Efficacy testing of novel antifouling coatings for pen nets in aquaculture: how good are alternatives to traditional copper coatings?

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Highlights

1. Copper-based antifouling coatings in salmon aquaculture have limited efficacy; there is a need for alternative coatings.
2. All tested novel coatings accumulated biofouling earlier and/or at higher abundances than the commercial copper coating.
3. One coating with a copper content <1% combined with a booster performed more similar to the commercial copper coating.
4. This study indicates that it is possible to substitute much of the copper in a coating while retaining functionality.

Abstract

Biofouling is a challenge in global sea-based salmon farming. Norway's salmon-growing industry relies primarily on copper-based antifouling coatings. However, copper is an increasingly recognised environmental hazard, and there is a need to develop alternative antifouling products to prevent biofouling in marine aquaculture. Using field experiments, this study compared the efficacy of six novel antifouling coatings for fish farm nets (two with reduced copper content, three with alternative biocides and one biocide-free coating) against a popular commercial copper coating and uncoated samples. The performance of one of the new coatings with lower copper content was more similar to the commercial copper control while the rest were colonised by biofouling faster and/or at higher abundances. However, none of the tested products were able to prevent biofouling entirely, underlining the importance of the search for alternative and improved antifouling technologies.

Keywords: Biofouling, Antifouling, *Salmo salar*, Cage nets, Copper

1 Introduction

Biofouling, the undesired growth of marine organisms on surfaces submerged at sea, is one of the major challenges in contemporary salmon farming. Biofouling can cover up to 100% of the mesh openings in cage nets, and growth of organisms such as algae, bivalves and hydroids can lead to net deformation and volume reduction (Cronin et al. 1999, Braithwaite et al. 2007, Lader et al. 2008, Guenther et al. 2010). The presence of biofouling is further suspected to reduce the efficacy of cleaner fish (employed to feed on salmon lice as biological control), by providing an alternative food source (Kvenseth 1996, Eliasen et al. 2018). Biofouling on farm nets may also affect fish health, by facilitating exposure to pathogens associated with biofouling organisms and by hindering water exchange and thus reducing oxygen levels and the removal of waste (reviewed in de Nys & Guenther 2009, Fitridge et al. 2012).

To limit the impacts of biofouling on salmon farming operations in Norway, most farmers use antifouling (AF) coatings to prevent settlement and reduce growth on their nets (Floerl et al. 2016, Bannister et al. 2019). The majority of current AF coatings contain cuprous oxide as a biocidal agent due to its toxicity to biofouling organisms. However, the current AF coatings are no optimal solution for two main reasons:

(1) Current copper-based coatings have highly limited efficacy and are not able to prevent biofouling for the entire production cycle. Nets are reported to be fouled after an average of three months, and sometimes as fast as after just four weeks (Guenther et al. 2010, Bloecher et al. 2015; SINTEF, unpublished data). This forces the farmers to combine copper coatings with cleaning of the nets using *in situ* high-pressure washers. High-pressure cleaning is time-consuming, costly, and poses a variety of risks. For example, mechanical damage to nets caused by abrasive cleaning can lead to the escape of fish, while biofouling washed off the net can irritate gills (Bloecher et al. 2018) or facilitate exposure to pathogens associated with biofouling (Hellebø et al. 2016, reviewed in Floerl et al. 2016). High-pressure cleaning also abrades the coating surface, further reducing antifouling performance.

(2) Copper is an environmental hazard. While being raised within intact copper coated nets does not harm Atlantic salmon (Solberg et al. 2002), acute exposure to dissolved copper or copper nano-particles, such as potentially released during net cleaning, can cause injuries to nerves and gill tissues in salmonids (Baldwin et al. 2011, Al-Bairuty et al. 2013). In addition, the released copper can be found in the surrounding water column and accumulates in the sediment beneath farms (Loucks et al. 2012, Nikolaou et al. 2014), where negative effects relating to embryonic development, movement, enzymatic activity and gill health have been reported for non-target organisms including fishes, invertebrates and algae (reviewed in Burridge et al. 2010).

Consequently, there is a need for novel AF products that contain less or no copper while offering better or at least similar protection from biofouling than currently available copper-based coatings. This is especially important for companies that farm organic salmon, and the increasing number of farmers interested in complying with certification standards such as the Aquaculture Stewardship Council (ASC) Salmon Standard that restrict the use of antifouling coatings (Aquaculture Stewardship Council 2012). Commercial alternatives to copper-based coatings include coatings that contain other biocides such as Tralopyril (e.g., 'Econea' by Janssen PMP). However, similar to copper, these alternative biocides can have negative impacts on non-target species (Oliveira et al. 2014, Oliveira et al. 2016). In contrast, biocide-free coatings mainly function by providing a smooth (Baum et al. 2017) and durable surface that is intended to reduce available settlement area and facilitate ease of removal and resistance to wear during net washing (e.g., 'Netpolish' by NetKem or 'NetCoating' by Steen Hansen). Variations of nets made of plastic are based on a similar (biocide-free) principle (e.g., 'KikkoNet' by Maccaferry). Many of these, however, result in increased cleaning requirements with the associated risks to net integrity and fish health.

In this study, the antifouling performance of two net coatings with reduced copper concentration (relative to a common commercial copper coating), three coatings based on alternative (non-copper) biocides, and one biocide-free coating were compared to a widely-used commercial copper coating and an uncoated control. The coatings examined were not commercially available in Norway at the time of the testing and the objective of the study was to evaluate their suitability as alternatives to current copper coatings.

2 Material and methods

2.1 Net sample preparation and placement

A total of six AF coatings were tested: two coatings with a copper content below 5% ('Low 1' and 'Low 2'), three coatings containing alternative biocides such as Econea ('Alt 1' and 'Alt 2') or a boron compound ('Alt 3'), and one biocide-free coating ('Free') (Table 1). Five of these coatings were developed for testing and are not commercially available, while 'Alt 3' is sold outside of the EU (e.g., Japan, Chile). The six coatings were compared to two controls: uncoated net samples ('Uncoated'), and net samples coated with a commercial copper-based AF coating featuring a cuprous oxide concentration of 22% ('CuCtrl'). Net samples (20 x 20 cm, n = 12 per coating and trial) were cut from a single piece of net material (Raschel knit nylon smolt netting, Egersund Net) and mailed to each coating manufacturer, who performed the coating. The coated samples varied in colour from white to black (Table 1). Each sample was individually numbered with a plastic tag (micro ear tags, OS ID), before they were attached to four metal frames (24 samples per frame) in random order using cable ties. Each individually marked frame held three replicates of each coating (including controls), with samples placed one sample size apart. The frames were distributed to four locations at the SINTEF ACE research farm site Rataren, in Mid-Norway. The test was conducted in two trials, aligned with the periods when hatchery-reared fish are transferred to sea. The samples of Trial 1 were deployed on 31.03.2017 ('Spring trial'), and the samples of Trial 2 were deployed on 28.06.2017 ('Autumn trial'). All samples were retrieved on 03.10.2017 after six and three months at sea, respectively. Permission for field testing of a boron-compound currently not permitted under the EU biocide regulation for antifouling paints (EU No 528/2012, art. 56) was granted by the Norwegian Ministry of Climate and Environment.

Table 1: Overview of the tested net coatings

ID	Colour	Active ingredients	Producer
Low 1	Gold	Low copper content (0.6% CuO + copper pyrithione)	Brynsløkken AS, NO
Low 2	Green	Low copper content (< 5% CuO + zinc/zinc pyrithione)	NetKem AS, NO
Alt 1	Olive	Alternative biocide (2% Econea + zinc pyrithione)	Brynsløkken AS, NO
Alt 2	Blue	Alternative biocide (2.9% Econea)	NetKem AS, NO
Alt 3	Yellow	Alternative biocide (boron compound) (= 'Nitto Boron Paint')	Nitto Seimo, Japan
Free	Black	Biocide-free, based on alternative substance	NetKem AS, NO
Cu Ctrl	Red	Commercial copper coating (22% CuO) (= 'Netwax NI 4')	NetKem AS, NO
Uncoated	White	Uncoated net	Egersund Net AS, NO

2.2 Sampling and analysis

Biofouling growth on net samples was monitored monthly via high-resolution photographs. Before photographing, the frames were cleared of biofouling and the samples were gently rinsed with a fine spray of sea water at a maximum rate of 5 L min^{-1} per panel to remove silt and entangled debris. The samples were then photographed vertically against a blue background under artificial light to optimise contrast. To limit air exposure and prevent desiccation, samples were processed in small batches of twelve. The experimental trials were terminated once biofouling accumulation on the copper control treatments exceeded 65% cover. For the 'Spring trial' and 'Autumn trial', this resulted in test durations of six and three months, respectively. Due to adverse weather conditions it was not possible to sample in June 2017.

Biofouling occurrence in the monthly photographs of the net samples was described using the Fouling Resistance (FR) scale, based the ASTM standard for antifouling coatings (D 3623–78a; American Society for Testing and Materials ASTM 1998), modified to evaluate nets. The analysis was based on the identification of taxonomic groups at 66 points on each net sample, defined by the intersection of the net with three randomly placed vertical and horizontal lines. Slimes and algal spores <3 mm were not counted as biofouling (FR score = 100 even in their presence), while the presence of larger biofouling organisms resulted in a default reduction of the FR score to 95, or lower depending on biofouling abundance (American Society for Testing and Materials ASTM 1998). Much of the recent research on finfish farm biofouling used Percentage Net-aperture Occlusion (PNO, Guenther et al. 2010) to quantify biofouling on net surfaces. However, the use of the FR scale was chosen here because it allowed to identify, and exclude from the analyses, exogeneous material (e.g. dead algae) that had drifted into the net panels. To ensure comparability of this study with previous research we also determined the PNO values for all samples in this study and found a significant positive relationship between the two metrics ($R^2 = 0.90$; $p < 0.001$; see Figs. S1 and S2 in supplement). Fouling resistance is presented as average ± standard error (SE).

For each sample, biofouling prevalence, species richness and community composition were estimated. Biofouling prevalence was calculated as the percentage of fouled replicates within one treatment per sampling event. Species richness was calculated for each treatment based on the sum of taxa found on all replicates per sampling event (S) and for the total duration of each trial (S_t). Community composition was calculated as percentage contribution of individual taxonomic groups to the biofouling community of each treatment for each sampling event and the total duration of each trial.

A statistical analysis of fouling resistance of the net coatings was conducted for data collected in the month of August, when surfaces associated with the 'Spring trial' and 'Autumn trial' had been immersed for a period of five and two months, respectively. This timeframe was chosen deliberately, because in August the copper control treatment still had some visible antifouling ability, but the uncoated control treatment was already fully fouled. The test was performed using permutational analysis of variance (PERMANOVA, PRIMER v.7.0) based on Euclidean distance with 9,999 unrestricted permutations of residuals under a reduced model and a significance level of 5%. The analyses included the factors "Trial" ('Spring' vs. 'Autumn trial'; fixed), "Location" (Location of the frame with samples on the farm site; four locations, random), and "Coating" (Coating type, fixed).

Principal Coordinate Analysis (PCO; PRIMER v.7.0) was used to explore and visualise the overall ‘performance’ of the antifouling coatings examined on the basis of a set of ‘performance indicators’: (i) the period for which the coatings stayed free of biofouling (rendering net cleaning unnecessary), (ii) total species richness associated with each treatment (the more species are able to colonise the coating the less effective its protection is), (iii) fouling resistance (FR) measured in August (the time when biofouling was first found on the copper control but before all coatings failed), and (iv) percent cover of the two most abundant taxonomic groups, 'hydroids' (whose presence can pose risks to gill health; Bloecher et al. 2018) and 'algae' (who are among the earliest settlers and most common biofouling organisms), in August. These five variables represent different but meaningful dimensions of the ‘antifouling performance’ of coatings or other treatments, since they are related to maintenance cost and/or potential health or welfare impacts. An integrated assessment of these variables is new and somewhat subjective, but we consider it a potentially useful approach for looking at performance in the context of factors that are relevant and important to fish farmers. The analysis was conducted individually for the 'Spring trial' and 'Autumn trial' using Euclidean distance based on normalised data with vector overlays based on Pearson correlations for the selected factors.

Of the 96 net panels deployed in the 'Spring trial', a total of twelve panels were lost due to adverse weather conditions, yet never more than two of the twelve replicates of each treatment were missing. In the 'Autumn trial', a single panel of the 'Alt 3' treatment was lost.

3 Results

Over the course of the two trials, 21 taxonomic groups were identified on the net panels. Hydroids, strongly dominated by *Ectopleura larynx*, were the most common biofouling organism on the net panels, accounting for 51% of biofouling cover observed. Hydroids were found on the samples as early as May, only two months after immersion (Figs. S3 and S4 in supplement). Algae, including green, brown, and red algae, were the second most abundant group (35%), and also present from May (Figs. S3 and S5). Ascidians, including the notorious biofouling species *Ciona intestinalis*, were found on the coated samples only in September, the final month of both trials, but at very high abundances, making them the third most abundant biofouling organisms (Fig. S3). Other invertebrates were present at lower numbers. This included erect and encrusting bryozoans, which also settled mainly in September (Fig. S3). Caprellid amphipods, in contrast, arrived together with the hydroids and were present throughout the experimental period (Fig. S3). In addition, low numbers of the tube-building amphipod *Jassa* sp., bivalve species such as *Mytilus edulis* and *Hiatella arctica*, one anemone species, and one stauromedusa species were encountered.

Biofouling development was slower in the 'Spring trial' than in the 'Autumn trial', but generally reached higher total abundance on the experimental surfaces. Initial biofouling development began on the uncoated samples in May, two months after immersion, and was found on 60% of the sample panels (Fig. 1). However, since it consisted mainly of very young hydroids, it had little impact on the fouling resistance (mean FR = 95±1 SE; Fig. 2). From the next month onwards, biofouling was found in abundance on all samples of the uncoated nets, resulting in a very low fouling resistance (FR = 9±9) in June, and FR = 0±0 in September, six months after immersion. In the 'Autumn trial', all uncoated control samples were heavily fouled (FR = 0±0) following two months of immersion (Fig. 2). The uncoated samples had the highest species richness (S) measured during the 'Spring trial' in August (S = 12, Fig. 3), as well as the highest total species richness (cumulative for all replicates of a given coating; $S_t = 14$, Fig. 3). The samples were dominated by hydroids for most of the time, with the exception of June, when algae were most abundant, and September in the 'Spring trial', when ascidians dominated the samples, followed by bryozoans (Fig. S3).

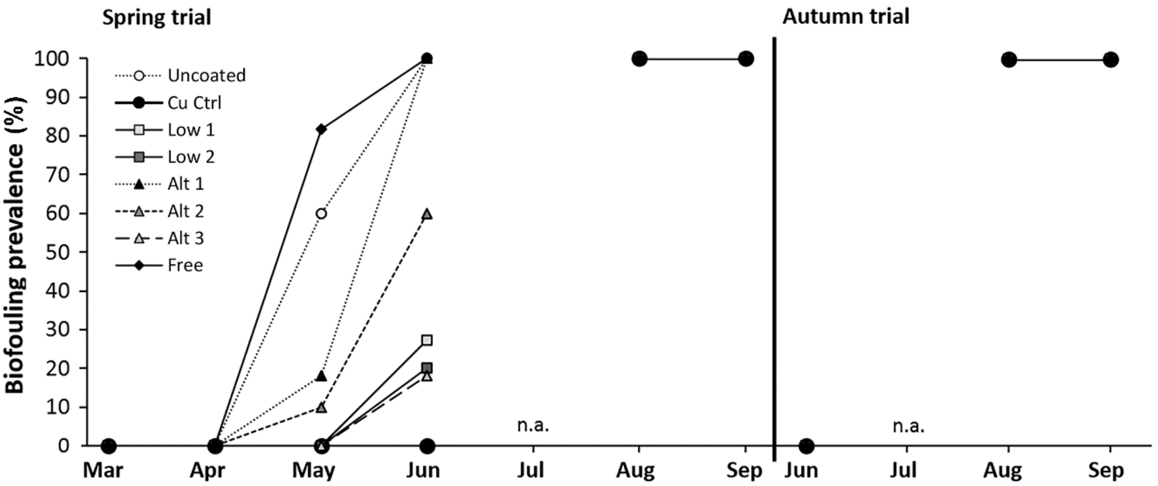

Fig. 1: Prevalence of biofouling on net samples treated with eight different coatings in the 'Spring' (six months duration) and the 'Autumn' (three months duration) trial. No data could be collected in July. As biofouling prevalence in August and September was 100% for all samples, data accumulated in the same position.

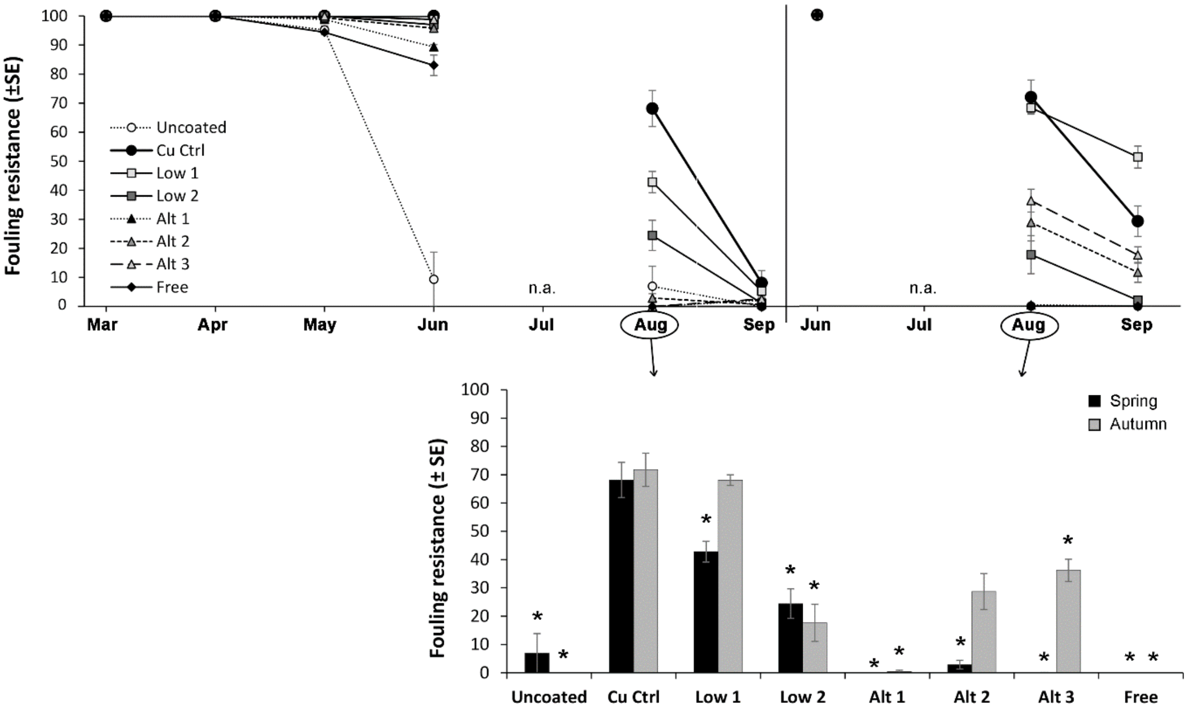

Fig. 2: Average fouling resistance (± standard error) of eight different coatings of net samples in the 'Spring' (six months duration) and the 'Autumn' (three months duration) trial, including a detailed depiction of the situation in August of both trials. Coatings with significantly lower fouling resistance than the copper control are indicated by an asterisk (Trial x Location x Coating: $F_{21;115}$= 2.279, p = 0.006). No data could be collected in July.

Samples coated with the commercial copper coating ('Cu Ctrl') remained free of biofouling for at least three months, longer than any of the other coating treatments. Although biofouling was found on 100% of the copper control panels in August and September of both trials, (Fig. 1), fouling resistance was highest in August of both trials (FR: spring = 68±6, autumn = 72±6; Fig. 2). In September, however, fouling resistance dropped sharply in both trials, to 8±4 ('Spring trial') and 29±5 ('Autumn trial'). This sudden increase in biofouling abundance was caused mainly by the hydroid *E. larynx*. Through most of both trials, the copper control samples had the lowest species richness (Fig. 3, $S_t = 6$) and were strongly dominated by hydroids in both trials. The only other taxa present on the copper coated samples were algae and caprellid amphipods (Fig. 4, Fig. S3).

August was the point in time where the uncoated control surfaces had been fully colonised, while panels coated with the copper control still showed considerable fouling resistance. From August onwards, performance of all coatings in both trials decreased considerably. Therefore, the comparison of the coatings to the copper and uncoated control was undertaken based on the August data. In that month, fouling resistance of the samples differed between the two trials and between coatings, and was not always consistent between locations (Trial x Location x Coating: $F_{21;115}= 2.279$, $p = 0.006$).

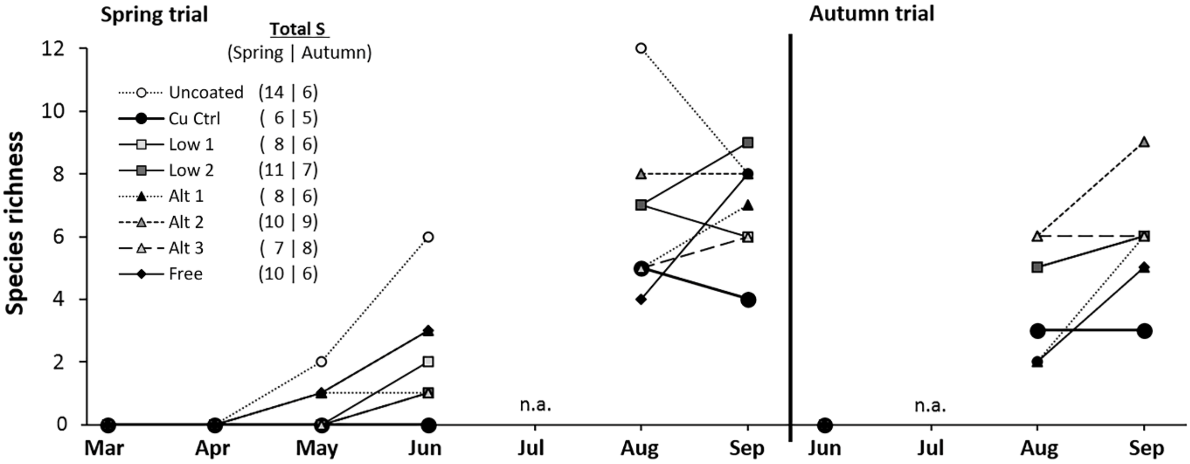

Fig. 3: Accumulated monthly species richness measured on eight different coatings in the 'Spring' (six months duration) and the 'Autumn' (three months duration) trial. In addition, the accumulated total species richness (Total S) for the duration of the trial is listed. No data could be collected in July.

Of the two coatings with low copper content, the 'Low 1' coating performed best relative to the copper control, while the 'Low 2' coating displayed inconsistent performance. Biofouling started to accumulate on the panels in June of the 'Spring trial', three months after immersion, later than on most other coatings, and with relatively low prevalence (27% and 20% of the panels of 'Low 1 and 2', respectively; Fig. 1). In August, after five months of immersion ('Spring trial'), fouling resistance of both coatings was below 50 ('Low 1': 43±4, Low 2: 24±5). While not as good as the copper control, the 'Low 1' coating performed significantly better than all other coatings (pairwise comparisons, $p < 0.05$; Fig. 2). In August of the 'Autumn trial', the 'Low 1' coating had fouling resistance values similar to the copper control and higher than all other coatings (pairwise comparison, $p < 0.05$; Fig. 2). In September, its fouling resistance did not decrease as rapidly as that of the other coatings. However, panels with the Low 2 coating were heavily colonised in the latter part of the trials and did not differ from the uncoated control by August. Species richness for the low copper coatings varied in both trials (Fig 3), with Low 2 having the second highest total species richness in the 'Spring trial' ($S_t = 11$, Fig 3). Although initial settlement on the 'Low 1' samples in June consisted of hydroids and caprellid amphipods, it was later dominated by algae (Fig. 4). In the 'Spring trial', the first settlers on the Low 2 coating were hydroids, but they were displaced by ascidians and algae by the end of the trial after six months at sea. In the 'Autumn trial', the biofouling on both coatings consisted mainly of algae.

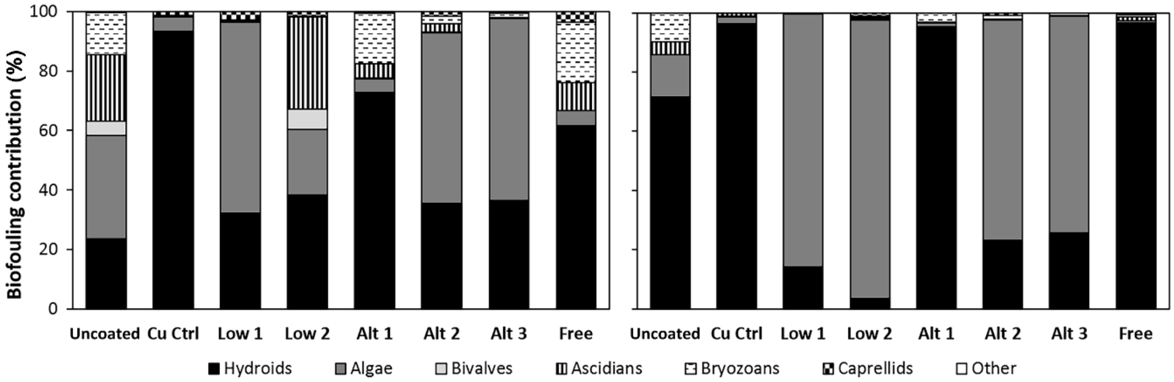

Fig. 4: Total biofouling composition on nets with eight different coatings after six months ('Spring trial') and three months ('Autumn trial') at sea, depicting percentage cover of hydroids, algae, bivalves, ascidians, bryozoans, and caprellids.

The antifouling performance of the coatings containing alternative biocides ('Alt 1 – 3') was in general inferior to the ones containing copper ('Cu Ctrl', 'Low 1 and 2'). 'Alt 1' and 'Alt 2' started accumulating biofouling in May, two months after immersion, though at lower prevalence than the uncoated samples (Fig. 1). 'Alt 3' remained free of biofouling until June, three months after immersion. During the 'Spring trial', fouling resistance started to decrease in June, and was 0 for all three coatings within five months following immersion (Fig. 2). 'Alt 1' and 'Alt 3' had significantly lower fouling resistance than the copper control in August of both trials (pairwise comparisons, $p < 0.05$; Fig. 2). While 'Alt 2' also had lower fouling resistance in the 'Autumn trial', it displayed large variability in the 'Spring trial' and did not differ from both copper control and uncoated control (Fig. 2). Species richness was relatively low on 'Alt 1' and 'Alt 3' in the 'Spring trial', and higher in the 'Autumn trial' (Fig. 3). 'Alt 2' reached the highest species richness of all coatings in the 'Autumn trial' ($S_t = 9$, Fig. 3). The species composition of 'Alt 1' coated samples was similar to the uncoated samples and was dominated by hydroids for most of the time (Figs. 3 and 4). Although initial biofouling on 'Alt 2 and 'Alt 3' in the 'Spring trial' was dominated by hydroids (and caprellid amphipods for Alt 2), the August and September samples of both trials were dominated by algal biofouling.

Colonisation of panels containing the biocide free coating ('Free') occurred in a manner similar to the uncoated samples throughout both trials. It was the first coating to reach high prevalence of biofouling on the samples (82%) after two months of immersion (Fig. 1) and fouling resistance was among the lowest of all tested coatings in both trials (Fig. 2). Species richness was below 5 in the 'Spring trial', and below 3 in the 'Autumn trial' on the biocide free coating until the last month of both trials, where it doubled (Fig. 3). The main biofouling species on this coating were hydroids and bryozoans, but also ascidians, caprellid amphipods, and algae occurred at moderate frequencies (Fig. 4).

The two dimensions of the PCO ordination as shown in Fig. 5 are a good representation of coating performance, capturing 81% and 98% of the information associated with the five performance indicator variables for the 'Spring trial' and 'Autumn trial', respectively.

For the 'Spring trial', the horizontal dimension of the PCO was correlated with fouling resistance values, time until biofouling occurred, and total species richness, and explained ~63% of the variation in the distance matrix. The copper control treatment performed most distinctly from the uncoated control on the basis of these variables, characterised by high fouling resistance and longer periods till first colonisation by biofouling organisms. The 'Alt 1', 'Alt 2' and biocide-free coatings showed little antifouling performance, with 'Alt 1' and biocide-free panels having a particularly high abundance of hydroids. Overall, the 'Low 1' coating achieved the best performance relative to the copper control. These patterns are similar for the 'Autumn trial', in that 'Low 1' achieved best performance relative to the copper control, although differences between coating are at times driven by the abundance of algae and hydroids.

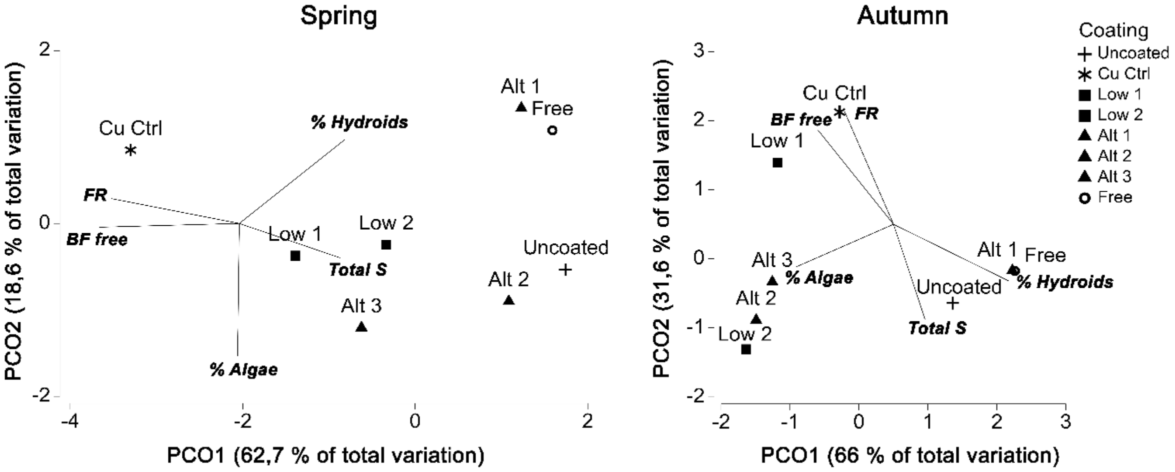

Fig. 5: Principal Coordinate Analysis (PCO) depicting the relation between the tested coatings based on selected performance indicators: Months until the first occurrence of biofouling (BF free), Total species richness (Total S), Fouling resistance measured in August (FR), and % cover represented by hydroids (% Hydroids) and algae (% Algae) in August.

4 Discussion

None of the novel antifouling coatings tested were able to prevent biofouling for the entire duration of the experiment or performed better than the established commercial copper coating. However, the coatings were able to delay the onset and reduce the abundance of biofouling for some time. Although the general trend in biofouling accumulation and community composition on the coatings was similar over the two trials, biofouling accumulation was much faster in the 'Autumn trial', when samples were immersed during the peak of the biofouling season. Compared to the 'Spring trial', however, the 'Autumn trial' had a lower species richness. Here, the fastest-growing species such as *Ectopleura larynx* and algae were able to monopolise the available space in the young community quickly, a common pattern in biofouling community development (Boero 1984, Migotto et al. 2001, Bloecher et al. 2013b).

The commercial copper coating prevented settlement of most organisms with the exception of hydroids and some algal species. Hydroids such as *E. larynx*, as well as some algae, are known to have a high tolerance to copper-based AF coatings (Barnes 1948, Pyefinch & Downing 1949, Hall 1980, Edwards et al. 2014), enabling their settlement on coated nets.

While the copper content of the 'Low 1' coating was substantially lower (0.6% CuO) than in the copper control coating (22% CuO), its performance was notably more similar to the copper control than that of the other tested coatings. In contrast, the second low-copper coating (Low 2), did not perform as well.

Since both coatings had a copper content <5%, the difference in performance was likely caused by the supplementary biocide: 'Low 1' contained the algaecide copper pyrithione in addition to copper, while in Low 2 the copper was supplemented with zinc/zinc pyrithione; indicating the copper pyrithione as a potential candidate for the superior performance.

Although none of the coatings containing alternative biocides were able to perform as well as the commercial copper coating, some differences between the coatings were apparent. 'Alt 3', containing a boron compound, showed a better overall performance than the other two coatings containing Econea (Tralopyril). Coatings containing boron are currently not approved in EU countries or Norway (European Chemicals Agency (ECHA) 2018a), but are used in Japan and Chile for finfish aquaculture.

The copper- alternatives and supplements tested in this study, copper pyrithione, zinc pyrithione, Econea, and an organic boron compounds, are collectively termed booster biocides. These compounds are often included in copper-based paints to complement and enhance the efficacy of the coating, but they are also available as the main active ingredient (Guardiola et al. 2012, Amara et al. 2018). While this study shows that they are indeed likely to enhance or partially substitute the antifouling abilities of cuprous oxide, it is not clear whether they are a true alternative to copper from an ecological standpoint, i.e. more beneficial towards non-target species and the environment at large. Research indicates that they, similar to copper, affect non-target species (Bao et al. 2011, Oliveira et al. 2014, Oliveira et al. 2016, Amara et al. 2018). Copper pyrithione, for example, has been shown to impact gill health of salmonids in laboratory assays (Borg & Trombetta 2010). In fact, some studies evaluated booster biocides as more toxic than copper or Tributyltin (TBT) for specific species (Bao et al. 2011), and in response the European Chemicals Agency has recently amended the hazard categories for zinc pyrithione (European Chemicals Agency (ECHA) 2018b).

While environmental concerns may indicate biocide-free coatings as a desirable solution, the one tested in this study was currently not able to perform equally well as the biocidal coatings. These coatings may, however, be able to delay settlement to some extent and offer a temporary advantage over uncoated nets. Baum et al. (2017) observed a delay in biofouling growth on nylon nets coated with a biocide-free coating compared to uncoated raschel knit nylon nets. The effect was attributed to the ability of the coating to alter the naturally rough and variable surface of a raschel knit nylon net, sealing and smoothing it which resulted in changed flow characteristics around it, thus creating a less favourable environment for biofouling organisms (Baum et al. 2017). Likely, a similar effect may have influenced settlement on the biocide-free coating. Unfortunately, this mechanism can only be effective for a limited time since hydroids such as *E. larynx* do not have topographic preferences (Bloecher et al. 2013a) and display very variable and plastic attachment methods (Carl et al. 2011). However, since manual net cleaning is a very costly process, even modest delays in biofouling development can provide significant cost savings for the fish farming industry.

The tested net coatings varied in colour from white to black due to manufacturer preferences. This colour variability between individual coatings may have influenced the species composition on the net samples, attracting some species while repelling others (Hodson et al. 2000, Satheesh & Wesley 2010). However, it is unlikely that the colour has influenced fouling resistance for an extended period of time, since other, less colour-sensitive species would likely have settled indiscriminately. Specifically, the hydroid *E. larynx*, one of the main biofouling organisms in this study that settled from early on, has been shown to settle indifferently to coating colour (Guenther et al. 2009). Thus, although coating colour may have influenced initial settlement and community composition, it is unlikely to have confounded the overall biofouling prevention assessment.

In the first two months of the 'Spring trial', considerable amounts of dead algal material drifted into the samples via local current regimes. Most of the material could be removed by gentle rinsing of the samples prior to photographing. The remaining material could largely be identified and thus excluded from the analysis for fouling resistance, underlining the advantage of fouling resistance analysis over a computerised analysis of net-aperture occlusion. The random arrangement of treatment replicates on the experimental frames furthermore ensured an ‘even’ distribution of any effects caused by drifting material. There was little drifting material in the successive months of the trial.

Interestingly, the producers of the individual coatings reported surprise upon seeing the study results, as they did not concur with their own observations for some of the coatings. It appears that the 'Low 1' coating had not previously performed as well as a classic copper coating, while the performance of the 'Alt 1' coating was below expectation. Part of the explanation for this discrepancy in test outcomes may be the temporal and spatial variation in biofouling species ready for settlement that is typical for marine waters (Olafsen 2006, Fitridge et al. 2012, Bloecher et al. 2013b). Furthermore, according to the producer of the 'Alt 3' coating, this product is usually used on nets other than raschel knit nylon nets and is reported to perform much better under such conditions. This indicates the interaction between the coating and the net material as another important factor to consider in biofouling management on aquaculture nets.

All other tested coatings gave inferior performance compared to the commercial copper coating in that they either became colonised sooner (all coatings were fouled after five months or earlier), attracted more biofouling, or both. As such, none of the other new coatings presents a better alternative to the *status quo*, from an antifouling perspective and without the added use of net washing. While environmental considerations associated with antifouling products (contamination, health risks) are justified and important, they have so far not been appropriately compared to the potential impacts of net cleaning operations. Net cleaning is expensive, and likely associated with considerable fuel consumption – and greenhouse gas emissions – by the growing number of cleaning service vessels. Net cleaning can also expose farmed (and wild) fish to harmful material that may have health or welfare effects (Floerl et al. 2016, Bloecher et al. 2018). As such, while the use of non-biocidal coatings is inherently desirable, if their antifouling efficacy is reduced compared to biocidal coatings then their net benefit needs to be derived from analyses that consider the factors above.

In conclusion, this study indicates that is it possible to design a coating that performs quite similar to commercial copper coatings despite containing only a fraction of the copper content, if paired with the 'right' booster biocide. This finding provides encouragement for the Norwegian aquaculture industry, who has had reservations regarding the use of reduced-copper antifouling over concerns around performance. A widely-adopted reduction to a copper content <5% has the potential to reduce the current annual amount of copper released in Norwegian salmon farming by approximately 870 tons (Skarbøvik et al. 2017). However, some booster biocides are known to pose serious environmental risks (Turner 2010). While the ability to reduce the use of copper-based antifouling coatings in fish farming can have tremendous environmental benefits, it is important that all effort is made that the incorporation of booster biocides into novel aquaculture antifouling products enhances rather than compromises these.

Acknowledgements

This work was supported by the Norwegian Seafood Research Fund (FHF; grant number 901289). The authors thank Egersund Net AS for supplying net material; as well as Brynsløkken AS, Netkem AS, and Nitto Seimo for supplying the coated net samples. In addition, we are thankful to Terje Bremvåg, Torbjørn Benjaminsen, Kari-Anne Ofstad, Eleni Kelasidi and Walter Caharija for assistance during sampling, and the anonymous reviewers for positive and constructive feedback.

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SUPPLEMENT to Bloecher and Floerl

Efficacy testing of novel antifouling coatings for net pens in aquaculture: how good are alternatives to traditional copper coatings?

Percentage Net-aperture Occlusion data

Percentage Net-aperture Occlusion (PNO), the total contribution of biofouling to the occlusion of the net aperture, was calculated as described in Guenther et al. (2010)[1].

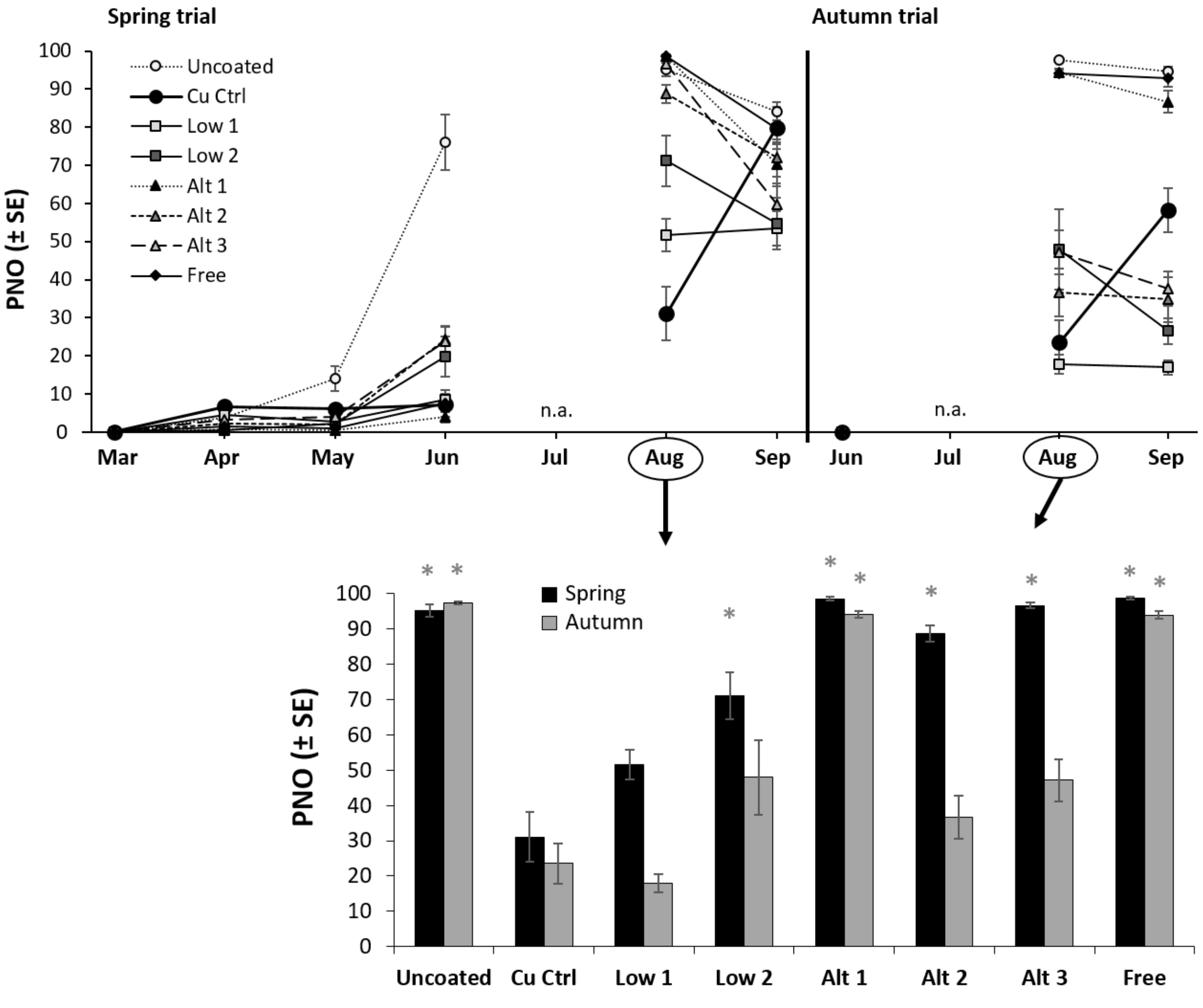

Fig. S1: Average Percentage Net-aperture Occlusion (PNO ± standard error) of eight different coatings of net samples in the 'Spring' (six months duration) and the 'Autumn' (three months duration) trial, including a detailed depiction of the situation in August of both trials. Coatings with significantly higher PNO values than the copper control are indicated by an asterisk (Trial x Location x Coating: $F_{21;115}$= 3.22, $p < 0.001$). No data could be collected in July.

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[1] Guenther J, Misimi E, Sunde LM. 2010. The development of biofouling, particularly the hydroid *Ectopleura larynx*, on commercial salmon cage nets in Mid-Norway. Aquaculture 300:120-127.

Correlation between fouling resistance and percentage net-aperture occlusion

To allow a linear regression going through 0, fouling resistance is presented inverted (= 100 – FR).

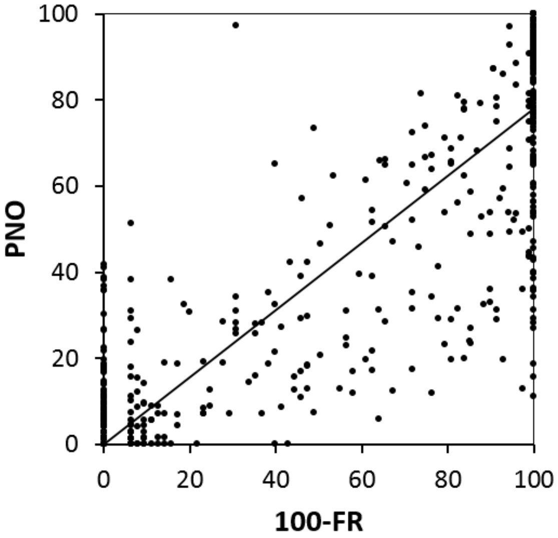

Fig. S2: Correlation between inverted fouling resistance (100-FR) and percentage net-aperture occlusion (PNO) with linear regression (100-FR = 0.781 x PNO; $R^2 = 0.90$, $p < 0.001$).

Biofouling composition

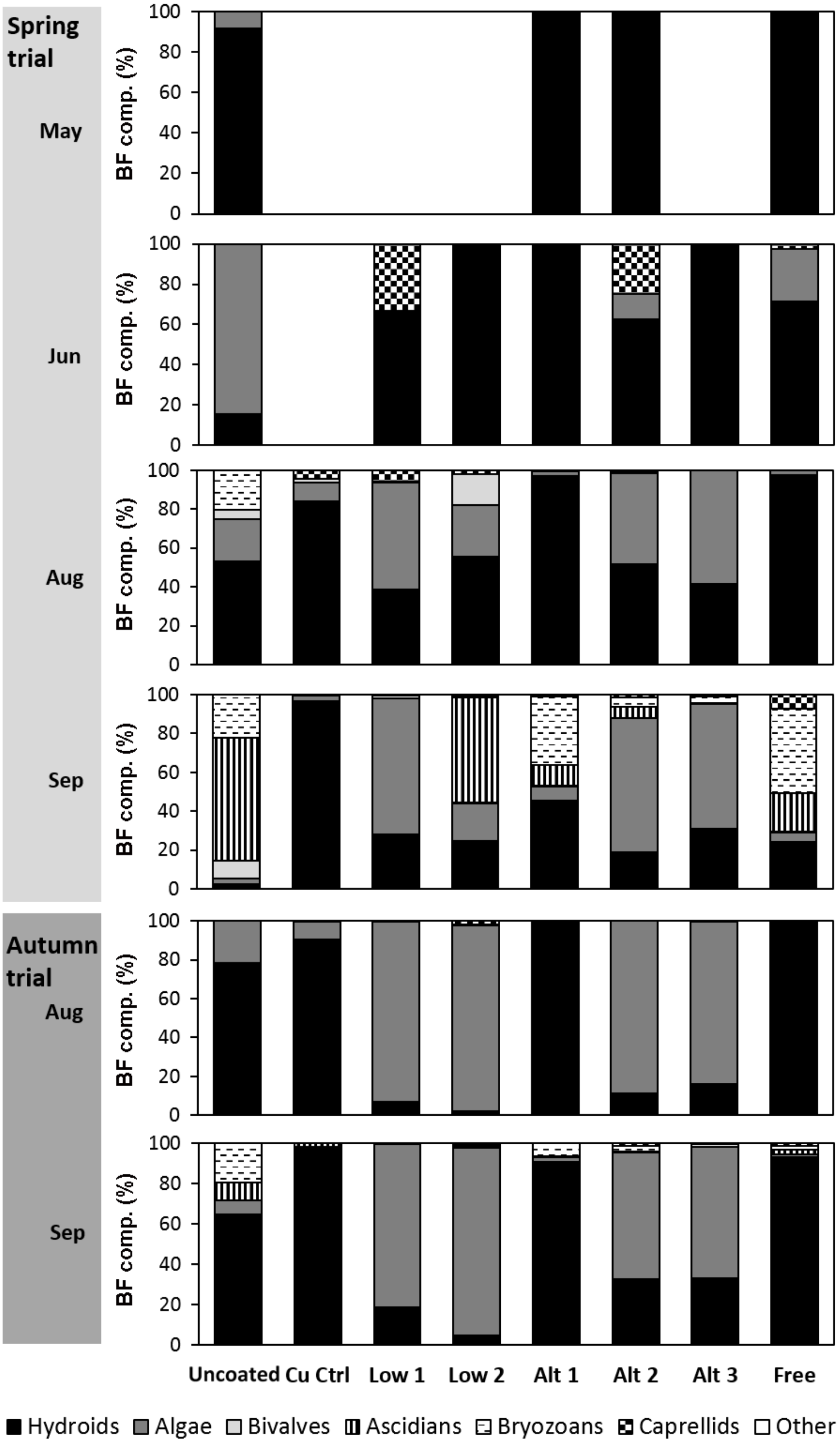

Fig. S3: Monthly biofouling composition (BF comp.) on nets with eight different coatings immersed for six months ('Spring trial') and three months ('Autumn trial'), depicting percentage cover of hydroids, algae, bivalves, ascidians, bryozoans and caprellids. There was no biofouling present in April. No data could be collected in July.

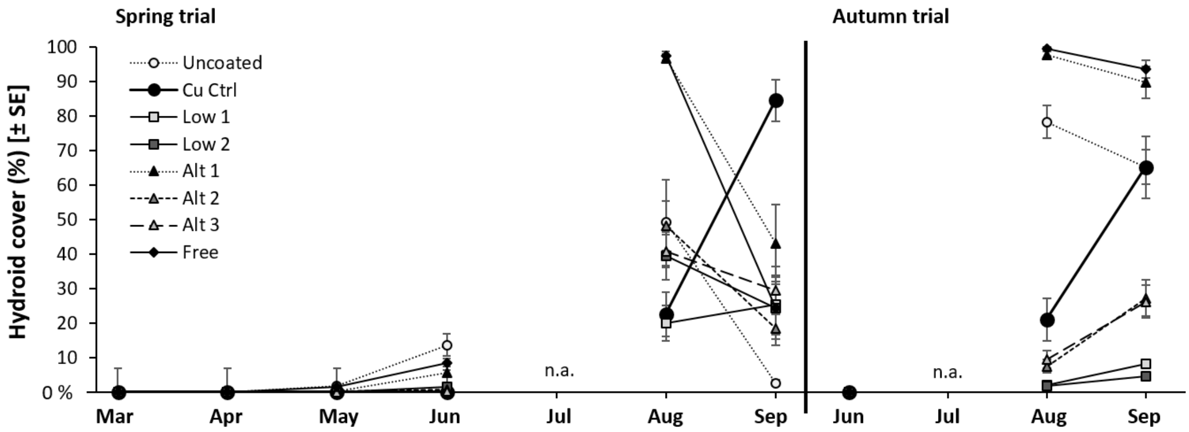

Fig. S4: Hydroid fouling measured as average % cover (± standard error) on eight different coatings in the 'Spring' (six months duration) and the 'Autumn' (three months duration) trial. No data could be collected in July.

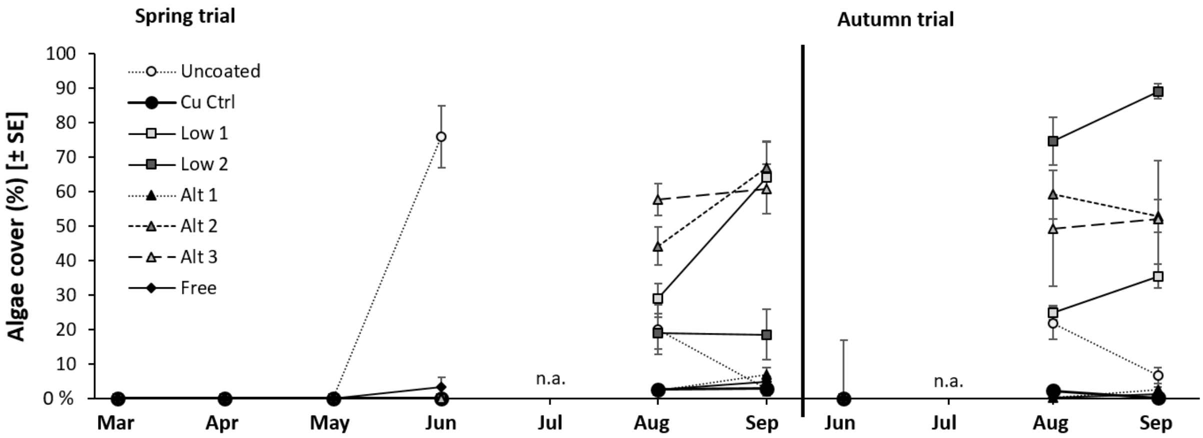

Fig. S5: Algal fouling measured as average % cover (± standard error) on eight different coatings in the 'Spring' (six months duration) and the 'Autumn' (three months duration) trial. No data could be collected in July.