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Perspective

A practical introduction to effective population size for fisheries management

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ABSTRACT

Objective: So far, many of the texts describing effective population size (N_e) offer technical descriptions that limit accessibility to nonspecialized audiences, hindering widespread uptake into natural resource management and biodiversity conservation. Our aim is to provide a practical introduction to N_e that is accessible to a wide audience, especially natural resource managers and other practitioners who lead decision-making efforts in fisheries contexts.

Methods: We summarize key concepts related to measuring and monitoring N_e in managed fish populations. We primarily focus on widely used linkage disequilibrium methods for measuring contemporary N_e. Among other topics, we review the value of N_e for conservation and management, basic assumptions of this parameter, common estimation methods, important study design considerations, and representative empirical examples. Additionally, we use simulations to explore two practical elements of studies aimed at inferring N_e: identifying an appropriate number of individuals to sample per population and a suitable number of molecular markers to survey across populations.

Results: Using accessible language, we illuminate the important role N_e plays in conserving and managing wild fish populations and describe practical elements related to estimating this parameter. Further, our simulations demonstrate the relationship among numbers of surveyed individuals and molecular markers on the accuracy and precision of N_e estimates, highlighting critical elements in the design of studies to measure N_e.

Conclusions: Together with more widely appreciated and implemented metrics, N_e forms a vital part of a complete tool kit for conserving and managing wild populations, including in fisheries contexts. While not a comprehensive review, our aim is that by providing a practical introduction to the complex concepts underlying N_e, we will facilitate increased conversation among practitioners with diverse areas of expertise and ultimately support expanded integration of this and related parameters into fisheries conservation and management efforts.

KEYWORDS: effective population size, fish conservation, fisheries management, genetic monitoring, natural resource management

LAY SUMMARY

For complex and nuanced topics like effective population size (N_e), nontechnical descriptions aimed at increasing accessibility to wide-ranging audiences are critical for expanding the role of genetic monitoring to support decision making in fisheries contexts.

INTRODUCTION

Effective population size (N_e; see Table 1 for definitions of terms used throughout this article) is one of the most important parameters in conservation biology (Allendorf & Luikart, 2007; Charlesworth, 2009; Luikart et al., 2010; J. Wang et al., 2016; R. S. Waples, 2022). Effective population size offers vital information on the conservation status and evolutionary trajectory of populations (Hare et al., 2011; R. S. Waples

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Table 1. Glossary of terms. The definitions provided here are largely adapted from Allendorf et al. (2022).

Term	Definition
Adaptive capacity	The capacity to evolve genetic-based changes in traits to respond to changing environmental conditions.
Alleles	Alternative forms of a gene; the different nucleotide variants found at a specific location in the genome.
Gene flow	The movement of genetic material from one population to another via migration followed by interbreeding.
Genetic drift	Changes to allele frequencies within a population due to random chance; such stochastic changes are more prevalent in small populations.
Genotype	The genetic composition of an organism; the alleles associated with a specific location in the genome.
Ideal population	A constant-size population in which external evolutionary forces are not present and individuals contribute equally to the next generation.
Inbreeding depression	The relative reduction in fitness in the progeny of related individuals compared to the progeny of unrelated individuals.
Linkage disequilibrium	Nonrandom associations (e.g., linkages) among alleles at different loci; the degree to which an allele at one locus is inherited together with an allele at another locus (e.g., linked loci). Sometimes referred to as "gametic disequilibrium" to distinguish from instances of unlinked loci that exhibit linkage disequilibrium.
Locus (plural: loci)	One (locus) or multiple (loci) molecular markers; a specific location or set of locations within the genome, including the location of a gene on a chromosome or a single nucleotide polymorphism within the genome.
Microsatellite	A short segment of DNA comprising a motif of two or more nucleotides, which is repeated several times; a type of molecular marker useful for assessing genetic variation within and among populations.
Mutation	Changes to genetic material that result in a new form; occurs via the altering of a single or multiple nucleotides.
N_b	Effective number of breeders; the number of breeding individuals that gave rise to a single cohort.
N_c	Population census size; the number of individuals in a population.
N_e	Effective population size; the size of the ideal population that would experience the same amount of genetic drift as the observed population.
Nucleotide	The molecular building blocks of DNA and RNA.
Physical linkage	Nonrandom associations among alleles at different loci due to the physical proximity of loci on a chromosome, where nearby loci are more likely to be inherited together.
Selection	Differential contributions of genotypes to the next generation due to differences in survival and reproduction.
Single nucleotide polymorphism (SNP)	A single nucleotide that exhibits different nucleotide variants; a type of molecular marker useful for assessing genetic variation within and among populations.

et al., 2014), primarily their degree of susceptibility to genetic diversity loss (Leroy et al., 2018; Luikart & Cornuet, 1999; Luikart et al., 2010). However, N_e is a complex and nuanced parameter and understanding how best to measure and monitor N_e requires specialized knowledge of key genetic principles based on diverse ecological and evolutionary relationships. For example, several analytical methods exist for inferring N_e (Luikart & Cornuet, 1999; Nadachowska-Brzyska et al., 2022; R. S. Waples, 2022; Whiteley et al., 2012; Wright, 1931), each with their own assumptions and constraints, making it difficult for nonexperts to gauge their utility and develop appropriate study designs. So far, many of the texts describing N_e primarily rely on technical language and in-depth explorations of specialized topics, limiting accessibility to audiences with differing areas of expertise. Collectively, these factors present barriers to a broader understanding of N_e and development of studies aimed at inferring this parameter, limiting the availability of important information about N_e to conservation and management practitioners.

Despite the challenges associated with measuring N_e, investigations of this concept in the context of fisheries management have yielded a rich repository of information to guide investigations in aquatic systems. For example, pioneering studies of N_e in populations of Pacific salmon have provided foundational knowledge on theoretical models, statistical methods, and experimental designs for estimating this parameter (R. S. Waples, 1990a, 1990b; R. S. Waples & Teel, 1990). Such studies have also elucidated important relationships among N_e, migration, genetic drift, and selection, as well as insights into the susceptibility of populations to genetic change relative to N_e (Gilbert & Whitlock, 2015; Novo et al., 2022; Ryman et al., 1981; Santiago & Caballero, 1995; R. S. Waples & England, 2011). Knowledge gained from studies of salmon and other species offers valuable guidance for inferring and monitoring N_e in a diversity of managed fish populations.

Our goal is to provide an accessible, nontechnical introduction to N_e for a wide audience but especially natural resource managers and other practitioners who lead decision-making efforts in fisheries contexts. Rather than provide a comprehensive review of the many topics related to N_e, we aim to create a resource that facilitates conversations among practitioners with wide-ranging expertise by highlighting the primary components of N_e. We accomplish this by (1) summarizing foundational topics related to N_e, including its definition, value, basic assumptions, related metrics, study design implications, and conservation applications, and (2) supplementing our review with simulation-based inferences aimed at exploring questions central to the development of effective study designs for measuring N_e. By drawing upon a wealth of information derived from comparatively well-studied systems, we endeavor to support broader understanding and engagement on studies of N_e while keeping content applicable to a range of fishery species. The key concepts described below are also summarized in Appendix A for convenient reference.

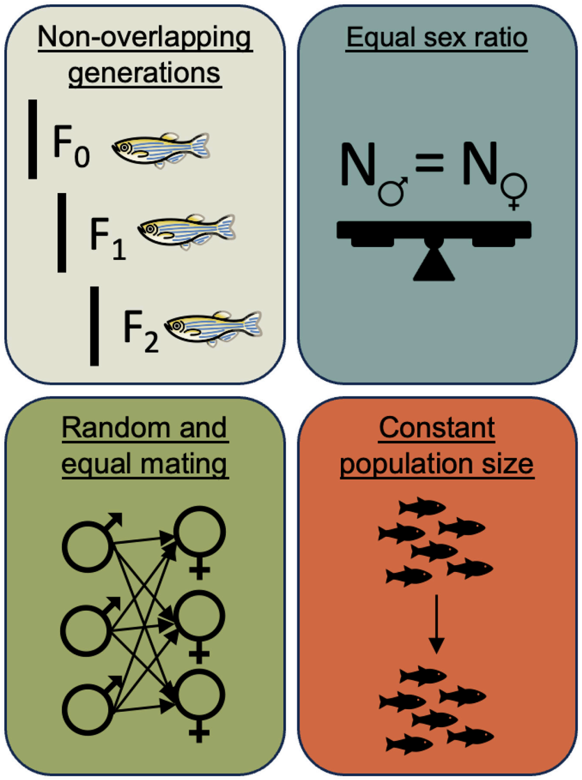
WHAT IS N_e AND WHY IS IT IMPORTANT?

N_e overview

Effective population size was introduced by Wright (1931) as the size of an ideal population (Box 1) that follows the same rate of genetic drift as the population under study. This parameter is thus inferred at the population level and reflects the rate of genetic drift within a population, which is particularly important to aquatic conservation and fisheries management given the potential for genetic drift to increase inbreeding and reduce genetic diversity. Effective population size is the evolutionary analog of census population size (N_c) and reflects the susceptibility of a population to neutral and selective evolutionary forces that increase genetic drift and cause genetic diversity to be lost over time (R. S. Waples et al., 2014; R. S. Waples, 2022). The rate at which this loss occurs depends on N_e rather than N_c, making N_e a parameter of fundamental importance. Populations with lower N_e exhibit greater susceptibility to genetic drift and the negative effects of genetic diversity loss, including inbreeding depression, deleterious mutations, and diminished adaptive capacity.

Box 1. Ideal population

The concept of an ideal population provides a simplified model for understanding basic population genetic principles and dynamics (Wright, 1931). In an ideal population, it is assumed that evolutionary mechanisms that affect the frequencies of alleles within a population are not occurring, and therefore, N_e is equal to N_c. Deviations from an ideal population result in an N_e that is different from N_c. Key assumptions of an ideal population include nonoverlapping generations, an equal sex ratio, random mating among individuals where all individuals are equally likely to reproduce, and equal reproductive output among individuals, resulting in a constant population size across generations. In an ideal population, all individuals have an equal opportunity to pass their genes on to the next generation. This idealized model provides a theoretical baseline for understanding genetic changes without the complexities of the real world, even if if these assumptions are rarely if ever met in nature.



Box Figure 1.1. Schematic depicting the key assumptions of an ideal population, a concept which provides a useful model for evaluating population-level genetic relationships.

Effective population size bridges the gap between the ecological and evolutionary processes affecting populations. Although N_c is broadly governed by ecological processes such as predation, competition, and population growth rate (Rockwood, 2015), these demographic parameters are inextricably linked to the underlying genotypes of the individuals comprising a population (Leffler et al., 2012); therefore, both ecological and evolutionary factors are important determinants of population-level conservation status. Effective populatoin size reflects how the major evolutionary forces (genetic drift, selection, mutation, and gene flow) shape populations (R. S. Waples, 2022; Whitlock, 2000). Considering these ecological and evolutionary factors together is thus vital for developing a comprehensive approach for monitoring the conservation status of populations.

In this context, N_e offers critical insights into evolutionary processes affecting wild populations and their demography. Effective population size is usually smaller than N_c because wild populations often violate the assumptions of an ideal population so that individuals do not contribute equally to the next generation (Box 1). However, N_e and N_c are often correlated (Kalinowski & Waples, 2002; Luikart et al., 2010; Shrimpton & Heath, 2003; Vucetich et al., 1997), and declines in N_c frequently correspond with declines in N_e. Effective population size and N_c have prescient implications in fisheries management given that lower values for these metrics generally indicate greater susceptibility of populations to demographic stochasticity and the negative effects of genetic drift. However, discordance between N_e and N_c is frequent; for example, in extensively harvested populations that exhibit high N_c but low N_e (Nadachowska-Brzyska et al., 2022; Palstra & Ruzzante, 2008). Similarly, many marine fishes exhibit small N_e to N_c ratios (Hoban et al., 2020). In some cases, N_c is difficult and resource-intensive to measure with adequate precision in wild fish populations (Fraser et al., 2013; Guschanski et al., 2009). Further, N_c alone does not reflect the sensitivity of a population to genetic drift and inbreeding depression or its ability to adapt to changing conditions. Inferences about these important evolutionary factors instead depend on N_e (Kalinowski & Waples, 2002; R. S. Waples, 2022; Whitlock, 2000), making N_e a critical tool for monitoring the health and persistence of managed fish populations.

HOW IS N_e MEASURED?

Prior to the now widespread availability of genetic data, early studies estimated N_e based on demographic information (Caballero, 1994; Lee et al., 2011; Nunney, 1995; Vucetich & Waite, 1998; R. S. Waples et al., 2013); however, the use of genetic data to estimate N_e is now commonplace. Since Wright's

Box 2. Methodological approaches for estimating effective population size

Effective population size (N_e) is notoriously difficult to accurately and precisely measure in wild populations, and efforts aimed at developing and applying new methods for inferring this parameter remain an active area of research. Currently available methods for estimating N_e primarily differ in the way they quantify genetic drift and in their assumptions about underlying population demography. For example, Wright's original formulation of N_e (Equation B1 in Appendix B) is based on the expected increase in common ancestry over one generation that results from genetic drift, where increasing common ancestry is reflective of increased inbreeding (and therefore increased homozygosity and decreased heterozygosity; Wright, 1931). This formulation of N_e is known as inbreeding N_e (Caballero & Hill, 1992; Crow & Denniston, 1988; Wright, 1931. In comparison, variance N_e (Equation B2 in Appendix B) is based on the expected increase in the variance of allele frequencies from one generation to the next due to genetic drift (Crow, 1954; Crow & Denniston, 1988; Hill, 1972; Kimura & Crow, 1963). However, temporal sampling is required to estimate variance N_e. Effective population size based on linkage disequilibrium (Equation B3 in Appendix B) measures the expected increase in correlations (e.g., linkages) of allele frequencies among loci due to genetic drift (Hill, 1981; R. S. Waples, 2006a; R. S. Waples & Do, 2010; for a comprehensive review of the linkage disequilbrium method for inferring N_e, see R. S. Waples, 2024a). Several additional definitions and estimators of N_e exist, including single sample estimators that incorporate information on sibship frequency (J. Wang, 2009; for comparisons to estimators based on linkage disequilibrium, see J. Wang, 2016, 2025; R. S. Waples, 2021), heterozygote excess (Luikart & Cornuet, 1999; Pudovkin et al., 1996, 2010), or molecular coancestry (Nomura, 2008). Although we focus on linkage disequilibrium N_e in this review, measures of N_e that quantify the rate of allele frequency change (e.g., variance N_e) and the accrual of shared ancestry (e.g., inbreeding N_e) are also important to fish conservation and fisheries management given that they also reflect relevant consequences of the genetic drift process. More detailed descriptions and comparisons of methods for estimating N_e can be found in Gilbert and Whitlock (2015) and Ryman et al. (2019, 2023).

original description of N_e, several formulations of this parameter have been developed that measure genetic drift in different ways (Box 2; Appendix B). These include genetic methods for inferring historical versus contemporary N_e (Box 3); in this review, we focus on genetic methods to assess contemporary N_e (see Nadachowska-Brzyska et al. [2022] for a review of methods for historical N_e). Contemporary estimators of N_e largely fall into two categories: (1) temporal methods (Krimbas & Tsakas, 1971; Nei & Tajima, 1981; J. Wang & Whitlock, 2003; R. S. Waples, 1989), which require at least two sampling events from the same population at different points in time, and (2) single-sample methods (Hill, 1981; Nomura, 2008; Pudovkin et al., 1996; R. S. Waples & Do, 2010), which are based on a single sampling event per population. Below, we provide a brief description of temporal methods for estimating N_e, but our primary focus is contemporary estimates of N_e based on single sample methods, particularly those that incorporate information on linkage disequilibrium.

Methodological approaches

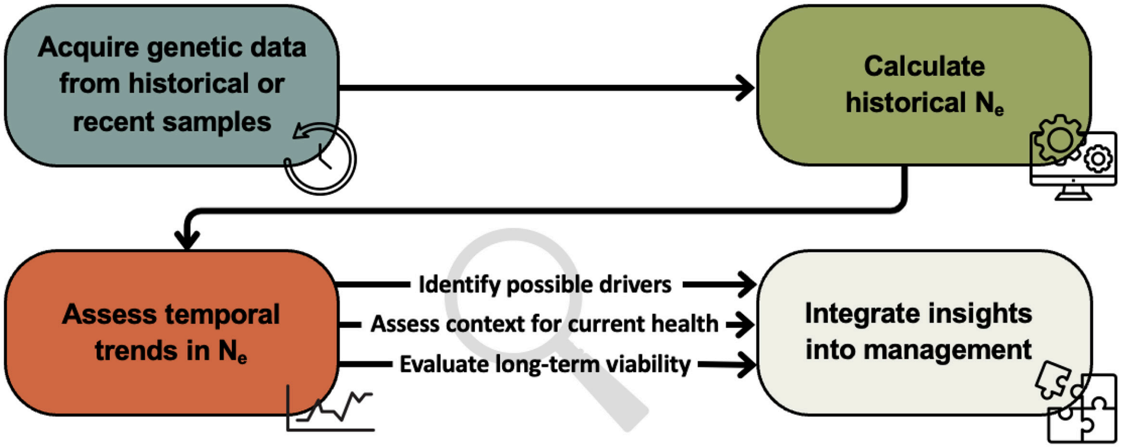
Temporal methods infer N_e based on changes in allele frequencies over time and involve calculating N_e using individuals sampled from a population at two or more time points separated by at least one generation, although a sampling interval of more than one generation is required for species with overlapping generations (Nei & Tajima, 1981). One potential benefit of temporal methods is the ability to account for fluctuating population size across generations (R. S. Waples, 1989). However, because repeated sampling of wild populations is often impractical, especially for rare species or those with long generation times, single-sample methods are more widely used. Additionally, in some scenarios, N_e cannot be reliably inferred using temporal methods (e.g., if the sampling interval is much longer than the interval over which N_e declined; J. Wang et al., 2016).

Several single-sample estimators of N_e exist and implement different approaches for measuring varying aspects of the genetic drift process (Box 2; Appendix B). Currently, one of the most widely implemented and evaluated single-sample methods for inferring contemporary N_e in populations targeted for conservation or management is the estimator based on linkage disequilibrium (Hill, 1981; R. S. Waples, 2006a; R. S. Waples & Do, 2010; Box 2). Linkage disequilibrium refers to nonrandom associations among genetic markers, which produce correlations in the frequencies of alleles across markers (Slatkin, 2008). Populations with lower N_e experience higher levels of genetic drift and exhibit greater linkage disequilibrium among loci. Measures of N_e based on linkage disequilibrium estimate N_e by quantifying the level of disequilibrium across genetic markers. In this context, linkage disequilibrium is assumed to result from genetic drift, though in reality other contributing factors may exist, including random deviations from expected levels due to sampling error or physical linkage among genetic markers (see sections on Marker Selection and How Is N_e Used in Fisheries Management?). Estimates of N_e inferred using linkage disequilibrium methods are presumed to correspond with the previous one-to-few generations (Luikart et al., 2010; R. S. Waples, 2005), offering contemporary insights into the recent genetic health of a population.

For the remainder of this review, we focus on inferences of contemporary N_e derived from linkage disequilibrium methods (and therefore single sampling events). These methods are well developed (R. S. Waples, 2024b, 2025) and have been integrated into broadly used software programs (e.g., NeEstimator [Do et al., 2014; Ovenden et al., 2007; R. S. Waples & Do, 2008], SNeP [Barbato et al., 2015], and LinkNe [Hollenbeck et al., 2016]; see Appendix C for a list of programs useful for estimating N_e). Additionally, estimates of N_e based on linkage disequilibrium have been widely applied to monitor the genetic health of wild populations (R. S. Waples, 2024b, 2025), including in the context of fisheries management. However, despite

Box 3. Historical versus contemporary effective population size

Estimates of historical effective population size (N_e) provide information on the past N_e of a population. Historical N_e can be calculated from population samples collected from historic or contemporary sampling efforts. Several methodologies exist for inferring historical N_e from contemporary samples (see review by Nadachowska-Brzyska et al., 2022), all of which require simplifying assumptions that may or may not accurately reflect the evolutionary history of a population depending on the data analyzed, methodology employed, and biological system. Nonetheless, it is useful to estimate historical N_e to explore temporal trends and provide historical context for estimates of contemporary N_e. Quantifying trends in N_e over time can help identify environmental or evolutionary factors that potentially underlie population fluctuations, such as genetic bottlenecks. Estimates of historical N_e also provide a more complete picture of the long-term health and viability of a population. For example, Lehnert et al. (2019) used contemporary samples of Atlantic Salmon *Salmo salar* to infer the historical N_e of populations across the native species range. The authors identified several populations that exhibited significant declines in N_e over time, as well as climate variables associated with observed declines. Historical estimates of N_e produced from contemporary samples were validated by making comparisons to estimates of N_e derived from a small number of historical sample collections.



Box Figure 3.1. Schematic depicting a basic workflow for estimating historical effective population size (N_e)and examples of how these estimates can inform fisheries management.

our focus on linkage disequilibrium N_e, other implementations of N_e that measure different aspects of genetic drift (Box 2) also provide important information on the evolutionary trajectories of populations. Similarly, measures of genetic diversity beyond N_e (e.g., allelic diversity; Allendorf et al., 2024) may be needed to fully understand population-level conservation status. We focus here on linkage disequilibrium N_e given that this concept has so far ranked among the most widely studied definitions of N_e in fisheries contexts, which we hope supports greater participation and understanding in discussions of N_e.

Key assumptions

All methods for estimating N_e require the same foundational understanding of underlying assumptions. Although N_e is defined relative to an ideal population (R. S. Waples, 2005, 2022; Wright, 1931; Box 1), this simplification is usually unrealistic for wild populations, which experience gene flow, mutation, and selection—all of which impact standing genetic variation (Slatkin, 1987; Harmon & Braude, 2010). Further, estimates of N_e are often based on models that assume discrete (e.g., nonoverlapping) generations and thus a lack of age structure (Nei & Tajima, 1981; R. S. Waples, 2024b, 2025). Calculating N_e without accounting for factors such as these can result in excessively biased estimates that do not reflect the true N_e of a population (Hill, 1972). For example, even weak deviations from random mating can lead to underestimates of N_e (Ryman et al., 2023). Similarly, samples comprising more than one generation (which is possible for species with overlapping generations) can yield downwardly biased estimates of N_e, particularly when linkage disequilibrium methods are used (R. S. Waples et al., 2014).

In practice, violating any one of the basic assumptions underlying N_e is often unavoidable, but awareness of which assumptions are violated and the likely impact on resulting estimates provides important context for interpreting results (see R. S. Waples [2024b] for a detailed review). For example, the assumption of nonoverlapping generations is routinely violated in studies of N_e in wild populations. Despite this, R. S. Waples et al. (2014) found that for estimates of N_e based on linkage disequilibrium, it is possible to produce largely unbiased estimates by applying a correction derived from basic life history information on age at maturity, adult lifespan, and age-specific fecundity. Similarly, R. S. Waples and England (2011) found that N_e derived from linkage disequilibrium methods is generally robust to low levels of gene flow among populations (migration rates <~5–10%). For populations experiencing high levels of gene flow, estimates of N_e are more likely to reflect the N_e of a metapopulation rather than a single population (R. S. Waples, 2024a). Studies that assess the impacts of violating the

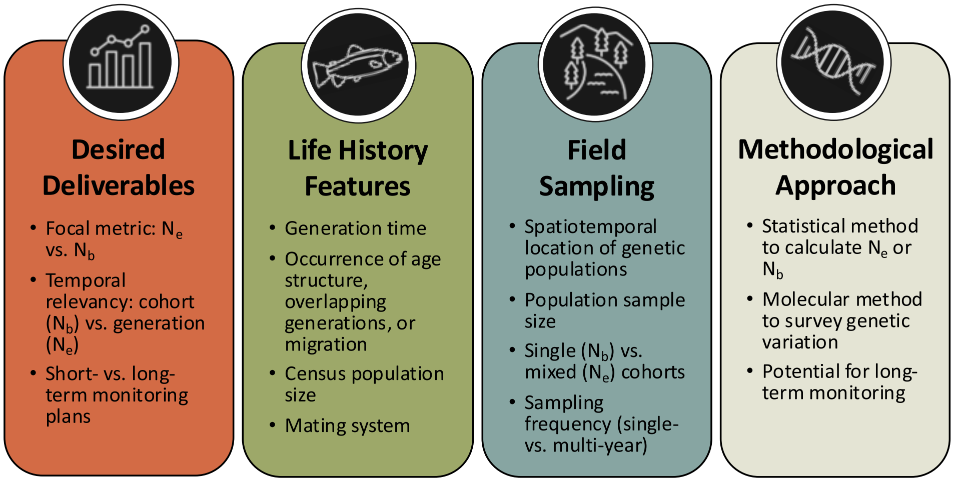


Figure 1. Schematic highlighting important factors to consider when designing empirical studies to estimate effective population size (N_e). The factors listed here are not exhaustive but rather highlight a set of key information needs likely to influence the study design. Many factors are interrelated, where information on one factor is relevant to one or more additional factors. For example, populations that exhibit larger population census size potentially (but not necessarily) exhibit larger N_e and may require larger population sample sizes to reliably estimate N_e. In this scenario, sampling a suitable number of individuals may only be practical via sampling mixed cohorts, including in more than one field season. If multi-season sampling is necessary, an important consideration is whether focal populations exhibit seasonal movement and mixing.

underlying assumptions of N_e elucidate important relationships that greatly assist the interpretation of results from wild systems.

Additional considerations apply to N_e estimated from populations that exhibit iteroparous versus semelparous life histories (R. S. Waples, 2025). Semelparous populations with discrete generations are relatively straightforward, as most N_e models assume nonoverlapping generations (R. S. Waples, 2005). However, age-structured semelparous species exhibit both discrete generations resulting from the die-off of parents following reproduction and overlapping generations resulting from differences in maturity across age-classes (R. S. Waples, 2005). Estimates of N_e for both iteroparous and semelparous species are further complicated by large interannual variation in reproductive success (R. S. Waples, 2002; R. S. Waples, Scribner, et al., 2018). Collectively, these characteristics contribute increased variance to N_e estimates (Nunney, 2002; R. S. Waples, 2006b), making it important to understand their context, including the biological system from which estimates were derived and the study design used to produce estimates.

HOW SHOULD STUDIES TO MEASURE N_e BE DESIGNED?

Generating reliable estimates of N_e requires careful consideration of the study design, particularly for factors related to the field sampling of genetically distinct populations. For natural resource managers, these factors must usually be balanced by decisions regarding desired deliverables and the molecular approach employed in the lab (Figure 1). In this section, we discuss key elements of the field sampling and molecular components of study design, including (1) sampling of genetically distinct populations, (2) the number of individuals to sample from each population, (3) which population segment to target for sampling, and (4) selecting appropriate molecular markers to survey in sampled individuals.

Population delimitation

Given that N_e is a population-level parameter, a critical first step in the experimental design of studies aimed at inferring N_e is to develop field plans for sampling genetically distinct populations. Accounting for population substructure by delimiting genetic populations is important given the sensitivity of linkage disequilibrium-based estimates of N_e to nonrandom mating (Ryman et al., 2023). Inadvertently sampling individuals from more than one genetic population can upwardly bias estimates of N_e due to a larger pool of potential parents (Nei & Lee, 1973; R. S. Waples, 2024b). However, if the combined populations are very genetically distinct, then N_e can be biased low (Nei & Lee, 1973; R. S. Waples, 2024b). Baseline knowledge on the location of genetic populations in space and time is valuable for informing field sampling, but this information is lacking or incomplete for many managed fish populations, making it necessary to both determine population structure and estimate N_e within the same study. In this scenario, the sample sizes needed for reliable estimation of N_e may be sufficient to also infer genetic population structure so that the same sampling scheme can be used (see section on Sample Sizes per Population). Even when genetically distinct populations are known, assignment of newly sampled individuals to source population is necessary prior to calculating N_e. Given these factors, it is typical to leverage the genetic data generated in studies of N_e to first assign individuals to genetic population.

When prior genetic information is limited, leveraging other biological information can inform decisions about the probable location of genetic populations in the field (Cramer et al., 2023; Reis-Santos et al., 2018). For example, abundance data may help identify areas of high abundance that potentially correspond with populations and areas of low abundance that may reflect breaks between populations (Collie & Sissenwine, 1983). Knowledge of barriers to movement (e.g., dams, waterfalls, road crossings, mountain ranges) may help in evaluating the likely presence of genetically distinct populations (Brunke et al., 2019; Funk et al., 2005; Mamoozadeh et al., 2023), as well as information on environmental heterogeneity (e.g., temperature gradients, habitat diversity, salinity clines) potentially affecting genetic connectivity between distinct regions of the landscape (Keyghobadi et al., 2005; Manel et al., 2003). Movement patterns inferred from studies that employ tagging or other similar methods can provide important information on the location and seasonal stability of local populations (Block et al., 2005; Lindley et al., 2011). Finally, traditional ecological knowledge can supply important local information indicative of potential genetic population structure (Adams et al., 1993; Bayliss & Hutton, 2017; Drew, 2005; Drew & Henne, 2006; Henson et al., 2021).

Sample sizes per population

Equally important to identifying genetic populations to target in field sampling is determining how many individuals to sample from each population. This per population sample size is critical for producing accurate and precise estimates of N_e that are informative for decision making in fisheries contexts. However, a universally appropriate sample size does not exist. This is because populations exhibit wide-ranging life histories and biological characteristics and have experienced different ecological and evolutionary histories (Clarke et al., 2024). Regardless, findings from previous empirical and simulation studies offer useful guidelines. For example, estimates of N_e generally exhibit lower levels of bias as sample size increases, and this bias may be negligible when sample size exceeds two times N_e (R. S. Waples, 2006a). An appropriate per-population sample size thus depends on the true N_e of a population, which is likely unknown, although estimates generated in previous studies (even if imprecise; R. S. Waples, 2024a) can help identify sampling bounds. A common approach is to target minimum sample sizes of 50–100 individuals per population (Marandel et al., 2020; Tallmon et al., 2010; R. S. Waples, 2006a; Whiteley et al., 2012). Smaller sample sizes may be possible for populations that exhibit low N_e, particularly if N_e is inferred using large numbers of molecular markers (see sections on Marker Selection and How Is N_e Used in Fisheries Management?); although smaller sample sizes may be insufficient for populations experiencing strong genetic drift (Ryman et al., 2019; J. Wang, 2005). Conversely, populations that exhibit high N_e require larger sample sizes to estimate N_e (J. Wang, 2025). In a practical context, this means that achieving sufficient sample sizes for accurately inferring N_e may become increasingly feasible in populations where N_e is declining. In populations where N_e is increasing, it may not be possible to collect enough samples to produce an accurate estimate.

System-specific factors may also constrain sample availability and impede reliable inference of N_e. For example, in rare, threatened, or cryptic populations, attaining suitable sample sizes may not be possible. Populations with extremely large N_c (and presumably large N_e), like in some pelagic marine fishes, may require sampling thousands of individuals to reliably infer N_e, exceeding the practical limitations of most sampling schemes (Hare et al., 2011; Marandel et al., 2019; R. S. Waples, 2016; R. K. Waples et al., 2016). In these systems, estimates of N_e can be substantially downwardly biased by factors such as extreme individual differences in reproductive success (R. S. Waples, 2016). However, robust inference of N_e may be possible by surveying both large numbers of individuals and molecular markers (J. Wang, 2025) and by assessing the consistency of N_e estimates across temporal replicates (R. S. Waples, Grewe, et al., 2018). Regardless of the exact system, sampling programs are often constrained by real-world factors such as cost, effort, and ease of obtaining samples.

Sample composition

In addition to delimiting genetic populations and determining appropriate sample sizes, sample composition is a crucial component of the field sampling design for studies of N_e. Analogous to other biological metrics inferred from wild fish populations, accurate inference of N_e requires a representative sample per focal population. Because N_e is a generation-scale metric, sampling efforts should focus on collecting population samples that capture the range of diversity reflected within a generation. If sampling occurs nonrandomly (for example, by sampling only juveniles), family groups may be overrepresented, particularly in fishes with limited dispersal capabilities, biasing N_e estimates (Luikart et al., 2010). However, if the goal is to measure effective size at the timescale of cohorts (rather than generations), then sampling a single cohort (e.g., young-of-year fish) is desirable (see section on Relationship of N_e to N_b). For less abundant populations, sampling a suitable number of individuals within a single field season may be difficult. In these cases, an alternative may be to pool individuals sampled across multiple field seasons, but care must be taken to ensure that the pool of sampled individuals is representative of the overall genetic diversity within a population (Marandel et al., 2019; Trask et al., 2017). Similarly, samples pooled across field seasons may comprise multiple generations, introducing bias to N_e estimates (see section on Key Assumptions; Kardos & Waples, 2024). Ideally, field efforts should be designed to minimize biases due to age structure, sex ratio, family relationships, and genetic population structure (R. S. Waples, 2025).

Marker selection

Selecting appropriate genetic markers is an important molecular element of the study design, as the number and type of marker affects the precision and accuracy of N_e estimates. Studies to infer N_e in the context of fisheries management have primarily relied on surveys of microsatellites or single nucleotide polymorphisms (SNPs). Robust estimates of N_e are possible using either type of genetic marker. Microsatellites typically contain several alleles per locus, conferring higher information content per marker compared to SNPs, which contain only two alleles per locus (see Morin et al. [2004] for a more detailed comparison of these marker types). This feature makes it possible to produce reliable estimates of N_e using a smaller number of microsatellites (e.g., the tens of loci typical of genetic data sets) compared to SNPs (e.g., the thousands or more loci typical of genomic data sets). However, this benefit is balanced by the ever-increasing ease and cost-effectiveness of producing genomic data sets that comprise large numbers of SNPs (e.g., thousands to hundreds of thousands of loci) and cover a greater portion of the genome, making robust SNP-based estimates of N_e that reflect genome-wide diversity widely practical. The information content of SNPs may be further increased through microhaplotypes, which comprise multi-allelic loci spanning multiple nearby SNPs (Baetscher et al., 2018) and have already been used to infer the N_e of some fish populations (Osborne et al., 2023). Ultimately, the decision of which molecular markers to employ is influenced by multiple factors, including the availability of baseline data sets that may offer a useful comparison to newly produced data sets.

Determining whether to control for the presence of rare alleles is another important factor in the molecular design of studies to infer N_e, particularly for efforts that employ linkage disequilibrium methods. When population sample sizes are low, it is difficult to determine if the infrequent occurrence of some alleles is due to sampling error or reflective of the true frequency of an allele within a population. These scenarios can also be difficult to differentiate when sequencing depth is low, which is possible for genomic data sets (see section on Considerations for Studies that Employ Genomic Methods). The inclusion of alleles that appear rare due to sampling error results in upwardly biased estimates of N_e (Do et al., 2014; Nunziata & Weisrock, 2018; R. S. Waples & Do, 2010). In studies with low population sample sizes, a common practice is to minimize bias from rare alleles by employing a critical threshold for excluding these alleles. For linkage disequilibrium methods, this threshold is ideally set to exclude alleles that occur only once in a population sample (Do et al., 2014). A correction for rare alleles is not needed in instances where allele frequencies have been accurately characterized, for example when a large fraction of individuals within a population were sampled.

SIMULATION-BASED EXPLORATION OF STUDY DESIGN

We conducted simulations to illustrate some of the relationships highlighted in the previous section by exploring factors central to the design of studies where a major goal is to infer the contemporary N_e of fish populations. We focused on linkage disequilibrium-based estimates of N_e derived from genome-wide SNPs to evaluate two questions: (1) what should the per population sample size be to facilitate accurate inference of N_e? and (2) how many SNPs should be surveyed to accurately infer N_e? Our first question directly affects the field sampling component of the study design. Because the number of individuals needed to reliably estimate N_e is expected to vary across populations (see section on Sample Sizes per Populations), a universally applied target sample size is unlikely to yield precise estimates for every population. However, practical constraints on field sampling necessitate striking a balance between the sample sizes needed to accurately infer N_e and the costs of superfluous sampling effort.

Our second question relates to the molecular component of the study design. Methods for surveying genome-wide SNPs (e.g., restriction site-associated DNA sequencing [RADseq; Baird et al., 2008] and low coverage whole genome sequencing [lcWGS; Lou et al., 2021]) are increasingly practical. Additionally, panels of genetic markers that enable standardized surveys (e.g., via RAD capture [Ali et al., 2016] or genotyping-in-thousands by sequencing [Campbell et al., 2015]) are frequently used to monitor managed fish populations. Such panels allow data to be easily combined across studies and further increase the cost-effectiveness of sequencing efforts by targeting an informative subset of the genome, among other potential benefits (Meek & Larson, 2019). In some instances, standardized panels may be preferable for inferring N_e, particularly for long term monitoring efforts. However, information on the number of loci needed to reliably estimate N_e is necessary for identifying an appropriate panel size.

Simulation approach

We explored both study questions using forward-time simulations conducted in SLiM (Haller & Messer, 2019; Messer, 2013). We used SNP genotypes simulated for a single population to create data subsets that differed in the number of SNPs and sampled individuals (see online Supplementary Material). We calculated linkage disequilibrium-based N_e in NeEstimator (Do et al., 2014) for each data subset, then compared N_e estimates across data sets to determine the optimal numbers of samples and SNPs needed to reliably infer N_e. We designed our SLiM model to simulate a population with a constant N_e of approximately 250. We also parameterized our SLiM model to reflect the life history of Brook Trout *Salvelinus fontinalis*, a salmonid that has been the target of considerable conservation and management effort across northeastern North America (including efforts to infer N_e; Bernos & Fraser, 2016; Kazyak et al., 2022; Mamoozadeh et al., 2023; Robinson et al., 2024; Ruzzante et al., 2016; Whiteley et al., 2012). Our aim is to provide fisheries managers with practical insights for investigating N_e in wild fish populations, although system-specific life history traits and other biological and ecological characteristics will differ across systems.

Simulation results

Our simulation results offer general insights into the field and molecular components of studies designed to measure N_e. Point estimates for N_e approximated our targeted N_e of 250 (Figure 2). Effective population size estimates stabilized at a population sample size of ≥30 individuals, with additional gains in precision possible at larger sample sizes. Estimates based on sample sizes of <30 individuals were often negative or infinite, indicating an unreliable estimate due to sampling error, and exhibited very wide confidence intervals. Results based on 500 versus 5,000 SNPs were similar, but at some sample sizes, estimates based on 5,000 SNPs exhibited greater degrees of precision. We compared our linkage disequilibrium-based estimates of N_e to temporal estimates derived from two sampling events separated by approximately 10 generations. The temporal estimates also approximated an N_e of 250 (Figure S1 [available in the online Supplementary Material]), indicating that our targeted N_e could be inferred using information on either linkage disequilibrium or temporal changes in allele frequencies.

Our results illustrate the relationship between population sample size and the resulting accuracy and precision of N_e estimates, where a minimum sample threshold must be met to yield estimates informative for decision making. Additionally, we found that numbers of loci reflecting molecular methods based on RADseq or a SNP panel (as represented by 5,000 versus 500 SNPs, respectively) both produced reliable estimates of N_e. This result indicates that N_e can be accurately inferred once a minimum number of loci have been surveyed (R. S. Waples, 2025), assuming that sufficient sequencing depth and per population sample sizes have also been achieved. These relationships are consistent with those reported from other simulation-based explorations of population sample size and locus count on precision and accuracy of N_e estimates (Luikart et al., 2021; R. K. Waples et al., 2016; Whiteley et al., 2012). We expect

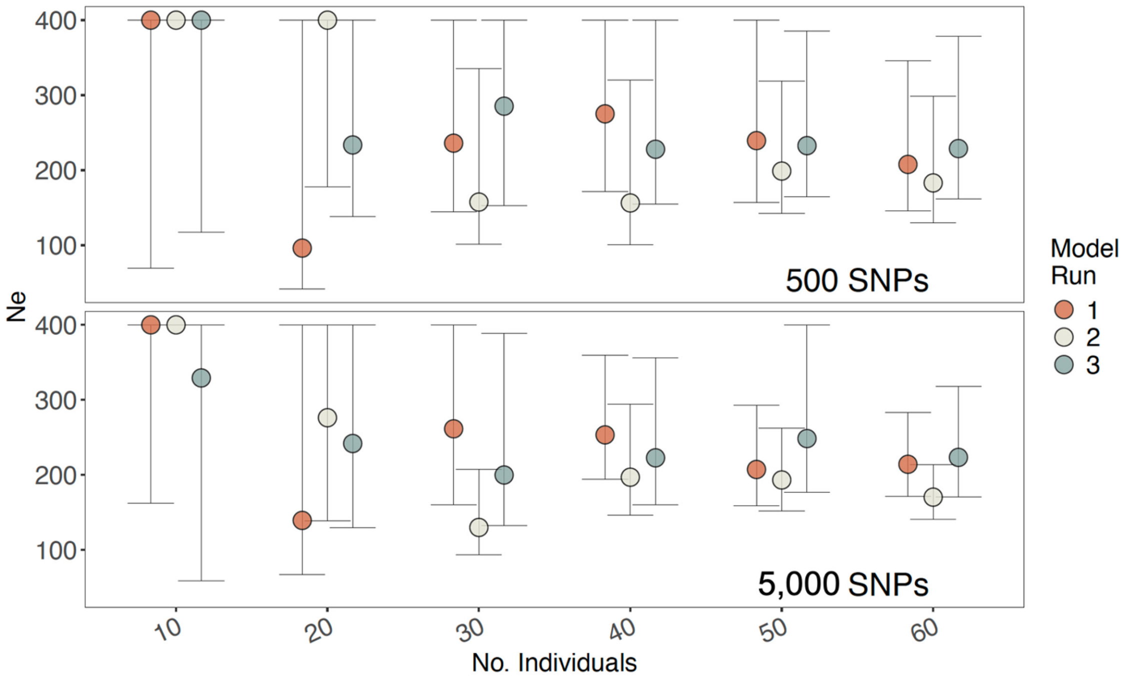


Figure 2. Results from simulations conducted in SLiM to explore the effects of population sample size and the number of surveyed single nucleotide polymorphisms (SNPs) on linkage disequilibrium estimates of contemporary effective population size (N_e). Point estimates for N_e and 95% jackknife confidence intervals are shown for three SLiM model runs. Scenarios with 10–60 (by increments of 10) individuals genotyped at 500 (top panel) or 5,000 (bottom panel) SNPs are shown. For clarity, N_e estimates and confidence intervals with large or negative (infinite) values were converted to a value of 400 for this figure. Effective population size was independently estimated for each scenario using the linkage disequilibrium method implemented in NeEstimator and a minimum minor allele frequency threshold of 5%.

that precise estimates of N_e may be feasible with fewer SNPs through the use of microhaplotypes (Baetscher et al., 2018).

RELATIONSHIP OF N_e TO N_b

A parameter related to but distinct from N_e is the effective number of breeders (N_b; Box 4). Effective number of breeders reflects the number of breeding individuals that gave rise to a single cohort (Luikart et al., 2021; Ruzzante et al., 2016; R. S. Waples & Antao, 2014; R. S. Waples et al., 2014). Like N_e, N_b is useful for inferring and monitoring conservation status (Luikart et al., 2021); however, the timescale associated with each parameter is different. Whereas N_e reflects generation-scale conservation status, N_b offers information on a single breeding cycle, enabling monitoring at an annual timescale. Effective number of breeders potentially facilitates earlier detection of declining conservation status, whereas declines across multiple reproductive cycles are presumed to produce a corresponding decline in N_e, which reflects longer term evolutionary trends (R. S. Waples et al., 2013, 2014). For species with long generation times, N_b offers information on a timescale more relevant to fisheries management (Ferchaud et al., 2016; R. S. Waples et al., 2013; Whiteley et al., 2012). Additionally, because N_b corresponds with a single cohort, this parameter offers a practical way to control for overlapping generations, provided that single cohort sampling is feasible (e.g., by sampling only age-0 individuals; J. Wang, 2009; R. S. Waples, 2005; R. S. Waples et al., 2014; see Whiteley et al. [2012] for an empirical example).

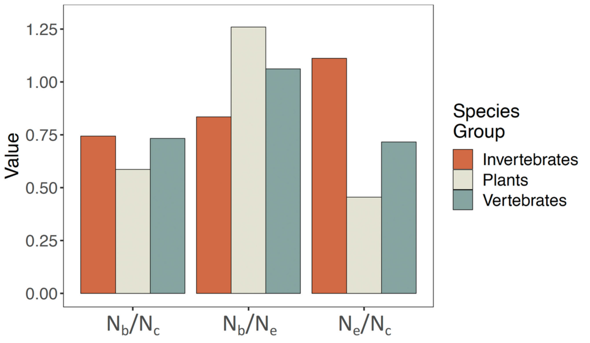
Depending on the sampling design employed, efforts aimed at inferring N_e may actually reflect N_b, and this distinction is important for interpreting results in fisheries contexts. For example, if field sampling targeted individuals from a single cohort, then estimates likely reflect the effective size of the parental generation (N_b). For estimates based on linkage disequilibrium methods, estimates of N_b can be used to calculate N_e, provided that the ratio of N_b to N_e is known (R. S. Waples et al., 2013, 2014). This ratio is based on age at maturity and adult lifespan (see R. S. Waples et al., 2013, 2014 for equation; for another empirical example, see Ruzzante et al., 2016). An important distinction is that N_e generally provides greater sampling flexibility (e.g., mixed cohorts versus a single cohort) and offers a better understanding of the evolutionary trajectories of populations (Luikart et al., 2021; R. S. Waples et al., 2013) but precludes decision making at the temporal scale of the reproductive cycle.

CONSIDERATIONS FOR STUDIES THAT EMPLOY GENOMIC METHODS

Genomic data sets based on surveys of genome-wide SNPs can greatly increase both power and precision for estimating N_e compared to genetic data sets that comprise much smaller numbers of molecular markers. Genomic data sets may also require fewer samples to reliably infer N_e. For example, in simulations conducted by Luikart et al. (2021), N_e estimates based on hundreds of SNPs generally exhibited greater precision at smaller per population sample sizes than estimates based on tens of microsatellites. In some systems, as few as 30 individuals may be sufficient for linkage disequilibrium estimates of contemporary N_e when large numbers of SNPs are surveyed (e.g., >2,000

Box 4. Relationship of effective number of breeders to effective population size and census population size

The effective size of a population over a reproductive cycle (N_b) is distinct from the effective size of a population over a generation (N_e) and from the census size of a population (N_c). All three parameters are informative of population stability but reflect different aspects of a population. Whereas N_c determines the consequences of ecological processes, such as competition and predation, N_b and N_e determine the consequences of evolutionary processes such as genetic drift and mutation (Ferchaud et al., 2016; R. S. Waples, 2022). Importantly, values for N_b, N_e, and N_c often differ from each other, as demonstrated by Clarke et al. (2024) and R. S. Waples et al. (2013), who compared these parameters across a wide range of taxa. Additionally, the effort needed to measure N_b, N_e, and N_c varies across parameters. For example, N_b may be easier to measure when single cohort sampling is possible (Ferchaud et al., 2016). Effective number of breeders is also more practical for populations that exhibit long generation times because the intergenerational sampling needed to monitor N_e may not occur on a timescale relevant to natural resource management.



Box Figure 4.1. Ratios of effective number of breeders (N_b), effective population sizes (N_e), and census population size (N_c) for 63 plant and animal species. Ratios are based on the mean values observed within each species group. Data reproduced from R. S. Waples et al. (2013).

loci, simulation results presented above; Nunziata & Weisrock, 2018). Given that genome-scale data sets are no longer impractical for wild populations, the most onerous element of studies aimed at estimating N_e has shifted from the molecular component to the field sampling component.

Linkage disequilibrium-based estimates of N_e assume that genetic markers do not exhibit physical linkage. However, genetic markers are packaged into a finite number of chromosomes, and for genome-scale data sets that survey increasingly large numbers of SNPs, the probability of surveying SNPs that are physically linked increases (R. K. Waples et al., 2016; R. S. Waples et al., 2022). This linkage causes estimates of N_e to be downwardly biased (R. K. Waples et al., 2016; R. S. Waples, 2024b). Physical linkage among SNPs can be readily controlled for by using a linkage map to gauge the degree of linkage among loci and by restricting N_e estimates to only those locus pairs located on different chromosomes (e.g., Weise et al., 2022). Similarly, the parametric confidence intervals used to reflect variance in microsatellite-based estimates of N_e are inappropriate for analyses based on >100 SNPs (Jones et al., 2016). This is because physical linkage among loci results in parametric confidence intervals that are excessively narrow (Jones et al., 2016; Luikart et al., 2021; R. S. Waples & Do, 2008). The use of jackknife confidence intervals (Jones et al., 2016), although potentially overly conservative, offers a more robust approach to easily control for bias in N_e estimated from genomic data.

Estimates of N_e based on linkage disequilibrium are also susceptible to bias from missing data, which tends to be more prevalent in genomic data sets due to the large number of loci comprising these data sets. For example, genomic data sets with large levels of missing data can upwardly bias estimates of N_e, particularly if missingness is nonrandom (Marandel et al., 2020). Similarly, low-coverage genomic data sets, where larger numbers of individuals are sequenced at a lower depth to increase cost efficiency, are increasingly common and have the potential to severely bias linkage disequilibrium estimates of N_e (Kardos & Waples, 2024; J. Wang, 2025). Bias in genomic data sets due to low coverage or missing data can be readily accounted for by excluding individuals and loci that exhibit excessively low coverage or large proportions of missing data (e.g., SNPs missing more than 30% of genotypes), as well as genotypes that are unable to be reliably inferred.

HOW IS N_e USED IN FISHERIES MANAGEMENT?

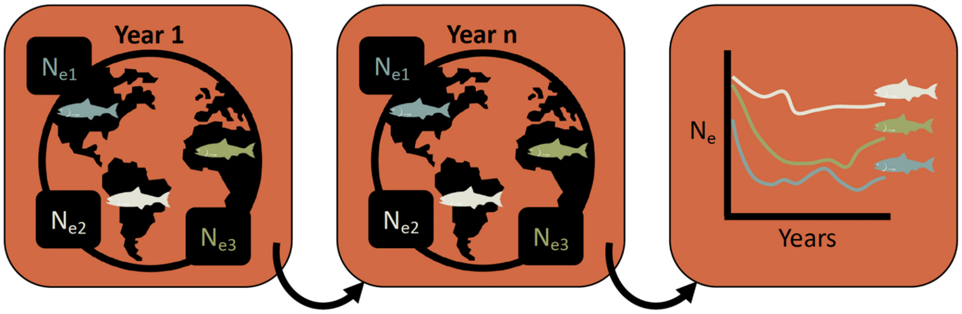
Factors influencing decision making

How is information on N_e integrated into the practical decision making central to managing and conserving wild populations? One avenue is through generalized thresholds proposed as a guide for natural resource managers, who are often required to make decisions on short timelines and with incomplete information. Lower thresholds of N_e <50 (Franklin, 1980) or <100 (Frankham et al., 2014) have been recommended to distinguish high-risk populations susceptible to genetic diversity loss due to accelerated genetic drift and inbreeding depression (Jamieson & Allendorf, 2012). Conversely, upper thresholds of N_e >500 (Franklin, 1980) or >1,000 (Frankham et al., 2014) have been used to distinguish low risk populations that presumably exhibit sufficient evolutionary potential. These 50/500 and 100/1,000 thresholds are widely cited in conservation decision making (Box 5). For example, the United Nations Convention on Biological Diversity recently adopted the 50/500 thresholds for monitoring genetic biodiversity (https://www.cbd.int/gbf/targets/4).

Approaches beyond the use of simplifying thresholds are important for reliably monitoring wild populations. Multiple life history factors such as reproductive rate and generation interval influence which thresholds are reasonable for a particular system (Clark et al., 2024; Clarke et al., 2024; Pérez-Pereira et al., 2022; Traill et al., 2007; T. Wang et al., 2019; R. S. Waples, 2024a), making generalized recommendations (such as the 50/500 and 100/1,000 thresholds) about N_e challenging and sometimes insufficient. For example, populations

Box 5. Integrating effective population size into biodiversity conservation

As biodiversity is increasingly impacted by anthropogenic stressors, new standards are needed to monitor conservation status at broad spatial scales. For example, intergovernmental organizations such as the United Nations Convention on Biological Diversity (CBD) promote national- to global-scale conservation efforts by creating frameworks that allow flexible application to various species (Secretariat of the CBD, 2011). One way the CBD incorporates information on effective population size (N_e) into global conservation standards is through genetic indicators (Hoban et al., 2022, 2023; Jetz et al., 2019; Pereira et al., 2013), including N_e (CBD, 2022), which standardize how biodiversity data are collected and offer practical methods for monitoring conservation status over time. Effective population size is ideally suited as an indicator because of its sensitivity to genetic diversity loss within populations, offering a reliable reflection of population-level conservation status (Hoban et al., 2022, 2024; O'Brien et al., 2022). Data accumulated from monitoring N_e within populations can be compared across populations to provide a view of conservation status within species. Monitoring N_e thus offers practical information on biodiversity at the genetic level.



Box Figure 5.1. Schematic portraying how information on effective population size (N_e) can be used to monitor the evolutionary trajectories of populations across time, providing valuable insights for biodiversity conservation at local to broad spatial scales.

with a large N_e but a short generation interval may lose genetic diversity faster than populations with a small N_e but long generation interval. Further, N_e does not always provide a complete picture of genetic diversity loss. For example, the number of alleles in a population (e.g., allelic diversity, an important component of genetic diversity) depends on both N_e and N_c. Populations with a smaller N_c tend to exhibit fewer alleles than populations with a larger N_c so that populations with the same N_e may lose allelic diversity at potentially very different rates due to differences in N_c (Allendorf et al., 2024). Finally, practical constraints on fisheries management may mean that decisions are influenced by factors including projected recovery timelines, the probability of population persistence, and the level of genetic diversity loss willing to be tolerated. Collectively, these insights indicate that a multifaceted approach is needed to monitor and conserve the genetic diversity vital for long-term population persistence.

Illustrative empirical examples

Combined with additional ecological and evolutionary parameters, N_e is a valuable part of a conservation and management tool kit that supports the ability of natural resource managers to monitor the conservation status of managed fish populations and infer their evolutionary potential. Here, we highlight illustrative empirical examples that demonstrate how N_e has been used to guide decision making in fisheries contexts. A recent status review of the Lahontan Cutthroat Trout *Oncorhynchus henshawi* (formerly *O. clarkii henshawii*), which is listed as threatened under the U.S. Endangered Species Act, found that most populations exhibited an $N_e < 50$, even in populations with a comparatively large N_c (U.S. Fish and Wildlife Service, 2023). Combined with other measures of population resiliency, this information was used to maintain the threatened classification of Lahontan Cutthroat Trout while also providing guidance to direct future rehabilitation efforts. In native populations of Brook Trout, Robinson et al. (2024) estimated N_b and identified associated environmental variables. This information is useful for prioritizing populations for management intervention as well as key habitat features for mitigation. Baillie et al. (2016) estimated N_e in Lake Trout *Salvelinus namaycush* sampled before, during, and after fishing-induced population collapses. These results supplied important insights into the effectiveness of fishing regulations at promoting the recovery of Lake Trout populations. In the Australasian Snapper *Chrysophrys auratus*, sampled young-of-year fish were used to calculate N_b while mixed-age adults were used to calculate N_e, with adjustments to account for adult lifespan and age at maturity (Bertram et al., 2024). This study demonstrates the possibility of using large sample sizes from a single cohort to reliably estimate N_b or N_e for abundant, iteroparous species.

Effective population size also offers a useful tool to monitor genetic diversity in captive populations and evaluate the genetic effects of hatchery supplementation on wild populations. For example, Christie et al. (2012) observed that hatchery supplemented populations of steelhead *Oncorhynchus mykiss* exhibited reduced N_e despite increased numbers of adult spawners. This was attributed to the reduced N_b of hatchery broodstock, providing an example of the Ryman–Laikre effect, where hatchery supplementation aimed at increasing genetic diversity yields a reduction in effective size (Ryman & Laikre, 1991). Tracking the Ryman–Laikre and other genetic effects of hatchery supplementation is an important use of N_e estimation in managed populations (R. K. Waples et al., 2016). Many additional empirical examples demonstrate the utility of N_e for guiding management efforts in fisheries contexts. The use of N_e for biodiversity monitoring is increasing (Garner et al., 2020; Hoban et al., 2020, 2021; Box 5) but is ripe for expansion (Garner et al., 2020; Schmidt et al., 2023), particularly given the fundamental role genetic diversity plays in long-term population persistence.

CONCLUDING REMARKS

Effective population size is a parameter central to conservation biology and natural resource management, providing essential information on population-level conservation status and evolutionary potential, including for managed fish populations. However, despite the importance of the information conveyed by N_e, this parameter frequently receives limited attention compared to more widely understood and appreciated metrics. One factor potentially contributing to this is the complexity of concepts underlying N_e and the presently limited availability of nontechnical resources on this topic. Our intent for this review is to offer an accessible introduction to N_e for a broad audience of practitioners with wide-ranging expertise. Further, we aim for the information shared here to facilitate practical discussions for designing empirical studies to measure N_e. Our goal is to support more widespread integration of N_e into decision-making efforts in fisheries contexts, thereby spurring increased genetic monitoring efforts that improve our ability to manage fish populations into the future.

SUPPLEMENTARY MATERIAL

Supplementary material is available at *Transactions of the American Fisheries Society* online.

DATA AVAILABILITY

The script used in the SLiM simulation analysis described here was adapted from Reid & Pinsky (2022) and is available in the Supplementary material.

ETHICS STATEMENT

There were no ethical guidelines applicable to this study.

FUNDING

The authors do not have any funding sources to declare for this work.

CONFLICTS OF INTEREST

The authors have no conflicts of interest to declare.

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REFERENCES

Adams, M., Frost, K. J., & Harwood, L. A. (1993). Alaska and Inuvialuit Beluga Whale Committee (AIBWC)—An initiative in "at home management." *Arctic*, *46*, 134–137. https://doi.org/10.14430/arctic1334

Ali, O. A., Jeffres, C., & Miller, M. R. (2016). RAD capture (rapture): Flexible and efficient sequence-based genotyping. *Genetics*, *202*, 389–400. https://doi.org/10.1534/genetics.115.183665

Allendorf, F. W., Funk, W. C., Aitken, S. N., Byrne, M., Luikart, G., & Antunes, A. (2022). *Conservation and the genomics of populations* (3rd ed.). Oxford University Press. https://doi.org/10.1093/oso/9780198856566.001.0001

Allendorf, F. W., Hössjer, O., & Ryman, N. (2024). What does effective population size tell us about loss of allelic variation? *Evolutionary Applications*, *17*, Article e13733. https://doi.org/10.1111/eva.13733

Allendorf, F. W., & Luikart, G. (2007). *Conservation and the genetics of populations*. Blackwell Publishing.

Babyn, J., Ruzzante, D., Bravington, M., & Mills Flemming, J. (2024). Estimating effective population size using close-kin mark–recapture. *Methods in Ecology and Evolution*, *15*, 2059–2073. https://doi.org/10.1111/2041-210X.14419

Baetscher, D. S., Clemento, A. J., Ng, T. C., Anderson, E. C., & Garza, J. C. (2018). Microhaplotypes provide increased power from short-read DNA sequences for relationship inference. *Molecular Ecology Resources*, *18*, 296–305. https://doi.org/10.1111/1755-0998.12737

Baillie, S. M., Muir, A. M., Scribner, K., Bentzen, P., & Krueger, C. C. (2016). Loss of genetic diversity and reduction of genetic distance among Lake Trout *Salvelinus namaycush* ecomorphs, Lake Superior 1959 to 2013. *Journal of Great Lakes Research*, *42*, 204–216. https://doi.org/10.1016/j.jglr.2016.02.001

Baird, N. A., Etter, P. D., Atwood, T. S., Currey, M. C., Shiver, A. L., Lewis, Z. A., Selker, E. U., Cresko, W. A., & Johnson, E. A. (2008). Rapid SNP discovery and genetic mapping using sequenced RAD markers. *PLoS ONE*, *3*, Article e3376. https://doi.org/10.1371/journal.pone.0003376

Barbato, M., Orozco-terWengel, P., Tapio, M., & Bruford, M. W. (2015). SNeP: A tool to estimate trends in recent effective population size trajectories using genome-wide SNP data. *Frontiers in Genetics*, *6*, Article 109. https://doi.org/10.3389/fgene.2015.00109

Bayliss, P., & Hutton, M. (2017). *Integrating Indigenous knowledge and survey techniques to develop a baseline for dugong* (Dugong dugon*) management in the Kimberley* (Final Report; Project 1.2.5). Western Australian Marine Science Institution, Kimberley Marine Research Program Node.

Bernos, T. A., & Fraser, D. J. (2016). Spatiotemporal relationship between adult census size and genetic population size across a wide population size gradient. *Molecular Ecology*, *25*, 4472–4487. https://doi.org/10.1111/mec.13790

Bertram, A., Bell, J., Brauer, C., Fairclough, D., Hamer, P., Sandoval-Castillo, J., Wellenreuther, M., & Beheregaray, L. B. (2024). Estimation of effective number of breeders and effective population size in an abundant and heavily exploited marine teleost. *Evolutionary Applications*, *17*, Article e13758. https://doi.org/10.1111/eva.13758

Block, B. A., Teo, S. L. H., Walli, A., Boustany, A., Stokesbury, M. J. W., Farwell, C. J., Weng, K. C., Dewar, H., & Williams, T. D. (2005). Electronic tagging and population structure of Atlantic Bluefin Tuna. *Nature*, *434*, 1121–1127. https://doi.org/10.1038/nature03463

Brunke, J., Radespiel, U., Russo, I.-R., Bruford, M. W., & Goossens, B. (2019). Messing about on the river: The role of geographic barriers in shaping the genetic structure of Bornean small mammals in a fragmented landscape. *Conservation Genetics*, *20*, 691–704. https://doi.org/10.1007/s10592-019-01159-3

Caballero, A. (1994). Developments in the prediction of effective population size. *Heredity*, *73*, 657–679. https://doi.org/10.1038/hdy.1994.174

Caballero, A., & Hill, W. G. (1992). A note on the inbreeding effective population size. *Evolution*, *46*, 1969–1972. https://doi.org/10.2307/2410048

Campbell, N. R., Harmon, S. A., & Narum, S. R. (2015). Genotyping-in-Thousands by sequencing (GT-seq): A cost effective SNP genotyping method based on custom amplicon sequencing. *Molecular Ecology Resources*, *15*, 855–867. https://doi.org/10.1111/1755-0998.12357

Charlesworth, B. (2009). Effective population size and patterns of molecular evolution and variation. *Nature Reviews Genetics*, *10*, 195–205. https://doi.org/10.1038/nrg2526

Christie, M. R., Marine, M. L., French, R. A., Waples, R. S., & Blouin, M. S. (2012). Effective size of a wild salmonid population is greatly reduced by hatchery supplementation. *Heredity*, *109*, 254–260. https://doi.org/10.1038/hdy.2012.39

Clark, M. I., Fitzpatrick, S. W., & Bradburd, G. S. (2024). Pitfalls and windfalls of detecting demographic declines using population genetics in long-lived species. *Evolutionary Applications*, *17*, Article e13754. https://doi.org/10.1111/eva.13754

Clarke, S. H., Lawrence, E. R., Matte, J.-M., Gallagher, B. K., Salisbury, S. J., Michaelides, S. N., Koumrouyan, R., Ruzzante, D. E., Grant, J. W. A., & Fraser, D. J. (2024). Global assessment of effective population sizes: Consistent taxonomic differences in meeting the 50/500 rule. *Molecular Ecology*, *33*, Article e17353. https://doi.org/10.1111/mec.17353.

Collie, J. S., & Sissenwine, M. P. (1983). Estimating population size from relative abundance data measured with error. *Canadian Journal of Fisheries and Aquatic Sciences*, *40*, 1871–1879. https://doi.org/10.1139/f83-217

Convention on Biological Diversity. (2022). *Decision adopted by the conference of the parties to the Convention on Biological Diversity* (CBD/COP/DEC/15/5). https://www.cbd.int/doc/decisions/cop-15/cop-15-dec-05-en.pdf

Coombs, J. A., Letcher, B. A., & Nislow, K. H. (2012). GONe: Software for estimating effective population size in species with generational overlap. *Molecular Ecology Resources*, *12*, 160–163. https://doi.org/10.1111/j.1755-0998.2011.03057.x

Cramer, A. N., Hoey, J. A., Dolan, T. E., Gatins, R., Toy, J. A., Chancellor, J. L., Palkovacs, E. P., Garza, J. C., & Beltran, R. S. (2023). A unifying framework for understanding ecological and evolutionary population connectivity. *Frontiers in Ecology and Evolution*, *11*, Article 1072825. https://doi.org/10.3389/fevo.2023.1072825

Crow, J. F. (1954). Breeding structure of populations. II. Effective population number. In O. Kempthorne, T. A. Bancroft, J. W. Gowen, & J. L. Lush (Eds.), *Statistics and mathematics in biology* (pp. 543–556). Iowa State College Press.

Crow, J. F., & Denniston, C. (1988). Inbreeding and variance effective population numbers. *Evolution*, *42*, 482–495. https://doi.org/10.2307/2409033

Do, C., Waples, R. S., Peel, D., Macbeth, G. M., Tillett, B. J., & Ovenden, J. R. (2014). Neestimator v2: Re-implementation of software for the estimation of contemporary effective population size (N_e) from genetic data. *Molecular Ecology Resources*, *14*, 209–214. https://doi.org/10.1111/1755-0998.12157

Drew, J. A. (2005). Use of traditional ecological knowledge in marine conservation. *Conservation Biology*, *19*, 1286–1293. https://doi.org/10.1111/j.1523-1739.2005.00158.x

Drew, J. A., & Henne, A. P. (2006). Conservation biology and traditional ecological knowledge: Integrating academic disciplines for better conservation practice. *Ecology and Society*, *11*, Article 34. https://doi.org/10.5751/ES-01959-110234

Ferchaud, A.-L., Perrier, C., April, J., Hernandez, C., Dionne, M., & Bernatchez, L. (2016). Making sense of the relationships between N_e, N_b and N_c towards defining conservation thresholds in Atlantic Salmon (*Salmo salar*). *Heredity*, *117*, 268–278. https://doi.org/10.1038/hdy.2016.62

Frankham, R., Bradshaw, C. J. A., & Brook, B. W. (2014). Genetics in conservation management: Revised recommendations for the 50/500 rules, Red List criteria and population viability analyses. *Biological Conservation*, *170*, 56–63. https://doi.org/10.1016/j.biocon.2013.12.036

Franklin, I. R. (1980). Evolutionary change in small populations. In M. E. Soule, & B. A. Wilcox (Eds.), *Conservation biology—An evolutionary-ecological perspective* (pp. 135–149). Sinauer Associates.

Fraser, D. J., Calvert, A. M., Bernatchez, L., & Coon, A. (2013). Multidisciplinary population monitoring when demographic data are sparse: A case study of remote trout populations. *Ecology and Evolution*, *3*, 4954–4969. https://doi.org/10.1002/ece3.871

Funk, W. C., Blouin, M. S., Corn, P. S., Maxell, B. A., Pilliod, D. S., Amish, S., & Allendorf, F. W. (2005). Population structure of Columbia spotted frogs (*Rana luteiventris*) is strongly affected by the landscape. *Molecular Ecology*, *14*, 483–496. https://doi.org/10.1111/j.1365-294X.2005.02426.x

Garner, B. A., Hoban, S., & Luikart, G. (2020). IUCN Red List and the value of integrating genetics. *Conservation Genetics*, *21*, 795–801. https://doi.org/10.1007/s10592-020-01301-6

Gilbert, K. J., & Whitlock, M. C. (2015). Evaluating methods for estimating local effective population size with and without migration. *Evolution*, *69*, 2154–2166. https://doi.org/10.1111/evo.12713

Guschanski, K., Vigilant, L., McNeilage, A., Gray, M., Kagoda, E., & Robbins, M. M. (2009). Counting elusive animals: Comparing field and genetic census of the entire mountain gorilla population of Bwindi Impenetrable National Park, Uganda. *Biological Conservation*, *142*, 290–300. https://doi.org/10.1016/j.biocon.2008.10.024

Haller, B. C., & Messer, P. W. (2019). SLiM 3: Forward genetic simulations beyond the Wright–Fisher model. *Molecular Biology and Evolution*, *36*, 632–637. https://doi.org/10.1093/molbev/msy228

Hare, M. P., Nunney, L., Schwartz, M. K., Ruzzante, D. E., Burford, M., Waples, R. S., Ruegg, K., & Palstra, F. (2011). Understanding and estimating effective population size for practical application in marine species management: Applying effective population size estimates to marine species management. *Conservation Biology*, *25*, 438–449. https://doi.org/10.1111/j.1523-1739.2010.01637.x

Harmon, L. J., & Braude, S. (2010). Conservation of small populations: Effective population sizes, inbreeding, and the 50/500 rule. In S. Braude, & B. S. Low (Eds.), *An introduction to methods and models in ecology, evolution, and conservation biology* (pp. 125–138). Princeton University Press. https://doi.org/10.1515/9781400835454-017

Henson, L. H., Balkenhol, N., Gustas, R., Adams, M., Walkus, J., Housty, W. G., Stronen, A. V., Moody, J., Service, C., Reece, D., vonHoldt, B. M., McKechnie, I., Koop, B. F., & Darimont, C. T. (2021). Convergent geographic patterns between grizzly bear population genetic structure and Indigenous language groups in coastal British Columbia, Canada. *Ecology and Society*, *26*, Article 7. https://doi.org/10.5751/ES-12443-260307

Hill, W. G. (1972). Effective size of populations with overlapping generations. *Theoretical Population Biology*, *3*, 278–289. https://doi.org/10.1016/0040-5809(72)90004-4

Hill, W. G. (1981). Estimation of effective population size from data on linkage disequilibrium. *Genetics Research*, *38*, 209–216. https://doi.org/10.1017/S0016672300020553

Hoban, S., Archer, F. I., Bertola, L. D., Bragg, J. G., Breed, M. F., Bruford, M. W., Coleman, M. A., Ekblom, R., Funk, W. C., Grueber, C. E., Hand, B. K., Jaffé, R., Jensen, E., Johnson, J. S., Kershaw, F., Liggins, L., MacDonald, A. J., Mergeay, J., Miller, J. M., … Hunter, M. E. (2022). Global genetic diversity status and trends: Towards a suite of Essential Biodiversity Variables (EBVs) for genetic composition. *Biological Reviews*, *97*, 1511–1538. https://doi.org/10.1111/brv.12852

Hoban, S., Bruford, M. W., Funk, W. C., Galbusera, P., Griffith, M. P., Grueber, C. E., Heuertz, M., Hunter, M. E., Hvilsom, C., Kalamujic

Stroil, B., Kershaw, F., Khoury, C. K., Laikre, L., Lopes-Fernandes, M., MacDonald, A. J., Mergeay, J., Meek, M., Mittan, C., Mukassabi, T. A., … Vernesi, C. (2021). Global commitments to conserving and monitoring genetic diversity are now necessary and feasible. *BioScience*, *71*, 964–976. https://doi.org/10.1093/biosci/biab054

Hoban, S., Bruford, M., Jackson, J. D., Lopes-Fernandes, M., Heuertz, M., Hohenlohe, P. A., Paz-Vinas, I., Sjögren-Gulve, P., Segelbacher, G., Vernesi, C., Aitken, S., Bertola, L. D., Bloomer, P., Breed, M., Rodríguez-Correa, H., Funk, W. C., Grueber, C. E., Hunter, M. E., Jaffe, R., … Laikre, L. (2020). Genetic diversity targets and indicators in the CBD post-2020 Global Biodiversity Framework must be improved. *Biological Conservation*, *248*, Article 108654. https://doi.org/10.1016/j.biocon.2020.108654

Hoban, S., da Silva, J. M., Hughes, A., Hunter, M. E., Kalamujic Stroil, B., Laikre, L., Mastretta-Yanes, A., Millette, K., Paz-Vinas, I., Ruiz Bustos, L., Shaw, R. E., & Vernesi, C., & the Coalition for Conservation Genetics. (2024). Too simple, too complex, or just right? Advantages, challenges, and guidance for indicators of genetic diversity. *BioScience*, *74*, 269–280. https://doi.org/10.1093/biosci/biae006

Hoban, S., da Silva, J. M., Mastretta-Yanes, A., Grueber, C. E., Heuertz, M., Hunter, M. E., Mergeay, J., Paz-Vinas, I., Fukaya, K., Ishihama, F., Jordan, R., Köppä, V., Latorre- Cárdenas, M. C., MacDonald, A. J., Rincon-Parra, V., Sjögren-Gulve, P., Tani, N., Thurfjell, H., & Laikre, L. (2023). Monitoring status and trends in genetic diversity for the Convention on Biological Diversity: An ongoing assessment of genetic indicators in nine countries. *Conservation Letters*, *16*, Article e12953. https://doi.org/10.1111/conl.12953

Hollenbeck, C. M., Portnoy, D. S., & Gold, J. R. (2016). A method for detecting recent changes in contemporary effective population size from linkage disequilibrium at linked and unlinked loci. *Heredity*, *117*, 207–216. https://doi.org/10.1038/hdy.2016.30

Hong, A., Cheek, R. G., Nihara De Silva, S., Mukherjee, K., Yooseph, I., Oliva, M., Heim, M., Funk, C. W., Tallmon, D., Boucher, C. (2024). ONeSAMP 3.0: Estimation of effective population size via single nucleotide polymorphism data from one population. *G3: Gene Genomes Genetics*, *14*, Article jkae153. https://doi.org/10.1093/g3journal/jkae153

Hui, T.-Y., & Burt, A. (2015). Estimating effective population size from temporally spaced samples with a novel, efficient maximum likelihood algorithm. *Genetics*, *200*, 285–293. https://doi.org/10.1534/genetics.115.174904

Jamieson, I. G., & Allendorf, F. W. (2012). How does the 50/500 rule apply to MVPs? *Trends in Ecology & Evolution*, *27*, 578–584. https://doi.org/10.1016/j.tree.2012.07.001

Jetz, W., McGeoch, M. A., Guralnick, R., Ferrier, S., Beck, J., Costello, M. J., Fernandez, M., Geller, G. N., Keil, P., Merow, C., Meyer, C., Muller-Karger, F. E., Pereira, H. M., Regan, E. C., Schmeller, D. S., & Turak, E. (2019). Essential biodiversity variables for mapping and monitoring species populations. *Nature Ecology & Evolution*, *3*, 539–551. https://doi.org/10.1038/s41559-019-0826-1

Jones, A. T., Ovenden, J. R., & Wang, Y.-G. (2016). Improved confidence intervals for the linkage disequilibrium method for estimating effective population size. *Heredity*, *117*, 217–223. https://doi.org/10.1038/hdy.2016.19

Jones, O. R., & Wang, J. (2010). COLONY: A program for parentage and sibship inference from multilocus genotype data. *Molecular Ecology Resources*, *10*, 551–555. https://doi.org/10.1111/j.1755-0998.2009.02787.x

Jorde, P. E., & Ryman, N. (2007). Unbiased estimator for genetic drift and effective population size. *Genetics*, *177*, 927–935. https://doi.org/10.1534/genetics.107.075481

Kalinowski, S. T., & Waples, R. S. (2002). Relationship of effective to census size in fluctuating populations. *Conservation Biology*, *16*, 129–136. https://doi.org/10.1046/j.1523-1739.2002.00134.x

Kardos, M., & Waples, R. S. (2024). Low-coverage sequencing and Wahlund effect severely bias estimates of inbreeding, heterozygosity and effective population size in North American wolves. *Molecular Ecology*. Advance online publication, https://doi.org/10.1111/mec.17415

Kazyak, D. C., Lubinski, B. A., Kulp, M. A., Pregler, K. C., Whiteley, A. R., Hallerman, E., Coombs, J. A., Kanno, Y., Rash, J. M., Morgan, R. P., II, Habera, J., Henegar, J., Weathers, T. C., Sell, M. T., Rabern, A., Rankin, D., & King, T. L. (2022). Population genetics of Brook Trout in the southern Appalachian mountains. *Transactions of the American Fisheries Society*, *151*, 127–149. https://doi.org/10.1002/tafs.10337

Keyghobadi, N., Roland, J., & Strobeck, C. (2005). Genetic differentiation and gene flow among populations of the alpine butterfly, *Parnassius smintheus*, vary with landscape connectivity. *Molecular Ecology*, *14*, 1897–1909. https://doi.org/10.1111/j.1365-294X.2005.02563.x

Kimura, M., & Crow, J. F. (1963). The measurement of effective population number. *Evolution*, *17*, 279–288. https://doi.org/10.2307/2406157

Krimbas, C. B., & Tsakas, S. (1971). The genetics of *Dacus oleae*. V. Changes of esterase polymorphism in a natural population following insecticide control—Selection or drift? *Evolution*, *25*, 454–460. https://doi.org/10.2307/2407343

Lee, A. M., Engen, S., & Saether, B.-E. (2011). The influence of persistent individual differences and age at maturity on effective population size. *Proceedings of the Royal Society B: Biological Sciences*, *278*, 3303–3312. https://doi.org/10.1098/rspb.2011.0283

Leffler, E. M., Bullaughey, K., Matute, D. R., Meyer, W. K., Ségurel, L., Venkat, A., Andolfatto, P., & Przeworski, M. (2012). Revisiting an old riddle: What determines genetic diversity levels within species? *PLoS Biology*, *10*, Article e1001388. https://doi.org/10.1371/journal.pbio.1001388

Lehnert, S. J., Kess, T., Bentzen, P., Clément, M., & Bradbury, I. R. (2019). Divergent and linked selection shape patterns of genomic differentiation between European and North American Atlantic Salmon (*Salmo salar*). *Molecular Ecology*, *29*, 2160–2175. https://doi.org/10.1111/mec.15480

Leroy, G., Carroll, E. L., Bruford, M. W., DeWoody, J. A., Strand, A., Waits, L., & Wang, J. (2018). Next-generation metrics for monitoring genetic erosion within populations of conservation concern. *Evolutionary Applications*, *11*, 1066–1083. https://doi.org/10.1111/eva.12564

Lindley, S. T., Erickson, D. L., Moser, M. L., Williams, G., Langness, O. P., McCovey, B. W., Jr., Belchik, M., Vogel, D., Pinnix, W., Kelly, J. T., Heublein, J. C., & Klimley, A. P. (2011). Electronic tagging of Green Sturgeon reveals population structure and movement among estuaries. *Transactions of the American Fisheries Society*, *140*, 108–122. https://doi.org/10.1080/00028487.2011.557017

Lou, R. N., Jacobs, A., Wilder, A. P., & Therkildsen, N. O. (2021). A beginner's guide to low-coverage whole genome sequencing for population genomics. *Molecular Ecology*, *30*, 5966–5993. https://doi.org/10.1111/mec.16077

Luikart, G., Antao, T., Hand, B. K., Muhlfeld, C. C., Boyer, M. C., Cosart, T., Trethewey, B., Al-Chockhachy, R., & Waples, R. S. (2021). Detecting population declines via monitoring the effective number of breeders (N_b). *Molecular Ecology Resources*, *21*, 379–393. https://doi.org/10.1111/1755-0998.13251

Luikart, G., & Cornuet, J.-M. (1999). Estimating the effective number of breeders from heterozygote excess in progeny. *Genetics*, *151*, 1211–1216. https://doi.org/10.1093/genetics/151.3.1211

Luikart, G., Ryman, N., Tallmon, D. A., Schwartz, M. K., & Allendorf, F. W. (2010). Estimation of census and effective population sizes: The increasing usefulness of DNA-based approaches. *Conservation Genetics*, *11*, 355–373. https://doi.org/10.1007/s10592-010-0050-7

Mamoozadeh, N., Goldsworthy, C., Miller, L., Peterson, N., & Meek, M. (2023). Sources of coaster Brook Trout (*Salvelinus fontinalis*) revealed by genomic analysis of Brook Trout populations along Minnesota's shoreline with Lake Superior. *Journal of Great Lakes Research*, *49*, 901–917. https://doi.org/10.1016/j.jglr.2023.06.005

Manel, S., Schwartz, M. K., Luikart, G., & Taberlet, P. (2003). Landscape genetics: Combining landscape ecology and population genetics. *Trends in Ecology & Evolution*, *18*, 189–197. https://doi.org/10.1016/S0169-5347(03)00008-9

Marandel, F., Charrier, G., Lamy, J., Le Cam, S., Lorance, P., & Trenkel, V. M. (2020). Estimating effective population size using RADseq:

Effects of SNP selection and sample size. *Ecology and Evolution*, *10*, 1929–1937. https://doi.org/10.1002/ece3.6016

Marandel, F., Lorance, P., Berthelé, O., Trenkel, V. M., Waples, R. S., & Lamy, J.-B. (2019). Estimating effective population size of large marine populations, is it feasible? *Fish and Fisheries*, *20*, 189–198. https://doi.org/10.1111/faf.12338

Meek, M. H., & Larson, W. A. (2019). The future is now: Amplicon sequencing and sequence capture usher in the conservation genomics era. *Molecular Ecology Resources*, *19*, 795–803. https://doi.org/10.1111/1755-0998.12998

Messer, P. W. (2013). SLiM: Simulating evolution with selection and linkage. *Genetics*, *194*, 1037–1039. https://doi.org/10.1534/genetics.113.152181

Morin, P. A., Luikart, G., & Wayne, R. K., & The SNP Workshop Group (2004). SNPs in ecology, evolution and conservation. *Trends in Ecology & Evolution*, *19*, 208–216. https://doi.org/10.1016/j.tree.2004.01.009

Nadachowska-Brzyska, K., Konczal, M., & Babik, W. (2022). Navigating the temporal continuum of effective population size. *Methods in Ecology and Evolution*, *13*, 22–41. https://doi.org/10.1111/2041-210X.13740

Nei, M., & Li, W.-H. (1973). Linkage disequilibrium in subdivided populations. *Genetics*, *75*, 213–219. https://doi.org/10.1093/genetics/75.1.213

Nei, M., & Tajima, F. (1981). Genetic drift and estimation of effective population size. *Genetics*, *98*, 625–640. https://doi.org/10.1093/genetics/98.3.625

Nomura, T. (2008). Estimation of effective number of breeders from molecular coancestry of single cohort sample. *Evolutionary Applications*, *1*, 462–474. https://doi.org/10.1111/j.1752-4571.2008.00015.x

Novo, I., Santiago, E., & Caballero, A. (2022). The estimates of effective population size based on linkage disequilibrium are virtually unaffected by natural selection. *PLOS Genetics*, *18*, Article e1009764. https://doi.org/10.1371/journal.pgen.1009764

Nunney, L. (1995). Measuring the ratio of effective population size to adult numbers using genetic and ecological data. *Evolution*, *49*, 389–392. https://doi.org/10.2307/2410351

Nunney, L. (2002). The effective size of annual plant populations: The interaction of a seed bank with fluctuating population size in maintaining genetic variation. *The American Naturalist*, *160*, 195–204. https://doi.org/10.1086/341017

Nunziata, S. O., & Weisrock, D. W. (2018). Estimation of contemporary effective population size and population declines using RAD sequence data. *Heredity*, *120*, 196–207. https://doi.org/10.1038/s41437-017-0037-y

O'Brien, D., Laikre, L., Hoban, S., Bruford, M. W., Ekblom, R., Fischer, M. C., Hall, J., Hvilsom, C., Hollingsworth, P. M., Kershaw, F., Mittan, C. S., Mukassabi, T. A., Ogden, R., Segelbacher, G., Shaw, R. E., Vernesei, C., & MacDonald, A. J. (2022). Bringing together approaches to reporting on within species genetic diversity. *Journal of Applied Ecology*, *59*, 2227–2233. https://doi.org/10.1111/1365-2664.14225

Osborne, M. J., Caeiro-Dias, G., & Turner, T. F. (2023). Transitioning from microsatellites to SNP-based microhaplotypes in genetic monitoring programmes: Lessons from paired data spanning 20 years. *Molecular Ecology*, *32*, 316–334. https://doi.org/10.1111/mec.16760

Ovenden, J. R., Peel, D., Street, R., Courtney, A. J., Hoyle, S. D., Peel, S. L., & Podlich, H. (2007). The genetic effective and adult census size of an Australian population of tiger prawns (*Penaeus esculentus*). *Molecular Ecology*, *16*, 127–138. https://doi.org/10.1111/j.1365-294X.2006.03132.x

Palstra, F. P., & Ruzzante, D. E. (2008). Genetic estimates of contemporary effective population size: What can they tell us about the importance of genetic stochasticity for wild population persistence? *Molecular Ecology*, *17*, 3428–3447. https://doi.org/10.1111/j.1365-294X.2008.03842.x

Pereira, H. M., Ferrier, S., Walters, M., Geller, G. N., Jongman, R. H. G., Scholes, R. J., Bruford, M. W., Brummitt, N., Butchart, S. H. M., Cardoso, A. C., Coops, N. C., Dulloo, E., Faith, D. P., Freyhof, J., Gregory, R. D., Heip, C., Höft, R., Hurtt, G., Jetz, W., … Wegmann, M. (2013). Essential biodiversity variables. *Science*, *339*, 277–278. https://doi.org/10.1126/science.1229931

Pérez-Pereira, N., Wang, J., Quesada, H., & Caballero, A. (2022). Prediction of the minimum effective size of a population viable in the long term. *Biodiversity and Conservation*, *31*, 2763–2780. https://doi.org/10.1007/s10531-022-02456-z

Pudovkin, A. I., Zaykin, D. V., & Hedgecock, D. (1996). On the potential for estimating the effective number of breeders from heterozygote-excess in progeny. *Genetics*, *144*, 383–387. https://doi.org/10.1093/genetics/144.1.383

Pudovkin, A. I., Zhdanova, O. L., & Hedgecock, D. (2010). Sampling properties of the heterozygote-excess estimator of the effective number of breeders. *Conservation Genetics*, *11*, 759–771. https://doi.org/10.1007/s10592-009-9865-5

Reid, B. N., & Pinsky, M. L. (2022). Simulation-based evaluation of methods, data types, and temporal sampling schemes for detecting recent population declines. *Integrative and Comparative Biology*, *62*, 1849–1863. https://doi.org/10.1093/icb/icac144

Reis-Santos, P., Tanner, S. E., Aboim, M. A., Vasconcelos, R. P., Laroche, J., Charrier, G., Pérez, M., Presa, P., Gillanders, B. M., & Cabral, H. N. (2018). Reconciling differences in natural tags to infer demographic and genetic connectivity in marine fish populations. *Scientific Reports*, *8*, Article 10343. https://doi.org/10.1038/s41598-018-28701-6

Robinson, Z. L., Coombs, J. A., Hudy, M., Nislow, K. H., & Whiteley, A. R. (2024). Estimates of effective number of breeders identify drivers of decline in mid-Atlantic Brook Trout populations. *Evolutionary Applications*, *17*, Article e13769. https://doi.org/10.1111/eva.13769

Rockwood, L. L. (2015). *Introduction to population ecology* (2nd ed.). Wiley Blackwell.

Ruzzante, D. E., McCracken, G. R., Parmelee, S., Hill, K., Corrigan, A., MacMillan, J., & Walde, S. J. (2016). Effective number of breeders, effective population size and their relationship with census size in an iteroparous species, *Salvelinus fontinalis*. *Proceedings of the Royal Society B: Biological Sciences*, *283*, Article 20152601. https://doi.org/10.1098/rspb.2015.2601

Ryman, N., Baccus, R., Reuterwall, C., & Smith, M. H. (1981). Effective population size, generation interval, and potential loss of genetic variability in game species under different hunting regimes. *Oikos*, *36*, 257–266. https://doi.org/10.2307/3544622

Ryman, N., & Laikre, L. (1991). Effects of supportive breeding on the genetically effective population size. *Conservation Biology*, *5*, 325–329. https://doi.org/10.1111/j.1523-1739.1991.tb00144.x

Ryman, N., Laikre, L., & Hössjer, O. (2019). Do estimates of contemporary effective population size tell us what we want to know? *Molecular Ecology*, *28*, 1904–1918. https://doi.org/10.1111/mec.15027

Ryman, N., Laikre, L., & Hössjer, O. (2023). Variance effective population size is affected by census size in sub-structured populations. *Molecular Ecology Resources*, *23*, 1334–1347. https://doi.org/10.1111/1755-0998.13804

Santiago, E., & Caballero, A. (1995). Effective size of populations under selection. *Genetics*, *139*, 1013–1030. https://doi.org/10.1093/genetics/139.2.1013

Santiago, E., Caballero, A., Köpke, C., & Novo, I. (2024). Estimation of the contemporary effective population size from SNP data while accounting for mating structure. *Molecular Ecology Resources*, *24*, Article e13890. https://doi.org/10.1111/1755-0998.13890

Schmidt, C., Hoban, S., Hunter, M., Paz-Vinas, I., & Garroway, C. J. (2023). Genetic diversity and IUCN Red List status. *Conservation Biology*, *37*, Article e14064. https://doi.org/10.1111/cobi.14064

Secretariat of the Convention on Biological Diversity. (2011). *Convention on biological diversity: Text and annexes*. United Nations Environment Programme. https://www.cbd.int/doc/legal/cbd-en.pdf

Shrimpton, J. M., & Heath, D. D. (2003). Census vs. effective population size in Chinook Salmon: Large- and small-scale environmental perturbation effects. *Molecular Ecology*, *12*, 2571–2583. https://doi.org/10.1046/j.1365-294X.2003.01932.x

Slatkin, M. (1987). Gene flow and the geographic structure of natural populations. *Science*, *236*, 787–792. https://doi.org/10.1126/science.3576198

Slatkin, M. (2008). Linkage disequilibrium—Understanding the evolutionary past and mapping the medical future. *Nature Reviews Genetics*, *9*, 477–485. https://doi.org/10.1038/nrg2361

Tallmon, D. A., Gregovich, D., Waples, R. S., Scott Baker, C., Jackson, J., Taylor, B. L., Archer, E., Martien, K. K., Allendorf, F. W., & Schwartz, M. K. (2010). When are genetic methods useful for estimating contemporary abundance and detecting population trends? *Molecular Ecology Resources*, *10*, 684–692. https://doi.org/10.1111/j.1755-0998.2010.02831.x

Traill, L. W., Bradshaw, C. J. A., & Brook, B. W. (2007). Minimum viable population size: A meta-analysis of 30 years of published estimates. *Biological Conservation*, *139*, 159–166. https://doi.org/10.1016/j.biocon.2007.06.011

Trask, A. E., Bignal, E. M., McCracken, D. I., Piertney, S. B., & Reid, J. M. (2017). Estimating demographic contributions to effective population size in an age-structured wild population experiencing environmental and demographic stochasticity. *Journal of Animal Ecology*, *86*, 1082–1093. https://doi.org/10.1111/1365-2656.12703

U.S. Fish and Wildlife Service. (2023). *Status review: Lahontan Cutthroat Trout* (Oncorhynchus clarkii henshawi). https://ecos.fws.gov/ecp/report/species-five-year-review

Vucetich, J. A., & Waite, T. A. (1998). Number of censuses required for demographic estimation of effective population size. *Conservation Biology*, *12*, 1023–1030. https://doi.org/10.1046/j.1523-1739.1998.97328.x

Vucetich, J. A., Waite, T. A., & Nunney, L. (1997). Fluctuating population size and the ratio of effective to census population size. *Evolution*, *51*, 2017–2021. https://doi.org/10.2307/2411022

Wang, J. (2005). Estimation of effective population sizes from data on genetic markers. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *360*, 1395–1409. https://doi.org/10.1098/rstb.2005.1682

Wang, J. (2009). A new method for estimating effective population sizes from a single sample of multilocus genotypes. *Molecular Ecology*, *18*, 2148–2164. https://doi.org/10.1111/j.1365-294X.2009.04175.x

Wang, J. (2016). A comparison of single-sample estimators of effective population sizes from genetic marker data. *Molecular Ecology*, *25*, 4692–4711. https://doi.org/10.1111/mec.13725

Wang, J. (2022). MLNe: Simulating and estimating effective size and migration rate from temporal changes in allele frequencies. *Journal of Heredity*, *113*, 563–567. https://doi.org/10.1093/jhered/esac039

Wang, J. (2025). Estimating current effective sizes of large populations from a single sample of genomic marker data: A comparison of estimators by simulations. *Population Ecology*, *67*, 96–108. https://doi.org/10.1002/1438-390X.12167

Wang, J., Santiago, E., & Caballero, A. (2016). Prediction and estimation of effective population size. *Heredity*, *117*, 193–206. https://doi.org/10.1038/hdy.2016.43

Wang, J., & Whitlock, M. C. (2003). Estimating effective population size and migration rates from genetic samples over space and time. *Genetics*, *163*, 429–446. https://doi.org/10.1093/genetics/163.1.429

Wang, T., Fujiwara, M., Gao, X., & Liu, H. (2019). Minimum viable population size and population growth rate of freshwater fishes and their relationships with life history traits. *Scientific Reports*, *9*, Article 3612. https://doi.org/10.1038/s41598-019-40340-z

Waples, R. S. (1989). A generalized approach for estimating effective population size from temporal changes in allele frequency. *Genetics*, *121*, 379–391. https://doi.org/10.1093/genetics/121.2.379

Waples, R. S. (1990a). Conservation genetics of Pacific salmon. II. Effective population size and the rate of loss of genetic variability. *Journal of Heredity*, *81*, 267–276. https://doi.org/10.1093/oxfordjournals.jhered.a110989

Waples, R. S. (1990b). Conservation genetics of Pacific salmon. III. Estimating effective population size. *Journal of Heredity*, *81*, 277–289. https://doi.org/10.1093/oxfordjournals.jhered.a110990

Waples, R. S. (2002). Effective size of fluctuating salmon populations. *Genetics*, *161*, 783–791. https://doi.org/10.1093/genetics/161.2.783

Waples, R. S. (2005). Genetic estimates of contemporary effective population size: To what time periods do the estimates apply? *Molecular Ecology*, *14*, 3335–3352. https://doi.org/10.1111/j.1365-294X.2005.02673.x

Waples, R. S. (2006a). A bias correction for estimates of effective population size based on linkage disequilibrium at unlinked gene loci. *Conservation Genetics*, *7*, 167–184. https://doi.org/10.1007/s10592-005-9100-y

Waples, R. S. (2006b). Seed banks, salmon, and sleeping genes: Effective population size in semelparous, age-structured species with fluctuating abundance. *The American Naturalist*, *167*, 118–135. https://doi.org/10.1086/498584

Waples, R. S. (2016). Tiny estimates of the N_e/N ratio in marine fishes: Are they real? *Journal of Fish Biology*, *89*, 2479–2504. https://doi.org/10.1111/jfb.13143

Waples, R. S. (2021). Relative precision of the sibship and LD methods for estimating effective population size with genomics-scale datasets. *Journal of Heredity*, *112*, 535–539. https://doi.org/10.1093/jhered/esab042

Waples, R. S. (2022). What is N_e, anyway? *Journal of Heredity*, *113*, 371–379. https://doi.org/10.1093/jhered/esac023

Waples, R. S. (2024a). The N_e/N ratio in applied conservation. *Evolutionary Applications*, *17*, Article e13695. https://doi.org/10.1111/eva.13695

Waples, R. S. (2024b). Practical application of the linkage disequilibrium method for estimating contemporary effective population size: A review. *Molecular Ecology Resources*, *24*, Article e13879. https://doi.org/10.1111/1755-0998.13879

Waples, R. S. (2025). The idiot's guide to effective population size. *Molecular Ecology*. Advance online publication, https://doi.org/10.1111/mec.17670

Waples, R. S., & Antao, T. (2014). Intermittent breeding and constraints on litter size: Consequences for effective population size per generation (N_e) and per reproductive cycle (N_b). *Evolution*, *68*, 1722–1734. https://doi.org/10.1111/evo.12384

Waples, R. S., Antao, T., & Luikart, G. (2014). Effects of overlapping generations on linkage disequilibrium estimates of effective population size. *Genetics*, *197*, 769–780. https://doi.org/10.1534/genetics.114.164822

Waples, R. S., & Do, C. (2008). Ldne: A program for estimating effective population size from data on linkage disequilibrium. *Molecular Ecology Resources*, *8*, 753–756. https://doi.org/10.1111/j.1755-0998.2007.02061.x

Waples, R. S., & Do, C. (2010). Linkage disequilibrium estimates of contemporary N_e using highly variable genetic markers: A largely untapped resource for applied conservation and evolution. *Evolutionary Applications*, *3*, 244–262. https://doi.org/10.1111/j.1752-4571.2009.00104.x

Waples, R. S., & England, P. R. (2011). Estimating contemporary effective population size on the basis of linkage disequilibrium in the face of migration. *Genetics*, *189*, 633–644. https://doi.org/10.1534/genetics.111.132233

Waples, R. S., Grewe, P. M., Bravington, M. W., Hillary, R., & Feutry, P. (2018). Robust estimates of a high N_e/N ratio in a top marine predator, Southern Bluefin Tuna. *Science Advances*, *4*, Article eaar7759. https://doi.org/10.1126/sciadv.aar7759

Waples, R. K., Larson, W. A., & Waples, R. S. (2016). Estimating contemporary effective population size in non-model species using linkage disequilibrium across thousands of loci. *Heredity*, *117*, Article 4. https://doi.org/10.1038/hdy.2016.60

Waples, R. S., Luikart, G., Faulkner, J. R., & Tallmon, D. A. (2013). Simple life-history traits explain key effective population size ratios across diverse taxa. *Proceedings of the Royal Society B: Biological Sciences*, *280*, Article 20131339. https://doi.org/10.1098/rspb.2013.1339

Waples, R. S., Masuda, M., & Pella, J. (2007). Salmonnb: A program for computing cohort-specific effective population sizes (N_b) in Pacific salmon and other semelparous species using the temporal method.

Molecular Ecology Notes, *7*, 21–24. https://doi.org/10.1111/j.1471-8286.2006.01567.x

Waples, R. S., Scribner, K. T., Moore, J. A., Draheim, H. M., Etter, D., & Boersen, M. (2018). Accounting for age structure and spatial structure in eco-evolutionary analyses of a large, mobile vertebrate. *Journal of Heredity*, *109*, 709–723. https://doi.org/10.1093/jhered/esy018

Waples, R. S., & Teel, D. J. (1990). Conservation genetics of Pacific salmon I. Temporal changes in allele frequency. *Conservation Biology*, *4*, 144–156. https://doi.org/10.1111/j.1523-1739.1990.tb00103.x

Waples, R. S., Waples, R. K., & Ward, E. J. (2022). Pseudoreplication in genomic-scale data sets. *Molecular Ecology Resources*, *22*, 503–518. https://doi.org/10.1111/1755-0998.13482

Weise, E. M., Scribner, K. T., Adams, J. V., Boeberitz, O., Jubar, A. K., Bravener, G., Johnson, N. S., & Robinson, J. D. (2022). Pedigree analysis and estimates of effective breeding size characterize Sea Lamprey reproductive biology. *Evolutionary Applications*, *15*, 484–500. https://doi.org/10.1111/eva.13364

Whiteley, A. R., Coombs, J. A., Hudy, M., Robinson, Z., Nislow, K. H., & Letcher, B. H. (2012). Sampling strategies for estimating Brook Trout effective population size. *Conservation Genetics*, *13*, 625–637. https://doi.org/10.1007/s10592-011-0313-y

Whitlock, M. C. (2000). Fixation of new alleles and the extinction of small populations: Drift load, beneficial alleles, and sexual selection. *Evolution*, *54*, 1855–1861. https://doi.org/10.1111/j.0014-3820.2000.tb01232.x

Wright, S. (1931). Evolution in Mendelian populations. *Genetics*, *16*, 97–159. https://doi.org/10.1093/genetics/16.2.97

Wright, S. (1969). *The theory of gene frequencies* (Evolution and the genetics of populations, Volume 2). University of Chicago Press.

APPENDIX A: N_e KEY CONCEPTS

Table A1. Summary of key concepts central to N_e described in the main text. Key references associated with each concept are provided. This summary is not an exhaustive list of all the concepts related to N_e, nor a detailed description of the complexity and nuance associated with many of these concepts. Instead, it offers a quick reference of those key concepts necessary to facilitate conversations among fisheries practitioners with diverse areas of expertise who may collectively make important decisions about the need, feasibility, and basic experimental design of studies to infer N_e. Concepts and references are not provided for the section on Simulation-Based Exploration of Study Design as this section comprised a worked example rather than a concept review.

Manuscript section	Key concepts	Key references
WHAT IS N_e AND WHY IS IT IMPORTANT?		
N_e Overview	Original description of N_e.	Wright (1931)
	Effective population size reflects the susceptibility of a population to neutral and selective evolutionary forces that increase genetic drift and cause genetic diversity to be lost over time.	R. S. Waples (2022), R.S. Waples et al. (2014), and Whitlock (2000)
	Effective population size is typically smaller than N_c. These parameters are often correlated, although discordance between N_e and N_c is not uncommon.	Kalinowski and Waples (2002), Leffler et al. (2012), Luikart et al. (2010), Shrimpton and Heath (2003), and Vucetich et al. (1997)
HOW IS N_e MEASURED?		
Methodological Approaches	Review of methods to estimate historical N_e.	Nadachowska-Bryska et al. (2022)
	Temporal methods for estimating contemporary N_e require at least two sampling events from the same population separated by at least one generation. This sampling scheme is impractical for some populations (e.g., species with long generation times).	Krimbas and Tsakas (1971), Nei and Tajima (1981), J. Wang and Whitlock (2003), and R. S. Waples (1989)
	Single-sample methods for estimating contemporary N_e include measures based on linkage disequilibrium. Linkage disequilibrium-based estimators for N_e are the most widely implemented and evaluated single-sample methods for inferring N_e in populations targeted for conservation and management.	Barbato et al. (2015), Do et al. (2014), Hill (1981), Hollenbeck et al. (2016), Ovenden et al. (2007), Slatkin (2008), R. S. Waples (2006a, 2024a, 2024b), and R. S. Waples and Do (2008, 2010)
	Single-sample methods for estimating contemporary N_e also include measures based on sibship frequency.	J. Wang (2009, 2016, 2025) and R. S. Waples (2021)
	Single-sample methods for estimating contemporary N_e also include measures based on heterozygote excess.	Luikart and Cornuet (1999) and Pudovkin et al. (1996, 2010)
	Single-sample methods for estimating contemporary N_e also include measures based on molecular coancestry.	Nomura (2008)
Key Assumptions	The simplifying assumptions of an ideal population (upon which much population genetic theory is based, including concepts foundational to N_e) are usually unrealistic for wild populations, which experience gene flow, mutation, and selection.	Nunney (2002), Slatkin (1987), R. S. Waples (2005, 2006b, 2022, 2024a, 2024b), and Wright (1931)
	Basic assumptions of an ideal population include: mating among individuals is random (e.g., reproductive output is equal across individuals, without bias due to irregular reproductive cycles or age structure), generations are discrete (e.g., non-overlapping generations), the population sex ratio is equal, and population size is constant from one generation to the next.	Hill (1972), Nei and Tajima (1981), Ryman et al. (2023), R. S. Waples (2002, 2005, 2024a, 2024b), R. S. Waples et al. (2014), and R. S. Waples, Scribner, et al. (2018)
	Estimates of N_e assume a single genetically distinct population, but estimates based on linkage disequilibrium are generally robust to low levels of gene flow among populations (migration rates $<\sim$5–10%).	R. S. Waples and England (2011)

(*Continued*)

Table A1. Continued.

Manuscript section	Key concepts	Key references
HOW SHOULD STUDIES TO MEASURE N_e BE DESIGNED?		
Population Delimitation	Field sampling efforts should be designed to sample genetically distinct populations to reduce bias in linkage disequilibrium estimates of N_e.	Cramer et al. (2023), Reis-Santos et al. (2018), and Ryman et al. (2023)
	Abundance data may help inform field sampling of genetically distinct populations by identifying areas of high abundance that potentially correspond with distinct genetic populations and areas of low abundance that may reflect breaks between genetic populations.	Collie and Sissenwine (1983)
	Knowledge of movement barriers (e.g., waterfalls, dams, culverts) may help in evaluating the likely presence of genetically distinct populations.	Brunke et al. (2019), Funk et al. (2005), and Mamoozadeh et al. (2023)
	Information on environmental heterogeneity (e.g., temperature gradients, habitat diversity, salinity clines) potentially affecting genetic connectivity between distinct regions of the landscape may help in evaluating the likely presence of genetically distinct populations.	Keyghobadi et al. (2005) and Manel et al. (2003)
	Tagging or other methods to infer individual-based movements can provide information on the probable location and seasonal stability of genetically distinct populations.	Block et al. (2005) and Lindley et al. (2011)
	Traditional ecological knowledge can supply valuable information potentially indicative of genetically distinct populations and their seasonal locations.	Adams et al. (1993), Bayliss and Hutton (2017), Drew (2005), Drew and Henne (2006), and Henson et al. (2021)
Sample Sizes per Population	Populations exhibit wide-ranging life histories and different ecological and evolutionary histories, meaning that the minimum sample size needed to reliably estimate N_e will differ from one population to the next.	Clarke et al. (2024)
	Estimates of N_e generally exhibit lower levels of bias as sample size increases. This bias may be negligible when sample size exceeds two times N_e.	R. S. Waples (2006a)
	A common approach is to sample 50–100 individuals per population. However, smaller sample sizes may be sufficient for populations with low N_e, particularly when genomic data sets are used. Conversely, larger sample sizes will be necessary for populations with large N_e.	Marandel et al. (2020), Tallmon et al. (2010), R. S. Waples (2006a), R. S. Waples, Grewe, et al. (2018), and Whiteley et al. (2012)
Sample Composition	Pooling individuals sampled across field seasons may be suitable, but the pool of sampled individuals should represent the overall genetic diversity within a single genetic population. Pooling individuals across populations and generations will bias (potentially severely) N_e estimates, making them uninformative for conservation and management decision making.	Kardos and Waples (2024), Marandel et al. (2019), and Trask et al. (2017)
	If sampling occurs nonrandomly (e.g., by targeting only juveniles), family groups may be overrepresented, particularly in populations with limited dispersal capabilities, biasing N_e estimates.	Luikart et al. (2010)
Marker Selection	Retaining alleles that appear rare due to sampling error will upwardly bias N_e estimates. Erroneously rare alleles should be excluded prior to estimating N_e.	Do et al. (2014), Nunziata and Weisrock (2018), R. S. Waples and Do (2010)
	Baseline data sets that already exist for some populations may offer a useful comparison to newly produced data sets and therefore may influence which genetic markers are surveyed in subsequent studies.	Do et al. (2014), and Kazyak et al. (2022)
RELATIONSHIP OF N_e TO N_b		
	Effective number of breeders reflects the number of breeding individuals that gave rise to a single cohort and is useful for inferring and monitoring conservation status at the timescale of cohorts (rather than generations). However, N_e generally provides greater sampling flexibility (e.g., mixed cohorts versus a single cohort) and offers a better understanding of the evolutionary trajectory of a population.	Luikart et al. (2021), Ruzzante et al. (2016), R. S. Waples et al. (2013, 2014), and R. S. Waples and Antao (2014)
	Effective number of breeders potentially facilitates earlier detection of declining conservation status, given that the timescale associated with this metric is that of a cohort, rather than a generation. Declines across multiple reproductive cycles are presumed to produce a corresponding decline in N_e, reflecting longer term evolutionary trends.	R. S. Waples et al. (2013, 2014)
	For species with long generation times, N_b offers information on a time scale more relevant to fisheries management.	Ferchaud et al. (2016), R. S. Waples et al. (2013), and Whiteley et al. (2012)
	Because N_b corresponds with a single cohort, this parameter offers a practical way to control for overlapping generations, provided single cohort sampling is feasible.	J. Wang (2009), R. S. Waples (2005), and R. S. Waples et al. (2014)
	For linkage disequilibrium methods, estimates of N_b can be used to calculate N_e, provided the ratio of N_b to N_e is known. This ratio can be calculated from information on adult lifespan and age-at-maturity.	Ruzzante et al. (2016) and R. S. Waples et al. (2013, 2014)
CONSIDERATIONS FOR STUDIES THAT EMPLOY GENOMIC METHODS		
	In general, N_e estimates based on hundreds of SNPs may exhibit greater precision at smaller sample sizes than estimates based on tens of microsatellites.	Luikart et al. (2021)
	When sample sizes are low, the information gained by sampling additional individuals is oftentimes greater than the information gained from additional loci.	R. K. Waples et al. (2016) and R. S. Waples et al. (2022)
	For genome-scale data sets, the probability of surveying SNPs that are physically linked increases, which downwardly biases estimates of N_e. Genomic data sets with low sequence coverage can also bias N_e estimates. Biases due to physical linkage and low coverage can be controlled for by removing unreliable samples, SNPs, and genotypes prior to estimating N_e. Further, precision in N_e estimates derived from genomic data sets should be reflected using jackknife (rather than parametric) confidence intervals.	Jones et al. (2016), Luikart et al. (2021); Marandel et al. (2020), R. K. Waples et al. (2016), R. S. Waples (2024a, 2024b), R. S. Waples and Do (2008), and R. S. Waples et al. (2022)

(*Continued*)

Table A1. Continued.

Manuscript section	Key concepts	Key references
HOW IS N_e USED IN FISHERIES MANAGEMENT?		
Factors Influencing Decision Making	Decision making thresholds of $N_e = 50/500$ and $N_e = 100/1{,}000$ have frequently been used to distinguish populations at high/low risk of extirpation due to genetic diversity loss.	Frankham et al. (2014) and Franklin (1980)
	Simplifying the complex relationships reflected by N_e into general thresholds (e.g., the 50/500 and 100/1,000 thresholds) may lead to erroneous inference of extirpation risk. At worst, this may result in failure to identify populations experiencing actively declining N_e and/or high-risk populations that already exhibit perilously low N_e.	Clark et al. (2024), Clarke et al. (2024), Pérez-Pereira et al. (2022), Traill et al. (2007), T. Wang et al. (2019), and R. S. Waples (2024b)
	Populations with the same N_e may lose allelic diversity (an important component of genetic diversity) at very different rates if they differ in N_c. Allelic diversity thus provides an important complementary metric for monitoring genetic diversity loss.	Allendorf et al. (2024)
Illustrative Empirical Examples	Inference of N_e has informed decision making in diverse fisheries management contexts, including for wild and captive populations.	Baillie et al. (2016), Bertram et al. (2024), Christie et al. (2012), Robinson et al. (2024), Ryman and Laikre (1991), and U.S. Fish and Wildlife Service (2023)

APPENDIX B: MATHEMATICAL EXPRESSIONS FOR N_e

Here, we supply mathematical expressions that describe N_e based on inbreeding (Equation B1), allele frequency variance (Equation B2), and linkage disequilibrium (Equation B3). These expressions are intended to provide readers with examples of the different definitions of N_e discussed in Box 2 of the main text. Several alternative formulations are available, including for populations with demographic characteristics that differ from those described here. Descriptions for the symbology used below are listed in Table B1.

Table B1. Descriptions for the parameters used in Equations B1–B3.

Parameter	Description
N_f	Number of breeding females per generation
N_m	Number of breeding males per generation
N_{t-1}	Size of the population in the parental generation
$\bar{k}$	The mean number of progeny contributed by parents to the next generation
V_k	Variance in the mean number of progeny contributed by parents to the next generation
r^2	Estimator for the squared correlations of allele frequencies at pairs of loci
S	Number of sampled and genotyped individuals

Inbreeding N_e as defined by Wright (1931, 1969) for dioecious populations that are randomly mating:

$$\widehat{N}_e = \frac{4N_f N_m}{N_f + N_m}. \tag{B1}$$

Variance N_e as approximated by Kimura and Crow (1963) for dioecious populations that are randomly mating:

$$\widehat{N}_e = \frac{(N_{t-1} - 1)\bar{k}}{1 + \frac{V_k}{\bar{k}}}. \tag{B2}$$

Linkage disequilibrium N_e as approximated by R. S. Waples (2006a) and R. S. Waples and Do (2010) for monoecious or dioecious populations that are randomly mating:

$$\widehat{N}_e = \frac{1}{3(\hat{r}^2 - \frac{1}{S})}. \tag{B3}$$

APPENDIX C: METHODS FOR ESTIMATING N_e AND CORRESPONDING SOFTWARE PROGRAMS

Table C1. List of methods for estimating N_e and their corresponding software programs. Though not an exhaustive list of available programs, many of the most widely used programs for estimating N_e in wild populations are shown.

Estimator category	Sampling scheme	Time period for estimate	Software	References
Linkage disequilibrium	Within cohort	Point estimate	NeEstimator	R. S. Waples and Do (2010)
Heterozygote excess	Within cohort	Point estimate	NeEstimator	Luikart and Cornuet (1999)
Molecular coancestry	Within cohort	Point estimate	NeEstimator	Nomura (2008)
Temporal change	Among ≥2 cohorts	Multigeneration average	NeEstimator	Jorde and Ryman (2007) and Nei and Tajima (1981)
ABC	Within cohort	Point estimate	ONeSAMP	Hong et al. (2024)
Genome-wide linkage	Within cohort	Recent trajectory	GONE; SNeP	Barbato et al. (2015) and Coombs et al. (2012)
Sibship frequency	Within and/or among cohorts	Point estimate	COLONY	Jones and Wang (2010) and J. Wang (2009, 2016)
Neural net	Single sample	Point estimate	currentNe	Santiago et al. (2024)
Close kin mark recapture	Within and/or among cohorts	Point estimate	R	Babyn et al. (2024)
Temporal change	Among ≥3 cohorts	Multiple point estimates	SalmonNb	R. S. Waples et al. (2007)
Temporal change	Among ≥2 cohorts	Multigeneration average	MLNe	Hui and Burt (2015) and J. Wang (2022)