



Environmentally responsive parasite prevention halves salmon louse burden in commercial marine cages

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ABSTRACT

Though a naturally occurring species throughout the Atlantic, parasitic salmon louse (*Lepeophtheirus salmonis*) population dynamics are increasingly intertwined with salmonid aquaculture. In contrast to reactive louse management, tools and strategies which reduce louse infestation success provide an off-ramp from the 'more farming equals more lice' feedback loop. This experiment tested the efficacy of a dynamic, environmentally responsive louse prevention strategy using common, commercially available tools throughout a full production cycle at commercial scale. By strategically luring salmon away from the halocline where concentrations of infective louse copepodids are highest using feeding and lights, and minimizing surface water flow through the cage with a protective skirt barrier when no halocline was present, both new louse infestations and mobile louse numbers were cut by half compared to control cages. The reduced louse numbers resulted in 25% fewer delousing events and improved fish welfare in dynamic cages, with no differences in gill condition or growth between treatments. With farmed salmon driving the ecology of salmon lice, this dynamic, environmentally responsive prevention strategy offers a way to work with nature, rather than against it, to reduce the parasite burden on both farmed and wild salmonids.

1. Introduction

The salmon (*Salmo salar*) farming industry has reached a turning point. The efficacy of reactive parasite control is failing (Jones et al., 1992; Besnier et al., 2014; Ljungfeldt et al., 2014; Helgesen et al., 2015; Aaen et al., 2015; Fjørtoft et al., 2019, 2020; Myhre Jensen et al., 2020), while populations of the parasitic salmon louse, *Lepeophtheirus salmonis*, are exploding (Dempster et al., 2021). Lice multi-resistant to chemotherapeutic treatments span the Atlantic (Fjørtoft et al., 2021), and with farmed salmon dramatically outnumbering wild it appears likely that lice will develop tolerance to non-medicinal treatment methods as well (Ljungfeldt et al., 2017; Groner et al., 2019; Andrews and Horsberg, 2021). Given the scale of salmon production and scope of the parasite populations, even if it were possible to improve treatment efficacy such a strategy would only have a limited effect (Mennerat et al., 2012; Jeong et al., 2021). The key to reducing the parasite burden and sustaining the growth of the salmon industry is prevention (Jeong et al., 2021).

The louse life-cycle can functionally be divided into four phases, (1) planktonic dispersal, (2) infective copepodid, (3) host attached and (4) mobile on host (Hamre et al., 2013). Mitigation measures which rely on

reactive treatment typically remove mobile lice from hosts, while preventative strategies focus on minimizing encounters between salmon and the infective copepodid stage during which the lice locate and attach to hosts (Overton et al., 2018; Barrett et al., 2020). Several preventative strategies have been developed and tested in recent years, including barriers which minimize surface water flow through cages (Stien et al., 2016, 2018; Geitung et al., 2019; Oppedal et al., 2019; Jónsdóttir et al., 2021), luring mechanisms to attract the salmon away from the depths where the highest densities of copepodids are expected (Juell et al., 2003; Frenzl et al., 2014; Wright et al., 2015; Nilsson et al., 2017), submergence of the entire cage (Dempster et al., 2009; Glaropoulos et al., 2019; Warren-Myers et al., 2022), functional feeds, louse repellents and traps, and selective breeding for resistance (Barrett et al., 2020). Each of these strategies have shown promise in some trials, but had little to no effect in others (Barrett et al., 2020).

Skirt barriers are one of the oldest and most effective prevention strategies (Vigen, 2008; Barrett et al., 2020). Because salmon lice are attracted to light and congregate near the waters' surface (Heuch et al., 1995; Crosbie et al., 2019), a semi-permeable barrier which allows water to pass but not copepodids is used to encircle the uppermost

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portion of the cage (Stien et al., 2012; Frank et al., 2015; Jónsdóttir et al., 2021). Skirt barriers range from 4 to 16 m deep, and have reduced louse infestation levels by 30–81% in commercial trials (Grøntvedt et al., 2018; Stien et al., 2018). Barrier protection, however, is not without cost. By reducing the water exchange in cages skirts not only reduce louse infestation, but also dissolved oxygen availability and potentially overall water quality (Stien et al., 2012; Oldham et al., 2017; Jónsdóttir et al., 2020). Although some tools have been developed which aim to improve water quality within barriers such as bubble-aeration devices which generate upwelling, their efficacy is yet to be confirmed (Wright et al., 2017).

Behavior modification is another of the few commercially tested louse prevention strategies. Lights, which are attractive to salmon at night (Juell et al., 2003; Juell and Fosseidengen, 2004; Wright et al., 2015), have been used seasonally to prevent sexual maturation in marine cages since the 1990's (Hansen et al., 1992, 2017; Oppedal et al., 1997, 2006; Taranger et al., 1999; Bromage et al., 2001). Typically such lights are placed near the waters' surface (Bromage et al., 2001; Migaud et al., 2007), which, given the surface oriented nature of lice, creates an obvious problem (Hevroy et al., 2003). Conversely, given their attractive nature, underwater lighting also presents an opportunity. By simply adjusting the vertical positioning of the lights they can be used to attract salmon away from the cage areas with highest louse density (Juell et al., 2003; Frenzl et al., 2014; Bui et al., 2019).

Positioning of food distribution is another potential behavior modification tool. During the day vertical distribution of salmon in cages can be explained by trade-offs between day-time surface avoidance, hunger and temperature (Juell et al., 1994; Fernö et al., 1995; Oppedal et al., 2011). Generally food acquisition over-rides other cues until satiation is reached (Oppedal et al., 2011). Thus, the industry-wide standard practice of scattering food pellets across the surface provides strong motivation for the fish to spend time in the risky shallows (Johansson et al., 2007). Submerging feed distribution removes the attraction to the surface during the daytime, theoretically reducing the likelihood of copepodid-salmon encounters. No trials have studied the louse preventative efficacy of submerged feed distribution in isolation; however, a few have tested the strategy in combination with submerged lights. In some conditions, during summer months, the average swimming depth of the salmon was consistently deeper than control cages resulting in less louse infestation; however, in other conditions, there was no observed effect (Frenzl et al., 2014; Nilsson et al., 2017).

In contrast to the methods described above which aim to reduce host-parasite encounters, functional feeds aim to reduce post-encounter infestation success by providing physiological benefits beyond those required for growth (Tacchi et al., 2011). A vast array of functional feed additives have been tested or are in development (Dawood et al., 2018), but those currently available commercially aim to boost immune function and optimize mucus production. So far the protective efficacy of functional feeds for salmon is highly variable with regards to parasitic infections, with a tendency to have small but beneficial effects on louse infestation (Barrett et al., 2020) and amoebic gill disease (AGD) severity (Bridle et al., 2005; Powell et al., 2007; Mullins et al., 2020).

Individually, each of these tools provide modest reductions in louse infestation. However, if they can be deployed concurrently and harnessed in a way which allows the additive effects of each tool to build on that of the others, these simple and relatively inexpensive strategies could significantly reduce the parasite burden in salmon sea cages. In a commercial trial where functional feed, submerged lights and feeding and a 6 m deep lice skirt were continuously deployed in combination for 13 months, louse infestation pressure was significantly reduced compared to control cages, but the reduction in louse attachment did not carry-over to later louse stages (Bui et al., 2020). Despite periodically fewer lice attaching, there was no difference in mobile louse numbers nor the number of delousing events required (Bui et al., 2020).

One key takeaway, which is true of all previous trials using depth-based prevention strategies, is that focusing on protecting salmon

from the uppermost few meters without considering environmental conditions is flawed. While louse copepodids do orient toward light, they also respond to salinity gradients and avoid brackish water, congregating instead at the halocline where full salinity water is nearest the surface (Samsing et al., 2016; Crosbie et al., 2019). Consequently, it was distance from the halocline rather than distance from the surface which correlated with louse attachment in Bui et al. (2020). Second, all cages in Bui et al. (2020) were stocked with cleanerfish, and wrasse in cages equipped with lice skirts consumed one-ninth as many lice as those in cages without skirts (Gentry et al., 2020). Thus, while a strategy which integrates several prevention measures is promising, refinements are required if the concept is to be of practical advantage.

The aim of this trial was to test a dynamic parasite management strategy whereby each tool is applied according to the local, real-time environmental conditions throughout a complete production cycle at commercial scale. Rather than focusing on minimizing exposure to and protection from surface waters, this trial focused on minimizing exposure to the water depths where predicted copepodid density was highest based on salinity. Test cages were equipped with a lice skirt, aeration device, adjustable depth lights and feeding, and provided functional feed, while control cages were fed standard commercial feed at the surface. All cages were stocked with cleanerfish. Efficacy was measured in terms of louse burden (infestation & mobile stages), delousing frequency, salmon growth and welfare.

2. Materials & methods

2.1. Experimental set-up

The trial was carried out on a commercial salmon farm (Fosså) equipped with six circular sea cages (cage circumference = 200 m, depth = 38 m) arranged in two parallel rows perpendicular to shore in Ombofjord outside Skiftun, southwestern Norway (59.2699° N, 6.14295° E). All fish were from a single batch of roe (Mowi breed) and vaccinated with Alpha Ject micro 1 PD and Alpha Ject Micro 6 (Pharmaq) prior to transfer from the Kvingo freshwater facility in September 2018. Approximately 167,000 smolt (range: 161,126–175,667) which weighed on average 94–101 g were transferred into each cage.

The experiment consisted of two treatment groups each replicated in three cages, (a) control cages which followed normal production procedures, and (b) test cages utilizing a dynamic parasite prevention strategy. Control cages were given size appropriate standard commercial feed (Skretting) throughout production. Cages utilizing the dynamic strategy were supplemented with commercially available functional feed as appropriate (Protec, Skretting, Norway) and equipped with adjustable depth lights (150 W/1200 W Aurora SubLED Combi light, AKVA group) and feeding (SubFeeder, AKVA group), a 6 m skirt barrier (Fiizk, Norway), and a bubble aeration device (Midt-Norsk ringen, NorseAqua, Norway). Treatment groups were distributed throughout the cages in a factorial design. In addition, all cages were periodically stocked with seasonally appropriate cleaner fish divided amongst the cages to maintain similar relative stocking densities (~5% of salmon number) in all cages throughout production. Cleaner fishes used included ballan wrasse (*Labrus bergylta*), corkwing wrasse (*Symphodus melops*), goldsinny wrasse (*Ctenolabrus rupestris*) and lumpfish (*Cyclopterus lumpus*). To support cleaner fish welfare each cage was equipped with kelp-type shelters placed in a 'corridor' style layout and species appropriate food. Cleaner-fish were fished from cages prior to all delousing treatments.

The industry partner managing the facility adheres to the national legislation limiting the numbers of adult female lice allowed per fish to 0.2 during migration season and 0.5 the rest of the year. When the farm-average louse levels reached threshold, delousing treatments were administered on a cage-basis according to standard commercial practice. Because this experiment was performed at an R & D concession site granted by the Norwegian Ministry of Trade, Industry and Fisheries

separate ethics approval was not required.

2.2. Dynamic strategy

The driving principle of the dynamic strategy was to utilize local, real-time environmental data to optimally deploy each tool for the prevailing conditions. When salinity was homogeneous throughout the cage volume and copepodids were expected at highest density in the surface, fish were lured away from the surface using eight submerged lights positioned at 8–10 m depth and submerged feeding at 8 m. To protect fish which chose to spend time near the surface anyway the influx of copepodids was minimized by shielding the uppermost 6 m of the cage with a lice barrier. Finally, to maintain optimal water quality within the barrier volume, an aeration mixing device was positioned centrally at 10 m to circulate deep water upwards. Conversely, when there was a brackish layer at the surface (defined as salinity ≤ 26 ppt at 3 m for at least three days), the louse barrier was removed, and aeration turned off to facilitate maintenance of the brackish layer within the cage. Additionally, because lice aggregate at highest density near the halocline (Crosbie et al., 2019), lights and feeding were moved to the surface to attract the fish into the brackish layer above the halocline.

2.3. Environmental monitoring

In total the site was equipped with 20 real-time environmental monitoring sensors which could be remotely viewed online (InnovaSea, USA). Two combined dissolved oxygen and temperature sensors were placed in each cage at depths of 3 m (mid-skirt) and 12 m (below skirt), as well as a salinity sensor at 3 m, all approximately 5 m from the cage wall. An additional set of dissolved oxygen, temperature and salinity sensors were placed at 3 m at a reference site at least 100 m from the nearest cage. A multi-sensor CTD (SD204, SAIV AS) was also used to collect a daily vertical profile of temperature, salinity and dissolved oxygen from the surface to 40 m.

2.4. Sampling protocol

Throughout production fish were regularly assessed for louse infestation, gill health and welfare. A baseline sampling was performed at the hatchery immediately prior to sea transfer, with sampling every 1 to 4 weeks thereafter until harvest for a total of 24 sample events. Fish were captured using either a seine net or 3 m diameter ring-net pulled from ~15 m to the surface by crane, depending on fish behavior at the time of sampling. The same catch method was used for all cages at each individual sampling event. During each sampling 30 fish per cage were euthanized in a sedative bath (Tricaine methanesulfonate) and all lice on each host and in the sedation vessel were counted and staged. Fish welfare was evaluated using 13 morphological indicators outlined in the FISHWELL scoring system, scored as 0 = ideal, 1 = light, 2 = moderate and 3 = extreme (Noble et al., 2018). Gill health was further evaluated by visually scoring each arch using the standard 0 to 5 scale for amoebic gill disease (AGD) (Taylor et al., 2016).

2.5. Data analyses

Louse stages were grouped as either new infestations (all attached stages: copepodid, chalimus 1 and 2) or existing infestations which could have been present at the previous sampling (all mobile stages: pre-adult 1, pre-adult 2 and adults). Total gill score was calculated as the mean AGD score of all eight arches (0–5) plus the separate proliferative gill inflammation (PGI) score (0–3). A factor was then created which separated total gill score as being either acceptable (≤ 2) or bad (> 2). Condition factor (K) was calculated as $K = \text{body weight (g)} / \text{fork length}^3 \text{ (cm)} \times 100$ (Ricker, 1975). Repeated measures anova were used to determine if there were differences in growth or condition factor between treatment groups. Repeated ordinal regression was used to

determine if there were differences in welfare score between treatment groups. Unless otherwise specified, data shown are mean \pm SD.

For the key parameters of interest, new louse infestation and gill condition, an information theoretic approach was used to find the optimal set of covariates which best fit the data (Burnham and Anderson, 2002). Existing and total louse infestations could not be statistically examined because delousing treatments, which affect all mobile louse stages, were differentially applied to cages on an as-needed basis. Standard procedures for data exploration were followed to identify any outlying observations and test for collinearity between potential explanatory variables for both the new louse infestation and gill condition datasets (Zuur et al., 2010). A list of all covariates considered for both models is provided in Table 1. To find the optimal set of covariates for both response variables a selection of candidate models was prepared a-priori based on specific hypotheses. Models were then compared using Akaike information criterion (AIC). As these data consist of observations of multiple fish from the same set of cages sampled repeatedly throughout a production cycle, mixed-effects models were applied with cage and day in year as random intercepts. All analyses were carried out in R version 3.6.1 (R Core Team 2018).

To examine the factors affecting new louse infestations, the total number of sessile lice on each fish was modelled using generalized additive mixed models (GAMMs). The mgcv package was used to estimate the parameters of the GAMMs (Wood, 2011, 2017). Because 56% of individuals measured in the trial had zero attached lice the zero-inflated poisson distribution was chosen. Table 2 shows the 13 models which were applied.

To evaluate the impacts of treatment and environmental variation on gill condition, total gill score was modelled using generalized linear mixed models (GLMMs) with a binomial distribution. The lme4 package was used to estimate the parameters of the GLMMs (Bates et al., 2015). Table 3 shows the 12 models which were applied.

3. Results

3.1. Environmental conditions

There was considerable temporal and spatial variation in environmental conditions (Fig. 1). From the surface down to 30 m depth salinity averaged 31.8 ± 2.3 ppt, with a minimum of 18.9 ppt in the upper 3 m and a maximum of 35 ppt below 25 m. In total, three periods displayed a

Table 1
List of covariates considered in the new infestation and gill condition models.

Covariate	Abbreviation	Continuous/categorical
New infestation model		
Treatment	treatment	Categorical (control or dynamic)
Halocline	halo	Categorical (present or absent)
Depth of optimal temperature	depth14	Continuous
Length	L	Continuous
Condition factor	K	Continuous
Medicated feed	feed	Categorical (yes or no)
Scale loss	scaless	Categorical (0,1,2,3)
Adult female louse count	female lice	Continuous
Gill condition model		
Treatment	treatment	Categorical (control or dynamic)
Halocline	halo	Categorical (present or absent)
Depth of optimal temperature	depth14	Continuous
Length	L	Continuous
Condition factor	K	Continuous
Mean dissolved oxygen (0–25 m)	DO	Continuous
Mean temperature (0–25 m)	temp	Continuous
De-lousing status	deloused	Categorical (yes or no - deloused within the previous 4 weeks)

Table 2

Explanation of the 13 models tested to determine the optimal set of covariates to explain variation in new louse infestations, as measured by sessile louse counts. Degrees of freedom, AIC, delta AIC and Akaike weight are shown for each model.

Model	Expression	df	AIC	Δ AIC	Akaike weight	Description
M1	intercept only	7	23,197	6780	0.000	null
M2	treatment	9	22,646	6229	0.000	Treatment only
M3	halo + depth14	11	20,398	3981	0.000	Environment only
M4	L + K	10	20,941	4524	0.000	Fish condition only
M5	feed + scale loss	11	22,357	5940	0.000	Handling only
M6	treatment: halo + treatment: depth14	29	17,153	736	0.000	Interaction between treatment & environment
M7	treatment: halo + treatment: depth14 + L + K	30	16,437	20	0.000	Interaction between treatment & environment + fish condition
M8	treatment: halo + treatment: depth14 + L + K + feed + scale loss	32	16,424	7	0.019	Interaction between treatment & environment + fish condition + handling
M9	treatment: halo + treatment: depth14 + L + K + feed + scale loss + female lice	33	16,417	0	0.708	Interaction between treatment & environment + fish condition + handling + adult density
M10	treatment: halo + treatment: depth14 + L + K: treatment	31	16,439	22	0.000	Interactions between treatment & environment + treatment & fish condition
M11	treatment: halo + treatment: depth14 + L + K: treatment + feed + scale loss	33	16,426	9	0.007	Interactions between treatment & environment + treatment & fish condition + handling
M12	treatment: halo + treatment: depth14 + L + K: treatment + female lice	32	16,432	15	0.000	Interactions between treatment & environment + treatment & fish condition + adult density
M13	treatment: halo + treatment: depth14 + L + K: treatment + feed + scale loss + female lice	34	16,419	2	0.266	Interactions between treatment & environment + treatment & fish condition + handling + adult density

strong enough brackish surface layer (salinity ≤ 26 ppt at 3 m for at least three days) to trigger the change in management strategy whereby the skirts were removed and fish were encouraged into the surface with lights and feeding (Fig. 1b). Temperatures were seasonally variable, averaging 10.7 ± 3.9 °C. The most extreme temperatures were observed in the surface layer above 5 m and ranged 4.0–20.2 °C, while the range at 30 m was smaller, 5.5–15.8 °C (Fig. 1a). Ambient DO saturation averaged $91 \pm 8.4\%$, with three periods when it dropped below 80% saturation, primarily in mid- or deep-water: Jan-Feb 2019, Nov-Dec 2019 and Feb-Mar 2020. The minimum ambient DO recorded was

Table 3

Explanation of the 13 models tested to determine the optimal set of covariates to explain gill condition, as measured by total gill score. Degrees of freedom, AIC, delta AIC and Akaike weight are shown for each model.

Model	Expression	df	AIC	Δ AIC	Akaike weight	Description
G1	intercept only	3	1503	13	0.000	null
G2	treatment	4	1504	14	0.000	Treatment only
G3	temp	4	1499	9	0.005	Temperature only
G4	DO	4	1502	12	0.000	Dissolved oxygen only
G5	halo + depth14	6	1508	18	0.000	Environmental preference
G6	L + K	5	1494	4	0.050	Fish condition
G7	treatment + deloused	5	1506	16	0.000	Handling
G8	DO + temp	5	1499	9	0.005	Environmental conditions
G9	L + K + DO + temp	7	1490	0	0.364	Fish condition + environmental conditions
G10	L + K + DO + temp + K: temp	8	1490	0	0.501	Importance of condition factor interacts with temperature
G11	L + K + treatment + deloused	7	1498	8	0.008	Fish condition + handling
G12	L + K + treatment + deloused + halo + depth14	10	1503	13	0.000	Fish condition + handling + environmental preference
G13	L + K + treatment + deloused + DO + temp	9	1494	4	0.060	Fish condition + handling + environmental conditions

49% saturation in Feb 2020, and maximum 121% during spring/summer 2019 (Fig. 1c). Real-time sensors within the dynamic strategy cages showed that one mixing device was sufficient to maintain DO at or near ambient concentrations throughout this experiment, but not higher (Fig. 2).

3.2. Growth and welfare

Overall, from sea transfer in September 2018 until harvest in March 2020, there were no significant differences in fish weight ($df = 4$, $F = 0.043$, $P = 0.85$) or condition factor ($df = 4$, $F = 0.638$, $P = 0.47$) between the control and dynamic treatment groups (Fig. 3). In contrast, welfare scores were significantly lower in the dynamic treatment group than control (estimate = -0.61 , SE = 0.25, $z = -2.47$, $P = 0.01$). Of a maximum potential welfare score of 39 (extremely damaged fish), welfare scores ranged from 2.9 (dynamic – Aug 2019) to 9.9 (control – Feb 2020), with average welfare scores throughout the trial for fish in control cages being 5.94 ± 2.43 and dynamic 5.38 ± 2.19 . The main factors which contributed to elevated welfare scores were scale loss, fin damage and cataracts which averaged between 1 and 1.5 throughout the trial, followed by skin haemorrhaging, eye haemorrhaging and mouth injuries which averaged between 0.3 and 0.8.

3.3. Preventative effect – louse attachment

On average, fish in control cages were host to 2.9 ± 4.5 sessile lice while fish in cages using the dynamic prevention strategy were host to 1.1 ± 2.1 sessile lice. Thus, overall louse infestation was reduced by 62% in cages utilizing the dynamic strategy. Further, out of 24 sampling events sessile louse counts were lower in dynamic cages compared to controls 21 times (by 12 to 87%), while there was no difference on three occasions (Fig. 3c). In control cages there were two infestation peaks,

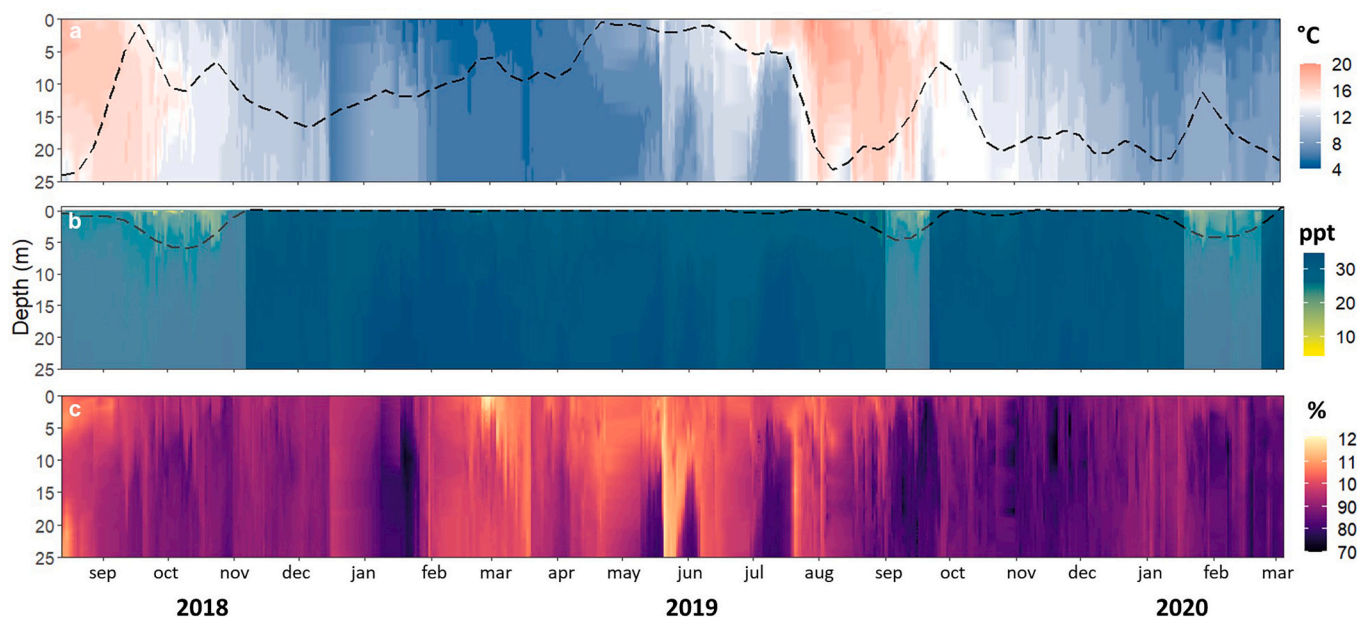


Fig. 1. Environmental conditions. (a) Ambient temperature ($^{\circ}\text{C}$), (b) salinity (ppt) and (c) dissolved oxygen saturation (%) at a reference location at least 100 m from nearest cage. The dashed black lines mark the shallowest depth of the optimal temperature (a) and the maximum depth of 26 ppt salinity (b). Shaded areas in (b) denote when a brackish layer was present.

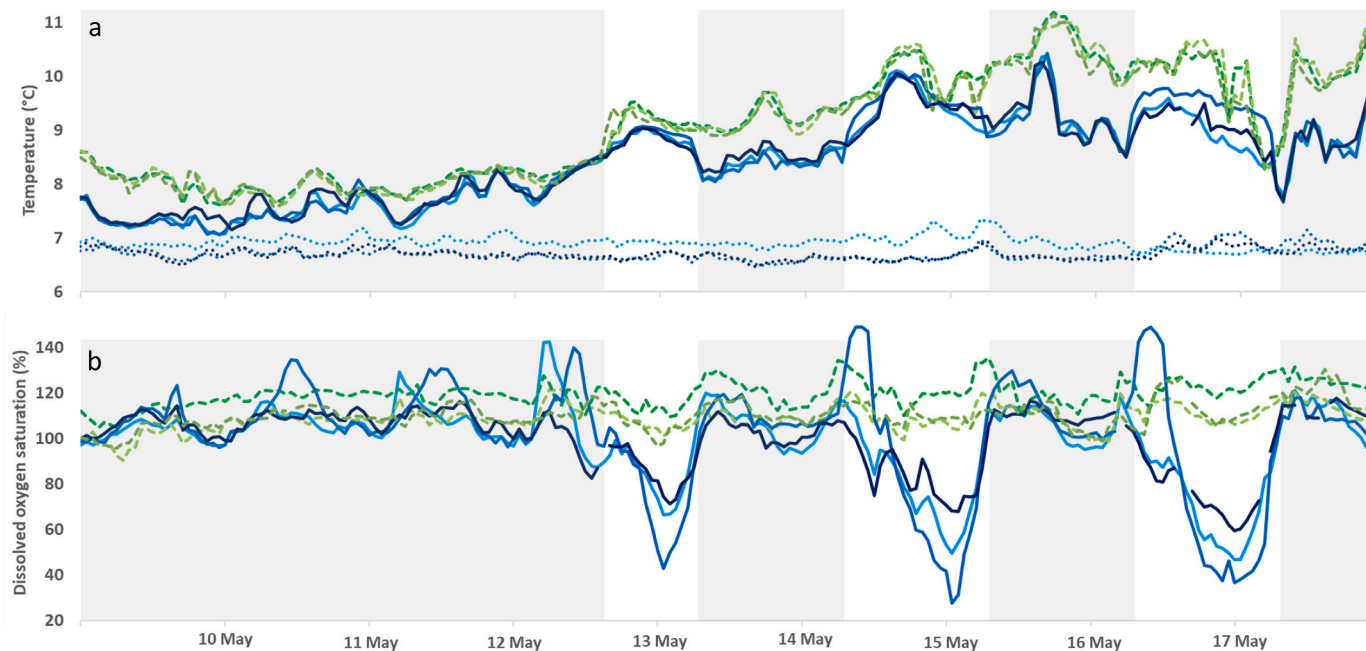


Fig. 2. Aeration effect. Effect of aeration devices on water quality within skirts. Dashed green lines present (a) temperature and (b) dissolved oxygen saturation at 3 m in the three replicate control cages, while solid blue lines present data for the three replicate cages using the dynamic louse prevention strategy, which during this period were each fitted with a 6 m skirt barrier. Dotted blue lines in the temperature plot present data for the three dynamic cages at 12 m. Shaded areas denote periods when the aeration devices were operating at maximum output, while the open windows denote periods when the aeration devices were turned off.

one in September 2019 (17.7 ± 7.3 sessile lice fish $^{-1}$) and one in January 2020 (11.9 ± 4.5 sessile lice fish $^{-1}$). The infestation pattern was different in cages using the dynamic strategy, with a single peak in new infestations in September 2019 (10.1 ± 4.5 sessile lice fish $^{-1}$) (Fig. 3c).

The number of regression parameters, AICs, differences in AIC values (ΔAIC) and Akaike weights for each model of sessile lice fish $^{-1}$ are presented in Table 2. Akaike weights can be directly interpreted as conditional probabilities, so a perfect model would have an Akaike weight of 1. For sessile lice one model stood out as far superior to the

rest, model M9, with an Akaike weight of 0.708, followed by M13 with an Akaike weight of 0.266. Therefore, in 71% of cases model M9 (Interaction between treatment & environment + fish condition + handling + adult density) is the most optimal, while in 27% of cases model M13 (all factors & interactions) is the most optimal. Both models contain interactions between treatment and halocline and treatment and depth of optimal temperature with a smoother, as well as all predictor variables: treatment, halocline, depth of optimal temperature, length, condition factor, scale loss, medicated feed, scale loss and adult female

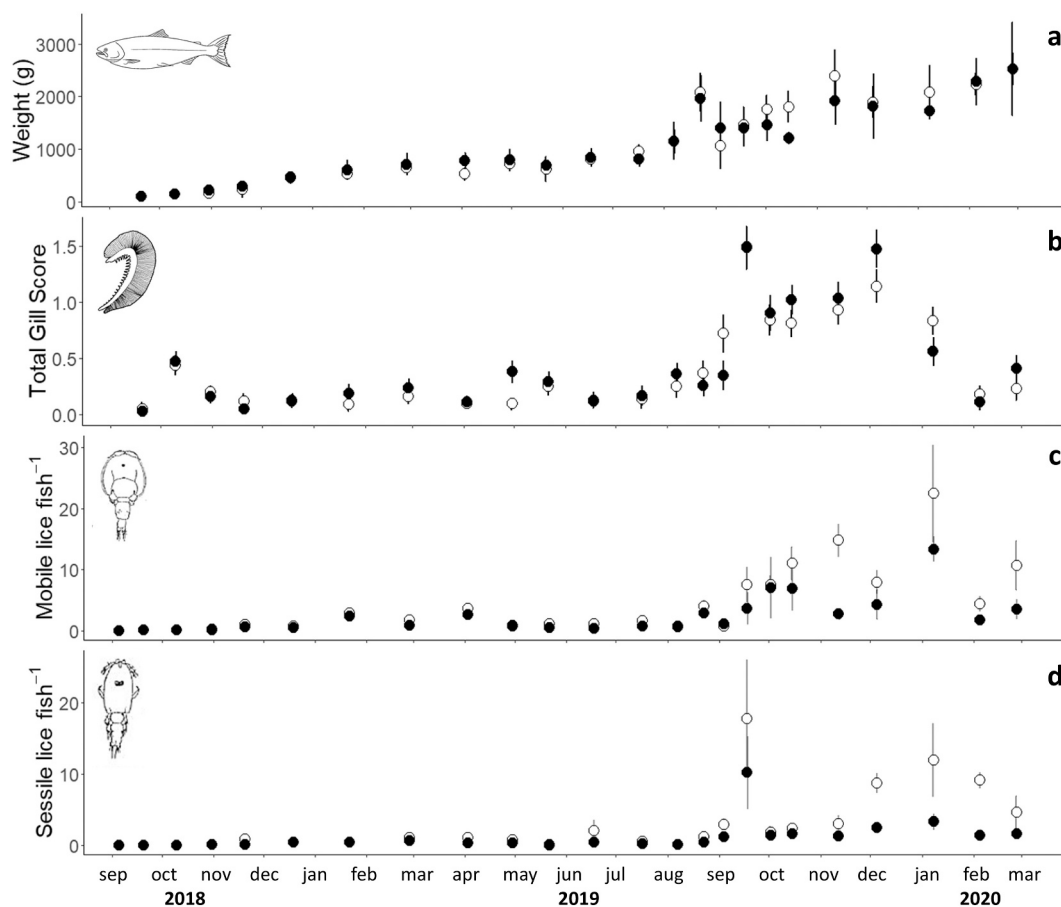


Fig. 3. Lice and welfare. Mean \pm 95% confidence interval of (a) weight, (b) total gill score, (c) mobile lice (pre adults, adults) and (d) sessile lice (copepod, chalimus I, chalimus II) in standard cages (open circle) and cages utilizing a dynamic lice prevention strategy (solid circles) on all sampling dates throughout the production cycle.

louse count. In addition, model M13 also includes an interaction between treatment and condition factor. In M9 the smoother on the interaction between treatment and depth of optimal temperature as well as the covariates treatment, halocline, length, medicated feed, scale loss and adult louse density are all important determinants of sessile louse numbers (Table 4). According to model M9, louse attachment is minimal in dynamic cages when optimal temperatures are in the upper portion of the cage, with a single peak in infestation pressure when optimal temperatures were around 20 m deep (Fig. 4). In contrast, infestation pressure in control cages was bimodal with a first peak when optimal temperatures were around 9 m, and a second peak when optimal temperatures were around 20 m (Fig. 4). Infestation pressure in both control and dynamic cages peaked when the halocline was 6 m deep (Fig. 5).

3.4. Preventative effect – mobile lice

Throughout the trial control cages averaged 4.4 ± 5.6 mobile lice fish⁻¹ (maximum = 22.4, Jan 2020), while the dynamic cages averaged 2.4 ± 3.1 mobile lice fish⁻¹ (maximum = 13.2, Jan 2020). On 21 of the 24 sampling events dynamic cages had fewer mobile lice than controls, and less than half as many lice on nine occasions (Fig. 3). On the three occasions where control cages had fewer mobile lice than dynamic, both groups averaged less than one mobile lice fish⁻¹. Further, the observed reduction in mobile lice infestation in dynamic cages relative to controls occurred despite dynamic cages also receiving 25% fewer de-lousing treatments. In total, control cages were deloused 24 times throughout production (mean = 8 cage⁻¹), including the early harvest of two cages due to high lice levels in conjunction with cardiomyopathy syndrome which precluded de-lousing, while the dynamic strategy cages required

Table 4

Estimate, standard error (SE), z-value and P-values of the explanatory variables in the minimum adequate models for (a) new louse infestation and (b) gill condition.

	Estimate	SE	z value	P
(a) New louse infestation				
Intercept	0.065	0.080	0.814	0.416
Treatment-Dynamic	-0.815	0.066	-12.397	<0.001
Halocline-Present	-0.458	0.056	-8.239	<0.001
Length	0.027	0.001	24.832	<0.001
Condition factor	-0.109	0.059	-1.827	0.068
Medicated feed	-0.301	0.100	-3.020	0.003
Scale loss	-0.051	0.017	-3.012	0.003
Adult louse density	-0.061	0.021	-2.973	0.003
Treatment-Dynamic: Halocline-Present	0.115	0.103	1.118	0.264
significance of smooth terms		df	chi.sq	P
Depth14: Treatment-Control		9	1351	<0.001
Depth14: Treatment-Dynamic		9	786	<0.001
(b) Gill condition				
Intercept	1.328	3.741	0.355	0.723
Length	0.028	0.009	2.965	0.003
Condition factor	-3.529	1.738	-2.031	0.042
Mean dissolved oxygen (0–25 m)	-0.044	0.030	-1.454	0.146
Mean temperature (0–25 m)	-0.090	0.169	-0.536	0.592
Condition factor: mean temperature	0.226	0.137	1.654	0.098

18 delousing events (mean = 6 cage⁻¹) and were able to be harvested according to schedule. At each delousing event, the same method was used all treated cages. Most of the delousing treatments were thermal

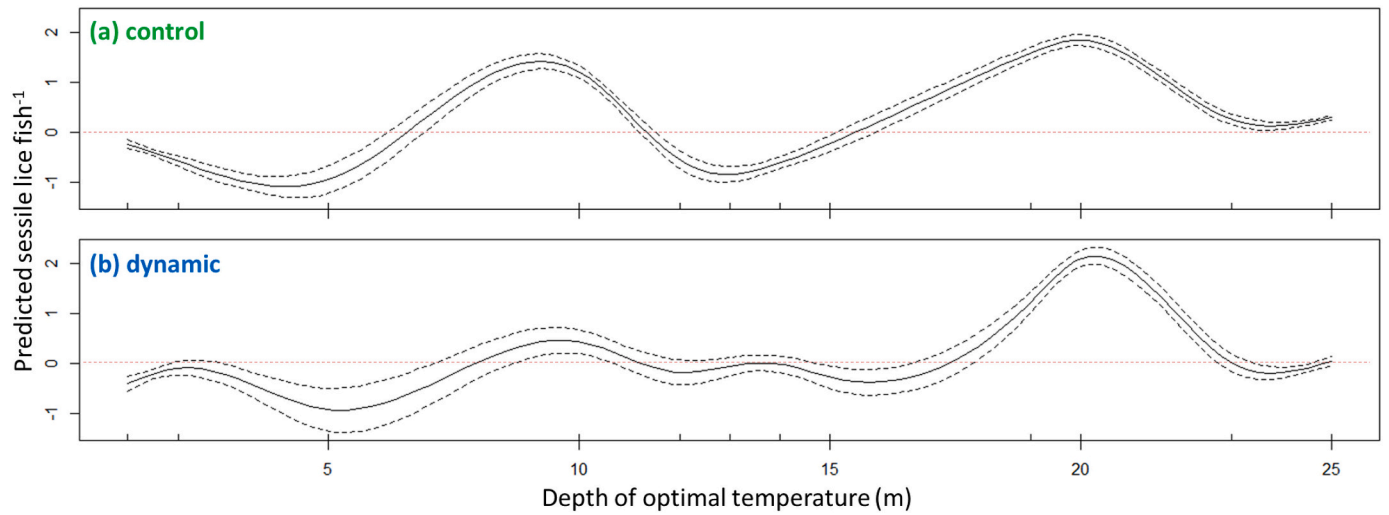


Fig. 4. Effect of depth of optimal temperature on sessile louse numbers. Modelled smoother effect of the interaction between treatment and depth of optimal temperature on sessile louse numbers (copepodid, chalimus I and chalimus II) in (a) standard cages and (b) cages utilizing a dynamic louse prevention strategy. The solid line depicts the smoother and dashed lines the 95% confidence intervals. While new louse attachments were unaffected by depth of optimal temperature in dynamic cages except at 20 m, it was an important determinant of new attachments in control cages.

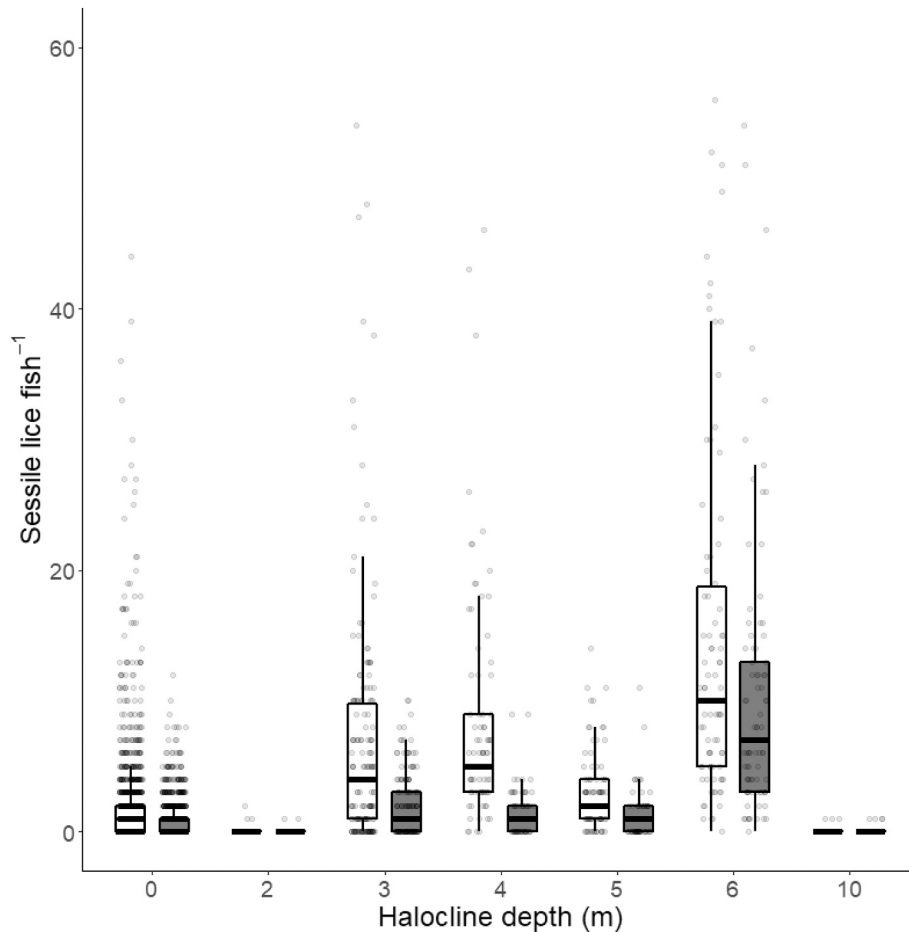


Fig. 5. Impact of halocline depth on sessile lice numbers. Boxplot illustrating the variation in sessile lice numbers (copepodid, chalimus I, chalimus II) in standard cages (open boxes) and cages utilizing a dynamic louse prevention strategy (filled boxes) in relation to halocline (26 ppt) depth. Points present louse counts from individual fish.

(heat), while two treatments were mechanical (hydrolicer, smir.no).
 In general, *Caligus elongatus* abundance was low (control: 0.2 ± 1.4 lice fish⁻¹, dynamic: 0.2 ± 1.4 lice fish⁻¹) and there was no significant

difference between treatment groups ($df = 1, F = 0.015, P = 0.9$). In both groups *C. elongatus* prevalence began to increase in June 2019. In the dynamic group abundance peaked once in August 2019 (0.9 ± 5.3 lice

fish⁻¹) and a second time in October 2019 (0.7 ± 3.6 lice fish⁻¹), after which they tapered off. In the control group there was a single peak in *C. elongatus* infestation in October 2019 (1.2 ± 5.4 lice fish⁻¹).

3.5. Gill condition

Total gill score ranged from 0.03 immediately after sea transfer to 1.49 in Sept 2019 (Fig. 3). Gill score followed the same general pattern in both treatments, rapidly increasing to 0.5 the first autumn after transfer before declining through winter, increasing again throughout the second autumn, and dropping off again through winter (Fig. 3). Overall, total gill score averaged 0.39 ± 0.63 in control cages and 0.41 ± 0.66 in cages using the dynamic strategy. Of the factors contributing to poor gill condition, proliferative gill inflammation (PGI) peaked in Sept 2019, while AGD peaked in Dec 2019. The number of regression parameters, AICs, differences in AIC values (Δ AIC) and Akaike weights for each model of gill condition are presented in Table 3. For gill condition, two models stood out as superior to the rest, G9 with an Akaike weight of 0.364 and G10 with an Akaike weight of 0.501. Therefore, in 50% of cases model G10 (Importance of condition factor interacts with temperature) is the most optimal, while in 36% of cases model G9 (Fish condition + environmental conditions) is the most optimal. Both models contain the same predictor variables: fish length, condition factor, mean dissolved oxygen and mean temperature, while model G10 also includes an interaction between condition factor and mean temperature. In G9 the covariates length, condition factor and mean temperature are significant determinants of gill condition, while in G10 only length and condition factor are significant. According to model G10, probability of poor gill condition increases with higher temperatures and larger fish size, while it decreases with higher condition factor (Fig. 6). The interaction between temperature and condition factor in model G10 demonstrates that the protective effect of higher condition factor is reduced at higher temperatures (Fig. 6). The treatment, halocline status, depth of optimal temperature and delousing status covariates were not important drivers of gill condition and did not improve fit in comparison to an intercept only model.

3.6. Cleanerfish

In total 75,887 lumpfish and 71,613 wrasse were added to control cages, while 80,217 lumpfish and 71,382 wrasse were added to cages utilizing the dynamic strategy. The first mortalities were observed on 13 Sept 2018, immediately after the first stocking of wrasse, in both control

and dynamic strategy cages. Overall, there were no differences in mortality between treatment groups for wrasse or lumpfish. At the time of harvest, 20% of the wrasse stocked in control cages and 28% of lumpfish had been recorded as mortalities, compared to 19% of wrasse and 23% of lumpfish in cages utilizing the dynamic prevention strategy.

4. Discussion

Despite decades of research and development, salmon lice remain the greatest impediment to the sustainable expansion of the salmon farming industry. Although not a silver bullet, these results demonstrate that the salmon louse burden can be significantly reduced in marine cages using a straightforward strategy and widely available tools. Further, given the environmentally responsive nature of the dynamic strategy tested herein, it is a method which can be adapted to suit most salmon farming sites.

All previous commercial tests of louse prevention strategies have focused on minimizing contact between salmon and surface waters, assuming that would be where louse copepodids were most abundant (Guragain et al., 2021). Such strategies have yielded inconsistent results (Barrett et al., 2020). In some cases, at locations where haloclines are rare or the protective barrier was deep enough to exceed the halocline depth, such strategies performed exceptionally well (Oppedal et al., 2017; Wright et al., 2017; Geitung et al., 2019). In other trials however, when haloclines were more common or the preventative tools did not provide sufficient protection from the halocline, reductions in louse burden were highly variable (Frenzl et al., 2014; Nilsson et al., 2017; Grøntvedt et al., 2018; Stien et al., 2018; Wright et al., 2018; Oppedal et al., 2019; Bui et al., 2020). For a louse prevention strategy to be successful, it must take site-specific environmental conditions into account (Samsing et al., 2016).

The dynamic strategy we describe used the same tools tested in previous work, but in contrast to those trials used site-specific environmental data collected in real-time to adapt the prevention strategy based on local salinity conditions. Overall, the dynamic strategy halved new louse infestations and mobile louse abundance compared to control cages, resulting in 25% fewer delousing treatments. Furthermore, these reductions in louse burden led to improved fish welfare and were achieved with no negative effects on growth.

Importantly, however, the reduction in louse infestation pressure was not entirely consistent throughout the trial (Fig. 7). As implemented here the dynamic strategy responded only to a halocline trigger by either (a) encouraging the fish deep and protecting the surface when salinity

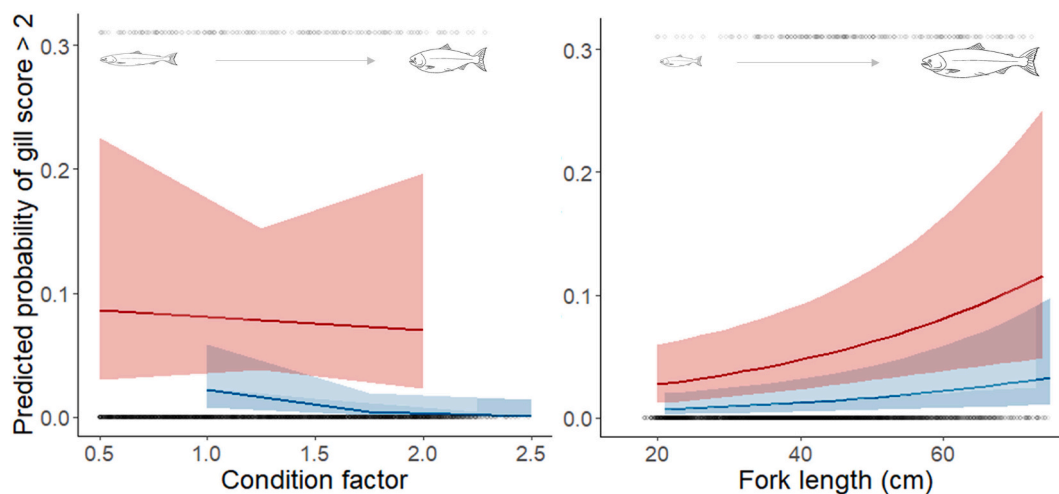


Fig. 6. Influential determinants of poor gill condition. Predicted probability of a total gill score greater than 2 as influenced by condition factor, fish length and mean sea temperature (0–25 m). Solid lines and shaded areas display a fitted GAMM with 95% confidence intervals, while dot points represent each individual measured. Red indicates model predictions at 15 °C, while blue indicates predictions for 6 °C.

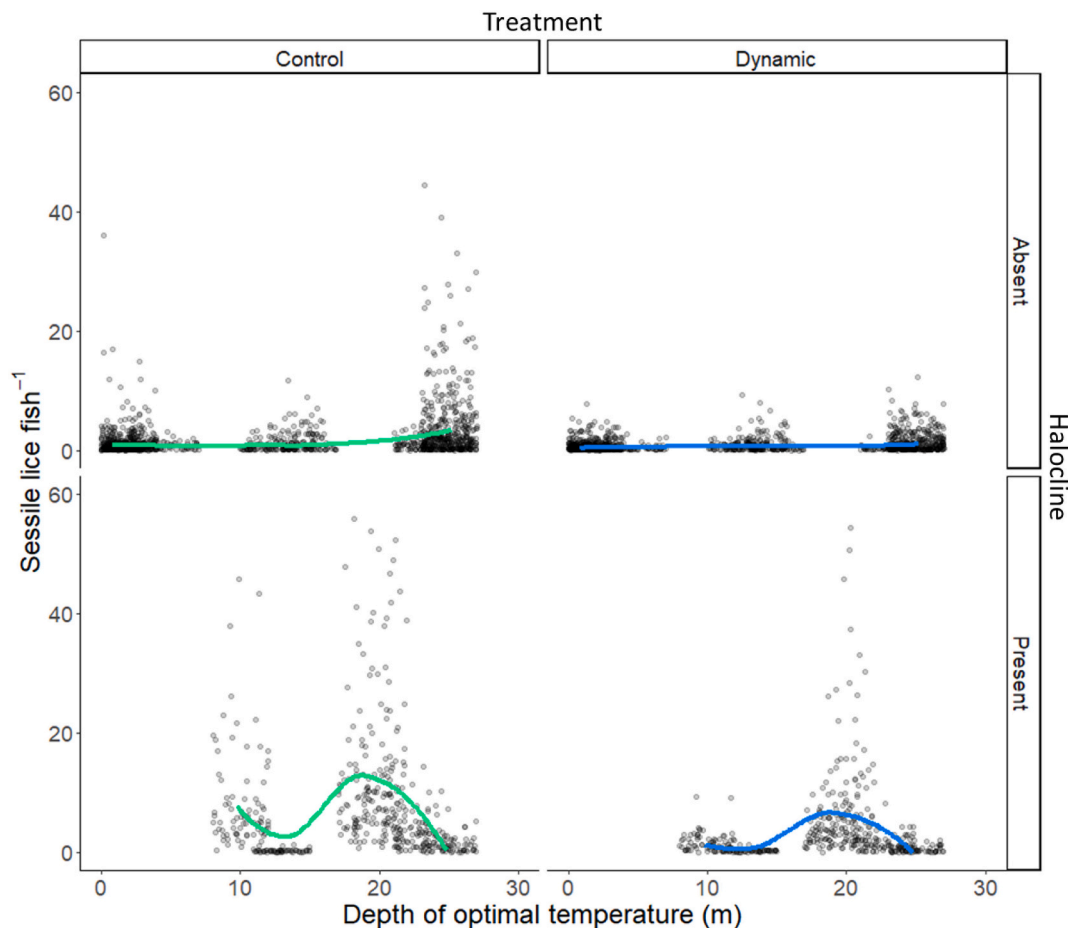


Fig. 7. Influence of the environment on louse infestation pressure. Points present sessile (copepodid, chalimus I, chalimus II) louse counts from each individual fish, while solid lines display a fitted GAM. The GAM is presented in green for standard cages and blue for cages utilizing the dynamic prevention strategy. The figure is divided into the control and dynamic treatment groups, as well as whether or not a halocline, defined as salinity ≤ 26 ppt at 3 m, was present or absent.

was relatively uniform, or (b) encouraging the fish into the surface brackish layer when present. Temperature was not included in the dynamic strategy framework. As such, new louse infestations were reduced in dynamic cages when the halocline was absent and preferred temperature deep, and when the halocline was present and preferred temperature relatively shallow. In short, reduced infestation was seen when the preferred temperature of the salmon matched the depth at which we were aiming to lure the salmon. However, when a halocline was present and preferred temperature was deep, new louse infestations were high in both groups. In hindsight, the reason for this appears clear. During daylight hours the behavior of farmed salmon is driven primarily by trade-offs between hunger, surface avoidance and temperature (Oppedal et al., 2011). By attempting to lure the salmon into the brackish layer with surface feeding when their preferred temperature was deep we inadvertently encouraged them to repeatedly swim through the halocline, where copepodid density is highest, until satiated. Future iterations of a dynamic louse prevention strategy should incorporate both temperature and salinity into the decision matrix to avoid such conflicts. In addition, by feeding at the depth of the salmon's preferred temperature a dynamic strategy could not only reduce louse burden further but also maximize feed intake and growth potential.

Also noteworthy was the lack of effect of the dynamic strategy on *C. elongatus* infestation. *Caligus* sp. occur in southern Norway, where the experiment was conducted, but are not considered a major pest like *L. salmonis*. While it is likely that an environmentally informed prevention strategy could also work for *Caligus* sp., further work is needed to understand the behavior and environmental preferences of *Caligus* sp. for such a strategy to be effective. It is clear from these data that

C. elongatus are distributed deeper in the water column than *L. salmonis* and respond differently to variations in salinity.

Beyond lice, a major concern regarding the use of barrier technologies is their effect on gill health. Previous tests of skirt (Stien et al., 2012, 2018; Oldham et al., 2017; Jónsdóttir et al., 2020) and snorkel (Oppedal et al., 2017; Wright et al., 2017) barriers have shown that they can cause dissolved oxygen levels to rapidly decline, sometimes even in the presence of mitigation measures. Additionally, though to our knowledge not documented, there are reports from farmers and veterinarians that gill diseases progress faster and have poorer prognosis in cages equipped with barriers. One possible cause for this could be the direct effects of poor dissolved oxygen conditions within barriers accelerating disease progression (Oldham et al., 2020). Alternatively, the reduced water flow through barriers may lead to more rapid accumulation of infectious particles once an infection is present in the population (Frank et al., 2015; Jónsdóttir et al., 2021). Whatever the cause in previous tests, the use of skirt barriers was not an important determinant of gill condition in this experiment. Gill condition did deteriorate in all cages in the autumn of 2019, with positive diagnoses of both amoebic gill disease and proliferative gill inflammation, but even though barriers remained in place it was condition factor, temperature and fish length which were the important determinants of disease severity.

Two manipulations within the dynamic strategy framework reduce the risk posed by barriers regarding gill related health concerns: (1) continuous use of aeration mixing devices to maintain water quality when barriers are in use, and (2) attraction to the surface when a brackish layer is present. The aeration devices used in this experiment do not work by directly increasing oxygen concentrations, but rather by

circulating deeper water into the barrier volume and generating vertical rather than horizontal water movement. There are pros and cons to this mode of aeration, evident in Fig. 2. On one hand the aeration devices not only maintain dissolved oxygen concentrations, but also overall water quality by creating circulation. On the other hand, however, such a strategy is unable to increase dissolved oxygen saturations beyond ambient levels, and as such does not entirely safeguard against poor oxygen conditions during periods when ambient oxygen levels are low. Additionally, when deeper temperatures are favourable mixing will improve feeding and growth, but when surface temperatures are favourable, such as during the period shown in Fig. 2, feeding may be lower than expected based on surface measurements. Regardless, these data clearly show, in agreement with previous work, that some form of supplemental aeration or oxygenation is required to maintain acceptable dissolved oxygen conditions within barriers, including even relatively shallow skirts. During the brief periods when the aeration devices were turned off dissolved oxygen saturation within the barriers rapidly dropped below 50% despite fully saturated ambient oxygen levels (Fig. 2).

Another benefit of the dynamic strategy relative to previous trials using similar tools was observed in cleanerfish performance. Bui et al. (2020) report that despite new louse infestations being reduced when continuously using a skirt barrier, cleaner fish, functional feed, deep feeding and deep lights, neither mobile lice numbers nor the number of required de-lousings differed from control cages. The authors hypothesized that because the cleaner fish hides were positioned within the barrier it was likely that chronic exposure to reduced oxygen concentrations led to poorer performance of cleaner fish, thus negating the reduction in louse attachment. Such a theory aligns with the findings of Gentry et al. (2020) in which corkwing wrasse in cages equipped with a barrier consumed one-ninth as many lice as those in cages without barriers. In this experiment, though stomach contents of cleaner fish were not checked, the observed reduction in louse infestations in dynamic cages carried through to reduced mobile lice numbers and fewer delousing events, suggesting that cleaner fish performance was not dramatically reduced in dynamic cages.

5. Conclusions

Farmed salmonids account for 99.6% of available hosts and produce 99.1% of adult female salmon lice present in Norwegian waters (Dempster et al., 2021). The only way to reduce the louse burden without reducing the number of available hosts is to reduce infestation success. While research continues in search of the perfect solution which negates the need for reactive delousing entirely, environmentally responsive, dynamic louse prevention offers a cost-effective, practical strategy which can significantly reduce the louse burden on farmed salmonids. Unlike other preventative strategies, implementation of a dynamic cage management strategy requires no specific conditions and can be deployed on almost any site where salmon are farmed. Here, when used only on half of the cages at a single site, the louse burden was reduced by 50%. If such a strategy were applied industry-wide, the cascading effects of significantly reduced louse infestation success and subsequent population reduction could radically reduce the louse burden on both farmed and wild salmonids.

Author statement

We have read through the comments from all 3 reviewers and revised the manuscript accordingly. A detailed outline of the changes made can be found in the attached 'Response to reviewers' file.

CRedit authorship contribution statement

Tina Oldham: Conceptualization, Methodology, Formal analysis, Investigation, Data curation, Writing – original draft, Visualization,

Project administration. **Benedicte Simensen:** Investigation, Resources, Data curation, Project administration, Writing – review & editing. **Henrik Trengereid:** Conceptualization, Resources, Project administration, Writing – review & editing. **Frode Oppedal:** Conceptualization, Methodology, Validation, Resources, Writing – review & editing, Supervision, Project administration, Funding acquisition.

Declaration of Competing Interest

Tina Oldham and Frode Oppedal declare that they have no known competing financial interests or personal relationships that could have influenced the work reported in this paper. Benedicte Simensen and Henrik Trengereid were employed by Mowi ASA at the time of the reported work. CAC was not involved in the analysis, interpretation or writing up of the results, and support the decision to submit the article for publication.

Data availability

Data will be made available on request.

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References

- Aaen, S.M., Helgesen, K.O., Bakke, M.J., Kaur, K., Horsberg, T.E., 2015. Drug resistance in sea lice: a threat to salmonid aquaculture. *Trends Parasitol.* 31, 72–81.
- Andrews, M., Horsberg, T.E., 2021. In vitro bioassay methods to test the efficacy of thermal treatment on the salmon louse, *Lepeophtheirus salmonis*. *Aquaculture* 532, 736013.
- Barrett, L.T., Oppedal, F., Robinson, N., Dempster, T., 2020. Prevention not cure: a review of methods to avoid sea lice infestations in salmon aquaculture. *Rev. Aquac.* 12 (4), 2527–2543. <https://doi.org/10.1111/raq.12456>.
- Bates, D., Mächler, M., Bolker, B., Walker, S., 2015. Fitting linear mixed-effects models using lme4. *J. Stat. Softw.* 67, 1–48.
- Besnier, F., Kent, M., Skern-Mauritzen, R., Lien, S., Malde, K., Edvardsen, R.B., Taylor, S., Ljungfeldt, L.E., Nilsen, F., Glover, K.A., 2014. Human-induced evolution caught in action: SNP-array reveals rapid amphiatlantic spread of pesticide resistance in the salmon ectoparasite *Lepeophtheirus salmonis*. *BMC Genomics* 15, 937.
- Bridle, A., Carter, C., Morrison, R., Nowak, B., 2005. The effect of beta-glucan administration on macrophage respiratory burst activity and Atlantic salmon, *Salmo salar* L., challenged with amoebic gill disease-evidence of inherent resistance. *J. Fish Dis.* 28, 347–356.
- Bromage, N., Porter, M., Randall, C., 2001. The environmental regulation of maturation in farmed finfish with special reference to the role of photoperiod and melatonin. In: Pages 63–98 *Reproductive Biotechnology in Finfish Aquaculture*. Elsevier.
- 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.
- Bui, S., Stien, L.H., Nilsson, J., Trengereid, H., Oppedal, F., 2020. Efficiency and welfare impact of long-term simultaneous in situ management strategies for salmon louse reduction in commercial sea cages. *Aquaculture* 520, 734934.
- Burnham, K.P., Anderson, D.R., 2002. *Model Selection and Multimodel Inference: A Practical Information Theoretic Approach*, 2.
- Crosbie, T., Wright, D., Oppedal, F., Johnsen, I., Samsing, F., Dempster, T., 2019. Effects of step salinity gradients on salmon lice larvae behaviour and dispersal. *Aquacult. Environ. Interact.* 11, 181–190.
- Dawood, M.A., Koshio, S., Esteban, M.Á., 2018. Beneficial roles of feed additives as immunostimulants in aquaculture: a review. *Rev. Aquac.* 10, 950–974.
- Dempster, T., Korsøen, O., Folkedal, O., Juell, J.-E., Oppedal, F., 2009. Submergence of Atlantic salmon (*Salmo salar* L.) in commercial scale sea-cages: a potential short-term solution to poor surface conditions. *Aquaculture* 288, 254–263.
- Dempster, T., Overton, K., Bui, S., Stien, L.H., Oppedal, F., Orjan Karlsen, A., Coates, B., Phillips, Barrett, L.T., 2021. Farmed salmonids drive the abundance, ecology and evolution of parasitic salmon lice in Norway. *Aquacult. Environ. Interact.* 13, 237–248.
- Fjørtoft, H.B., Nilsen, F., Besnier, F., Stene, A., Bjørn, P.A., Tveten, A.-K., Aspehaug, V.T., Finstad, B., Glover, K.A., 2019. Salmon lice sampled from wild Atlantic salmon and sea trout throughout Norway display high frequencies of the genotype associated with pyrethroid resistance. *Aquacult. Environ. Interact.* 11, 459–468.

- Fernö, A., Huse, I., Juell, J.E., Bjordal, Å., 1995. Vertical distribution of Atlantic salmon (*Salmo solar* L.) in net pens: trade-off between surface light avoidance and food attraction. *Aquaculture* 132, 285–296.
- Fjortoft, H.B., Nilsen, F., Besnier, F., Espedal, P.G., Stene, A., Tveten, A.-K., Bjørn, P.A., Aspehaug, V.T., Glover, K.A., 2020. Aquaculture-driven evolution: distribution of pyrethroid resistance in the salmon louse throughout the North Atlantic in the years 2000–2017. *ICES J. Mar. Sci.* 77, 1806–1815.
- Fjortoft, H.B., Nilsen, F., Besnier, F., Stene, A., Tveten, A.-K., Bjørn, P.A., Aspehaug, V.T., Glover, K.A., 2021. Losing the “arms race”: multiresistant salmon lice are dispersed throughout the North Atlantic Ocean. *R. Soc. Open Sci.* 8, 210265.
- Frank, K., Gansel, L., Lien, A., Birkevold, J., 2015. Effects of a shielding skirt for prevention of sea lice on the flow past stocked salmon fish cages. *J. Offshore Mech. Arctic Eng.* 137, 011201.
- Frenzl, B., Stien, L., Cockerill, D., Oppedal, F., Richards, R., Shinn, A., Bron, J., 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, 183–188.
- Geitung, L., Oppedal, F., Stien, L.H., Dempster, T., Karlsbakk, E., Nola, V., Wright, D.W., 2019. Snorkel sea-cage technology decreases salmon louse infestation by 75% in a full-cycle commercial test. *Int. J. Parasitol.* 49 (11), 843–846.
- Gentry, K., Bui, S., Oppedal, F., Dempster, T., 2020. Sea lice prevention strategies affect cleaner fish delousing efficacy in commercial Atlantic salmon sea cages. *Aquacult. Environ. Interact.* 12, 67–80.
- 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., Laurin, E., Stormoen, M., Sanchez, J., Fast, M.D., Revie, C.W., 2019. Evaluating the potential for sea lice to evolve freshwater tolerance as a consequence of freshwater treatments in salmon aquaculture. *Aquacult. Environ. Interact.* 11, 507–519.
- 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.
- Guiragain, P., Tkachov, M., Båtnes, A.S., Olsen, Y., Winge, P., Bones, A.M., 2021. Principles and methods of counteracting harmful Salmon-arthropod interactions in Salmon farming: addressing possibilities, limitations, and future options. *Front. Mar. Sci.* 9, 65.
- Hamre, L.A., Eichner, C., Caipang, C.M.A., Dalvin, S.T., Bron, J.E., Nilsen, F., Boxshall, G., Skern-Mauritzen, R., 2013. The Salmon louse *Lepeophtheirus salmonis* (Copepoda: Caligidae) life cycle has only two Chalimus stages. *PLoS One* 8, e73539.
- Hansen, T., Stefansson, S., Taranger, G., 1992. Growth and sexual maturation in Atlantic salmon, *Salmo salar* L., reared in sea cages at two different light regimes. *Aquac. Res.* 23, 275–280.
- Hansen, T.J., Fjellidal, P.G., Folkedal, O., Vågseth, T., Oppedal, F., 2017. Effects of light source and intensity on sexual maturation, growth and swimming behaviour of Atlantic salmon in sea cages. *Aquacult. Environ. Interact.* 9, 193–204.
- Helgesen, K.O., Romstad, H., Aaen, S.M., Horsberg, T.E., 2015. First report of reduced sensitivity towards hydrogen peroxide found in the salmon louse *Lepeophtheirus salmonis* in Norway. *Aquacult. Rep.* 1, 37–42.
- 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.
- Hevroy, 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.
- Jeong, J., Stormoen, M., McEwan, G.F., Thakur, K.K., Revie, C.W., 2021. Salmon lice should be managed before they attach to salmon: exploring epidemiological factors affecting *Lepeophtheirus salmonis* abundance on salmon farms. *Aquaculture* 541, 736792.
- Johansson, D., Juell, J.-E., Oppedal, F., Stiansen, J.-E., Ruohonen, K., 2007. The influence of the pycnocline and cage resistance on current flow, oxygen flux and swimming behaviour of Atlantic salmon (*Salmo salar* L.) in production cages. *Aquaculture* 265, 271–287.
- Jones, M., Sommerville, C., Wootton, R., 1992. Reduced sensitivity of the salmon louse, *Lepeophtheirus salmonis*, to the organophosphate dichlorvos. *J. Fish Dis.* 15, 197–202.
- Jónsdóttir, K.E., Volent, Z., Alfreðsen, J.A., 2020. Dynamics of dissolved oxygen inside salmon sea-cages with lice shielding skirts at two hydrographically different sites. *Aquacult. Environ. Interact.* 12, 559–570.
- Jónsdóttir, K.E., Klebert, P., Volent, Z., Alfreðsen, J.A., 2021. Characteristic current flow through a stocked conical sea-cage with permeable lice shielding skirt. *Ocean Eng.* 223, 108639.
- Juell, J.E., Fernö, A., Furevik, D., Huse, I., 1994. Influence of hunger level and food availability on the spatial distribution of Atlantic salmon, *Salmo salar* L., in sea cages. *Aquac. Res.* 25, 439–451.
- Juell, J.-E., Fosseidengen, J.E., 2004. Use of artificial light to control swimming depth and fish density of Atlantic salmon (*Salmo salar*) in production cages. *Aquaculture* 233, 269–282.
- Juell, J.-E., Oppedal, F., Boxaspen, K., Taranger, G.L., 2003. Submerged light increases swimming depth and reduces fish density of Atlantic salmon *Salmo salar* L. in production cages. *Aquac. Res.* 34, 469–478.
- Ljungfeldt, L.E.R., Espedal, P.G., Nilsen, F., Skern-Mauritzen, M., Glover, K.A., 2014. A common-garden experiment to quantify evolutionary processes in copepods: the case of emamectin benzoate resistance in the parasitic sea louse *Lepeophtheirus salmonis*. *BMC Evol. Biol.* 14, 108.
- Ljungfeldt, L.E.R., Quintela, M., Besnier, F., Nilsen, F., Glover, K.A., 2017. A pedigree-based experiment reveals variation in salinity and thermal tolerance in the salmon louse, *Lepeophtheirus salmonis*. *Evol. Appl.* 10, 1007–1019.
- Mennerat, A., Hamre, L., Ebert, D., Nilsen, F., Dávidová, M., Skorpjng, A., 2012. Life history and virulence are linked in the ectoparasitic salmon louse *Lepeophtheirus salmonis*. *J. Evol. Biol.* 25, 856–861.
- Migaud, H., Cowan, M., Taylor, J., Ferguson, H.W., 2007. The effect of spectral composition and light intensity on melatonin, stress and retinal damage in post-smolt Atlantic salmon, *Salmo salar*. *Aquaculture* 270, 390–404.
- Mullins, J., Nowak, B., Leef, M., Oyvind Røn, T.B., Eriksen, and C. McGurk., 2020. Functional diets improve survival and physiological response of Atlantic salmon (*Salmo salar*) to amoebic gill disease. *J. World Aquacult. Soc.* 51 (3), 634–648.
- Myhre Jensen, E., Horsberg, T.E., Sevatdal, S., Helgesen, K.O., 2020. Trends in de-lousing of Norwegian farmed salmon from 2000–2019—consumption of medicines, salmon louse resistance and non-medical control methods. *PLoS One* 15, e0240894.
- Nilsson, J., Stien, L., Oppedal, F., 2017. Environment, lice levels, welfare and salmon swim depth at Kobbavika site with surface or deep feeding combined with artificial light. In: Havforskningstutttet Internale Report.
- Noble, C., Gismervik, K., Iversen, M.H., Kolarovic, J., Nilsson, J., Stien, L.H., Turnbull, J. F., 2018. Welfare Indicators for Farmed Atlantic Salmon: Tools for Assessing Fish Welfare.
- Oldham, T., Dempster, T., Fosse, J.O., Oppedal, F., 2017. Oxygen gradients affect behavior of caged Atlantic salmon *Salmo salar*. *Aquaculture Environment Interactions* 9, 145–153.
- Oldham, T., Dempster, T., Crosbie, P., Adams, M., Nowak, B., 2020. Cyclic hypoxia exposure accelerates the progression of amoebic gill disease. *Pathogens* 9, 597.
- Oppedal, F., Taranger, G.L., Juell, J.-E., Fosseidengen, J.E., Hansen, T., 1997. Light intensity affects growth and sexual maturation of Atlantic salmon (*Salmo salar*) postsmolts in sea cages. *Aquat. Living Resour.* 10, 351–357.
- Oppedal, F., Berg, A., Olsen, R.E., Taranger, G.L., Hansen, T., 2006. Photoperiod in seawater influence seasonal growth and chemical composition in autumn sea-transferred Atlantic salmon (*Salmo salar* L.) given two vaccines. *Aquaculture* 254, 396–410.
- 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.
- Oppedal, F., Bui, S., Stien, L.H., Overton, K., Dempster, T., 2019. Snorkel technology to reduce sea lice infestations: efficacy depends on salinity at the farm site, but snorkels have minimal effects on salmon production and welfare. *Aquacult. Environ. Interact.* 11, 445–457.
- Overton, K., Dempster, T., Oppedal, F., Kristiansen, T.S., Gismervik, K., Stien, L.H., 2018. Salmon lice treatments and salmon mortality in Norwegian aquaculture: a review. *Rev. Aquac.* 11 (4), 1398–1417.
- Powell, M.D., Ransome, J., Barney, M., Duijff, R.M., Flik, G., 2007. Effect of dietary inclusion of N-acetyl cysteine on mucus viscosity and susceptibility of rainbow trout, *Oncorhynchus mykiss*, and Atlantic Salmon, *Salmo salar*, to amoebic gill disease. *J. World Aquacult. Soc.* 38, 435–442.
- R Core Team, 2018. R: A language and environment for statistical computing. Foundation for Statistical Computing, Vienna, Austria. <https://www.R-project.org/>.
- Ricker, W.E., 1975. Computation and interpretation of biological statistics of fish populations. *Bull. Fish. Res. Bd. Can.* 191, 1–382.
- Samsing, F., Johnsen, I., Stien, L.H., Oppedal, F., Albretsen, J., Asplin, L., Dempster, T., 2016. Predicting the effectiveness of depth-based technologies to prevent salmon lice infection using a dispersal model. *Prev. Vet. Med.* 129, 48–57.
- Stien, L.H., Nilsson, J., Hevroy, E.M., Oppedal, F., Kristiansen, T.S., Lien, A.M., Folkedal, O., 2012. Skirt around a salmon sea cage to reduce infestation of salmon lice resulted in low oxygen levels. *Aquac. Eng.* 51, 21–25.
- 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.
- Tacchi, L., Bickerdike, R., Douglas, A., Secombes, C.J., Martin, S.A., 2011. Transcriptomic responses to functional feeds in Atlantic salmon (*Salmo salar*). *Fish & Shellfish Immunol.* 31, 704–715.
- Taranger, G.L., Haux, C., Hansen, T., Stefansson, S.O., Björnsson, B.T., Walther, B.T., Kryvi, H., 1999. Mechanisms underlying photoperiodic effects on age at sexual maturity in Atlantic salmon, *Salmo salar*. *Aquaculture* 177, 47–60.
- Taylor, R., Huynh, C., Cameron, D., Evans, B., Cook, M., Ritchie, G., Asa, H., 2016. Gill Score Guide-Amoebic Gill Disease (AGD) Management Training Document.
- Vigen, J., 2008. Oxygen Variation within a Seacage. Master’s thesis. University of Bergen, Norway.
- Warren-Myers, F., Vågseth, T., Folkedal, O., Stien, L.H., Fosse, J.O., Dempster, T., Oppedal, F., 2022. Full production cycle, commercial scale culture of salmon in submerged sea-cages with air domes reduces lice infestation, but creates production and welfare challenges. *Aquaculture* 548, 737570.
- Wood, S.N., 2011. Fast stable restricted maximum likelihood and marginal likelihood estimation of semiparametric generalized linear models. *J. Royal Stat. Soc. Ser. B (Stat. Methodol.)* 73, 3–36.
- Wood, S.N., 2017. Generalized Additive Models: An Introduction with R, 2nd edition. Chapman and Hall/CRC.

- Wright, D.W., Glaropoulos, A., Solstorm, D., Stien, L.H., Oppedal, F., 2015. Atlantic salmon *Salmo salar* instantaneously follow vertical light movements in sea cages. *Aquacult. Environ. Interact.* 7, 61–65.
- Wright, D., Stien, L., 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. *Aquacult. Environ. Interact.* 10, 255–265.
- Zuur, A.F., Ieno, E.N., Elphick, C.S., 2010. A protocol for data exploration to avoid common statistical problems. *Methods in Ecol. Evol.* 1, 3–14.