



Succinctus

Snorkel sea-cage technology decreases salmon louse infestation by 75% in a full-cycle commercial test



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ABSTRACT

Methods to prevent parasite infestations in farmed fish are becoming widespread, yet tests of their effectiveness often lack commercial relevance and statistical power, which may lead to technology misuse. Here, we examined salmon louse infestation on Atlantic salmon in triplicate commercial snorkel louse barrier and standard cages over a 12 month production cycle. Barrier cages reduced newly settling lice on Atlantic salmon by 75%, with variability in parasite reduction over time depending upon environmental variables. The commercial, triplicate, long-term study design serves as a template to validate performance and detect weaknesses in anti-parasite techniques in fish mariculture.

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Intensive animal farming systems are susceptible to parasite outbreaks. However, understanding host–parasite interactions creates opportunities to prevent parasite encounters and infestations in these systems (Bui et al., 2019). In fish mariculture, novel sea-cage designs or host behaviour manipulations that mismatch host and parasite environments have been developed (Wright et al., 2017; Stien et al., 2018) in attempts to overcome the normal free-flow of rapidly spreading marine parasites onto fish stocked in open enclosures (McCallum et al., 2003). These preventive methods appear fruitful against the salmon louse, *Lepeophtheirus salmonis*, the primary parasite causing issues for the world's largest finfish mariculture industry, sea-cage Atlantic salmon, *Salmo salar*, farming. In 2015, Norway produced NOK 49 billion of farmed salmon, but spent > NOK 5 billion to control the parasite (Brooker et al., 2018). Adding a layer of complexity, wild salmonids dying at unacceptable rates from farm-magnified salmon louse populations (Kristoffersen et al., 2018) have triggered the Norwegian government to enforce production volume limits and treatments when

salmon louse infestation levels are too high in salmon farms (Lovdata, 2012, 2017).

For management of salmon louse infestation, prophylactic depth-based technologies are emerging (Bui et al., 2019). These include barrier cages (a skirt or snorkel tarpaulin wrapped around the upper depths), submerged cages (repeatedly submerged or submerged with an air dome), semi-enclosed cages (with deep water pumped in), and deep lighting and feeding (motivating salmon to swim deeper). They work by uncoupling salmon from surface-dwelling salmon louse larvae but provide surface air access required for salmon swim bladder re-inflation, buoyancy control and optimal welfare. Several trials and case studies report prophylactic depth-based technologies reduce salmon louse infestation levels, however their short-term, research-scale, or sub-optimally replicated nature increases uncertainty surrounding the results (Table 1). Short-term studies will not capture how seasonal variations in louse larvae development and dispersal (Samsing et al., 2016, 2017) and environmental factors that influence host or parasite depths (Heuch et al., 1995; Stien et al., 2016) affect depth-based technologies over full production cycles. In addition, research-scale studies could suffer from scale-dependent differences such as fish numbers and cage volumes that mean their results are not directly transferable to the salmon farming industry

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Table 1

The scale, replication and seasonal coverage of studies assessing salmon louse infestation in preventive depth-based cage designs versus standard cages.

Depth-based preventive cage	Study	Commercial scale	≥3 replicates	Seasons covered			
				Autumn	Winter	Spring	Summer
Snorkel	Stien et al. (2016)		x	x			
	Oppedal et al. (2017)		^a		x		
	Wright et al. (2017)	x				x	x
	Wright et al. (2018)		x	x			
Skirt	This study	x	x	x	x	x	x
	Stien et al. (2018)	x	x				x
	Grøntvedt et al. (2018)	x	x	^d	^d	^d	^d
Floating enclosed	Nilsen et al. (2017)	x	^b	^c	^c	^c	^c
Deep light	Hevrøy et al. (2003)				x	x	x
Deep feed and light	Frenzl et al. (2014)	x				x	x
Submerged	Korsøen et al. (2009)		x		x		
	Sievers et al. (2018)		x			x	
	Glaropoulos et al. (2019)		x		x		

The current study is indicated in bold.

^a Regression design.^b Different sites used, with different louse infestation pressures.^c Cages were stocked over inconsistent periods using different fish cohorts with variable louse infestation dynamics.^d Seasons were not known, but farm sites were tracked for 2–5 months.

(Wright et al., 2017). To have sufficient statistical power, experiments should also use at least three replicate cages to account for expected random environmental variation (Ling and Cotter, 2003), with all cages at the same farm site so they experience similar louse infestation pressures. In a best practice experiment, we compared salmon louse infestation between three commercial-scale snorkel and three standard cages at a single site over 12 months (Fig. 1). Environmental conditions were monitored to assess the influence of periodic brackish water and high surface temperatures, respectively, expected to push lice and fish in standard cages deeper, on snorkel technology effectiveness.

The study was conducted at a commercial salmon sea-cage farm at Låva, Jelsafjorden, Finnøy commune, Norway (59.1° N, 5.6° E). Data were collected through most of a production cycle from sea transfer to harvest, from June 2016 to August 2017. Atlantic salmon (*S. salar*, autumn transferred smolts, Salmobreed strain in four cages and Mowi strain in two cages; the strains were split evenly between cage types) were stocked in triplicate standard and snorkel cages (Fig. 1). The snorkels of 10 m depth were deployed before fish arrival. Two snorkel and two standard cages were stocked between 11–14 June, while one snorkel and one standard cage were stocked on 22 September. At transfer, the number of fish per cage ranged between 147,149–159,775 with an average weight of 82–155 g. Co-stocked cleaner fish were in equal numbers between cage types.

At fortnightly sampling events, we randomly netted and lethally dosed (Benzoak vet., Benzocaine, 200 mg/ml, VESO Vikan,

Namsos, Norway) 20 fish per cage, and counted their sessile salmon lice stages (copepodid, chalimus I, chalimus II) while submerged in seawater-filled trays. Sessile lice stages were used to represent new lice encounters when determining the effect of snorkel technology on louse infestation because: (i) they were expected to develop within the fortnightly sampling interval and (ii) were less likely to be influenced by de-lousing measures and cleaner fish compared with mobile stages. We monitored water salinity and temperature between 0–20 m depth daily by profiling a Conductivity, Temperature and Depth (CTD) recorder (SD208, SAIV-AS, Bergen, Norway) at the feed barge. When louse infestations at the farm exceeded the maximum allowed limit of 0.5 adult female lice per fish or 0.2 adult females during weeks 16–21 (Lovdata, 2012), cages over the limit were deloused with hydrogen peroxide or thermolicer treatments. Delousing events that occurred before a sampling event could have reduced the sessile lice numbers recorded to some extent, but as the standard cages were deloused more often, our results on louse reduction in snorkel compared with standard cages are conservative.

Data analyses were performed using R software v.3.1.0 (Copyright 2009, The R Foundation for Statistical Computing, Vienna, Austria). We compared square-root-transformed newly attached lice (copepodid to chalimus II) counts between cage types using linear mixed-effect models, setting cage type as a fixed factor and sampling time as a random effect. Square-root transformed newly attached lice numbers were also compared between cage types at individual times via a Welch's *t*-test. Correlations between

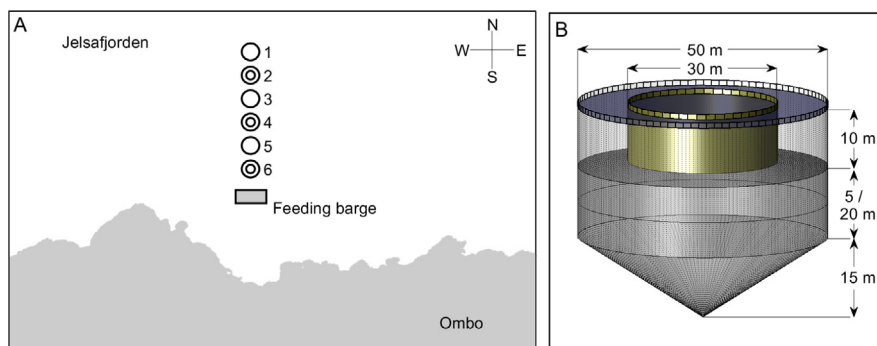


Fig. 1. Schematic of commercial farm used in the study; (A) Låva fish farm, Norway and (B) commercial snorkel sea-cage. The fish farm had six circular cages on a line from the feeding barge and perpendicular to the shoreline. The rectangle represents the feeding barge, the circles represent standard cages and the double circles represent snorkel cages. All cages were 50 m in diameter and 30–50 m deep, while three cages were also fitted with a 30 m diameter and 10 m deep snorkel.

the significance of cage type effects based on P values from t -tests at individual times and the corresponding magnitude of salinity (depth of 28 ppt contour) or temperature stratification (depth of 16 °C contour) in the preceding fortnight were assessed using Pearson's product-moment correlation tests. Error distributions were checked for variance and normality and the significance level was set at $P < 0.05$.

Research data for this article are available in Mendeley Data, DOI: <https://doi.org/10.17632/3jn84ngx9t.1>.

Overall, throughout the study period newly attached lice were on average 75% lower in snorkel relative to standard cages (mean of 0.17 ± 0.03 versus 0.71 ± 0.07 ; $\chi^2 = 104,18$, $P < 0.001$). When compared at individual times, counts of new infestations were significantly lower in snorkel than in standard cages, 15 of 28 times when 50–100% less lice were observed (Fig. 2). The significance of snorkel effects on newly attached lice was negatively correlated with the intensity of surface brackish water ($t = -2.52$, $P = 0.018$) and surface warm water events ($t = -3.38$, $P = 0.002$) (Fig. 2). Louse bath treatments were reduced by a factor of almost 2 in the three snorkel cages (treated zero, two and two times) in relation to the three standard cages (treated zero, four and three times).

In this study we demonstrate the effectiveness of spatially separating Atlantic salmon from infective salmon louse larvae using depth-based technologies in commercial-scale sea-cages. Over 12 months, approximating a full seawater phase production cycle, installing a 10 m deep snorkel in sea-cages reduced louse infestation by a factor of 4 and louse bath treatments by a factor of almost 2, relative to standard cages (Fig. 2). The reductions are consistent with previous snorkel cage studies at commercial- and research-scales (Stien et al., 2016; Oppedal et al., 2017; Wright et al., 2017). The salmon louse develops through both free-swimming and host-attached stages, and initial host infection involves the infective free-swimming copepodid stage. Infective copepodids vertically migrate into surface waters using positive phototaxis and possibly geotaxis (Bron et al., 1993), using average swimming speeds of 1.55 mm s^{-1} (Heuch et al., 1995). Sea-caged Atlantic salmon typically spend extensive periods in surface waters due to a combination of abiotic and biotic factors and sea-cage structures (Oppedal et al., 2011) which typically expose them to infective lice, and likely explains the success of depth-based prophylactic strategies.

However, depth-based technology effects were weakest when surface brackish water (salinities < 28 ppt) and warm surface

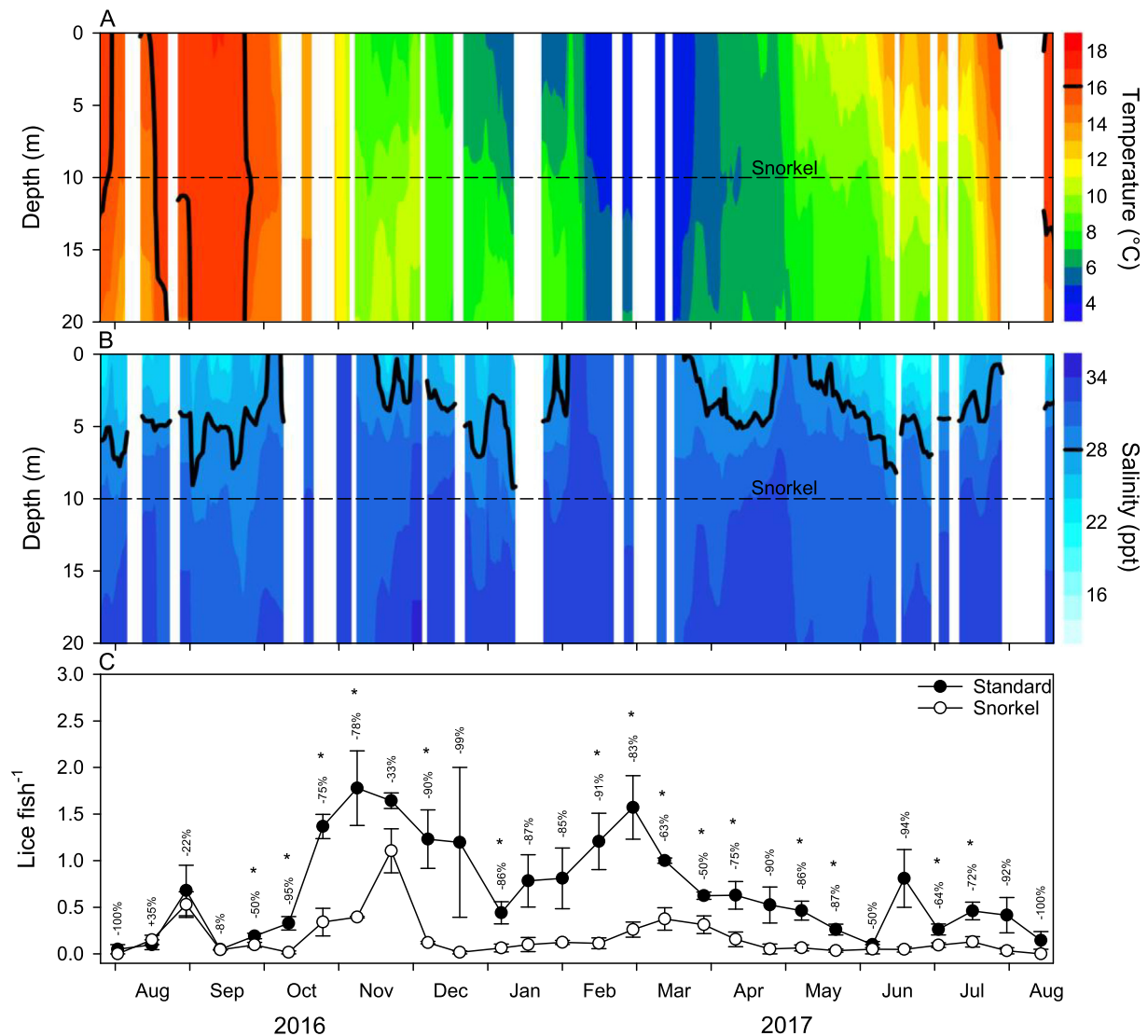


Fig. 2. Daily depth profiles between 0–20 m of (A) temperature (with a black line tracing 16 °C levels) and (B) salinity (with a black line tracing 28 ppt levels) from a reference location at the feed barge at Låva fish farm, Norway. The dashed black line indicates snorkel depth (10 m). Also shown is the mean number (\pm S.E.) of newly attached lice fish⁻¹ (copepodite, chalimus I and chalimus II) per cage type (snorkel and standard cage) for each sampling point (C). The percentage differences between cage types are displayed above each sampling time and significance is indicated with an asterisk when $P < 0.05$.

waters (temperatures >16 °C) occurred. Others have also reported that snorkel sea-cages can make little to no difference in louse infestation in the presence of a strong vertical salinity gradient (Oppedal et al., unpublished data) and at times when fish in standard cages swim deeper and thus both control and snorkel cage fish avoid lice equally (Stien et al., 2016). Infective copepodids have reduced survival at <29 ppt and tend to sink out of low salinity layers to aggregate at haloclines (Heuch et al., 1995; Crosbie et al., 2019), threatening encounters with snorkel fish when these layers penetrate deep enough. Atlantic salmon prefer depths nearest 16 °C for thermoregulation (Oppedal et al., 2011), and likely swim deeper in standard cages when surface temperatures are above this threshold, avoiding infective copepodid encounters. Weak or no effects on louse infestation from snorkel cages during pycnoclines with warm brackish upper layers over late summer and early autumn indicate that depth-based technologies could be abandoned in these situations.

Our experimental design expands on previous studies investigating depth-based technology effects on salmon louse infestation in its combined scale, replication and duration (Table 1). While long-term controlled manipulative experiments in commercial fish production systems are logistically difficult, they are the ultimate test of effectiveness and feasibility for this type of technology. Other fish parasite control methods preventively applied over entire production cycles and lacking data, such as continuous vision-based laser systems (www.stingray.no) and the use of many species of cleaner fish, warrant investigations similar to ours to conclusively reveal performance and weaknesses. Only then can integrated parasite management strategies involving treatments and preventive measures at individual farm and regional scales (Groner et al., 2016) be effectively and adaptively prescribed.

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References

- Bron, J., Sommerville, C., Rae, G., 1837. Aspects of the behaviour of copepodid larvae of the salmon louse *Lepeophtheirus salmonis* (Krøyer, 1837). In: Boxshall, G.A., Defaye, D. (Eds.), *Pathogens of Wild and Farmed Fish: Sea Lice*. Ellis Horwood, Chichester, pp. 125–142.
- Brooker, A.J., Skern-Mauritzen, R., Bron, J.E., 2018. Production, mortality, and infectivity of planktonic larval sea lice, *Lepeophtheirus salmonis* (Krøyer, 1837): current knowledge and implications for epidemiological modelling. *ICES J. Mar. Sci.* 75, 1214–1234.
- Bui, S., Oppedal, F., Sievers, M., Dempster, T., 2019. Behaviour in the toolbox to outsmart parasites and improve fish welfare in aquaculture. *Rev. Aquac.* 11, 168–186.
- Crosbie, T., Wright, D.W., Oppedal, F., Johnsen, I.A., Samsing, F., Dempster, T., 2019. Effects of step salinity gradients on salmon lice larvae behaviour and dispersal. *Aquac. Environ. Interac.* 11, 181–190.
- Frenzl, B., Stien, L.H., Cockerill, D., Oppedal, F., Richards, R.H., Shinn, A.P., Bron, J.E., Migaud, H., 2014. Manipulation of farmed Atlantic salmon swimming behaviour through the adjustment of lighting and feeding regimes as a tool for salmon lice control. *Aquaculture* 424–425, 183–188.
- Glaropoulos, A., Stien, L.H., Folkedal, O., Dempster, T., Oppedal, F., 2019. Welfare, behaviour and feasibility of farming Atlantic salmon in submerged cages with weekly surface access to refill their swim bladders. *Aquaculture* 502, 332–337.
- Groner, M.L., Rogers, L.A., Bateman, A.W., Connors, B.M., Frazer, L.N., Godwin, S.C., Krkošek, M., Lewis, M.A., Peacock, S.J., Rees, E.E., Revie, C.W., Schlägel, U.E., 2016. Lessons from sea louse and salmon epidemiology. *Philos. Trans. R. Soc. B. Biol. Sci.* 371, 20150203.
- Grøntvedt, R.N., Kristoffersen, A.B., Jansen, P.A., 2018. Reduced exposure of farmed salmon to salmon louse (*Lepeophtheirus salmonis* L.) infestation by use of plankton nets: Estimating the shielding effect. *Aquaculture* 495, 865–872.
- Heuch, P.A., Parsons, A., Boxaspen, K., 1995. Diel vertical migration: a possible host-finding mechanism in salmon louse (*Lepeophtheirus salmonis*) copepodids? *Can. J. Fish. Aquat. Sci.* 52, 681–689.
- Høvøy, E., Boxaspen, K., Oppedal, F., Taranger, G., Holm, J., 2003. The effect of artificial light treatment and depth on the infestation of the sea louse *Lepeophtheirus salmonis* on Atlantic salmon (*Salmo salar* L.) culture. *Aquaculture* 220, 1–14.
- Korsøen, Ø.J., Dempster, T., Fjellidal, P.G., Oppedal, F., Kristiansen, T.S., 2009. Long-term culture of Atlantic salmon (*Salmo salar* L.) in submerged cages during winter affects behaviour, growth and condition. *Aquaculture* 296, 373–381.
- Kristoffersen, A.B., Quiller, L., Helgesen, K.O., Vollset, K.W., Viljugrein, H., Jansen, P. A., 2018. Quantitative risk assessment of salmon louse-induced mortality of seaward-migrating post-smolt Atlantic salmon. *Epidemics* 23, 19–33.
- Ling, E.N., Cotter, D., 2003. Statistical power in comparative aquaculture studies. *Aquaculture* 224, 159–168.
- Lovdata, 2012. Regulations on combating salmon lice in aquaculture facilities (in Norwegian: Forskrift om bekjempelse av lakselus i akvakulturanlegg) <https://lovdata.no/dokument/SF/forskrift/2012-12-05-1140> (accessed July 2018).
- Lovdata, 2017. Forskrift om produksjonsområder for akvakultur av matfisk i sjø av laks, ørret og regnbueørret (produksjonsområdeforskriften) https://lovdata.no/dokument/SF/forskrift/2017-01-16-61#KAPITTEL_2 (accessed July 2018).
- McCallum, H., Harvell, D., Dobson, A., 2003. Rates of spread of marine pathogens. *Ecol. Lett.* 6, 1062–1067.
- Nilsen, A., Nielsen, K.V., Biering, E., Bergheim, A., 2017. Effective protection against sea lice during the production of Atlantic salmon in floating enclosures. *Aquaculture* 466, 41–50.
- Oppedal, F., Dempster, T., Stien, L.H., 2011. Environmental drivers of Atlantic salmon behaviour in sea-cages: a review. *Aquaculture* 311, 1–18.
- Oppedal, F., Samsing, F., Dempster, T., Wright, D.W., Bui, S., Stien, L.H., 2017. Sea lice infestation levels decrease with deeper 'snorkel' barriers in Atlantic salmon sea-cages. *Pest Manag. Sci.* 73, 1935–1943.
- Samsing, F., Oppedal, F., Dalvin, S., Johnsen, I., Vågseth, T., Dempster, T., 2016. Salmon lice (*Lepeophtheirus salmonis*) development times, body size, and reproductive outputs follow universal models of temperature dependence. *Can. J. Fish. Aquat. Sci.* 73, 1841–1851.
- Samsing, F., Johnsen, I., Dempster, T., Oppedal, F., Trembl, E.A., 2017. Network analysis reveals strong seasonality in the dispersal of a marine parasite and identifies areas for coordinated management. *Landscape Ecol.* 32, 1953–1967.
- Sievers, M., Korsøen, Ø., Dempster, T., Fjellidal, P., Kristiansen, T., Folkedal, O., Oppedal, F., 2018. Growth and welfare of submerged Atlantic salmon under continuous lighting. *Aquac. Environ. Interac.* 10, 501–510.
- Stien, L.H., Dempster, T., Bui, S., Glaropoulos, A., Fosseidengen, J.E., Wright, D.W., Oppedal, F., 2016. 'Snorkel' sea lice barrier technology reduces sea lice loads on harvest-sized Atlantic salmon with minimal welfare impacts. *Aquaculture* 458, 29–37.
- Stien, L.H., Lind, M.B., Oppedal, F., Wright, D.W., Seternes, T., 2018. Skirts on salmon production cages reduced salmon lice infestations without affecting fish welfare. *Aquaculture* 490, 281–287.
- Wright, D.W., Stien, L.H., Dempster, T., Vågseth, T., Nola, V., Fosseidengen, J.E., Oppedal, F., 2017. 'Snorkel' lice barrier technology reduced two co-occurring parasites, the salmon louse (*Lepeophtheirus salmonis*) and the amoebic gill disease causing agent (*Neoparamoeba perurans*), in commercial salmon sea-cages. *Prev. Vet. Med.* 140, 97–105.
- Wright, D.W., Geitung, L., Karlsbakk, E., Stien, L.H., Dempster, T., Oldham, T., Nola, V., Oppedal, F., 2018. Surface environment modification in Atlantic salmon sea-cages: effects on amoebic gill disease, salmon lice, growth and welfare. *Aquac. Environ. Interac.* 10, 255–265.