REPLICATE DIVERGENCE BETWEEN AND WITHIN SOUNDS IN A MARINE FISH: THE COPPER ROCKFISH (*SEBASTES CAURINUS*)

by

Stefan James Dick

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ABSTRACT

Understanding the factors that influence larval dispersal and connectivity among marine populations is critical to the conservation and sustainable management of marine resources. Whereas marine populations were once thought to be panmictic through extensive larval exchange, recent studies are increasingly uncovering fine-scale substructure driven by contemporary features of the seascape such as coastal topography, bathymetry, and nearshore currents. In this study I tested the degree of genetic subdivision among ten populations of copper rockfish (*Sebastes caurinus*) representing paired samples collected from inlets and adjacent outer coast habitats in five replicate sounds on the west coast of Vancouver Island, British Columbia, using 17 microsatellite DNA loci. Overall, subdivision (F_{ST}) was low (F_{ST} = 0.031), but consistently higher between paired inlet and coast sites (mean F_{ST} = 0.047) compared to among the five coast sites (F_{ST} = -0.001) or among the five inlet sites (F_{ST} = 0.026). Heterozygosity, allelic richness, and estimates of effective population size were also consistently lower in inlet sites, suggesting local inbreeding due to limited larval delivery from coastal habitats. Bayesian analysis of population structure identified two genetic groups across all samples, a single genetic group amongst only coast sites, two genetic groups amongst only inlet sites, and two genetic groups within each sound analysed separately. Copper rockfish collected from inlets were also consistently shorter, had lower condition factors, and grew more slowly than fish collected from coast sites. My results implicate inlet-coast seascape transitions in driving the evolution of population structure, likely resulting from inlet topography and estuarine circulation patterns that act to retain

pelagic larvae within inlets and reduce physical connectivity with coastal habitats. Phenotypic differences between inlet and coast fish suggest differential growth environments across the inlet-coast seascape transition, which may promote genetic differentiation through selection against maladapted immigrant genotypes. Coast sites appear to be well served by the existing Rockfish Conservation Areas (RCAs), which have a coastally biased distribution. By contrast, inlet sites may require more specific local conservation measures as they appear to be less well connected to adjacent coast sites and may be more susceptible to overexploitation.

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INTRODUCTION

The study of landscape genetics focuses on the role of habitat features (topography, distance, environmental gradients) in structuring patterns of genetic variation within and among localities (Manel *et al*. 2003; Holderegger and Wagner 2010). While the same processes (drift, differential selection and barriers to migration) influence genetic variation in all habitats, the study of seascape genetics has lagged behind genetic studies in the terrestrial realm (Selkoe *et al.* 2008; Manel and Holderegger 2013). From recent studies in seascape genetics, it is evident that in addition to historical processes (McCusker and Bentzen 2010) and natural selection (Nielsen *et al*. 2009), contemporary features of the seascape such as coastline shape, bathymetry, and circulation patterns can heavily influence the structure of natural populations (Banks *et al.* 2007). Such factors, however, need not act independently. For instance, physical features of coastlines or island chains can create physical retention zones that influence the extent to which marine populations are connected by the exchange of pelagic eggs and larvae and, through their influence on water temperature and salinity, help to create environmental gradients that generate distinct selective regimes that may influence population structure (*e.g.*, Johanessonn and André 2006; Schmidt *et al*. 2008).

Understanding how seascape features influence larval dispersal and connectivity among marine fish populations is central to fisheries management and conservation (Palumbi 2003; Selkoe *et al.* 2008). Increasingly, studies are uncovering substructure in populations that were thought to be panmictic through extensive larval exchange (*e.g.*, Rocha-Olivares and Vetter 1999; Cunningham *et al.* 2009). Physical features of the

seascape such as coastline topography, bathymetry, currents and upwelling zones are often identified as key drivers of population structure, as these have the potential to act as barriers to larval dispersal, limiting connectivity between proximate geographic locations (*e.g.*, Buonaccorsi *et al.* 2002; Johansson *et al.* 2008). Failure to account for population substructure can lead to the rapid overexploitation of fish stocks, particularly in cases where there is limited larval delivery from outside sources (Carvalho and Hauser 1994; Stephenson 1999). While marine protected area (MPA) networks can be effective at restoring the abundance of depleted stocks, their successful establishment requires a thorough understanding of larval transport (Botsford *et al.* 2003; Shanks *et al.* 2003). This is particularly important for MPAs designed to enhance fisheries, as larval export is necessary to increase fishable biomass in unprotected areas (Botsford *et al.* 2003; Shanks *et al.* 2003).

In the northeast Pacific Ocean, rockfishes (genus *Sebastes*) are one of the most diverse and abundant groups of marine fishes. The genus consists of about 107 species, all but six of which are part of a species flock endemic to the North Pacific Ocean (Johns and Avise 1998). Rockfishes range from the intertidal zone to several thousands of metres in depth, are generally long-lived and slow to mature (some may exceed 200 years of age), and are live-bearing (Lea *et al.* 1999; Love *et al.* 2002). The copper rockfish (*Sebastes caurinus*) is a common nearshore species found from the Gulf of Alaska south to the Baja Peninsula where it occupies habitats from the shallow subtidal zone to depths of about 180 metres (Love *et al.* 2002). In British Columbia (BC), it is one of the most abundant rockfishes and is typically found in inshore waters over rocky reefs and other high relief habitats (Richards 1987; Love *et al.* 2002). Copper rockfish reach sexual maturity between six and

eight years of age and live to a maximum age of about 50 (Love *et al.* 2002). Like many other nearshore species, adult copper rockfish have very small home ranges (~100-4,000 m^2) and demonstrate strong site fidelity, especially those occupying high-value habitats (Matthews 1990; Tolimieri *et al.* 2009). Dispersal occurs almost exclusively during the pelagic larval phase, which lasts between one and six months for most rockfishes (Moser and Boehlert 1991; Love *et al.* 2002). On the west coast of Vancouver Island, BC, copper rockfish parturition (*i.e.*, larval release) occurs from March to April, followed by a pelagic phase lasting approximately two months (Markel 2011). Juvenile copper rockfish settle in shallow, nearshore habitats and typically associate with canopy-forming kelp, boulder fields, eelgrass beds, or any other structurally complex habitats (Love *et al.* 2002).

The inshore distribution of the copper rockfish, preference for areas of complex rocky substrate, and high degree of site fidelity, coupled with a relatively short pelagic larval duration may act to limit gene flow resulting in substantial population structure. In a previous study of copper rockfish, Buonaccorsi *et al*. (2002) examined microsatellite DNA variation at six loci across six localities, and documented significant population subdivision, particularly between the semi-enclosed waters of Puget Sound, Washington (WA), and coastal samples from BC, Oregon and California. Estimates of F_{ST}, a measure of the proportion of genetic variance attributable to division among subpopulations (Weir and Cockerham 1984), were about ten times higher for comparisons between Puget Sound and coastal locations than among all coastal locations combined. The authors suggested that physical watermass retention within Puget Sound was responsible for limiting larval exchange with coastal habitats. For the coastal collections, the study revealed a pattern of

isolation-by-distance (IBD), whereby genetic differentiation was positively correlated with geographic distance (Wright 1938). Johansson *et al.* (2008) expanded on the study by Buonaccorsi *et al.* by conducting fine-scale sampling of copper rockfish along the Oregon coast. Analysis of genetic variation at 11 microsatellite loci revealed significant genetic breaks at two locations within Oregon corresponding to an offshore jet at Cape Blanco and a 100-km stretch of sand habitat. The authors concluded that local seascape features such as oceanographic breaks and segments of unsuitable habitat could be responsible for the pattern of IBD observed across the species' range. In the yelloweye rockfish (*S. ruberrimus*), another sedentary demersal species with pelagic larvae, significant genetic differentiation at nine microsatellite loci occurred between samples taken from the semi-enclosed waters of the Strait of Georgia and outside waters on the west coast of Vancouver Island (Siegle *et al.* 2013). Taken together, these studies suggest that a major factor driving population subdivision in nearshore rockfish is coastline complexity and the potential for local retention zones to form owing to the interaction between topography, circulation, and life history.

Understanding how geographic features influence contemporary patterns of gene flow has important implications for rockfish conservation and management. The life history characteristics common to most rockfish, which include slow growth, late maturation and high site fidelity, make them particularly susceptible to overfishing (Parker *et al.* 2000; Berkeley *et al.* 2004). Indeed, decades of intense fishing pressure have led to the collapse of numerous rockfish stocks (Parker *et al.* 2000; Berkeley *et al.* 2004). In BC, four species of rockfish are listed as Threatened by the Committee on the Status of Endangered Wildlife in

Canada (COSEWIC) and three are listed as Special Concern (COSEWIC 2002; 2007a,b; 2008; 2009a,b; 2010). In an effort to protect vulnerable nearshore rockfishes from further exploitation and promote population recovery, Fisheries and Oceans Canada established 164 Rockfish Conservation Areas (RCAs) throughout the coast of BC (Yamanaka and Logan 2010). The RCAs were located in areas with high seafloor complexity (*i.e.*, steep, rocky habitats) and high historical commercial landings (Yamanaka and Logan 2010) in order to protect valuable rockfish habitat. However, there was little or no consideration of how well the RCAs would be connected by larval exchange, or whether or not they would protect populations of rockfishes that are especially vulnerable to overfishing due to limited recruitment from outside sources. Key to assessing the effectiveness of the current configuration of RCAs is an understanding of how BC's complex coastal topography and variable patterns of ocean circulation influence larval dispersal and population structure in nearshore rockfishes.

One of the most prominent features of the BC coast is the abundance of deeply incised inlets or fjords, of which about 79 have been documented (Thomson 1981). These inlets, especially on the west coast of Vancouver Island, have a characteristic, deep U-shaped basin, with a glacially derived mud bottom (Pickard 1963; Thomson 1981). There is usually a river at the head of the inlet and an underwater ridge, or sill, at the mouth (Pickard 1963; Thomson 1981). The mouths of such inlets empty onto the continental shelf where there is typically a seasonally variable longshore current (Thomson 1981). Together, the shelf area and inlets constitute a series of "sounds" along the west coast of Vancouver Island. The topography of these inlets creates unique circulation and environmental

gradients from the head to the mouth (Pickard 1963; Thomson 1981). For instance, freshwater flows from rivers help to create a surface outflow and a low to high surface salinity gradient from head to mouth (Pickard 1963; Thomson 1981). A deeper inflow of high salinity, oxygenated water typically occurs across the sill up towards the head of the inlet (Pickard 1963; Thomson 1981). These circulation patterns, coupled with the partial depth constriction created by the sills, and the longitudinal environmental gradients within the inlets, may strongly influence on the extent of genetic exchange between populations of rockfishes whose distributions encompass both the inlets themselves and the adjacent continental shelf. In addition, differential selective pressures at the head and the mouth of inlets may present the possibility for local adaptation to also promote genetic isolation.

In this study, I investigated the population genetic structure of copper rockfish along the west coast of Vancouver Island. Specifically, I tested the influence of inlets as putative barriers to gene flow by sampling across five replicate sets of inlet–coast seascape transitions (Figure 1). I predicted that genetic variation at 17 microsatellite DNA loci would be most pronounced among samples collected at the heads of the five inlets, followed by differences between the inlets and their adjacent outer coast (continental shelf) areas. I further predicted that, given the pronounced longshore currents that occur across the continental shelf, fish sampled from adjacent outer coast areas would be much less genetically differentiated from one another. To determine whether differences between the selective environments of inlets and outer coast sites could potentially be responsible for driving population differentiation, I measured phenotypic correlates of fitness, including length, weight and age of individual copper rockfish. I predicted that fish sampled from

inlets would be smaller and have lower growth rates than outer coast fish as a result of the greater variability in abiotic factors such as temperature, salinity and oxygen availability, which would create a more stressful growth environment (Love *et al.* 1991; Landaeta *et al.* 2012).

MATERIALS AND METHODS

Fish Collections

Copper rockfish were collected from five sounds on the west coast of Vancouver Island: Barkley Sound, Clayoquot Sound, Nootka Sound, Kyuquot Sound and Quatsino Sound (Figure 1). Within each sound, two sites were selected for sampling: an 'inlet' site at the terminal end of the longest inlet, and a 'coast' site on the exposed coastline, immediately adjacent to the opening of the sound. Sampling locations were paired in this fashion to allow for comparisons between proximate inlet and coast collections, which were separated from each other by an average of 48.0 km (range = 29-70 km). The average distance between adjacent coast sites was 70.5 km (range = 63–84 km), and between adjacent inlet sites was 129.5 km (119–140 km). All sites were named using the first letters of the sound (e.g., B, C, N, K, Q) and location (e.g., I, C). For example, the Barkley Sound inlet site was named BI.

A total of 605 copper rockfish were collected between June 2009 and October 2010 (Table 1). Two sites (Clayoquot Sound inlet and Kyuquot Sound coast) were sampled in both 2009 and 2010 to assess inter-annual stability in allelic variation. All fish were caught by hook and line from a small rigid-hull inflatable vessel. In order to maximize the distance between paired inlet and coast collection sties, fishing effort was restricted to very small geographic areas, typically on the order of 0.5 to 1 km^2. Due to low catch per unit effort at the Barkley Sound inlet site, the sampling area was broadened to approximately 2 km^2. All fishing took place at depths between 8 and 20 m over rocky-bottom habitats.

Copper rockfish were landed, euthanized immediately and placed in a cooler with ice for further processing. All other species were released immediately. At the end of each day of fishing, all copper rockfish were measured (total length) to the nearest millimeter and weighed to the nearest centigram. Fin clips were taken from the anal fins, washed in deionized water, and placed in 1.5 mL vials with 95% ethanol. Sagittal otoliths were removed from each fish, washed in deionized water and stored dry for subsequent ageing.

Age, Growth and Condition Factor

Otoliths are small calcareous structures within the inner ear of fish used in maintaining equilibrium and sound reception (Weatherley and Gill 1987). As a fish grows, its otoliths accrete layers of calcium carbonate, the rate of which varies with the growth rate of the fish (Weatherley and Gill 1987). In species like copper rockfish that live in seasonally variable thermal environments (colder in winter, warmer in summer), annual changes in somatic growth result in the formation of concentric rings or 'annuli' that are optically different from the surrounding tissue (Weatherley and Gill 1987). The age of a fish (in years) can be determined by counting these annuli, much in the same way a tree can be aged by counting its rings.

A commonly employed method used to age adult fish otoliths is the burnt section technique (or otolith break and burn technique; MacLellan 1997; Andrews *et al.* 2007). Briefly, the otoliths are snapped in half along the short axis and the exposed surface gently burned to a uniform light brown colour. The annuli become dark brown and are easily distinguished from the surrounding matrix, which retains a lighter colour. A drop of mineral oil is typically added to the burnt surface to increase clarity. The otolith burnt section

technique was used to age a total of 274 copper rockfish. The number of annual rings was counted on digital images captured using a JVC 3-CCD camera mounted on an inverted transmission light microscope. The digital imagery software Auto-Montage Pro (version 5.03, Syncroscopy, Beacon House, Cambridge) was used to record the locations of annuli and other 'checks' (marks on the otolith that do not correspond to annual rings), which aided in the estimation of age.

In age estimation of fishes, it is important to validate that annual rings actually represent approximately one year of growth (Beamish and MacFarlane 1983). Although age validation has not been completed for copper rockfish, annual growth rings have been validated for several closely related rockfish species that inhabit the North Pacific Ocean, including quillback rockfish (*S. maliger*; Kerr *et. al.* 2005), yelloweye rockfish (*S. ruberrimus*; Andrews *et al.* 2002), and canary rockfish (*S. pinniger*; Andrews *et al.* 2007).

The von Bertalanffy growth model is the most widely used length-age model in fisheries research (Bertalanffy 1938; Chen *et al.* 2002). It has been shown to accurately represent fish growth across a wide variety of taxa (Chen *et al.* 2002; Katsanevakis 2006) and has been employed in numerous studies of rockfish growth (Boehlert and Kappenman 1980; Laidig *et al*. 2003). The equation for the von Bertalanffy growth model is:

$$L(t) = L\infty * [1 - \exp(-K*(t-t_0))]$$

where L(t) = total length (cm) at age t (years),

$L\infty$ = average total maximum length (cm)

K = growth completion rate ($years^{-1}$)

t_0 = theoretical age when fish is length 0 (years)

To test for differences in growth rate between inlet and coast collections, the von Bertalanffy growth model was fitted separately to length-age data for inlet fish and coast fish using the iterative method described by Schnute (1981). Resulting growth functions were compared using likelihood ratio tests, as described by Kimura (1980). Tests were implemented in R (R Core Team, 2013).

Condition factor is a measure of the relative health of a fish, and assumes that the heavier a fish is at a given size, the better its condition (Ricker 1975). Condition factor was calculated for each fish as $100[\text{weight}/\text{length}^3]$ (Ricker 1975). Differences in mean length, weight and condition factor between sounds and between coast and inlet sites within sounds were tested using a two-way analysis of variance (ANOVA) with "sound" (N = 5) and "location" (coast, inlet, N = 10) as factors. Tests were implemented in R.

Microsatellite Assays

Total genomic DNA was extracted from fin clip tissue using a standard phenol-chloroform extraction procedure. Polymerase chain reaction (PCR) was used to amplify 17 microsatellite loci developed for *Sebastes maliger*, *S. pinniger*, and *S. rastelliger*: *Sma*2, *Sma*3, *Sma*4, *Sma*10, and *Sma*11 (Wimberger *et al*. 1999); *Spi*4, *Spi*6, *Spi*10, *Spi*12 and *Spi*18 (Gomez-Uchida *et al*. 2003); and *Sra*5-9, *Sra*7-7, *Sra*7-25, *Sra*11-103, *Sra*15-8, and *Sra*16-5 (Westerman *et al*. 2005). All loci have been previously tested on *S. caurinus* and have been shown to be polymorphic (Wimberger *et al.* 1999; Gomez-Uchida *et al.* 2003; Westerman *et al. 2005*).

Forward PCR primers were fluorescently labeled to allow for multiplexing (see Table A1 in Appendix A for additional details). Each PCR reaction was performed using a Qiagen™

Multiplex PCR Kit following the manufacturer's protocols, with approximately 150 ng template DNA in 15 μl total volume. Typical cycling conditions included an initial denaturation at 94°C for 2 minutes, followed by 30 cycles of 94°C for 1 minute, 56°C for 30 seconds, and 72°C for 1 minute. Final extension was carried out at 72°C for 10 minutes. Annealing temperatures were adjusted to optimize PCR conditions (see Table A1 in Appendix A for additional details).

The PCR products were analyzed using a CEQ™ 8000 Genetic Analysis System (Beckman Coulter™). Internal DNA size standards were included in each well, and fragments were sized using CEQ™ 8000 software (Beckman Coulter™).

Analysis of Population Structure

Genetic diversity within sampling locations was characterized using expected heterozygosity (H_E; Nei 1987) and allelic richness. Expected heterozygosity is the probability that two alleles sampled randomly from a population will be different (Nei 1987). Allelic richness was calculated for each locus at the smallest collection size (2N = 60), using the rarefaction index approach computed in the program FSTAT version 2.9.3 (Goudet 2002) that accounts for the tendency for larger samples to exhibit a greater number of alleles. Differences in mean expected heterozygosity and mean allelic richness between coast and inlet collections were tested for statistical significance using 5,000 permutations in FSTAT.

Genetic divergence within and among sampling locations was characterized using Weir and Cockerham's (1984) unbiased *F*-statistics as implemented in the program GENEPOP version 4.2 (Raymond and Rousset 1995; Rousset 2008). The *F*-statistics (F_{IS}, F_{ST}, F_{IT}) describe the amount of heterozygosity at different hierarchical levels of population

structure. More specifically, F_{IS} is the proportion of genetic variance attributable to inbreeding within a subpopulation and F_{ST} is the proportion of genetic variance attributable to division among subpopulations. Estimates of F_{IS} were calculated for each locus-population combination to measure deviation from conformance to Hardy-Weinberg equilibrium (HWE). Estimates of F_{ST} were calculated for: 1) all sites combined (global F_{ST}); 2) the five coast sites; 3) the five inlet sites; and 4) the five coast-inlet site combinations (pairwise F_{ST}).

Exact-probability tests were used to evaluate conformance to Hardy-Weinberg equilibrium (HWE) and linkage equilibrium, and to assess the homogeneity of spatial distributions of genetic variance. Estimates of exact significance probabilities were obtained using the Markov-chain algorithm as implemented in GENEPOP (1000 batches, 5000 iterations per batch). The following null hypotheses were tested:

1) Hardy-Weinberg equilibrium: alleles are randomly distributed among individuals at each locus ($F_{IS} = 0$).
2) Linkage equilibrium: alleles are randomly distributed among loci at each sampling location.
3) Allele frequency homogeneity: alleles are randomly distributed among sampling locations ($F_{ST} = 0$).

Significance levels were adjusted for multiple tests using the sequential Bonferroni correction method (Rice 1989). Significance probabilities over multiple loci were combined using Fisher's method (Sokal and Rohlf 2012).

In cases where loci did not conform to Hardy-Weinberg equilibrium, the program MICRO-CHECKER was used to detect the presence of non-amplifying (null) alleles or dropout of large alleles (Van Oosterhout *et. al.* 2004). Null alleles are alleles that fail to amplify in PCR, either because PCR conditions are not ideal or because the primer-binding region contains a mutation that inhibits binding (Selkoe and Toonen 2006). This can result in a homozygote excess (more homozygotes than would be expected under HWE) or the failure of some individuals to amplify any alleles (if the individual is a homozygote for the null allele). Large allele dropout occurs when the longer allele in a heterozygote does not amplify as well as the shorter allele, causing it to appear too faint to detect during the allele scoring process (Selkoe and Toonen 2006). Because the PCR process is more efficient for shorter sequences than longer ones, large allele dropout is more likely when two alleles are very different in size (Selkoe and Toonen 2006).

To assess the assumption of selective neutrality of the microsatellite loci used in this study, the data were analyzed using the program LOSITAN (Antao *et al.* 2008), which implements the F_{ST} outlier approach of FDIST (Beaumont and Nichols 1996). This approach works by computing a null distribution of locus-specific F_{ST} values in a plot of F_{ST} vs. expected heterozygosity using an island model of migration, and then identifying any outlying values (Beaumont and Nichols 1996; Antao *et al.* 2008). A locus that lies outside of the null distribution may be influenced by selection (divergent or balancing), either at the locus itself or at a closely linked locus.

The amount of genetic variance attributable to geographic partitioning among samples was quantified using a hierarchical analysis of molecular variance (AMOVA) in the

computer program ARLEQUIN version 3.1 (Excoffier *et al.* 2005). AMOVA uses non-parametric permutation procedures to test the significance of covariance components associated with different levels of population structure (e.g., among groups, among populations within groups, within populations). By modifying the grouping of populations (i.e., sampling locations), the user can identify the structure that maximizes among group variance. This approach is particularly useful for testing the influence of putative barriers to gene flow. In this analysis, AMOVAs were run for two different groupings of samples: 1) inlet and coast samples combined into two separate groups; and 2) paired inlet and coast samples combined into five groups corresponding to sounds.

The genetic clustering program STRUCTURE version 2.3 (Pritchard *et al.* 2000) was used to determine the most likely number of distinct genetic clusters (*K*) among the sampling locations. STRUCTURE differs from ARLEQUIN in that it uses no *a priori* information on the grouping of collections. STRUCTURE uses a Bayesian clustering method to assign individuals to genetic clusters based solely on their genotypes. An individual may be assigned to more than one cluster if its genotype indicates that it is an admixture of two or more genetic groups. A Markov chain Monte Carlo method (MCMC) is used to estimate posterior probability distributions for each possible number of clusters.

Simulations in STRUCTURE were performed using values of *K* between 1 and 15 for analyses of all 10 sampling locations, between 1 and 10 for analyses of all coast sites or all inlet sites, and between 1 and 5 for analyses of paired coast and inlet sites. All simulations were run under a model that allowed admixture and did not include a location prior. Each run consisted of a 100,000 step burn-in followed by an additional 500,000 steps. Ten

iterations were run for each value of *K*. In cases where the highest likelihood value was for a value of $K > 1$, the method of Evanno *et al.* (2005) was used. This method calculates the second order rate of change in the log probability of successive *K* values (ΔK) and is used to evaluate different solutions for *K*.

Given the relatively small spatial scale of study and relatively low levels of genetic divergence, no isolation-by-distance (IBD), identified as a significant correlation between F_{ST} and geographic distance, was apparent among coast localities. Furthermore, given the structural complexity of inlet localities relative to coast localities, no attempt was made to derive IBD relationships across all locations, as distance would be confounded by the geomorphological complexity of the coastline.

Effective Population Size

Effective population size (N_e) is defined as the number of breeding individuals in an ideal population of organisms (i.e., a hypothetical population under Hardy Weinberg equilibrium) that would experience the effects of drift or inbreeding (e.g., loss of heterozygosity) to the same degree as the observed population (Wright 1938). Estimates of N_e were derived for each of the ten copper rockfish samples using the linkage disequilibrium method of LDNe (Waples and Do 2008). The LD method operates on the principle that departures from random association between alleles across loci (linkage disequilibrium) will be inversely proportional to N_e. Because all copper rockfish samples consisted of multiple cohorts (generations), the estimates derived are more properly referred to as the effective number of breeders (N_b) rather than N_e *per se.* (see Waples 2005).

RESULTS

Length, Condition Factor, Age and Growth

Phenotypic data was collected for a total of 466 copper rockfish. Average length was 30.5 cm (range: 13.2–49.8 cm), average weight was 583 g (range: 20–2,100 g) and average condition factor was 1.73 (range: 1.05–2.28; Table 2). Inlet fish were on average 9.7 cm shorter, 530 g lighter, and had a 7.3% lower condition factor than fish from the outer coast (Table 2). Results of the two-way ANOVA with sound and location (i.e., inlet, coast) as factors indicated that there was a significant effect of both sound ($P = 0.0004$) and location ($P < 0.0001$) on mean length, as well as a significant sound*location interaction ($P < 0.0001$). Fish collected from inlet sites were consistently shorter than those collected from coast sites, especially for Clayoquot, Kyuquot, and Quatsino Sounds (Figure 2). Similarly, there was as significant effect of both sound ($P = 0.001$) and location ($P < 0.0001$) on mean weight, and a significant sound*location interaction ($P < 0.0001$). In all cases, inlet collections showed consistently lighter weights than coast collections, especially for Clayoquot, Kyuquot and Quatsino Sounds (Figure 2). In contrast, mean condition factor did not differ by sound ($P = 0.719$), but there was a significant effect of location ($P < 0.0001$) as well as an interaction between sound and location ($P < 0.0001$). Except for Kyuquot, all samples collected in inlets had lower average condition factors than those collected on the outer coast (Figure 2).

Ages of 274 copper rockfish were determined from their otoliths, including 127 fish collected from inlet sites and 147 fish collected from coast sites. The mean age of inlet fish was 10.0 years (range: 2–43 years) and the mean age of coast fish was 10.9 years (range: 4–

33 years; Table 3). The overall von Bertalanffy growth equation (i.e., fit to all data) produced an asymptotic total length ($L\infty$) of 38.2 cm (95% CI = 36.0–40.8 cm) and a growth coefficient (K) of 0.17 (95% CI = 0.15-0.21). The size-at-age clearly consisted of two groupings of points; fish collected on the outer coast were consistently longer at a given age than were fish collected from inlets (Figure 3). This same pattern of faster growth at coast sites was consistent across the four sounds for which there was comparable data.

Fitting the von Bertalanffy growth function to inlet and coast fish separately produced two markedly different growth curves (Figure 3). For inlet fish, $L\infty$ was estimated to be 30.7 cm (95% CI = 28.4–33.7 cm) compared to 42.2 cm (95% CI = 41.2–43.3 cm) for coast fish. Values of K were estimated to be 0.18 (95% CI = 0.15–0.23) for inlet fish and 0.20 (95% CI = 0.19–0.22) for coast fish. Likelihood ratio tests comparing the two growth curves confirmed that coast fish grow more quickly (H_o: $K_{coast} = K_{inlet}$, $P = 0.021$) and reach a larger maximum size (H_o: $L\infty_{coast} = L\infty_{inlet}$, $P = 0.003$) than inlet fish. Differences in t_0, the theoretical age when a fish has 0 length, were not significant (H_o: $t_{0coast} = t_{0inlet}$, $P = 0.06$). Overall, the differences between the two growth curves were highly significant (H_o: $K_{coast} = K_{inlet}$, $L\infty_{coast} = L\infty_{inlet}$, $t_{0coast} = t_{0inlet}$, $P < 0.0001$).

Microsatellite Diversity

All seventeen microsatellite loci were successfully amplified in all 605 individuals and all were polymorphic. The average number of alleles per locus was 13.1, with a range of 3 to 36 (Table 4). A total of 223 alleles were detected across all loci and collections. Expected heterozygosity at individual loci ranged from 0.08 to 0.94 (Table 4). MICRO-CHECKER found no evidence of scoring error due to stutter or large allele dropout. Average expected

heterozygosity over all collections was 0.61, with a range of 0.57 to 0.94 (Table 5). Expected heterozygosity was significantly lower (P = 0.008; Table 5) for inlet collections (mean = 0.59 ± 0.01) than coast collections (mean = 0.64 ± 0.01). Allelic richness calculated at a minimum sample size of 2N = 60 was also significantly lower (P = 0.007; Table 5) for inlet collections (mean = 6.8 ± 0.4) than coast collections (mean = 8.0 ± 0.1). Average allelic richness over all collections was 7.4 (Table 5).

Three single-locus departures from Hardy-Weinberg equilibrium (HWE) were significant after corrections for multiple tests: the *Spi*4 locus in the BC collection (F_{IS} = 0.121, P = 0.0018); the *Sma*10 locus in the KI collection (F_{IS} = 0.362, P = 0.0004); and the *Sra*16-5 locus in the QI collection (F_{IS} = 0.088, P = 0.0029). When probabilities were combined over all collections for each locus (Fisher's method), the *Sma*10 locus displayed a significant departure from HWE (F_{IS} = 0.047, P = 0.001; Table 4). When probabilities were combined over all loci for each collection, there were no significant departures from HWE (Table 5). The inbreeding coefficient (F_{IS}) for all coast collections combined was low and not significant (F_{IS} = 0.005; P = 0.353), but F_{IS} was significant for all inlet sites combined (F_{IS} = 0.027; P = 0.004; Table 5). The global inbreeding coefficient for all loci in all collections was low, but significant (F_{IS} = 0.015, P = 0.014; Table 5).

MICRO-CHECKER results suggested that the three single-locus departures from HWE were the result of null alleles. Allele frequencies at these loci were adjusted using the Brookfield (1996) null allele estimator 1 model. This method assumes that there are no null-null homozygotes, which is valid for this study because all individuals amplified at least one allele. The results of allele-frequency based analyses (i.e., F_{ST} and significance of F_{ST}) were

very similar between the corrected and uncorrected data sets; therefore, all analyses were performed using the uncorrected data set.

Allele frequencies at all loci conformed to linkage equilibrium after corrections for multiple tests, confirming the assumption of independent assortment. Tests for selective neutrality in LOSITAN found evidence of selection at two loci: *Sra*11-103 and *Spi*18. Both loci had F_{ST} values that fell above the neutral distribution of F_{ST}, suggesting possible divergent selection at these loci. Removing *Sma*10 and *Spi*18 from the dataset did not affect the results of allele-frequency based analyses (i.e., F_{ST} and significance of F_{ST}); therefore, both loci were included in subsequent analyses.

Effective Population Size

The linkage disequilibrium method produced estimates of N_b from about 100 in the BI and NI samples to over 2,800 in the CC sample (Table 6). Several localities produced negative estimates of N_b owing to insufficient linkage disequilibrium within the sample. Point estimates of N_b averaged 1,181 (SD = 1,418) for the coast samples and 401 (SD = 526) for the inlet samples.

Population Structure

Genetic variance was strongly partitioned among sampling locations. Comparisons between sample years (2009 and 2010) were not significant either for the CI location (F_{ST} = 0.0023, $P > 0.10$) or for the KC location (F_{ST} = 0.0018, $P > 0.15$). Consequently both years' samples were pooled for each sample. When all ten collections were contrasted, sixteen of the seventeen loci exhibited significant heterogeneity in allele frequency (Table 7). The

global F_{ST} for all loci over all collections was 0.031 and highly significant (P <0.0001; Table 7). This variance was largely driven by differentiation between coast and inlet collections. Pairwise F_{ST} values for the five coast-inlet comparisons ranged from 0.042 to 0.054 (mean = 0.047) and all were highly significant (P < 0.0001; Table 8). When only the coast collections were contrasted, allele frequencies were homogeneous at all loci (Table 7). Overall F_{ST} was small and not significant (F_{ST} = -0.001, P = 0.737; Table 7), suggesting a single panmictic population among coast sites. By contrast, among the inlet collections significant heterogeneity in allele frequency was detected at thirteen of the seventeen loci (Table 7). Overall F_{ST} was significant (F_{ST} = 0.026, P < 0.0001; Table 7) and higher than that among the coast collections, but only about half of the average coast-inlet value.

Consistent with the F_{ST} values, analysis of molecular variance suggested that the most significant axis of genetic divergence was between samples grouped into coast and inlet localities. Here, variation between coast and inlet groups was significant (3.5%, P = 0.006) and higher than variation between samples within these two groupings (1%, P < 0.0001), with the majority of variation residing within individual samples (95.5%, P < 0.0001; Table 9). An alternative arrangement where coast and inlet samples were grouped into the five distinct sounds resulted in non-significant variation among groups (-1.8%, P = 0.93), but significant variation between coast and inlet samples within sounds (4.7%, P < 0.0001; Table 10). Again, the majority of variation resided within individual samples (97.1%, P < 0.0001).

The pronounced genetic differentiation between coast and inlet samples was strongly supported by the STRUCTURE analyses. Across all samples, the most likely number

of populations (i.e., genetic clusters) was K =2. Although Ln probability estimates were similar for values of K between 2 and 7, ΔK, calculated using the Evanno (2005) method, clearly found greatest support for two populations (Figure 4). For runs where K =2, coast and inlet samples showed strikingly different proportions of the two genetic groups (Figure 4), indicting that the majority of individuals were assigned to a cluster corresponding to the location from which they were sampled (i.e., coast vs. inlet). When the coast samples were examined by themselves, the most likely number of populations was K = 1 (Figure 5). By contrast, when the inlet samples were analyzed by themselves, K = 2 was the most likely number of populations, with the Clayoquot Sound inlet sample distinct from the others (Figure 6). In each of the analyses conducted separately by sound, the most likely number of populations was also K = 2 (Figures 7 to 11). Consistent with the F_{ST} values and AMOVA results, inlet and coast samples showed strikingly different proportions of the two genetic groups (Figures 7 to 11), providing further support for genetic differentiation between coast and inlet sites.

DISCUSSION

In this study I found strong evidence for repeated genetic divergence between samples of copper rockfish collected from coast and inlet sites across relatively small spatial scales in five sounds on the west coast of Vancouver Island. I also found significant genetic differentiation among the five samples collected within the inlets, but no differentiation among coastal samples. These results suggest that physical features of the seascape, in this case shallow sills and narrow channels characteristic of the five study inlets, may limit gene flow between proximate geographic locations, resulting in fine-scale population structure in copper rockfish. The lack of genetic differentiation among coastal samples suggests that strong coastal currents promote strong inter-site dispersal, at least at the scale of my investigation. Further, my study has revealed consistent differences in size and growth rate between coast and inlet collections, with inlet fish showing lower condition factors and slower growth rates. These phenotypic differences suggest that the outer coast is a more favourable growth environment, possibly due higher productivity or lower variability in abiotic factors (e.g., temperature, salinity, dissolved oxygen; Pickard *et al.* 1963; Thomson 1981; Love *et al.* 1991; Landaeta *et al.* 2012). Consequently, selection against maladapted immigrant phenotypes in contrasting environments may also play a role in driving the population genetic structure of copper rockfish (Marshall *et al.* 2010).

The Role of Coastal Topography and Hydrography in Structuring Marine Populations

Contemporary oceanographic and seascape factors are increasingly recognized for their role in structuring current patterns of population subdivision in marine species

exposed to natural selection, migration, drift, and their interactions across both space and time (*e.g.*, Planes *et al*. 2001; Helleberg *et al*. 2002; Galindo *et al*. 2006; Hedgecock *et al*. 2007). In this context, oceanographic factors influencing the ability and direction of movement during dispersive phases of the life cycle of marine species (*e.g.*, upwelling, currents and gyres) are recognized as potentially important influences on marine connectivity and the evolution of population structure (Sinclair and Iles 1989; Stepien 1999; White *et al.* 2010, but see Benzie and Williams 1997). Patterns of ocean circulation may be heavily influenced by physical landform features such as depth profiles, islands, the continental shelf, or coastline complexity (*e.g.*, Farmer and Freeland 1983; Maier-Reimer *et al*. 1990; Ridgway and Dunn 2003) and such influence may be an important driver in the evolution of population genetic structure (*e.g.*, Banks *et al.* 2007; Fontaine *et al*. 2007; Schultz *et al*. 2008) and more general patterns of marine diversification (*e.g.*, Avise 1992; Bernardi *et al.* 2003; Jacobs *et al.* 2004; Banks *et al.* 2007).

The results of my study suggest that coastal topography is an important factor influencing population structure in copper rockfish. Despite finding no genetic differentiation among coastal collections sampled over approximately 300 km, I found significant differentiation between coast and inlet collections spaced as little as 29 km apart. This suggests that inlets, through their influence on currents and circulation patterns, restrict larval dispersal between adjacent coast and inlet localities. This finding is consistent with studies of other marine fishes and invertebrates that have been shown to exhibit substantial differentiation in phenotype and genetic traits between populations located in coastal areas and those within and among inlets (*e.g.*, Jørstad and Nævdal 1989; Girever

and Stien 1998; Suneetha and Nævdal 2001, Perrin *et al.* 2004). In Norwegian Atlantic cod (*Gadus morhua*), Atlantic herring (*Clupea harengus*), and pearlside (*Maurolicus muelleri*), morphological, biochemical and molecular data have demonstrated significant differentiation among fjords and between fjords and adjacent coastal areas (Jørstad and Nævdal 1989; Girever and Stien 1998; Suneetha and Nævdal 2001, respectively). In their study of pearlside, Suneetha and Nævdal (2001) found significant differences in allele frequencies at twelve allozyme loci among samples collected in five west Norwegian fjords and one offshore area in the North Sea. Analysis of four meristic and body proportion measures (number of gill rakers, body depth, head length and relative eye diameter) were found to be similar among fjords but significantly different between fjords and the offshore area. The authors concluded that fjord topography, specifically the presence of shallow sills, combined with differences in the selective environments of fjords and offshore habitats were likely the primary drivers of population structure in Norwegian pearlside. Perrin *et al.* (2004) found a similar pattern of genetic divergence between fjord and open coast populations of the eleven-arm seastar (*Coscinasterias muricata*) collected from fourteen fjords and three open coast locations in New Zealand. Significant differentiation at mitochondrial D-loop sequences was observed among fjords and between fjords and open coast populations separated by as little as 10 km (range 10-200 km), suggesting a strong isolating mechanism limiting gene flow. The authors concluded that estuarine circulation patterns likely act to retain larvae within fjords, resulting in population divergence through random genetic drift.

Several population genetic studies of marine fishes distributed in the northeast Pacific Ocean have also found patterns of differentiation that support the role of nearshore marine topography in limiting larval dispersal between semi-enclosed inland waters and the open coast. In their study of Pacific cod (*Gadus macrocephalus*), Cunningham *et al.* (2009) reported greater genetic differentiation when samples collected from fjord-like areas of the inner Strait of Georgia, BC, and Puget Sound, WA, were compared to open coast samples (F_{ST} of 0.012) than when open coast samples, separated by comparable or even much greater distances, were compared to each other (F_{ST} of ~ 0.003 to 0.004). Siegle *et al*. (2013) also noted marked differentiation between Straight of Georgia and outer coast samples of yelloweye rockfish (*Sebastes ruberrimus*), but no differentiation among coastal samples collected over approximately 2,000 km from the southwest coast of Vancouver Island to Alaska. A previous study on copper rockfish also identified heightened divergence between the Strait of Georgia and Puget Sound and open coastal areas of the northeastern Pacific Ocean (F_{ST} of ~ 0.025 to 0.098 for Puget Sound / Strait of Georgia vs. coastal, compared to -0.001 to 0.022 for coastal comparisons; Buonaccorsi *et al.* 2002). Buonaccorsi *et al.* (2005) found a similar pattern in brown rockfish (*S. auriculatus*), whereby genetic differentiation was substantially higher for comparisons between Puget Sound and coastal localities in California and Mexico (F_{ST} of 0.048 to 0.138) than comparisons among coastal sites only (F_{ST} of 0.014).

The results of my study add to a growing body of literature that suggests coastal topography and local circulation patterns are important factors influencing the population structure of nearshore marine fishes. By replicating my comparisons across five adjacent

pairs of inlet and open coast sites, I have found compelling evidence of parallel divergence across a common seascape transition (inlet to coast). My results are consistent with those of other studies that have investigated population structure of marine organisms in fjord and adjacent open coast habitats, suggesting that a common set of processes are responsible for driving the evolution of population structure in these species.

Genetic Divergence Among Coastal Sites

The absence of divergence among the five coastal collection sites I sampled (F_{ST} = -0.001) is consistent with studies of copper rockfish over even larger distances in coastal environments. Johansson *et al.* (2008) reported slight, but significant isolation-by-distance (IBD) and an F_{ST} of only 0.0042 among 12 localities spanning more than 2,000 km of coastline in Washington, Oregon, and California. Over a small range (450 km, seven localities) in Oregon, $F_{ST,}$ although statistically significant, was only 0.001 and IBD was not detected (Johansson *et al.* 2008). The inshore, shallow-water nature of copper rockfish habitat use, the sedentary nature of adults, behavioural attributes of larvae, and habitat breaks along the Oregon coast (rocky versus sandy areas) may reduce offshore advection of larvae and help generate some population structure despite a larval duration period of 2-3 months (Buonaccorsi *et al.* 2002; Johansson *et al*. 2008). These processes appeared to act together to generate a mesoscale dispersal pattern on the northwest US coast, in which populations are self-recruiting on a regional scale, but habitat disjunctions may produce genetic breaks or distinctions (Gunderson and Vetter 2006; Johansson *et al*. 2008). In my study, copper rockfish sampled across less than 300 km of coastline also appear to exhibit a

mesoscale pattern of dispersal, as I detected no IBD among the coastal localities, but significant genetic differentiation between each inlet-coast comparison.

The widespread dispersal of copper rockfish among the five coastal localities sampled in my study is likely facilitated by the predominant pattern of alongshore currents on the west coast of Vancouver Island. The dominant offshore current is the west – east Subarctic Current, a large, slow trans-Pacific current that bifurcates into the northward flowing Alaska Current and the southward flowing California Current between 45-50^{o}N latitude and 130-150^{o}W longitude (Thomson 1981). Seasonal variation in the strength of offshore atmospheric pressure systems moves the bifurcation point of the Subarctic Current as far south as the coasts of Washington and Oregon in the winter and as far north as the Alaskan Panhandle in the summer (Thomson 1981; Crawford *et al.* 2007). This results in a seasonal reversal of the prevailing coastal currents along the west coast of Vancouver Island, with the northward flowing Davidson Current dominating during the winter months (October-March), and the southward flowing California Current most prominent during the summer months (April-September; Thomson 1981). Copper rockfish larvae released by females at coastal spawning sites in the late winter and early spring may be advected considerable distances by the predominant alongshore currents during their 2-3 month larval phase, explaining the lack of population structure among my five coastal localities.

Genetic Divergence Between Inlets and Coastal Sites

My analyses indicated that F_{ST} between coast and inlet collections of copper rockfish averaged 0.047, which is comparable to that reported by Buonaccorsi *et al*. (2002) for differentiation between Puget Sound and coastal collections (F_{ST} = 0.063), although their

analysis included localities that were separated by far greater distances (>2000 km) than in this study (maximum 70 km). Paired comparisons between coast and inlet sites within individual sounds were also remarkably consistent with one another, varying by a maximum of only about 11% from the maximum pairwise comparison. This argues that the processes influencing genetic divergence between inlet and coastal sites are relatively consistent among the five sounds I investigated. On a geological time scale, this divergence is recent, as the inlets would have been colonized following the retreat of glaciers at the end of the last ice age, about 10,000 years before present (Thomson 1981). This suggests that contemporary processes, perhaps in combination with historical vicariant events (i.e., deglaciation), are acting to restrict gene flow between the inlets and coastal sites.

Inlets typically have an estuarine-type circulation pattern where low salinity surface flow generated by freshwater inputs from rivers at the head of each inlet is replaced by deeper, higher salinity water inflow from coastal localities (Thomson 1981; Farmer and Freeland 1983). Such a circulation pattern might facilitate at least some exchange of pelagic copper rockfish larvae between coastal and inlet sites. The level of exchange would, however, be influenced by the residence time of water within the inlet. Buonaccorsi *et al.* (2002) argued that because the residence time of water within Puget Sound, WA (50% retention over 7 months), exceeded that of the typical copper rockfish larval duration period (2-3 months) there would be limited exchange of larvae between Puget Sound and outside waters. Residence times for inlets on the west coast of Vancouver Island may be considerably less than that reported for Puget Sound, given their smaller size and direct openings to the continental shelf (Pickard 1963). Drinkwater and Osborn (1975) estimated

that it would take approximately one month for 85% of the water in the Rupert-Holberg basin of Quatsino Sound (one of my collection locations) to be replaced under average tidal conditions. Although estimates of residence times are not available for the other four study inlets, their similarity in terms of size, depth and general hydrography suggests that water replacement would occur at a similar rate as in Rupert-Holberg Inlet (Pickard 1963). Consequently, physical watermass retention alone may not explain the significant genetic divergence I found between inlet and coastal samples of copper rockfish.

Icthyoplankton surveys conducted along the coasts of Washington, Oregon and California have found larval rockfish to be most concentrated in the upper mixed layer of the water column above the thermocline (Ahlstrom 1959; Boehlert *et al.* 1985; Lenarz *et al.* 1991; Moser and Boehlert 1991). For example, Boehlert *et al.* 1985 sampled two stations off the coast of Oregon and reported that 95% of rockfish larvae (not identified to species) occurred at depths shallower than 40 m (70% were collected within the 5-30 m range). Similarly, in a study of the larval distribution of *Sebastes capensis* in the fjords of southern Chile, Landaeta and Castro (2006) found larval abundance to be highest in the upper 50 m. Although no studies have specifically investigated the larval distribution of copper rockfish, recruitment rates are highest in shallow nearshore habitats (Love *et al.* 1991), suggesting that pelagic larvae are most abundant in near-surface waters. In the five inlets I sampled, the net outflow of low-salinity surface water characteristic of estuarine circulation would be expected to entrain copper rockfish larvae distributed in the upper water column and transport them out of the inlets. Conversely, the net inflow of deep, saline water from the continental shelf would be ineffective at transporting larvae into the inlets, as the bulk flow

would occur at depths greater than the vertical distribution of larvae. This pattern of one-way transport out of the inlets is consistent with my findings of low allelic richness and low expected heterozygosity in inlet samples of copper rockfish, which suggest local inbreeding and limited larval delivery from the outer coast. High levels of effective dispersal from inlets to adjacent coastal habitats would, however, be expected to homogenize allele frequencies over time. This is at odds with the significant divergence I found between inlet and coast collections of copper rockfish, and suggests that other processes may be acting in combination with inlet topography and hydrography to limit gene flow.

For instance, larval behaviours that limit dispersal away from natal sites may significantly reduce connectivity among marine populations (discussed in Miller and Shanks 2004; Cowen and Sponaugle 2009). These behaviours, which include rheotaxis, substrate orientation, schooling, and vertical migration in the water column, may contribute to local recirculation and retention of larvae despite strong dispersal vectors (Marliave 1986; Miller and Shanks 2004; Cowen and Sponagule 2009). At least some rockfishes appear capable of olfaction-based specific habitat recognition (Mitamura *et al.* 2005). Spawning choice by adults that reduces exposure of larvae to coastward currents has also been proposed as a possible explanation for the limited dispersal distances observed in several nearshore rockfishes, including copper rockfish (Buonaccorsi *et al.* 2002, 2004). Larval behaviours that limit dispersal would be adaptive if passive advection away from natal sites resulted in high rates of larval mortality (Cowen *et al.* 2000). For copper rockfish larvae spawned in inlets, the primary dispersal vector is the outflow of low-salinity surface water generated by freshwater inputs from rivers. During peak parturition of copper rockfish (March-April;

Markel 2011), spring rains and melting snowpacks result in high discharges of freshwater into Vancouver Island's inlets (Pickard 1963; Thomson 1981). Surface water salinity values during this time may be as low as zero parts per thousand near the heads of inlets, increasing gradually down-inlet as the freshwater layer absorbs deeper, saltier water (Thomson 1981). Although there are no studies on salinity tolerance for any species of rockfish, it is presumed that mortality rates would be higher for larvae entrained in low-salinity surface waters than those that remained at depth. This would both reduce effective dispersal out of the inlets (*i.e.*, due to high mortality) and promote behavioural adaptations that limit passive advection.

Genetic differentiation between inlet and coast collections of copper rockfish appears to be driven by a combination factors, which likely include physical retention of water within the inlets due to constricted openings to the continental shelf (*e.g.*, narrow channels and shallow sills), estuarine circulation that favours net transport of larvae out of the inlets but may result in high larval mortality in the low-salinity surface flow, and behavioural adaptations that favour larval retention. Moreover, selection against immigrant genotypes in contrasting environments may also play an important role in limiting effective dispersal between inlets and outer coast habitats (Marshall *et al.* 2010). While the relative contribution of these factors to the observed pattern of genetic differentiation is unknown, my study provides strong evidence that their influence is similar across multiple inlet-coast comparisons. This suggests that the processes acting to limit gene flow between inlets and outer coast populations of copper rockfish may also drive the evolution of population

structure in other nearshore rockfishes, and perhaps even unrelated taxa with similar distributions.

Genetic Divergence Among Inlets

A surprising result of my study was that divergence among inlet collections (F_{ST} = 0.026) was lower than divergence between paired inlet-coast collections (F_{ST} = 0.047). Bayesian clustering analysis in STRUCURE also supported the similarity among inlet collections, as it found only the Clayoquot Sound sample to be genetically distinct from the others. This result cannot be explained by physical or environmental limitations on larval dispersal, as the distances between inlet sites are considerably greater than the distances between adjacent inlet and coast sites. Considering that the inlets on the west coast of Vancouver Island were glaciated until about 10,000 years ago, it is probable that colonization occurred at approximately the same time in all of the inlets (Thomson 1981; Buonaccorsi *et al.* 2002). A post-Pleistocene founder event, followed by limited gene flow from the outer coast, may explain the genetic similarity of inlet populations of copper rockfish. Low allelic diversity is considered to be indicative of historical founder events (Leberg 1992), and all inlet samples I analyzed had significantly lower allelic richness than adjacent coastal samples. Buonaccorsi *et al.* (2002) proposed a similar explanation for the significant divergence they found between Puget Sound and coastal populations of copper rockfish. They used computer simulations to show that high gene flow (N_Em = 200) since the Pleistocene would rapidly erode genetic divergence that may have resulted from a founder event, which supported their conclusion that historical divergence followed by

contemporary limitations to gene flow was responsible for the present-day population structure of copper rockfish.

The genetic distinctiveness of the Clayoquot Sound inlet collection suggests that larval exchange with the outer coast is more limited in this inlet than it is in others. Geographic distance alone does not explain this result, as the distance between the Clayoquot inlet and coast sites (36 km) is the second shortest after Kyuquot Sound (29 km). However, oceanographic investigations of Clayoquot Sound report that of all the inlets on Vancouver Island, those of Clayoquot Sound exhibit the greatest difference in open ocean water characteristics, likely because of their extremely shallow sills (Pickard 1963). In addition, my inlet sample of copper rockfish in Clayoquot Sound was collected at the head of Tofino Inlet, which is considered to be a permanently anoxic basin (Pickard 1963; Nuwer and Keil 2005). This suggests that there is little or no inflow of deep, oxygen-rich water into Tofino Inlet, which would provide the vector for larval delivery from the open coast. The isolation of copper rockfish inhabiting Tofino Inlet would lead more rapid differentiation through random genetic drift, and this is consistent with my results.

Phenotypic Variation and Natural Selection

My data show that copper rockfish collected from inlet sites are smaller and have lower condition factors, a general measure of body robustness (Ricker 1975), than fish sampled from coast sites. Further, my length at age analyses indicate that fish inhabiting the open coast environment grow much faster than those living within inlets. These results strongly suggest that conditions for growth are better at coastal sites than at inlet sites and, therefore, that the two habitats are not ecologically equivalent. Like all inlets on the BC

coast, the five inlets I investigated are influenced by seasonally variable freshwater inflows from rivers, which affect water temperature, salinity, suspended sediment levels and water clarity, with resultant effects on biotic elements (Pickard 1963; Landaeta *et al.* 2012). In addition, shallow sills at the mouths of the inlets restrict the inflow of oxygen-rich water from the continental shelf, resulting in prolonged periods of anoxia within the inlet basins (Pickard 1963). This variability in abiotic conditions is likely responsible for the slower growth and lower condition factors of inlet copper rockfish, either through direct physiological effects or by way of changes to the composition and abundance of prey (Love *et al.* 1991; Landaeta *et al.* 2012).

The consistent phenotypic differences between copper rockfish from inlet and coast sites suggest the possibility for variation in selection between habitats. Divergent natural selection acting in contrasting marine environments is common (e.g., Nielsen *et al.* 2006, 2009; Galindo *et al.* 2010), and may contribute to the strong genetic differentiation I observed between inlet and coast collections. In fact, larval dispersal between inlet and coast sites may be much more extensive than my data suggest, but strong selection against maladapted immigrant genotypes reduces effective connectivity and promotes genetic differentiation (Marshall *et al.* 2010). My study did not use loci thought to be under selection, as the objective of my research was to investigate population structure using neutral genetic markers. However, a variety of genetic tools are available for identifying and isolating genes under selection (*e.g.*, Beaumont and Balding 2004; Foll and Gaggiotti 2008; Schmidt *et al.* 2008; Nielsen *et al.* 2009). Future research into the role of natural selection in

driving genetic divergence across the inlet-coast seascape transition is needed to further our understanding of the evolution of population structure in copper rockfish.

Implications for Rockfish Conservation and Management

Decades of heavy fishing pressure have led to significant population declines in numerous species of rockfish in both Canada and the US (Parker *et al.* 2000; Berkeley *et al.* 2004; Yamanaka and Logan 2010). The life history characteristics common to most rockfish, which include slow growth, late maturation and high site fidelity, make them particularly susceptible to overexploitation, and slow to recover even in the absence of fishing (Parker *et al.* 2000; Berkeley *et al.* 2004). In 2002, in response to mounting concern for inshore rockfish in BC, Fisheries and Oceans Canada (DFO) introduced a Rockfish Conservation Strategy aimed at reversing long-term declines in abundance and rebuilding depleted populations (Yamanaka and Logan 2010). A key component of this strategy was the implementation of Rockfish Conservation Areas (RCAs), which are areas of inshore habitat closed to commercial and recreational fishing for rockfishes (DFO 2007). Given that RCAs are part of an explicit conservation strategy to help rebuild populations coastwide, some understanding of the level of connectivity amongst RCAs and intervening habitat is essential to their evaluation.

The results of my study have three important implications for rockfish conservation and management. First, my analyses document previously unknown spatial genetic diversity in a nearshore rockfish: a repeated pattern of divergence across inlet-coast seascape transitions. Such biodiversity should be considered in conservation plans for copper rockfish (i.e., the potential existence and recognition of ecotypes) to ensure adequate

representation of distinct populations within RCAs. My results may also be applicable to other species of rockfishes that occupy both inlet and coastal habitats. Quillback rockfish (*S. maliger*) and brown rockfish (*S. auriculatus*) are closely related to copper rockfish and both were collected in abundance during my inlet sampling. Quillback rockfish have experienced significant population declines in BC (Yamanaka and Logan 2010), and are listed as a species of Special Concern by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC 2009b). In addition, yelloweye rockfish (*S. ruberrimus*) are reported to occur in high abundance in the large inlets on the mainland coast of BC, including Knight Inlet (Lynne Yamanaka, DFO, Nanaimo, BC, *pers. comm.*), and are also listed as Special Concern by COSEWIC (COSEWIC 2008). Future population genetic studies of these and other inshore rockfishes may resolve a similar pattern of genetic divergence across the inlet-coast seascape transition.

Second, my analyses provide insight into the patterns of larval dispersal in a common nearshore rockfish, and can be used to qualitatively assess the level of connectivity among RCAs along the west coast of Vancouver Island. Of the 164 RCAs distributed throughout BC, 15 are found on the west coast of Vancouver Island (DFO 2007). Thirteen of these are located along the outer coast, while only two are found within inlets (Holberg Inlet in Quatsino Sound and Bedwell Sound in Clayoquot Sound; DFO 2007). Given that my data suggest widespread effective dispersal amongst coastal sites, inferred from the genetic similarity of coastal collections, all the coastally distributed RCAs are likely well connected to each other and to intervening areas, at least when considered over multiple generations. This suggests that coastal RCAs may function to not only reduce rockfish mortality within

each RCA, but also provide some export of within-RCA production to non-RCA localities, and thus serve as an effective "network" of reserves (Gaines *et al.* 2010). By contrast, the inlet sites appear to be much less well served by RCAs. Of the five inlet sites I sampled, only the Quatsino Sound site (Holberg Inlet) is in close proximity of an RCA (~10 km; DFO 2007). In addition, my data suggest that connectivity between coast and inlet sites is restricted and that the coastally biased distribution of RCAs may not function well in terms of exporting larvae into the inlets. Thus, inlet sites may not be part of an effective reserve network (*i.e.*, they have few RCAs themselves and are more isolated from the coastal RCA network). This suggests that the size of individual inlet RCAs (to ensure self-sustainability, Botsford *et al*. 2003; Shanks *et al.* 2003) and conservation regulations not related to RCA establishment (*e.g.*, fishing regulations in adjacent areas) are especially important for the inlet sites.

Finally, my data show that copper rockfish inhabiting inlets grow significantly more slowly and have lower condition factors than fish living on the exposed outer coast. Because fecundity in rockfishes is positively related to fish size (Haldorson and Love 1991), this means that larval production is lower per individual in inlets than it is on the coast. While copper rockfish abundance estimates are not available for the five study inlets, the steep inlet walls combined with abiotic constraints on distribution (*e.g.*, low salinity near the surface, low oxygen at depth; Pickard 1963), suggest suitable rockfish habitat is more limited in inlets than it is on the coast. My estimates of effective numbers of breeders (N_b) also support lower population sizes in inlets compared to the outer coast. Low abundance and low fecundity combined with limited larval delivery from outside sources make inlet populations of copper rockfish particularly susceptible to overfishing. Indeed, low catch per

unit effort at the Barkley Sound inlet site (near Port Alberni) can likely be attributed to the popularity of this area as a destination for recreational anglers. Recovery of depleted inlet populations is expected to be slow, given their lack of connectivity with coastal populations and their low intrinsic growth rates. This further emphasizes the need for increased representation of inlets in the RCA network, as well as conservation regulations specific to inlet areas.

Conclusion

Several recent studies have measured population structure of Pacific coast marine fishes (*e.g.*, Rocha-Olivares and Vetter 1999; Bernardi *et al*. 2003; Buonaccorsi *et al.* 2002, 2005; Johansson *et al.* 2008; Cunningham *et al.* 2009; Sivasundar and Palumbi 2010). These studies commonly reveal patterns of isolation-by-distance across large geographic scales (*e.g.*, Buonaccorsi *et al*. 2002, 2005), relatively sharp "breaks" in the distribution of genetic variants associated with major physiographic features (*e.g.*, Rocha-Olivares & Vetter 1999; Bernardi *et al.* 2003; Sivasundar and Palumbi 2010), or some combination of such effects (*e.g.*, Buonaccorsi *et al.* 2002; Johansson *et al.* 2008). My study illustrates how seascape topography and hydrography, in particular the incision of coastlines by inlets and the associated currents and circulation patterns, are important drivers of population structure in common nearshore rockfish. My analyses suggest that in addition to physical restriction of larval dispersal between inlets and outer coast habitats, selection against immigrant genotypes in divergent selective environments may further reduce connectivity across the inlet-coast seascape transition. While more research is needed to fully understand the complex processes that are responsible for the evolution of population structure in marine

fishes, my results represent an important contribution to our current state of knowledge. In addition, my results have direct implications for the conservation of rockfishes in BC, and will hopefully be used to inform future management actions to protect these remarkable creatures.

TABLES

Table 1. Locations and sample sizes for collections of copper rockfish (*Sebastes caurinus*) from five sounds on the west coast of Vancouver Island, British Columbia.

Region	Location	Abbreviation	Latitude	Longitude	*N*
Barkley Sound	Inlet	BI	49°11'26" N	124°49'24" W	30
	Coast	BC	48°47'35" N	125°13'00" W	51
Clayoquot Sound	Inlet	CI	49°13'32" N	125°35'55" W	105
	Coast	CC	49°07'29" N	125°58'05" W	60
Nootka Sound	Inlet	NI	49°38'03" N	126°04'30" W	49
	Coast	NC	49°34'45" N	126°40'15" W	52
Kyuquot Sound	Inlet	KI	50°11'00" N	127°18'05" W	53
	Coast	KC	49°59'20" N	127°25'11" W	100
Quatsino Sound	Inlet	QI	50°38'02" N	127°56'54" W	52
	Coast	QC	50°27'04" N	128°03'45" W	53

Table 2. Phenotypic characteristics of ten samples of copper rockfish (*Sebastes caurinus*) collected from the west coast of Vancouver Island, British Columbia. Sample size (*N*), mean total length (cm) and standard error (*SE*), mean weight (g) and standard error (*SE*), and mean condition factor (100[weight/length^3]) and standard error (*SE*). BI: Barkley inlet; BC: Barkley coast; CI: Clayoquot inlet; CC: Clayoquot coast; NI: Nootka inlet; NC: Nootka coast; KI: Kyuquot inlet; KC: Kyuquot coast; QI: Quatsino inlet; and QC: Quatsino coast.

Collection	*N*	Total Length (cm)		Weight (g)		Condition Factor	
		Mean	*SE*	Mean	*SE*	Mean	*SE*
BI	10	34.3	1.53	713	74	1.71	0.04
BC	51	35.4	0.74	855	58	1.78	0.02
CI	50	22.8	0.41	200	11	1.61	0.02
CC	49	36.8	0.73	956	57	1.82	0.02
NI	50	27.8	0.56	382	22	1.69	0.03
NC	50	30.3	0.98	572	52	1.79	0.02
KI	54	23.3	0.75	247	24	1.72	0.03
KC	48	35.7	0.70	816	46	1.70	0.02
QI	50	25.6	0.39	278	13	1.61	0.03
QC	54	36.4	0.69	930	51	1.81	0.02
All Inlet	214	25.3	0.33	297	12	1.66	0.01
All Coast	252	34.9	0.37	827	25	1.78	0.01
Overall	466	30.5	0.34	583	19	1.73	0.01

Table 3. Mean ages of nine samples of copper rockfish (*Sebastes caurinus*) collected from the west coast of Vancouver Island, British Columbia. No fish from the Nootka Sound inlet site were aged. Sample size (*N*), mean age (years), and standard error of mean age (*SE*). BI: Barkley inlet; BC: Barkley coast; CI: Clayoquot inlet; CC: Clayoquot coast; NI: Nootka inlet; NC: Nootka coast; KI: Kyuquot inlet; KC: Kyuquot coast; QI: Quatsino inlet; and QC: Quatsino coast.

Collection	***N***	**Age (years)**	
		Mean	***SE***
BI	10	10.4	1.1
BC	48	11.2	0.9
CI	49	9.4	0.2
CC	10	10.9	1.6
NC	24	8.7	0.8
KI	53	10.0	1.0
KC	49	11.4	0.7
QI	14	11.4	1.1
QC	16	11.3	1.7
All Inlet	127	10.0	0.4
All Coast	147	10.9	0.5
Overall	274	10.4	0.3

Table 4. Single locus summary statistics for copper rockfish (*Sebastes caurinus*) assayed at 17 microsatellite DNA loci. Sample size (*N*), allele number (*A*), expected heterozygosity (H_E), observed heterozygosity (H_O), local inbreeding coefficient (F_{IS}) and significance of F_{IS} (*P*). Adjusted significance level (α) for multiple comparisons = 0.003.

Locus	***N***	***A***	H_E	H_O	F_{IS}	***P***
*Sma*2	605	5	0.34	0.33	-0.004	0.102
*Sma*3	605	9	0.56	0.55	0.008	0.459
*Sma*4	605	8	0.56	0.54	0.035	0.464
*Sma*5	605	6	0.08	0.08	-0.025	0.999
*Sma*10	605	21	0.67	0.63	0.047	0.001
*Sma*11	605	8	0.58	0.58	0.000	0.236
*Spi*4	605	20	0.87	0.87	0.003	0.009
*Spi*6	605	24	0.88	0.87	0.006	0.573
*Spi*10	605	5	0.59	0.57	0.029	0.443
*Spi*12	605	5	0.48	0.46	0.022	0.834
*Spi*18	605	23	0.80	0.77	0.031	0.050
*Sra*5-9	605	3	0.50	0.52	-0.023	0.459
*Sra*7-7	605	16	0.58	0.56	0.027	0.073
*Sra*7-25	605	12	0.79	0.77	0.024	0.393
*Sra*11-103	605	5	0.44	0.42	0.026	0.299
*Sra*15-8	605	14	0.79	0.79	0.007	0.940
*Sra*16-5	605	39	0.94	0.93	0.008	0.158
Average	605	13.1	0.61	0.60	0.015	0.014

Table 5. Summary statistics for ten samples of copper rockfish (*Sebastes caurinus*) assayed at 17 microsatellite DNA loci. Sample size (*N*), average allele number at 2*N* = 60 (*A*), average expected heterozygosity (H_E), observed heterozygosity (H_O), local inbreeding coefficient (F_{IS}) and significance of F_{IS} (*P*). BI: Barkley inlet; BC: Barkley coast; CI: Clayoquot inlet; CC: Clayoquot coast; NI: Nootka inlet; NC: Nootka coast; KI: Kyuquot inlet; KC: Kyuquot coast; QI: Quatsino inlet; and QC: Quatsino coast. Adjusted significance level (α) for multiple comparisons = 0.005.

Collection	*N*	*A*	H_E	H_O	F_{IS}	*P*
BI	30	7.3	0.57	0.55	0.044	0.201
BC	51	8.1	0.64	0.63	0.020	0.082
CI	105	6.2	0.61	0.61	-0.005	0.197
CC	60	7.9	0.64	0.65	-0.018	0.586
NI	49	6.8	0.59	0.56	0.047	0.023
NC	52	8.2	0.64	0.64	0.002	0.919
KI	53	6.9	0.59	0.55	0.068	0.007
KC	100	7.9	0.64	0.63	0.012	0.459
QI	52	6.9	0.59	0.58	0.025	0.536
QC	53	8.1	0.63	0.62	0.006	0.195
All Inlet	289	6.8	0.59	0.58	0.027	0.004
All Coast	316	8.0	0.64	0.63	0.005	0.353
Overall	605	7.4	0.61	0.60	0.015	0.014

Table 6. Estimates of effective number of breeders (N_b) in inlet and coast collections of copper rockfish (*Sebates caurinus*) assayed at 17 microsatellite DNA loci. Estimates were derived using the linkage disequilibrium method with the lowest allele frequency class of 0.01 used in the calculations. An estimate of ∞ denotes that insufficient linkage disequilibrium was present to estimate N_b. BI: Barkley inlet; BC: Barkley coast; CI: Clayoquot inlet; CC: Clayoquot coast; NI: Nootka inlet; NC: Nootka coast; KI: Kyuquot inlet; KC: Kyuquot coast; QI: Quatsino inlet; and QC: Quatsino coast.

Collection	N_b	*95% CI*
BI	90.2	60.6 – 164.7
BC	304.5	177.5 – 932.7
CI	1,185.4	463.2 – ∞
CC	2,818.0	406.3 – ∞
NI	105.9	80.0 – 149.7
NC	421.5	210.2 – 7,485.0
KI	224.1	137.0 – 547.1
KC	1,611.7	677.4 – ∞
QI	∞	392.2 – ∞
QC	2,818.1	405.6 – ∞
All Inlet*	401	526 (SD)
All Coast	1,181	1,418 (SD)

* excludes QI collection

Table 7. Genetic differentiation (F_{ST}) estimates for all collections combined, coast collections only, and inlet collections only for copper rockfish (*Sebastes caurinus*) assayed at 17 microsatellite DNA loci. Proportion of genetic variance attributable to division among samples (F_{ST}) and significance of F_{ST} (P). Adjusted significance level (α) for multiple comparisons = 0.003.

Locus	All Collections		Coast Collections Only		Inlet Collections Only	
	F_{ST}	P	F_{ST}	P	F_{ST}	P
*Sma*2	0.0170	<0.001	-0.004	0.940	0.032	<0.001
*Sma*3	0.0328	<0.001	0.003	0.166	0.054	<0.001
*Sma*4	0.0256	<0.001	-0.019	0.458	0.019	<0.001
*Sma*5	0.0106	0.005	0.000	0.713	-0.002	0.099
*Sma*10	0.0297	<0.001	0.000	0.404	0.003	0.003
*Sma*11	0.0153	<0.001	0.003	0.178	0.030	<0.001
*Spi*4	0.0313	<0.001	0.002	0.007	0.035	<0.001
*Spi*6	0.0303	<0.001	-0.002	0.720	0.028	<0.001
*Spi*10	0.0331	<0.001	-0.004	0.694	0.042	<0.001
*Spi*12	0.0099	<0.001	-0.006	0.679	0.008	0.036
*Spi*18	0.0540	<0.001	0.000	0.540	0.015	<0.001
*Sra*5-9	0.0170	<0.001	-0.005	0.946	0.030	<0.001
*Sra*7-7	0.0179	<0.001	0.002	0.628	0.025	<0.001
*Sra*7-25	0.0228	<0.001	-0.001	0.789	0.041	<0.001
*Sra*11-103	0.0943	<0.001	0.000	0.454	0.011	0.025
*Sra*15-8	0.0422	<0.001	-0.002	0.889	0.012	<0.001
*Sra*16-5	0.0180	<0.001	-0.003	0.794	0.020	<0.001
Overall	0.0307	<0.001	-0.001	0.737	0.026	<0.001

Table 8. Pairwise genetic differentiation (F_{ST}) estimates for coast-inlet comparisons of copper rockfish assayed at 17 microsatellite DNA loci. Proportion of genetic variance attributable to division among samples (F_{ST}), significance of F_{ST} (P), and distance in kilometers between paired coast-inlet sites (Distance in km). BI: Barkley inlet; BC: Barkley coast; CI: Clayoquot inlet; CC: Clayoquot coast; NI: Nootka inlet; NC: Nootka coast; KI: Kyuquot inlet; KC: Kyuquot coast; QI: Quatsino inlet; and QC: Quatsino coast. Adjusted significance level (α) for multiple comparisons = 0.01.

Pairwise comparison	F_{ST}	P	*Distance (km)*
BF vs. BC	0.044	<0.001	57
CF vs. CC	0.043	<0.001	36
NF vs. NC	0.051	<0.001	48
KF vs. KC	0.042	<0.001	29
QF vs. QC	0.054	<0.001	70
Average	0.047	<0.001	48

Table 9. Analysis of molecular variance (AMOVA) results for samples of copper rockfish (*Sebastes caurinus*) combined into coast and inlet groups (group 1 = Barkley inlet, Clayoquot inlet, Nootka inlet, Kyuquot inlet, and Quatsino inlet; group 2 = Barkley coast, Clayoquot coast, Nootka coast, Kyuquot coast, and Quatsino coast). *P*-values represent the significance of the variation observed at each hierarchical level of population structure.

Source of variation	***d.f.***	***Sum of squares***	***Variance components***	***Percentage of variation***	***P***
Among groups	1	127.9	0.189	3.45	0.00087
Among samples within groups	8	98.3	0.060	1.10	< 0.0001
Within samples	1200	6294.8	5.246	95.46	< 0.0001

Table 10. Analysis of molecular variance (AMOVA) results for samples of copper rockfish (*Sebastes caurinus*) combined into five groups corresponding to sounds (group 1 = Barkley inlet and Barkley coast; group 2 = Clayoquot inlet and Clayoquot coast; group 3 = Nootka Inlet and Nootka coast; group 4 = Kyuquot inlet and Kyuquot coast; group 5 = Quatsino inlet and Quatsino coast). P-values represent the significance of the variation observed at each hierarchical level of population structure.

Source of variation	***d.f.***	***Sum of squares***	***Variance components***	***Percentage of variation***	***P***
Among groups	4	54.2	-0.099	-1.83	0.92803
Among samples within groups	5	171.9	0.254	4.71	< 0.0001
Within samples	1200	6294.8	5.246	97.12	< 0.0001

FIGURES

Figure 1. Copper rockfish (*Sebastes caurinus*) collection locations along the west coast of Vancouver Island, British Columbia. Blue circles show coast sites located at the seaward opening of each sound. Red circles show inlet sites located at the head of the longest inlet within each sound.

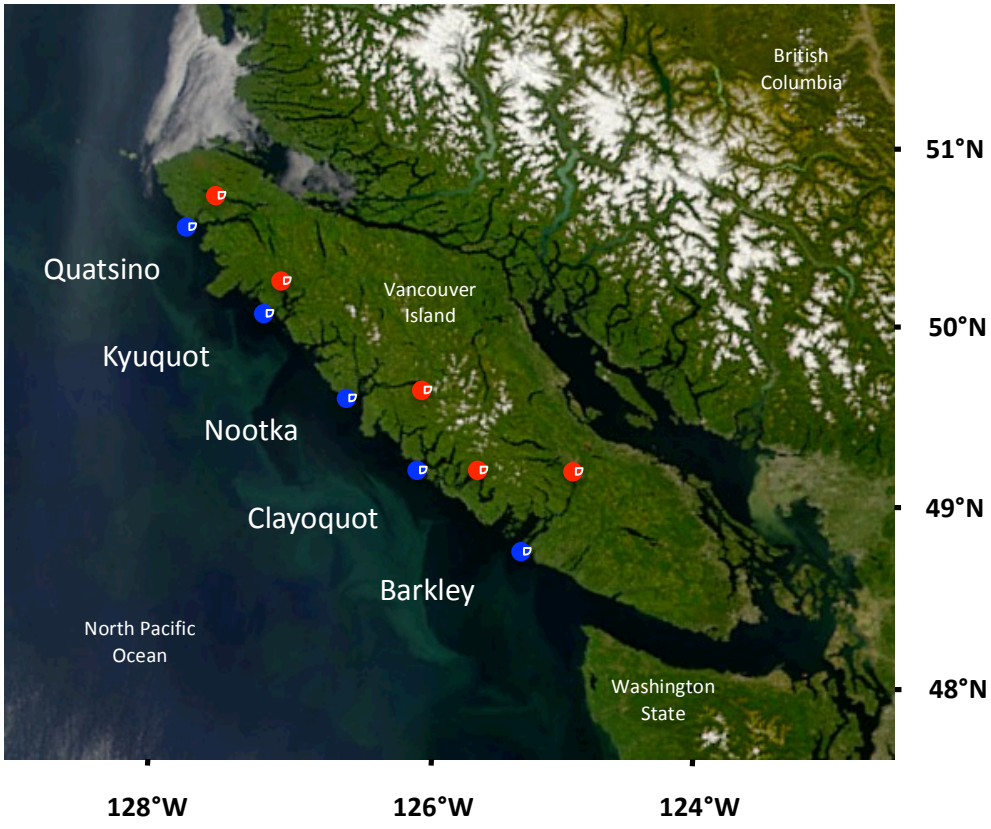


Figure 2. Variation in A) total length (cm); B) weight (g); and C) condition factor (100[g/cm]^3) in copper rockfish (*Sebastes caurinus*) collected from five sounds along the west coast of Vancouver Island, British Columbia. Blue bars are coastal sites and red bars are inlet sites within each of the five sounds (Barkley, Clayoquot, Nootka, Kyuquot and Quatsino). Small open circles are outliers, heavy horizontal lines indicate the median, dashed vertical lines represent the range, and the box represents the second (upper box border) and third (lower box border) quartiles.

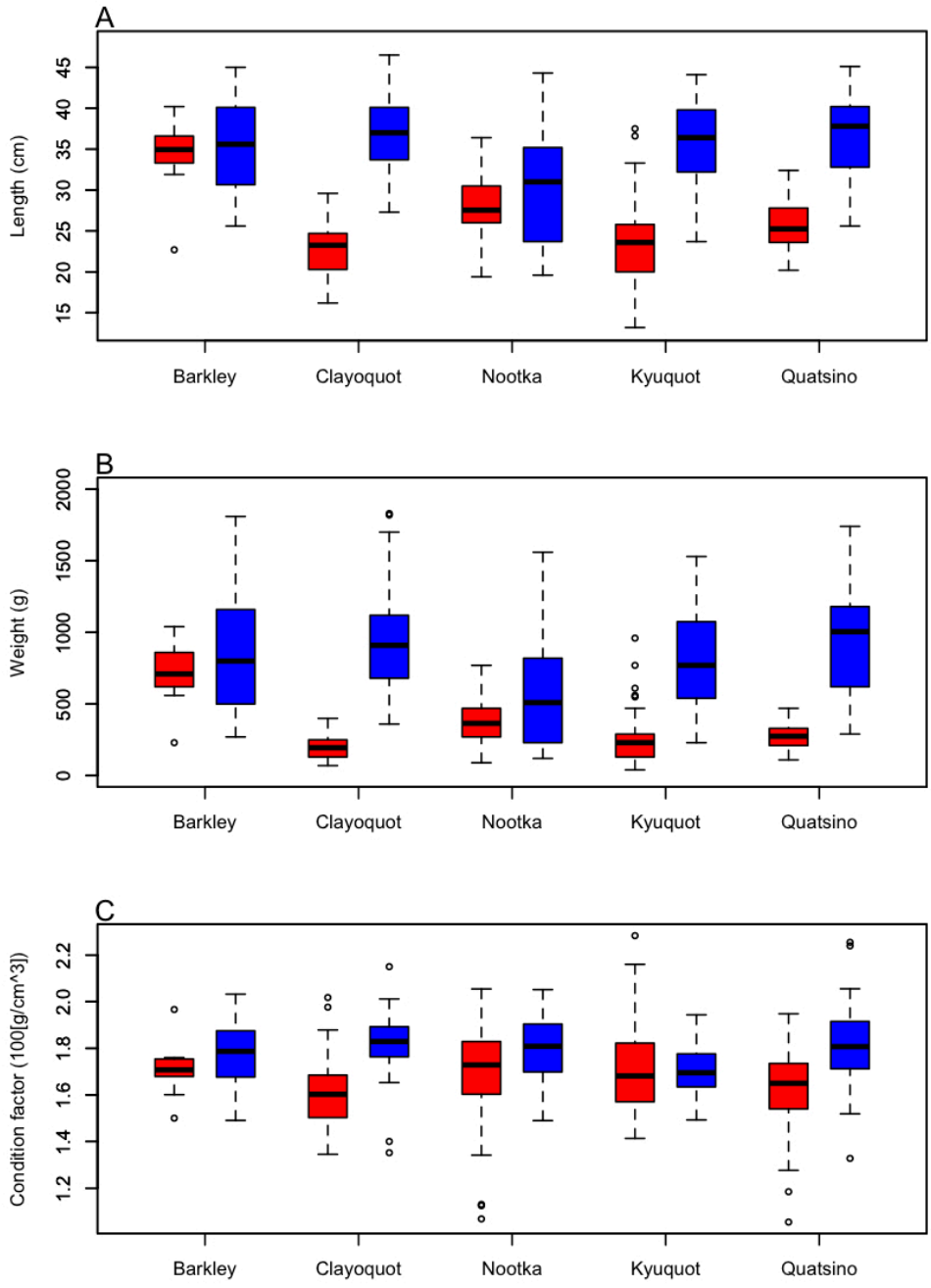


Figure 3. Length-at-age data for copper rockfish (*Sebastes caurinus*) collected from coast sites (blue, n = 147) and inlet sites (red, n = 127) on the west coast of Vancouver Island, British Columbia. Lines represent fitted von Bertalanffy growth functions.

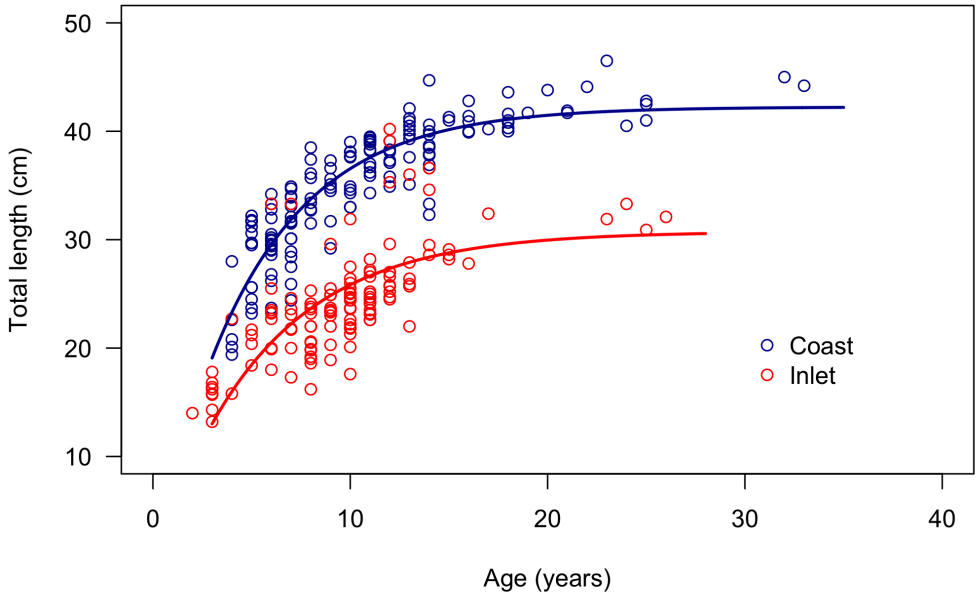


Figure 4. Results of STRUCTURE simulations testing values of K between 1 and 15 for all ten coast and inlet samples of copper rockfish (*Sebastes caurinus*) combined. A) Mean Ln probability of K (± SD) calculated from 10 replicate runs for each value of K. B) Second order rate of change of mean Ln probability of K (Delta K) calculated for successive values of K using the method of Evanno (2005). C) STRUCTURE output for K = 2 averaged over 10 runs. Each fish's genotype is represented by a thin vertical line, the blue and red portions of which represent the proportional contribution of one or two genetic groups to the admixture coefficient, Q (Pritchard et al. 2000). BI: Barkley inlet; BC: Barkley coast; CI: Clayoquot inlet; CC: Clayoquot coast; NI: Nootka inlet; NC: Nootka coast; KI: Kyuquot inlet; KC: Kyuquot coast; QI: Quatsino inlet; and QC: Quatsino coast.

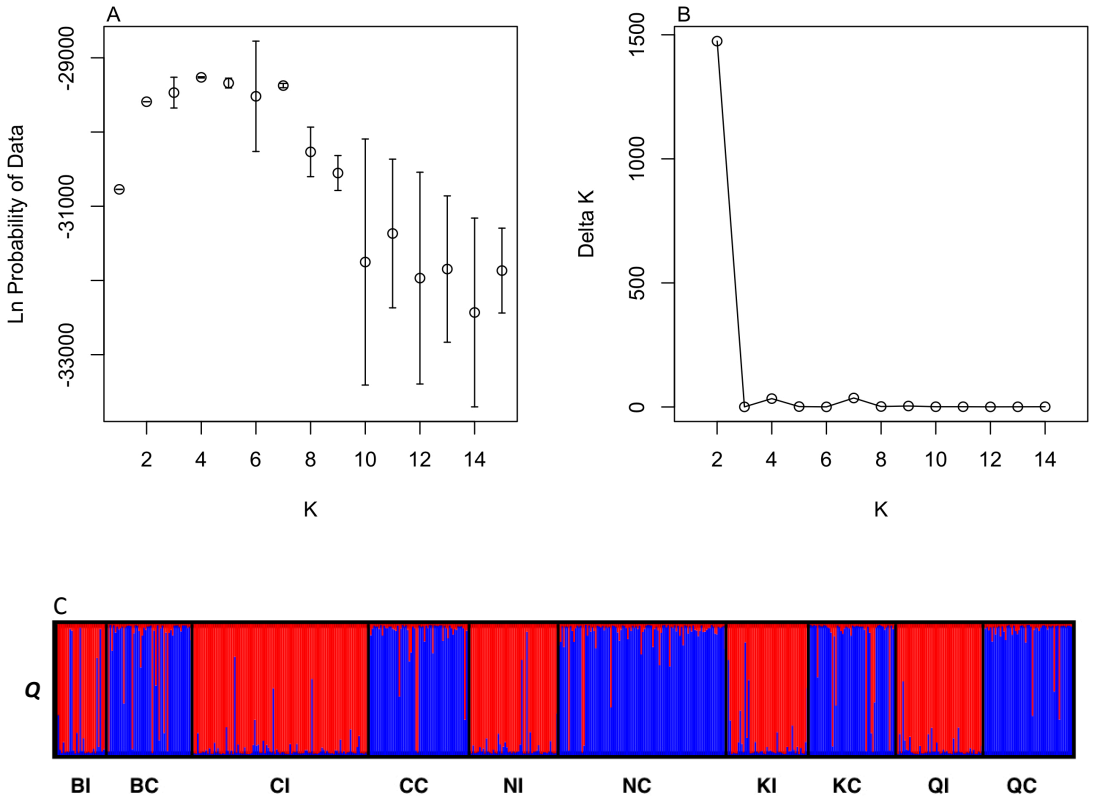


Figure 5. Results of STRUCTURE simulations testing values of K between 1 and 10 for the five coast samples of copper rockfish (*Sebastes caurinus*). Mean Ln probability of K (± SD) calculated from 10 replicate runs for each value of K.

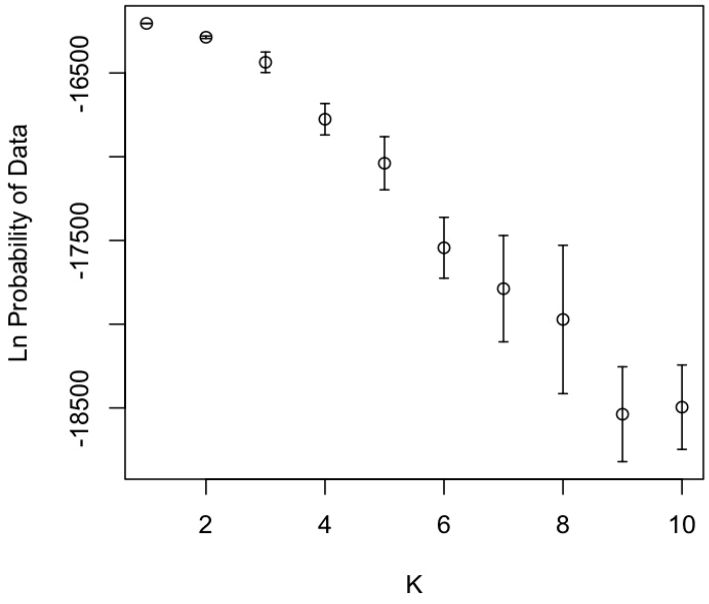


Figure 6. Results of STRUCTURE simulations testing values of K between 1 and 10 for the five inlet samples of copper rockfish (*Sebastes caurinus*). A) Mean Ln probability of K (± SD) calculated from 10 replicate runs for each value of K. B) Second order rate of change of mean Ln probability of K (Delta K) calculated for successive values of K using the method of Evanno (2005). C) STRUCTURE output for K = 2 averaged over 10 runs. Each fish's genotype is represented by a thin vertical line, the blue and red portions of which represent the proportional contribution of one or two genetic groups to the admixture coefficient, Q (Pritchard et al. 2000). BI: Barkley inlet; CI: Clayoquot inlet; NI: Nootka inlet; KI: Kyuquot inlet; and QI: Quatsino inlet.

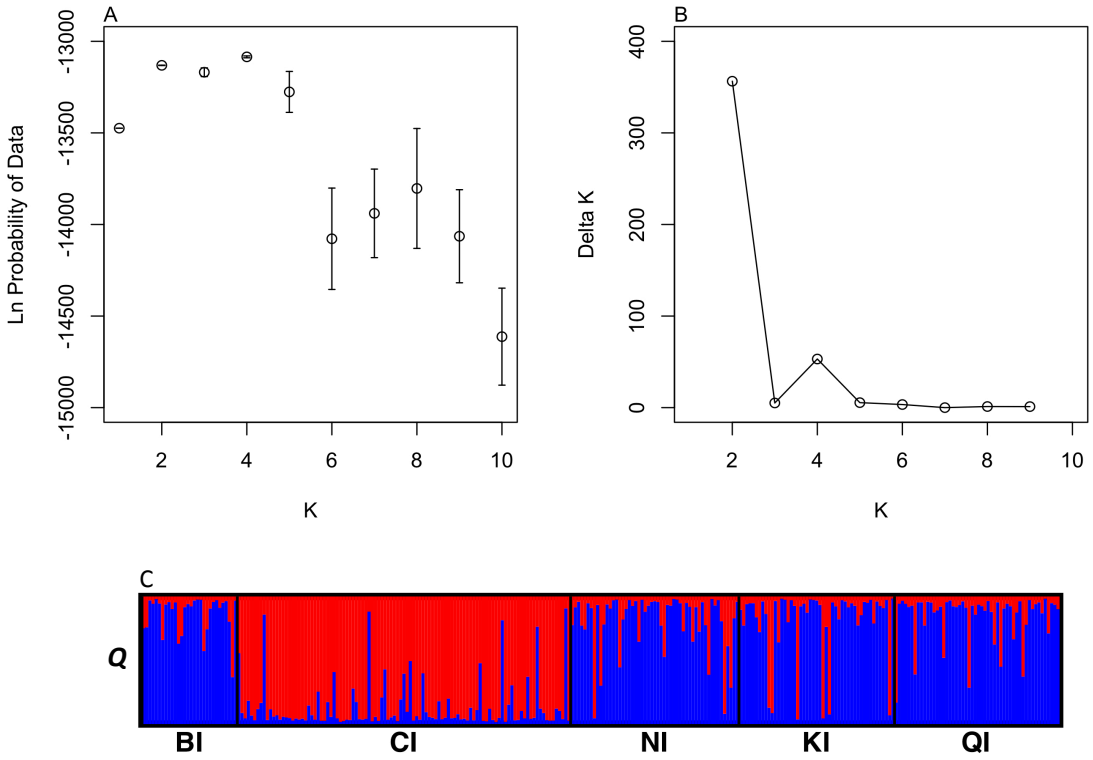


Figure 7. Results of STRUCTURE simulations testing values of K between 1 and 5 for Barkley Sound coast and inlet samples of copper rockfish (*Sebastes caurinus*). A) Mean Ln probability of K (± SD) calculated from 10 replicate runs for each value of K. B) Second order rate of change of mean Ln probability of K (Delta K) calculated for successive values of K using the method of Evanno (2005). C) STRUCTURE output for K = 2 averaged over 10 runs. Each fish's genotype is represented by a thin vertical line, the blue and red portions of which represent the proportional contribution of one or two genetic groups to the admixture coefficient, Q (Pritchard et al. 2000).

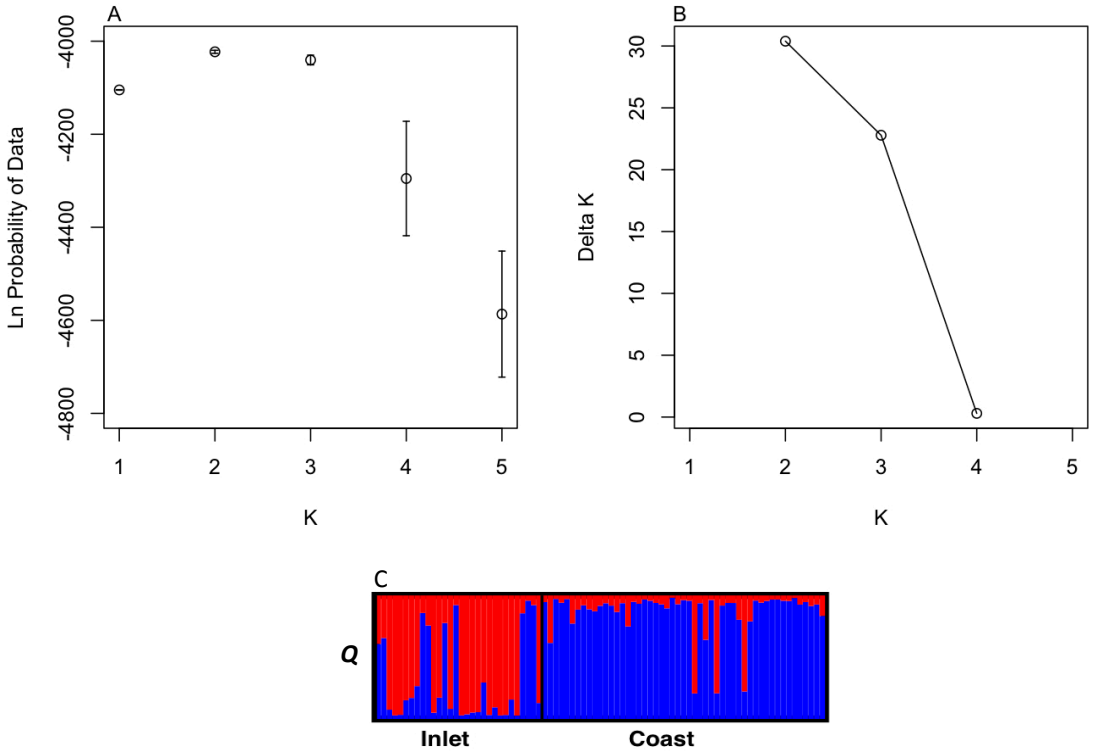


Figure 8. Results of STRUCTURE simulations testing values of K between 1 and 5 for Clayoquot Sound coast and inlet samples of copper rockfish (*Sebastes caurinus*). A) Mean Ln probability of K (± SD) calculated from 10 replicate runs for each value of K. B) Second order rate of change of mean Ln probability of K (Delta K) calculated for successive values of K using the method of Evanno (2005). C) STRUCTURE output for K = 2 averaged over 10 runs. Each fish's genotype is represented by a thin vertical line, the blue and red portions of which represent the proportional contribution of one or two genetic groups to the admixture coefficient, Q (Pritchard et al. 2000).

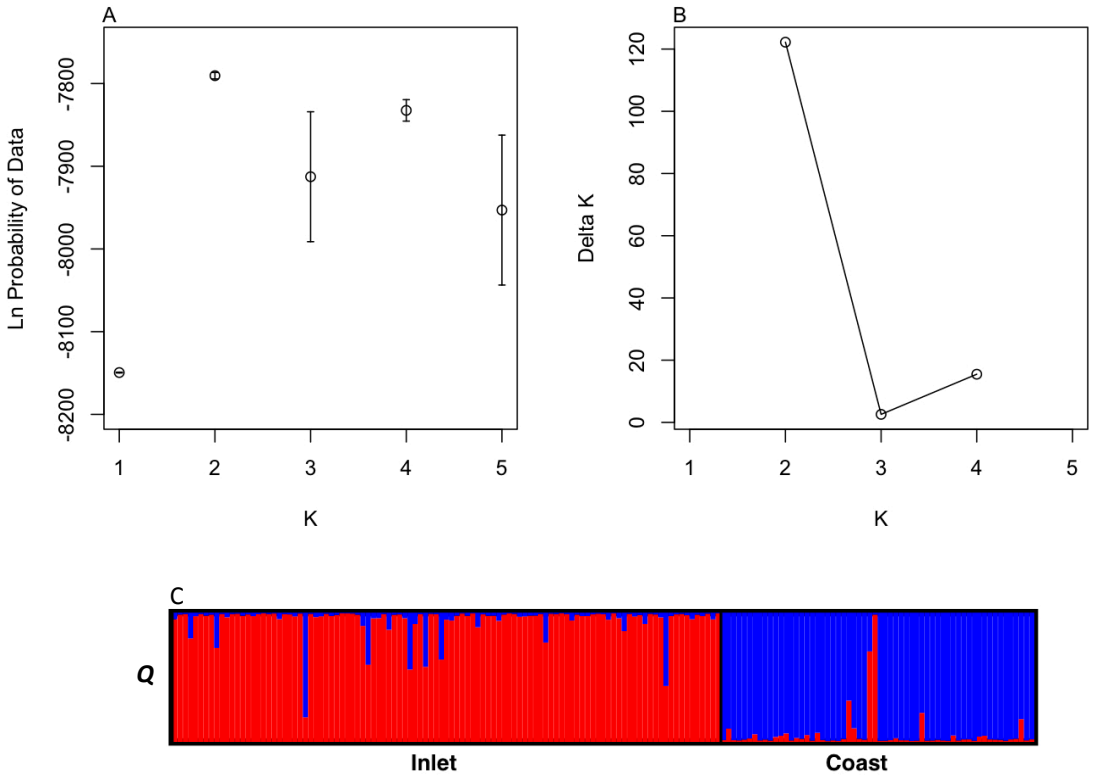


Figure 9. Results of STRUCTURE simulations testing values of K between 1 and 5 for Nootka Sound coast and inlet samples of copper rockfish (*Sebastes caurinus*). A) Mean Ln probability of K (± SD) calculated from 10 replicate runs for each value of K. B) Second order rate of change of mean Ln probability of K (Delta K) calculated for successive values of K using the method of Evanno (2005). C) STRUCTURE output for K = 2 averaged over 10 runs. Each fish's genotype is represented by a thin vertical line, the blue and red portions of which represent the proportional contribution of one or two genetic groups to the admixture coefficient, *Q* (Pritchard *et al.* 2000).

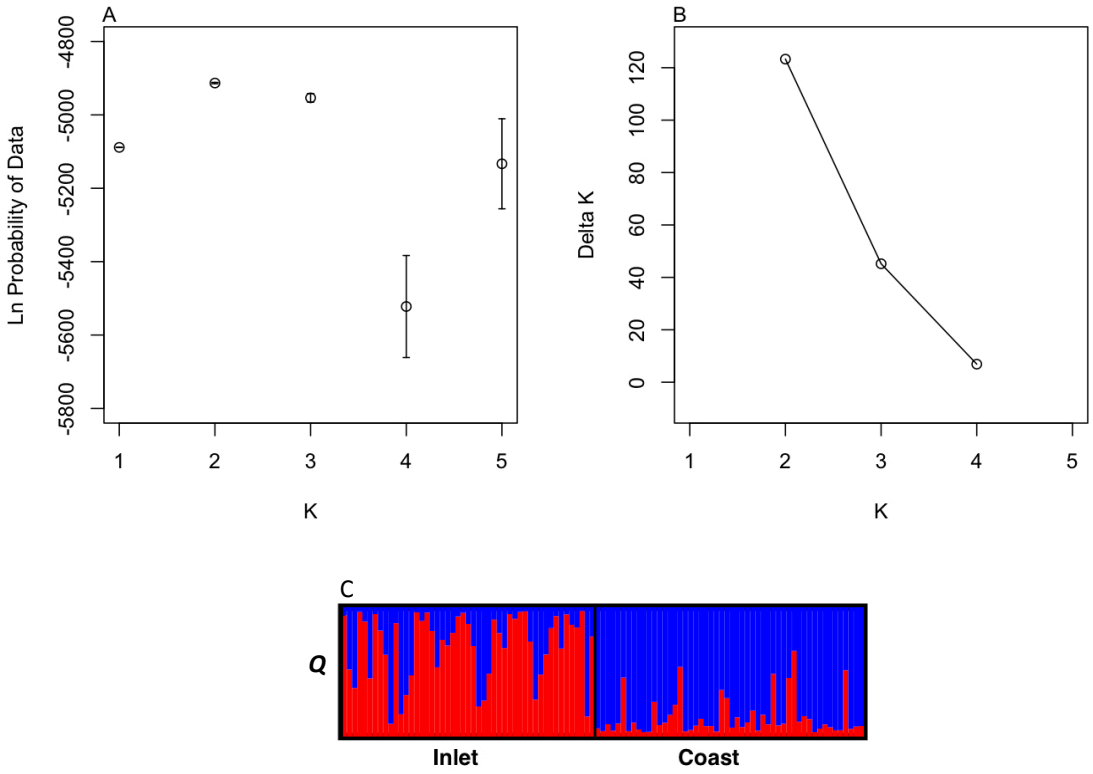


Figure 10. Results of STRUCTURE simulations testing values of K between 1 and 5 for Kyuquot Sound coast and inlet samples of copper rockfish (*Sebastes caurinus*). A) Mean Ln probability of K (± SD) calculated from 10 replicate runs for each value of K. B) Second order rate of change of mean Ln probability of K (Delta K) calculated for successive values of K using the method of Evanno (2005). C) STRUCTURE output for K = 2 averaged over 10 runs. Each fish's genotype is represented by a thin vertical line, the blue and red portions of which represent the proportional contribution of one or two genetic groups to the admixture coefficient, *Q* (Pritchard *et al.* 2000).

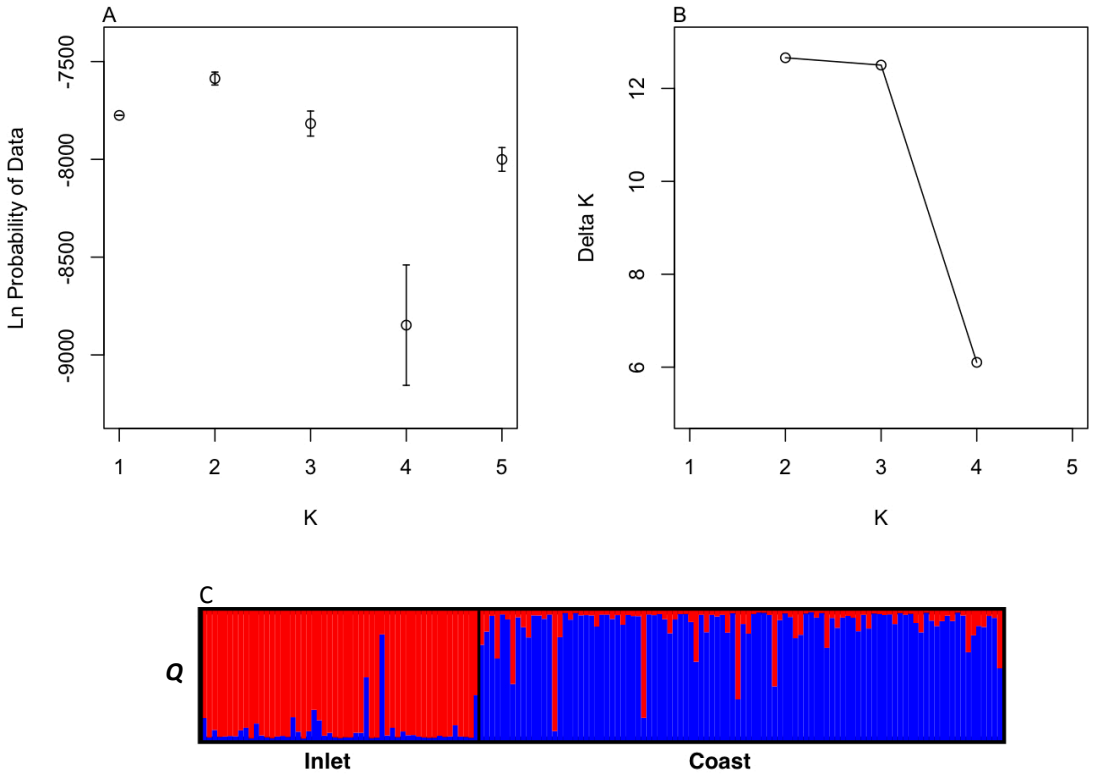
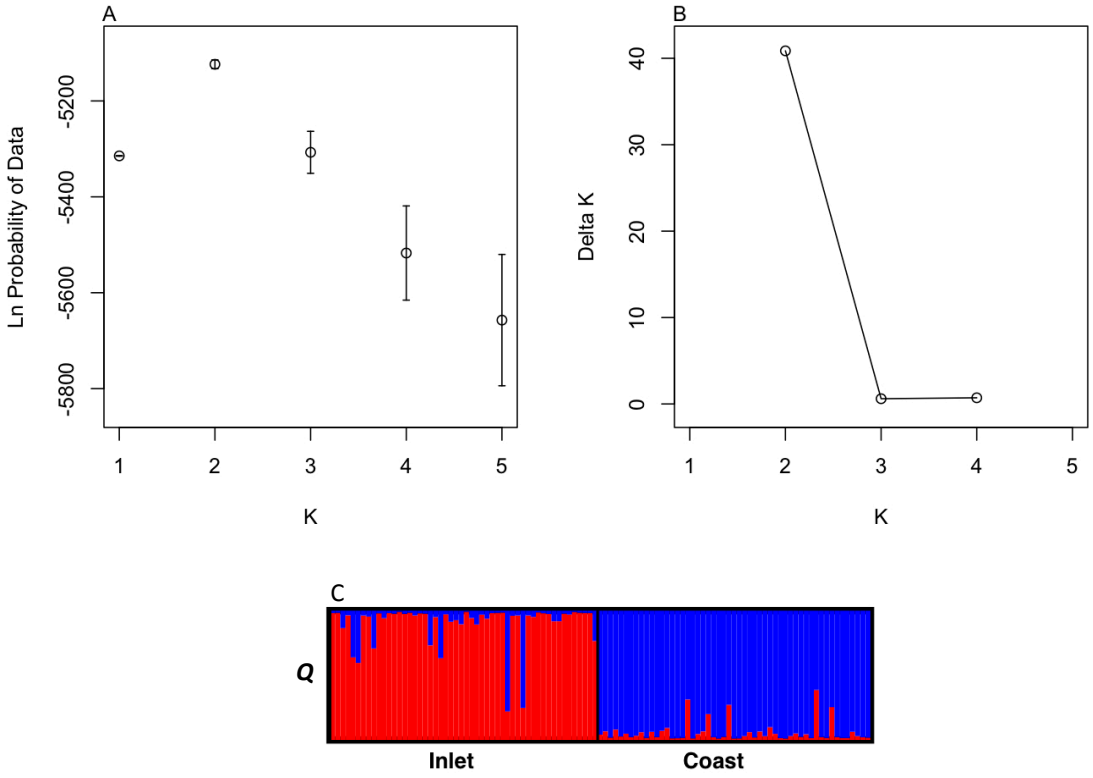


Figure 11. Results of STRUCTURE simulations testing values of K between 1 and 5 for Quatsino Sound coast and inlet samples of copper rockfish (*Sebastes caurinus*). A) Mean Ln probability of K (± SD) calculated from 10 replicate runs for each value of K. B) Second order rate of change of mean Ln probability of K (Delta K) calculated for successive values of K using the method of Evanno (2005). C) STRUCTURE output for K = 2 averaged over 10 runs. Each fish's genotype is represented by a thin vertical line, the blue and red portions of which represent the proportional contribution of one or two genetic groups to the admixture coefficient, *Q* (Pritchard *et al.* 2000).



REFERENCES

Ahlstrom, E.H. 1959. Vertical distribution of pelagic fish eggs and larvae off California and Baja California. *U.S. Fish and Wildlife Service Fishery Bulletin* 60:106-146.

Andrews, A.H., G.M. Cailliet, K.H. Coale, K.M. Munk, M.M. Mahoney and V.M. O'Connell. 2002. Radiometric age validation of the yelloweye rockfish (*Sebastes ruberrimus*) from southeastern Alaska. *Marine and Freshwater Research* 53:139-146.

Andrews, A., L.A. Kerr, G.M. Cailliet, T.A. Brown, C.C. Lundstrom and R.D. Stanley. 2007. Age validation of canary rockfish (*Sebastes pinniger*) using two independent otolith techniques: lead-radium and bomb radiocarbon dating. *Marine and Freshwater Research* 58:531-541.

Antao, T., A. Lopes, R.J. Lopes, A. Beja-Pereira and G. Luikart. 2008. LOSITAN: a workbench to detect molecular adaptation based on a F_{ST}-outlier method. *BMC Bioinformatics* 9:323.

Avise, J.C. 1992. Molecular population structure and biogeographic history of a regional fauna: a case history with lessons for conservation biology. *Oikos* 63:62-76.

Banks, S.C., M.P. Piggot, J.E. Williamson, U. Bové, N.J. Holbrook and L.B. Beheregaray. 2007. Oceanic variability and coastal topography shape genetic structure in a long-dispersing sea urchin. *Ecology* 88:3055-3064.

Beamish, R.J. and G.A. MacFarlane. 1983. The forgotten requirement for age validation in fisheries biology. *Transactions of the American Fisheries Society* 112:735-743.

Beaumont, M.A. and D.J. Balding. 2004. Identifying adaptive genetic divergence among populations from genome scans. *Molecular Ecology* 13(4):969-980.

Beaumont, M.A. and R.A. Nichols. 1996. Evaluating loci for use in the genetic analysis of population structure. *Proceedings of the Royal Society B* 363:1619-1626.

Benzie, J.A.H. and S.T. Williams. 1997. Genetic structure of giant clam (*Tridacna maxima*) populations in the West Pacific is not consistent with dispersal by present-day ocean currents. *Evolution* 51:768-783.

Berkeley, S.A., M.A. Hixon, R.J. Larson and M.S. Love. 2004. Fisheries sustainability via protection of age structure and spatial distribution of fish populations. *Fisheries* 29(8):23-32.

Bernardi, G., L. Findley and A. Rocha-Olivares. 2003. Vicariance and dispersal across Baja California in disjunct marine fish populations. *Evolution* 57:1599-1609.

Bertalanffy, von L. 1938. A quantitative theory of organic growth (Inquiries on growth laws II). *Human Biology* 10:181-213.

Brookfield, J.F.Y. 1996. A simple new method for estimating null allele frequency from heterozygote deficiency. *Molecular Ecology* 5(3):453-455.

Boehlert, G.W., D.M. Gadomski and B.C. Mundy. 1985. Vertical distribution of icthyoplankton off the Oregon coast in spring and summer months. *Fishery Bulletin* 83(4):611-621.

Boehlert, G.W. and R.F. Kappenman. 1980. Variation of growth with latitude in two species of rockfish (*Sebastes pinniger* and *S. diploproa*) from the Northeast Pacific Ocean. *Marine Ecology Progress Series* 3:1-10.

Botsford, L.W., F. Micheli and A. Hastings. 2003. Principles for the design of marine reserves. *Ecological Applications* 13:S25-S31.

Buonaccorsi, V.P., C.A. Carol, E.A. Lynn and R.D. Vetter. 2002. Population structure of copper rockfish (*Sebastes caurinus*) reflects postglacial colonization and contemporary patterns of larval dispersal. *Canadian Journal of Fisheries and Aquatic Sciences* 59:1374-1384.

Buonaccorsi, V.P., C.A. Kimbrell, E.A. Lynn and R.D. Vetter. 2005. Limited realized dispersal and introgressive hybridization influence genetic structure and conservation genetic strategies for brown rockfish, *Sebastes auriculatus*. Conservation Genetics 6:697-713.

Buonaccorsi, V.P., M. Westerman, J. Stannard, C.A. Kimbrell, E.A. Lynn and R.D Vetter. 2004. Molecular genetic structure suggests limited larval dispersal in grass rockfish, *Sebastes trelliger*. *Marine Biology* 145:779-788.

Carvalho, G.R. and L. Hauser. 1994. Molecular genetics and the stock concept in fisheries. *Reviews in Fish Biology and Fisheries* 4:326-350.

Chen, Y., D.A. Jackson and H.H. Harvey. 1992. A comparison of von Bertalanffy and polynomial functions in modeling fish growth data. *Canadian Journal of Fisheries and Aquatic Sciences* 49:1228-1235.

COSEWIC. 2002. COSEWIC assessment and status report on the Bocaccio *Sebastes paucispinis* in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. vii + 43 pp.

COSEWIC. 2007a. COSEWIC assessment and status report on the Canary Rockfish *Sebastes pinniger* in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. vii + 71 pp.

COSEWIC. 2007b. COSEWIC assessment and status report on the Rougheye Rockfish *Sebastes* sp. type I and *Sebastes* sp. type II in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. viii + 36 pp.

COSEWIC. 2008. COSEWIC assessment and status report on the Yelloweye Rockfish *Sebastes ruberrimus*, Pacific Ocean inside waters population and Pacific Ocean outside waters population, in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. vii + 75 pp.

COSEWIC. 2009a. COSEWIC assessment and status report on the Darkblotched Rockfish *Sebastes crameri* in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. vii + 48 pp.

COSEWIC. 2009b. COSEWIC assessment and status report on the Quillback Rockfish *Sebastes maliger* in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. vii + 71 pp.

COSEWIC. 2010. COSEWIC assessment and status report on the Yellowmouth Rockfish *Sebastes reedi* in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. vii + 57 pp.

Cowen, R.K., K.M.M. Lwiza, S. Sponagule and C.B. Paris. 2000. Connectivity of marine populations: open or closed? *Science* 287:857-859.

Cowen, R.K. and S. Sponagule. 2009. Larval dispersal and marine population connectivity. *Annual Review of Marine Science* 1:443–466.

Crawford, W., D. Johannessen, F. Whitney, R. Birch, K. Borg, D. Fissel, and S. Vagle. 2007. Appendix C: Physical and chemical oceanography. *In* Ecosystem overview: Pacific North Coast Integrated Management Area (PNCIMA). Edited by Lucas, B.G., S. Verrin, and R. Brown. Canadian Technical Report of Fisheries and Aquatic Sciences 2667: vii + 77 p.

Cunningham, K.M., M.F. Canino, I.B. Spies and L. Hauser. 2009. Genetic isolation by distance and localized fjord population structure in Pacific cod (*Gadus macrocephalus*): limited effective dispersal in the northeastern Pacific Ocean. *Canadian Journal of Fisheries and Aquatic Sciences* 66:153-166.

Drinkwater, K.F. and T.R. Osborn. 1975. The role of tidal mixing in Rupert and Holberg Inlets, Vancouver Island. *Limnology and Oceanography* 20(4):518-529.

Evanno, G., S. Regnaut and J. Goudet. 2005. Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. *Molecular Ecology* 14:2611-2620.

Excoffier, L.G., G. Laval and S. Schneider. 2005. Arlequin version 3.0: An integrated software package for population genetics data analysis. *Evolutionary Bioinformatics Online* 1:47-50.

Farmer, O.M. and H.J Freeland. 1983. The physical oceanography of fjords. *Progress in Oceanography* 12:147-220.

Fisheries and Oceans Canada (DFO). 2007. Rockfish Conservation Areas: Protecting British Columbia's Rockfish. Fisheries and Oceans Canada, Pacific Region. v + 174 pp.

Foll, M. and O. Gaggiotti. 2008. A genome-scan method to identify selected loci appropriate for both dominant and codominant markers: a Bayesian perspective. *Genetics* 180:977-993.

Fontaine, M., S.J.E. Baird, S. Piry, N. Ray, K.A. Tolley, S. Duke, A. Bikrun, M. Ferreira, T. Jauniaux, Á. Llavona, B. Öztürk, A.A. Öztürk, V. Ridoux, E. Rogan, M. Sequeira, U. Siebert, G.A. Vikingsson, J-M. Bouquegneau and J.R. Michaux. 2010. Rise of oceanographic barriers in continuous populations of a cetacean: the genetic structure of harbour porpoises in Old World waters. BMC Biology 5(30). doi:10.1186/1741-7007-5-30.

Gaines, S.D., C. White, M.H. Carr and S.R. Palumbi. 2010. Designing marine reserve networks for both conservation and fisheries management. *Proceedings of the National Academy of Sciences of the USA*, 107:18286–18293.

Galindo, H.M., D.B. Olson and S.R. Palumbi. 2006. Seascape genetics: A coupled oceanographic-genetic model predicts population structure of Caribbean corals. *Current Biology* 16:1622–1626.

Girever, M. and J. Stien. 1998. Population genetic substructure in blue whiting based on allozyme data. *Journal of Fish Biology* 52:782-795.

Gomez-Uchida, D., E.A. Hoffman, W.R. Arden and M.A. Banks. 2003. Microsatellite markers for the heavily exploited canary (*Sebastes pinniger*) and other rockfish species. *Molecular Ecology Notes* 3:387-389.

Goudet, J. 2002. FSTAT, a program to estimate and test gene diversities and fixation indices (version 2.9.3.2).

Gunderson, D.R. and R. D. Vetter. 2006. Temperate rocky reef fishes. *In*: Marine Metapopulations (eds Sale P.F. and J.P. Kritzer). Academic Press, San Diego, California.

Haldorson, L. and M. Love. 1991. Maturity and fecundity in rockfishes, *Sebastes* spp., a review. *Marine Fisheries Review* 53(2):25-31.

Hedgecock D., P.H. Barber and S. Edmands. 2007. Genetic approaches to measuring connectivity. *Oceanography* 20:70-79.

Hellberg M.E., R.S. Burton, J.E. Neigel and S.R. Palumbi. 2002. Genetic assessment of connectivity among marine populations. *Bulletin of Marine Science* 70:273–290.

Holderegger, R. and H.H Wagner. 2010. A brief guide to landscape genetics. *Landscape Ecology* 21:793-796.

Jacobs, D.K., T.A. Haney and K.D. Louie. 2004. Genes, diversity and the geologic process on the Pacific coast. *Annual Review of Earth and Planetary Science* 32:601–652.

Johanessonn, K. and K. André. 2006. Life on the margin: genetic isolation and diversity loss in a peripheral marine ecosystem, the Baltic Sea. *Molecular Ecology* 15:2013-2029.

Johansson, M.L., M.A. Banks, K.D. Glunt, H.M. Hassel-Finnegan and V.P. Buonaccorsi. 2008. Influence of habitat discontinuity, geographical distance, and oceanography on fine-scale population genetic structure of copper rockfish (*Sebastes caurinus*). *Molecular Ecology* 17:3051-3061.

Johns, G.C. and J.C. Avise. 1998. Tests for ancient species flocks based on molecular phylogenetic appraisals of *Sebastes* rockfishes and other marine fishes. *Evolution* 52:1135-1146.

Jørstad, K.E. and G. Nævdal. 1989. Genetic variation and population structure of cod, *Gadus morhua* L., in some fjords in northern Norway. *Journal of Fish Biology* 35(Supplement A):245-252.

Katsanevakis, S. 2006. Modeling fish growth: model selection, multi-model inference and model selection uncertainty. *Fisheries Research* 81:229-235.

Kerr, L.A., A.H. Andrews, K. Munk, K.H. Coale, B.R. Frantz, G.M. Cailliet and T.A. Brown. 2005. Age validation of quillback rockfish (*Sebastes maliger*) using bomb radiocarbon. *Fishery Bulletin* 103:97-107.

Kimura, D.K. 1980. Likelihood methods for the von Bertalanffy growth curve. *Fishery Bulletin* 77(4):765-776.

Laidig, T.E., D.E. Pearson and L.L. Sinclair. 2003. Age and growth of blue rockfish (*Sebastes mystinus*) from central and northern California. *Fishery Bulletin* 101:800-808.

Landaeta, M. F. and L.R. Castro. 2006. Larval distribution and growth of the rockfish, *Sebastes capensis* (Sebastidae, Pisces), in the fjords of southern Chile. *ICES Journal of Marine Science* 63:714-724.

Landaeta, M.F., G. Lopez, N. Suarez-Donoso, C.A. Bustos and F. Balbontin. 2012. Larval fish distribution, growth and feeding in Patagonian fjords: potential effects of freshwater discharge. *Environmental Biology of Fishes* 93:73-87.

Lea, R.N. R.D. McAllister and D.A. Ventresca. 1999. Biological aspects of the nearshore rockfishes of the genus *Sebastes* from central California. *California Department of Fish and Game Fish Bulletin* 177:1-109.

Leberg, P.L. 1992. Effects of population bottlenecks on genetic diversity as measured by allozymes electrophoresis. *Evolution* 46:477-494.

Lenarz, W.H., R.J. Larson and S. Ralston. 1991. Depth distributions of late larvae and pelagic juveniles of some fishes of the California Current. *CalCOFI Report* 32:41-46.

Love, M.S., M.H. Carr and L.J. Haldorson. 1991. The ecology of substrate-associated juveniles of the genus *Sebastes*. *Environmental Biology of Fishes* 30:225-243.

Love M.S., M. Yoklavich and L.K. Thorsteinson. 2002. *The rockfishes of the Northeast Pacific.* University of California Press, Berkeley and Los Angeles, CA. 414 pp.

MacLellan, S.E. 1997. How to age rockfish (*Sebastes*) using *S. alutus* as an example – the otolith burnt section technique. *Canadian Technical Report of Fisheries and Aquatic Sciences* 2146:1-42.

Maier-Reimer, E., U. Mikolajewicz and T. Crowley. 1990. Ocean general circulation model sensitivity experiment with an open Central American isthmus. *Paleoceanography* 5:349–366.

Manel, S., M.K. Schwartz, G. Luikart and P. Taberlet. 2003. Landscape genetics: combining landscape ecology and population genetics. *Trends in Ecology and Evolution* 18:189-197.

Manel, S. and R. Holeregger. 2013. Ten years of landscape genetics. *Trends in Ecology and Evolution*. In press.

Markel, R.W. 2011. Rockfish recruitment and trophic dynamics on the west coast of Vancouver Island: fishing, ocean climate and sea otters. Ph.D. Thesis. University of British Columbia, Vancouver, B.C.

Marlieve, J.B. 1986. Lack of planktonic dispersal of rocky intertidal fish larvae. *Transactions of the American Fisheries Society* 115(1):149-154.

Marshall, D.J., K. Monro, M. Bode, M.J. Keough and S. Swearer. 2010. Phenotype-environment mismatches reduce connectivity in the sea. *Ecology Letters* 13:128-140.

Matthews, K.R. 1990. A telemetric study of the home ranges and homing routes of copper and quillback rockfishes on shallow rocky reefs. *Canadian Journal of Zoology* 68:2243-2250.

McCusker, M.R. and P. Bentzen. 2010. Historical influences dominate the population genetic structure of a sedentary marine fish, Atlantic wolfish (*Anarhichas lupus*), across the North Atlantic Ocean. *Molecular Ecology*. 19:4228-4241.

Miller, J.A. and K. Shanks. 2004. Evidence for limited larval dispersal in black rockfish (*Sebastes melanops*): implications for population structure and marine-reserve design. *Canadian Journal of Fisheries and Aquatic Sciences* 61:1723–1735.

Mitamura, H., N. Arai, W. Sakamoto, Y. Mitsunaga, H. Tanaka, Y. Mukai, K. Nakamura, M. Sasaki and Y. Yoneda. 2005. Role of olfaction and vision in homing behaviour of black rockfish *Sebastes inermi*. *Journal of Experimental Marine Biology and Ecology* 322:124-134.

Moser, H.G. and G.W. Boehlert. 1991. Ecology of pelagic larvae and juveniles of the genus *Sebastes*. *Environmental Biology of Fishes*: 30:203-224.

Nei, M. 1987. *Molecular Evolutionary Genetics.* University of Columbia Press. New York, USA. 513 pp.

Nielsen, E.E., M.M. Hansen and D. Meldrup. 2006. Evidence of microsatellite hitch-hiking election in Atlantic cod (*Gadus morhua* L.): implications for inferring population structure in nonmodel organisms. *Molecular Ecology* 15:3219–3229.

Nielsen, E.E., J. Hemmer-Hansen, P.F. Larsen and D Bekkevold. 2009. Population genomics of marine fishes: identifying adaptive variation in space and time. *Molecular Ecology* 18:3128-3150.

Nuwer, J.M. and R.G. Keil. 2005. Sedimentary organic matter geochemistry of Clayoquot Sound, Vancouver Island, British Columbia. *Limnology and Oceanography* 50(4):1119-1128.

Palumbi, S.R. 2003. Population genetics, demographic connectivity, and the design of marine reserves. *Ecological Applications* 13(1):S146-S158.

Parker, S.J., S.A. Berkeley, J.T. Golden, D.R. Gunderson, J. Heifetz, M.A. Hixon, R. Larson, B.M. Leaman, M.S. Love, J.A. Musick, V.M. O'Connell, S. Ralston, H.J. Weeks and M.M. Yaklovich. 2000. Management of Pacific rockfish. *Fisheries* 25(3):22-30.

Perrin, C., S.R. Wing and M.S. Roy. 2004. Effects of hydrograchic barriers on population genetic structure of the sea star *Coscinasterias muricata* (Echinodermata, Asteroidea) in the New Zealand fiords. *Marine Ecology* 13(8):2183-2195.

Pickard, G.L. 1963. Oceanographic characteristics of inlets of Vancouver Island, British Columbia. *Journal of the Fisheries Research Board of Canada* 20(5):1109-1144.

Planes S., P.J. Doherty and G. Bernardi. 2001. Strong genetic divergence among populations of a marine fish with limited dispersal: *Acanthochromis polyacanthus*, within the Great Barrier Reef and the Coral Sea. *Evolution* 55:2263–2273.

Pritchard, J.K., M. Stephens and P. Donnelly. 2000. Inference of population structure using multilocus genotype data. *Genetics* 155:945-959.

Raymond, M. and Rousset, F. 1995. GENEPOP (version 1.2): population genetics software fro exact tests and ecumenicism. *Journal of Heredity* 86:248-249.

Rice, W.R. 1989. Analyzing tables of statistical tests. *Evolution* 43(1):223-225.

Richards, L.J. 1987. Copper rockfish (*Sebastes caurinus*) and quillback rockfish (*Sebastes maliger*) habitat in the Strait of Georgia, British Columbia. *Canadian Journal of Zoology* 65:3188-3191.

Ricker, W.E. 1975. Computation and interpretation of biological statistics of fish populations. *Bulletin of the Fisheries Research Board of Canada* 191:1-382.

Ridgway, K.R. and J.R. Dunn. 2003. Mesoscale structure of the mean East Australian Current system and its relationship with topography. *Progress in Oceanography* 56:189–222.

Rocha-Olivares, A. and R.D. Vetter. 1999. Effects of oceanographic circulation on the gene flow, genetic structure, and phylogeography of the rosethorn rockfish (*Sebastes helvomaculatus*). *Canadian Journal of Fisheries and Aquatic Sciences* 56:803–813.

Rousset, F. 2008. Genepop'007: a complete reimplementation of the Genepop software for Windows and Linux. *Molecular Ecology Resources* 8:103-106.

Schmidt, P.S., E.A. Serrão, G.A. Pearson, C. Riginos, P.D. Rawson, T.J. Hilbish, S.H. Brawley, G.C. Trussell, E. Carrington, D.S. Wethey, J.W. Grahame, F. Bonhomme and D.M. Rand. 2008. Ecological genetics in the North Atlantic: environmental gradients and adaptation at specific loci. *Ecology* 89:S91-S107.

Schnute, J. 1981. A versatile growth model with statistically stable parameters. *Canadian Journal of Fisheries and Aquatic Sciences* 38:1128-1140.

Schultz, J.K., J.A. Feldheim, S.H. Gruber, M.V. Ashley, T.M. McGovern and B.W. Bowen. 2008. Global phylogeography and seascape genetics of the lemon shark (Genus *Negaprion*). *Molecular Ecology* 17:5336–5348.

Selkoe, K.A., C.M. Henzler and S.D. Gaines. 2008. Seascape genetics and the spatial ecology of marine populations. *Fish and Fisheries* 9:363-377.

Selkoe, K.A. and R.J. Toonen. 2006. Microsatellites for ecologists: a practical guide to using and evaluating microsatellite markers. *Ecology Letters* 9(5):615-629.

Shanks, A.L., B.A. Grantham and M.H. Carr. 2003. Propagule dispersal distance and the size and spacing of marine reserves. *Ecological Applications* 13(1):S159-S169.

Siegle, M.R., E.B. Taylor, K.M. Miller, R.E. Withler and K.L. Yamanaka. 2013. Subtle population genetic structure in yelloweye rockfish (*Sebastes ruberrimus*) is consistent with a major oceanographic division in British Columbia, Canada. PLoS ONE 8(8): e71083. doi:10.1371/journal.pone.0071083.

Sinclair, M. and T.D. Iles. 1989. Population regulation and speciation in the oceans. *Journal Conseil International pour l'Exploration de la Mer* 45:165-175.

Sivasundar, R. and S.R. Palumbi. 2010. Life history, ecology and the biogeography of strong genetic breaks among 15 species of Pacific rockfish, *Sebastes*. *Marine Biology* 157:1433-1452.

Stephenson, R.L. 1999. Stock complexity in fisheries management: a perspective of emerging issues related to population sub-units. *Fisheries Research*: 247-249.

Stepien, C.A. 1999. Phylogeographical structure of the Dover sole *Microstomus pacificus*: the larval retention hypothesis and genetic divergence along the deep continental slope of the northeastern Pacific Ocean. *Molecular Ecology* 8:923–939.

Sokal, R.R. and F.J. Rohlf. 2012. Biometry: the principles and practice of statistics in biological research. 4th edition. W.H. Freeman and Co., New York. 937 pp.

Suneetha, K.B. and G. Nævdal. 2001. Genetic and morphological stock structure of the pearlside, *Maurolicus muelleri* (Pisces, Sternoptychidae), among Norwegian fjords and offshore area. *Sarsia* 86:191-201.

Thomson, R.E. 1981. Oceanography of the British Columbia coast. *Canadian Special Publications of Fisheries and Aquatic Sciences* 56:1-291.

Tolimieri, N., K. Andrews, G. Williams, S. Katz and P.S. Levin. 2009. Home range size and patterns of space use by lingcod, copper rockfish and quillback rockfish in relation to diel and tidal cycles. *Marine Ecology Progress Series* 380:229-243.

Van Oosterhout, C., W.F. Hutchinson, D.P.M. Wills and P. Shipley. 2004. MICRO-CHECKER: software for identifying and correcting genotyping errors in microsatellite data. *Molecular Ecology Notes* 4:535-538.

Waples, R.S. 2005. Genetic estimates of contemporary effective population size: to what time periods do the estimates apply? *Molecular Ecology* 14:3335-3352.

Waples, R.S and C. Do. 2008. LDNE: a program for estimating effective population size from data on linkage disequilibrium. *Molecular Ecology Notes* 8:753-756.

Weatherley, A.H. and H.S. Gill. 1987. The biology of fish growth. Academic Press, London. 443 pp.

Weir, B.S. and C.C. Cockerham. 1984. Estimating F-statistics for the analysis of population structure. *Evolution* 38(6):1358-1370.

Westerman, M.E., V.P. Buonaccorsi, J.A. Stannard, L. Galver, C. Taylor, E.A. Lynn, C.A. Kimbrell and R.D. Vetter. 2005. Cloning and characterization of novel microsatellite DNA markers for the grass rockfish, *Sebastes rastrelliger,* and cross-species amplification in 10 related *Sebastes* spp. *Molecular Ecology Notes* 5:74-76.

White, C., K.A. Selkoe, J. Watson, D.A. Siegel, D.C. Zacherl and R.J. Toonen. 2010. Ocean currents help explain population genetic structure. *Proceedings of the Royal Society of London Series B* 277:1685–1694.

Wimberger, P., J. Burr, A. Gray, A. Lopez and P. Bentzen. 1999. Isolation and characterization of twelve microsatellite loci for rockfish (*Sebastes*). *Marine Biotechnology* 1:311-315.

Wright, S. 1938. Size of population and breeding structure in relation to evolution. *Science* 87(2263):430-431.

Yamanaka, K.L. and G. Logan. 2010. Developing British Columbia's inshore rockfish conservation strategy. *Marine and Coastal Fisheries: Dynamics, Management and Ecosystem* 2010:28-46.

APPENDIX A

Table A1. Microsatellite loci multiplexed for polymerase chain reaction (PCR) amplification. All forward primers were labeled (5') with black, green or blue fluorescent dyes. Thermocycling conditions (temperature, time) are given for each multiplex: initial denaturation (*I.D.*), denaturation (*D.*), annealing (*A.*), elongation (*E.*), number of cycles (*# Cyc.*), and final extension (*F.E.*). Primer sequences are given in Wimberger *et. al.* 1999 (*Sma*); Gomez-Uchida *et al.* 2003 (*Spi*) and Westerman *et al.* 2005 (*Sra*).

Multiplex #	***Loci***	***PCR Thermocycling Conditions***					
		I.D.	***D.***	***A.***	***E.***	***# Cyc.***	***F.E.***
1	*Sra*5-9, *Sra*7-7, *Sra*7-25, *Sma*3, *Sma*5	94°C 2min	94°C 1min	56°C 30sec	72°C 1min	30	72°C 10min
2	*Spi*6, *Sra*11-103, *Spi*10, *Spi*4	94°C 2min	94°C 1min	57°C 30sec	72°C 1min	30	72°C 10min
3	*Sma*2, *Sma*10, *Sma*11	94°C 2min	94°C 1min	57°C 30sec	72°C 1min	30	72°C 10min
4	*Spi*12, *Sma*4	94°C 2min	94°C 1min	56°C 30sec	72°C 1min	30	72°C 10min
5	*Spi*18, *Sra*15-8, *Sra*16-5	94°C 2min	94°C 1min	52°C 30sec	72°C 1min	30	72°C 10min