American Genetic Association

Journal of Heredity, 2022, 121–144
https://doi.org/10.1093/jhered/esab069
Invited Review
Advance Access publication March 19, 2022

OXFORD

Invited Review

Implications of Large-Effect Loci for Conservation: A Review and Case Study with Pacific Salmon

Robin S. Waples, Michael J. Ford, Krista Nichols, Marty Kardos, Jim Myers, Tasha Q. Thompson, Eric C. Anderson, Ilana J. Koch, Garrett McKinney, Michael R. Miller, Kerry Naish, Shawn R. Narum, Kathleen G. O'Malley, Devon E. Pearse, George R. Pess, Thomas P. Quinn, Todd R. Seamons, Adrian Spidle, Kenneth I. Warheit, and Stuart C. Willis

From the Northwest Fisheries Science Center, National Marine Fisheries Service, 2725 Montlake Blvd. East, Seattle, WA 98112, USA (Waples, Ford, Nichols, Kardos, Myers, McKinney, and Pess); the University of Montana, Missoula, MT, USA (Kardos); the Michigan State University, East Lansing, MI, USA (Thompson); the Southwest Fisheries Science Center, National Marine Fisheries Service, Santa Cruz, CA, USA (Anderson and Pearse); the Columbia River Inter-Tribal Fish Commission, Hagerman, ID, USA (Koch, Narum, and Willis); the Washington Department of Fish and Wildlife, Olympia, WA, USA (McKinney, Seamons, and Warheit); the University of California, Davis, CA, USA (Miller); the School of Aquatic and Fishery Sciences, University of Washington, Seattle, WA USA (Naish and Quinn); the Oregon State University, Newport, OR, USA (O'Malley); and the Northwest Indian Fisheries Commission, Olympia, WA, USA (Spidle).

Address correspondence to R. S. Waples at the address above, or e-mail: robin.waples@noaa.gov.

Received June 1, 2021; First decision September 17, 2021; Accepted November 7, 2021.

Corresponding Editor: C Scott Baker

Abstract

The increasing feasibility of assembling large genomic datasets for non-model species presents both opportunities and challenges for applied conservation and management. A popular theme in recent studies is the search for large-effect loci that explain substantial portions of phenotypic variance for a key trait(s). If such loci can be linked to adaptations, 2 important questions arise: 1) Should information from these loci be used to reconfigure conservation units (CUs), even if this conflicts with overall patterns of genetic differentiation? 2) How should this information be used in viability assessments of populations and larger CUs? In this review, we address these questions in the context of recent studies of Chinook salmon and steelhead (anadromous form of rainbow trout) that show strong associations between adult migration timing and specific alleles in one small genomic region. Based on the polygenic paradigm (most traits are controlled by many genes of small effect) and genetic data available at the time showing that early-migrating populations are most closely related to nearby late-migrating populations, adult migration differences in Pacific salmon and steelhead were considered to reflect diversity within CUs rather than separate CUs. Recent data, however, suggest that specific alleles are required for early migration, and that these alleles are lost in populations where conditions do not support early-migrating phenotypes. Contrasting determinations under the US Endangered Species Act and the State of California's equivalent legislation illustrate the complexities of incorporating genomics data into CU configuration decisions. Regardless how CUs are defined, viability assessments

should consider that 1) early-migrating phenotypes experience disproportionate risks across large geographic areas, so it becomes important to identify early-migrating populations that can serve as reliable sources for these valuable genetic resources; and 2) genetic architecture, especially the existence of large-effect loci, can affect evolutionary potential and adaptability.

Keywords: conservation, genetic architecture, Pacific salmon, phenology, steelhead

I. Introduction

Efforts to conserve biodiversity at the species or population level often wrestle with 3 complex questions: Q1: What are the most appropriate units of conservation (aka conservation units, CUs)? Q2: What is the status of each unit? Q3: What conservation measures will best ensure persistence of these units into the future? These questions are generally addressed in sequence but they are not independent, and uncertainty in one propagates into the others. This review focuses on recent advances in genome science that have challenged previous approaches to Q1, as well as the consequences for Q2 and Q3.

The 20th century witnessed increasing need and interest in protecting threatened and endangered species, and recent decades have seen a greater appreciation of the role that populations and other subspecific units play in species persistence (Rojas 1992; Houlahan et al. 2000; Ceballos and Ehrlich 2002; Gustafson et al. 2007). Defining CUs within species is generally a 2-step process: 1) Describe the (often hierarchical) biological relationships among populations; and 2) Choose an appropriate hierarchical level for defining separate units (Waples 1995). The first step is to characterize the continuum from individuals to species, and the attributes that define the intermediate levels. In theory, this process is objective, but in reality it is subject to continual discussion, refinement, and debate (Sanderson and Shaffer 2002; Lotterhos and Whitlock 2014; Shirk et al. 2017). On the second issue—where should the threshold level for distinctiveness be—there is no single "correct" answer; instead, the best approach for any particular application depends on several factors, including goals, legal mandates, and available resources (Waples 1995, 2006). Even after considering these factors, it is inevitable that some reasonable people will be inclined to combine units that others might view as separate.

In these ongoing discussions, considerable interest has centered on the concept of evolutionarily significant units (ESUs), which represent major components of diversity within species (Ryder 1986; Waples 1991; Moritz 1994; Crandall et al. 2000; Fraser and Bernatchez 2001). The most widely used ESU frameworks can be mapped onto 2 axes of diversity: isolation and adaptation (Figure 1). The isolation axis reflects the strength and duration of reproductive isolation, typically quantified using data from putatively neutral genetic markers. The adaptation axis is more difficult to evaluate directly, and for this reason conservation practitioners have typically relied on proxies, such as phenotypic traits that might reflect adaptations (e.g., morphology, behavior, life history) or ecological features of the species' habitat, which might create selective pressure for adaptations to local environments. These proxies have important limitations, however. Phenotypic traits are influenced to varying degrees by both genes and the environment; inferring selective pressures from habitat features is fraught with difficulties; and common-garden experiments that might document adaptations are difficult or impossible to conduct for most species of conservation interest.

Following major technological advances around 2007, the explosive increase in availability and affordability of genomic data for non-model species (Primmer 2009; Allendorf et al. 2010; Narum et al. 2013; Supple and Shapiro 2018) has raised a new possibility: Is it now feasible to use genomics data to help parameterize the adaptation axis, without relying entirely on proxies (Funk et al. 2012, 2019; Pearse 2016; Waples et al. 2020)? If so, how might this affect division of species into CUs? And do new genomics data suggest different weightings for the isolation and adaptation axes (Ralls et al. 2020; Fernandez-Fournier et al. 2021)? Here, we explore these and related questions, using a case study involving 2 species of anadromous Pacific salmonids (*Oncorhynchus* spp.) for which a strong association has recently been found between a key life-history trait (timing of adult migration to fresh water) and a small genomic region on one chromosome. This topic has attracted a great deal of general attention because of its broad relevance to conservation and management (Langin 2018; Waples and Lindley 2018; McKinney 2020).

This review is organized as follows. For context, in Section II, we review key features of evolutionary ecology of Pacific salmonids that are relevant to themes discussed later, and we explain the legal framework that allows for federal protection of these species under the US Endangered Species Act (ESA). Section III reviews theory and empirical data for large-effect loci in other species. Recent genomics studies that have reported strong associations between adult migration timing and specific alleles are reviewed in Section IV. Finally, in Section V (Discussion), we 1) summarize major conclusions; 2) identify critical uncertainties; 3) discuss implications for conservation and management; and 4) identify research priorities for the future.

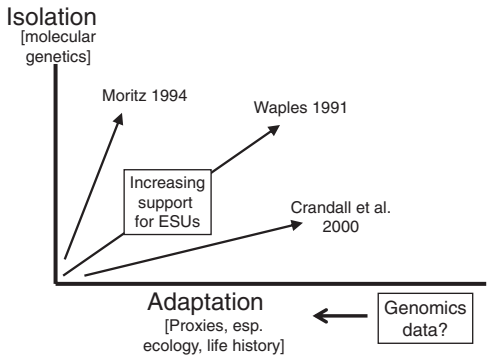

Figure 1. Two major axes of diversity that characterize the most widely used ESU concepts. Moritz's (1994) method based on reciprocal monophyly of mtDNA places almost all emphasis on the isolation axis. Crandall et al.'s (2000) method based on exchangeability places more emphasis on adaptations. The NMFS salmon ESU framework (Waples 1991) places relatively equal weight on the 2 axes. Isolation is most easily documented using molecular genetic methods, whereas inferences about adaptations have traditionally had to rely on proxies like ecology and life history. The advent of new genomics tools raises the possibility that it will be possible to study adaptations directly at the DNA level. Modified from Waples and Lindley (2018).

Box 1. Salmonid Life History

The term salmonid fishes, as used here, refers primarily to the genera *Salmo*, *Oncorhynchus*, and *Salvelinus*, native to the Northern Hemisphere. These species show exceptional levels of within-species life-history diversity (Figure 1.1). All spawn in freshwater (stream and lake) habitats, where females select and prepare a nest site, bury their eggs after fertilization, and in some species guard them as well. Eggs are unusually large for freshwater fishes, and have a long incubation period, emerging as free-living fish in the spring or early summer. Depending on species and population, they migrate immediately to sea; feed and grow for some weeks, months, or years in fresh water before migrating to sea; or remain in freshwater habitats (with either limited or extensive movements) before returning to spawn at or near their natal site years later. Anadromous and nonanadromous populations can be genetically distinct from one another, even when breeding in the same river, but some or considerable interbreeding often occurs between life-history variants; genetic and environmental controls over anadromy are complicated, and variable. Finally, most species are capable of breeding in more than 1 year (i.e., are iteroparous), but the Pacific salmon (a subset of the *Oncorhynchus* species) all die after spawning.

Salmonids differ markedly from most migratory animals in several important ways related to the evolution of migratory timing. Many species such as birds, butterflies, and whales make regular seasonal migrations between breeding and feeding grounds at different latitudes (e.g., north in the spring and south in the fall in the northern hemisphere). In intervening seasons the animals experience a changing photoperiod that helps synchronize their circannual rhythms, as well as the common ecological pressures that caused migration to evolve. In contrast, salmon migrate into rivers to spawn at every month of the year, commencing during increasing or decreasing photoperiod while at sea. Moreover, their distributions at sea are very broad, such that some individuals will migrate southward to get to the mouth of their natal stream, while others are simultaneously migrating northward, to reach the same river mouth at the same time, or a completely different time, of year.

In addition to the complexities introduced by the diversity in timing and direction of migration from the ocean to the freshwater habitats for breeding, spatial and temporal connections between entry into freshwater and actual breeding are equally diverse. Some populations enter a river and spawn within a few km of salt water only a few days later, so timing of river entry and breeding are

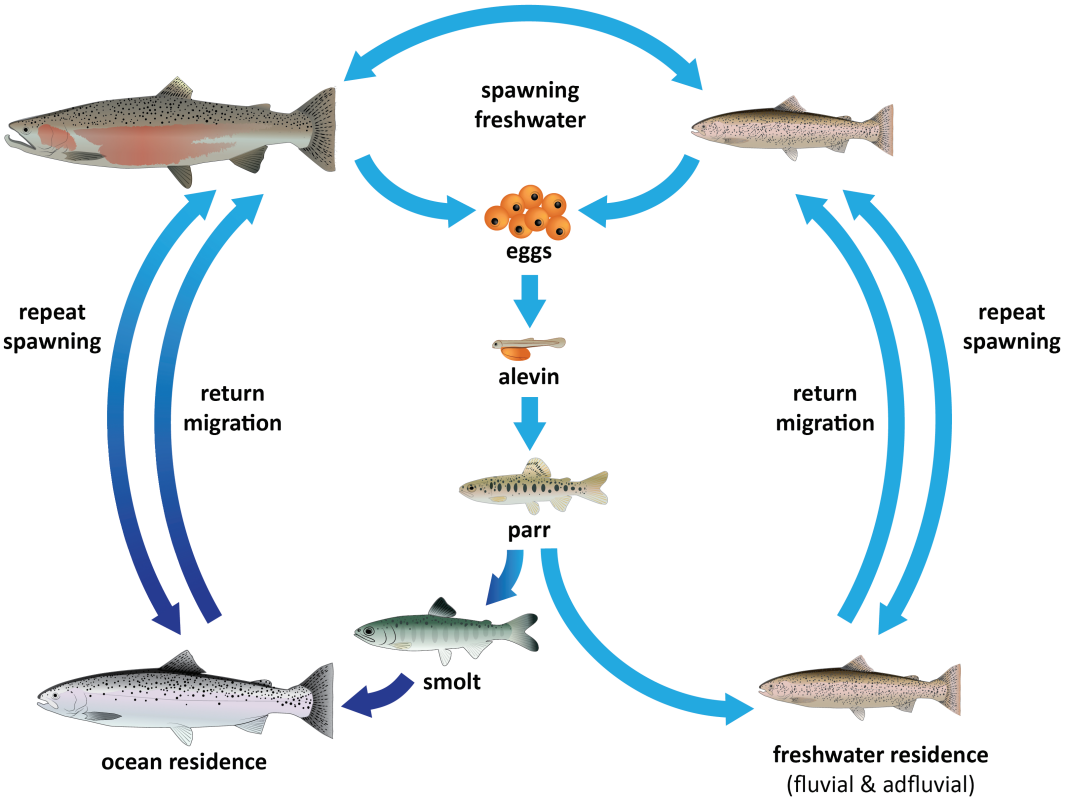

Figure 1.1. Common life-history patterns within *Oncorhynchus mykiss*. Adults that go to sea as juveniles (anadromous steelhead) and those that mature in fresh water (resident rainbow trout) both spawn and lay eggs in freshwater habitats. Juveniles (alevin, fry [not shown], and parr) grow in fresh water until they either undergo a physiological transformation and migrate to sea as smolts, or remain in fresh water until they mature. Unlike true Pacific salmon, *O. mykiss* is iteroparous so adults can spawn in multiple years, although repeat spawning by steelhead is uncommon. Return migration timing by steelhead can occur in any month of the year, generally categorized as summer (April–September) or winter (October–March), but spawning consistently occurs in the spring. The life cycle of Chinook salmon (Oncorhynchus *tshawytscha*) is simpler in 2 major ways: 1) all Chinook salmon die after spawning; 2) apart from some rare exceptions (introduced populations in the US Great Lakes; precocious males that mature as parr and never migrate to sea), Chinook salmon are always anadromous.

essentially the same, in which case sexual maturation was largely completed while they were at sea. In other cases, populations breeding within a large watershed might enter many months apart but breed at overlapping times (e.g., enter in spring and fall and breed in fall, or enter in summer and winter and breed in winter and spring). In their natal rivers, these life-history types would thus experience entirely different patterns of photoperiod and river temperatures prior to breeding. Within a single river system, it is not uncommon for some individuals to spend 6–8 months in fresh water prior to breeding, and thus undergo sexual maturation in an entirely different osmotic environment than other individuals that largely completed maturation at sea but nevertheless spawn nearby at the same time of year.

II. Background

II.1 Speciation of Pacific Salmonids

Lineages leading to Pacific (*Oncorhynchus* spp.) and Atlantic salmonids (*Salmo* spp.) diverged an estimated 15–27 MYA (Behnke 1992; Stearley 1992; Devlin 1993; Montgomery 2000; Macqueen and Johnston 2014). By the late Miocene (6–8 MYA), the extant species of North American Pacific salmon and trout had appeared (Smith 1992; Wilson and Turner 2009), leaving a great deal of time for diversification within species. Of the anadromous Pacific salmonids, intraspecific genetic and life-history differentiation is greatest in sockeye salmon (*Oncorhynchus nerka*), Chinook salmon (Oncorhynchus *tshawytscha*), and steelhead (the anadromous form of rainbow trout, Oncorhynchus *mykiss*) (Waples et al. 2001; Quinn 2018; Box 1). Here, we focus on associations between genomic variation and timing of spawning migrations from the ocean into rivers in the latter 2 species, as examples of the broader concept of the application of phenotypic and genomic data to conservation problems.

II.2 Life-History Diversity

Compared to their relatives that live exclusively in fresh water, anadromous salmonids undergo a complex series of transitions to successfully complete their life cycle (Figure 1.1 in Box 1). Our understanding of their life cycle and migrations has grown from observations by early naturalists, including patterns that were widely recognized by indigenous peoples of the region (Swezey and Heizer 1977), to experimental demonstrations of the heritability of phenological traits (related to seasonal timing), and recent identification of specific alleles correlated with adult run timing. From all these perspectives, the timing of return from the ocean to breed defines salmon in meaningful ways. Most salmon life-history traits have at least a partial genetic basis, and traits related to phenology generally have the highest heritabilities (median 0.51; Carlson and Seamons 2008). Adult migration timing (departure from the ocean to initiate upriver migration prior to spawning) is particularly important because it transitions fish from the sea (where they must balance opportunities for growth against risks of predation) to fresh water (where they have to contend with anorexia and energetic demands of migration and spawning). A genetic influence on adult migration timing was initially demonstrated by Rich and Holmes (1928); formal heritability studies of within-run adult migration/spawn timing include Quinn et al. (2000, 2011) for Chinook salmon and Abadia-Cardoso et al. (2013) for steelhead.

In western North America, both Chinook salmon and steelhead return to fresh water in every month of the year (Figure 2), but the range of return times within a given drainage is shorter and often multimodal. No consensus has been reached in the scientific literature regarding the most appropriate terminology to describe adult migration timing (see Healey 1983 and Quinn et al. 2016 for prominent examples). Here, we use the terms "early" or "early-migrating" to refer to spring-returning Chinook salmon and summer-returning steelhead and "late" or "late-migrating" to refer to fall-returning Chinook salmon and winter-returning steelhead. Early-migrating fish enter rivers many months prior to spawning, whereas late-migrating fish enter shortly before spawning. The "early" and "late" terms are simplistic, given that adult migration timing occurs along a continuum, but they capture major modes in migration timing.

Early-migrating forms exist in many anadromous fishes, but the phenomenon is especially common in salmonids (Hearsey and Kinziger 2014; Quinn et al. 2016). Early-returning Chinook salmon and steelhead are prized by fishers for their flesh quality and high fat content, and spring Chinook salmon are of special cultural significance to Native American tribes, who value them as the "First Fish" to return each year (Lord 1866, Powers 1877). The early-migration strategy entails several costs, including foregone opportunity for growth in the ocean, and exposure to predation, pathogens, and extreme temperature and flow regimes in freshwater habitats where they hold while fasting prior to spawning (Quinn et al. 2016). Because the early-migration strategy is nevertheless widespread, it must generate benefits to offset these costs. Several potential benefits have been identified, but for Pacific salmonids it is generally thought that the crucial factor is access to specific spawning and rearing habitats that temperature/flow conditions make unavailable (or less available) to later-migrating fish (Quinn et al. 2016).

Diverse entry times allow anadromous salmonids to more fully exploit suitable habitats for spawning and rearing (Beechie et al. 2008; Quinn et al. 2016). Environmental conditions that differ significantly between headwater and lower mainstem spawning habitats can limit gene flow between fish spawning in separate areas, particularly where spawning habitat is discontinuous. Gene flow is also limited by flow-dependent barriers to migration (Withler 1966; Smith 1969). Prior to anthropogenic modifications, rivers such as the Klickitat in Washington, Willamette in Oregon, and Salmon in California had waterfalls or cascades that prevented upstream migration, except during high flows that are common in winter and spring (Fulton 1968; Howell et al. 1985; Olson and Dix 1993), whereas other cascades or falls are passable only during low flows that are common in the summer and early fall (WDF et al. 1993; Kostow 2012). Low flows on the southern Oregon-California coast in summer/fall result in barrier sandbars at river entrances that block migration and can disrupt timing patterns (Shapovalov and Taft 1954, Nicholas and Hankin 1988). These barrier bars are a selective effect across a large geographic region, but waterfalls are more localized and depend on specific geologic features (Myers et al. 2006).

Temperature also plays an important role in diversifying adult migration timing. If suitable habitat is continuously distributed, spawning generally occurs earliest in the cooler upper reaches of a given river (Hard et al. 1996; Doctor and Quinn 2009), although exceptions do occur (Olsen et al. 2008). However, many rivers have temporal modes of migration and spawning, which typically are spatially segregated to reflect thermal conditions experienced by spawning adults and developing embryos. These thermal patterns tend to reduce gene flow between early and late components of populations.

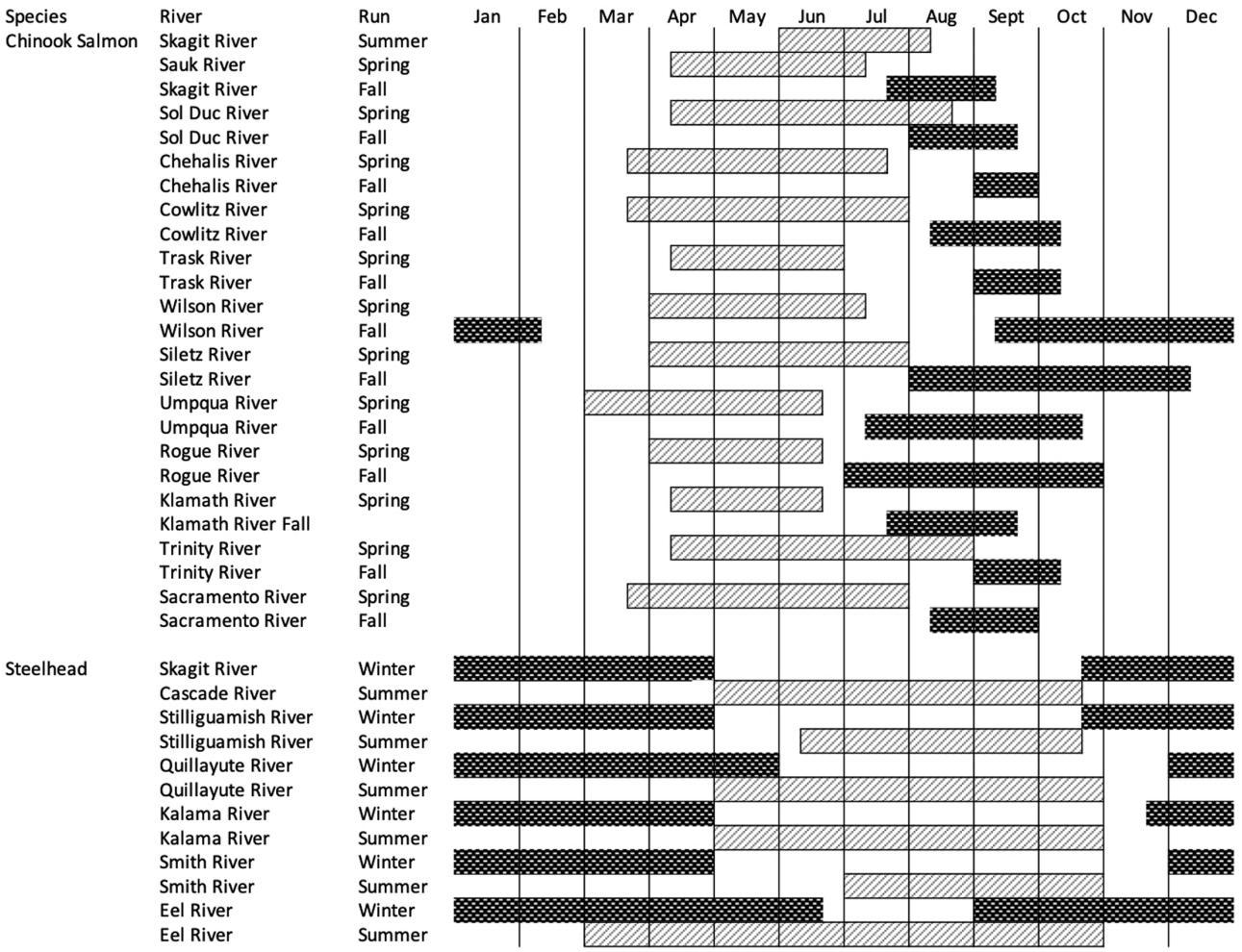

Figure 2. Freshwater entry times for different migration times of returning adult Chinook salmon and steelhead in Puget Sound, Washington Coast, Oregon Coast, and California rivers. Within each species, rivers are arranged from North to South. Light shaded bars are early-migrating populations, dark shading is for late-migrating populations. Data are from Myers et al. (1998) and Busby et al. (1996).

Managers have long used adult migration timing, combined with other kinds of information, to define management units for salmon and steelhead, regulate harvest, and operate hatchery programs (Kostow 2009). These management units are generally considered to be demographically independent, meaning that their dynamics are determined more by local births and deaths than by immigration (McElhany et al. 2000; Box 2). As part of this review, we consider whether genomics data provide new insights regarding demographic independence.

II.3 Population-Genetic Diversification

By 1950, it was widely recognized in the scientific community that homing to natal sites for spawning—and the resulting population differentiation—were essential features of salmon biology and management (e.g., Moulton 1939; Thompson 1959), but a detailed understanding of population structure had to await development of suitable genetic markers. Population-genetic studies in Chinook salmon and steelhead now span 6 decades and cover a succession of technologies, including allozymes (e.g., Utter et al. 1973), microsatellites (e.g., Beacham et al. 2006), single nucleotide polymorphisms (SNPs; e.g., Narum et al. 2008; Hecht et al. 2015) and whole-genome sequences (e.g., Narum et al. 2018; Thompson et al. 2020).

A series of large-scale studies of North American Chinook salmon have included both early- and late-returning fish from the same set of watersheds (Table 1). In coastal drainages and in the lower Columbia River, the following pattern has consistently been found: different life-history types of Chinook salmon and steelhead within the same stream are genetically more similar to each other than either is to the same life-history type in another stream (Figure 3). This pattern, which is consistent with repeated divergence of the more specialized early-migrating forms from the more generalized later-migrating forms, is limited to coastal rivers and was initially described with allozymes and subsequently confirmed with microsatellites (Moran et al. 2013) and SNPs (Narum et al. 2008; Hecht et al. 2015; Arciniega et al. 2016; Prince et al. 2017). A different pattern is seen in the interior Columbia and Snake river basins, where early- and late-returning Chinook salmon are strongly diverged genetically (F_{ST} ~0.1 or higher), while relatively modest differences are found among geographically separated populations of the same run type (Waples et al. 2004; Moran et al. 2013).

II.4 Anthropogenic Influences

Native Americans have harvested anadromous Pacific salmonids for at least the last 10 000 years (Chatters et al. 1995; Campbell and

Box 2. Demographic Independence

It is becoming increasingly clear that most problems in conservation biology are not strictly ecological or evolutionary, but rather are eco-evolutionary in nature. The concept of a population is central to many aspects of applied conservation and management, so it is important that populations are defined in a way that reflects both ecological and evolutionary processes. At a minimum, individuals within a population must co-occur in space and time for at least part of their life cycle, so that they can interact ecologically (competition, predation, etc.) and interbreed. To be biologically meaningful, populations also should have somewhat limited interactions with other conspecific groups of individuals—but how strong must isolation be? A criterion often applied in this context is "demographic independence"—roughly speaking, different units are considered demographically independent if population trajectories are driven more by local births and deaths than by immigration/emigration. But what level of migration is consistent with demographic independence? Considering the importance of this fundamental question, it has received remarkably little rigorous evaluation. The most comprehensive analysis still appears to be a nearly 3-decades-old study by Hastings (1993), which suggested that migration rates above about 10% generally produce correlated trajectories in 2 population units. Ten percent migration is very high in genetic terms and leads to very low levels of neutral genetic differentiation unless effective population size is low enough that genetic drift is strong (Figure 2.1). Furthermore, because of the nonlinear relationship between F_{ST} and N_e, statistical power here is very asymmetrical: It is easy to demonstrate demographic independence if you get a moderate or large F_{ST}, but with a small F_{ST} it will generally be very difficult to determine from genetic data alone which side of the threshold you are on.

In salmon conservation and management, the concept of demographic independence has been used to identify salmon and steelhead populations for which it is meaningful to conduct separate viability analyses (McElhany et al. 2000). These evaluations have used a variety of criteria in addition to genetics, including population size, natural barriers to migration, distance between watersheds, and ecological characteristics of natal watersheds. Even in coastal drainages where genetic differences between early- and late-run populations are typically modest (F_{ST} of a few percent or less), spring and fall Chinook salmon (and summer and winter steelhead) have generally been considered to reflect closely related but demographically independent populations. Furthermore, another major uncertainty is whether the transition from demographic dependence to independence is gradual or sharp (Figure 2.2). In conjunction with the limited statistical power of genetic data alone to evaluate demographic independence, these uncertainties make it challenging to determine whether early- and late-migrating types within a given river basin should be considered separate populations, or simply life-history diversity within a single population.

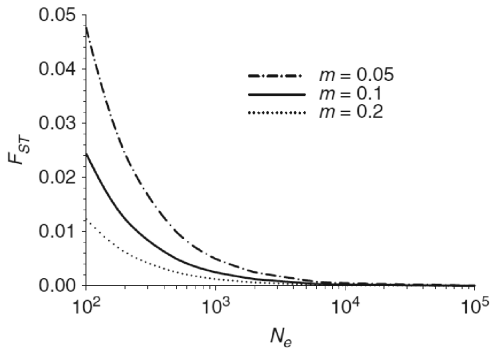

Figure 2.1. Relationship between F_{ST} (a measure of genetic differentiation) and effective population size (N_e), based on Wright's (1943) island model of migration. F_{ST} depends on the product of migration rate (m) and N_e, and expected results are plotted for 3 migration rates that are equal to a generally accepted threshold for demographic independence (m = 0.1; solid line) and twice or half as large as the presumed threshold. Any given F_{ST} value is consistent with a range of migration rates, and (unless N_e is small) rates that are and are not consistent with demographic independence lead to F_{ST} values that are very small and hard to distinguish. Reproduced from Waples et al. (2008).

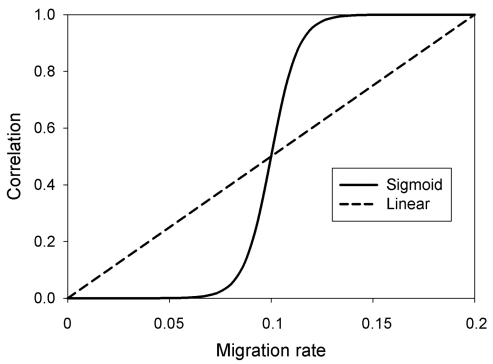

Figure 2.2. Two hypothetical relationships between migration rate (fraction of individuals exchanged per time unit, m) and the strength of the correlation between trajectories of 2 interacting populations. Even if m = 0.1 is a useful general guideline for assessing demographic independence (as suggested by results of Hastings 1993), the shape of the relationship has important implications for practical application, as well as for the feasibility of distinguishing between levels of connectivity that are and are not consistent with demographic independence.

Butler 2010; Morin et al. 2021), but anthropogenic effects on salmon ecosystems greatly accelerated following major European settlement and land development in the last 2 centuries (Waples et al. 2009). Status assessments typically focus on anthropogenic stressors such as habitat loss and modification, blockage of migratory routes, harvest, and artificial propagation (Myers et al. 1998; McClure et al. 2003), all of which can affect salmon life histories and population resilience (NRC 1996) and can interact with effects of climate change (Irvine and Fukuwaka 2011; Crozier et al. 2019; Tillotson et al. 2021). Several types of anthropogenic modifications to salmonid ecosystems can influence adult migration timing (Tillotson et al. 2021). Blockage of specific up-stream breeding habitats due to migration barriers (e.g., dams) can lead to the eradication of early-migrating adult salmonids (Beechie et al. 2006; McClure et al. 2008; Pess et al. 2014). This may be a key reason why in both Chinook salmon and steelhead, early-migrating populations have been extirpated at

Table 1. Population genetic studies of Chinook salmon that encompass multiple populations of early and late returning fish throughout much of the species' North American range

Study	Number and type of loci	Comments
Myers et al. (1998)	31 allozymes	
Waples et al. (2004)	32 allozymes	Largely the same samples as Myers et al. (1998)
Beacham et al. (2006)	13 microsatellites	Some samples likely the same as earlier studies
Seeb et al. (2007)	13 microsatellites	
O'Malley et al. (2008)	13 microsatellites, 2 circadian rhythm genes	
Narum et al. (2008)	13 microsatellites, 37 SNPs	Same samples as Seeb et al. (2007)
Moran et al. (2013)	13 microsatellites	Same samples as Seeb et al. (2007)
Clemento et al. (2014)	96 SNPs	Same samples as Seeb et al. (2007)
Hecht et al. (2015)	19 703 SNPs	
Davis et al. (2017)	21 microsatellites and 96 SNPs	
Prince et al. (2017)	215 354 SNPs (posterior prob >.8 in >50% inds)	

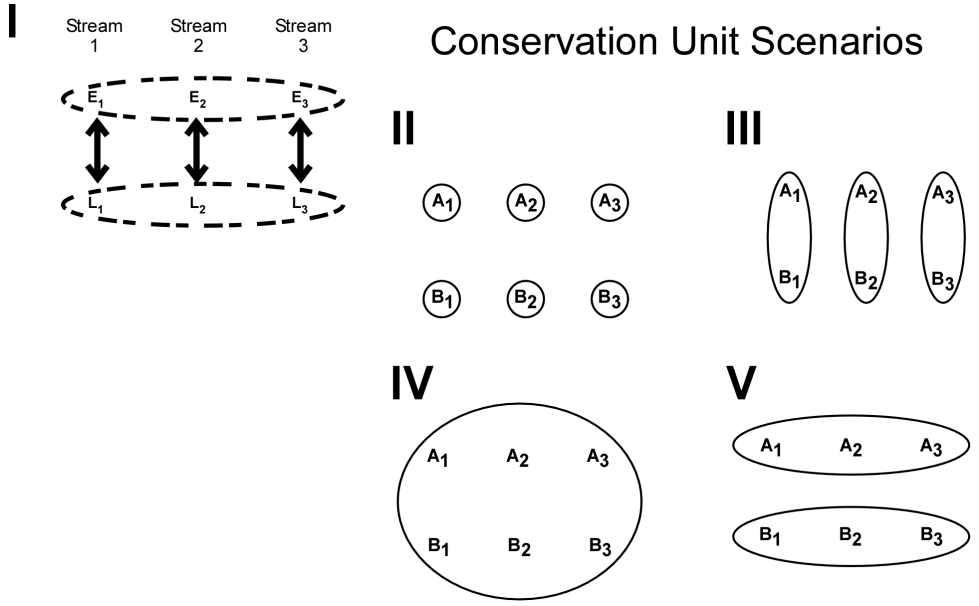

Figure 3. Schematic representation of some possible ways of configuring conservation units (Panels II–V), given patterns of genetic relationship depicted in Panel I. In Panel I, 2 ecotypes (E, L) occur in each of 3 locations (streams, in this example). Overall patterns of genetic similarity (bold arrows) reflect geography rather than ecotype, but at one large-effect locus specific alleles are strongly associated with specific ecotypes (ellipses). Depending on whether one is inclined to be a lumper or a splitter, a CU concept based on evolutionary lineages could produce units as depicted in Panels II, III, or IV. The scenario in Panel V would be consistent with a lineage-based approach only if the large-effect gene were weighted more heavily than all other genes combined, but this scenario could be consistent with approaches that use other frameworks to define CUs. Modified from Ford et al. (2020).

higher rates than late-migrating populations (Gustafson et al. 2007). Where flow-dependent and/or thermal migration barriers have been removed or their temporal passage windows altered (e.g., falls are laddered or cascades altered), early-migrating populations can be displaced by their late-migrating counterparts (Hemstrom et al. 2018; Thompson et al. 2019a). Flow regulation by dams also can affect adult migration timing. Changes to downstream flow and temperature conditions are thought to have selected against spring-run Chinook salmon in the Rogue River by allowing for expansion of the fall-run Chinook salmon distribution into habitat that was previously accessible primarily by spring-run (Thompson et al. 2019a). Hatchery operations can artificially impose selection on timing (Tillotson et al. 2021), and propagation of populations with different migration timings from the same facility has resulted in interbreeding and changes in migration timing (Kinziger et al. 2008; Hess et al. 2011; Fraser et al. 2020). In some cases at least, removal of these anthropogenic impacts can also alter adult migration timing and "reawaken" historical life-history types (Quinn et al. 2017), provided the underlying genetic diversity still persists. For instance, summer steelhead that migrate prior to winter steelhead and spawn in an entirely different location have re-emerged recently in the Elwha River following removal of 2 dams (Fraik et al. 2021).

II.5 Salmonids under the US ESA

The 1990s was a decade of reckoning for Pacific salmonids in the Pacific Northwest and California. A key report (Nehlsen et al. 1991) documented over 200 at-risk wild stocks, and Native American tribes and conservation groups filed petitions for legal protection under the US ESA. Although the species in their entirety were not threatened with extinction, the ESA allows for protection of Distinct Population Segments of vertebrate species, and this DPS provision has been used to protect US populations of iconic species like grizzly bears, wolves, and bald eagles that are more abundant elsewhere (Scott et al. 2006). During the 1990s, the US National Marine Fisheries Service (NMFS)—the agency primarily

responsible for implementing the ESA for marine and anadromous species—conducted a series of status reviews of North American anadromous Pacific salmonids. Collectively, these status reviews identified over 50 salmon and steelhead DPSs, of which about half are now protected under the ESA (https://www.fisheries.noaa.gov/species-directory/threatened-endangered). A retrospective analysis estimated that, of all the US populations of salmon outside Alaska that existed prior to major European contact, about one-third had been extirpated, another third were considered Threatened or Endangered under the ESA, and the final third were not known to be at risk (Gustafson et al. 2007).

Salmon and steelhead DPS evaluations were conducted using both prongs of the ESU concept depicted in Figure 1. According to a policy developed by NMFS (NMFS 1991; Waples 1991), a group of salmon populations is considered an ESU and hence a DPS if it satisfies both of 2 criteria: 1) substantial reproductive isolation, and 2) substantial contribution to the evolutionary legacy of the species. In this context, "reproductive isolation" refers to restricted gene flow, and "evolutionary legacy" is both backward looking (the product of past evolutionary events) and forward looking (raw material for future evolution). Thus, salmon DPSs are major evolutionary components of the species as a whole. Together with data on straying and inferences based on geography, population-genetic data were used to evaluate the degree of reproductive isolation among populations. Evolutionary legacy was evaluated using the adaptation axis; the most important proxies used were ecological features of the habitat (e.g., freshwater migration distance; water flow and temperature; main source of precipitation—rain or snow) and life-history traits (esp. timing of juvenile and adult migrations; age and size at maturity; ocean distribution patterns). Canada has adopted a similar framework for identifying "designatable units" (DUs) of Canadian species and is in the process of evaluating the status of Pacific salmon DUs for potential listing under their Species at Risk Act (COSEWIC 2018, 2019).

Most salmon and steelhead DPSs cover relatively large spawning and rearing areas (e.g., Oregon Coast; Puget Sound; Snake River; Figure 4) and include up to several dozen demographically independent populations, which nevertheless share common ecological, phenotypic, and genetic characteristics (Weitkamp et al. 1995; Busby et al. 1996; Myers et al. 1998). Adult migration timing was one of the most important life-history traits considered in making DPS/ESU determinations. Based on the genetic and life-history data described above, it was concluded that adult migration differences had evolved independently many times within both Chinook salmon and steelhead (Thorgaard 1983; Busby et al. 1996; Myers et al. 1998; Waples et al. 2004). Furthermore, it was thought that the early-migration phenotype could evolve from late-migrators on relatively short timescales (perhaps around 100 years; Waples et al. 2004). Therefore, in defining ESUs of coastal and lower Columbia River populations in both species, it was concluded that adult migration differences reflected diversity within ESUs (as illustrated in Figure 3).

These ESU configurations are consistent with the conventional paradigm that most quantitative genetic traits are due to many genes of small effect (Falconer and Mackay 1996; Mackay et al. 2009). Many studies support this paradigm; for example, height in humans is associated with many thousands of SNPs spread throughout the genome (Wood et al. 2014). However, recent findings—in both steelhead and Chinook salmon—of strong associations between specific alleles and adult migration timing pose the following questions:

- Should existing conservation units be reconfigured in response to these new data?

Figure 4. Distribution and size of Steelhead ESUs in the western USA. as defined by Busby et al. 1996.

- Should methods for viability assessment be modified (and if so, how)?
- Should new/different conservation measures be implemented to ensure persistence of important components of biodiversity?

In the following sections, we first review the empirical details and then return to consideration of these questions.

III. Review of Theory and Empirical Data for Large-Effect Loci

III.1. A Quantitative-Genetics View of Trait Variation

Most phenotypic traits, including salmon life-history traits, are influenced by both genetic and environmental variation. Quantitative genetics is the theoretical foundation for describing the genetic component of variation in such traits within a population (e.g., the

heritability of the trait), as well as predicting responses to selection (Falconer and Mackay 1996; Walsh and Lynch 2018).

The adaptive phenotypic response (R) to a single generation of selection is predicted by the breeder's equation $R = h^2S$, where h^2 is the narrow sense heritability (Falconer and Mackay 1996). h^2 is the fraction of phenotypic variance in a population (V_P) that is due to additive genetic effects (V_A—the component of genetic variation that is directly inherited). The selection differential S is the difference between the population mean phenotype and the mean phenotype among parents of the next generation. Traditional quantitative genetics approaches assume that V_A within a population arises from additive genetic effects at many loci, each with a small phenotypic effect (the polygenic or "infinitesimal" model of inheritance; Barton et al. 2017). This model predicts that adaptation results in extremely small changes in allele frequency at each locus, and little or no loss in h^2 over time. The maintenance of h^2 for many traits in domesticated species despite many generations of selection (Hill 2008; Havenstein et al. 2003; Dudley and Lambert 2003) suggests that the polygenic model is often a useful approximation.

It is important to consider outcomes of selection when assumptions underlying the polygenic model are not met (Lande 1983; Chevalet 1994; Hospital and Chevalet 1996). In some cases, genetic variation underlying a quantitative trait might be explained by one or more loci with large effects, combined with many small-effect loci (Lande 1983; Schielzeth et al. 2012; Walsh and Lynch 2018; see Table 2). When large-effect loci are involved, response to multiple generations of selection is expected to deviate from predictions of the polygenic model because evolution caused by changes in large effect allele frequencies causes temporal changes in V_A (Walsh and Lynch 2018). V_A, h^2, and the rate of adaptive phenotypic change can increase substantially as a large-effect allele approaches intermediate frequency and decrease when frequency approaches 1 or 0 (Figure 5). Fixation or loss of a large-effect allele can erase much of the initial V_A, leaving only the small-effect component to respond to future selection.

The genetic architecture of a trait describes the number of loci involved, the variants at those loci, and their component effects on phenotypic variation. Effects of genetic architecture on evolutionary responses to selection can translate into effects on population dynamics when selection is sustained over multiple generations (Kardos and Luikart 2021). For example, holding initial h^2 constant while varying the underlying genetic architecture shows that evolutionary potential and population viability are often higher when the selected trait is explained by many loci of small effect than when large-effect loci are involved (Kardos and Luikart 2021). Therefore, an improved understanding of the relative contribution of different loci to the genetic architecture underlying fitness-related traits has important implications for our understanding of adaptation and for the conservation of natural populations.

Widespread availability of genomic data has led to new insights into the genetic basis of phenotypic variation and adaptation in non-model organisms. The genetic architecture of a trait is frequently characterized by detecting associations between specific loci and variable traits through pedigree-based analyses (of quantitative trait loci, QTLs; Schielzeth and Husby 2014) or population surveys (genome-wide association studies, GWAS; Visscher et al. 2012). Combining information from GWAS with genomic relatedness among individuals can provide a more complete understanding of how loci across the genome contribute to trait variation (Sella and Barton 2019). Termed "genomic selection" or "genomic prediction," implementation of these approaches in agricultural taxa for genetic improvement has evolved into sophisticated methods for predicting how both small- and large-effect loci contribute to complex traits and to phenotypic variation (Meuwissen et al. 2001). These approaches show great promise for understanding contributions of small- and large-effect loci in nature (e.g., Ashraf et al. 2020). In genomic-based studies on natural populations, there has been some emphasis to date on detecting molecular signatures of natural selection, and using these to pinpoint genomic regions involved with historical adaptation in natural populations (Zhan et al. 2014; Epstein et al. 2016; Jones et al. 2018). These signatures of selection can be driven by historical selection favoring variants at protein coding loci, or loci related to gene regulation. It is important to recognize that genomic signatures of selection arise from historical selection events (Pritchard et al. 2010; Le Corre and Kramer 2012), and they might not contribute to variation in a trait under current conditions (Pritchard and Di Rienzo 2010; Brieuc et al. 2015; Ashraf et al. 2020).

Describing the genetic architecture underlying conservation-relevant traits is a developing area of research, so current understanding is limited. Large-effect loci are easier to detect and therefore likely to be identified earlier. Detectability is influenced by experimental design, including the number of variable loci surveyed, the additive variance underlying the trait, the evolutionary history of a population, and the number of individuals and frequency of recombination events in the survey population (e.g., Wang and Xu 2019). For example, by using a larger study design, Sinclair-Waters et al. (2020) describe a more complex architecture comprising a mixture of large and small effect loci underlying age at maturity in Atlantic salmon than previously reported (Barson et al. 2015). Effect sizes can be reported either as the proportion of phenotypic (Barson et al. 2015) or additive genetic variance (Johnston et al. 2013) explained, or phenotypic values for specific genotypic classes or vice versa might be given (Barson et al. 2015; Thompson et al. 2019a). Many studies report statistically significant associations between a trait and one or more loci, but not effect sizes. In these cases, the loci might be simply designated as loci of large effect because they were detected in studies with relatively low power. In natural populations, reporting effect size can be challenging because the association analyses themselves might require statistical approaches that make interpretation difficult. For example, Waters et al. (2021) report significant association between a locus *Six6* and age at maturity in 2 Pacific salmon species, but study design and corrections for factors such as population structure meant that a measure of effect size was not reported. Finally, the underlying biology of the genetic architecture itself can influence interpretation (Oomen et al. 2020). For example, large effect loci might represent 2 or more loci that are linked with each other along a continuum of distances along the chromosome, and the strength of statistical association among such loci (linkage disequilibrium) can vary by population. Such variation creates uncertainty in predicting evolutionary change (Oomen et al. 2020).

Albeit challenging, studies that characterize the relative contributions of large-effect versus small-effect loci would improve understanding of evolutionary potential as well as the evolutionary and conservation consequences of changes in large-effect allele frequencies. Changes in the frequency of an allele that underlies a substantial proportion of the additive genetic variance could have important ecological effects, including removing a large fraction of the additive trait variance. Conversely, when the polygenic variance is high, fixation of any specific allele would be less important ecologically, even if the allele was considered to be large-effect. Considering the phenotypic effects of polygenic variation given the varying effect sizes of explanatory loci would improve our understanding of phenotypic

Table 2. Examples of large-effect loci detected in natural populations

Trait	Summary	Citation
Atlantic salmon age at maturity	SNPs in VGLL3 six6 associated with age at maturity in Atlantic salmon. VGLL3 exhibits sex specific dominance. Polygenic variation also present.	Barson et al. (2015); Sinclair-Waters et al. (2020)
Presence of burrows in old field mice	Presence of an escape tunnel is controlled by single locus, length of tunnel is complex trait controlled by at least 3 genomic regions, suggests that complex behaviors (extended phenotypes) can evolve through multiple genetic changes each affecting distinct behavior modules.	Sinclair-Waters et al. (2020)
Stationary vs. migratory Atlantic cod	Migratory life-history influenced by 2 adjacent inversions.	Kirabukaran et al. (2016)
Beak size in Darwin's finches	Beak size in finches is influenced by 2 distinct haplotypes in ~500 kb region of HMGA2 gene.	Lamichhaney et al. (2016a)
Mating strategies in ruffs	Different male mating strategies are influenced by chromosomal inversions.	Lamichhaney et al. (2016b)
Steelhead resident vs. migrant	Resident vs. migrant life-history influenced by double inversion.	Pearse et al. (2019a)
Chinook male age at maturity	Chinook salmon male age at maturity associated with Y-chromosome haplotypes. Presumed to be due to heterochiasmy but could be caused by inversion.	McKinney et al. (2020) McKinney et al. (2021)
Winter coat color in snowshoe hares	Winter coat color in snowshoe hares is influenced by variation in Agouti gene, including ~1 kb insertion in white coat haplotype and 4.3 kb deletion in brown coat haplotype. Brown winter coat haplotype likely arose from introgression with jackrabbit.	Jones et al. (2018)
Coat color in deer mice	Coat color in deer mice is influenced by multiple mutations that form haplotypes in Agouti gene.	Linnen et al. (2013)
Horn morphology in Soay sheep	Horn morphology (normal vs. scurred vs. polled) is influenced by SNPs in the RXFP2 gene. Further fine-mapping is needed to determine causal variant, but gene as a whole explained 76% of variation in horn morphology.	Johnston et al. (2011)
Ecotypic variation in sunflowers	Several haplotypes spanning tens of megabases were associated with traits involved with local adaptation in sunflowers (e.g., seed size and flowering time).	Todesco et al. (2020)

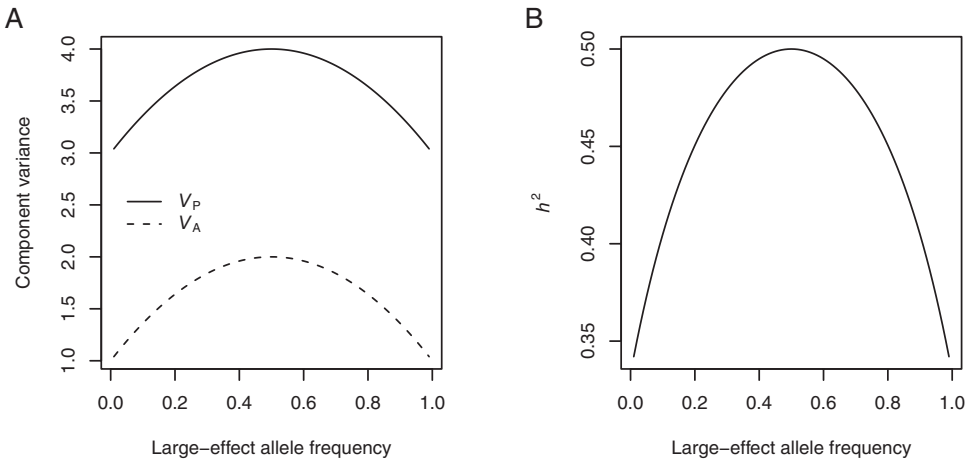

Figure 5. Relationship between large-effect allele frequency (*x* axis) and phenotype variance components (**A**) and heritability (**B**) (*y* axis). In this hypothetical example, the phenotypic variance due to environmental effects is $V_E = 2$ (in arbitrary units), the additive genetic variance contributed by a polygenic (infinitesimal) background is constant at a value of $V_A = 1$, and there is a single large-effect locus with a phenotypic effect of $a = 2$ (again in arbitrary units). V_A, V_P, and h^2 are all maximized when the large-effect allele is at an intermediate frequency.

responses to selection, and the consequences of management actions over time.

IV. Genomic Studies of Adult Migration Timing in Pacific Salmonids

IV.1 Identification of the *GREB1L*/*ROCK1* Region

Several studies have reported that a single genomic region has a strong statistical association with adult migration timing in steelhead (Hess et al. 2016; Prince et al. 2017; Micheletti et al. 2018; Collins et al. 2020; Willis et al. 2020) and Chinook salmon (Prince et al. 2017; Narum et al. 2018; Thompson et al. 2019a, 2019b; Koch and Narum 2020, Thompson et al. 2020; Willis et al. 2021). These studies have identified one approximately 200 Kb region of chromosome 28, centered on the regulatory region between 2 genes called *GREB1L* and *ROCK1* and containing part of the coding region of each gene (Figure 6). The strong association between this region and various measures of adult-migration phenotypes has been found in multiple populations of both species, from coastal California and Oregon, to the interior Columbia River, the Strait of Juan de

Fuca, and Puget Sound (Supplementary Tables S1 and S2). Initial studies (Hess et al. 2016; Prince et al. 2017) were based on relatively sparse (<1%) genome coverage using restriction-site associated DNA sequencing (RAD-seq) markers and identified only a handful of associated markers in the *GREB1L/ROCK1* region. Subsequent studies with more complete genome coverage, either for the whole genome or targeted at the *GREB1L/ROCK1* region (Micheletti et al. 2018; Narum et al. 2018; Thompson et al. 2019a; Thompson et al. 2020) found even stronger statistical associations between genomic markers and adult migration phenotypes. The exact causal genetic variants within this region remain unknown, and the peak of statistical association with adult migration timing spans *GREB1L* and the intergenic region between the *GREB1L* and *ROCK1* genes (Figure 6), suggesting that the causal variant might be regulatory.

IV.2 Strength of Association and Phenotype Complexity

Precisely quantifying the strength of association or the variance explained by variation in the *GREB1L/ROCK1* region is challenging, primarily due to the difficulty of accurate and consistent migration phenotyping. Ideally, a study examining association with adult migration timing would directly measure freshwater entry time (at a river's mouth) across a broad geographic range. Resources necessary to do this are rarely available, so proxies for freshwater entry time (e.g., passage date at a facility farther upstream, carcass recovery date/location) usually are employed. The correlation between such proxies and actual freshwater entry time tends to erode with distance upstream and as the migration season progresses (Thompson et al. 2019a; Willis et al. 2020, 2021), so associations based on such proxies can appear substantially weaker than they would be if timing were assessed at a river's mouth. Associations with other aspects of adult migration are also likely to be affected by phenotyping methodology, but not necessarily in the same way as migration time (e.g., associations with spawning-ground arrival timing of interior Columbia River stocks can become more apparent with distance upstream; Narum et al. 2018).

To date, only a single study has presented a time series of fish sampled upon (or within a few days of) freshwater arrival, and only in a single river system. Thompson et al. (2020) directly measured freshwater entry time by collecting Chinook salmon samples from the Yurok tribal fishery in the estuary of the Klamath River and calculated that *GREB1L/ROCK1* genotypes accounted for 85% of the variance in sample collection date, with nearly complete separation between entry-time distributions of the 2 homozygous genotypes (Figure 7). Analyses of Chinook salmon in the Columbia Basin directly comparing phenotypes of freshwater passage timing at 235 river kilometers (Rkm) versus spawning ground arrival timing found the strongest association of markers in the GREB1L/ROCK1 region with freshwater passage timing for all lineages (Willis et al. 2021). An analysis of Klickitat River steelhead collected at a fish ladder approximately 280 Rkm from the mouth of the Columbia River estimated that *GREB1L/ROCK1* genotypes accounted for 46% of the variance in ladder passage date (Hess et al. 2016). A study in Hood River steelhead collected approximately 235 Rkm from the mouth of the Columbia River found a similar result (Willis et al. 2020), although results varied between coastal and inland lineages elsewhere in the Columbia Basin. Regardless of challenges associated with phenotyping, it is clear that variation in the *GREB1L/ROCK1* region is strongly associated with adult migration timing and explains a large degree of the variance in both species.

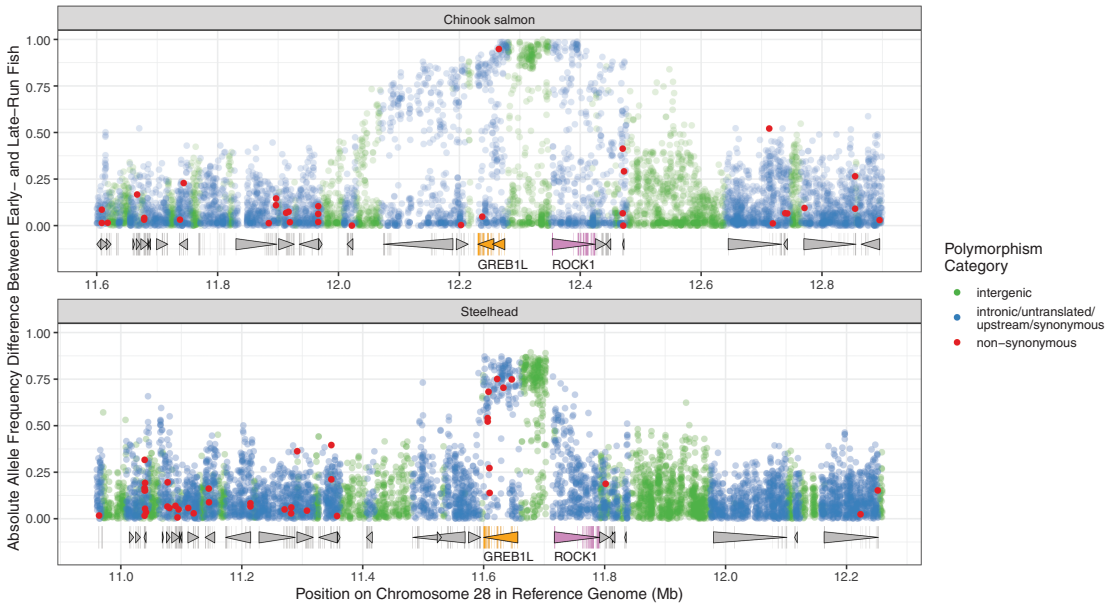

Figure 6. Genomic location of variation associated with adult migration timing within 1.3 Mb surrounding the *GREB1L/ROCK1* region in Chinook salmon and steelhead. The *x* axes show genomic location and the *y* axes show the difference in allele frequency between early- and late-migrating fish. Top panel: difference in allele frequency between 64 spring-run and fall-run Chinook salmon from California (modified from Thompson et al. 2020). Bottom panel: allele frequency differences from pool-Seq between summer and winter steelhead from the Kalama and Klickitat Rivers, Columbia River, Washington (using data from Micheletti et al. 2018). Each point represents a variant, and color indicates the type of genome region each variant occurs in (genic, intergenic, etc.). Bottom track in each panel shows location of genes (triangles = direction of transcription, wide to narrow; vertical whiskers = exons) from each species' genome annotation. *GREB1L* is colored orange, *ROCK1* is colored violet. Note: although by some conventions names of genes appear in lower-case italics, here we use upper-case italics to be consistent with nomenclature in several recent salmonid publications for these genes.

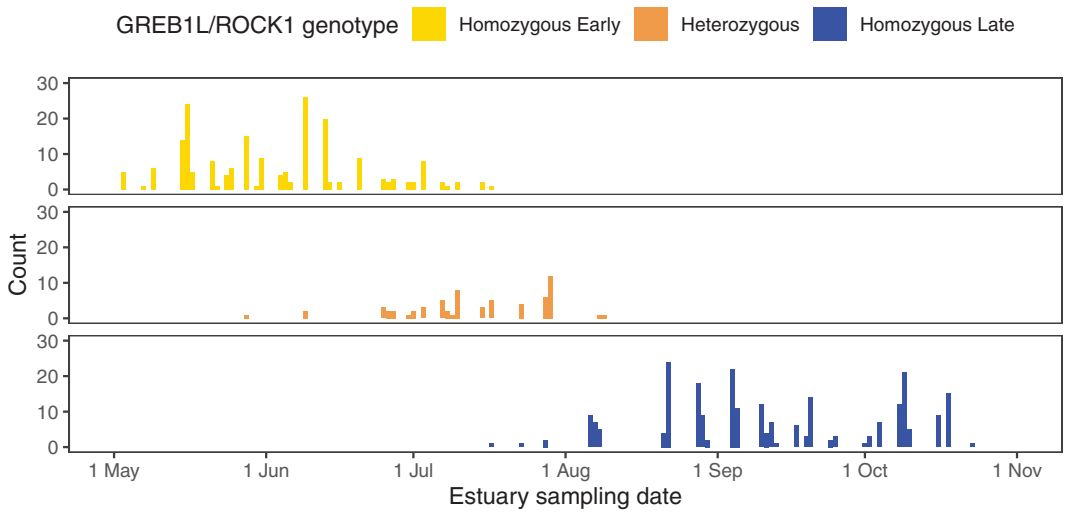

Figure 7. Freshwater entry dates and *GREB1L/ROCK1* region genotype of 502 Klamath River Chinook salmon Modified from Thompson et al. (2020).

The relationship between *GREB1L/ROCK1* and other traits associated with migration time (responses to photoperiod, endocrine controls over osmoregulation and sexual maturity, adiposity, spawn timing, etc.) might be less direct than the relationship with return time, or associated with it in complex ways. In their analysis of Klamath River Chinook, Thompson et al. (2020) measured the gonadosomatic index [GSI: (gonad mass)/(somatic mass)] of fish entering fresh water. They found that, after controlling for differences in sampling date, there was no detectable direct effect of *GREB1L/ROCK1* genotype on GSI. In other words, prior to and at the time of freshwater entry, the sexual maturation process of both spring- and fall-run Chinook salmon in the Klamath River appears to proceed along a single, common trajectory (Figure 7). In that study, *GREB1L/ROCK1* genotype accounted for 67% of the variance in spawn time at the Trinity River Hatchery, but the authors suggest this difference between genotypes might be due to the relatively warm river environment accelerating maturation of early migrators, rather than a direct effect of genotype on spawning time. Thompson et al. (2020) also concluded that, in the Klamath River, differences in adiposity (measured as liver non-water fraction) are also better explained by sampling date than genotype, and that the association between *GREB1L/ROCK1* variation and other traits associated with run type may be indirect and mediated by the direct influence of *GREB1L/ROCK1* variation on migration timing.

Variation in the *GREB1L/ROCK1* region appears to have a more complex relationship to adult migration characteristics in populations from the interior Columbia River basin. For example, some interior spring/summer Chinook salmon populations, such as Johnson Creek in the Salmon River basin (Idaho), segregate for variants at *GREB1L/ROCK1* that are evolutionarily related to "early" and "late" alleles in coastal populations (Narum et al. 2018). The Johnson Creek population has a unimodal "early" freshwater time of entry (compared to any coastal or Columbia River fall-run population), but exhibits a bimodal return timing to the spawning grounds that is associated with early and late alleles at *GREB1L/ROCK1* (Narum et al. 2018; Koch and Narum 2020). Results from these studies indicate that freshwater entry timing and arrival at spawning grounds might be 2 different phenotypes that are highly correlated in coastal lineages, but which can be uncoupled for interior populations. Distinctive patterns of linkage disequilibrium also occur in this region of chromosome 28, which suggests 2 haplotype blocks for

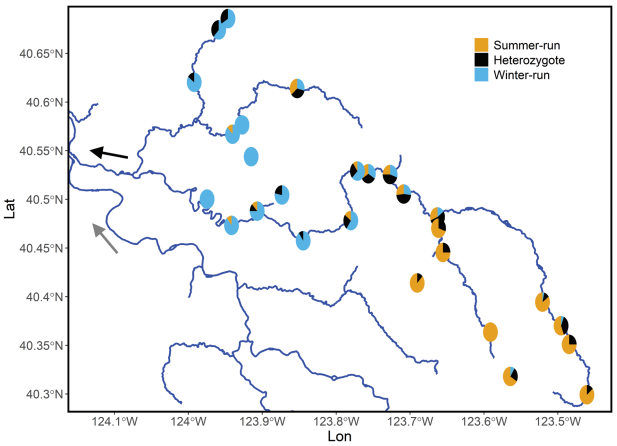

Figure 8. Distribution of *GREB1L/ROCK1* genotypes in juvenile steelhead sampled in the Van Duzen River, a major tributary of the Eel River, CA. Genotype calls were made using the combined likelihoods from snp649286 and snp649467. The black arrow indicates direction of flow of the Van Duzen River and the grey arrow indicates direction of flow of the Eel River. Modified from Kannry et al. (2020).

the interior spring/summer lineages instead of one block as seen in other lineages of Chinook salmon and steelhead (Collins et al. 2020; Koch and Narum 2020; Willis et al. 2020, 2021). Thus, *GREB1L* and *ROCK1* (and/or their regulatory regions) might have a different effect on each of these 2 phenotypic traits, with the portion of the region proximal to *ROCK1* more directly associated with timing of arrival to spawning grounds than freshwater entry.

Boundaries of these haplotype blocks appear to coincide with the duplicated genomic region identified by Thompson et al. (2020). This is indicated by the fact that the interior spring/summer lineages [represented by the Johnson Creek fish used for the CHI06 reference genome (Narum et al. 2018)] share nucleotide variation with California spring-run near *GREB1L*, but share nucleotide variation with California fall-run fish within the duplicated region (Figure S2 in Thompson et al. 2020). Thus, within the *GREB1L/ROCK1* region in Chinook salmon there appear to be adjacent haplotypic blocks with distinct evolutionary histories, including histories of introgression and spread through a large swath of the species' range.

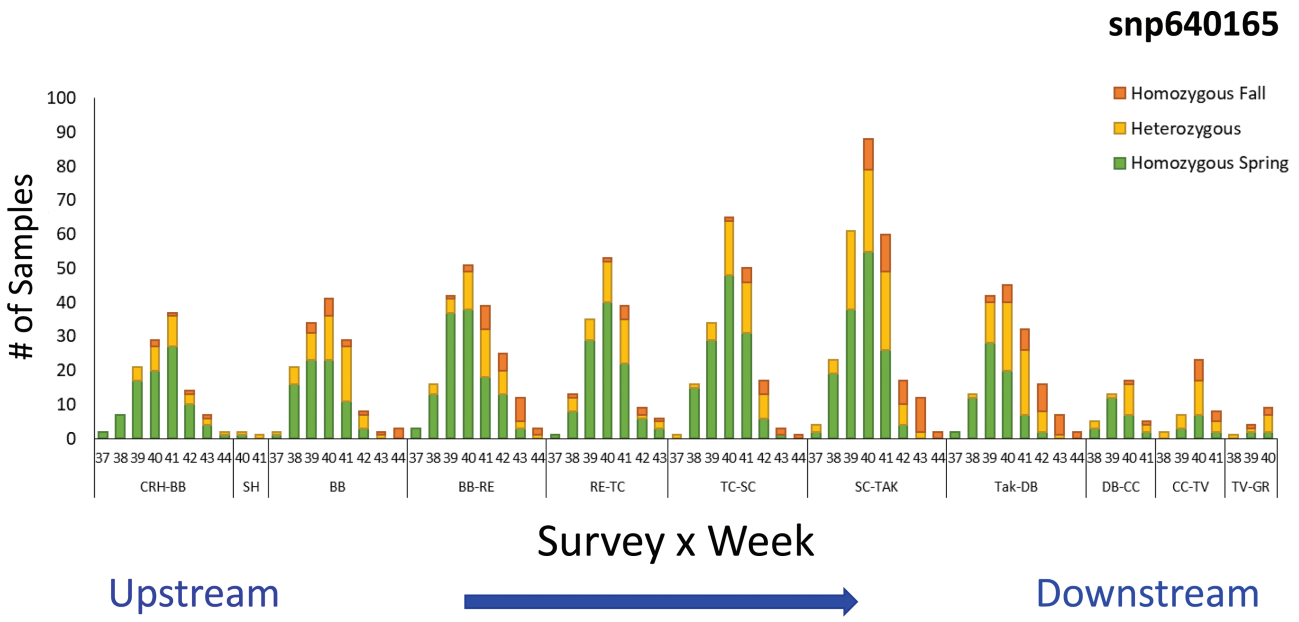

Figure 9. Distribution of *GREB1L/ROCK1* SNP1 genotypes across survey reaches and time in Chinook salmon from the Rogue River, Oregon (combined data for 2016–2018). *GREB1L/ROCK1* SNP1 (snp640165) is more diagnostic of adult migration phenotype in Rogue River Chinook salmon than SNP2 (snp670329) (T. Thompson, unpublished data). The Julian week when carcass samples were collected is on the *x* axis and ranges from 37 (10–16 September) to 44 (28 October–4 November), grouped by survey reach. The most upstream survey location is Cole Rivers Hatchery (CRH) and the furthest downstream location is the old Gold Ray Dam site (GR). Number of carcass samples collected is on the *y* axis. Reproduced from O'Malley et al. (2020b).

Table 3. Summary of determinations for state and federal petitions requesting changes to Chinook salmon or steelhead CUs and/or listing status, based primarily on new genomics data

CU	Type	Request	Determination	Citation
SONCC Chinook	Federal	Split CU by run type and list spring run	Splitting not warranted	NMFS (2021)
Oregon Coast Chinook	Federal	Split CU by run type and list spring run	Splitting not warranted	NMFS (2021)
N Cal Steelhead	Federal	Split CU by run type and list summer steelhead as endangered	Splitting not warranted[a]	NMFS (2020)
N Cal Steelhead	State	Split CU by run type and list summer steelhead as endangered	List summer steelhead separately as endangered	CFGC (2021a)
UKTR Chinook	Federal	Split CU by run type and list spring Chinook, or list current CU based on declines of spring-run	Pending	NA
UKTR Chinook	State	Split CU by run type and list spring Chinook, or list current CU based on declines of spring-run	List spring Chinook separately as threatened	CFGC (2021b)

N Cal = Northern California; SONCC = Southern Oregon–Northern California Coast; UKTR = Upper Klamath-Trinity Rivers.
[a]The CU is currently federally-listed as threatened.

Finally, it is clear that variation at the *GREB1L/ROCK1* region contributes to variation both within and between the interior Columbia and coastal Chinook salmon lineages, but current studies do not rule out the possibility that other genomic regions also play a role in some of the major adult migration difference between these lineages. Interior Columbia steelhead (which all have relatively early freshwater entry times) also appear to have a complex and not-fully-resolved relationship between adult migration phenotypes, related traits, and variation in the *GREB1L/ROCK1* region (Micheletti et al. 2018; Willis et al. 2020).

IV.3 Evolution of Early-Migration Alleles

The evolutionary history of the *GREB1L/ROCK1* region is complex and has not been well characterized throughout each species' entire range. But it is clear that the early and late haplotypes evolved long ago in each species' evolutionary history (Prince et al. 2017; Thompson et al. 2020). Based on available data, it is also clear that allelic variants for early migration have not arisen independently via new mutations from the genomic background of late migration individuals in each watershed. Rather, the dominant evolutionary feature of these alleles appears to have been migration, which has spread the alleles throughout most of the range of both species, followed by repeated and parallel increases in frequency in habitats that support early-run ecotypes.

Maintenance of variation at *GREB1L/ROCK1* is as interesting as its genesis. Large-effect alleles that are beneficial for all individuals, in all environments, and at all times are expected to rapidly sweep

to fixation. Maintenance of large-effect polymorphisms is therefore thought to require that fitness effects vary among individuals, environments, or through time. For example, large-effect polymorphisms can be maintained when one allele confers higher juvenile survival, and another confers a reproductive advantage Johnston et al. 2013, when the phenotypic optimum and large-effect allelic dominance are both sex-dependent (Barson et al. 2015), or when the ends of the phenotypic distribution have a fitness advantage over intermediate phenotypes (Narum et al. 2018). Large-effect polymorphisms also can be maintained in populations experiencing immigration from other population(s) with different phenotypic optima (Savolainen et al. 2013). It seems likely that at least some of these factors have helped to maintain the large-effect polymorphism at *GREB1L/ROCK1*.

IV.4 Dominance

An important consideration is the fact that although salmonid phenotypes are typically characterized as dichotomous—e.g., early vs. late run-timing—a locus of major effect with 2 alleles has 3 possible genotypes: 2 homozygotes and 1 heterozygous. However, the relative phenotypes of these 3 genotypes—i.e., the dominance pattern—can be sensitive to when and how phenotypes are measured. For both Chinook salmon and steelhead, robust, generalizable conclusions regarding dominance can be difficult in light of uncertainties associated with accurately and consistently measuring run-timing phenotypes. For example, Thompson et al. (2019a) found that heterozygous Chinook salmon in the Rogue River overlapped more with homozygous-late than homozygous-early fish when collected relatively high in the system, but that this was likely an artifact of phenotyping methodology, as the heterozygotes had entered earlier but held lower in the river before migrating to the collection site during the spawning season (O'Malley et al. 2020b). In the California Central Valley, Thompson et al. (2020) found that heterozygous Chinook salmon were more likely to be classified as fall run than as spring run when phenotyping was based on spatio-temporal patterns related to spawning; however, in the Klamath River, they found that heterozygotes entering fresh water overlapped in timing slightly more with homozygous-early than with homozygous-late fish (Figure 7). Similarly, in the interior Columbia River Basin, heterozygous steelhead appeared more intermediate with respect to passage date lower in the system (Prince et al. 2017; Willis et al. 2020), but more similar to homozygous-late individuals with respect to spawning-ground arrival date, potentially because environmental conditions at intermediate times affect the rate of upstream movement (Willis et al. 2020). Consistent with this, Pearse et al. (2019b) found that heterozygotes had complete overlap with late-migrating individuals in Eel River steelhead collected near the spawning grounds. Conversely, in all examined coastal locations where spring-run Chinook salmon are not currently present and were either extirpated (Shasta River, Scott River, Iron Gate, and Wynoochee Rivers) or were never known to exist (Eel and Russian Rivers), early alleles are absent or extremely rare (Thompson et al. 2019a, 2019b; Thompson et al. 2020), which suggests that the early allele is not completely recessive. Similarly, in steelhead, early alleles are absent from the South Fork Eel River, which is not currently occupied by summer steelhead, suggesting that the early allele is not completely recessive (Kannry et al. 2020). Finally, some evidence supports the idea that dominance relationships in the *GREB1L/ROCK1* region might be dependent upon the specific migration phenotypes, specific lineages, and marker proximity to the 2 genes in this region: alleles near *ROCK1* associated with early arrival migration timing appeared to have strong dominance effects across the 3 Columbia Basin lineages, but effects differed by lineage for markers near *GREB1L* (Koch and Narum 2020; Willis et al. 2021). Thus, while empirical data so far show that heterozygotes in general have a run-timing phenotype intermediate between the 2 homozygotes (i.e., an additive or codominant pattern), the apparent dominance relationships can vary depending on how the phenotype is measured and/or the dynamics of the habitat in a given population.

IV.5 Spatial Distribution

For Chinook salmon, the *GREB1L/ROCK1* region and its association with adult migration timing has been best characterized in coastal and Columbia River watersheds (Supplementary Table S1), including the Sacramento/San Joaquin, Klamath, Rogue, Nooksack, Puyallup, and Chehalis rivers on the coast and the Cowlitz, Lewis, McKenzie, Clearwater, Deschutes, Yakima, Methow rivers, Johnson Creek, and Priest Rapids Hatchery in the Columbia; smaller sample sets have also been analyzed from some other rivers. Characterization of *GREB1L/ROCK1* variation in steelhead has also largely focused on coastal and Columbia River watersheds (Supplementary Table S2). Although populations in Canada, Alaska, and Russia are yet to be studied in detail (and those from Alaska in particular have a smaller range of river-entry timing than do southern populations), it is clear that *GREB1L/ROCK1* variants are strongly associated with adult-migration phenotypes across at least a substantial portion of each species' range, as well as in diverse geographic and ecological environments.

Several studies have examined distributions of early- and late-migration alleles within individual watersheds (Thompson et al. 2019a, 2019b, 2020; Collins et al. 2020; Ford et al. 2020; Kannry et al. 2020; O'Malley et al 2020a, 2020b). Three general patterns have emerged from studies conducted within coastal drainages. First, alleles associated with early migration tend to be found at higher elevations (Figures 8 and 9), consistent with what is known about the ecology of adult migration timing (Section II). Second, in locations where early and late migrators overlap in spawning, heterozygotes are relatively common (Figures 8 and 9). This suggests that individuals of different migration times readily interbreed when the opportunity is present (either through natural or anthropogenic causes), and provides an explanation for the low overall genetic differentiation observed between run-types in coastal watersheds (Waples et al. 2004; Prince et al. 2017; Thompson et al. 2019b, Thompson et al. 2020; Ford et al. 2020). Third, in locations where the early-migration phenotype does not exist—either naturally or as a result of recent extirpation—early-migration alleles are rare or absent, and when they are seen, might be due to recent migration from nearby reservoirs of the allele (Thompson et al. 2019a, 2019b, 2020; Collins et al. 2020; Kannry et al. 2020). This suggests that early-migration alleles tend to be lost in locations that lack suitable habitat to support the early-migration phenotype, which cannot be expected to arise from late-migrating populations that lack early alleles. For *O. mykiss*, resident (non-anadromous) populations above barriers to anadromy can carry the early-migration allele (Pearse et al. 2019b; Kannry et al. 2020), consistent with the historical presence of early-run phenotypes in these areas. In these cases, residents could provide an important source of genetic variation for the early phenotype, which has apparently occurred recently following dam removal in the Elwha River (Fraik et al. 2021).

In Chinook salmon from the interior Columbia River, the early (nominally spring- and summer run, depending on the location) and late (nominally summer- and fall-run) lineages remain strongly differentiated in both genetics and life history (Waples et al. 2004; Narum et al. 2008; Moran et al. 2013), but phenological diversity

within the 2 major lineages is associated with adult migration alleles (Hecht et al. 2015; Fraser et al. 2020).

IV.6 Demographic Relationships between Early- and Late-Migrating Fish

Interbreeding between individuals with different adult migration timings has occurred historically, is expected, and likely varies depending on environmental conditions (Prince et al. 2017; Ford et al. 2020; Thompson et al. 2020). However, the degree of interbreeding in some watersheds has undoubtedly increased over the past century, as habitat alterations and other human actions have increased the potential for spatiotemporal overlap between early and late runs (see Section II.4). But precisely estimating natural/historical levels of interbreeding is challenging. For example, an analysis of recombination patterns in the Klamath River rejected the hypothesis that no interbreeding occurred between spring and fall runs prior to 200 years ago, but did not distinguish among a wide range of historical interbreeding scenarios (e.g., 1% vs. 25%; Thompson et al. 2020). In addition, salmon habitat is dynamic over a variety of temporal scales even in pristine watersheds (Waples et al. 2008), and as a consequence natural levels of interbreeding must have varied over time as well.

In many locations, there are indications that human-driven habitat modifications have increased opportunities for interbreeding. In Chinook salmon, many heterozygotes have been observed in contemporary samples from the Salmon (Klamath, CA), Rogue (OR), and Chehalis (WA) River basins, indicating high levels of current and/or recent interbreeding among fall and spring-run fish (Thompson et al. 2019a, 2019b, 2020; Ford et al. 2020; O'Malley et al. 2020b). In the Salmon River, the homozygous-early, heterozygous, and homozygous-late genotypes were found in nearly Hardy–Weinberg equilibrium proportions in one data set, suggesting spring and fall Chinook salmon are currently interbreeding at a high rate (Thompson et al. 2020). Documented anthropogenic alterations in that watershed (e.g., modification of low flow barriers that previously hindered fall-run migration) have likely increased opportunities for interbreeding (Olson and Dix 1993). In the Rogue River, 1942–2009 data from an upper-basin fish-counting station indicate that the frequency of fall-run fish accessing historical spring-run habitat increased sharply after a dam was constructed, with a concomitant increase in intermediate migrators (putative heterozygotes; Thompson et al. 2019a; ODFW 2000). Importantly, the frequency of fall-run and intermediate migrators in the Upper Rogue was consistently low across almost 40 years, before a substantial increase corresponding to construction of the dam. Despite this increase, there is currently a significant degree of temporal and spatial separation among homozygous-early and homozygous-late genotypes, and, to a lesser extent, among homozygous-early and heterozygotes (O'Malley et al. 2020b), likely due to improvements in flow management in recent years. However, the current extent of reproductive isolation between spring and fall Chinook salmon in the Rogue is below predam levels (Thompson et al. 2019a; ODFW 2000; O'Malley et al. 2020b). Similarly, US Fish and Wildlife surveys noted a reduction in spatiotemporal segregation between spring- and fall-run spawning in the Chehalis basin after a dam was built (Hiss et al. 1985), and a substantial proportion of heterozygotes observed in the Chehalis were sampled near this dam (Thompson et al. 2019b).

To date, comparable genomic studies of steelhead have not been published, but steelhead undoubtedly face the same issues. High frequencies of heterozygotes in some steelhead populations indicate recent and likely ongoing interbreeding (Pearse et al. 2019a, 2019b; Kannry et al. 2020), and one non-genomic study in the Siletz River (OR) found summer-winter hybrids in a summer-run population after a fish ladder installed at a barrier falls gave winter-run steelhead access to habitat that had previously been accessible only to early migrators (Hemstrom et al. 2018). Therefore, although the degree of demographic interaction between early- and late-run fish naturally varies over time and some degree of interbreeding is normal, habitat modifications undoubtedly have increased opportunities for interbreeding in many locations.

V. Discussion

V.1 Conclusions Emerging from the Review

Major conclusions from the previous sections can be summarized as follows:

- Loci of large effect are not uncommon, in salmon or other species. Changes in large-effect allele frequency, either by adaptation or random genetic drift, can substantially affect adaptive potential. For traits with little background polygenic variation, loss of polymorphism at a large-effect locus can preclude future adaptation to a shifting phenotypic optimum.
- Evolutionary potential for polygenic traits is much more temporally stable through bouts of adaptation than is the case for traits controlled by large-effect genes.
- In genomics-scale datasets for Chinook salmon and steelhead, overall patterns of population genetic differentiation continue to support relationships found in earlier studies. However, at a single small region of chromosome 28 in both species, a strong association is found between specific alleles and adult migration timing.
- Strongest associations have been found in coastal drainages, with more complex patterns evident in the interior Columbia River.
- The strength of these associations is positively correlated with marker density, indicating the importance of marker choice in interpretation of results.
- Dominance patterns in both species are largely consistent with an additive (codominant) model, with perhaps slight partial dominance for the early allele in some populations.
- In many cases, difficulty in precisely defining migration phenotypes for individual fish creates uncertainty in evaluating dominance patterns and strength of association.
- It does not appear that "early" alleles can persist indefinitely in systems that do not support the early-migrating phenotype.
- Interbreeding between alternate adult migration homozygotes is common in many streams. Interbreeding almost certainly occurred historically to some degree; however, various anthropogenic modifications within the last century or so have increased opportunities for genetic exchange, to a degree that varies by location and is difficult to quantify precisely due to lack of robust historical data.
- Early-migrating populations have been adversely affected by anthropogenic changes that both increase and decrease migratory capabilities. Impassable dams have precluded access by early-migrating fish to preferred upriver spawning and rearing areas. Conversely, habitat modifications that reduce flow and/or temperature barriers have allowed late-migrating fish to access areas that were previously available primarily to early-migrating populations.
- In some cases at least, resident *O. mykiss* trapped above impassable barriers can provide a reservoir of early-run alleles that could be tapped for restoration/recovery efforts.

V.2 Conservation Implications

V.2.1 Identification of CUs

For coastal Chinook salmon and steelhead, federal ESA status reviews conducted in the 1990s concluded that differences in adult run timing represent diversity contained within larger CUs. Part of the rationale for these conclusions was genetic data suggesting that early-migrating populations had evolved many times independently from nearby late-run populations. Subsequent genomic data have confirmed the close overall genetic similarity between early- and late-migrating fish in these systems, but have also shown that adult migration timing is strongly associated with specific alleles in a single genomic region. What are some of the potential implications for CU designations? Answering this question requires consideration of inherent tradeoffs between lumping and splitting in defining CUs, as well as the objectives one is trying to accomplish. Different management goals can result in different CU configurations, so there is unlikely to be a single "correct" way to identify CUs. Our literature review indicated that large-effect loci are not uncommon, in salmonids as well as other species, so identifying CUs based on these small genomic regions could potentially lead to an unmanageable plethora of small CUs, or situations in which different large-effect loci suggest conflicting CU configurations. Conversely, including multiple life-history types within a single CU could also be potentially problematic by making it more difficult to implement separate conservation and management regimes. Furthermore, because not all large-effect loci warrant equal consideration from a conservation perspective, due to their highly variable characteristics (e.g., effect size, trait importance, dominance pattern), potential problems associated with fine-scale splitting of CUs could be alleviated by developing stringent criteria for evaluating if/when large-effect loci might be useful in CU identification (Kardos and Shafer 2018). A recent workshop reviewed these issues and concluded that "using patterns of genetic variation throughout the genome remains important for identifying CUs, rather than identifying units based solely on small genomic regions associated with specific traits" (Ford et al. 2020, p. 35).

After publication of Prince et al. (2017) and subsequent articles, a number of petitions were filed seeking legal protection of early-run populations of Chinook salmon and steelhead under the federal ESA or the state of California's similar legislation (CESA 1970). The petitions shared the primary argument that new genomics data indicate that previous listing determinations should be revised, with respect to CU configuration and/or listing status. As of December 2021, determinations had been made for all but one of these petitions (Table 3).

The Northern California steelhead CU, which includes both summer and winter steelhead, has been listed as threatened under the ESA (NMFS 2000, 2006) but not listed under the CESA, and both federal and California state petitions asked that the summer steelhead be placed in a separate CU and listed as endangered. Following a scientific review (Pearse et al. 2019b), NMFS (2020) determined that the request to split the current federal CU was not warranted, and that both summer and winter steelhead would remain together in a single unit listed as threatened. In response to the state petition, the California Department of Fish and Wildlife (CDFW) conducted a status review and concluded that Northern California summer steelhead do not qualify as a listable unit under CESA (CDFW 2021). However, after considering the CDFW status review as well as information presented by the petitioners and through public comments, the California Fish and Game Commission (CFGC), which implements CESA, concluded that the petitioned actions were warranted: Northern California summer steelhead constitute their own CU and warrant an endangered listing under the CESA CFGC 2021a.

Federal and CESA petitions were also filed for Chinook salmon from the upper Klamath River basin, where NMFS (1998) identified an Upper Klamath-Trinity Rivers (UKTR) CU that includes both early- and late-run populations but concluded that listing under the ESA was not warranted. These petitions asked that either 1) early-run populations be assigned to a separate CU and listed as threatened or endangered, or 2) the entire CU be listed as threatened or endangered. Although a NMFS scientific review panel concluded that separation of the spring run into a separate CU was not appropriate (Anderson et al. 2018), the agency had not made a final determination on listing status of UKTR Chinook salmon at the time this article was finalized. The state determination for UKTR Chinook salmon under CESA paralleled that for Northern California steelhead: the status review (CDFW 2020) concluded that the federally defined CU should not be split, and that listing the entire unit was not warranted, but the CFGC agreed with the petitioners that UKTR spring Chinook salmon constitute a separate CU, and concluded that a threatened listing for the early-run CU was warranted under the CESA (CFGC 2021b).

The remaining 2 petitions requested ESA listing of spring Chinook salmon from the Oregon Coast and Southern Oregon–Northern California Coast (SONCC) CUs; both CUs include early and late migrators and neither has been listed under the ESA. Both petitions requested that early-run populations be considered a separate CU and listed as threatened or endangered under the ESA. A scientific review panel concluded that spring-run Chinook salmon from the Oregon Coast or SONCC do not meet the criteria to be a CU as it is defined by NMFS policy (Ford et al. 2021), and NMFS subsequently found that listing of the spring-run populations was therefore not warranted (NMFS 2021).

The biological arguments for and against separate CU status for early-run populations, including considerations of recent genomics studies, are complex, and the reader should look to the referenced petitions and status reviews for details. In brief, those favoring separate CU status based on the genomics findings 1) emphasize the monophyletic nature of early-run alleles in the GREB1L region instead of average patterns of variation throughout the genome; 2) argue the early-run life-history depends upon unique alleles that are at risk of being lost from a large portion of the species, and if that occurs it would be difficult if not impossible to regain those alleles; and 3) argue that new genomics data further underscore the distinct biological characteristics of the early-run life-history. Arguments against changing CU status tend to focus on 1) the lack of reproductive isolation between the early- and late-run forms, and 2) concerns about defining noncongruent CUs based on the average patterns of variation throughout the genome.

Additional factors, not directly related to genomic architecture, that are potentially relevant in considering these federal and state listing decisions include the following:

- Although a provision to list units smaller than species or subspecies was included in the original 1973 version of the ESA, the current DPS language comes from 1978 amendments (Endangered Species Act Amendments of 1978, 16 U.S.C. § 1531, Definition 16). The ability to list DPSs provides agencies with considerable flexibility to determine what subspecific groups merit legal protection under the ESA (e.g., 619 F.3d 1024 (C.A.9 (Cal.), 2010); Alagona 2016). On the other hand, Congress recognized that the ability to list units smaller than formally-defined species or subspecies also provides "great potential for abuse," and accordingly they directed that the DPS provision be used "sparingly" (Sen. Rep.151, 96th Cong., 1st Sess., 1979).

- Unlike the federal ESA, the CESA has no formal provision for listing DPSs or similar units below the species/subspecies level. The 1984 amendments to the CESA define an "endangered species" to be "a native species or subspecies. However, on a number of occasions, the CFGC has listed units at a lower level than a taxonomic subspecies, and this approach has been upheld by the State Court of Appeals (California Forestry Association v. California Fish and Game Commission, 2007). The CFGC concluded that UKTR spring Chinook salmon qualify as a 'subspecies' according to CESA (CFGC 2021b), and (as this paper goes to press) a similar formal determination regarding Northern California summer steelhead is expected early in 2022.
- In their listing determinations, agencies implementing the ESA "shall make determinations … solely on the basis of the best scientific and commercial data available" (Sec. 4(b)(1)(A)). Among many other types of data, this can include traditional ecological knowledge (TEK). Whereas the recent NMFS scientific reviews and listing determinations for Chinook salmon (Anderson et al. 2018; Ford et al. 2021; NMFS 2021) did not specifically reference TEK, the CFGC clearly considered TEK to be an important source of information in their listing determination for UKTR spring Chinook salmon under CESA (CFGC 2021b). The Karuk (and other indigenous peoples) have considered spring- and fall-run Chinook distinct since time immemorial, using a different name for each run type and reserving for the spring-run a profound sociocultural significance (Langin 2018). Canada has developed a formal process under their Species At Risk Act to consider Aboriginal Traditional Knowledge (ATK), a close analogue of TEK (https://cosewic.ca/index.php/en-ca/assessment-process/atk-guidelines.html).
- The California rulings do not change the number or boundaries of federally identified CUs, but they differ from the federal framework in one important respect: within each of the federally listed CUs, the state listings group all early-run populations into a unit separate from all late-run populations. That is, the recent state listings follow Panel V in Figure 3. In contrast, federally defined ESUs generally follow Panel IV, grouping run-timing life-history types within CUs, consistent with overall genetic affinities following geography more than life history (as in Figure 3, Panel I).

Regardless of the challenges and uncertainties outlined above, there is widespread agreement that diversity of adult migration timing is important to conserve, so even if that diversity exists within a single CU, an approach like that proposed by Funk et al. (2012) might be used to emphasize adaptively important groups of populations within larger CUs. In fact, most recovery teams for ESA-listed Pacific salmon and steelhead have adopted a similar approach by identifying diversity strata, including adult migration variation, within ESUs or DPSs that reflect important components of eco-evolutionary diversity (Myers et al. 2006; Ruckelshaus et al. 2006; Lawson et al. 2007; Spence et al. 2008).

V.2.2 Population Viability

Even if CU configurations remain unchanged, it is important to consider ramifications of the new genomics data for assessments of population viability. Important considerations include the following:

- Spring Chinook salmon and summer steelhead occupy a specialized ecological niche—upstream areas accessible primarily during spring flow events—and status review and recovery planning teams have consistently concluded that viable populations of both early- and late-migrating forms are necessary for the larger ESUs as a whole to be considered viable (Busby et al. 1996; Myers et al. 1998; McElhany et al. 2006; Shared Strategy Development Committee 2007; Dornbush 2013; Hard et al. 2015; Pearse et al. 2019b).
- These specialized habitat requirements make early-run populations particularly vulnerable to decline or extirpation due to habitat degradation, blockage of migratory routes, climate change, and interactions with hatchery and harvest management (Nehlsen et al. 1991; Gustafson et al. 2007; Tillotson et al. 2021).
- Discovery that specific alleles in one genomic region are associated with the early-run trait implies that the trait is at greater risk than if it were highly polygenic, because loss of the "early" allele(s) equates to loss of the phenotype. If early allele(s) were recessive, a genetic reservoir might persist in late-run populations, but that does not appear to be the case. Available evidence indicates that early allele(s) are codominant or partially dominant, and surveys in multiple Chinook salmon populations indicate that early alleles disappear or become extremely rare after extirpation of the early phenotype. This indicates that, although hatchery propagation might be an important conservation strategy and source of early-run alleles in the short term, the only reliable way to conserve early-run genes in nature is by maintaining habitat that supports early-run phenotypes.
- Evolutionary potential for polygenic traits is much more temporally stable through bouts of adaptation (Barton and Keightley 2002; Oomen et al. 2020; Kardos and Liukart 2021). Accelerated genetic drift can further compound the loss of rare alleles in declining populations experiencing limited gene flow of adapted alleles. If adaptive large-effect alleles are lost from a population under such circumstances, the phenotypes they are associated with are unlikely to re-evolve in timescales relevant to conservation, unless the large-effect alleles are restored through immigration. Conservation monitoring of ecologically important genes of large effect (i.e., keystone genes; Skovmand et al. 2018) can identify populations where allele frequencies have shifted to favor a particular phenotype (e.g., Thompson et al. 2019a) and adaptive alleles have become rare across a broad geographic region (e.g., Collins et al. 2020).

These developments in turn have important ramifications for conservation planning and management. The most reliable strategy is to conserve adult migration diversity in situ by providing conditions that can support early-run phenotypes at sustainable levels. If an early-migrating population is lost, the original polygenic paradigm suggested that, providing suitable habitat conditions were available, an early run might re-evolve from a local late-run population within ecological time scales. New genomics data, by contrast, suggest that restoration of an extirpated early-run population would have to rely on natural immigration of fish carrying the early allele(s), or active anthropogenic intervention (enhancing connectivity to allow natural recolonization, or actively through translocations or assisted migration).

If restoration were as simple as finding some fish with the desired *GREB1L/ROCK1* genotypes, this could be good news from a conservation perspective. However, 2 major factors also need to be considered. First, empirical data demonstrate that attempts to transplant Pacific salmon within their historical range have rarely been successful in producing sustainable populations (Withler 1982), with most exceptions being related to recolonization events involving newly accessible habitat (Pess et al. 2014; Pitman et al. 2020). Reasons for these failures appear to be related to disruption of the complex series of adaptive life-history events required to complete the anadromous life cycle (Allendorf and Waples 1996; Wood

and Foote 1996; Figure 1.1 in Box 1). Therefore, success of such efforts cannot be taken for granted, and any potential benefits would have to be weighed against risks, including disease transfer and unintended interactions with non-target species/populations.

Second, it will be important to expand the geographic scale for assessing viability. Under the polygenic paradigm, conservation and management of adult migration diversity could focus on a local scale (within CUs or even within watersheds). However, since early-run phenotypes appear to require early-run alleles, and early-run populations appear to be at relatively high risk everywhere, it becomes important to consider which populations can be considered viable sources for these valuable genetic resources into the foreseeable future. This broader perspective could be challenging to implement under the US ESA, as salmon ESUs are considered separate ESA "species" and there is little precedent for conducting risk assessments for multiple interacting ESA "species."

V.3 Uncertainties and Complications

One critical area of uncertainty is the degree to which adult migration diversity is partitioned among populations versus among individuals within populations. In many cases, federal, state, tribal, and local salmon management strategies consider early and late migration timing to be characteristic of demographically independent populations, albeit with substantial variation among individuals. However, heterozygous genotypes at the *GREB1L/ROCK1* region are common in contemporary samples of Chinook salmon and steelhead from many coastal drainages. Thus, distinguishing between "natural" levels of interbreeding between the 2 life-history types and recent increases caused by anthropogenic modifications to salmon habitats is a pressing, albeit challenging, problem. In some situations, high-quality monitoring data from prior to major anthropogenic habitat alteration might provide targets for future management and/or restoration actions (e.g., Rogue River fish counts from Gold Ray Dam; Thompson et al. 2019a).

Whether human-mediated or not, interbreeding between run types presents challenges for status monitoring, recovery planning, and other management actions. If early and late runs are not demographically independent (and making that determination can be very challenging; Box 2), current population modeling approaches for assessing risk are flawed and potentially could be improved by incorporating genomic information to allow estimation of run-type mixture proportions. Limited interbreeding with late-run fish might help maintain viability of the early phenotype, especially for smaller, more isolated populations, but this "genetic rescue" benefit generally can be achieved with even very low levels of interbreeding (Mills and Allendorf 1996). High levels of interbreeding currently reported in some areas indicate that early and late individuals substantially overlap in space and time. In some locations that might have occurred historically, but in others this pattern suggests that early individuals have lost access to their historic habitats, or late individuals are now able to access them. In either case, it is likely that the late-run phenotype would experience a competitive advantage because they would not have to face the same risk-benefit tradeoffs as early-migrating individuals.

Another important unknown is how much genetic variance for adult migration timing is present in the genome outside of *GREB1L/ROCK1*. There is clearly a large phenotypic variance among individuals with the same *GREB1L/ROCK1* genotypes (Thompson et al. 2020). This remaining variation is either attributable to random environmental differences among individuals, or to genetic variants elsewhere in the genome with substantially smaller effects than at *GREB1L/ROCK1*. Obtaining a broad, comprehensive understanding of the genetic basis of adult migration timing, and of the evolutionary potential of populations devoid of genetic variation at *GREB1L/ROCK1*, will require determining what proportion of the h^2 is due to genome-wide polygenic variation.

Dominance affects our understanding of conservation implications and optimal management strategies. In the Klamath, the genetic effect on time of freshwater return appears to be additive (Thompson et al. 2020)—heterozygous fish return earlier than homozygous-late fish, but not as early as homozygous-early fish. Such codominant inheritance of the trait might have facilitated the original dispersal of early alleles: under additivity the heterozygous offspring of rare early-run strays into late-migrating populations would have returned earlier than most existing late-run fish in the basin, giving heterozygotes the potential to mate assortatively, greatly increasing the chance of producing homozygous early-run offspring to colonize any habitat suitable for early-run fish in the new basin. In the present day, however, persistence of early-run alleles in basins with early-run fish would be most likely if the allele were recessive. If the allele were recessive (which does not appear to be the case based on available data), then heterozygous fish and late-run fish would have the same phenotype, allowing heterozygotes to serve as a long-term reservoir of the allele, even if conditions in the basin changed to disadvantage earlier-arriving fish.

Another key uncertainty involves the optimal way to define adult migration phenotypes. Applying the most widely-used criterion (time of entry into fresh water on the spawning migration) is difficult because most fish are not monitored at that point. This requires a backward extrapolation from the date and location where a fish is first encountered, such that variation in true migration time can be difficult to distinguish from estimation error. Furthermore, when a salmon or steelhead physiologically commits to mature the following year, that "decision" is typically made many months before it begins to migrate (Thorpe et al. 1998), and that migration might cover thousands of kilometers at sea before the fish enters fresh water (Groot and Margolis 1991). A complete migratory phenotype thus might include timing of these other key maturation-related events, which are even more challenging to measure.

Regarding one empirical observation, there appears to be little uncertainty or disagreement: across large geographic areas in both Chinook salmon and steelhead, early-run populations have been extirpated at a disproportionate rate, and remaining populations are under greater stress than late-run populations. Science can provide insights into likely consequences of alternative conservation and management actions to respond to this observation, and some of the suggestions in the next section could be useful in that regard. But science alone cannot determine which action(s) "should" be taken, because doing so requires consideration of myriad social, cultural, economic, legal, and ethical tradeoffs that arise when natural resource decisions also affect society as a whole (Lackey 2004). For example, the observation of a disproportionate decline in early-run phenotypes can be viewed from at least 2 fundamentally different perspectives. In one view, this indicates that strong conservation measures focused on early-run populations are needed, because genetic change and biodiversity loss can compromise both the current viability and the future evolutionary potential of the species. Conversely, if major human-mediated changes in the Anthropocene are accepted as a given, decline of early-migrating phenotypes could be viewed as the natural consequence of a species adapting to its current, anthropogenically modified environmental conditions.

V.4 Research to Address Uncertainties

In this rapidly moving field, the vast majority of the information reviewed here has emerged within the last 5 years. Priority research needs for the future include the following:

- More thorough marker development and validation, including identification of functional variant(s) in the *GREB1L/ROCK1* region associated with migration phenotypes.
- Better standardization and characterization of adult migration phenotypes in multiple populations and lineages, including when the "decision" to migrate is made, how it relates to timing of sexual maturity, and relationships between date of freshwater entry and subsequent upstream movements. Populations with intermediate migration patterns should be examined as well as those with extreme phenotypes.
- More thorough characterization of patterns of dominance and effect sizes and how they vary in space and time.
- Greater understanding of physiological mechanisms leading to alternative migration phenotypes, including circannual rhythms, responses to photoperiod, fat deposition, onset of anorexia, osmoregulation, sexual maturation, and other processes crucial for successful migration from marine to freshwater habitats.
- Tests for association of *GREB1L/ROCK1* variation with other phenotypic traits, such as juvenile migration and timing of sexual maturity relative to freshwater entry.
- Replication across different genetic lineages of Chinook salmon and steelhead throughout their ranges. Adult phenotypes of interest include dates of freshwater entry, arrival to spawning grounds, and spawning, state of maturation (GSI) at various migration stages (ocean, freshwater entry, spawning grounds), and lipid content and composition in different tissues. Future research should carefully consider the effect of phenotyping methodology on interpretation of results.
- Comparative analyses on systems with both run types that have been differentially affected by human activities, resulting in differing levels of interbreeding between life-history types, to improve our understanding of how interbreeding affects persistence of run-type alleles.

V.5 Broader Relevance

Although this review was inspired by and focused on new genomics data for Pacific salmonids, all of the issues discussed here are likely to resonate with many other species of conservation interest. Large-effect loci have been identified in many taxa, and genetic architecture has predictable consequences for species persistence and biodiversity conservation. Similarly, how best to define and/or assess the viability of CUs is a generic problem that arises in a wide variety of regulatory frameworks, and several recent articles have discussed the relative importance of neutral versus adaptive variation, whether genome wide or in a few large-effect loci, in making conservation decisions (Ralls et al. 2020; Xuereb et al. 2020; Teixeira and Huber 2021; Fernandez-Fournier et al. 2021). However, regardless of how the CUs are defined, we suggest that the best overall conservation strategy is to maintain the ecological and evolutionary processes necessary to sustain endemic life-history variation. Our review also indicates that, when large-effect loci influence an important trait, viability assessments should take a broader geographic view to ensure that reservoirs of the required genes are conserved across the landscape.

Supplementary Material

Supplementary data are available at *Journal of Heredity* online.

Glossary

Units of Conservation

Conservation Unit (CU): a general term for a group of one or more populations considered to be a useful unit for organizing and/or prioritizing conservation efforts.

Evolutionarily Significant Unit (ESU): a CU that is defined with special attention to evolutionary significance.

Distinct Population Segment (DPS): a subspecific CU that is considered a "species" under the U.S. ESA and can be listed if determined to be threatened or endangered.

Sequencing Methods

Whole-genome sequencing (WGS): General term for methods that sequence the entire genome of an organism, as opposed to methods that sequence subsets of the genome (see reduced-representation sequencing).

Pooled WGS: A cost-saving approach whereby pools of DNA from multiple individuals are sequenced to estimate allele frequencies of the pool, rather than obtaining individual genotypes. Typically used as a variant of whole-genome sequencing.

Reduced-representation sequencing (RRS): General term describing methods that sequence subsets of the genome. Commonly used to economically genotype thousands to tens of thousands of SNPs.

Restriction-site associated DNA sequencing (RAD-seq): Reduced-representation sequencing method that sequences DNA adjacent to restriction sites. This method has many variants that differ in details of sample preparation and in number of loci generated, which typically ranges from thousands to tens of thousands.

Genetic Variants

Locus: Context dependent term used to describe a specific location in the genome. This can encompass single SNPs or larger genomic regions, particularly when these regions are inherited as a non-recombining block (i.e., inversions).

Allele: One of the genetic variants at a locus.

Haplotype: A set of DNA variants that are inherited together. A haplotype can refer to a combination of alleles or to a set of SNPs found on the same chromosome.

Allozymes (allo + enzymes): Variant sites at protein-coding loci that are detected with protein electrophoresis based on differences in amino acid sequence.

Microsatellites: Noncoding regions of DNA that contain variable numbers of short (usually 2-4 base pairs), repeated DNA sequences.

Single-nucleotide polymorphisms (SNPs): Single DNA base pairs that are variable within the target population; most SNPs only have two variant alleles.

Genome-wide association study (GWAS): Study of genetic variation spanning the genome to detect variants associated with specific phenotypic traits.

Quantitative Genetics

Additive genetic variance: Genetic variance attributed to the average effects of substituting one allele for another at a given locus. It is the component of variance that allows prediction of the rate of response for selection of quantitative traits.

Dominance: The relative importance of different alleles at a locus in determining the phenotype. A single copy of a dominant allele is sufficient to determine the phenotype (as in heterozygotes). If the phenotype of heterozygotes is intermediate, the alleles are said to be co-dominant with additive effects.

Phenotypic variance: The observed variance in the trait of interest.
Genetic architecture: The underlying genetic basis of a phenotypic trait; the number and effect sizes of genes, their interactions within and between each other, and their inheritance pattern.

Acknowledgments

Most of the authors participated in a workshop, "Reviewing and synthesizing the state of the science regarding associations between adult migration timing and specific genotypes in Chinook salmon and steelhead," held on 27–28 February 2020 in Seattle, WA, USA. A report of that workshop is available at: https://repository.library.noaa.gov/view/noaa/24980. Damon Holzer, Su Kim, and Kathleen Neely assisted with figures. Three anonymous reviewers provided useful comments that improved the manuscript. Original data used in this review are available from the primary publications. The authors declare no conflict of interest.

References

Abadía-Cardoso A, Anderson EC, Pearse DE, Garza JC. 2013. Large-scale parentage analysis reveals reproductive patterns and heritability of spawn timing in a hatchery population of steelhead (*Oncorhynchus mykiss*). *Mol Ecol*. 22:4733–4746.

Alagona PS. 2016. Species complex: classification and conservation in American environmental history. *Isis*. 107:738–761.

Allendorf FW, Hohenlohe PA, Luikart G. 2010. Genomics and the future of conservation genetics. *Nat Rev Genet*. 11:697–709.

Allendorf FW, Waples RS. 1996. Conservation and genetics of salmonid fishes. In: Avise JC and Hamrick JL, editors. *Conservation genetics: case histories from nature*. New York: Chapman and Hall. p. 238–280.

Anderson EC, Ford MJ, Garza JC, Kiernan JD. 2018. *Upper Klamath and Trinity River Chinook Salmon ESU-Configuration Review-Panel Report*. Report from the Southwest Fisheries Science Center to the NMFS West Coast Region, Protected Resources Division. Update of 15 June 2018 report.

Arciniega M, Clemento A, Miller MR, Peterson M, Garza JC, Pearse DE. 2016. Parallel evolution of the summer steelhead ecotype in multiple populations from Oregon and northern California. *Conserv Genet*. 17:165–175.

Ashraf B, Hunter DC, Bérénos C, Ellis PA, Johnston SE, Pilkington JG, Pemberton JM, Slate J. 2020. Genomic prediction in the wild: A case study in Soay sheep. Molecular Ecology. Available from: https://doi.org/10.1111/mec.16262

Barson NJ, Aykanat T, Hindar K, Baranski M, Bolstad GH, Fiske P, Jacq C, Jensen AJ, Johnston SE, Karlsson S, *et al.* 2015. Sex-dependent dominance at a single locus maintains variation in age at maturity in salmon. *Nature*. 528:405–408.

Barton NH, Etheridge AM, Véber A. 2017. The infinitesimal model: Definition, derivation, and implications. *Theor Popul Biol*. 118:50–73.

Barton NH, Keightley PD. 2002. Understanding quantitative genetic variation. *Nat Rev Genet*. 3:11–21.

Beacham TD, Candy JR, Jonsen KL, Supernault J, Wetklo M, Deng L, Miller KM, Withler RE, Varnavskaya N. 2006. Estimation of stock composition and individual identification of Chinook salmon across the Pacific Rim by use of microsatellite variation. *Trans Am Fish Soc*. 135:861–888.

Beechie T, Buhle E, Ruckelshaus M, Fullerton A, Holsinger L. 2006. Hydrologic regime and the conservation of salmon life history diversity. *Biol Conserv*. 130:560–572.

Beechie TJ, Moir H, Pess GR. 2008. Hierarchical physical controls on salmonid spawning location and timing. In salmonid spawning habitat in rivers: physical controls, biological responses, and approaches to remediation. *American Fisheries Society, Symposium*. 65:83–102.

Behnke RJ. 1992. *Native trout of Western North America*. Bethesda (MD): American Fisheries Society Monographs 6.

Brieuc MS, Ono K, Drinan DP, Naish KA. 2015. Integration of random forest with population-based outlier analyses provides insight on the genomic basis and evolution of run timing in Chinook salmon (*Oncorhynchus tshawytscha*). *Mol Ecol*. 24:2729–2746.

Busby PJ, Wainwright TC, Bryant GJ, *et al.* 1996. *Status Review of West Coast Steelhead from Washington, Idaho, Oregon, and California*. NOAA Technical Memorandum NMFS-NWFSC-27. Seattle, WA.

California Forestry Association v. California Fish and Game Commission (2007). *156 Cal.App.4th 1535*, 1541–1542.

Clemento AJ, Crandall ED, Garza JC, Anderson EC, 2014. Evaluation of a single nucleotide polymorphism baseline for genetic stock identification of Chinook Salmon (Oncorhynchus tshawytscha) in the California Current large marine ecosystem. *Fishery Bullet*. 112:112–130.

Campbell SK, Butler VL. 2010. Archaeological evidence for resilience of Pacific Northwest salmon populations and the socioecological system over the last ~ 7,500 years. *Ecol Soc*. 15:17. Available from: http://www.ecologyandsociety.org/vol15/iss1/art17/

Carlson SM, Seamons TR. 2008. A review of quantitative genetic components of fitness in salmonids: implications for adaptation to future change. *Evol Appl*. 1:222–238.

CDFW (California Department of Fish and Wildlife). 2020. *California Endangered Species Act Status Review for Upper Klamath and Trinity Rivers Spring Chinook Salmon (Oncorhynchus tshawytscha)*. Sacramento (CA): A Report to the California Fish and Game Commission. California Department of Fish and Wildlife. 211 pp., plus appendices.

CDFW. 2021. *California Endangered Species Act Status Review for Northern California Summer Steelhead (Oncorhynchus mykiss)*. Sacramento (CA): A Report to the California Fish and Game Commission. California Department of Fish and Wildlife. 188 pp., with appendices.

Ceballos G, Ehrlich PR. 2002. Mammal population losses and the extinction crisis. *Science*. 296:904–907.

CESA (California Endangered Species Act). 1970 (as amended 1984 & 1997). The full text is included within the California Fish and Game Code. Available from: https://www.animallaw.info/statute/ca-endangered-species-chapter-15-endangered-species

CFGC (California Fish and Game Commission). 2021a. Determinations regarding petitions for protection of Northern California summer steelhead and Upper Klamath-Trinity rivers spring Chinook salmon under the California state Endangered Species Act (CESA). June 16–17, 2021. Available from: https://fgc.ca.gov/Meetings/2021

CFGC (California Fish and Game Commission). 2021b. Notice of findings for Upper Klamath-Trinity River Spring Chinook Salmon (*Oncorhynchus tshawytscha*). *California Regulatory Notice Register 2021. 53-Z*.

Chatters JC, Butler VL, Scott MJ, Anderson DM, Neitzel DA. 1995. A paleoscience approach to estimating the effects of climatic warming on salmonid fisheries of the Columbia River basin. In Beamish RJ, editor. *Climate change and Northern fish populations*. Ottawa CA: Canadian Special Publication, Fisheries and Aquatic Sciences, 121. p. 48–96.

Chevalet C. 1994. An approximate theory of selection assuming a finite number of quantitative trait loci. *Genet Sel Evol*. 26:379–400.

Collins EE, Hargrove JS, Delomas TA, Narum SR. 2020. Distribution of genetic variation underlying adult migration timing in steelhead of the Columbia River basin. *Ecol Evol*. 10:9486–9502.

COSEWIC (Committee on the Status of Endangered Wildlife in Canada) 2018. *Appendix F5. Guidelines for recognizing designatable units. COSEWIC operations and procedures manual*. Ottawa: Committee on the Status of Endangered Wildlife in Canada. Retrieved from https://www.canada.ca/en/environment-climate-change/services/committee-status-endangered-wildlife/guidelines-recognizing-designatable-units.html

COSEWIC. 2019. *COSEWIC assessment and status report on the Chinook salmon (Oncorhynchus tshawytscha) in Canada 2018*. Ottawa: Committee on the Status of Endangered Wildlife in Canada. Available from: https://www.canada.ca/en/environment-climate-change/services/species-risk-public-registry/cosewic-assessments-status-reports/chinook-salmon-2018.html

Crandall KA, Bininda-Emonds OR, Mace GM, Wayne RK. 2000. Considering evolutionary processes in conservation biology. *Trends Ecol Evol*. 15:290–295.

Crozier LG, McClure MM, Beechie T, Bograd SJ, Boughton DA, Carr M, Cooney TD, Dunham JB, Greene CM, Haltuch MA, *et al.* 2019. Climate

vulnerability assessment for Pacific salmon and steelhead in the California Current Large Marine Ecosystem. *PLoS One*. 14:e0217711.

Davis CD, Garza JC, Banks MA (2017). Identification of multiple genetically distinct populations of Chinook salmon (*Oncorhynchus tshawytscha*) in a small coastal watershed. *Environmental Biology of Fishes*. 100:923–933.

Devlin RH. 1993. Sequence of sockeye salmon type 1 and 2 growth hormone genes and the relationship of rainbow trout with Atlantic and Pacific salmon. *Can J Fish Aquat Sci*. 50:1738–1748.

Doctor KK, Quinn TP. 2009. The potential for adaptation-by-time in sockeye salmon (*Oncorhynchus nerka*): the interactions of body size and in-stream reproductive lifespan with date of arrival and breeding location. *Can J Zool*. 87:708–717.

Dornbush P. 2013 *ESA recovery plan for: Lower Columbia River coho salmon, Lower Columbia River Chinook salmon, Lower Columbia River Chum Salmon, and Lower Columbia River steelhead*. Portland, OR: National Marine Fisheries Service. 503 pp.

Dudley J, Lambert R. 2003. 100 generations of selection for oil and protein in corn. In Plant Breeding Reviews. Janick, J. (Ed.). Hoboken, New Jersey: Wiley, pp. 79-110.

Epstein B, Jones M, Hamede R, Hendricks S, McCallum H, Murchison EP, Schönfeld B, Wiench C, Hohenlohe P, Storfer A. 2016. Rapid evolutionary response to a transmissible cancer in Tasmanian devils. *Nat Commun*. 7:1–7.

Falconer DS, Mackay TFC. 1996 *Introduction to quantitative genetics*. 4th ed. Essex: Longman Limited.

Fernandez-Fournier P, Lewthwaite JMM, Mooers AØ. 2021. Do we need to identify adaptive genetic variation when prioritizing populations for conservation? *Conserv Genet*. 22:205–216.

Ford MJ, Anderson E, Garza JC, Myers J, Williams TH, Wapes R. 2021. Report on a review of the Oregon coast and Southern Oregon Northern California Coastal spring Chinook salmon ESU configuration. Northwest Fisheries Science Center, National Marine Fisheries Service. Available from: https://media.fisheries.noaa.gov/2021-08/OC_SONCC%20spring%20Chinook%20ESU%20report%20060221.pdf

Ford MD, Nichols KM, Waples RS, *et al.* 2020. Reviewing and synthesizing the state of the science regarding associations between adult run timing and specific genotypes in Chinook salmon and steelhead: report of a workshop held in Seattle, Washington, 27–28 February 2020. Seattle, WA: NWFSC processed report 2020-06. Available from: https://repository.library.noaa.gov/view/noaa/24980

Fraik AK, McMillan JR, Liermann M, Bennett T, McHenry ML, McKinney GJ, Wells AH, Winans G, Kelley JL, Pess GR, Nichols KM. 2021. The impacts of dam construction and removal on the genetics of recovering steelhead (*Oncorhynchus mykiss*) populations across the Elwha River watershed. *Genes*. 12:89.

Fraser DJ, Bernatchez L. 2001. Adaptive evolutionary conservation: towards a unified concept for defining conservation units. *Mol Ecol*. 10:2741–2752.

Fraser GS., DeHaan PW, Smith CT, Von Bargen JF, Cooper MR, Desgrosseillier TJ. 2020. Overlap of spatial and temporal spawning distributions of spring and summer Chinook salmon results in hybridization in the upper Columbia River. *Trans Am Fish Soc*. 149:517–531.

Fulton LA. 1968. *Spawning areas and abundance of Chinook salmon (Oncorhynchus tshawytscha) in the Columbia River basin—past and present*. Washington, D.C.: US Dept. Commer., NMFS, Special Sci. Rep. No. 571.

Funk W, Forester BR, Converse SJ, Darst C, Morey S. 2019. Improving conservation policy with genomics: a guide to integrating adaptive potential into US Endangered Species Act decisions for conservation practitioners and geneticists. *Conserv Genet*. 20:1–20.

Funk WC, McKay JK, Hohenlohe PA, Allendorf FW. 2012. Harnessing genomics for delineating conservation units. *Trends Ecol Evol*. 27:489–496.

Groot C, Margolis L. 1991. *Pacific salmon life histories*. Vancouver (Canada): UBC Press.

Gustafson RG, Waples RS, Myers JM, Weitkamp LA, Bryant GJ, Johnson OW, Hard JJ. 2007. Pacific salmon extinctions: quantifying lost and remaining diversity. *Conserv Biol*. 21:1009–1020.

Hard JJ, Kope RG, Grant WS, Waknitz FW, Parker LC, Waples RS. 1996. *Status review of pink salmon from Washington, Oregon, and California*. Seattle: Department of Commerce.

Hard JJ, Myers JM, Connor EJ, Hayman RA, Kope RG, Lucchetti G, Marshall AR, Pess GR, Thomson BE. 2015. *Viability criteria for steelhead within the Puget Sound distinct population segment*. U.S. Dept. Commer., NOAA Tech. Memo. NMFS-NWFSC-129, 332 p.

Hastings A. 1993. Complex interactions between dispersal and dynamics: lessons from coupled logistic equations. *Ecology*, 74:1362–1372.

Havenstein GB, Ferket PR, Qureshi MA. 2003. Growth, livability, and feed conversion of 1957 versus 2001 broilers when fed representative 1957 and 2001 broiler diets. *Poult Sci*. 82:1500–1508.

Healey MC. 1983 Coastwide distribution and ocean migration patterns of stream- and ocean-type Chinook salmon, *Oncorhynchus tshawytscha*. *Can Field Nat*. 97:427–433.

Hearsey JW, Kinziger AP. 2014. Diversity in sympatric Chinook salmon runs: timing, relative fat content and maturation. *Environ Biol Fish*. 98:413–423.

Hecht BC, Matala AP, Hess JE, Narum SR. 2015. Environmental adaptation in Chinook salmon (*Oncorhynchus tshawytscha*) throughout their North American range. *Mol Ecol*. 24:5573–5595.

Hemstrom W, van de Wetering S, Banks M. 2018. Fish ladder installation across a historical barrier asymmetrically increased conspecific introgressive hybridization between wild winter and summer run steelhead salmon in the Siletz River, Oregon. *Can J Fish Aquat Sci*. 75:1383–1392.

Hess JE, Matala AP, Zendt JS, Frederiksen CR, Sharp B, Narum SR. 2011. Introgressive hybridization among major Columbia River Chinook salmon (*Oncorhynchus tshawytscha*) lineages within the Klickitat River due to hatchery practices. *Can J Fish Aquat Sci*. 68:1876–1891.

Hess JE, Zendt JS, Matala AR, Narum SR. 2016 Genetic basis of adult migration timing in anadromous steelhead discovered through multivariate association testing. *Proc Biol Sci*. 283:20153064.

Hill WG. 2008. Estimation, effectiveness and opportunities of long term genetic improvement in animals and maize. *Lohmann Information*. 43:3–20.

Hiss JM, Meyer JH, Boomer RS. 1985. *Chehalis spring Chinook progress report 1984*. Olympia (WA): US Fish and Wildlife Service.

Hospital F, Chevalet C. 1996. Interactions of selection, linkage and drift in the dynamics of polygenic characters. *Genet Res*. 67:77–87.

Houlahan JE, Findlay CS, Schmidt BR, Meyer AH, Kuzmin SL. 2000. Quantitative evidence for global amphibian population declines. *Nature*. 404:752–755.

Howell P, Jones K, Scarnecchia D, LaVoy L, Kendra W, Ortmann D. 1985. *Stock assessment of Columbia River anadromous salmonids. Volume I (chinook, coho, chum, and sockeye salmon stock summaries) and Volume II (Steelhead stock summaries, stock transfer guidelines—information needs)*. 1,023 p. Contract No. Portland, OR: DEA179-84BP12737 with US Department of Energy, Bonneville Power Administration, Division of Fish and Wildlife.

Irvine JR, Fukuwaka MA. 2011. Pacific salmon abundance trends and climate change. *ICES J Mar Sci*. 68:1122–1130.

Johnston SE, Gratten J, Berenos C, Pilkington JG, Clutton-Brock TH, Pemberton JM, Slate J. 2013. Life history trade-offs at a single locus maintain sexually selected genetic variation. *Nature*. 502:93–95.

Johnston SE, McEwan JC, Pickering NK, Kijas JW, Beraldi D, Pilkington JG, Pemberton JM, Slate J. 2011. Genome-wide association mapping identifies the genetic basis of discrete and quantitative variation in sexual weaponry in a wild sheep population. *Mol Ecol*. 20:2555–2566.

Jones MR, Mills LS, Alves PC, Callahan CM, Alves JM, Lafferty DJR, Jiggins FM, Jensen JD, Melo-Ferreira J, Good JM. 2018. Adaptive introgression underlies polymorphic seasonal camouflage in snowshoe hares. *Science*. 360:1355–1358.

Kannry SH, O'Rourke SM, Kelson SJ, Miller MR. 2020. On the ecology and distribution of steelhead (*Oncorhynchus mykiss*) in California's Eel River. *J Hered*. 111:548–563.

Kardos M, Luikart G. 2021. The genetic architecture of fitness drives population viability during rapid environmental change. *Am Nat*. 197:511–525.

Kardos M, Shafer ABA. 2018. The peril of gene-targeted conservation. *Trends Ecol Evol*. 33:827–839.

Kinziger AP, Loudenslager EJ, Hankin DG, Anderson EC, and Garza JC. 2008. Hybridization between spring- and fall-run Chinook salmon returning to the Trinity River, California. *North Am J Fish Manage*. 28:5, 1426–1438.

Kirubakaran TG, Grove H, Kent MP, Sandve SR, Baranski M, Nome T, De Rosa MC, Righino B, Johansen T, Otterå H, Sonesson A, Lien S,

Andersen Ø. 2016. Two adjacent inversions maintain genomic differentiation between migratory and stationary ecotypes of Atlantic cod. *Molecular Ecology* 25:2130–2143.

Koch IJ, Narum SR. 2020. Validation and association of candidate markers for adult migration timing and fitness in Chinook salmon. *Evol Appl*. 13:2316–2332.

Kostow K. 2009. Factors that contribute to the ecological risks of salmon and steelhead hatchery programs and some mitigating strategies. *Rev Fish Biol Fish*. 19:9–31.

Kostow K. 2012. Strategies for reducing the ecological risks of hatchery programs: case studies from the Pacific Northwest. *Environ Biol Fish*. 94:285–310.

Lackey RT. 2004. Societal values and the proper role of restoration ecologists. *Front Ecol Environ*. 2:45–46.

Lamichhaney S, Fan G, Widemo F, Gunnarsson U, Thalmann DS, Hoeppner MP, Kerje S, Gustafson U, Shi C, Zhang H, *et al.* 2016a. Structural genomic changes underlie alternative reproductive strategies in the ruff (*Philomachus pugnax*). *Nat Genet*. 48:84–88.

Lamichhaney S, Han F, Berglund J, Wang C, Almén MS, Webster MT, Grant BR, Grant PR, Andersson L. 2016b. A beak size locus in Darwin's finches facilitated character displacement during a drought. *Science*. 352:470–474.

Lande R. 1983. The response to selection on major and minor mutations affecting a metrical trait. *Heredity* 50:47–65.

Langin K. 2018. A different animal. *Science*. 360:590–592.

Lawson PW, Bjorkstedt EP, Chilcote MW, Huntington CW, Mills JS, Moores KM, Nickelson TE, Reeves GH, Stout HA, Wainwright TC, Weitkamp LA. 2007. *Identification of historical populations of coho salmon (Oncorhynchus kisutch) in the Oregon coast evolutionarily significant unit*. NOAA Technical Memorandum NMFS-NWFSC-79. p. 129.

Le Corre V, Kremer A. 2012. The genetic differentiation at quantitative trait loci under local adaptation. *Mol Ecol*. 21:1548–1566.

Linnen CR, Poh YP, Peterson BK, Barrett RD, Larson JG, Jensen JD, Hoekstra HE. 2013. Adaptive evolution of multiple traits through multiple mutations at a single gene. *Science*. 339:1312–1316.

Lord JK. 1866. *The naturalist in Vancouver Island and British Columbia*. Vol. 1. *Elibron Classics*. Boston (MA): Adamant Media Corporation. p. 358.

Lotterhos KE, Whitlock MC. 2014. Evaluation of demographic history and neutral parameterization on the performance of FST outlier tests. *Mol Ecol*. 23:2178–2192.

Mackay TF, Stone EA, Ayroles JF. 2009. The genetics of quantitative traits: challenges and prospects. *Nat Rev Genet*. 10:565–577.

Macqueen DJ, Johnston IA. 2014. A well-constrained estimate for the timing of the salmonid whole genome duplication reveals major decoupling from species diversification. *Proc Biol Sci*. 281:20132881.

McClure MM, Carlson SM, Beechie TJ, Pess GR, Jorgensen JC, Sogard SM, Sultan SE, Holzer DM, Travis J, Sanderson BL, *et al.* 2008. Evolutionary consequences of habitat loss for Pacific anadromous salmonids. *Evol Appl*. 1:300–318.

McClure MM, Holmes EE, Sanderson BL, Jordan CE. 2003. A large-scale, multispecies status assessment: anadromous salmonids in the Columbia River basin. *Ecol Appl*. 13:964–989.

McElhany P, Busack C, Chilcote M, Kolmes S, McIntosh B, Myers J, Rawding D, Steel E, Steward C, Ward D, *et al*. 2006. Revised viability criteria for salmon and steelhead in the Willamette and Lower Columbia Basins. *Review Draft*. April 1, 2006. p. 178.

McElhany P, Ruckelshaus MH, Ford MJ, Wainwright TC, Bjorkstedt EP. 2000. Viable salmonid populations and the recovery of evolutionarily significant units. *US Dept. Commer. NOAA Tech. Memo. NMFS-NWFSC* 42:156.

McKinney G. 2020. Should I stay or should I go? *Science*. 370:526–527.

McKinney GJ, Nichols KM, Ford MJ. 2021. A mobile sex-determining region, male-specific haplotypes and rearing environment influence age at maturity in Chinook salmon. *Mol Ecol*. 30:131–147.

McKinney GJ, Seeb JE, Pascal CE, Schindler DE, Gilk-Baumer SE, Seeb LW. 2020. Y-chromosome haplotypes are associated with variation in size and age at maturity in male Chinook salmon. *Evol Appl*. 13:2791–2806.

Meuwissen TH, Hayes BJ, Goddard ME. 2001. Prediction of total genetic value using genome-wide dense marker maps. *Genetics*. 157:1819–1829.

Micheletti SJ, Hess JE, Zendt JS, Narum SR. 2018. Selection at a genomic region of major effect is responsible for evolution of complex life histories in anadromous steelhead. *BMC Evol Biol*. 18:140.

Mills LS, Allendorf FW. 1996. The one-migrant-per-generation rule in conservation and management. *Conserv Biol*. 10:1509–1518.

Montgomery DR. 2000. Coevolution of the Pacific salmon and Pacific Rim topography. *Geology*. 28:1107–1110.

Moran P, Teel DJ, Banks MA, Beacham TD, Bellinger MR, Blankenship SM, Candy JR, Garza JC, Hess JE, Narum SR, *et al*. 2013 Divergent life-history races do not represent Chinook salmon coast wide: the importance of scale in Quaternary biogeography. *Can J Fish Aquat Sci*. 70:415–435.

Morin J, Royle TC, Zhang H, Speller C, Alcaide M, Morin R, Ritchie M, Cannon A, George M, George M, Yang D. 2021. Indigenous sex-selective salmon harvesting demonstrates pre-contact marine resource management in Burrard Inlet, British Columbia, Canada. *Scientific reports* 11:1–13.

Moritz C. 1994. Defining "evolutionarily significant units" for conservation. *Trends Ecol Evol*. 9:373–375.

Moulton FR, editor. 1939. *The migration and conservation of salmon*. Lancaster (PA): American Association for the Advancement of Science.

Myers JM, *et al*. 1998. Status review of chinook salmon from Washington, Idaho, Oregon, and California. Seattle, WA: U.S. Dept. Commer., NOAA Tech. Memo. NMFS_NWFSC_35.

Myers J, Busack C, Rawding D, Marshall A, Teel D, Van Doornik DM, Maher MT. 2006. *Historical population structure of Pacific salmonids in the Willamette River and lower Columbia River basins*. Seattle, WA: U.S. Dept. Commer., NOAA Tech. Memo. NMFS-NWFSC-73. p. 311.

Narum SR, Banks M, Beacham TD, Bellinger MR, Campbell MR, Dekoning J, Elz A, Guthrie CM 3rd, Kozfkay C, Miller KM, *et al*. 2008. Differentiating salmon populations at broad and fine geographical scales with microsatellites and single nucleotide polymorphisms. *Mol Ecol*. 17:3464–3477.

Narum SR, Buerkle CA, Davey JW, Miller MR, Hohenlohe PA. 2013. Genotyping-by-sequencing in ecological and conservation genomics. *Mol Ecol*. 22:2841–2847.

Narum SR, Di Genova A, Micheletti SJ, Maass A. 2018. Genomic variation underlying complex life-history traits revealed by genome sequencing in Chinook salmon. *Proc Biol Sci*. 285:20180935.

National Research Council. 1996. *Upstream: salmon and society in the Pacific Northwest*. Washington (DC): The National Academies Press.

Nehlsen W, Williams JE, Lichatowich JA. 1991. Pacific salmon at the crossroads: stocks at risk from California, Oregon, Idaho, and Washington. *Fisheries*. 16:4–21.

Nicholas JW, Hankin DG. 1988. *Chinook salmon populations in Oregon coastal river basins: Description of life histories and assessment of recent trends in run strengths*. Salem, OR: Oregon Department of Fish and Wildlife, Fish. Div. Info. Rep., No. 88-1. p. 359.

NMFS (National Marine Fisheries Service). 1991. Notice of policy: policy on applying the definition of species under the Endangered Species Act to Pacific salmon. *Fed Regist*. 56:58612–58618.

NMFS. 1998. Endangered and threatened species: proposed endangered status for two Chinook salmon ESUs and proposed threatened status for five Chinook salmon ESUs; proposed redefinition, threatened status, and revision of critical habitat for one Chinook salmon ESU; proposed designation of Chinook salmon critical habitat in California, Oregon, Washington, Idaho. *Fed Regist*. 63:11482–11520.

NMFS. 2000. Endangered and threatened species: threatened status for one steelhead evolutionarily significant unit (ESU) in California. *Fed Regist*. 65:36074–36094.

NMFS. 2006. Endangered and threatened species: final listing determinations for 10 distinct population segments of west coast steelhead. *Fed Regist*. 71:833–862.

NMFS. 2020. Listing endangered and threatened wildlife and plants; notice of 12-month finding on a petition to list summer-run steelhead in Northern California as endangered under the Endangered Species Act. *Fed Regist*. 85:6527–6531.

NMFS. 2021. Listing endangered and threatened wildlife; 12-month findings on petitions to list spring-run Oregon Coast Chinook salmon and spring-run Southern Oregon and Northern California Coastal Chinook salmon as threatened or endangered under the Endangered Species Act. *Fed Regist*. 86:45970–45974.

ODFW (Oregon Department of Fish and Wildlife). 2000. *Effects of Lost Creek Dam on spring Chinook salmon in the Rogue River*. Salem (OR): Oregon Dept of Fish and Wildlife (Phase II completion report).

Olson AD, Dix OJ. 1993. *Lower Salmon River sub-basin fish habitat condition and utilization assessment 1990/1991 final report*. Yreka, CA: USDA—Forest Service, Klamath National Forest.

Olsen JB, Flannery BG, Beacham TD, Bromaghin JF, Crane PA, Lean CF, Dunmall KM, Wenburg JK. 2008. The influence of hydrographic structure and seasonal run timing on genetic diversity and isolation-by-distance in chum salmon (*Oncorhynchus keta*). *Can J Fish Aquat Sci*. 65:2026–2042.

O'Malley KG, Banks MA. 2008. A latitudinal cline in the Chinook salmon (Oncorhynchus tshawytscha) Clock gene: evidence for selection on PolyQ length variants. *Proceedings of the Royal Society B: Biological Sciences*. 275:2813–2821.

O'Malley KG, Mazur S, Green L, Bohn S, Wells A. 2020a. Evaluating the genetics of naturally produced Chinook salmon (*Oncorhynchus tshawytscha*) captured in the Lower Rogue River, Oregon fishery. Information report 2020-02. Salem (OR): Oregon Department of Fish and Wildlife. Available from: https://nrimp.dfw.state.or.us/web%20stores/data%20libraries/files/ODFW/ODFW_42004_2_O'Malley%20et%20al.%20(2020)%20ODFW%20Information%20Report%202020-02.pdf

O'Malley KG, Van Dyke D, Samarin PA, Bohn S, Clements S. 2020b. An evaluation of "early" and "late" run alleles in Rogue River Chinook salmon (*Oncorhynchus tshawytscha*). Information report 2020-06. Salem (OR): Oregon Department of Fish and Wildlife. Available from: https://nrimp.dfw.state.or.us/web%20stores/data%20libraries/files/ODFW/ODFW_42006_2_O'Malley%20et%20al.%20(2020)%20ODFW%20Information%20Report%202020-06.pdf

Oomen RA, Kuparinen A, Hutchings JA. 2020. Consequences of single-locus and tightly linked genomic architectures for evolutionary responses to environmental change. *J Hered*. 111:319–332.

Pearse DE. 2016. Saving the spandrels? Adaptive genomic variation in conservation and fisheries management. *J Fish Biol*. 89:2697–2716.

Pearse DE, Barson NJ, Nome T, Gao G, Campbell MA, Abadía-Cardoso A, Anderson EC, Rundio DE, Williams TH, Naish KA, *et al*. 2019a. Sex-dependent dominance maintains migration supergene in rainbow trout. *Nat Ecol Evol*. 3:1731–1742.

Pearse DE, Garza JC, Myers J, Spence B. 2019b. Northern California steelhead DPS-Configuration Review-Panel Report. Available from: https://www.fisheries.noaa.gov/action/12-month-finding-petition-list-summer-run-steelhead-northern-california-endangered-under

Pess GR, Quinn TP, Gephard SR, Saunders R. 2014. Re-colonization of Atlantic and Pacific rivers by anadromous fishes: linkages between life history and the benefits of barrier removal. *Rev Fish Biol Fish*. 24:881–900.

Pitman KJ, Moore JW, Sloat MR, Beaudreau AH, Bidlack AL, Brenner RE, Hood EW, Pess GR, Mantua NJ, Milner AM, *et al*. 2020. Glacier retreat and Pacific salmon. *Bioscience*. 70:220–236.

Powers S 1877. *Tribes of California. Department of the Interior. U.S. Geographical and Geological Survey of the Rocky Mountain Region*. Washington, D.C.: Government Printing Office. p. 740.

Primmer CR. 2009. From conservation genetics to conservation genomics. *Ann NY Acad Sci*. 1162:357–368.

Prince DJ, O'Rourke SM, Thompson TQ, Ali OA, Lyman HS, Saglam IK, Hotaling TJ, Spidle AP, Miller MR. 2017. The evolutionary basis of premature migration in Pacific salmon highlights the utility of genomics for informing conservation. *Sci Adv*. 3:e1603198.

Pritchard JK, Pickrell JK, Coop G. 2010. The genetics of human adaptation: hard sweeps, soft sweeps, and polygenic adaptation. *Curr Biol*. 20:R208–R215.

Quinn TP. 2018. *The behavior and ecology of Pacific salmon and trout*. 2nd ed. Seattle: University of Washington Press.

Quinn TP, Bond MH, Brenkman SJ, Paradis R, Peters RJ. 2017. Re-awakening dormant life history variation: stable isotopes indicate anadromy in bull trout following dam removal on the Elwha River, Washington. *Environ Biol Fishes*. 100:1659–1671.

Quinn TP, McGinnity P, Reed TE. 2016. The paradox of "premature migration" by adult anadromous salmonid fishes: patterns and hypotheses. *Can J Fish Aquat Sci*. 73:1015–1030.

Quinn TP, Unwin MJ, Kinnison MT. 2000. Evolution of temporal isolation in the wild: genetic divergence in timing of migration and breeding by introduced Chinook salmon populations. *Evolution*. 54:1372–1385.

Quinn TP, Unwin MJ, Kinnison MT. 2011. Contemporary divergence in migratory timing of naturalized populations of Chinook salmon, *Oncorhynchus tshawytscha*, in New Zealand. *Evol Ecol Res*. 13:45–54.

Ralls K, Sunnucks P, Lacy RC, Frankham R. 2020. Genetic rescue: a critique of the evidence supports maximizing genetic diversity rather than minimizing the introduction of putatively harmful genetic variation. *Biol Conserv*. 251:108784.

Rich WH, Holmes HB 1928. Experiments in marking young Chinook salmon on the Columbia River, 1916 to 1927. *US Bur Fish Bull*. 44:215–264.

Rojas M. 1992. The species problem and conservation: what are we protecting? *Conserv Biol*. 6:170–178.

Ruckelshaus MH, Currens KP, Graeber WH, Fuerstenberg RR, Rawson K, Sands NJ, Scott JB. 2006. *Independent populations of Chinook salmon in Puget Sound*. Seattle, WA: NOAA Technical NMFS-NWFSC-78 Memorandum. p. 125.

Ryder OA. 1986. Species conservation and systematics: the dilemma of subspecies. *Trends Ecol Evol*. 1:9–10.

Sanderson MJ, Shaffer HB. 2002. Troubleshooting molecular phylogenetic analyses. *Annu Rev Ecol Syst*. 33:49–72.

Savolainen O, Lascoux M, Merilä J. 2013. Ecological genomics of local adaptation. *Nat Rev Genet*. 14:807–820.

Schielzeth H, Husby A. 2014. Challenges and prospects in genome-wide quantitative trait loci mapping of standing genetic variation in natural populations. *Ann NY Acad Sci*. 1320:35–57.

Schielzeth H, Kempenaers B, Ellegren H, Forstmeier W. 2012. QTL linkage mapping of zebra finch beak color shows an oligogenic control of a sexually selected trait. *Evolution*. 66:18–30.

Scott JM, Goble DD, Davis FW, editors. 2006. The Endangered Species Act at thirty: conserving biodiversity in human-dominated landscapes. Washington (DC): Island Press.

Seeb LW, Antonovich A, Banks MA, Beacham TD, Bellinger MR, Blankenship SM, Campbell MR, Decovich NA., Garza, J.C., Guthrie Iii, C.M. and Lundrigan, T.A., 2007. Development of a standardized DNA database for Chinook salmon. *Fisheries*. 32:540–552.

Sella G, Barton NH. 2019. Thinking about the evolution of complex traits in the era of genome-wide association studies. *Annu Rev Genomics Hum Genet*. 20:461–493.

Shapovalov L, Taft AC. 1954. The life histories of the steelhead rainbow trout (*Salmo gairdneri gairdneri*) and silver salmon (*Oncorhynchus kisutch*) with special reference to Waddell Creek, California and recommendations regarding their management. *Calif Dep Fish Game Fish Bull*. 98:275.

Shared Strategy Development Committee. 2007. Puget Sound salmon recovery plan. Vol. 1. Plan adopted by the National Marine Fisheries Service (NMFS) January 19, 2007. Available from: http://www.westcoast.fisheries.noaa.gov/protected_species/salmon_steelhead/recovery_planning_and_implementation/puget_sound/puget_sound_chinook_recovery_plan.html

Shirk AJ, Landguth EL, Cushman SA. 2017. A comparison of individual-based genetic distance metrics for landscape genetics. *Mol Ecol Resour*. 17:1308–1317.

Sinclair-Waters M, Ødegård J, Korsvoll SA, Moen T, Lien S, Primmer CR, Barson NJ. 2020. Beyond large-effect loci: large-scale GWAS reveals a mixed large-effect and polygenic architecture for age at maturity of Atlantic salmon. *Genet Sel Evol*. 52:9.

Skovmand LH, Xu CCY, Servedio MR, Nosil P, Barrett RDH, Hendry AP. 2018. Keystone Genes. *Trends Ecol Evol*. 33:689–700.

Smith SB 1969. Reproductive isolation in summer and winter races of steelhead trout. In: Northcote TG, editor. *Proceedings of the H. R. MacMillan Lectures Symposium on Salmon and Trout in Streams, February 22–24, 1968*, Vancouver (Cananda): Institute of Fisheries, Univ. British Columbia, Vancouver. p. 21–38.

Smith GR. 1992. Introgression in fishes: significance for paleontology, cladistics, and evolutionary rates. *Syst Biol*. 41:41–57.

Spence BC, Bjorkstedt EP, Garza JC, Smith JJ, Hankin DG, Fuller DW, Jones WE, Macedo R, Williams TH, Mora E. 2008. *A framework for assessing the viability of threatened and endangered salmon and steelhead in the North-Central California Coast Recovery Domain*. NOAA Technical Memorandum NMFS-SWFSC-423. p. 173.

Stearley RF. 1992. Historical ecology of Salmoninae, with special reference to *Oncorhynchus*. In Mayden RL, editor. *Systematics, historical ecology, and North American freshwater fishes*. Stanford (CA): Stanford University Press. pp. 622–658.

Supple MA, Shapiro B. 2018. Conservation of biodiversity in the genomics era. *Genome Biol*. 19:131.

Swezey SL, Heizer RF. 1977. Ritual management of salmonid fish resources in California. *J Calif Anthropol*. 4:6–29.

Teixeira JC, Huber CD. 2021. The inflated significance of neutral genetic diversity in conservation genetics. *Proc Natl Acad Sci USA*. 118:e2015096118.

Thompson WF. 1959. An approach to population dynamics of the Pacific red salmon *Trans Am Fish Soc*. 88:206–209.

Thompson NF, Anderson EC, Clemento AJ, Campbell MA, Pearse DE, Hearsey JW, Kinziger AP, Garza JC. 2020. A complex phenotype in salmon controlled by a simple change in migratory timing. *Science*. 370:609–613.

Thompson TQ, Bellinger MR, O'Rourke SM, Prince DJ, Stevenson AE, Rodrigues AT, Sloat MR, Speller CF, Yang DY, Butler VL, *et al.* 2019a. Anthropogenic habitat alteration leads to rapid loss of adaptive variation and restoration potential in wild salmon populations. *Proc Natl Acad Sci USA*. 116:177–186.

Thompson TQ, O'Rourke SM, Brown SK, Seamons T, Zimmerman M, Miller MR. 2019b. *Run-type genetic markers and genomic data provide insight for monitoring spring-run Chinook Salmon in the Chehalis Basin, WDFW contract #18-11697*. Final report submitted to Washington Department of Fish and Wildlife. p. 26.

Thorgaard GH. 1983. Chromosomal differences among rainbow trout populations. *Copeia*. 1983:650–662.

Thorpe JE, Mangel M, Metcalfe NB, Huntingford FA. 1998. Modelling the proximate basis of salmonid life-history variation, with application to Atlantic salmon, *Salmo salar* L. *Evol Ecol*. 12:581–599.

Tillotson MD, Arostegui MC, Austin CS, Lincoln AE, Matsubu W, McElroy KN, Quinn TP. 2021. Challenges in the identification and interpretation of phenological shifts: anthropogenic influences on adult migration timing in salmonids. Rev Fish Sci Aquac. 29:769–790.

Todesco M, Owens GL, Bercovich N, Légaré JS, Soudi S, Burge DO, Huang K, Ostevik KL, Drummond EBM, Imerovski I, *et al.* 2020. Massive haplotypes underlie ecotypic differentiation in sunflowers. *Nature*. 584:602–607.

Utter FM, Allendorf FW, Hodgins HO. 1973. Genetic variability and relationships in Pacific salmon and related trout based on protein variations. *Syst Zool*. 22:257–270.

Visscher PM, Brown MA, McCarthy MI, Yang J. 2012. Five years of GWAS discovery. *Am J Hum Genet*. 90:7–24.

Walsh B, Lynch M. 2018, *Evolution and selection of quantitative traits*. Oxford, UK: Oxford University Press.

Wang M, Xu S. 2019. Statistical power in genome-wide association studies and quantitative trait locus mapping. *Heredity (Edinb)*. 123:287–306.

Waples RS. 1991. Pacific salmon, *Oncorhynchus* spp., and the definition of "species" under the Endangered Species Act. *Mar Fish Rev*. 53:11–22.

Waples RS. 1995. Evolutionarily significant units and the conservation of biological diversity under the Endangered Species Act. *Am Fish Soc Symp*. 17:8–27.

Waples RS. 2006. Distinct population segments. In: Scott JM, Goble DD, Davis FW, editors. *The Endangered Species Act at thirty: conserving biodiversity in human-dominated landscapes*. Washington (DC): Island Press. p. 127–149.

Waples RS, Beechie T, Pess GR. 2009. Evolutionary history, habitat disturbance regimes, and anthropogenic changes: what do these mean for resilience of Pacific salmon populations? *Ecol Soc*. 14:3. http://www.ecologyandsociety.org/vol14/iss1/art3/

Waples RS, Gustafson RG, Weitkamp LA, Myers JM, Johnson O.W, Busby PJ, Hard JJ, Bryant GJ, Waknitz FW, Neely K, Teel D, Grant WS, Winans GA, Phelps S, Marshall A, Baker B. 2001. Characterizing diversity in Pacific salmon. *J. Fish Biol*. 59(Suppl A):1–41.

Waples RS, Lindley ST. 2018. Genomics and conservation units: the genetic basis of adult migration timing in Pacific salmonids. *Evol Appl*. 11:1518–1526.

Waples RS, Naish KA, Primmer CR. 2020. Conservation and management of salmon in the age of genomics. *Annu Rev Anim Biosci*. 8:117–143.

Waples RS, Pess GR, Beechie T. 2008. Evolutionary history of Pacific salmon in dynamic environments. *Evol Appl*. 1:189–206.

Waples RS, Teel DJ, Myers JM, Marshall AR. 2004. Life-history divergence in Chinook salmon: historic contingency and parallel evolution. *Evolution*. 58:386–403.

Waters CD, Clemento A, Aykanat T, Garza JC, Naish KA, Narum S, Primmer CR. 2021. Heterogeneous genetic basis of age at maturity in salmonid fishes. *Mol Ecol*. 30:1435–1456.

WDF, WDG, and WWTIT (Washington Dept. Fisheries, Washington Dept. Game, and Western Washington Treaty Indian Tribes). 1993. *1992 Washington salmon and steelhead stock inventory (SASSI)*. Olympia: Washington Dept. Fisheries.

Weitkamp LA, Wainwright TC, Bryant GJ, Milner GB, Teel DJ, Kope RG, Waples RS. 1995. *Status review of Coho salmon from Washington, Oregon, and California*. U.S. Dep. Commer., NOAA Tech. Memo. Seattle, WA: NMFS-NWFSC-24. p. 258.

Willis SC, Hess JE, Fryer JK, Whiteaker JM, Brun C, Gerstenberger R, Narum SR. 2020. Steelhead (*Oncorhynchus mykiss*) lineages and sexes show variable patterns of association of adult migration timing and age-at-maturity traits with two genomic regions. *Evol Appl*. 13:2836–2856.

Willis SC, Hess JE, Fryer JK, Whiteaker JM, Narum SR. 2021. Genomic region associated with run timing has similar haplotypes and phenotypic effects across three lineages of Chinook salmon. *Evol Appl*. 14:2273–2285.

Wilson WD, Turner TF. 2009. Phylogenetic analysis of the Pacific cutthroat trout (*Oncorhynchus clarki* ssp.: Salmonidae) based on partial mtDNA ND4 sequences: a closer look at the highly fragmented inland species. *Mol Phylogenet Evol*. 52:406–415.

Withler F. 1982. Transplanting Pacific salmon. *Can Tech Rep Fish Aquat Sci*. 1079:27.

Withler IL. 1966 Variability in life history characteristics of steelhead trout (*Salmo gairdneri*) along the Pacific coast of North America. *J Fish Res Board Can*. 23:365–393.

Wood AR, Esko T, Yang J, Vedantam S, Pers TH, Gustafsson S, Chu AY, Estrada K, Luan J, Kutalik Z, *et al.*; Electronic Medical Records and Genomics (eMEMERGEGE) Consortium; MIGen Consortium; PAGEGE Consortium; LifeLines Cohort Study. 2014. Defining the role of common variation in the genomic and biological architecture of adult human height. *Nat Genet*. 46:1173–1186.

Wood CC, Foote CJ. 1996. Evidence for sympatric genetic divergence of anadromous and nonanadromous morphs of sockeye salmon (*Oncorhynchus nerka*). *Evolution*. 50:1265–1279.

Wright S. 1943. Isolation by distance. *Genetics*. 28:114–138.

Xuereb A, d'Aloia CC, Andrello M, Bernatchez L, Fortin MJ. 2021. Incorporating putatively neutral and adaptive genomic data into marine conservation planning. *Conservation Biology* 35:909–920.

Zhan S, Zhang W, Niitepõld K, Hsu J, Haeger JF, Zalucki MP, Altizer S, de Roode JC, Reppert SM, Kronforst MR. 2014. The genetics of monarch butterfly migration and warning colouration. *Nature*. 514:317–321.