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GENETIC RISKS ASSOCIATED WITH SABLEFISH AQUACULTURE IN BRITISH COLUMBIA, CANADA

CONTEXT

Sablefish (*Anoplopoma fimbria*) aquaculture is currently limited to three marine facilities in British Columbia (BC) with six-year licences that were issued on July 1st, 2024. As for any cultured species, removal of wild Sablefish individuals from a population for broodstock and inadvertent release of aquaculture individuals back into a population have the potential to pose genetic risks to the long-term sustainability of the species. For instance, captive and wild fish may be genetically or epigenetically differentiated, such that gene flow from captive-bred or captive-reared populations into natural populations may adversely affect fitness in the wild. The magnitudes of these potential genetic risks are often context- and species-specific, and genetic risks have yet to be formally evaluated for Sablefish aquaculture in BC. Understanding the genetic risks associated with marine Sablefish aquaculture is an important precursor to the continuation of this practice in a scientifically defensible manner within the province.

Current aquaculture activities for Sablefish in BC involve collecting wild-sourced individuals for broodstock and producing either first- or second-generation captive-reared offspring with no intentional release of captive individuals back into the wild. In captive environments, natural selection that is typically experienced in the wild is relaxed, and artificial or domestication selection may cause traits to shift in a manner that would be sub-optimal in the natural environment. As a result, these differences can be harmful to wild populations if inadvertent escapees introduce disadvantageous genetic variation into natural habitats. The size of the genetic risks associated with the collection of broodstock and domestication are dependent on both the specific aquaculture practices and the biology of the cultured species. Numerous studies have demonstrated that gene flow from captive to wild populations is often to the detriment of fitness in the wild. However, the majority of this work in finfish has focused on salmonids, particularly Atlantic Salmon (*Salmo salar*), which have populations that are highly genetically structured. Unlike salmonids, Sablefish populations are widespread and relatively unstructured, suggesting that the genetic risks associated with aquaculture of this species may be comparatively low. In 2022, the stock status for Sablefish in BC, was found to be in the healthy zone (DFO 2009; DFO 2023).

Fisheries and Oceans Canada (DFO) Aquaculture Management Division has requested that Science Branch provide advice on the genetic risks associated with marine aquaculture of Sablefish under both current practices and a possible future scenario in which broodstock are no longer wild-sourced. The assessment and advice arising from this Canadian Science Advisory Secretariat (CSAS) Science Response (SR) process will be used to support future decision-making related to Sablefish aquaculture.

The specific objectives of this review are to:

1. Review the current understanding of wild Sablefish population genetics in British Columbia.
2. Document the potential genetic risks posed to wild Sablefish due to captive-rearing of individuals for aquaculture harvest.

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3. Evaluate these potential genetic risks to wild populations in the context of current Sablefish aquaculture practices in British Columbia.
4. Assess how genetic risks may change if current practices for Sablefish aquaculture transition from the use of wild-sourced broodstock to captive broodstock.

The intent of this report is not to provide a risk assessment of Sablefish aquaculture. Instead, this report serves to summarize the current understanding of Sablefish genetics and population structure in British Columbia, to identify the direct genetic risks of aquaculture activities to wild populations of the species, and to compare those genetic risks to those associated with the culture of other finfish species (e.g., Atlantic salmon). Finally, this report highlights data deficiencies that limit the certainty to which genetic risks of aquaculture can be assessed for this species.

This Science Response Report results from the regional peer review of October 30, 2024, on the Genetic Risks to Wild Sablefish from the Escape of Farmed Sablefish.

BACKGROUND

Sablefish (*Anoplopoma fimbria*) are deep-water marine fish with a northern amphi-Pacific distribution (Figure 1) and high fisheries value, estimated at approximately $27.9 million per year between 2012 and 2022 (DFO 2024a). Outside of Canada’s exclusive economic zone Sablefish tend to aggregate at seamounts, although individual seamounts are not believed to represent distinct populations and this species is distributed more homogenously throughout areas where commercial harvest is undertaken (DFO 2013). As such, Sablefish are managed in Canada as a single unit major fish stock under Canada’s renewed *Fisheries Act* (DFO 2013; Cox et al. 2023). Consequently, the stock is subjected to assessments and fisheries evaluations in adherence with the precautionary approach (DFO 2009). Such management approaches have begun to reverse nearly 50 years of declining stock biomass and recent analyses indicate above-average stock recruitment over the past decade. For example, female spawning stock biomass was clearly above the level of biomass associated with maximum sustainable yield in 2022 (DFO 2023). However, consumer demand for this species continues to rise, suggesting that other means of production, such as aquaculture, may help to subsidize wild fisheries (Hartley et al. 2020).

Since 2005, the harvest of wild populations has been complemented by commercial aquaculture. Currently, there are three marine aquaculture facilities for Sablefish in Canada, two of which are active and situated in Kyuquot Sound, British Columbia and a single inactive facility in Barkley Sound (Figure 1). The two active facilities, Charlie’s Place and Centre Cove, are currently licensed for 2,700 and 2,202 metric tons of Sablefish, respectively, whereas Jane Bay is licensed for 550 metric tons (Kerra Shaw, *personal communications*). These aquaculture facilities are owned and operated by Golden Eagle Sablefish in partnership with Kyuquot-Checleseht First Nations and produce Sablefish products for both local and international markets.

Sablefish aquaculture in Canada is currently based on the collection and rearing of a combination of wild-sourced and first-generation (F1) hatchery broodstock. Individuals are captured in the wild, transported to the hatchery, and held under low-light and low-temperature conditions to emulate their typical deep-water rearing environment. Fish are monitored during maturation and a combination of hormonal treatments as well as environmental cues (i.e., photoperiod and water temperature) are used to induce fish to spawn. After capture, adult Sablefish are held for several years, and may be used in spawning across more than one season (Derek Price, *personal communications*). Larval Sablefish are incubated in the hatchery and initially fed live diets (i.e., rotifers and artemia) before being transitioned to formulated diets.

Survival during larval rearing is reported to be between 10 and 40% (Cook et al. 2015). After approximately four months in the hatchery, juvenile Sablefish are transferred to marine net-pen installations to complete grow-out. The marine net-pens used for Sablefish grow-out are organized in a grid and are surrounded by a secondary net designed to prevent predation and act as an additional layer of containment. Grow-out can also occur in land-based recirculating systems, but this is less relevant to the subject of this report as cultured fish in these systems are unlikely to escape into the environment. After approximately two years of growth, fish are harvested and processed for distribution at weights averaging between 1.8 and 3.6 kg.

Recent developments in the broodstock management of Sablefish aquaculture in Canada include the use of sex-reversed females, referred to by the industry as “neomales”. Successful application of a hormonal treatment during development creates genetically female fish (i.e., fish carrying two X chromosomes) that express male gametes (i.e., milt containing sperm). Crossing these fish with egg-producing female fish produces an all-female production population for grow-out that is free of the hormonal treatment. The all-female production population results in faster growth and larger fish due to the known sex-specific differences in growth rates for this species (Morita et al. 2012).

The genetic risks posed to wild populations due to these aquaculture activities are dependent on the specific culture practices, and the biology of Sablefish. Sablefish biology has been thoroughly reviewed elsewhere (e.g., Beamish & Mcfarlane, 1988; Head et al. 2014; Morita et al. 2012; Wilkins & Saunders, 1997), and it is beyond the scope of the current report to repeat an in-depth review here. Instead, we focus on key aspects of Sablefish biology that influence how genetic variation is distributed within this species, which are relevant for contextualizing potential direct genetic interactions between escapees from aquaculture and the wild population.

Sablefish are a highly mobile species that can disperse great distances (>1,000 km). In mark-recapture studies, fewer than half of tagged fish are recovered within 50 km of their release site, with a proportion of those tagged in British Columbia dispersing either north or south as far as Alaska or Mexico, respectively (Figure 2) (Morita et al. 2012; DFO 2013). Both sexes display this high capacity for dispersal, but females have been shown to grow larger and faster than males, and there is evidence to suggest they also disperse across longer distances than their male counterparts (Morita et al. 2012).

Otolith analyses indicate Sablefish can live for more than 100 years and typically reproduce annually (although some skip years) (Beamish and McFarlane 2000; Rodgveller et al. 2015; Guzmán et al. 2017). In the wild, Sablefish mature at around five years of age and a size of 50-60 cm. Fecundity of females ranges from approximately 60,000 at 58 cm to over 1,000,000 for fish over 1 m of length, with a majority of mature females exhibiting annual fecundity between 100,000 – 500,000 (Mason et al. 1983). Sablefish spawn throughout the winter at depths greater than 300 m along the Pacific coast (Mason et al. 1983), and mature individuals are found at depths greater than 183 m (Head et al. 2014). Many demersal fishes, like Sablefish, exhibit sweepstakes reproduction, wherein a small proportion of the total population successfully reproduces (Christie et al. 2010). While these data are lacking in the wild, cultured Sablefish have been observed to undergo sweepstakes reproduction in captivity, with high reproductive variance among individuals and families (Rubi et al. 2022).

Several studies have considered genetic variation across the range of Sablefish. Throughout the eastern Pacific Ocean, Sablefish appear to exhibit a general lack of genetic population structure (Gharrett et al. 1982; Tripp-Valdez et al. 2012; DFO 2013; Jasonowicz et al. 2017; Orozco-Ruiz et al. 2023; Timm et al. 2024). In other words, the majority of genetic variation occurs among individuals within sampling locations rather than between individuals sampled at different locations, which is consistent with the high levels of movement detected by tagging studies (Figure 2) (Morita et al. 2012; DFO 2013). Recent whole genome resequencing efforts have detected a pair of large inversions on chromosome 22, but even these inversions do not display differential spatial distributions (Timm et al. 2024), and any link between these inversions and distinct lineages remains to be identified. However, there is some evidence for subtle differentiation at the extremes of the species' range, as nuclear and mitochondrial data have identified divergent lineages between the far east (Kamchatka) and a unique population in Baja, respectively (Orozco-Ruiz et al. 2023). While there are only a few samples from British Columbia that have contributed to these population genetics studies, fish from Alaska to Washington consistently form one genetic group (Gharrett et al. 1982; Tripp-Valdez et al. 2012; DFO 2013; Jasonowicz et al. 2017; Orozco-Ruiz et al. 2023; Timm et al. 2024). Thus, it would be surprising to observe differentiation among locations in BC. Furthermore, similar patterns of low levels of genetic differentiation across large spatial scales are commonly observed in many other demersal fishes with long migratory patterns and life-long reproductive capacity (e.g., Halibut, *Hippoglossus sp.*; Drinan et al. 2016; Kess et al. 2021; Ferchaud et al. 2022).

Despite the lack of genetic differentiation among locations in Sablefish, numerous factors can result in genetic differences between cultured and natural populations of an aquaculture species. Breeding and rearing animals in an aquaculture environment is known to cause both intentional and unintentional changes to traits of fish reared in culture (Hallerman 2008; Lorenzen et al. 2012; Koch et al. 2023). For species with closed or partially closed broodstocks, intentional selection for traits that improve performance (e.g., growth rate, low precocial maturation, flesh quality, feed conversion efficiency, etc.) can produce substantial deviations in the phenotypic trait optima between aquaculture and wild populations (Teletchea and Fontaine 2014). Likewise, unintentional selection in the aquaculture environment (e.g., due to high density, disease outbreaks, formulated diets) and relaxed selection compared to natural environments (e.g., lack of predators) can also result in genetic changes in aquaculture populations (Glover et al. 2012; Kristjánsson and Arnason 2016). The rate of differentiation between aquaculture and wild populations can be influenced by a number of factors, including aspects of the culturing facility (e.g., does the artificial environment attempt to mimic the natural environment?) and the nature of the husbandry protocols (e.g., are animals bred randomly, selective, or not at all?). Indeed, a variety of artificial environments and protocols have been shown to illicit rapid differentiation between captive and wild populations in many fishes, with differences ranging from variation in epigenetic programming to broodstock populations with highly differentiated regions of the genome (e.g., Le Luyer et al. 2017; Liu et al. 2017; Habibi et al. 2024).

Culture of animals may also result in random effects on genetic variation such that differentiation between aquaculture and wild populations is not necessarily the result of differences in selection. For species exhibiting population structure due to restricted gene flow among natural populations, the movement and culture of non-local populations will cause genetic differentiation between the cultured population and local natural populations (Skaala et al. 2004; Sylvester et al. 2018). Population structure in a species suggests the potential for local adaptation, which may indicate adaptive differences between local natural and non-local cultured populations. Additionally, the maintenance of genetic variation within a population is dependent on the effective population size (N_e), which reflects an idealized population that allows calculation of the rate at which genetic variation is lost from the population (Wright 1990). An effective population size is generally lower than the census population size (Frankham 1995), and there is only an approximate relationship between the two measures (Clarke et al. 2024; Waples 2024). Regardless, as the effective population size decreases, stochastic changes in genetic variation due to a small number of breeding individuals increases. Consequently, at low effective population sizes, the likelihood that genetic variation is lost from a population is increased. Aquaculture broods tend to involve a relatively low number of spawners, and coupled with the possibility for sweepstakes reproduction, there is the potential for increased random loss of genetic variation compared to a wild population (Ryman and Laikre 1991; Hallerman 2008; Waples et al. 2016). These random losses affect both neutral and adaptive genetic variation, and therefore can also result in the loss of genetic variation that is adaptive in natural habitats. Furthermore, negative fitness effects associated with inbreeding increase at low effective populations sizes as well (Naish et al. 2013; Pekkala et al. 2014; Lohr and Haag 2015). Thus, a combination of random and nonrandom genetic effects can rapidly produce differentiation between aquaculture and wild fish, leading to reduced fitness of artificially-reared fish in natural settings (Milot et al. 2013; Johnsson et al. 2014; Solberg et al. 2020).

In addition to inheritance of genetic sequence variation associated with aquaculture, other structural aspects of DNA may also be affected by aquaculture and passed on to the next generation (reviewed by Angers et al. 2020, and Bossdorf et al. 2008). These structural changes are known as epigenetic modifications, and perhaps the best characterized example is the addition of methyl groups to cytosine base pairs within DNA (i.e., DNA methylation) (Laine et al. 2023). Even in the absence of sequence variation, epigenetic variants can affect traits related to fitness, particularly through changes in gene expression. These epigenetic modifications offer a rapid, potentially heritable, mechanism for altering gene expression, and there is a growing body of evidence to suggest that rearing finfish in an artificial environment induces epigenetic changes relative to wild epigenetic patterns in the same species (Christie et al. 2012, 2016; Le Luyer et al. 2017; Wellband et al. 2021; Koch et al. 2023; Venney et al. 2023). In the context of aquaculture, epigenetics is a relatively new area of study, and the extent to which aquaculture-associated epigenetic variants are inherited remains unresolved. In general, epigenetic variation is reprogrammed during development (Feng et al. 2010; Ksenia et al. 2018; Wellband et al. 2021); however, there is at least some evidence for transmission of variants from parents to offspring in cultured species (Anastasiadi et al. 2021; Wellband et al. 2021; Venney et al. 2023). Therefore, similar to genetic variants, there is potential that epigenetic variants associated with an aquaculture environment may be transmitted to wild fish (Koch et al. 2023; Venney et al. 2023) posing a risk to the fitness of wild populations.

The extent of risk posed by genetic and epigenetic differences between aquaculture and wild finfish is highly influenced by the rate at which aquaculture individuals interbreed with their wild counterparts, and the likelihood that variation associated with aquaculture becomes established in wild populations. Opportunities for interbreeding and introgression are created via the escape of fish from aquaculture facilities to the natural environment. In general, escapes are often categorized as small-scale inadvertent releases (i.e., “trickle” escapes), or as large-scale escape events typically associated with equipment failure or handling errors (Leggatt et al. 2010; Yang et al. 2019). In either case, direct genetic interactions between the escaped fish and the wild population can shift traits away from their optimal values in the natural habitat (Yang et al. 2019). The salmonid literature provides numerous examples where introgression from aquaculture to wild populations is to the detriment of the wild stock (Bourret et al. 2011; Christie et al. 2014; Stringwell et al. 2014; Davison and Satterthwaite 2017; Bradbury et al. 2020; McMillan et al. 2023), and putative loss of adaptive variation is particularly common given high levels of local adaptation and strong population structure in salmonids (Fraser et al. 2011). For example, introgression among aquaculture and wild Atlantic Salmon (*Salmo salar*) in the Magaguadavic River coincided with considerable population declines and the loss of genetic variation underlying an adaptive trait in the wild population (Donnelly and Dill 1984; Bourret et al. 2011). Loss of genetic variants as a result of introgression from aquaculture is less well characterized in large populations, but evidence from different populations of Atlantic Salmon indicate that larger populations are more resistant to introgression from aquaculture fish (Glover et al. 2012) and some examples of highly abundant fish (e.g., Atlantic Cod) have failed to detect signatures of introgression following escape events (Varne et al. 2015). Taken together, this suggests that escape rates of aquaculture fish, and the sizes and genetic structure of the receiving wild populations will play a large role in determining both the likelihood and consequence of introgression of genetic or epigenetic variation associated with aquaculture (Hallerman 2008).

Species-specific population characteristics, reproductive behaviours, and culturing practices are key components in determining the potential consequences of direct genetic interactions between escaped aquaculture fish and wild populations. As such, this report outlines these factors for Sablefish in the context of aquaculture in BC given current culturing practices and the available information on Sablefish life-history and genetics. Sources of potential uncertainty in this assessment are also explored and the possibility of development of a captive broodstock is discussed. At the time of publication, data limitations preclude a formal genetic risk assessment; knowledge gaps identified in this report provide guidance for the data collection efforts necessary to support a risk assessment in the future.

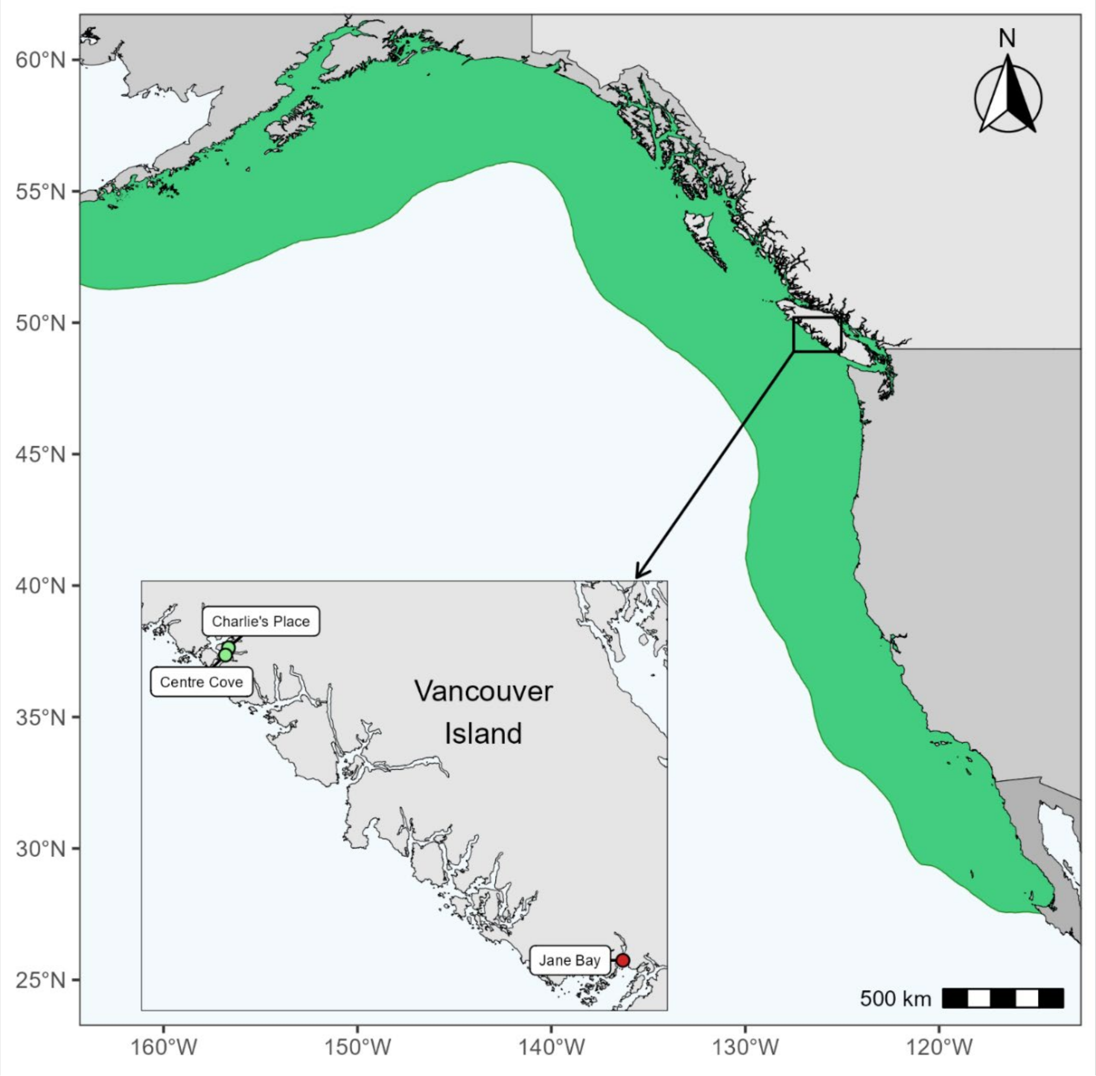

Figure 1. Locations of active (green) and inactive (red) sablefish aquaculture facilities in Canada. The green area in the Pacific Ocean indicates the approximate adult range of sablefish (adapted from Orlova et al. 2019 and Tokranov et al. 2005).

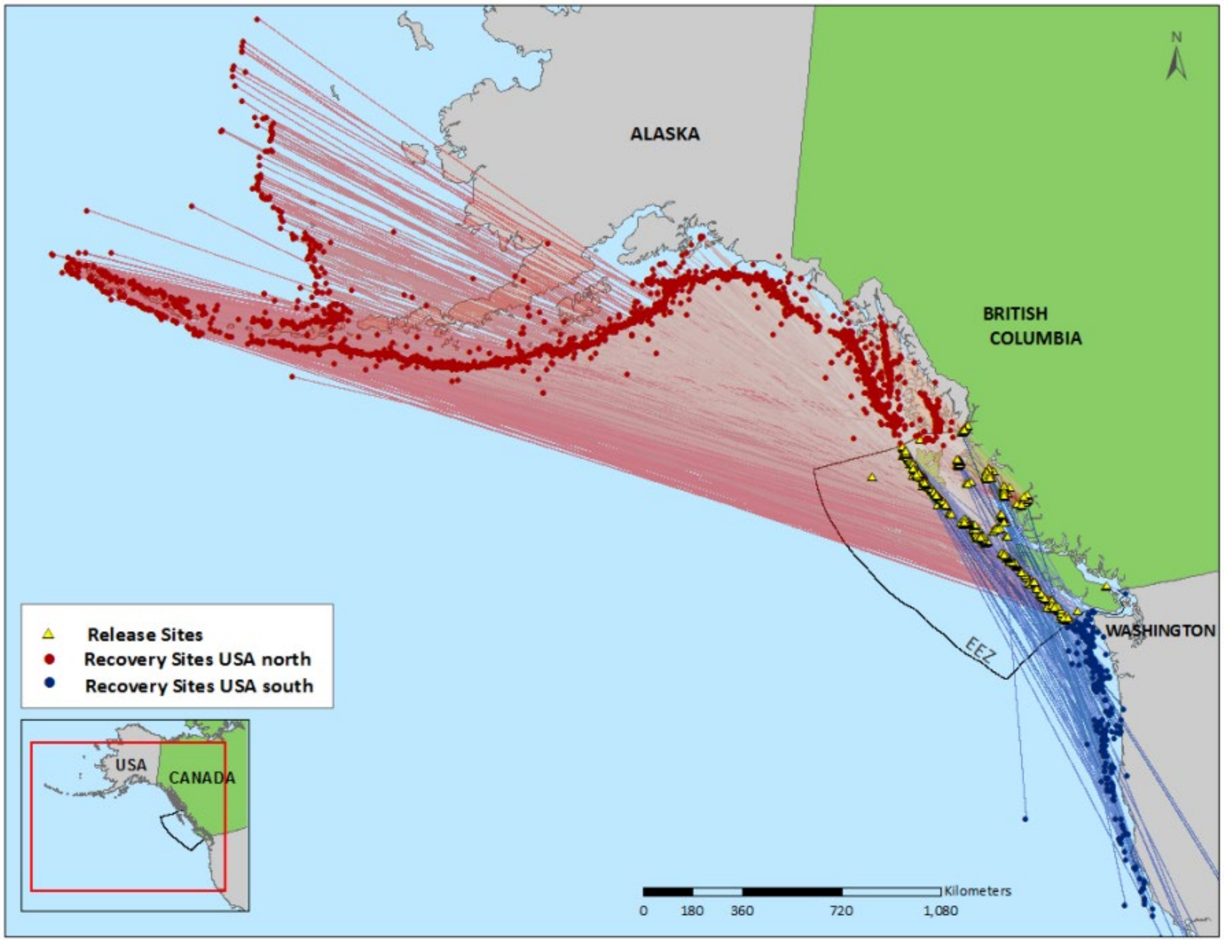

Figure 2. Release and recovery locations of tagged Sablefish in British Columbia between 1991 and 2012 (DFO 2013).

ANALYSIS AND RESPONSE

Direct genetic interactions between aquaculture and wild populations require the following steps:

1. fish or their gametes must escape from an aquaculture facility,
2. escapees must survive in the wild and disperse to natural spawning habitats, and
3. escapees must breed successfully with wild individuals (Figure 3).

These genetic interactions have the potential for negative consequences for the wild population by altering genetic diversity, fitness and productivity of the wild population (e.g., DFO 2024b). Here, these points are considered in the context of Sablefish aquaculture by considering all available escape information for Sablefish in BC, examining evidence for introgression from aquaculture facilities in other species of finfish, and highlighting empirical examples where introgression has been associated with negative impacts on natural populations.

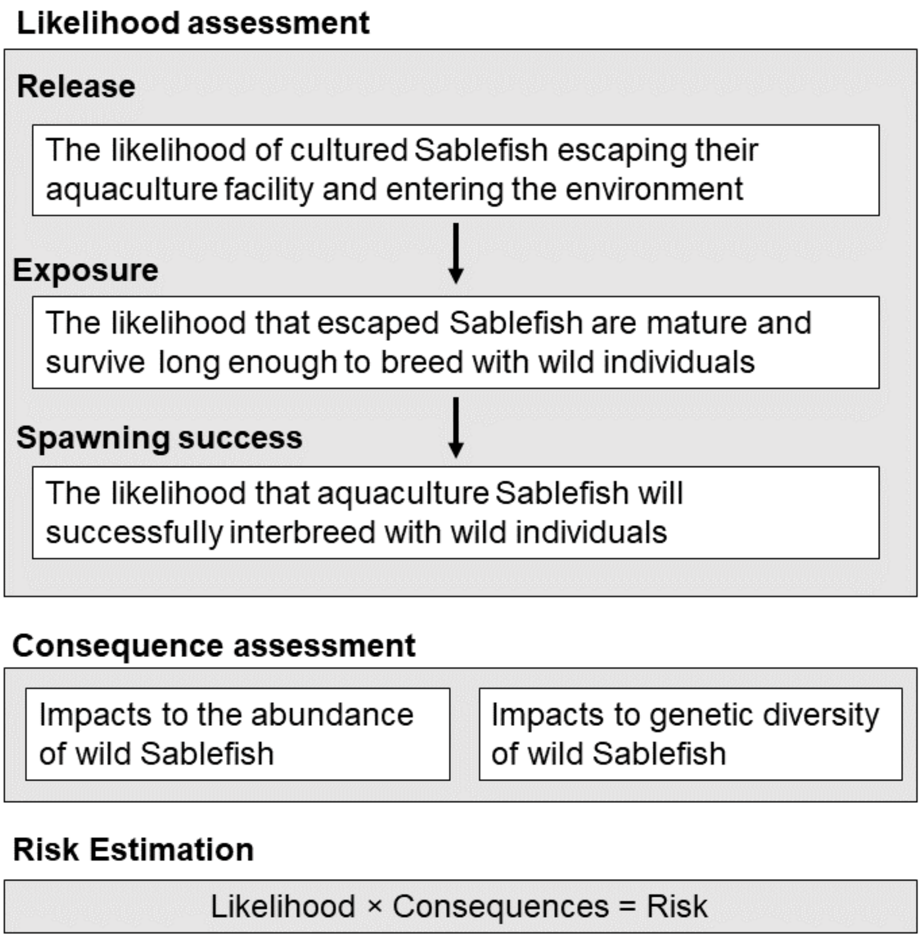

Figure 3. Conceptual model to assess the risks associated with direct genetic interactions between wild and escaped aquaculture Sablefish (modified from a previous genetic risk assessment for Atlantic Salmon; DFO 2024b).

Likelihood of Genetic Interactions

Gene flow from aquaculture populations to wild populations first requires that fish escape their enclosure. In British Columbia, Sablefish producers must make reasonable efforts to prevent escapes from occurring and attempt to control and contain fish that escape. Producers are also required to report all escapes when they are identified and data for all escapes are published online by Fisheries and Oceans Canada: Escapes of cultured finfish from BC aquaculture sites. To date there have not been any reported Sablefish escapes, which indicates that large-scale release events from Sablefish facilities have not occurred since the inception of aquaculture of this species in BC. Additionally, the lack of reported Sablefish escapees suggests that the rate of trickle escapes is low. Assessments of escape rates from aquaculture facilities for other fish species have consistently concluded that trickle escapes are consistently underestimated due to unintentional underreporting, because not all small escape events are observed (Carr and Whoriskey 2006; Skilbrei et al. 2015). To address this possibility, we considered open net-pen escape rates from other finfish species. There is considerable variation in estimated escape rates among species driven in part by species-specific differences in circumstances leading to escape. For instance, some groundfish bite through netting and have relatively high escape rates (e.g., Atlantic Cod, *Gadus morhua*), whereas salmonid escapes are more reliant on damage to the nets themselves (Føre and Thorvaldsen 2021). In comparison, Sablefish is a groundfish species, but their potential mechanism of escape is unknown. Although no escaped Sablefish have been reported in BC which precludes a direct estimate of a species-specific escape rate, reports from other species suggest that escape rates generally range from <1% to 6% of total stock (Leggatt et al. 2010).

In species that display broadcast spawning, aquaculture individuals may also escape from facilities through the release of fertilized embryos (e.g., Atlantic Cod; Jørstad et al. 2008). Although Sablefish are broadcast spawners (Sogard and Berkeley 2017), the possibility of this pathway of escape is likely mitigated by current culturing practices in this species. First, production fish are typically harvested at two years old, whereas Sablefish in nature reach sexual maturity at the age of five. Second, only female fish are stocked into net-pens for grow-out, and mature individuals in nature are found at depths greater than ~180 m (Head et al. 2014). Together, these facts suggest it is unlikely that either gametes or fertilized embryos would be released from Sablefish aquaculture facilities.

To our knowledge there is no species-specific data available to assess the ability of escaped Sablefish to survive, disperse or spawn successfully in the natural habitat. For genetic interactions with the wild population to occur, escaped Sablefish would need to migrate to spawning locations at depth during the winter spawning season. However, the demonstrated ability of first-generation hatchery Sablefish (i.e., those that have been subjected to culture) to successfully reproduce in the hatchery environment in response to cues mimicking the natural environment (Rubi et al. 2022) suggests that escaped Sablefish would be capable of spawning in the wild. There is an abundance of evidence in other finfish species demonstrating that escapees from aquaculture facilities are sufficiently fit to survive in the wild and to spawn with wild fish (Karlsson et al. 2016; Bradbury et al. 2022). A precautionary approach would assume escaped Sablefish are capable of surviving and dispersing in the natural habitat, and that they have the ability to spawn with wild individuals. Furthermore, the relative introgression associated with escaped individuals may be higher in Sablefish than expected based on data from semelparous species like salmonids; as Sablefish display annual or biannual spawning over lifespans that can reach 100 years, the genetic impacts of aquaculture escapees may compound over time.

In aggregate, the data available to consider the likelihood of exposure of wild Sablefish to genetic effects from aquaculture are extremely limited. None of the escape, survival or natural reproduction of aquaculture Sablefish have been observed. However, evidence from other cultured finfish would suggest all three occur in other species, and that escape rates are often underestimated. Given requirements in BC, the lack of reported escapes supports that the rate of escape of Sablefish from aquaculture is low, but dedicated data collection efforts would be necessary to accurately assess all three components contributing to the likelihood of direct genetic interactions between aquaculture and wild Sablefish (Figure 3).

Consequences of Genetic Interactions

Once individuals from an aquaculture population escape into the natural habitat and successfully interbreed with wild individuals, the consequences for the genetic state of the wild population will depend on the amount of differentiation between the captive and wild populations, and the extent to which the wild population is buffered against perturbations from influx of variation associated with aquaculture. Population differentiation will be impacted by both broodstock management and the number of generations in the captive environment, whereas wild population structure and genetic diversity will influence the resilience of the species to genetic perturbations.

Population differentiation

No genetic data are available to assess genetic differentiation between Sablefish aquaculture broodstock and their wild counterparts directly. However, using a combination of first principles, information about wild Sablefish populations, and evidence from other cultured species, it is possible to make inferences about the likelihood of genetic differentiation between cultured Sablefish and their wild counterparts.

A large proportion of Sablefish broodstock is sourced from the wild and this poses a different set of considerations for genetic differentiation of aquaculture broodstock from wild sources compared with salmonid aquaculture species. Specifically, founder effects during capture have the potential to generate genetic differentiation based on behavioural variation and gear types used for broodstock capture. The use of long-lines and baited traps, the most common gear types used in the commercial fishery, may inadvertently select for bolder fish during broodstock collection as seen in several species of bass (Wilson et al. 2015; Andersen et al. 2018). Consequently, fish captured for breeding purposes may not be a random sample of the population at large, but rather a non-random subset of the boldest individuals depending on the implemented method of capture.

After capture, differences between aquaculture and natural environments for Sablefish (e.g., absence of predators, prevalence of food, shallow depths, restricted spaces) comprise a selective landscape wholly different than that experienced by wild members of the species. Through both relaxed natural selection and domestication selection, these differences can also produce substantial changes in genetic variation and trait distributions between wild and aquaculture fish (e.g., Johnson and Abrahams 1991). Selection-mediated differentiation between aquaculture and wild individuals would be predicted to shift aquaculture populations away from the optimal traits in wild habitats, such that aquaculture-mediated genetic change could represent a risk to the fitness of wild populations (Leggatt et al. 2010). As discussed in the Background section, this mechanism of genetic differentiation and associated fitness effects has been frequently observed in other cultured finfish species (Houde et al. 2010; Christie et al. 2012, 2014; O'Sullivan et al. 2020; Dayan et al. 2024), and has the potential to occur even over a single generation in captivity (Christie et al. 2012, 2014, 2016; Habibi et al. 2024). The likelihood and extent of differentiation would be expected to increase with the number of generations over which the cultured species is held in captivity. The majority of Sablefish broodstock are currently wild-sourced (e.g., Rubi et al. 2022), but the use of sex-reversed neomales indicates that production fish have at least one parent that has spent its entire life in captivity. The bulk of evidence across species demonstrating that genetic differentiation between aquaculture broodstocks and wild populations is common and occurs rapidly suggests that differentiation between aquaculture and wild Sablefish is likely, although it has yet to be directly assessed. The extent of differentiation may be limited, especially in comparison to other species with longstanding captive broodstock (e.g., Atlantic Salmon), given the low number of captive generations in current Sablefish aquaculture and the lack of deliberate trait selection.

Population structure and genetic diversity

The likelihood of negative impacts associated with introgression from aquaculture populations is also dependent on the receiving wild population. Evidence from Atlantic Salmon shows that the effective population size and population structure of the wild population both influence the magnitude of these impacts (Heino et al. 2014; Sylvester et al. 2018). Small highly structured populations are particularly vulnerable even to relatively small influxes of genetic variation associated with aquaculture. In a small population, introgression is more likely to directly produce a large shift in the proportions of genetic variants present among the spawners in the natural habitat. In contrast, large populations with high effective population sizes and low genetic structure among locations are buffered against these effects. Not only are similarly sized influxes proportionally smaller in a larger population, but also selection operates more efficiently in large populations, such that suboptimal genetic variants are less likely to become widely established and adaptive variation is less likely to be lost via random processes such as genetic drift (Charlesworth 2009). As discussed above, it appears Sablefish in BC reflect a large population with limited population structure. This also suggests that any non-local origins or movements of fish used in aquaculture operations in BC are unlikely to introduce novel genetic variation into local habitats via escapees.

It is challenging to estimate the effective population size of a wild population (N_e) based on a census count of individuals (N_c) with accuracy (e.g., Palstra and Fraser 2012; Ferchaud et al. 2016), but with respect to potential thresholds for the maintenance of additive genetic variation (e.g., >500 or >1,000; Frankham 2005; Waples 2024) it is most probable that the large population of Sablefish in BC reflects an effective population size well above a genetically vulnerable level. For instance, empirical estimates of effective population size relative to census population size (N_e/N_c) in sea bass (*Atractoscion nobilis*) range from 0.27 to 0.40 (Bartley et al. 1992). In contrast, estimated values of N_e/N_c for marine fishes may be as low as 0.017 on average (Clarke et al. 2024). However, despite this low estimate, modeled populations have a high probability of N_e exceeding 500 individuals (Clarke et al. 2024). Thus, Sablefish may be well buffered against negative consequences of introgression from aquaculture due to relatively large numbers of wild spawners with limited differentiation among locations throughout the species range.

Negative consequences associated with aquaculture escapees on the effective population sizes of wild populations are primarily dependent on two factors: the fraction of escapees in the wild population, and the ratio of the effective size of the wild population to that of the aquaculture population (Waples et al. 2016). Results from modeling efforts indicate that negative consequences are mitigated when the hatchery fraction is small, and the effective population size of the wild population is substantially greater than that of the aquaculture population (e.g., 1,000-fold higher; Waples et al. 2016). Although data to assess this ratio for Sablefish are not available, it is likely that both of these conditions are the case under current production levels.

Although limited population structure has been detected in Sablefish, one limitation of the available data is that the majority of studies considered putatively neutral genetic variation at relatively few loci across the genome (DFO 2013; Jasonowicz et al. 2017; Orozco-Ruiz et al. 2023). The Sablefish genome is approximately 650,000,000 base pairs (Flores et al. 2023), meaning even thousands of single-nucleotide polymorphisms (SNPs) represent a lower density of markers than is often necessary to resolve adaptive patterns (Manel et al. 2016; Ahrens et al. 2018). As a result, there may be unresolved adaptive genetic variation among Sablefish in the natural habitat. For instance, a recent study assessed variation at >7,000,000 locations within the genome, and resolved a pair of previously undetected inversions on chromosome 22 (Timm et al. 2024). It is possible these inversions represent adaptive patterns or a component of population structure yet to be understood (e.g., life-history related traits such as migration timing, etc.). Indeed, similar studies using high marker densities in other marine taxa have detected subtle patterns of local adaptation despite high gene flow and low population structure (Gagnaire et al. 2012; Gleason and Burton 2016; Liu et al. 2022; Fuentes-Pardo et al. 2024). Thus, introgression of genetic variation associated with aquaculture may yet pose a threat to unresolved adaptive variation in Sablefish. However, it is likely that the conclusions above with respect to large population sizes and widespread genetic variation in the species will hold as an overall pattern even in the face of future data collection efforts.

The use of an all-female production population also introduces considerations for genetic effects associated with interbreeding between aquaculture escapees and wild Sablefish. The accumulation of selection in these fish which have an additional generation spent in the hatchery was addressed above, but escaped individuals will also transfer maternal effects associated with aquaculture at a proportionally higher rate into the wild population than would be expected from production with typical sex ratios (Mousseau et al. 2009). On the other hand, transfer of any possible paternal effects will be correspondingly reduced. Given the lack of information regarding maternal and paternal effects in Sablefish, it not possible to evaluate the net impact of biasing towards maternal effects with currently available information. Additionally, another possible consequence of all-female production is that escaped Sablefish could shift local sex ratios in spawning groups away from 50:50, which can also reduce effective population sizes and alter patterns of genetic variation (Frankham 1995). Despite this possibility, it is likely that wild Sablefish are buffered against this consequence given the large and widespread population of the species as discussed above.

The information considered in this report suggests that genetic differentiation to some extent is likely between the aquaculture and wild populations of Sablefish in BC, and that the variation present in the aquaculture population is most likely suboptimal in the natural habitat. Taken together, these suggest that interbreeding of aquaculture escapees with wild Sablefish would have genetic consequences for the wild population, but the wild population may be naturally buffered against these consequences by Sablefish population structure, particularly compared to species like salmonids. Again, the data available for Sablefish are limited, and addressing knowledge gaps on the extent of genetic differentiation and fitness impacts of aquaculture-mediated genetic changes will be necessary to assess the consequences on interbreeding in the future.

Development of a captive brood population

In many hatchery and aquaculture environments, an isolated breeding population is maintained for multiple generations to continually provide offspring for production purposes without reliance on natural populations. This provides a benefit for natural populations as there is not consistent removal of individuals for broodstock. Yet, this benefit comes at a potential cost, as the likelihoods of differentiation and domestication are higher as the number of generations in captivity increase (e.g., Fraser 2008). Thus, captive broodstocks may result in more widespread genomic differentiation between captive and wild populations (Howe et al. 2024). For example, in Atlantic Salmon some broodstocks have been sustained in captivity for decades (e.g., 6-10 generations), and introgression from these broodstocks into wild populations has been shown to have negative fitness consequences as a result of early maturation, delayed run timing and increased predator susceptibility (Stringwell et al. 2014; Solberg et al. 2020; Besnier et al. 2022; Bekkevold et al. 2024). Furthermore, brood populations that are maintained and bred in captivity over a long period may be sufficiently depleted of genetic variation that inbreeding depression is observed (Kincaid 1983; Gallardo et al. 2004; Venney et al. 2016), increasing the potential hazard associated with introgression. As a result, the development of a captive brood program for Sablefish aquaculture is a reasonable approach to reduce the reliance on wild fish for brood, but this approach would likely come at the cost of increased genetic or epigenetic differentiation between aquaculture and wild populations. This, in turn, would place an even greater importance on mitigating the likelihood of wild Sablefish interbreeding with fish from aquaculture populations (i.e., minimization of escapees, sterility of cultured populations).

KNOWLEDGE GAPS

There are several critical data gaps and sources of uncertainty surrounding the biology and aquaculture of Sablefish in BC (Table 1):

- The lack of observations of escapees from Sablefish aquaculture facilities precludes accurate estimation of the escape rate.
- It is currently unknown if escaped aquaculture Sablefish survive and interbreed with wild Sablefish.
- There are no data describing genetic or epigenetic differentiation between aquaculture and wild Sablefish as a result of current culturing practices.
- The genetic effective population size of Sablefish in the eastern Pacific has not been directly estimated.
- The fitness of aquaculture Sablefish or aquaculture-mediated genetic variation in the natural habitat has not been examined.

Table 1. Uncertainties associated with aquaculture Sablefish being exposed to wild populations and the potential consequences of aquaculture and wild individuals interbreeding.

	Assessment component	*Uncertainty*
Likelihood	*Sablefish escape from their aquaculture facility and enter the environment*	*"Trickle" escapes may not be observed in all cases* *No data on presence of aquaculture Sablefish in the natural habitat*
	Escaped Sablefish survive to breed with wild individuals	*Survival rate of aquaculture Sablefish in the wild is unknown* *Timing of maturation of aquaculture Sablefish outside of the aquaculture facilities is unknown* *Dispersal patterns of aquaculture Sablefish are unknown*
	Aquaculture Sablefish interbreed with wild individuals	*It is unknown if aquaculture and wild Sablefish interbreed in the natural habitat*
Consequence	*Impacts to the abundance of wild Sablefish*	*Relative fitness of aquaculture Sablefish in the natural habitat is unknown* *Effects of any fitness decrement on the productivity of the wild population of Sablefish are unknown*
	Impacts to genetic diversity of wild Sablefish	*The extent of genetic differentiation between populations of aquaculture and wild Sablefish has not been examined* *Estimates of the effective population size for both aquaculture and wild Sablefish are unavailable*

CONCLUSIONS

Despite a lack of species-specific data, a precautionary approach would assume:

1. there is genetic differentiation between aquaculture and wild Sablefish,
2. there is at least a low rate of escape from Sablefish aquaculture facilities, and
3. Sablefish from aquaculture are capable of interbreeding with wild fish.

Genetic evidence supports a single, large population of Sablefish in BC, which implies the impact of genetic introgression from aquaculture may be naturally mitigated under current levels of licensed production. However, there are substantial knowledge gaps for Sablefish, such that a more detailed assessment of the impacts of genetic interactions is not currently possible. Additionally, to assess the risk posed by aquaculture activities to wild Sablefish, interactions beyond the scope of this report, such as ecological and health risks, would also need to be considered (DFO 2010).

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