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Mortality Rates for Juvenile Pink *Oncorhynchus gorbuscha* and Chum *O. keta* Salmon Infested with Sea Lice *Lepeophtheirus salmonis* in the Broughton Archipelago

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ABSTRACT: Wild juvenile pink *Oncorhynchus gorbuscha* and chum salmon *O. keta* were captured and sorted by the number of sea lice *Lepeophtheirus salmonis* infecting each fish. These fish were placed in groups of 60 in flow-through containers immersed in seawater near the site of capture. There were 3–4 replicates for each infection category and 3 consecutive trials or Series run to assess the impact of sea lice on short-term fish mortality. Control groups of lice-free fish were included to measure handling and containment effects. In each trial or Series, significantly more fish died in the categories with sea lice than in the lice-free category. The majority of fish infected with motile-stage sea lice died. These observations indicate that short-term mortality of wild juvenile pink and chum salmon is increased by infestations of 1–3 sea lice.

INTRODUCTION

Although the salmon-specific parasite, *Lepeophtheirus salmonis*, is common on adult Pacific salmon *Oncorhynchus* spp., it has only recently been reported on wild juvenile pink *O. gorbuscha* and chum salmon *O. keta* (Morton et al. 2004; Krkosek et al. 2005). Infestation of *L. salmonis* on juvenile salmon appears causally related to salmon farms (Heuch et al. 2004; Morton et al. 2004; Morton et al. 2005; Krkosek et al. 2005). Therefore, an expectation exists that management of sea lice on farmed salmon could reverse specific wild salmon population declines, as per the Norwegian Action Plan Against Lice on Salmonids (Eithun 2000; Heuch et al. 2004) and the British Columbia Sea Lice Action Plan (Ministry of Agriculture, Food, and Fisheries 2003). Essential to the efficacy of this strategy is answering the question: How many sea lice can young Pacific salmon tolerate? This is a preliminary study to address this uncertainty.

Lepeophtheirus salmonis settle on salmon as copepodids, then molt, attach, and pass through 4 chalimus stages, followed by further motile stages (Johnson and Albright 1991). In laboratory-infected Atlantic salmon *Salmo salar* postsmolts, onset of mortality occurs at 0.75 motile lice \times g host mass⁻¹ (Bjørn and Finstad 1997; Finstad et al. 2000). The less mature, chalimus-stage lice can cause distinctive black scarring centered where the chalimus sea louse's filament attaches to the fish host (Grimnes and Jakobsen 1996).

The 2 species of sea lice commonly found on salmonids in British Columbia can be reliably distinguished (Galbraith 2005). However, due to constraints in the field when counting sea lice on living fish, i.e. no ability to stain, lesser magnification and limited time, we assume the potential for misidentification at the chalimus III and IV stage exists when live-sampling fish.

METHODS

Three trials were conducted, identified as Series I, II and III. Fish were caught in a 30 m \times 1.8 m beach seine with 1.6-cm mesh. There were at least 2 sets per location. Collections were made in Tribune Channel of the Broughton Archipelago—an area known for presence of sea lice on juvenile salmon (Morton et al. 2004; Krkosek 2005). The capture sites were situated between 2.5 and 5 km away from the study site.

Each fish was identified to species and measured (fork-length), and the sea lice were counted and identified by stage and species as per Krkosek et al. (2005). The maximum sea lice intensity used in this study was 10 chalimus-stage lice, and so examination of both infested and lice-free fish required an average of 30 s.

The fish were sorted into flow-through barrels (Figure 1) based on their number of sea lice. Each barrel received 60 fish. Where the species were held separately, the chum salmon barrels were 189 L and

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the pink salmon barrels were 132 L. We ran 3 consecutive trials beginning 14 March 2004 labeled Series I, II and III. Pink salmon were held for 35 d, chum salmon, for 47 d, except where the 2 species were mixed within barrels (Series I). This group was released after 31 d.

Each Series consisted of replicate statistical blocks, consisting of 3–4 barrels of increasing infestation levels. Each incrementally distinct infestation level was defined as a “category” 0–3 with category “0” being lice-free. All barrels within each replicate set were identical in size and orientation and were randomly sequenced (Figure 1). The barrels were grouped into blocks to control for potential differences due to location within the containment facility. For logistical reasons, some blocks could not contain barrels in each infestation category. Hence, we used an incomplete block design (Freeman 1983). Because the fish were sourced from the natural environment, the sea lice available for each Series were increasingly mature. The stocking criteria for each barrel are given in Tables 1 and 2.

The fish were fed commercial pellets to satiation every 2–4 hours during daylight. Wild food also entered through the mesh at the barrel ends. Barrels were scrubbed and gently sluiced daily to prevent fouling from restricting the water flow. Wild fry schooled outside the barrels. Water temperature and salinity were recorded (Table 1). Infestation from sea lice larvae adrift in the surrounding waters was possible.

Mortalities were removed frequently, measured, and weighed, and sea lice and scarring were recorded. At the end dates, surviving fish were measured, weighed, inspected for sea lice and released. There

were some fish escapes particularly in Series I due to occasional extreme wave action.

The fish were observed regularly during the husbandry routine and timed observation periods for potential effects of sea lice on behavior and appearance.

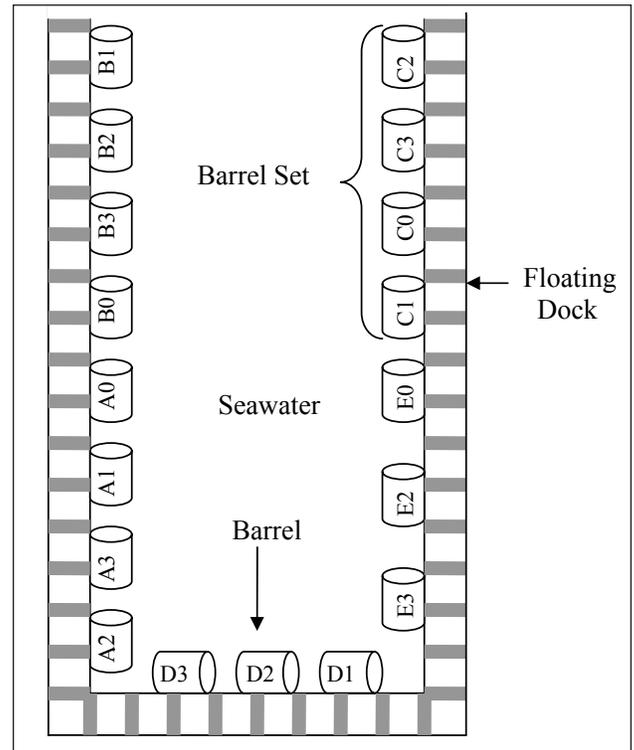


Figure 1. The fish containment (barrels) arrangement using Series II as an example.

Table 1. The date each Series was started (barrels loaded with sorted fish), with the number and stage of sea lice per category (infestation level). Cope=copepodid, chal. I/II=chalmus stage I and/or II. Mean salinity and temperature presented per Series.

Series	Date loaded	Category 0	Category 1	Category 2	Category 3	Salt (SD)	Temp°C (SD)
I	14–25 March	0 lice-free	1 cope/chal I	2 cope/chal. I	n/a	30‰ (2.1)	9.2 (0.9)
II	17–23 April	0 lice-free	1–3 chal I/II	4 chal. I/II	5 chal. I/II	29‰ (1.9)	12.2 (0.1)
III	18–21 May	0 lice-free	1–5 chal. I/II or 1 motile	6–9 chal. I/II or 2 motiles	10 chal. I/II or 3 motiles	28‰ (3.5)	16.7 (2.6)

Table 2. The number of replicate barrel sets per Series and the percentage of species of fish in each barrel. Categories were based on number of sea lice per fish with 0 being lice-free and 3 being the most infested (Table 1). Each Series is a trial of 31–47 days.

Barrel replicate sets	Series I categories and species	Series II categories and species	Series III categories and species
A	0, 1, 2 50% pink/chum	0, 1, 2, 3 100% chum	
B	0, 1, 2 50% pink/chum	0, 1, 2, 3 100% chum	
C	0, 1, 2 50% pink/chum	0, 1, 2, 3 100% pink	0, 1, 2, 3 100% pink
D	0, 1, 2 100% pink	0, 1, 2 100% pink	0, 1, 2 100% pink
E	0, 1, 2 100% pink	0, 2, 3 100% pink	0, 2, 3 100% pink
F	0, 1, 2 100% pink		

STATISTICS

Differences in mortality rates between infestation categories were tested for significance within each series by logistic regression using the R function, “glm” with infestation category specified as a nominal factor as opposed to a continuous variable. This analysis can be viewed as an extension of the standard χ^2 -test for equality of several proportions—the extension incorporating both the blocking by barrel sets and extra-binomial variation to account for potential within-barrel dependencies (Collett 1991). Without this allowance for extra-binomial variation, the fate of different fish within the same barrel would have to have been viewed as independent. The logistic regression model required independence only between separate barrel sets. (In Series I, with a total of only 6 chum salmon deaths, this test was replaced with an exact conditional test producing a mid-*p*-value.) This test assessed the potential for a mortality effect at any infestation level. To test specifically for a potential mortality effect at the lowest infestation level, a single, pre-planned test was made (for each series and species) of the null hypothesis that there was no increase in mortality between barrels with lice-free fish (category 0) and the lowest infestation level (category 1).

Fish were assigned to treatments (categories 0–3) according to the number of lice that were attached to them. This had the advantage of natural age distribution among the sea lice and the disadvantage that the fish were not assigned to treatments at random. We cannot exclude the possibility that there was some systematic difference between fish that had more lice and fish that had fewer, but the fish appeared healthy in behavior and appearance while the lice were still small.

RESULTS

In all 3 Series, the death rate for sea-lice infected fish was higher than for uninfected fish. Few fish survived with motile sea lice attached (Table 3) and sea lice loads had a significant effect on mortality cases (Table 4).

In Series I, sea lice loads had a significant effect on pink salmon mortality despite the fact that only copepodid sea lice, not yet permanently affixed, were present on fish entering this trial and many lice were shed (Table 3). The death rate for pink salmon increased in this trial with sea-lice infection rates (Figure 2a), and 75% of the mortalities had motile-stage sea lice vs. only 1.1% of the survivors.

The increase in chum salmon mortality through infection categories in Series I was not significant (Table 4). Of the 6 that died, 3 were infested with motile lice, and the other 3 with advanced chalimus stage III/IV lice. Only 4.9% of the surviving chum salmon were infested with motile lice, none with more than one. Throughout this project 5.6% of the fish escaped; most were lost during Series I.

In Series II, sea lice loads had a clearly significant effect on mortality for both species, though impact differed significantly between species ($p < 0.001$). The size difference between pink and chum salmon was not significant ($p > 0.5$). The death rate for pink and chum salmon climbed with each increasing infestation category (Figure 2b). In this Series, 97.8% of the pink salmon infected with a motile sea louse died. The 4 survivors with a motile louse had only one. Among the chum salmon, 88.4% with motile sea lice died. Both species also died with only chalimus-stage lice. Among fish with no surviving motile lice, but at least

Table 3. Sea lice retention and prevalence among dead and surviving fish for each Series trial and species.

Series	Species	% Lice Retained	Dead All infestation levels			Survived All infestation levels		
			Total	With Motile Lice	Percent	Total	With Motile Lice	Percent
I	Pink	24%	64	48	75%	726	8	1%
	Chum	23%	6	3	50%	203	10	5%
II	Pink	32%	262	107	41%	351	14	4%
	Chum	49%	282	176	62%	199	4	2%
III	Pink	16%	354	62	18%	226	0	0%

Table 4. *P*-values for increase in mortality between the louse-free category “0” and least the infested category “1,” for all Series.

Series	Pink	Chum
I	0.034	not significant
II	< 0.002	< 0.002
III	0.020	na

one chalimus III/IV stage louse, 94.1% of pink salmon died and 96.0% of chum salmon died.

In Series III (pink salmon only), mortality rate again climbed with each increasing infestation category (Figure 2c). Lice retention and salinity were lowest

in this Series. No fish survived with any remaining lice or even with lice scarring.

The number of fish dying per day varied between the series (Figure 3). Chum salmon daily mortality consistently peaked later than pink salmon in both trials where the 2 species were observed.

The presence of wild juvenile pink and chum salmon outside the barrels suggests water quality at the study site was suitable for salmon survivorship. Of 2,044 sea lice recorded on fish exiting the barrels, only 50 were of the most juvenile life stages (copepodid, chalimus I/II) and therefore recruited while the fish were in the barrels.

Infested pink salmon behavior changed predictably just prior to death. While louse-infested fish generally fed actively, in fact voraciously, they stopped feeding 24–36 hours prior to death, abandoned the school and became “loners.” Loners exhibited darkened skin and sculled slowly at the surface, nonresponsive to movement above them. At the time of death, sea-lice infested fish appeared emaciated (Figure 4). “Loner” behavior was rare among the chum salmon.

Salmon were occasionally observed picking adult sea lice off schoolmates. On several occasions fish were observed biting and pressing recipient fish downwards. Gravid sea lice were the primary targets of these “attacks” and damage to the lice often resulted. These sea lice exhibited patchy gray areas, and their eggstrings became crooked. “Attacks” also altered orientation of sea lice.

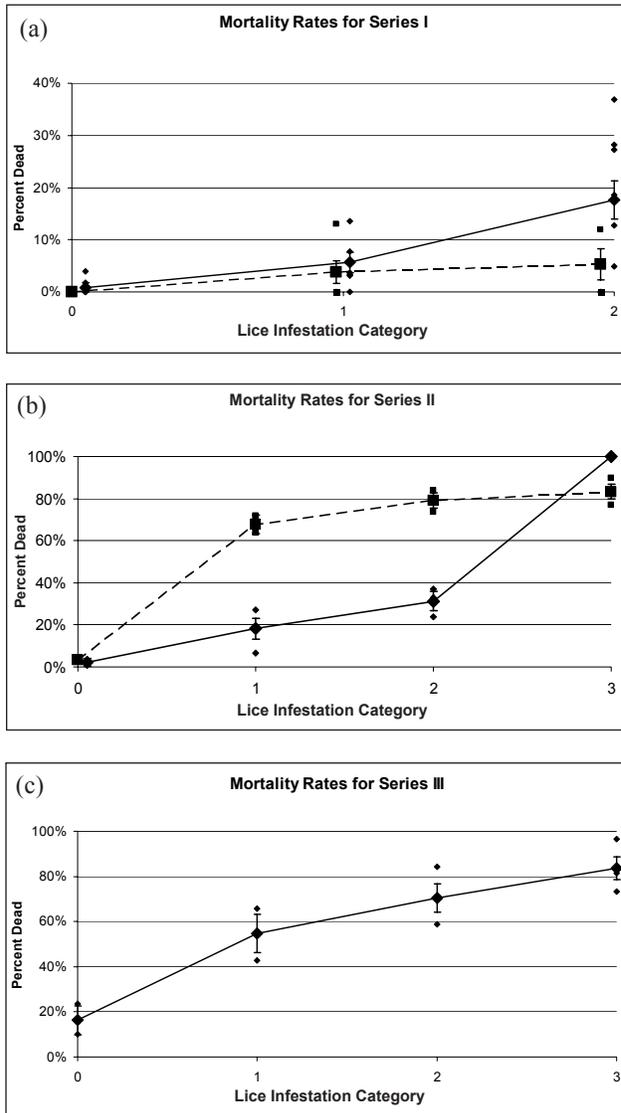


Figure 2. Death rates by infestation category (not individual barrels) for (a) Series I, (b) Series II, and (c) Series III. Overall category proportions are represented by larger symbols joined by lines: (—◆—) for pink salmon, (---■---) for chum salmon. Proportions within individual barrels are designated by smaller symbols. Infestation categories (offset marginally for some categories to reduce clutter) were determined according to abundances of chalimus stage I/II sea lice on fish as the barrels were stocked, and are described in Table 1. No lice at that time were beyond these stages. Standard error bars are for the overall category proportions with extra-binomial variation estimated from the logistic regression analysis.

DISCUSSION

The salmon infested with sea lice died at a higher rate than those without sea lice in all 3 trials. While handling and containment likely had some affect on fish and sea lice survivorship, fish survival was clearly related to sea lice abundance. There is statistically significant evidence from Series I of an impact on short-term pink salmon mortality even from a single copepodid or early chalimus-stage louse.

Nonetheless, this was not a controlled experiment. The purpose was merely to hold fish at a variety of sea lice infestation levels to observe, in as natural an environment as possible, their short-term mortality. Experiments with controlled infestation levels would be an important complement to this observational study. In addition to temperature and salinity, we also could not control recruitment of new sea lice on fish in the barrels during this study. However, the low number of juvenile sea lice at termination of each series suggested that few lice were acquired while the fish were in the barrels.

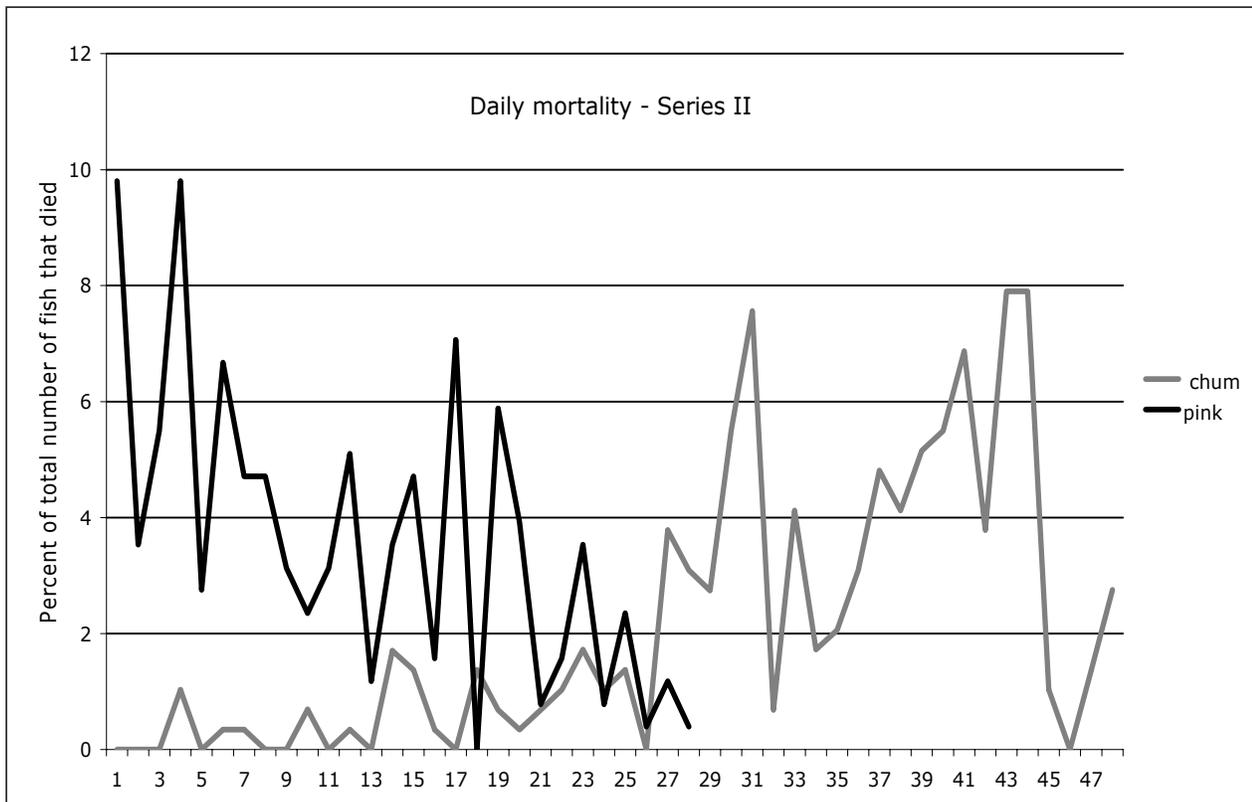
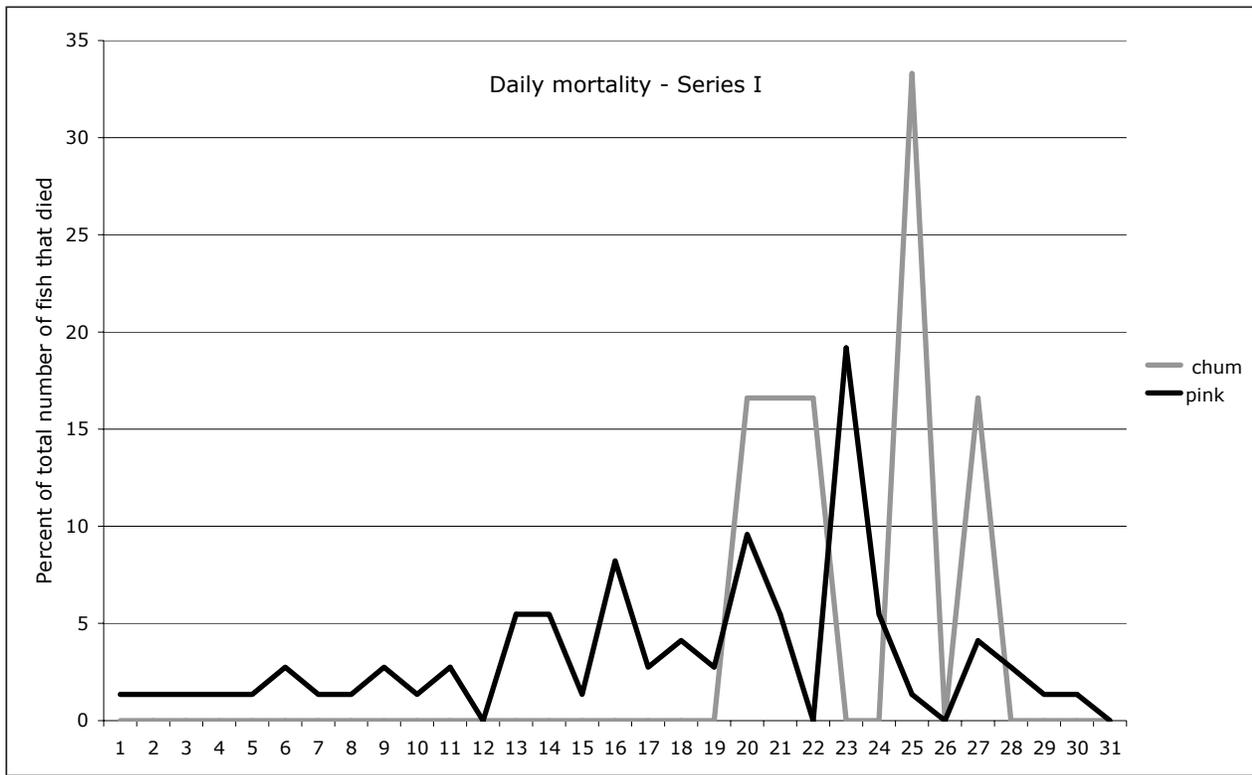


Figure 3. The daily percentage of total number of fish that died in each Series I, II, III. Note differences in scale between Y-axis of each Series.

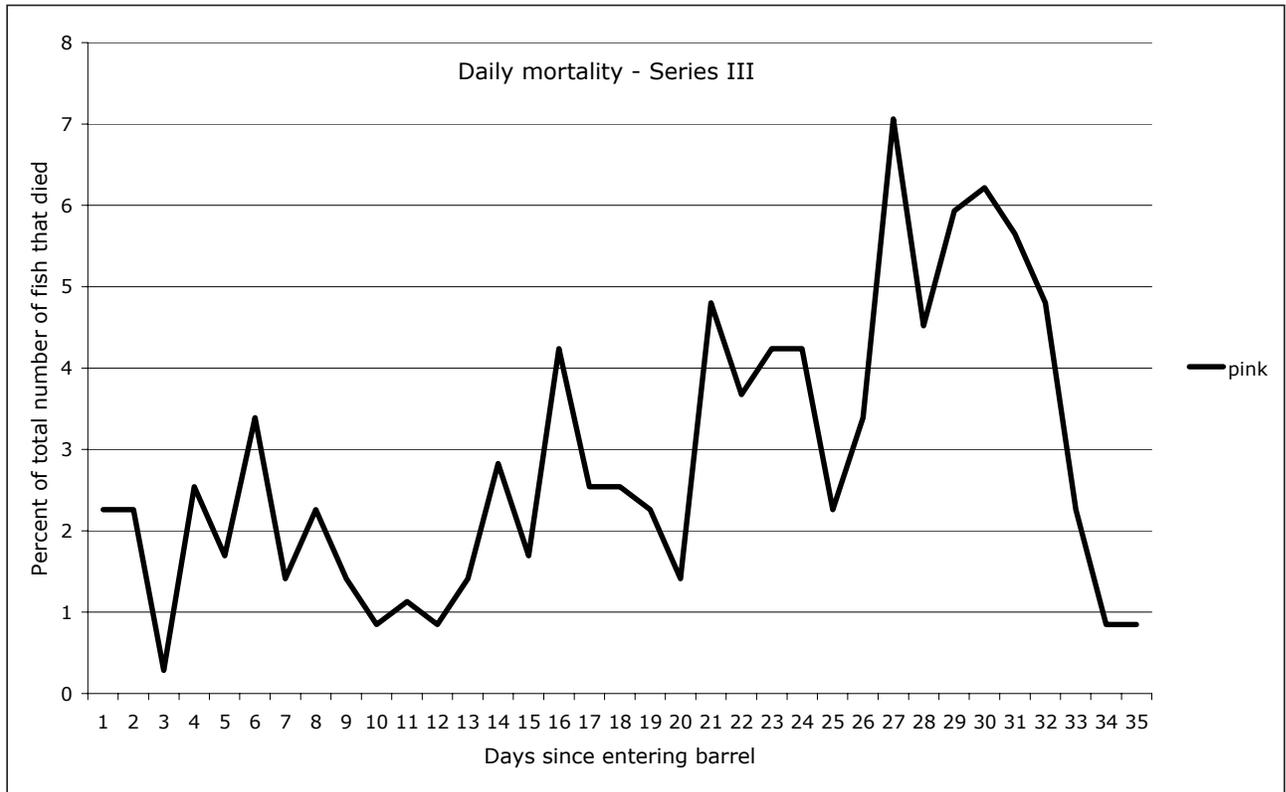


Figure 3. Continued.

Many sea lice disappeared. The retention rate varied from 49% to 16%. Low lice retention in Series III was possibly attributable to low salinity. By contrast, Grimnes and Jakobsen (1996) report that 53% of *L. salmonis* reach maturity under laboratory conditions. If retention rates in this study were less than in the wild, then the actual impact of sea lice on short-term mortality would be higher than our observations would suggest.

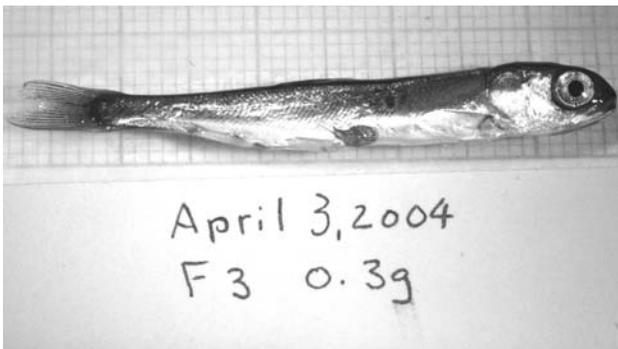


Figure 4. Photograph of a typical juvenile pink salmon at removal from a barrel after death with sea lice.

Of particular interest is lack of fish survival immediately following the development of lice to the motile stage—a period of intense mortality for Atlantic salmon (Grimnes and Jakobsen 1996). While this study was not designed to track events for individual fish, the relatively high prevalence of motile lice on the dead fish points to the same conclusion. Furthermore, we observed that sea lice carapaces appear translucent immediately following a molt and darkened over time. From such observations, it appears that, while pink salmon often died at the onset of hosting motile stage sea lice, chum salmon survived motile-stage sea lice for a week or more, despite being of equivalent size. This agrees with the differences observed in daily mortality between species.

We cannot explain why so many chalimus-stage sea lice were present at the end of each series when sea lice growth rates determined in the Atlantic suggest that more of these parasites should have progressed past the chalimus phase. Our observations may have been due to reduced light availability in the barrels slowing lice development, subtle differences between Atlantic and Pacific sea lice, the suboptimal nature of small salmon fry as hosts compared to adult salmon

and larger smolts, or a cause not identified. This certainly warrants further research.

Despite further research needs, the mortality impact of sea lice on the remarkably small juvenile pink and chum salmon would appear acute, with a single, copepodid louse having significant impact on short-term mortality. Thus, this contribution to the available evidence of sea lice effect on wild juvenile Pacific salmon is consistent with studies on Atlantic salmonids. However, this work firmly suggests pink and chum salmon will die from much lower infestation rates than reported for Atlantic salmon and sea

trout (anadromous brown trout, *Salmo trutta*). As a result, while Europeans may reduce impacts on wild salmonids adequately by treating farm salmon with chemicals to reduce sea lice loads, this management strategy may not be sufficient to promote survivorship of juvenile pink and chum salmon near salmon farms. The particular vulnerability of the diminutive pink and chum salmon fry indicates that management restrictions aimed at protecting these species will have to be particularly stringent. It may not be possible to site salmon farms in areas where Pacific salmon weigh less than several grams.

LITERATURE CITED

- Bjørn, P. A. and B. Finstad 1997. The physiological effects of salmon lice infection on sea trout post smolts. *Nordic Journal of Freshwater Research* 73:60–72.
- Collett, D. 1991. *Modeling Binary Data*. Chapman and Hall, London
- Eithun I. 2000. Measures to control sea lice in Norwegian fish farms. *Caligus* 6:4–5 <<http://ecoserve.ie/projects/sealice>> Accessed December 2, 2005.
- Finstad, B., P. A. Bjørn, A. Grimnes and N. A. Hvidsten. 2000. Laboratory and field investigations of salmon lice (*Lepeophtheirus salmonis* Krøyer) infestation on Atlantic salmon (*Salmo salar* L.) postsmolts. *Aquaculture Research* 31:1–9.
- Freeman, G. H. 1983. Incomplete block designs. Pages 40–46 in S. Kotz, N. L. Johnson, and C. B. Read, editors. *The Encyclopedia of Statistical Sciences*, Volume 4.
- Galbraith, M. 2005. Identification of larval stages of *Caligus clemensi* and *Lepeophtheirus salmonis* from the Broughton Archipelago. Technical report of Fisheries and Aquatic Sciences 2548.
- Grimnes, A. and P. J. Jakobsen. 1966. The physiological effects of salmon lice infection on post-smolt of Atlantic salmon. *Journal of Fish Biology* 48:1179–1194.
- Heuch, P. A., P. A. Bjørn, B. Finstad and J. C. Holst. 2004. A review of The Norwegian Plan Against Salmon Lice on Salmonids: the effect on wild salmonids. *Aquaculture* 246: 79–92.
- Johnson, S. C., and L. J. Albright. 1991. The development stages of *Lepeophtheirus salmonis* (Krøyer, 1837) (Copepoda: Caligidae). *Canadian Journal of Zoology* 69:929–950.
- Krkosek, M., M. A. Lewis and J. P. Volpe. 2005. Transmission dynamics of parasitic sea lice from farm to wild salmon. *Proceedings of the Royal Society, Series B*. 272(1564): 689–696.
- Krkosek, 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(3):711–716.
- MAFF (Ministry of Agriculture, Food and Fisheries). 2003. <http://www2.news.gov.bc.ca/nrm_news_releases/2003AGF0004-000137-Attachment1.htm> Accessed December 13, 2005.
- Morton, A., R. Routledge, C. Peet, and A. Ladwig. 2004. Sea lice *Lepeophtheirus salmonis* infection rates on juvenile pink *Oncorhynchus gorbuscha* and chum salmon *Oncorhynchus keta* in the nearshore marine environment of British Columbia, Canada. *Canadian Journal of Fisheries and Aquatic Science* 61:147–157.
- Morton, A.B., R. Routledge and R. Williams. 2005. Temporal patterns of sea lice infestation on wild Pacific salmon in relation to the fallowing of Atlantic salmon farms. *North American Journal of Fisheries Management* 25:811–821.
- Todd, C. D., A. M. Walker, M. G. Ritchie, J. A. Graves and A. F. Walker. 2004. Population genetic differentiation of sea lice (*Lepeophtheirus salmonis*) parasitic on Atlantic and Pacific salmonids: analysis of microsatellite DNA variation among wild and farm hosts. *Canadian Journal of Fisheries and Aquatic Science* 61:1176–1190.

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