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Rubber net mesh reduces scale loss during routine handling of farmed Atlantic salmon (Salmo salar)

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Abstract

*Atlantic salmon (*Salmo salar*) are an economically and ecologically important fish species that interact with humans during farming, fishing and research operations. Routine handling in nets exposes fish to mesh and causes scale loss. To promote welfare and experimental refinement, a study was performed in a controlled environment to investigate the effect of net mesh type (rubber-coated or standard knotless, both bag volumes* circa *7 l; mesh size: 6 mm) and the number of fish per net (capture density) on scale loss. Up to three large adult salmon (mean weight: 900 g) or 15 small smolts (mean weight: 145 g) were briefly captured in hand-nets during routine immersed-stock movement between tanks. Scales were recovered and counted from transportation containers, to establish a simple and rapid methodology. For both size grades, scale loss was generally proportional to capture density. For large adult salmon, scale loss significantly increased with capture density when knotless mesh was used, however the increase was less marked and not statistically significant for adults handled in rubber mesh. Small smolts also demonstrated significantly reduced scale loss when handled with rubber mesh, which increased gradually with capture density. In contrast, small smolts handled in knotless mesh showed greater scale loss as capture density increased. An overall reduction in scale loss with increased capture density was not shown, although the biomass loading per net used in this study were intentionally low (< 3.5 kg). This method suggests a low-tech and rapid approach to quantitatively compare net types and husbandry techniques and suggests a fundamental but simple improvement to salmonid handling in recreational and commercial operations. However, any correlation to conventional stress assays or behavioural observations remains to be established.*

Keywords: *animal welfare, aquaculture, fish health, mesh, salmonid, skin damage*

Introduction

Global production of farmed Atlantic salmon (*Salmo salar*) has doubled since 2000, and from 2012 has exceeded 2 million tonnes per annum, originating from at least 16 producer countries (Food and Agriculture Organisation [FAO] 2020). During this period, overall farm animal and fish welfare research has increased (Friere & Nicol 2019), including definition of welfare indicators specifically for farmed *S salar* (Santurtun *et al* 2018). In addition to aquaculture and associated research, wild salmonids such as *S salar* are a popular species for recreational angling (Olaussen 2016), interact with commercial fishing gear (Veneranta *et al* 2018; Cook *et al* 2019) and represent a commonly sampled environmental indicator species (eg Malcolm *et al* 2019).

These activities involve capture, handling and transport, usually requiring interaction with net mesh which causes the loss of protective mucus and scales. This may lead to pathogen invasion and osmoregulatory stress (Brydges *et al* 2009; Cook *et al* 2019) and, potentially, internal bruising (Bord Iascaigh Mhara [BIM] 2020). Commercial fishing (Olsen *et al* 2012), recreational angling (Butcher *et al* 2010), handling in aquaria (Brydges *et al* 2009), holding and transportation of stock (Portz *et al* 2006) and man-made barriers in watercourses (eg Brackley *et al* 2018) have all contributed to skin damage in fish. Studies have commissioned experimental scale and mucus removal treatments, demonstrating a correlation with the severity of osmoregulatory stress, infection and mortality rate in herring (*Clupea harengus*; Olsen *et al* 2012) and *S salar* smolts (Svendsen & Bøgwald 1997; Zydlewski *et al* 2010). This suggests that handling techniques which minimise scale loss during routine handling of *S salar* would be beneficial to fish health and welfare. Additionally, optimising handling is an example of experimental refinement which can improve the outcome of experiments by reducing stress, disease and injury, and hence variation (Brydges *et al* 2009).

Optimisation of handling techniques has included investigation of different net mesh types in field studies, working alongside recreational angling of wild fish. Subsequently, this has informed husbandry techniques by recording the presence of fin and skin damage (including grading the levels of scale loss). Study species have included brook trout (*Salvelinus fontinalis*; Lizeé *et al* 2018), largemouth bass (*Micropterus salmoides*) and northern pike (*Esox lucius*) (both Colotelo & Cook 2011), eastern sea garfish (*Hyporhamphus australis*; Butcher *et al* 2010), dusky flathead (*Platycephalus fuscus*; Butcher *et al* 2008), barramundi (*Lates calcifer*; De Lestang *et al* 2008) and bluegill (*Lepomis macrochirus*; Barthel *et al* 2003). There appears to be additional factors defining the extent of injury, such as species-specific effects (due to behaviour or morphology; Barthel *et al* 2003; Colotelo *et al* 2011) and different net mesh size (impacting upon fin damage; Lizeé *et al* 2018). Indeed, salmonids appear more sensitive to handling stressors compared to other taxa (Brydges *et al* 2009), particularly during smoltification (Carey & McCormick 1998; Zydlewski *et al* 2010; Santurtun *et al* 2018) with the species requiring several weeks to complete wound healing (Sveen *et al* 2019).

Measuring scale loss quantitatively by direct counting could be a simple and rapid method to evaluate husbandry techniques. In addition to studying the effect of mesh type on scale loss in a controlled environment using stock of similar size, studies would benefit from investigating more than one fish per net for applications other than recreational angling. Finally, species-specific studies need to confirm results are applicable to *S salar*, a particularly important and valuable species that interacts with humans across several commercial and recreational sectors, in both fresh and marine environments. Therefore, the aims of this study were: i) to establish methodology of scale recovery and analysis; ii) to compare scale loss between net capture densities (the number of fish per net scoop); and iii) to compare scale loss between mesh types (rubber-coated or standard knotless) of similar mesh size and net bag volume.

Materials and methods

Ethical statement

Modest scale loss in Atlantic salmon is routinely observed during stock management (ie movement, counting and grading of non-experimental stock between origin and destination tanks) at the Marine Environmental Research Laboratory (MERL) at the Institute of Aquaculture, Stirling University, UK. Under a standard internal protocol, salmon are removed from water in hand nets and immersed for less than 2 s prior to immersion and sedation in circular containers. Stock are swiftly moved to destination tanks and monitored until recovery (a regain of balance and normal swimming behaviour). The current study simply counted lost scales at opportune moments during routine fish stock management (ie the primary purpose of fish handling was never to collect data). Ethical approval was approved by the University of Stirling AWERB (application [18/19] 184) as a 'non-ASPA' study. The capture densities used were low, not exceeding normal practice with relation to fish size or biomass (less than *circa* 3.5 kg in a *circa* 7 l net bag volume), whilst the rubber net mesh was likely to meet or exceed the performance of the conventional knotless mesh.

Salmon stock

Diploid Atlantic salmon smolts were routinely transported to MERL using a commercial fish transporter courier following a confirmatory saltwater tolerance assay. Adult smolts used in this study arrived at *circa* 130 g (Inchmore Hatcheries, Marine Harvest McConnell Ltd, Glenmoriston, UK) in August 2019, whilst small smolts arrived at *circa* 80 g (Niall Bromage Freshwater Research Unit, NBFRU, Institute of Aquaculture, Stirling University, UK) in February 2020. Feed was provided by automated feeders during the hours of daylight (0500–2300h) at 1.4% decreasing to 0.8% biomass using successive grades of 2.0, 3.0 or 4.5 mm feed (Incio Plus or BioMar Pearl, BioMar UK Ltd, Grangemouth, UK) as smolts underwent full smoltification and increased in weight.

Aquarium system and salmon husbandry

On arrival, up to 1,000 smolts were housed in circular 3 m diameter tanks (*circa* 6,000 l) with a slight clockwise current. Seawater was pumped to shore and passed through a drum filter into a header tank, and subsequently gravity-fed through a sand filter consisting of 100 µm membrane filters with final UV sterilisation. Salinity and temperature varied gradually throughout the year (31–33‰, 7–15°C, respectively), with dissolved oxygen and photoperiod maintained within strict limits (85–105% and 18 h light: 6 h dark, respectively). Stock smolts were transferred periodically to other tanks to manage stock density and facilitate size grading, so that the total length of smolts never exceeded half a tank radius, and stocking density did not exceed 20 kg m^{-3}.

Stock movement and data collection

Salmon that had not been handled for at least 80 days and had undergone smoltification were removed from tanks using hand-nets during April–May 2020 (salinity 32–33‰, temperature 9–11°C). Nets consisted of either a knotless mesh typically used by the aquaculture sector (FIAP Auminium Profinet, Sterner AquaTech UK, Inverness, UK) or a rubber-coated mesh designed for recreational angling (MAP scoop landing net, H Young [Operations] Ltd, Newbury, UK), both 6 mm mesh size, *circa* 40 cm depth, attached to a net frame of *circa* 150 cm circumference.

Adult salmon (total: n = 144; weight: 902 [± 19.1] g; range of capture densities: 1–3 per net scoop) or small smolts (total: n = 376; weight: 139 [± 5.0] g from a subsample of 25 individually netted fish; range of capture densities: 1–15 per scoop) were removed from tanks in a rapid, fluid motion, remaining emersed in the net for *circa* 2 s. Alternate mesh types were used between each net scoop to reduce bias. Salmon were placed into meticulously cleaned cylindrical containers, containing 12–150 l seawater, to provide small salmon with at least 10 l, and large adults at least 15 l of physical space to minimise collision. Salmon were sedated during movement (Tricaine methanesulfonate, 100 mg l^{-1} seawater, Pharmaq Ltd, Fordingbridge, UK) and swiftly moved in containers on trolleys from origin to destination tanks, as sedation occurred. To assist calculation of accurate feed ration and aid recovery from anaesthesia,

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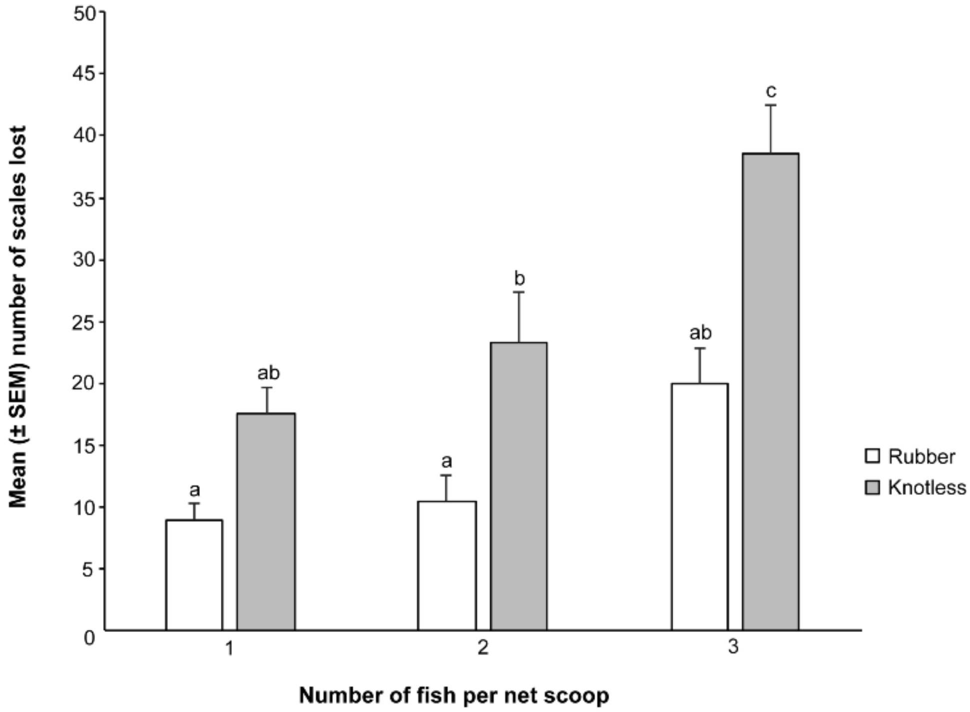
Figure 1



Mean (± SEM) scale loss in adult salmon (*Salmo salar*) following brief handling in hand-nets, composed of either rubber or knotless mesh, at capture densities of 1, 2 or 3 salmon per net. Differing superscripts denote significant differences between treatments at $P < 0.05$, (n = 12).

salmon were additionally weighed (adults individually, small smolts individually or batch weighed) in a tared circular container containing seawater. Finally, salmon were placed gently and counted into destination tanks and monitored until recovery. No salmon died or required culling following routine movement between tanks.

To count scales, the net was agitated in the unstocked container to dislodge any adhering material. The container was inspected under bright illumination and all scales removed and counted using a large pipette. The anaesthetic water was refreshed frequently to maintain water quality and clarity. The number of scales lost per fish was then calculated according to mesh type, capture density, fish number and biomass.

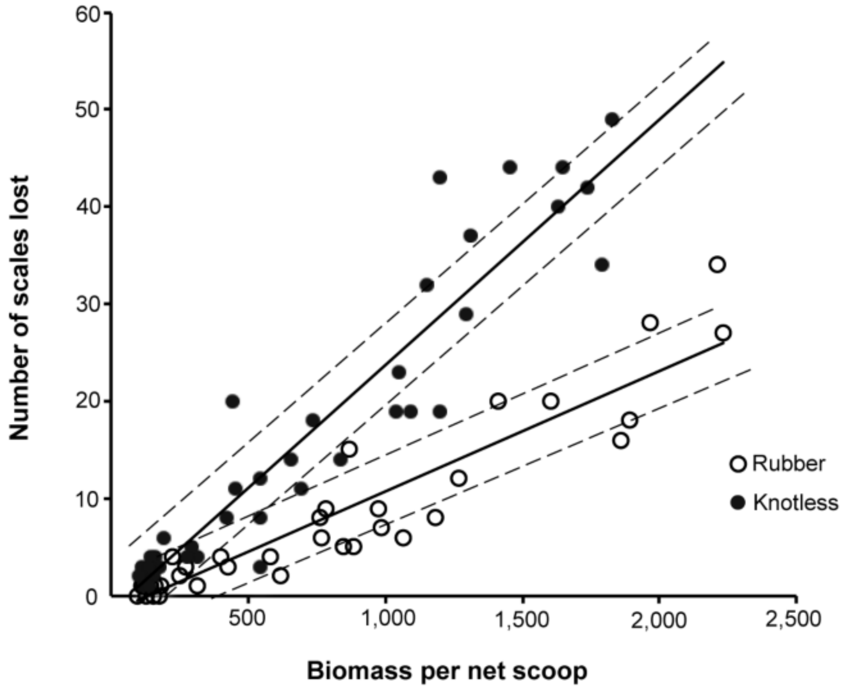
Data analysis

All data were analysed using GraphPad-Prism (GraphPad Software Inc, San Diego, USA) and are shown as means (± SEM) (other than regression where a 5% confidence limit is shown surrounding regression lines). Data were confirmed for normality and homogeneity of variances (Kolmogorov-Smirnov test; Bartlett's test, respectively) prior to further analysis. Large adults, with a limited range of capture densities, showed a respectable correlation between fish number and biomass ($r^2 = 0.6779$) whilst for smaller salmon with a wider range of capture densities, a very strong correlation was confirmed ($r^2 = 0.9626$) prior to further analysis. Capture density was recorded as net biomass, which was highly correlated to scale loss (linear equation for rubber mesh, $r^2 = 0.88$, $y = 0.01243 \times x - 1.730$; knotless mesh, $r^2 = 0.89$, $y = 0.02531 \times x - 1.652$). For larger adult salmon, the impact of mesh type, capture density (and interaction between both independent variables) upon scale loss (continuous dependent variable) were analysed using two-way ANOVA with a Tukey *post hoc* test. For smaller smolts, data were analysed using regression (scale loss vs biomass) to ascertain the equation of a straight line, 95% confidence limit and goodness of fit (r^2). An F-statistic was employed to ascertain slopes were significantly different between mesh types. Individual large and small salmon were not compared within mesh types due to the difference in stock origin, life history and possible 'tank effect.' However, scale loss for individual small smolts (n = 12–13 within the same data set) were compared between mesh types using an unpaired Student's t-test.

Results

For large adult salmon, scale loss increased with capture density for both mesh types (Figure 1). Whilst this was not significant for rubber mesh, comparing capture densities of 1, 2 or 3 fish per net ($P > 0.05$), the numbers of scales lost for knotless mesh was significantly higher for three fish per net compared to other capture densities ($P < 0.01$–0.001). Between mesh types, rubber mesh reduced scale loss compared to knotless mesh, with respect to discrete capture densities. While this was not significantly different for individual fish ($P > 0.05$), it was significant for two fish per net and highly significant for three fish per net ($P < 0.05$ and

Figure 2



Scale loss in salmon smolts (*Salmo salar*) following brief handling in hand-nets, composed of either rubber or knotless mesh, at capture densities of between 1 and 15 fish per net (corresponding to a biomass between 0.1, and 2.3 kg). Dashed lines represent 95% confidence limit, (n = 39).

$P < 0.001$, respectively). Mesh type and capture density were responsible for a similar proportion of the total variation in scale loss (*circa* 24% each; $F = 16.48$ and 30.99, respectively; $P < 0.001$) with insignificant interaction between either variable (*circa* 2%; $F = 1.42$, $P > 0.05$).

For smaller smolts, the number of lost scales increased with capture density (Figure 2; rubber and knotless slopes both significantly non-zero; $F = 265.7$ and 307.0, respectively; $P < 0.001$). However, significantly fewer scales were lost for smolts handled using rubber, compared to knotless mesh (Figure 2; difference between slopes extremely significant; $F = 66.14$, $P < 0.001$). Individual smolts (ie capture density of one) showed no significant difference in scale loss between mesh types (rubber: mean 0.77 scales per fish; knotless: mean 2.83 scales per fish; Student's *t*-test; $P > 0.05$).

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Discussion

This study suggests a simple, relatively rapid method to recover and analyse scales from *S salar* and suggests a low tech, quantitative comparison of subjective handling methods for salmon (although, ethically, these should reasonably meet or exceed current practice). This method is minimally invasive, in the respect that scales are unavoidably removed during routine husbandry or sampling of this species.

Previous studies demonstrated that individually angled fish, handled in rubber-coated or knotless mesh, reduced skin damage compared to more obsolete knotted mesh. This was likely due to the less abrasive nature and improved physical support in rubber and knotless types (Barthel *et al* 2003; Colotelo & Cooke 2011; Lizeé *et al* 2018). This idea is further supported as fish variously exposed to non-netted (control) treatments (eg Barthel *et al* 2003), manipulated only using hands (Lizeé *et al* 2018) or handled immersed in a scoop (Brydges *et al* 2009) demonstrated less skin damage and a reduced stress response compared to fish handled in nets. In the current study, we sourced two nets of differing mesh type, but of the same mesh size and similar bag volume, using graded salmon maintained in a controlled environment. Having removed any confounding factors as far as is reasonable, the current study illustrates that for individual salmon handled in nets, rubber mesh significantly reduced scale loss (compared to knotless mesh). This is an important finding for activities that involve routinely handling individual salmon (eg angling, environmental monitoring, manipulation of brood stock in hatcheries or aquaria) or indeed other uses of mesh (eg jump-out mesh over tanks, seine-netting in ponds).

Since both large adults and small smolts had different stock origin and life history in this study, they were not compared. However, further investigation of the relationship between mesh size and fish size would be of merit, almost certainly

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corresponding to fish weight and strength of movement in the net (Barthel *et al* 2003; Olsen *et al* 2012). In terms of net mesh, for instance, angling in fast-flowing rivers necessitates the use of large mesh sizes, to reduce drag during operation (eg salmonid 'Gye nets') whilst manipulation of stock for aquaculture may require movement of an array of different fish sizes commensurate with stock growth. More recently, the type of plastic compound used in a variety of knotless meshes has shown the potential to reduce skin damage in yellowtail tetra (*Astyanax altiparanae*; Alvarez-Rubio *et al* 2020) which is of interest to inform overall net construction and design. To inform practical use and uptake it would also be of interest to compare cost, longevity, mesh degradation and disinfection efficacy of rubber and knotless meshes, alongside public and commercial dissemination of the results.

The current study is apparently novel since it has investigated handling of multiple fish in hand-nets. Loss of scales, perhaps predictably, was proportional and increased with capture density (number of fish per net) for both size grades. However, for rubber mesh, the observed increase in scale loss was much reduced compared to knotless mesh. Overall, this suggests that at the point of capture in nets, the effect of mesh abrasion on scales is reduced with single or low fish numbers (or biomass). The observation of 'net roll' (escape behaviour during brief immersion) exposes and abrades fish flanks against net mesh, causing scale loss. Netting single fish (or low capture densities) apparently reduced, whilst higher capture densities promoted, this abrasive effect. Perhaps this can be explained by the increased pressure or interaction with other fish, as capture density increased. This may be more marked with larger adults, as they are physically heavier, more powerful and demonstrate more aggressive net roll. The capture densities were quite modest with respect to net bag volume (under 3.5 kg), and further studies could investigate the relationship between scale loss at higher capture densities, or the effect of water temperature on the intensity of net roll. The practical implications suggest that for angling or environmental monitoring, where it is probable that only one fish will be handled in nets, rubber mesh could somewhat reduce the impact of handling stress and promote survival following release. For aquaculture or research operations that are likely to handle more than one fish, it becomes increasingly important to use rubber mesh for stock welfare and experimental refinement.

Animal welfare implications

This study supports a fundamental change to salmonid husbandry techniques, by suggesting the use of rubber meshes during routine handling, to reduce scale loss. Use of rubber mesh can safeguard salmon integument, and thus promote resilience to infection or osmoregulatory stressors. Procurement of rubber mesh is a relatively reasonable and cost-effective approach that could be adopted internationally, across several commercial and recreational sectors that routinely interact with potentially millions of salmonids. The scale counting technique also suggests a relatively rapid, low-tech approach to compare subjective handling protocols, and could inform further improvements in net design, construction or use.

Conclusion

This study has met the outlined aims by suggesting a simple scale recovery technique for *S salar*. Albeit in terms of scale loss only, rubber-coated mesh was found to be superior to knotless mesh for individual salmon, and this became more marked at higher capture densities for both size grades examined in this study. Although these initial findings support the routine practice of brief, high capture densities in hand-nets as a reasonable husbandry technique, these data only refer to scale loss. The findings would benefit from establishing any correlation with existing stress assays or physical impacts associated with high capture densities in nets, such as mucus removal or fin and eye damage (fluroscein, scanning techniques and image analysis, eg Butcher *et al* 2010; Colotelo & Cooke 2011; Alvarez-Rubio *et al* 2020), bruising or bleeding (BIM 2020; Uhlmann *et al* 2020) and other conventional methods of measuring systemic stress (quantitative behavioural observations or cortisol stress assays, eg Brydges *et al* 2009; Sadoul & Geffroy 2019).

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