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# Prediction of goldsinny wrasse (*Ctenolabrus rupestris*) minimum size required to avoid escape through salmon (*Salmo salar*) farm nets

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#### ABSTRACT

The salmon (Salmo salar) aquaculture industry has used various practices to treat salmon louse (Lepeophtheirus salmonis) infections, including mechanical, thermal, and chemical treatments. However, due to the environmental and fish welfare issues, and limitations associated with these practices, the use of cleaner fish has become a popular complementary delousing method. The goldsinny wrasse (Ctenolabrus rupestris) is the smallest among the delousing species used, which makes it most susceptible to escape through the salmon cage netting. Escape of goldsinny wrasse into the wild would lead to additional costs for farmers and could spread diseases and genetically contaminate wild stocks, as these cleaner fish are often transported over large distances from the capture site to the aquaculture site. The Norwegian aquaculture industry sets the goldsinny wrasse minimum size for use with different salmon cage mesh sizes using guidelines based on simple penetration tests that do not consider the potential compressibility of fish or changes in mesh state due to factors such as sea conditions and maintenance operations. Using morphological measurements of 100 fish, results of 47,800 penetration tests, and computer simulation, we predicted the minimum sizes required to avoid escape of goldsinny wrasse through and array of salmon farm nets. Our results show that the minimum stocking sizes for goldsinny wrasse in the current guidelines used by the industry may pose escape risk. Further, our findings illustrate the importance of considering fish compressibility and mesh state when estimating the fish minimum sizes required to avoid escape via cage netting mesh penetration. Finally, the results presented here provide the salmon farming industry with new guidelines that will contribute to reducing goldsinny wrasse escape from the cages and ultimately lessen the potential environmental consequences of such escape. For the two most typical mesh-size used by the aquaculture industry in Norway i.e. 30 and 50 mm and under the worst-case scenario i.e. slack state, the minimum recommended lengths for goldsinny wrasse would be  $\sim$ 128 ( $\sim$ 28) and  $\sim$  206 mm ( $\sim$ 123 g). These sizes represent an underestimation of 16 and 24% in the sizes of goldsinny wrasse necessary to avoid escape compared to the current guidelines based on stiff meshes.

#### 1. Introduction

Aquaculture of Atlantic salmon (*Salmo salar*) has become an important industry for several countries globally and in the North Atlantic in particular. In Norway (Olaussen, 2018), salmon production in 2019 reached 845,000 t and a value of 5 billion Euros (Norwegian Seafood Council, 2019). However, the rapid development and expansion of the industry over the last 30 years has led to questions about its effects on marine ecosystems (Naylor et al., 2000), sustainability (Olesen et al., 2011), and fish welfare (Hvas et al., 2021). Issues such as the spreading of parasites and diseases (Aaen et al., 2015) and escapees that interbreed with local fish populations (Karlsson et al., 2016) are specific challenges that the industry has not yet fully resolved (Taranger et al., 2015).

The salmon louse (*Lepeophtheirus salmonis*) is a common parasite of wild Atlantic salmon and has become a major problem for the development of the salmon aquaculture industry over the last two decades. At low densities, infection by this parasite can have negative effects on fish, such as lower growth, but more heavily infected salmon experience

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severe skin damage and stress that can eventually lead to downgrading at harvest, rapid on-site multiplication to unacceptable/damaging levels for fish health and welfare, or even death (Glover et al., 2004; Ugelvik et al., 2017). Thus, the salmon farming industry's efforts to control lice densities and the associated economic costs have been considerable (Costello, 2009; Torrissen et al., 2013; Abolofia et al., 2017). Interventions (remedies) to control lice infections of salmon include thermal and/or mechanical treatments and chemical baths. However, the majority of these remedies have associated problems, such as drug resistance in sea lice, negative impacts on fish welfare like mechanical/ metabolic damage, and environmental issues. Therefore, the use of cleaner fish that feed on lice has become a widespread delousing method in countries such as Ireland, Canada, the Faroe Islands, Norway, and Scotland (Bolton-Warberg, 2017; Gonzalez and de Boer, 2017; Brooker et al., 2018; Foss et al., 2020).

The Norwegian salmon farming industry uses lumpfish (*Cyclopterus lumpus*) and four species of wrasses (Ballan wrasse (*Labrus bergylta*), goldsinny wrasse (*Ctenolabrus rupestris*), corkwing wrasse (*Symphodus melops*), and rock cook wrasse (*Centrolabrus exoletus*)) as cleaner fish (Skiftesvik et al., 2015; Gonzalez and de Boer, 2017; Overton et al., 2018). The use of wrasses for salmon lice control in Norway began in the late 1980s (Bjordal, 1988, 1990, 1991), and their use became prevalent in the early 2000s. In 2017, the industry used over 21 million wrasses for delousing purposes, and goldsinny wrasse was the most applied species with over 11 million individuals released in salmon farming pens (Norwegian Directorate of Fisheries, 2021). Currently, large efforts are made to farm some of the cleaner fish species (e.g. ballan wrasse and lumpfish) (Leclercq et al., 2013; Skiftesvik et al., 2013) with the aim of minimizing the exploitation of wild resources. However, the industry still relies on wild fish to cope with the prevailing large demand.

The goldsinny wrasse is the smallest of the wrasse species used by salmon farmers. It ranges in length from 60 to 200 mm and normally reaches 100-120 mm (Skiftesvik et al., 2015). This species is relatively abundant along the coast of Europe (the northern limit is in central Europe), and it was identified in the early 1990s as an effective delouser (Bjordal, 1991). In addition to its delousing efficiency and availability, its adaptability to changes in water temperature (Sayer and Davenport, 1996) and small size make it one of the species preferred by salmon farmers for salmon smolt delousing (Gonzalez and de Boer, 2017). However, the small size of goldsinny wrasse allows it to more easily escape through salmon farming cage netting compared to other wrasse species. Escape leads to economic losses for salmon farmers, and escapees from farming cages can spread diseases (Aaen et al., 2015; Taranger et al., 2015). Escapees would also lead to lack of efficiency and predictability of the delousing method. Furthermore, if escapees are individuals that have been translocated over substantial distances, which is quite a common practice between the south and central coastal areas in Norway (Jansson et al., 2017), their mixture with the local fish communities can pose a serious environmental threat via genetic introgression (Karlsson et al., 2016; Faust et al., 2018).

Previous studies suggested that wrasses can escape through cage netting panels (Deady et al., 1995; Faust et al., 2018). To avoid this potential fish loss, farmers use guidelines that determine the smallest fish sizes to be used for each specific mesh size (Sigstadstø, 2017). However, these guidelines are based on only a few specific mesh sizes. Although commonly used nettings consist of square meshes of 30–50 mm (Moe et al., 2007), mesh size can vary greatly between salmon aquaculture sites because it is not regulated. Moreover, these guidelines are based on mesh penetration tests that do not account for potential variability in condition and compressibility of fish of different sizes (Harboe and Skulstad, 2013) or the variability in mesh state.

The meshes in the salmon cage netting can change shape and/or state (from stiff to slack) during netting manipulation in maintenance operations or due to variation in sea conditions (e.g., currents, waves) (Huang et al., 2006; Lader et al., 2008; Sistiaga et al., 2020). The latter has become especially relevant in recent years, as there is a growing interest in moving salmon farming sites to more exposed areas (Jónsdóttir et al., 2019). Alterations in mesh state and mesh shape can affect the penetrability and consequently the escape risk of fish through netting meshes (Herrmann et al., 2016a; Sistiaga et al., 2020), but mesh penetration tests conducted to date for goldsinny wrasse have not considered any of these aspects. Ignoring these scenarios can lead to underestimation of the sizes of this species that can be used safely in salmon aquaculture cages without risk of escape. Therefore, the aims of this study were to assess and predict how mesh size and mesh state affect the sizes of goldsinny wrasse that can be used safely by the aquaculture industry without risking escape and to provide the industry with complete guidelines for a wide range of square mesh sizes and mesh states.

#### 2. Materials and methods

## 2.1. Effect of mesh shape and state vs. goldsinny wrasse size and morphology on potential escape through cage netting

If a goldsinny wrasse is to pass through a netting panel, two conditions must be met. First, the fish needs to contact the netting at an orientation that gives it a size-dependent possibility of passing through the mesh of the netting (Sistiaga et al., 2010). Second, the fish needs to be morphologically able to pass through the mesh. Therefore, the main factors to consider when assessing the escape risk of goldsinny wrasse from salmon farming cages are mesh size, shape, and state in relation to fish size, morphology, and tissue compressibility.

The industry carries out penetration tests to investigate the minimum size at which fish cannot escape from certain net mesh sizes (Harboe and Skulstad, 2013). In these trials, individuals of a range of sizes are tested on the stretched (stiff) square meshes (Fig. 1a) of the cage to see if they are able to pass through them. However, the meshes in the netting of a salmon cage are flexible, which means that they can be deformed and adopt different shapes depending on the magnitude and direction of the forces to which they are exposed. These forces depend on factors such as weather and sea currents (Huang et al., 2006; Lader et al., 2003, 2008), meaning that the mesh state in the netting of cages in exposed locations changes frequently, and the meshes often tend to be in semi-slack and slack states (Fig. 1). Furthermore, many of the operations carried out in fish farming involve manipulation of the cage netting, which again results in the meshes adopting semi-slack or slack states. In a square mesh net panel hanging at sea, the load in the netting is on the vertical bars due to gravity, meaning that the horizontal bars are to a certain extent tensionless and therefore deformable. When the meshes are semi-slack, the fish in the cages could potentially deform the horizontal bars in the meshes while squeezing through them and finally escaping (Fig. 1b). Alternatively, when the sea state and water currents are strong enough to deform the netting, the load on the vertical bars would disappear, making the meshes slack and deformable in all directions (Fig. 1c). It is likely that slack meshes and at least some states of semi-slack meshes would lead to a higher risk of escape for goldsinny wrasse simply because the mesh totally (slack) or partially (semi-slack) deforms when adjusting to the shape of the wrasse trying to squeeze through it. Therefore, assuming a stable stiff state of the meshes in cage netting could lead to a serious underestimation of the minimum fish size required to avoid the risk of escape.

Two factors determine the maximum size at which a goldsinny wrasse individual would be able to squeeze itself through a mesh. The first is the deformability of the meshes in the netting and the second is the tissue deformability or compressibility. In Fig. 1, only a goldsinny wrasse with a compressibility level illustrated by the green cross section (CS) would be able to pass through the square meshes in each of the mesh states (Fig. 1a–c). Thus, to quantify the potential risk of escape for a goldsinny wrasse through a specific netting, it is necessary to consider different potential netting scenarios in combination with the morphology and cross-sectional compressibility of the fish.



Fig. 1. Mesh penetration of a goldsinny wrasse represented by its cross-section (red = uncompressed, green = maximum compression) through a (a) stiff, (b) semislack, and (c) slack mesh.

#### 2.2. FISHSELECT methodology and data collection

FISHSELECT (Herrmann et al., 2009, 2012) is a framework of methods, tools, and software developed to determine if a fish can penetrate a certain mesh or defined shape. The method has been widely used to study fishing gear size selectivity (the size-dependent probability for escape/retention of fish) (Krag et al., 2011; Sistiaga et al., 2011; Herrmann et al., 2016a, 2016b; Tokaç et al., 2016; Tokaç et al., 2018; Cuende et al., 2020). In this study, we applied this method for the first time to predict the escape risk of goldsinny wrasse through salmon farming cages.

To study the size selectivity of a species using this method, both the FISHSELECT software and specific measuring tools are needed (Fig. 2). Through computer simulation, the FISHSELECT methodology estimates the risk of escape by comparing the morphological characteristics of a particular fish species and the shape and size of the selection devices of interest. The following subsections briefly describe the different steps needed to use FISHSELECT. A more thorough description of the method can be found in Herrmann et al. (2009, 2012).

#### 2.2.1. FISHSELECT morphometric data collection

In addition to measuring the length of each goldsinny wrasse individual included in the study, we measured the cross-sectional morphology of each fish. To obtain the correct morphometric measures, it is important that the shape of the fish measured is not affected by dehydration, depressurization, rigor mortis, or any other factor that could alter the measurements. Therefore, the goldsinny wrasse used in the trials were handpicked in batches of 4 or 5 fish and killed with an overdose of MS 222 anaesthetic just before use. The aim of FISHSELECT is to make predictions about mesh penetration probability for the widest possible range of fish sizes. Thus, the method requires that the morphometric characteristics of the largest possible size range are measured. In the current study, apart from the condition of the goldsinny wrasse selected, the only other selection criteria for fish was that the fish included in the trials covered the widest possible size range.

The two cross-sections measured for each goldsinny wrasse were selected for their potential to determine fish passage through a mesh: cross section 1 (CS1) was located directly behind the operculum, and cross section 2 (CS2) was located at the point of the maximum transverse perimeter (i.e., the foremost point of the dorsal fin) (Fig. 2). CS1 represents the point at which the bony structure in the head has its maximum girth, whereas CS2 was selected because it represents the point with maximum girth of the fish overall. Thus, these two CSs were expected to be the decisive CSs for mesh penetration of goldsinny wrasse. The two cross-sections were measured using a sensing tool called a morphometer. The shapes formed in the morphometer were then scanned to obtain digital images of the contours using a flatbed scanner (Fig. 2).

Models (i.e., numerical representations of parametric shapes) of the digitized cross-sectional images obtained for each wrasse were developed. For each CS, we considered five different shape models: ellipse, flexellipse1, flex drop, super drop, and ship (see Sistiaga et al. (2020) for further information about these five models). The models were selected based on previous experience with other fish species. The Akaike information criterion (AIC) value (Akaike, 1974) and R<sup>2</sup>-values were calculated for each of the five models for both CS1 and CS2 (see Tokaç et al. (2016) for further details about this process). For each of the two cross-sections, the shape model with the lowest mean AIC value was



Fig. 2. The first and second row describe CS1 and CS2. The third row shows the process of digitization of the shapes measured by the morphometer.

chosen. The mean  $R^2$ -value was applied to judge how well the selected models on average described the cross-sectional shapes of goldsinny wrasse. The relationship between total length and cross-section shape parameters was modelled for the most suitable shapes found for CS1 and CS2, respectively.

#### 2.2.2. Fall-through experiments

Immediately after the morphology of each fish was measured, fallthrough experiments were conducted to determine whether the fish could physically pass through an array of stiff mesh shapes perforated in 5 mm nylon-plate templates (Fig. 3). Only the force of gravity was used to simulate the attempted penetration of goldsinny wrasse through the mesh (Fig. 3). The set of mesh templates used in this experiment consisted of 478 different shapes representing mesh sizes from 20 to 245 mm. The shapes included diamonds (252 meshes), hexagons (98 meshes), and rectangles (128 meshes) and were identical to those described by Tokaç et al. (2016). All fish were presented at an optimal orientation for mesh penetration for each of the 478 meshes in the templates. Penetration (Yes) or retention (No) was recorded for each fish (see Herrmann et al. (2009) for further details about the procedure). The purpose of the fall-through experiments was to estimate the maximum compressibility for a fish trying to squeeze itself through a mesh (see Herrmann et al. (2009) for further details).

#### 2.2.3. Simulation of mesh penetration and selection of a penetration model

The shape and compressibility of a fish determine whether it will be able to pass through a mesh. The penetration models implemented in FISHSELECT simulate the compressibility of each fish at each crosssection. Visual and tactile inspection of the deformability of goldsinny wrasse revealed that the dorsal and ventral compressibility of this species may differ. Therefore, a model that allows asymmetrical compression for both CS1 and CS2 was applied. This model was previously used for redfish (*Sebastes* spp.) by Herrmann et al. (2012) and includes the estimation of three parameters representing the dorsal, lateral, and ventral compressibility of the fish. The potential compressibility of the fish at an arbitrary angle around the fish cross-section was then modelled by linear interpolation between the potential compressibility (dorsally, laterally, and ventrally) of the fish at each cross-section (see Herrmann et al. (2012) for further details).

To establish an optimal penetration model for goldsinny wrasse, each CS1 and CS2 measurement, both individually and in combination, were tested with different compression models using different values for the assumed dorsal, lateral, and ventral compression. The penetration of the modelled CS1 and CS2 shapes of each fish through the 478 different mesh templates used in the fall-through trials was simulated using the FISHSELECT software. The purpose of these simulations was to estimate the compression potential of the cross-sections and to assess which crosssection combinations needed to be considered when estimating the potential for goldsinny wrasse to pass through meshes of different sizes and shapes. Models considering one cross-section at a time were created. For CS1 modelling, the dorsal, lateral, and ventral compression varied from 0 to 20%, 0 to 30%, and 0 to 30%, respectively, in increments of 5%. This resulted in a total of 245 penetration models for CS1. For CS2 modelling, the dorsal, lateral, and ventral compression varied from 0 to 30%, 0 to 20%, and 0 to 40%, respectively, in increments of 5%. This resulted in a total of 315 penetration models for CS2. In addition to the models run for each cross-section, 77,175 models in which CS1 and CS2 were combined were tested. Each compression model was used to simulate fall-through results for each of the meshes and fish used in the experimental fall-through data collection (Section 2.2.2) Using the FISHSELECT software, the results obtained from all penetration models were compared with the experimental fall-through results. This evaluation produced a value for the degree of agreement (DA) for each tested model, which expresses the percentage fraction of the fall-through



Fig. 3. Photo (a) shows a sample of two goldsinny wrasse used in the fall-through tests, (b-c) and (e-f) illustrate the fall-through procedure for different shaped openings, and (d) shows the different templates employed in the fall-through tests.

results in which the simulated process yielded the same result ("yes" or "no"). Among all of the models tested, the model with the highest DA was chosen for further analysis in FISHSELECT.

### 2.2.4. Modelling of mesh shapes for square meshes in fish farm cages during potential goldsinny wrasse escape attempts

Before we could predict escape risk for goldsinny wrasse through square meshes in salmon farm cages using the FISHSELECT methodology, an appropriate model for the semi-slack mesh state (Fig. 1b) and for the fully slack mesh state (Fig. 1c) was required. In the FISHSELECT simulation, the latter is directly modelled by the condition that a fish can escape if the circumference of its cross-section under maximum compression is less than the inner circumference of the mesh (twice the mesh size) it attempts to pass through. This is because in this mesh state the mesh will be fully distorted while the fish is passing through it. In semi-slack and partly open square meshes (Fig. 1b), the shape the mesh will become when a fish attempts to pass through it was approximated by a hexagonal shape in which the tensionless horizontal mesh bars are bent upwards and downwards (Fig. 4a-c). This approximation previously was applied successfully when modelling fish escape through square mesh codends in trawl and demersal seine fisheries for several species including cod (Herrmann et al., 2016a, 2016b), haddock (Krag et al., 2011; Herrmann et al., 2016b), red mullet (Tokaç et al., 2016), and hake (Tokac et al., 2018). It also was recently applied to estimate the escape risk of salmon smolt and lumpfish through salmon cage nettings (Sistiaga et al., 2020; Herrmann et al., 2021).

Two related measures are applied to describe the openness of a hexagonal modelled distorted semi-slack square mesh: the opening angle (OA) and the relative openness (OP). The latter quantifies the horizontal opening of the mesh (B) relative to the vertical opening (A) (Fig. 4a). Fig. 4d shows the relationship between OA and OP for hexagonal distorted square meshes. The relationship between OP and OA is:

$$OP = 100 \times \frac{B}{A} = 100 \times \sin\left(\frac{OA}{2}\right) \tag{1}$$

The stiff mesh scenario (Fig. 1a) is a special case for the hexagonal approximation of the semi-slack mesh when  $OA = 180^{\circ}$ , which

corresponds to an OP of 100%.

#### 2.2.5. Quantifying the escape risk

Based on the morphological description of CS1 and CS2 (Section 2.2.1), a virtual population of 2000 goldsinny wrasse with uniformly distributed length of up to 250 mm was created for the simulation of size selection. This upper size limit was selected because predictions for meshes up to 100 mm were desired. For all three mesh scenarios (Fig. 1), the risk of goldsinny wrasse escape was simulated for square meshes with a mesh size between 10 and 100 mm in increments of 5 mm. For the semi-slack scenario, which was approximated by a hexagon, OP values from 50 to 100% were used in increments of 5%. Using the identified goldsinny wrasse penetration model, a simulation was created to determine whether each individual in the virtual population could pass through the mesh in each of the mesh scenarios (stiff, semi-slack, slack). Likewise, for the standard application of the FISHSELECT method (Herrmann et al., 2009), a virtual size selection dataset for each mesh was obtained, which consisted of wrasse size-dependent counts of individuals (in 10 mm wide length classes) from the virtual population being retained (not able to pass through) and released (able to pass through), respectively. The traditional logit size selection model (2) then was fitted to the size selection data by maximum likelihood estimation to obtain the values for the model parameters L50 and SR (Wileman et al., 1996):

$$logit(l; L50, SR) = \frac{exp\left(\frac{ln(9)}{SR} \times (l - L50)\right)}{1 + exp\left(\frac{ln(9)}{SR} \times (l - L50)\right)}$$
(2)

where *L50* is the length corresponding to the goldsinny wrasse that have 50% probability of being retained, and the selection range (*SR*) is the difference between *L75* and *L25* (Wileman et al., 1996). Based on the obtained size selection curves, the size of goldsinny wrasse with 99% retention probability (L99; maximum 1% escape risk) was calculated and used as a measure for the minimum safe size that could be kept in the salmon cages. For a logit size selection model, L99 can be directly calculated from Eq. (3) as in Krag et al. (2014):



Fig. 4. Hexagonal mesh shape approximation for fish escape through a semi-slack square mesh. (a) Details of hexagonal mesh. (b) Illustration of fish escape through a semi-slack square mesh. (c) Approximation of the distorted semi-slack square mesh with a hexagonal shape. (d) Examples of hexagonal shapes approximating distorted semi-slack square meshes with different levels of openness (see Eq. (1)).

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$$L99 = L50 + \frac{SR}{\ln(9)} \times \ln(99)$$
(3)

However, besides length, the farming industry is interested in the weight of goldsinny wrasse that can safely be used in the cages. Therefore, the weight length relationship for this species is given in Eq. (4). Parameters a and b were established based on least square estimation on the experimental data (length L (mm) versus weight W (g)) collected for goldsinny wrasse individuals acquired for the FISHSELECT analysis:

$$W = a \times L^b \tag{4}$$

Based on the above, the weight of goldsinny wrasse with maximum 1% escape risk (W99) for each individual mesh was obtained by:

$$W99 = a \times L99^{\circ} \tag{5}$$

#### 3. Results

#### 3.1. Data collection

The goldsinny wrasse morphology data collection and subsequent fall-though trials were conducted at a small research station located on the coast of the island of Hitra (Mid-Norway) in September 2017. The fish used were collected by a local fisherman and maintained alive in a small net fixed to the quay, which gave us continuous access to live fish and allowed us to select individuals necessary to cover the widest possible length span of goldsinny wrasse during the trials. The FISH-SELECT procedure was applied to 100 goldsinny wrasse with sizes varying from 78 to 160 mm and 6 to 56 g (Fig. 5).

#### 3.2. Cross-section model choice and compressibility of goldsinny wrasse

The contours registered for the CS1 and CS2 of each fish were modelled for all five models using computer simulation (Section 2.2.1). The model with the lowest AIC value was chosen as the best model in each case (Akaike, 1974). The ship model, which is a three-parameter model, best represented both the CS1 and CS2 of goldsinny wrasse. In both cases the  $R^2$  was >0.96, meaning that the model was able to describe both CSs well (Table 1).

Each of the 100 goldsinny wrasse included in the trials was tested for its ability to fall through 478 meshes of different sizes during the experimental period, thus 47,800 fall-through trials were conducted. The highest DA between the experimental and simulated fall-through results when considering only the compressibility at CS1 was 98.06%, whereas the highest DA when considering only the compressibility at



Fig. 5. Weight vs. length relationship for the 100 lumpfish included in the study (W =  $a \times L^b$ ). a = 0.00000826 and b = 3.10.  $R^2 = 0.9635$ . The stippled lines show 95% confidence intervals.

CS2 was 97.76%. However, when both CS1 and CS2 where considered, the highest DA achieved was 98.10%. Therefore, this combined compression model was chosen for further analysis and mesh penetration predictions for goldsinny wrasse in FISHSELECT. The model had a dorsal compression of 20%, lateral compression of 15%, and ventral compression of 0% for CS1 and a dorsal compression of 0%, lateral compression of 35% for CS2 (Fig. 6).

#### 3.3. Predictions for mesh penetration and escape risk

Based on the virtual population of 2000 fish, we made predictions for the escape risk of goldsinny wrasse through square meshes of 30 and 50 mm. Despite the large variety of mesh sizes used in Norwegian salmon farms, these are the two most commonly used mesh sizes in cage nets for smolt and adult salmon, respectively (Moe et al., 2007). The results showed that for perfectly square and stiff cage netting, goldsinny wrasse of up to 103 (14) and 162 mm (58 g) would be able to escape (>1% risk) through meshes of 30 and 50 mm, respectively (Fig. 7). If the meshes in the cage were completely slack and fully deformable, the escape risk for goldsinny wrasse would be higher, and fish of up to  $\sim$ 126 ( $\sim$ 27) and  $\sim$ 206 mm ( $\sim$ 123 g) would be able to escape (> 1% risk) through meshes of 30 and 50 mm, respectively (Fig. 7). If the meshes in the cage were semi-slack, meaning that only the horizontal bars in the meshes were deformable, the fish sizes with escape risk <1% would vary depending on the mesh size and mesh openness (deformation level of the horizontal bars). For square meshes of 30 mm, the fish size with <1% escape risk would increase first monotonously to  $\sim 106 \text{ mm}$  ( $\sim 16 \text{ g}$ ) with a mesh openness of  $\sim$ 60% and further to  $\sim$ 120 mm (23 g) for a mesh openness of ~80%, followed by a decrease to ~103 mm (~14 g) when the meshes reached 100% openness (perfectly square meshes) (Fig. 7a). For square meshes of 50 mm, the size with <1% escape risk would increase monotonously to  $\sim$ 187 mm (91 g) with a mesh openness of  $\sim$ 70% and then decrease to  $\sim 162 \text{ mm}$  ( $\sim 58 \text{ g}$ ) when the meshes were 100% open (Fig. 7b).

The plot in Fig. 8 illustrates the minimum size of goldsinny wrasse that can be used for meshes of different sizes and four different states (stiff, semi-slack with 75% mesh openness, semi-slack with 90% mesh openness, and slack meshes). Square meshes in the stiff state allow safe use of the smallest legal sizes of goldsinny wrasse, whereas the meshes need to be substantially smaller in size to maintain the same safety level if the meshes in the cage netting are slack (Fig. 8). For example, to safely retain the goldsinny wrasse of ca. 200 mm (112 g), the meshes in the cage netting would have to be  ${\sim}50$  mm or smaller if there is risk that the meshes can be completely slack at times. However, if there is assurance that the meshes are stiff at all times, this mesh size could be increased to ~65 mm with certainty of that size would escape. Regarding semi-slack meshes, escape risk with mesh openness >75% would be higher than that for stiff meshes but below that of slack meshes. The risk with semislack meshes is closest to the risk with the slack meshes when the semislack meshes have an openness of ca. 90% (Fig. 8).

The isolines in the design guide (DG) (Fig. 9) show the smallest sizes of goldsinny wrasse that can be used safely (escape risk <1%) for a given mesh size and mesh openness. It is obvious from the DG that independent of the mesh openness, the larger the mesh size the larger the wrasse that need to be used to avoid escape risk. As illustrated in Fig. 8, the DG also shows that the escape risk for semi-slack netting with a high degree of mesh openness is larger than that for perfectly square meshes (100% openness). For all mesh sizes considered, escape risk increased with mesh openness was reached, which was the same result obtained for the square stiff mesh. The results show, for example, that if the netting in the cages is changed from 30 mm square meshes to 50 mm square meshes (see dashed vertical lines in Fig. 9), the minimum size of goldsinny wrasse stocked in the cage should be increased by  $\sim$ 50–60 mm to maintain an escape risk <1% independent of mesh openness.

#### Table 1

Comparison of the performance of the five different models fitted to observed CS1 and CS2 of goldsinny wrasse (all models are described in <u>Sistiaga et al.</u>, 2020). AIC value, number of parameters, and R<sup>2</sup> value are given for each of the tested models. AIC for the best model is shown in bold.

CS <u>models</u>			$\bigcirc$		$\bigcirc$		$\bigcirc$		$\bigcirc$	
	Ellipse		Flex Ellipse 1		Super Drop		Flex Drop		Ship	
	CS1	CS2	CS1	CS2	CS1	CS2	CS1	CS2	CS1	CS2
AIC	180.48	186.22	192.26	194.01	206.44	207.37	239.69	236.55	174.83	170.55
Nr parameters	2	2	3	3	3	3	3	3	3	3
R <sup>2</sup>	0.9584	0.9617	0.9523	0.9580	0.9475	0.9533	0.9297	0.9382	0.9603	0.9662



**Fig. 6.** Modelled CS1 and CS2 for a goldsinny wrasse randomly selected from the test population of 100 fish. The estimation was based on the combined compression model that provided the highest DA. The red contour represents the uncompressed CS, and the green line represents the CS with maximum compression.

#### 4. Discussion

For delousing purposes, salmon farmers use different species of cleaner fish simultaneously in their cages. Goldsinny wrasse is one of the most widely used species, and it is the smallest of them (Skiftesvik et al., 2015). Therefore, estimation of the escape risk of this species is especially relevant when considering the mesh sizes to be used in salmon farming aquaculture cages.

We determined the minimum size of goldsinny wrasse that would have an escape risk <1% for an array of square meshes between 10 and 100 mm in size. These mesh sizes cover the majority of those used by the Norwegian aquaculture industry, which is the largest salmon farming industry in the world (FAO, 2016; Ellis and Tiller, 2019). As the mesh sizes and shapes allowed in salmon farming in Norway are not regulated by law and can vary greatly, generalizing or making recommendations about the minimum sizes of salmon and/or cleaner fish to be used in the cage netting to avoid escape is challenging. In the guidelines used by the industry (Sigstadstø, 2017), the recommendations for goldsinny wrasse are to use fish no smaller than 110 (13), 120 (23), 130 (30), and 135 mm (33 g) for meshes of 31, 33, 36, and 39 mm, respectively. A comparison of the recommendations in the guidelines with the results obtained in our study shows that for a mesh size of 30 mm, the recommendation in the guidelines (which is given for 31 mm mesh) of using fish >110 mm (18 g) agrees well with the FISHSELECT predictions if the meshes in the cage netting retain a square shape. However, if the meshes in the cage netting are distorted, resulting in a fully slack state, sizes of goldsinny wrasse of up to 128 mm (28 g) would be able to pass through the meshes, which represents an underestimation of 16%. The same applies for the recommendation for 40 mm netting (which is given for 39 mm mesh); if the meshes are distorted and become slack, goldsinny wrasse of up to 167 mm (64 g) would be able to pass through the meshes, which represents an underestimation of 24% compared to the recommendations in the guidelines. The differences between the FISHSELECT predictions and the recommendations in the guidelines illustrate the importance of considering circumstances that can lead to mesh distortion and not assuming that the meshes in a salmon aquaculture cage netting are always square. Additionally, FISHSELECT provides predictions for square meshes larger than 40 mm and fish longer than 135 mm (33 g), which the current guidelines do not provide. For the two most typical meshsize used by the aquaculture industry in Norway i.e. 30 and 50 mm and under the worst-case scenario i.e. slack state, the minimum recommended lengths for goldsinny wrasse would be  $\sim$ 128 ( $\sim$ 28) and  $\sim$ 206 mm (~123 g), respectively.

The escape of fish from salmon aquaculture cages is a well-known problem for the Norwegian salmon aquaculture industry (Aronsen et al., 2020). In 2019, almost 300,000 salmon reportedly escaped from Norwegian farms due to accidents (Norwegian Directorate of Fisheries, 2021). Cleaner fish can also escape from the farms when accidents occur, but they have also been intentionally released to the marine environment after they have been used for one season (Espeland et al., 2010). In addition, as reported in earlier studies (Treasurer and Feledi, 2014) and demonstrated in the present study, if the size of the meshes required to avoid escape is not properly estimated or the potential changes in mesh shape is not properly accounted for, wrasse can escape from the aquaculture cages and into the wild via mesh penetration.

Wrasses naturally occur as part of the fish fauna along the Norwegian coast. However, many of the fish used at salmon aquaculture sites are transported over long distances (often several hundred kilometers of coastline) before they are released in the salmon cages for delousing purposes. In the case of goldsinny wrasse, translocations of fish from the Skagerrak coast of Norway and Sweden to locations in the central



Fig. 7. Sizes of goldsinny wrasse with escape risk <1% (Y axis) as a function of mesh openness (X axis) for semi-slack meshes (solid line). The dashed lines represent the results for slack meshes of 30 mm ( $\sim$ 128 mm or  $\sim$  28 g) and 50 mm ( $\sim$ 206 mm or  $\sim$  123 g), respectively, and the stippled lines represent the results for stiff meshes of 30 mm (103 mm or 14 g) and 50 mm (162 mm or 58 g), respectively. Plot (a) shows the results for 30 mm square meshes and plot (b) shows the results for 50 mm square meshes.

regions of Norway are common (Gonzalez and de Boer, 2017). Translocation of fish and potential interbreeding (Faust et al., 2018) with autochthonous populations can lead to genetic introgression in these populations, especially for species such as goldsinny wrasse, for which genetic isolation by distance has been reported due to limited adult migration and spawning site fidelity (Jansson et al., 2017). The consequences of genetic introgression on the autochthonous populations of fish can be hard to predict and can range from extinctions of the local populations to increased genetic diversity (Hindar et al., 1991). Therefore, interbreeding between autochthonous fish and the translocated fish used in the aquaculture cages should be avoided to the highest possible extent. Escape of wrasses also poses the risk of spreading of diseases (Taranger et al., 2015; Karlsson et al., 2016) and economic losses for salmon farmers, who constantly need to add new wrasses to replaces escapees during the delousing period. These issues highlight the importance of choosing the correct mesh size and shape for use in the salmon cages to avoid escape through mesh penetration.

In addition to environmental consequences, unintentional release of wrasses from aquaculture cages has important implications for their welfare. Incorrect estimation of the mesh sizes required to retain wrasses results in high numbers of escapees, which fuels the demand to capture and culture additional new fish. High demand keeps the fishing effort for the species high, and the same fish likely are captured and released multiple times (Skiftesvik et al., 2014). Furthermore, in high demand scenarios the fishermen operating near a specific aquaculture site may not be able to supply enough cleaner fish to meet the local demand, leading to translocation of cleaner fish from other locales. Concerns around the welfare of cleaner fish that are transported over large distances have been raised in the literature (Espeland et al., 2010; Treasurer and Feledi, 2014), and avoiding the escape of cleaner fish from aquaculture cages is an important measure to reduce the need for translocations.

Accidents that lead to the release of fish from aquaculture cages will always occur, but correctly estimating the size and shape of the meshes required to avoid escape via mesh penetration and using the appropriate netting in the cages is a preventive measure that can minimize escape. As Sistiaga et al. (2020) and Herrmann et al. (2021) previously illustrated for salmon smolt and lumpfish, and as demonstrated in the present study, prediction of the escape risk of different species in the aquaculture industry based on different mesh sizes and states is relevant, especially for small species such as the goldsinny wrasse. Further, the approach used here is likely relevant for other cleaner fish species (e.g. farmed ballan wrasse) and it should be considered to optimize the exploitation of wild and farmed cleaner fish.



Fig. 8. Maximum square mesh size limits that guarantee <1% escape risk of goldsinny wrasse of different sizes. The lines in the plot show the limits for stiff meshes (black) and slack meshes (lightest grey), and semi-slack meshes with 75% and 90% mesh openness are shown in different tones of grey.



#### Minimum length to avoid escape risk versus mesh size and openness

Fig. 9. Isolines of minimum size of goldsinny wrasse required to prevent escape (< 1% chance) from square mesh net cages of given mesh size and mesh openness. The dashed lines show the estimates for the 30 and 50 mm meshes, the most commonly used mesh sizes for smolt and larger salmon, respectively.

#### **Declaration of Competing Interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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