteria for identifying reservoirs for American cutaneous leishmaniasis. *Trends in Parasitology* 23:311–316.
- Connors, B. M., N. B. Hargreaves, S. R. M. Jones, and L. M. Dill. 2010. Predation intensifies parasite exposure in a salmonid food chain. *Journal of Applied Ecology* 47:1365–1371.

- Connors, B. M., M. Krkošek, and L. M. Dill. 2008. Sea lice escape predation on their host. *Biology Letters* 4:455–457.
- Connors, B. M., C. Lagasse, and L. M. Dill. 2011. What's love for to do with it? Ontogenetic drivers of dispersal in a marine ectoparasite. *Behavioural Ecology* 22:588–593.
- Cornell, S. J., V. S. Isham, and B. T. Grenfell. 2004. Stochastic and spatial dynamics of nematode parasites in farmed ruminants. *Proceedings of the Royal Society B: Biological Sciences* 271:1243–1250.
- Costello, M. J. 2009. How sea lice from salmon farms may cause wild salmonid declines in Europe and North America and be a threat to fishes elsewhere. *Proceedings of the Royal Society B: Biological Sciences* 276:3385–3394.
- Dittmar, K., S. Morse, M. Gruwell, J. Mayberry, and E. DiBlasi. 2011. Spatial and temporal complexities of reproductive behavior and sex ratios: a case from parasitic insects. *PLoS ONE* 6:e19438.
- Fengyang, L., and R. Poulin. 2011. Effects of salinity on multiplication and transmission of an intertidal trematode parasite. *Marine Biology* 158:995–1003.
- Gottesfeld, A. S., B. Proctor, L. D. Rolston, and C. Carr-Harris. 2009. Sea lice, *Lepeophtheirus salmonis*, transfer between wild sympatric adult and juvenile salmon on the north coast of British Columbia, Canada. *Journal of Fish Diseases* 32:47–57.
- Grant, A. N. 2002. Medicines for sea lice. *Pest Management Science* 58:521–527.
- Groner, M. L., R. Cox, G. Gettinby, and C. W. Revie. 2013. Use of agent-based modelling to predict benefits of cleaner fish in controlling sea lice, *Lepeophtheirus salmonis*, infestations on farmed Atlantic salmon, *Salmo salar* L. *Journal of Fish Diseases* 36:195–208.
- Groner, M. L., G. Gettinby, M. Stormoen, C. W. Revie, and R. Cox. 2014. Modelling the impact of temperature-induced life history plasticity and mate limitation on the epidemic potential of a marine ectoparasite. *PLoS ONE* 9:e88465.
- Harrison, A., and N. C. Bennett. 2012. The importance of the aggregation of ticks on small mammal hosts for the establishment and persistence of tick-borne pathogens: an investigation using the R_0 model. *Parasitology* 139:1605–1613.
- Hull, M. Q., A. W. Pike, A. J. Mordue, and G. H. Rae. 1998. Patterns of pair formation and mating in an ectoparasitic caligid copepod *Lepeophtheirus salmonis* (Kroyer 1837): implications for its sensory and mating biology. *Philosophical Transactions of the Royal Society of London B: Biological Sciences* 353:753–764.
- Jacobsen, J. A., and E. Gaard. 1997. Open-ocean infestation by salmon lice (*Lepeophtheirus salmonis*): comparison of wild and escaped farmed Atlantic salmon (*Salmo salar* L.). *ICES Journal of Marine Science* 54:1113–1119.
- Jansen, P. A., A. B. Kristoffersen, H. Viljugrein, D. Jimenez, M. Aldrin, and A. Stien. 2012. Sea lice as a density-dependent constraint to salmonid farming. *Proceedings of the Royal Society B: Biological Sciences* 279:2330–2338.
- Johnson, S. C., and L. J. Albright. 1991. Development, growth, and survival of *Lepeophtheirus salmonis* (Copepoda: Caligidae) under laboratory conditions. *Journal of the Marine Biological Association of the United Kingdom* 71:425–436.
- Jones, S. R. M., and N. B. Hargreaves. 2007. The abundance and distribution of *Lepeophtheirus salmonis* (Copepoda: Caligidae) on pink (*Oncorhynchus gorbuscha*) and chum (*O. keta*) salmon in coastal British Columbia. *Journal of Parasitology* 93:1324–1331.
- Krkošek, M., B. M. Connors, M. A. Lewis, and R. Poulin. 2012. Allee effects may slow the spread of parasites in a coastal marine ecosystem. *American Naturalist* 179:401–412.
- Krkošek, M., A. Morton, and J. P. Volpe. 2005. Non-lethal assessment of juvenile pink and chum salmon for parasitic sea lice infections and fish health. *Transactions of the American Fisheries Society* 134:711–716.
- Krkošek, M., C. W. Revie, P. G. Gargan, O. T. Skilbrei, B. Finstad, and C. D. Todd. 2013. Impact of parasites on salmon recruitment in the Northeast Atlantic Ocean. *Proceedings of the Royal Society B: Biological Sciences* 280:2012–2359.
- Lothian, A. J., M. Newton, J. Barry, M. Walters, R. C. Miller, and C. E. Adams. 2017. Migration pathways, speed of migration and mortality of Atlantic salmon (*Salmo salar*) smolts in a Scottish river and the near-shore coastal environment. *Ecology of Freshwater Fish* 2017:1–10.
- Lysne, D. A., and A. Skorping. 2002. The parasite *Lernaeocera branchialis* on caged cod: Infection pattern is caused by differences in host susceptibility. *Parasitology* 124:69–76.
- Macdonald, G. 1965. The dynamics of helminth infections, with special reference to schistosomes. *Transactions of the Royal Society of Tropical Medicine and Hygiene* 59:489–506.
- Margolis, L., G. W. Esch, J. C. Holmes, A. M. Kuris, and G. A. Schad. 1982. The use of ecological terms in parasitology (report of an ad hoc committee of the American Society of Parasitologists). *Journal of Parasitology* 68:131–133.
- May, R. M. 1977. Togetherness among schistosomes: its effects on the dynamics of infection. *Mathematical Biosciences* 35:301–353.

- May, R. M., and M. E. Woolhouse. 1993. Biased sex ratios and parasite mating probabilities. *Parasitology* 107:287–295.
- Morand, S., J. P. Pointier, G. Borel, and A. Theron. 1993. Pairing probability of schistosomes related to their distribution among the host population. *Ecology* 74:2444–2449.
- Morand, S., R. Poulin, K. Rohde, and C. Hayward. 1999. Aggregation and species coexistence of ectoparasites of marine fishes. *International Journal for Parasitology* 29:663–672.
- Murray, A. G. 2002. Using observed load distributions with a simple model to analyse the epidemiology of sea lice (*Lepeophtheirus salmonis*) on sea trout (*Salmo trutta*). *Pest Management Science* 58:585–594.
- Parker, R. R., and L. Margolis. 1964. A new species of parasitic copepod, *Caligus clemensi* sp. novo (Caligoida, Caligidae) from pelagic fishes in the coastal waters of British Columbia. *Journal of the Fisheries Research Board of Canada* 21: 873–889.
- Patanasatienkul, T., J. Sanchez, E. E. Rees, M. Krkošek, S. R. Jones, and C. W. Revie. 2013. Sea lice infestations on juvenile chum and pink salmon in the Broughton Archipelago, Canada, from 2003 to 2012. *Diseases of Aquatic Organisms* 105: 149–161.
- Patanasatienkul, T., J. Sanchez, E. E. Rees, D. Pfeiffer, and C. W. Revie. 2015. Space-time cluster analysis of sea lice infestation (*Caligus clemensi* and *Lepeophtheirus salmonis*) on wild juvenile Pacific salmon in the Broughton Archipelago of Canada. *Preventive Veterinary Medicine* 120:219–231.
- Peacock, S. J., A. W. Bateman, M. Krkošek, B. Connors, S. Rogers, L. Portner, Z. Polk, C. Webb, and A. Morton. 2016. Sea-lice parasites on juvenile wild salmon in the Broughton Archipelago, British Columbia, Canada. *Ecology* 97:1887.
- Pert, C. C., et al. 2014. Using sentinel cages to estimate infestation pressure on salmonids from sea lice in Loch Shiel, Scotland. *Aquaculture Environment Interactions* 5:49–59.
- Poulin, R. 1997. Population abundance and sex ratio in dioecious helminth parasites. *Oecologia* 111:375–380.
- Poulin, R. 2008. Parasite aggregation: causes and consequences. Pages 134–159 in R. Poulin, editor. *Evolutionary ecology of parasites*. Princeton University Press, Princeton, New Jersey, USA.
- Pulliam, H. R. 1988. Sources, sinks and population regulation. *American Naturalist* 132:652–661.
- Reiczigel, J., L. Rozsa, A. Reiczigel, and I. Fabian. 2013. Quantitative parasitology (QPweb). <http://www2.univet.hu/qpweb>
- Revie, C. W. 2006. The Development of an Epi-informatics approach to increase understanding of the relationships between farmed fish and their parasites. Dissertation. University of Strathclyde, Strathclyde, UK.
- Revie, C. W., C. Robbins, G. Gettinby, L. Kelly, and J. W. Treasurer. 2005. A mathematical model of the growth of sea lice, *Lepeophtheirus salmonis*, populations on farmed Atlantic salmon, *Salmo salar* L., in Scotland and its use in the assessment of treatment strategies. *Journal of Fish Diseases* 28:603–613.
- Ritchie, G. 1997. The host transfer ability of *Lepeophtheirus salmonis* (Copepoda: Caligidae) from farmed Atlantic salmon, *Salmo salar* L. *Journal of Fish Diseases* 20:153–157.
- Ritchie, G., A. J. Mordue, A. W. Pike, and G. H. Rae. 1996. Observations on mating and reproductive behaviour of *Lepeophtheirus salmonis*, Kroyer (Copepoda: Caligidae). *Journal of Experimental Marine Biology and Ecology* 201:285–298.
- Robbins, C., G. Gettinby, F. Lees, M. Baillie, C. Wallace, and C. W. Revie. 2010. Assessing topical treatment interventions on Scottish salmon farms using a sea lice (*Lepeophtheirus salmonis*) population model. *Aquaculture* 306:191–197.
- Schmickl, T., and I. Karsai. 2010. The interplay of sex ratio, male success and density independent mortality affects population dynamics. *Ecological Modelling* 221:1089–1097.
- Shaw, D. J., and A. P. Dobson. 1995. Patterns of macro-parasite abundance and aggregation in wildlife populations: a quantitative review. *Parasitology* 111(Suppl):S111–S127.
- Shaw, D. J., B. T. Grenfell, and A. P. Dobson. 1998. Patterns of macroparasite aggregation in wildlife host populations. *Parasitology* 117:597–610.
- Stephenson, J. F. 2012. The chemical cues of male sea lice *Lepeophtheirus salmonis* encourage others to move between host Atlantic salmon *Salmo salar*. *Journal of Fish Biology* 81:1118–1123.
- Stien, A., P. A. Bjorn, P. A. Heuch, and D. A. Elston. 2005. Population dynamics of salmon lice *Lepeophtheirus salmonis* on Atlantic salmon and sea trout. *Marine Ecology Progress Series* 290:263–275.
- Stormoen, M., E. Skjerve, and A. Aunsmo. 2013. Modelling Salmon lice (*Lepeophtheirus salmonis*, Kroyer) reproduction on farmed Atlantic salmon (*Salmo salar* L.). *Journal of Fish Diseases* 36:25–33.
- Todd, C. D., R. J. Stevenson, H. Reinardy, and M. G. Ritchie. 2005. Polyandry in the ectoparasitic copepod *Lepeophtheirus salmonis* despite complex pre-copulatory and postcopulatory mate-guarding. *Marine Ecology Progress Series* 303:225–234.
- Todd, C. D., A. M. Walker, J. E. Hoyle, S. J. Northcott, A. F. Walker, and M. G. Ritchie. 2000. Infestations

- of wild adult Atlantic salmon (*Salmo salar* L.) by the ectoparasitic copepod sea louse *Lepeophtheirus salmonis* Kroyer: prevalence, intensity and the spatial distribution of males and females on the host fish. *Hydrobiologia* 429:181–196.
- Todd, C. D., B. D. M. Whyte, J. C. MacLean, and A. M. Walker. 2006. Ectoparasitic sea lice (*Lepeophtheirus salmonis* and *Caligus elongatus*) infestations of wild, adult, one sea-winter Atlantic salmon *Salmo salar* returning to Scotland. *Marine Ecology Progress Series* 328:183–193.
- Treasurer, J. W. 2002. A review of potential pathogens of sea lice and the application of cleaner fish in biological control. *Pest Management Science* 58:546–558.
- Trivers, R. L., and D. E. Willard. 1973. Natural selection of parental ability to vary the sex ratio of offspring. *Science* 179:90–92.
- Ugelvik, M. S., T. Mo, A. Mennerat, and A. Skorping. 2017. Atlantic salmon infected with salmon lice are more susceptible to new lice infections. *Journal of Fish Diseases* 40:311–317.
- Wilson, K., O. N. Bjornstad, A. P. Dobson, S. Merler, G. Pogliayen, S. E. Randolph, A. F. Read, and A. Skorping. 2001. Heterogeneities in macroparasite infections: patterns and processes. Pages 6–44 in P. J. Hudson, A. Rizzoli, B. T. Grenfell, H. Heesterbeek, and A. P. Dobson, editors. *The ecology of wildlife diseases*. Oxford University Press, Oxford, UK.

SUPPORTING INFORMATION

Additional Supporting Information may be found online at: <http://onlinelibrary.wiley.com/doi/10.1002/ecs2.2040/full>