Ecological Effects of Blue LED Lights Used in Aquaculture

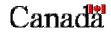
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ECOLOGICAL EFFECTS OF BLUE LED LIGHTS USED IN AQUACULTURE

by

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ABSTRACT

Stewart, H.L., Nomura, M., Piercey, G.E., Dunham, A. and Lelliott, T.L. 2013. Ecological effects of blue LED lights used in aquaculture. Can. Tech. Rep. Fish. Aquat. Sci. 3057: iv + 26 p.

Specific wavelength light emitting diodes (LEDs) are a technology increasingly used in finfish farms to delay grilsing and increase production of finfish, although their potential ecological impacts to adjacent habitats are not well understood. We investigated the ecological effects of blue (450 nm) LED lights in an experimental setting away from a finfish operation bi-weekly for eight months in 2011-2012. Our findings show that, similar to reports examining the effects of white metal halide lights, blue LEDs attract fishes, zoo- and phytoplankton, compared to ambient (no artificial light) conditions. No effect of blue lights, however, was found for the rate of settlement or species composition of sessile organisms adjacent to the lights. Preliminary data collected at a fish farm site suggested that blue LEDs did not delay grilsing as expected, and may have resulted in higher sea lice loads on salmon, although due to small sample size these results were not statistically significant.

RÉSUMÉ

Stewart, H.L., Nomura, M., Piercey, G.E., Dunham, A. et Lelliott, T.L. 2013. Effects écologiques des ampoules à DEL utilisées en aquaculture. Can. Tech. Rep. Fish. Aquat. Sci. 3057: iv + 26 p.

Des diodes électroluminescentes (DEL) à longueur d'onde spécifique sont de plus en plus utilisées dans les centres de pisciculture marine pour retarder la maturation précoce et augmenter le production, bien que les répercussions possibles sur l'écologie des habitats adjacents ne soient pas bien comprises. Nous avons étudié les effets écologiques de DEL bleues (450 nm) dans un milieu expérimental à distance d'une pisciculture marine deux fois par semaine pendant huit mois en 2011-2012. Selon nos conclusions, qui ressemblent à celles des rapports concernant les luminaires à halogénures métalliques blancs, les DEL bleues attirent les poissons, le zooplancton et le phytoplancton, comparé aux conditions ambiantes (pas d'éclairage artificiel). Toutefois, aucun effet de l'éclairage bleu n'a été constaté pour l'accumulation de compositions d'espèces d'organismes sessiles près des luminaires. Les données préliminaires recueillies au centre de pisciculture marine suggèrent que les DEL bleues n'ont pas retardé la maturation précoce comme on le pensait, et ont pu donner lieu à des charges plus élevées de pou du poisson sur les saumons, bien que, en raison de l'échantillon peu nombreux, ces résultats n'étaient pas statistiquement significatifs.

INTRODUCTION

In the past century, the extent and intensity of artificial night lighting has increased significantly both in terrestrial and aquatic ecosystems (Longcore and Rich 2004). In the marine environment, sources of artificial light include lighted fishing fleets, offshore oil platforms, and coastal man-made structures. More recently, artificial night lighting has been adopted by marine finfish aquaculture facilities. Northern hemisphere countries that employ continuous lighting around finfish aquaculture cages include Norway, Scotland, Ireland, Iceland and Canada. By artificially extending the photoperiod through the use of lights, the ability of fish to recognize shortening day length is masked, the physiological processes that initiate gonad development and gametogenesis are delayed, and growth rate is maintained (Davie et al. 2007, Harmon et al. 2003, Porter et al. 1999, Taranger et al. 2006) resulting in economic benefits that include higher yield and better quality product compared to that produced under natural maturation processes (Endal et al. 2000, Leclercq et al. 2010).

Alteration in light is one of the stressors associated with aquaculture that may impact aquatic ecosystems via their effect on non-target organisms. These affects may be dependent on the type and intensity of the light, and may be species specific (Fermin and Seronay 1997, Keenan et al. 2007, McConnell et al. 2010, Wickham 1973, Purvis et al. 1985). For example, in a study on the effects of hydroelectric dams on out-migrating salmonids, juvenile chinook and coho salmon were found to avoid strobe and fullintensity constant mercury lights, but chinook salmon exhibited an attraction to dim mercury light (Nemeth and Anderson 1992). Low irradiance light emitting diodes (LEDs) have been shown to enhance coral growth in coral aguaculture (Wijgerde and Laterveer 2013), and zooplankton abundance was increased in the field by blue, green and violet LEDs, but not by amber LEDs in a tropical lagoon (Alldredge et al. 2013). Studies have suggested that rainbow trout (Onchorhynchus mykiss) and sea bass (Dicentrarchus *labrax*) are most sensitive to wavelengths peaking at 450–500 nm (Max and Menaker 1992, Bayarri et al. 2002). McConnell et al. (2010) found that certain gastropod and bivalve invertebrate larvae, and pacific herring, sand lance, stickleback and some species of sculpins were attracted to white metal halide lights used in salmon farming in British Columbia.

White metal halide lights are routinely used in finfish aquaculture (Hay et al. 2004). These involve high operating costs and are comprised of many wavelengths including longer wavelength yellow-red light which are rapidly absorbed in the water column and not detected by fish (Loew and McFarland 1990, Migaud et al. 2006). Recently, the aquaculture industry has begun using new technologies, specifically blue light (450 nm) emitting diode (LED) lights in finfish aquaculture. Blue-green spectrum light is more suitable for use in seawater as these wavelengths have higher energy content and have been reported to penetrate up to 28 m through clear distilled water (Duntley 1963). LED lights can also be tuned to specific wavelengths, making them more speciesspecific than white lights. Economically, LED lights are relatively energy efficient, having lower power requirements, electrical operating costs and a longer life span than standard metal halide bulbs. However, their efficacy in delaying maturation and increasing fish growth rates, and their biological impact to the surrounding habitat is not fully understood.

The goal of this project was to determine the pathways of biological effects of blue LED grow lights that are currently in use in open pen finfish aquaculture. We evaluated, bimonthly, from October 2011-May 2012, the biological impacts of blue LED lights relative to unlit controls to evaluate specifically their effects on 1) local small and juvenile fish abundance, 2) zoo- and phytoplankton composition and abundance, and 3) settlement and species composition of sessile invertebrate communities. We also quantified photosynthetically active radiation light profiles around blue LED lights. In partnership with Grieg Seafood BC Ltd., preliminary data were also collected to determine the efficacy of these blue LED lights on growth and maturation of Atlantic salmon in aquaculture net pens, and to determine their effect on sea lice load.

MATERIALS AND METHODS

Blue light emitting diode (LED) lights

Six Idema underwater lights, model Blue LED 100W by AqvaSmart (AKVA Group North America), were used for this experiment. Each light consisted of blue light emitting diodes (LEDs) arranged in rows and encased in a borosilicate sheath filled with oil for temperature regulation.

Site and setup

The experiment was conducted on the floating dock at the West Vancouver Laboratory, Fisheries and Oceans Canada's Centre for Aquaculture and Environmental Research (CAER), West Vancouver, British Columbia. Lights were oriented as shown in Figure 1, and deployed on the dock as illustrated in Figure 2. Six blue light units were suspended on 3/8" galvanized chains that were shackled to the dock railing. Each light unit also had an artificial substrate apparatus, consisting of four clear plastic Petri dishes attached to a pail lid with cable ties and suspended 20 cm below the bottom of the light, along with a 2.3 to 4.5 kg lead weight to keep the apparatus upright and stable in the water column (Figure 1).

Lights were spaced 10 m apart, which was determined to be further than the penetration of the lights as measured as photosynthetically active radiation (PAR) (see Figure 3) and Light Attenuation section below). Our spacing of 10 meters was also chosen as it is 4x further than the distance of light attenuation of 50W blue LED light (2.5 m) in seawater to the threshold of effective irradiance (0.016 W m⁻² (Migaud et al. 2006) for Atlantic salmon as determined by Leclercq et al. (2011)). Effective irradiance is the threshold above which circadian melatonin production is suppressed to the extent that artificial light is perceived as daylight and the physiological processes that promote maturation are inhibited. Our spacing of 10 m was also longer than distances (2.5-3.5 m) away from 100W blue LED lights that Migaud et al. (2007) characterized as 'low-intensity,' and also further than the distance (0.9 m) over which light attenuation of

100W blue LED lights was reduced to 50% of initial intensities through seawater (Migaud et al. (2007).

The lights were set up with a timer so that they came on 15 minutes before sunset and shut off 15 minutes after sunrise, and this was adjusted every two weeks. Of the six lights, only three were on at any one time, and the treatments alternated light position (for example, #1, #3, #5 were on (Lit) and #2, #4, #6 off (Control), Figure 2). The sequence of lights that were lit and control was changed every three months. At this time, the artificial substrate apparatus was moved by one light counterclockwise so that each particular artificial substrate apparatus was continuously exposed to the same lighting conditions (either Lit or Control). The light apparatus was also scrubbed every two weeks. Prior to deployment, the cap portion of the lights were wrapped in black electrical tape to facilitate cleaning at the end of the deployment.

Sampling schedule

Sampling took place every two weeks from October 2011 to May 2012. On sampling days, the following measurements were made: turbidity, light attenuation, fish samples via minnow traps, and plankton tows adjacent to each light. One substrate sampling plate was collected from each apparatus every three months. See details below.

Turbidity and light attenuation

Water column turbidity was measured using a Secchi disk in the water adjacent to Light #2, at approximately noon on each sampling day. A measurement was taken on the way down and on the way up, and the average was used to calculate an attenuation coefficient using:

Equation 1: Attenuation Coefficient = (1.7 / Secchi depth) (Idso and Gilbert 1974)

Light penetration from the LED light through the water column at night was measured as photosynthetically active radiation (PAR) in µmol s⁻¹ m⁻² using a Li-Cor Spherical Quantum Sensor and 1400 Datalogger (Li-Cor Instruments, Nebraska, USA). PAR sensors can underestimate the response of particular wavelengths that they are not specifically tuned to, and although this sensor attempts to have an equal response to all photons in the 400-700 nm wavelength range, it has a less than 100% relative response to blue light (<u>http://www.licor.com/env/products/light/quantum_sensors/index.html</u>). As such, the field of light that we measured was the distance over which photosynthetically active radiation could be measured by this sensor, and is biologically relevant for photosynthetic responses of phytoplankton that may have been in the vicinity of the light and potentially drawn to it.

The sensor was fixed to a telescoping boat hook, and was used to take measurements at either Light #1 or Light #2, whichever was on at the time of sampling. The first measurement was made at the surface, away from any external light, with the sensor facing up in order to establish baseline irradiance. The 0 m measurement was made with the sensor touching the light at the depth of the mid-portion of the blue light apparatus. The procedure was repeated at one meter distances up to 10 meters

horizontally away from the light. Ten replicates of the light reading were recorded at each distance.

Plankton

The plankton community around each light was sampled at night between 4-7 hours after sunset. Collections were made adjacent to each light using a 100 micron plankton net of 150 cm length, with a round mouth of 50 cm in diameter. The cod end of the net was approximately 10 cm in diameter. The plankton net was carefully lowered so as not to touch the light or the sides of the dock to 5 m depth. Once the net was raised the contents of the net were rinsed into the collecting end and transferred into a pre-labeled 500 ml sample jar. Seawater filtered through a 63 micron mesh was used to rinse out any remaining organisms into the sample jar. The samples were then preserved by adding enough formalin to equal 10% formalin and split using a plankton splitter. Half of each plankton sample was sent to EcoAnalysts Inc. (Moscow, Idaho, USA) where all zooplankton and phytoplankton were identified and counted.

Fish

Collapsible minnow traps measuring 26 cm x 26 cm x 44 cm with 0.3 cm square mesh were used to sample small and juvenile fish in the vicinity of the lights. The bait pouch in each trap was filled with 50 g of 1/4 inch BioBrood fish pellets then suspended from the railing at the depth of the light, with a small lead weight at one end to keep the trap upright in the water column. One trap was deployed adjacent to each of the six lights approximately 30 minutes before sunset, then retrieved 15 to 30 minutes before sunrise. Upon retrieval, the contents were identified, counted, photographed and released. Individuals that could not be identified on site were preserved in 10% formalin in a 500 ml sample jar for subsequent processing.

Settlement

To determine if the blue LED lights had any effect on recruitment and settlement of sessile organisms, four plastic substrate plates were hung from the bottom of each light (see Figure 1). One substrate plate from each light apparatus was removed for sampling every three months. Plates from the same position were taken each time, starting with the northeast quadrat and working counterclockwise. Plates were removed and rinsed with filtered sea water to remove any loose organisms, then placed in individual jars and preserved in 10% formalin then transferred to 70% ethanol until analysis. Photographs were taken with a Pentax W90 digital camera on both black and white backgrounds and images were later analyzed for percent coverage of organisms using ImageJ software (version 1.47, National Institute of Health USA).

Effects of blue LED lights on salmon maturity, growth and sea lice load

In conjunction with the experiment to determine the effects of blue LED lights on the biology of the water column described above, data was also collected at farm sites operated by Grieg Seafood BC Ltd. (Figure 5) to determine the effect of blue LED lights on growth, maturity and sea lice (*Lepeophtheirus salmonis* and *Caligus clemensi*) load of Atlantic salmon, compared to 1000W white metal halide lights.

Approximately 470,000 smolted salmon from the same brood, average mass 78.2 grams were split into two groups after an initial period in pens exposed to white metal halide lights. Half of the group remained exposed to metal halide lights (Culloden site), and the other half was transferred to an adjacent site but exposed to blue LED lights (Ahlstrom). Each pen was 30.5 m square, and a light was suspended at a depth of 8 m in the centre of each 15.25 m square quadrant, so that each pen had four lights spaced evenly in a grid formation. Harvest began on July 7, 2011 at Culloden and August 4, 2011 at Ahlstrom, and continued alternately at each site until January 31, 2012. The sexual maturity, weight and sea lice load of each fish was recorded.

Analyses

Statistical analyses and graph production were done using statistical program R (version 2.15.0). Details of statistical analyses for each dataset can be found in the corresponding results section.

RESULTS

Turbidity

Turbidity measurements were converted into attenuation coefficients, and grouped by season as shown in Figure 4. A one-way ANOVA was used to test for differences among the three seasons, and we found a statistically significant difference between them (F = 8.03, p = 0.005). Tukey post-hoc comparisons indicate that spring (mean = 0.58, SD = 0.14) had significantly higher turbidity than either fall (mean = 0.40, SD = 0.60, p = 0.01) or winter (mean = 0.38, SD = 0.03, p = .008), which were not statistically different from one another (p = 0.92)

Light penetration through water column

Light measurements taken at set distances from a lit blue 100W LED light at night were converted into proportions of the measurement at 0 m (directly adjacent to the light) (Figure 3). These were then grouped into seasons as we did for turbidity. A Friedman Rank Sum Test showed that there was no significant effect of season on light intensity at any distance from the light source, although we did see a trend of reduced light penetration through the water column during winter and spring.

<u>Plankton</u>

The median counts of total zooplankton caught during the plankton tows for lit and control treatments were 3867 and 3140, respectively. As a Shapiro-Wilks Test showed that the data were not normally distributed, a Wilcoxon Signed-rank Test was conducted to test for differences in zooplankton abundance between treatments across dates. This showed that there was significantly more zooplankton caught around lit LED lights, than around the controls (W = 36, Z = 2.52, p =0.008, r = 0.63, Figure 6a).

Crustaceans comprised 97% of zooplankton in each tow and 95% of that was copepods (Figure 6b). When we examined the effect of light on non-crustacean zooplankton alone median counts for lit and control were 72 and 39, respectively, and a Wilcoxon Signed-

rank Test showed that there was a small but significant positive effect of blue LED lights on non-crustacean zooplankton abundance as well (W = 3, Z = -2.10, p = 0.04, r = 0.46). Details of the zooplankton collected in samples are contained in Table 1.

A statistical analysis of phytoplankton abundance between lit and unlit controls across all dates did not indicate significant differences (W = 21, Z = 0.42, p = 0.74, r = 0.11), likely due to our small sample size and the seasonality of the distribution of phytoplankton in our tows. However, in April and May substantially more phytoplankton was collected around the lit treatments (Figure 7).

Fish

There was an obvious seasonality in the species and abundance of fish at the sample site (Figure 8), and more fish were caught around blue LED lights than at unlit control lights (Wilcoxon Signed-rank Test W = 120, Z = 3.41, p = 6.1×10^{-5} , r = 0.62). Sticklebacks were the most abundant fish caught in our minnow traps throughout the study, except for one sampling event (April 12 – data not shown on graphic) when they were outnumbered by 36 juvenile pink salmon. Pink salmon were not found on any other sampling event.

Settlement of sessile organisms

There was very little diversity of organisms that settled on our settlement plates. A film of diatoms appeared in March 2012, followed by barnacles in April 2012. By the end of the experiment in May 2012, settlement plates were almost completely covered by barnacles, which were covered by diatoms (producing a cover of over 100% in the final plate collection) (Figure 9). A few solitary mussels were found on the settlement plates in May 2012. A Wilcoxon Signed-rank Test indicated that there were no statistically significant settlement differences for plates exposed to blue lights or unlit controls for either diatoms (W = 0, Z = -1.61, p = 0.25, r = 0.65) or barnacles (W = 2, Z = 0.45, p = 1, r = 0.22).

Effect of blue LED lights on farmed Atlantic salmon

We obtained data from Grieg Seafood on fish growth and maturity, as well as sea lice counts, for Atlantic salmon at two of their farm sites using different light types. The Ahlstrom site (equipped with blue LEDs) appeared to have a very high percentage of 'downgraded' fish – meaning that sexual maturity had not been delayed as desired by the producers. Statistical comparisons between the Ahlstrom site (blue LED) and the Culloden site (white metal halide site) are not possible for logistical reasons as the fish were not harvested on the same dates. Sea lice counts per fish at each farm site between November 2010 and November 2011 were higher for fish exposed to blue LED light at the Ahlstrom site (Figure 10), but statistical analysis was not possible on these data due to small sample size and the fact that these two sites, although fairly close to one another, vary in many physical and biological factors that were not accounted for in this study.

DISCUSSION

Our results indicate that the blue LED 100W lights that we examined attract fish, and zoo- and phytoplankton when blooms of these organisms occur. Our findings are similar to those by McConnell et al. (2010) on the effect of white metal halide lights in British Columbia, who reported increased abundance of larval, juvenile and adult fish, and zooplankton in the vicinity of lights, when compared to unlit controls.

The most dramatic result in our study is that fish were virtually non-existent in our sampling around unlit control lights, while many fish were sampled around the lit blue LED lights, primarily stickleback and one occurrence of many pink salmon, which were likely on migration. The high numbers found in our traps may be explained by the schooling behaviour of stickleback, and their apparent attraction to light, as has been shown for Pacific (*Clupea pallasi*) and Atlantic herring (*C. harengus*) (Blaxter and Batty 1990). McConnell et al. (2010) also recorded higher numbers of sticklebacks, in addition to sand lance (*Ammodytes* spp.) and sculpins around white metal halide lights.

A concern with the attraction of wild organisms drawn to the vicinity of the grow lights is that they will be consumed by captive fish. Even organisms that are not drawn to the light will be illuminated in its presence and more visible to predators (Blaxter and Batty 1990). We were not able to sample farmed salmon gut contents in this study, but Hay et al. (2004) found little evidence of wild organisms in the stomachs of farmed salmon, concluding that lights had no apparent effect on the consumption of wild food.

In our study, we noted a seasonal difference in plankton abundance, with spring blooms occurring in March-May, resulting in a corresponding increase in water column turbidity, as expected. Phytoplankton abundance was higher at lit treatments during the bloom in April and May, but very low during the other sampling times. Crustaceans, primarily copepods, were the most abundant plankter to be attracted to the blue LED lights across all sampling times, but were three times more abundant during spring. This differs from samples collected around white metal halide lights for which larval gastropods and bivalves dominated samples and copepods were not a major component (McConnell et al. 2010). This difference may be explained by the reported photophobic response of copepods that may be wavelength specific (Buskey et al. 1987), and light intensity as zooplankton have been shown to be attracted to low light intensities (Forward 1988). Our 100W blue LEDs were much less intense than the 400W metal halide lights used by McConnell et al. (2010). This high abundance of copepods, which are an important prey item for many fish, may in part explain the abundance of fish collected around our lit treatments, as they may have taken advantage of a cascading trophic subsidy that may have been based on zooplankton responding to increased phytoplankton around the lit treatments.

We found some adult harpacticoid copepods in our plankton samples, while these organisms, in adult form, are generally associated with the benthos (Bell et al. 1987). Given our experimental set-up, lights hung off a floating dock in relatively shallow water (4-8 m), it is possible our samples could include some benthic species stirred up by

wind events. These species would not be indicative of the organisms attracted to aquaculture finfish lights, which are deployed in much deeper water. However, harpacticoid copepods are not the bulk of the copepod sample, and the patterns we found are not driven by inclusion of benthic species.

Despite the differences we found in zoo- and phytoplankton around our lit and control lights, we did not see a difference in the species or rate of colonization of sessile organisms on the settlement plates. Diversity was low on these plates. They were initially colonized by films of diatoms, followed by barnacles, and the % cover of these was similar between treatments. By the end of our sampling, plates were covered with barnacles, which were coated by a thin layer of diatoms. It is noteworthy that, despite this evidence of arrival and settlement of barnacles on our plates, we did not have barnacle larvae in any of our plankton tows, at any sampling time, potentially a result of timing offset between barnacle settlement processes and our night time sampling. Shanks (1986) found that barnacle settlement was correlated with maximum daily tidal range at lags of +1 to +4 days; peak settling occurring several days before the spring tide. As our sampling was not based on moon phase, but rather a two week time interval, we could easily have missed such moon-phase settlement pulses.

Concern about the attraction of invertebrates to grow lights includes the possibility of an increase in the number of sea lice into pens and onto farmed salmon (Hevrøy et al. 2003), and that these may be transferred to migrating wild fish that pass by the open net farm pens, and ultimately impact wild stocks (Krkošek et al. 2005). We did not find sea lice larvae in our samples, although we did encounter an abundance of sticklebacks which are known to be hosts to both *Caligus* sp. and *Lepeiophtheirus* sp. (Jones et al. 2006). Data collected by Grieg Seafood at two of their finfish facilities indicate that sea lice load was slightly higher on salmon exposed to blue LED lights compared to fish exposed to white metal halide lights, although the data are limited and did not allow for statistical analysis.

Maturation data collected by Grieg Seafood BC Ltd. indicate that the blue 100W LED lights used in the salmon farm did not delay maturation or increase growth rates as expected, and this particular aquaculture operation has ceased to use them after this study. AKVA 100W blue LED lights are recommended by their manufacturer for tank use, not larger sea pen use. The model of this light recommended for larger net pens is a 360W model, which would be expected to have a larger light field and a potentially correspondingly larger biological impact. Migaud et al. (2007) report supressed levels of melatonin in post-smolt Atlantic salmon when exposed to a range of intensities of blue LED light, indicating that they can perceive this light even at low levels. A study by Leclercg et al. (2011) found that while exposure to 50W blue LED lights did suppress melatonin production in Atlantic salmon at high and low intensities tested, it failed to reduce maturation rates at low intensities relative to natural ambient light. Leclercg et al. (2011) note that the intensity of artificial light is the main light property influencing biological potency, and is more important than the light spectrum of the light, suggesting that appropriate tuning of the arrangement, intensity and schedule of light regime to the sensitivities and behaviour of target fish may determine their success (Trippel 2010).

Factors such as turbidity, ambient light levels, depth, and the size and arrangement of the artificial lights in pens also interact to affect the light field created by these lights and may present additional challenges with seasonal fluctuations. As LED technology is a relatively new application for finfish aquaculture, the particulars of the most appropriate methodologies are still being researched. Blue LED lights are in use in other operations in British Columbia.

For logistical reasons, our study was not conducted at an open pen finfish facility; it enabled us to isolate and examine the ecological implications of the lights themselves, without potential confounding effects associated with a farm. As our lit and unlit control treatments both used lights hung in identical configurations, and because we rotated the treatments throughout our study, we can be confident that our results are due to the effects of the blue LED lights and not due to the dock itself, light location, currents or other biological or physical factors. Increases in pelagic and demersal fish around finfish aquaculture operations (Dempster et al. 2002) have been attributed to availability of excess feed, chemical cues from captive fish and the abundance of fouling organisms on cages (Dempster et al. 2009, Akyol and Ertosluk 2010), but our study did not contain these variables and we thus attribute the presence of fish to the lights.

However, it is also important to note our study's limitations: (1) the lighting arrays' configuration, as well as their vertical and horizontal spacing differ from those typically used at finfish aquaculture facilities, and (2) environmental variables at our study site (*e.g.*, depth, temperature, currents) significantly differ from those at a typical finfish aquaculture facility. Thus, our findings have limited applicability to finfish aquaculture settings. To fully assess ecological effects of LEDs (and other types of lights) used at finfish farms and their impacts on farmed fish, the following study design is recommended:

- Year 1: farm A, lit with LED lights, stocked with fish; farm B, lit with LED lights, fallowed; farm C, unlit, stocked with fish; and farm D, unlit, fallowed.
- Years 2, 3, and 4: study parameters rotate between farms each year to allow adequate replication and account for potential spatial and temporal (annual) variability.

This study design, although difficult to apply due to logistical reasons, may be considered in the future.

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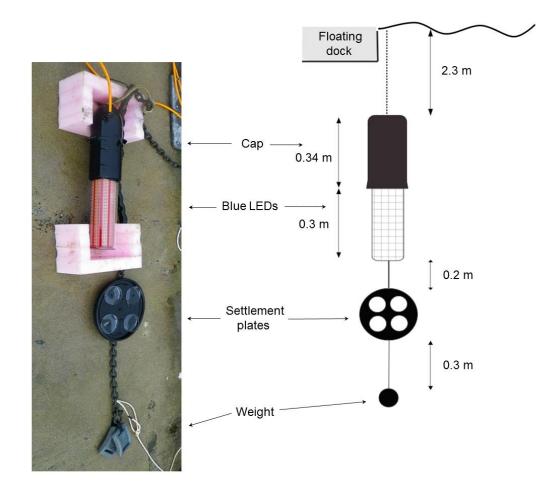


Figure 1. An example of the blue LED light set-ups deployed from the dock at West Vancouver Laboratory - CAER.

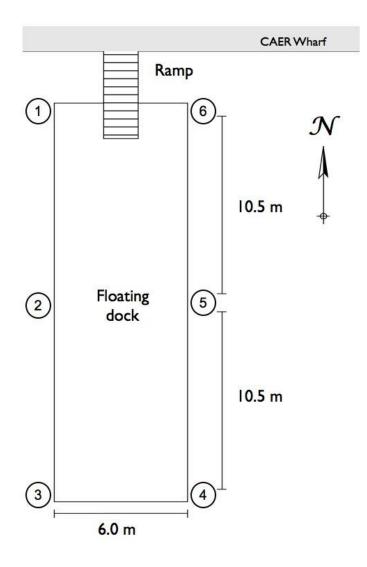


Figure 2. Experimental setup on the dock at West Vancouver Laboratory - CAER. Of the six light units installed, three were on (lit) at any one time (1, 3 and 5 or 2, 4 and 6), and the other three were unlit (control).

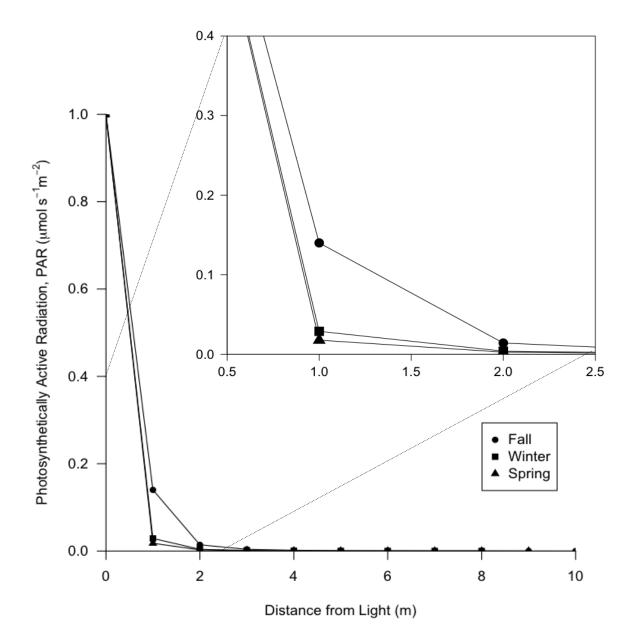


Figure 3. Light attenuation from blue LED lights measured as photosynthetically active radiation (PAR) at 1 m intervals from the blue 100W LED light during night sampling. The data are pooled into three seasons (Mean, n=3).

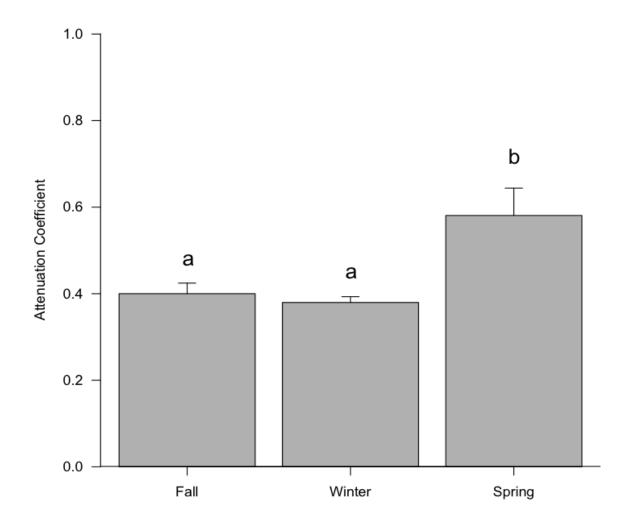


Figure 4. Turbidity in the water at the experimental site at West Vancouver Laboratory - CAER. Secchi disk measurements were taken at noon on sampling days and were converted into attenuation coefficients. Data were collected every two weeks and pooled into three seasons. Dissimilar letters indicate statistically significant differences ($\alpha = 0.05$) (Mean ± SE, n=3).

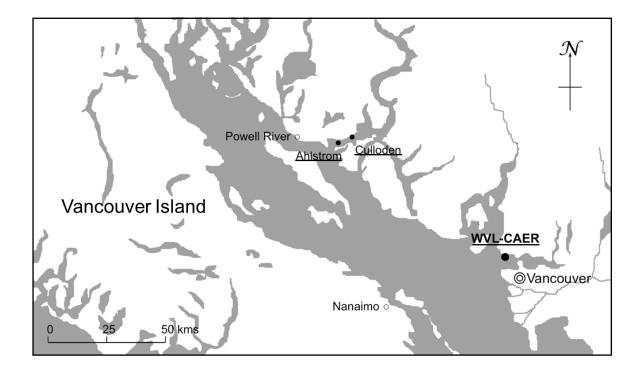


Figure 5. Map of experimental sites (underlined). The blue LED light deployment experiment was at West Vancouver Laboratory – CAER. Grieg Seafood BC Ltd. farm site Ahlstrom was the equipped with blue LED lights, and Culloden was equipped with white metal halide lights.

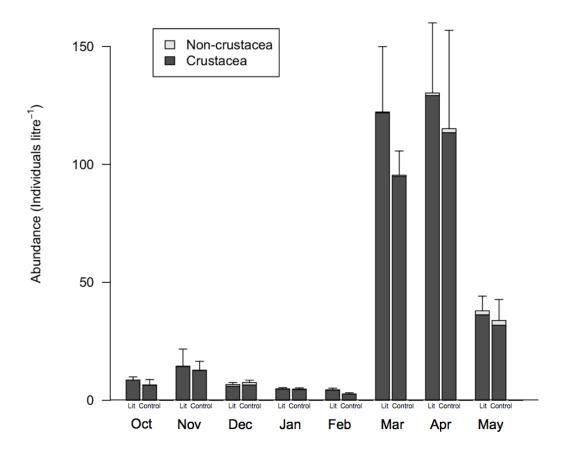


Figure 6a. Zooplankton abundance from plankton hauls conducted adjacent to each light during sampling from Oct 2011 – May 2012. For each date, the bar on the left is lit and the bar on the right is the unlit control. A Wilcoxon Signed-rank Test showed that there was a significant effect of light across all sampling dates (W = 36, Z = 2.52, p = 0.008, r = 0.63) (Mean ± SE, n=3 per date).

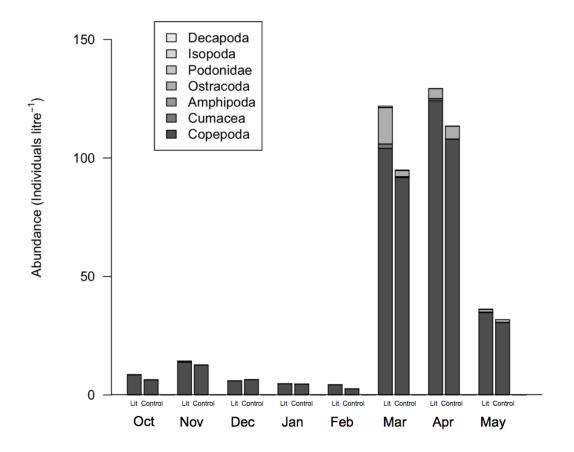


Figure 6b. Total abundance of crustaceans from plankton tows from Oct 2011 – May 2012. Each tow averaged 97% crustacean, and of those, 95% were copepods. For each date, the bar on the left is lit and the bar on the right is the unlit control (Mean, n=3 per date).

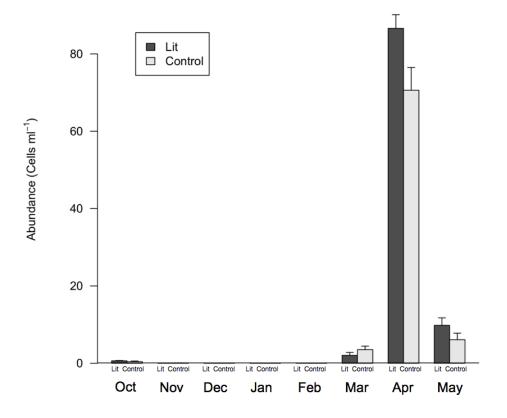


Figure 7. Total phytoplankton abundance from plankton tows from Oct 2011 – May 2012. For each date, the bar on the left is lit and the bar on the right is the unlit control (Mean \pm SE, n=3 per date).

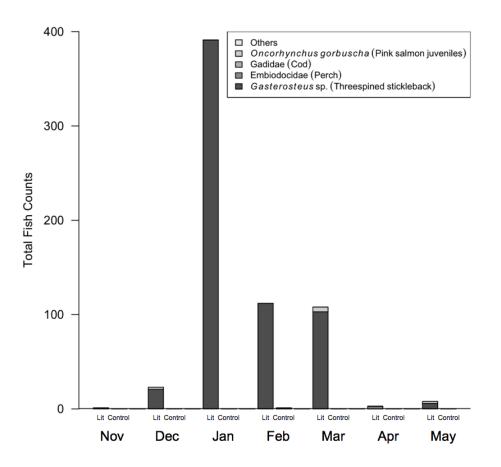


Figure 8. Total fish caught in minnow traps set overnight adjacent to each light during sampling from Oct 2011 – May 2012. For each date, the bar on the left is the lit blue LED light and the bar on the right is the unlit control (Mean, n=3 per date).

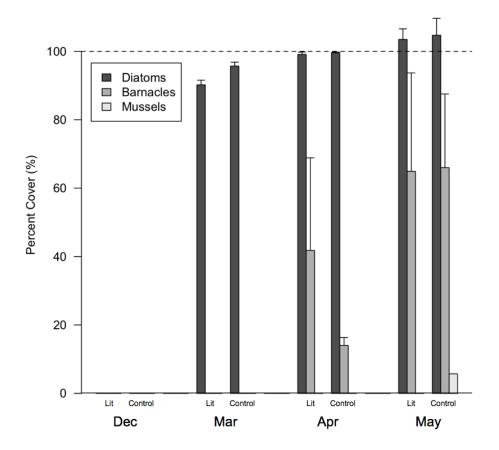


Figure 9. Percent coverage of settlement plates by organism from Dec 2011 – May 2012. There were no statistically significant settlement differences on plates adjacent to lit blue LED lights or unlit controls for either diatoms (A Wilcoxon Signed-rank Test W = 0, Z = -1.61, p = 0.25, r = 0.65) or for barnacles (W = 2, Z = 0.45, p = 1, r = 0.22). For each date, the bar on the left is lit and the bar on the right is the unlit control (Mean, n=3 per date).

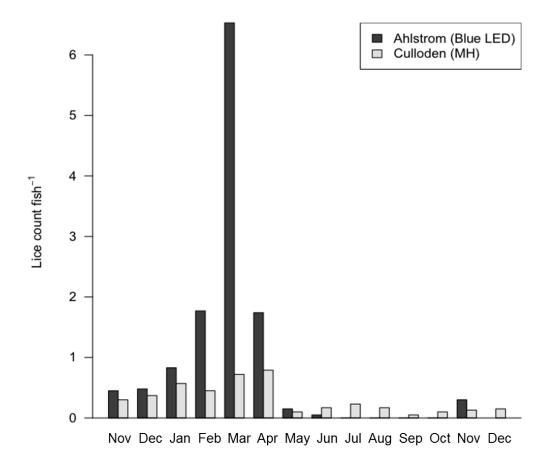


Figure 10. Total sea lice counts on salmon sampled at Grieg Seafood Ltd sites Ahlstrom (blue LED lights) and Culloden (white metal halide lights - MH) from November 2010 to December 2011. Statistical analysis was not possible for these data due to lack of replication of sites and limited data. Data provided by Grieg Seafood BC Ltd.

Table 1. Details of zooplankton collected in plankton tows at lit and unlit controls across all sampling events.

Sampling Date		27-Oct	27-Oct	22-Nov	22-Nov	20-Dec	20-Dec	18-Jan	18-Jan	15-Feb	15-Feb	28-Mar	28-Mar	25-Apr	25-Apr	30-May	30-May
Status		Lit	Control														
Crustaceans	Amphipoda	0.0054		0.0301	0.0163		0.0243	0.005	0.0036	0.0088	0.0044	0.097	0.0509	0.0815		0.0357	
	Copepoda	8.51	6.3913	13.84	12.598	5.9628	6.4834	4.6972	4.5998	4.3345	2.6312	104.1	91.712	124.09	107.93	34.734	30.494
	Cumacea	0.0326	0.0027	0.4333	0.057	0.0129	0.0243	0.1096	0.0092	0.0088	0.0309	1.7304	0.3989	0.8756	0.0855	0.0789	0.0438
	Decapoda											0.5452	0.2479			0.1485	
	Isopoda			0.0078								0.0679				0.0326	
	Ostracoda	0.0027		0.0272								15.316	2.5112	4.1252	5.3377	0.1688	0.2063
	Podonidae													0.0247	0.1137	1.0546	1.0915
Non-crustaceans	Hemiptera			0.0078													
	Bivalvia								0.0036					0.0129			
	Nematoda								0.0146								
	Cnidaria		0.0027	0.0078							0.0044				0.0647		
	Polychaeta	0.0054	0.0516		0.0054	0.0323	0.0388				0.0044	0.3441	0.2224	0.1875	0.3345	0.2262	0.2947
	Tunicata	0.0326	0.0978	0.1306	0.0978	0.6791	0.983	0.0747	0.1589	0.1116	0.0546	0.0291	0.2856	0.8467	1.2855	1.4589	1.7051
	Ctenophora	0.0027															
Total Zooplankton Abundance (Individuals litre ⁻¹)		8.59	6.55	14.48	12.77	6.69	7.55	4.89	4.79	4.46	2.73	122.23	95.43	130.25	115.15	37.94	33.84

Table 2. Details of phytoplankton in plankton tows at lit and unlit controls across all sampling events.

Sampling Dat	e	27-Oct	27-Oct	22-Nov	22-Nov	20-Dec	20-Dec	18-Jan	18-Jan	15-Feb	15-Feb	28-Mar	28-Mar	25-Apr	25-Apr	30-May	30-Ma
Status		Lit	Control	Lit	Control	Lit	Control	Lit	Control	Lit	Control	Lit	Control	Lit	Control	Lit	Contr
Diatom	Asterionellopsis sp.			.0003													
	Chaetoceros convolutus	.1314	.0653	.0219	.0507	.0299	.0264	.0042	.0089	.0076	.0067	.6559	.6997		.1857	.1248	.157
	Chaetoceros curvisetus	.3935	.4473	.0045	.0072	.0007	.0026				.0004	.1871	.4312	.0051	.0638	2.8074	1.49
	Chaetoceros decipiens	.2397	.1581	.0297	.0467	.0224	.0176	.0032	.0041	.0093	.0093	.073	.2627			2.8843	.815
	Chaetoceros didymus	.0233		.0008													
	Chaetoceros laciniosus	.1023	.0858	.0027	.0046	.0026	.0016			.0012		.1965	.3764		.0017	2.1069	1.37
	Chaetoceros radicans															5.1518	2.96
	Chaetoceros similis											.0501	.1882			.2187	.17
	Chaetoceros socialis																.33
	Chaetoceros sp.			.0016		.0008				.0014							
	Coscinodiscus sp.	.0037	.0065	.0036	.0042	.0016	.0014	.0034	.0044	.0019	.0018	.0002	.0004	.0021	.0019	.0002	.00
	Cyclotella sp.		.0007	.0008	.0031	.0006	.0012	.0011	.002	.0005	.0003						
	Cylindrotheca closterium						.0002	.0001	.002		.0001					.0174	.01
	Dactyliosolen fragilissimus						.0002	.0001					.0163				
	Detonula pumila											.1949	.0100	.0035	.0042	.4245	
	Ditylum brightwellii	.0146	.0323	.0004		.00005	.0004			.00004		.0001		.0055	.0042	.4243	.00
		.0140	.0020	.0004		.0001	.0004	.00005	.00005	.00004		.0001			.0004		.00
	Grammatophora marina		.0004			.0001		.00005	.00005				.0085				
	Gyrosigma sp.		.0004			.0001							.0085			0750	
	Lauderia annulata	0000	0004			0000	0004	0004	0000	0007	0000	0000	0005			.0756	.41
	Licmophora abbreviata	.0009	.0004		.0003	.0002	.0001	.0001	.0006	.0007	.0002	.0003	.0085				
	Melosira nummuloides			.0002				The second second									
	Melosira varians	.0486	.0092	.0025		.0066	.0043	.0021	.0078	.0005	.0027	.0008	.0345				.06
	Navicula sp.		.0104	.0013	.0003	.0064	.0049	.0029	.0213	.0027	.0183	.0037	.0336				.00
	Nitzschia paradoxa			.0011		.001			.0012		.0072		.0009				
	Odontella aurita					.0007	.0007	.0001	.0013	.0002	.0006	.0004	.0013				
	Odontella longicruris			.0001										.0002	.0003		
	Pseudo-nitzschia cf. delicatissima															1.4724	1.05
	Pseudo-nitzschia cf. pungens	.0313	.0315	.0024	.0011	.0015				.0008	.0002	.0008	.0327			2.2394	1.74
	Rhizosolenia setigera			.0003	.0002	.001	.0002	.0003	.0006	.0002	.0002	.0068	.0114				
	Skeletonema costatum	.2762	.2283	.0108	.0177	.0182	.0143	.0028	.0016	.0067	.0085	.5155	1.0935	173,498	141,189	2,4302	1.52
	Stephanopyxis turris	.0001											.0002	.0075	.0413	.0001	.00
	Striatella unipunctata								.0002			.0002	.0002				
	Synedra ulna	.0009	.0007	.0006		.0016	.0003	.0005	.0004	.0004	.0001						
	Thalassionema nitzschioides	.0121	.0108	.0083	0069	.0103	.0125	.0008	.0024	.0011	.0003	.097	.2318	.0034	.0494	.346	.46
	Thalassiosira nordenskioeldii		.0100	.0003							.0000	.9392	1,9134	.0079	.0531	.6126	.25
	Thalassiosira sp. (small)			.0005								1.2874	2.3713	.0013	.0001	.0120	.02
Dinoflagellate		.0033	.0036	.0007	.0003	.0001	.0005		.0003	.0001		1.2014	2.0110		.0001	.0001	.000
Dinonagellate	Ebria tripartita	.0033	.0030	.0007	.0003	.0001	.0005		.0005	.0001					.0001	.0001	.000
	Protoperidinium sp.			.0002						.0001		.0001	.0004	.0003	.0004		.000
Coldon algo				.0002		.00005						.0001	.0004	.0005	.0004		.000
Golden alga	Dinobryon sp.	.0079	.0578	.0023	.0039	.00005	.0045	.0075	.0161	.0035	.0026	.0003					
Green alga	Actinastrum sp. Unknown bluegreen filament	.0079	.05/6	.0023	.0039	.0065	.0045	.0075	.0161	.0033	.0026	.1713					
Bluegreen Unknown bluegreen filament				.0296	.0152	.0022	.0117	.0052	.011		.0108	.1/13					
Total Phytoplankton Abundance (Cells ml ¹)		1.29	1,149	.127	.162	.115	.105	.034	.084	.039	.07	4.382	7,717	173.528	141.591	20.913	12.8