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Quantitative traits

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Summary

Quantitative traits are phenotypic traits that are determined, jointly with the environment, by many genes of small effect. Most of the directly observable individual characteristics are quantitative traits. Differences in quantitative traits can give valuable signals on existence of separate stock components. Compared to neutral genetic markers, quantitative traits are more sensitive to population structure, and may suggest differences where neutral genetic markers fail to show any. However, quantitative traits may also show differences where demographic separation is weak. An important challenge is to understand how the environment influences quantitative traits. Representing quantitative traits as reaction norms makes their dependence on the environment explicit and thereby facilitates their use in stock identification.

Key Words

Environmental, phenotypic plasticity, population genetics, reaction norm, stock structure

Scope of the chapter

Variation in quantitative traits can provide us with information about stock structure and the basis for distinguishing stock components. However, quantitative traits also display variability that has little to do with stock structure. Therefore, it is important to understand the sources of phenotypic variability, and how traits may differ in the extent to which they are influenced by factors that are not related to stock structure.

1 Introduction

Quantitative traits are phenotypic traits that are determined by many genes of small effect, in contrast to Mendelian traits where the phenotypic state is controlled by just one gene (Stearns, 1992). Most of the individual characteristics we can directly observe, such as body size and shape, fin ray and scale counts and many aspects of colouration, are quantitative traits, whereas classic Mendelian traits are comparatively rare.

Quantitative traits are measurable on an interval scale with many showing essentially continuous variability within a population whereas others are discrete but countable (i.e., the meristic traits). Some traits, such as age at maturation for species living in seasonal environments, are characterized by only few possible states (e.g., alternative male life histories; Gross, 1984). Such traits are threshold traits, which, despite their seemingly simple phenotypic expression reminiscent of Mendelian traits, are usually best understood as manifesting continuous variability in an underlying liability trait (Hazel et al., 1990; Roff, 1996). Liability traits are quantitative traits that determine, together with the environment, an individual's propensity to develop a certain, discrete phenotype.

Phenotypic differences in quantitative traits may allow us to visually distinguish biological populations even when their geographic ranges overlap. For example, Dannevig (1953) reported that fishermen on the Norwegian south coast traditionally distinguished three types of Atlantic cod (Gadus morhua) based on colouration and body proportions, namely the 'dyptorsk', 'taretorsk', and 'fjordtorsk' (deep-water, kelp, and fjord cod, respectively). However, Dannevig also pointed out a critical problem in using such characteristics to identify population structure, namely that phenotypic differences could result from differences in life history and environment, in addition to genetic differences. Dannevig (1953) did not try to resolve whether the different cod types represented 'only' ecotypes or different cod populations. Interestingly, later research using molecular genetic methods has been able to demonstrate that cod along the southern coast of Norway show complex population structure (e.g., Knutsen et al., 2003; Jorde et al., 2007), perhaps best characterized as a metapopulation. Nevertheless, the possible genetic basis of Dannevig's three cod types has not been specifically addressed. In other cases, population structure might be more cryptic and not manifested in any readily observable external characteristic. For example, herring in the North Sea look much the same irrespective of their origin. However, differences in characteristics such as vertebral number, spawning time and age distributions, and more recently, genetic markers, have shed light on herring population structure (Ruzzante et al., 2006; Bekkevold et al., 2011).

Whether variability in quantitative traits is obvious to a casual observer or only visible to a trained eye or detectible using advanced methods, interpreting differences in quantitative traits requires understanding the causative mechanisms. In this chapter, the sources of variability in quantitative traits are discussed in the context of stock identification.

2 Nature of variation in quantitative traits

In broad terms, variability in quantitative traits can be due to demographic, genetic, or environmental differences, or any combination of these. Any consistent difference between groups can be used to separate stocks (Waldman, 2005), but genetic differences represent a deeper and more permanent source of separateness than those due to demography and the environment that could, at least in principle, change rapidly. It is therefore important to understand how different processes contribute to variability in quantitative traits.

Variability due to demography

Individual characteristics change as individuals age. This happens most dramatically early in the ontogeny, which in most fish involves metamorphosis from larval stage to juvenile/adult stage (Chambers and Trippel, 1997). Sexual maturation may also involve marked external changes in the body and lead to sexual dimorphism (Parker, 1992). Today, these differences may be obvious to us, but in the past they have led to incorrect conclusions about population structure and, in more extreme cases, species assignment. For example, what are now recognized as juvenile male and female specimens of whalefish (family Cetomimidae), had been assigned to different fish families (Johnson et al., 2009). In the case of eels, 'leptocephalus' was originally a species name and only later became recognised as the larvae of eels and their relatives (Miller, 2009).

Even in the absence of drastic changes associated with metamorphosis, demographic composition of a population will influence the population-level averages of traits. For example, two populations that are otherwise identical but experience different overall mortality rates will have different mean ages, and consequently, have different means and variances for traits that are correlated with age. For example, comparing mean length in two groups of individuals can be misleading unless age differences are accounted for. Similarly, a trait such as parasite load often increases with fish age and size, reflecting the longer time older individuals have had to accumulate infections (Poulin, 2000).

The bottom line is that it is advisable to base comparisons of quantitative traits between groups of fish on metrics where the confounding effects of demography are removed or at least lessened. The simplest way of achieving this is to study specimens within some age or size bracket that is common for all groups. Another option is to use statistical techniques, i.e., regression analyses. For example, morphometric condition indices, such as the Fulton's condition factor, allow for comparing the shape of fish in a way that accounts for differences

in size. Of course, such statistical approaches will rarely be perfect in removing the confounding effects of demography. For example, Fulton's condition factor will be affected by differences in mean size whenever growth is not isometric.

Phenotypically plastic variability

Individuals that reside in different environments are generally expected to exhibit differences, even if they are genetically identical, due to of the unique conditions they experienced during development. This is because many individual characteristics are phenotypically plastic whereby a single genotype can develop different phenotypes in different environments (Sultan and Stearns, 2005). Phenotypic plasticity has profound implications for the use of quantitative traits to identify stocks and populations (Swain and Foote, 1999; Swain et al., 2005). Because the environment can induce differences in trait means and variances, these differences may not be indicative of population separateness and would vanish if the populations were exposed to identical conditions.

Thus, as with demography, phenotypic plasticity can have dramatic manifestations that can confound the interpretation of population structure.. For example, plasticity in brown trout Salmo trutta lead Linnaeus to describe stream trout, river trout and sea trout as three different species (respectively Salmo fario, S. trutta, and S. eriox); nowadays these are recognized as ecotypes of a single species (Jonsson and Jonsson, 2011). Trophic polymorphisms, where a single body of water hosts two or more morphs showing adaptations to different niches, are common in some freshwater fishes, notably in salmonids and in threespine stickleback (Gasterosteus aculeatus) and represent a mixture of plastic and genetic effects (Lavin and McPhail, 1986; Gíslason et al., 1999; Robinson and Parsons, 2002; Proulx and Magnan, 2004). In these cases, major differences in morphology may or may not indicate presence of reproductively isolated populations (Svanbäck and Schluter, 2012).

Types of phenotypic plasticity

Two types of phenotypic plasticity are distinguished: developmental conversion and phenotypic modulation (reviewed by Smith-Gill, 1983). Developmental conversion refers to a situation in which an environmental cue determines which genetic program of development an individual will follow. Developmental conversions typically occur during a specific stage of development, termed a 'developmental switchpoint', and are irreversible. The temperature effect on sex determination is an example of developmental conversion. Phenotypic modulation refers to a situation in which the environment influences rates or degrees of

expression of the developmental program. This can happen at any stage during development and may be reversible. The resulting phenotypic variability is typically continuous (although in the case of threshold or meristic traits it can also be discrete), and therefore of direct relevance for understanding variation in quantitative traits. Therefore, phenotypic modulation will therefore be the focus here, even though in practice it may not be easy to distinguish between these two types of plasticity.

Phenotypic plasticity may or may not be adaptive (Smith-Gill, 1983; Stearns, 1989; Via et al., 1995). Consider for example phenotypic effects of temperature. In crucian carp Carassius carassius, the respiratory surface area of the gills is positively affected by temperature (Sollid et al., 2005). Because meeting the oxygen demand is harder in warmer water, this plastic response appears adaptive. However, temperature can have phenotypic effects simply because biochemical reactions are temperature-dependent. In this latter case, plasticity would not be adaptive; rather, it would reflect the inability of an organism to buffer development against environmental change.

Drivers of plasticity

Almost any aspect of the environment can trigger plastic changes in the phenotype. The broad term "environment" can be split into micro-environments, unique to each individual, and to macro-environments that represents conditions shared by many individuals (Gavrilets and Hastings, 1994). An individual's micro-environment consists of its immediate external environment with random differences in nutrition, temperature, or other factors, and developmental noise due to the vagaries of embryonic development. As a consequence, even genetically identical individuals will show some phenotypic differences (though these may not be obvious to an observer). Because full characterization of an individual's micro-environment is impossible, and even its partial characterization is difficult, micro-environment can usually be seen as a source of unaccountable noise in the phenotype.

What we usually refer to with "environment" is the macro-environment. This environment has both physical and biological characteristics that influence phenotypes through plasticity. Many aspects of the physical environment are obvious and important: temperature, salinity, oxygen concentration, light, etc. Temperature and oxygen in particular have major influences on growth in aquatic organisms (Pauly, 2010), with further ramifications on other aspects of the phenotype. While these variables often correlate with many other aspects of the environment and teasing out their phenotypic effects in field data can be difficult, under experimental conditions they have separable effects. For example, in

sailfin mollies Poecilia latipinna, salinity and temperature influence growth and maturation (Trexler et al., 1990).

The biological environment is also powerful in triggering phenotypically plastic variability. An obvious source of plasticity is the feeding conditions: abundant food supply ensures rapid growth, and may also influence morphology (Tonn et al., 1994; Olsson et al., 2006). However, there are also more intricate effects, involving quality rather than quantity of the food. For instance, the type of available food influences the body morphology (Wimberger, 1992) and the feeding apparatus of cichlids (Bouton et al., 2002; Muschick et al., 2011). These are intriguing findings, given that such phenotypic differences are used in differentiating the species. Another aspect of the biological environment is the presence of potential predators. Crucian carp develop high-bodied morphology in presence of piscivorous fish (Brönmark and Miner, 1992; Holopainen et al., 1997). Nevertheless, while common in some other animal groups, inducible defences have only rarely been documented in fish (Chivers et al., 2008).

Environmental influences causing plastic changes can even occur before an individual is conceived; plastic effects can carry from parents to their offspring because of maternal effects, or more generally, trans-generational plasticity (Sultan and Stearns, 2005; Jablonka and Raz, 2009). Experimenters are well aware of this caveat. However, trans-generational plasticity can also be important for understanding phenotypic variability in the wild. For example, sheepshead minnow Cyprinodon variegatus parents can influence the phenotype of their offspring such that the growth of their offspring is the highest in the temperature the parents encountered prior to spawning (Salinas and Munch, 2012). In other words, parents seem able to adaptively 'program' their progeny to the environment they will likely encounter, whether this is common in nature remains to be seen.

Reaction norms

Reaction norms describe how a single genotype gives rise to different phenotypes in different environments. In other words, they describe phenotypic plasticity. Reaction norms describe genetically determined traits, and differences in reaction norms suggest genetic differences. This suggests that if we were able to measure reaction norms from wild fish, reaction norms would greatly ease the use of phenotypic data in stock identification. Notice that the role of environmental variability here is not a nuisance confounding interpretation of observations—it is the prerequisite for being able to observe reaction norms (Heino and Dieckmann, 2008).

In reality, however, there are two major challenges in estimating reaction norms from wild organisms. First, reaction norms are not directly observable at the individual level. A single individual can only be exposed to one particular set of environmental conditions over its lifetime, and it can only reveal part of a reaction norm. Reaction norms are therefore typically measured at the group or population level. The second challenge is that, in practice, reaction norms measured under uncontrolled (field) conditions cannot be interpreted as being purely genetically determined. Differences in reaction norms can always be caused by differences in those aspects of the environment that were not considered. Indeed, measuring reaction norms of wild populations is challenging. One would need to observe the phenotypes of individuals together with the environmental conditions they have experienced (Heino and Dieckmann, 2008). With data storage tags (DST), such coupled observations are becoming increasingly available. However, as not all salient environmental dimensions can be recorded by a DST, and because conditions before their implantation may also have been important, our ability to observe the 'full' environment of wild fish will remain limited.

One type of reaction norms, however, is often readily estimated from wild populations. Reaction norms for age and size at life-history transitions, such as maturation, are less influenced by some key problems that generally complicate reaction-norm estimation in the wild (Heino and Dieckmann, 2008). Reaction norms for age and size at maturation describe how age and size at maturation depend on growth, with the environmental variability being manifested as variations in growth trajectories. Information on the environment is obtained as a by-product of measuring age and length, because the joint measurement of these defines an individual's mean growth; growth is interpreted as a proxy of overall environmental quality that integrates many different influences. Thus, maturation reaction norms help to account for one aspect of environmental variability, and an important one (Alm, 1959), although other aspects can remain important.

As with other types of reaction norms, differences in reaction norms for age and size at maturation suggest genetic differences, although in field studies it is always possible that differences are caused by unaccounted aspects of the environment. For example, Morita et al. (2009) transplanted white-spotted charr Salvelinus leucomaenis fry of common origin to a number of locations, and observed that their reaction norms for age and size at maturation estimated from age 1+ fish were significantly different. While these differences could be due to viability selection after transplantation, it is likely that the description of the environment

inherent in reaction norms for age and size at maturation did not capture all important aspect of the environment.

Methods to estimate reaction norms for age and size at maturation with commonly available types of data are now readily available (reviewed in Heino and Dieckmann, 2008). While most studies have focused on temporal changes in maturation reaction norms (i.e., putative evolution), a number of studies have compared maturation reaction norms of adjacent stocks. For example, Barot et al. (2004) and Olsen et al. (2005) identified differences among cod stocks in the Northwest Atlantic, and Wang et al. (2008) found differences among lake whitefish Coregonus clupeaformis in the Laurentian Great Lakes. In these cases, maturation reaction norms corroborated prior understanding of stock structure. Vainikka et al. (2009), however, showed differences in maturation reaction norms of Baltic herring Clupea harengus membras at a finer scale than the commonly accepted stock structure.

Reaction norms of any kind are more readily estimated under experimental conditions. In an experiment one can choose to vary only certain aspects of the environment while keeping other aspects constant. Differences in reaction norms in a suitably designed experiment are therefore readily interpreted as manifesting genetic differences. For example, Haugen and Vøllestad (2000) demonstrated significant differences in temperature reaction norms for growth and survival during the period of first feeding among grayling Thymallus thymallus populations of recent common origin, suggesting local adaptation. The issue of experimentation is elaborated upon in Section 3.

Genetic variability

Genetic differences in neutral loci are usually considered as the 'true' indicators of stock structure. Here I briefly review the theoretical background necessary for understanding the utility of both neutral and non-neutral genetic markers for stock identification.

In the short term, there are three processes that are potentially important in influencing genetic variability over space: genetic drift, selection, and gene flow (e.g., Endler, 1986). Selection and drift are processes that occur within populations and subpopulations, whereas gene flow occurs among them. While drift tends to enhance differences among subpopulations, gene flow reduces differences; selection can act in both ways. In the long term, mutations will also contribute to genetic variability between subpopulations (of course, all genetic variability ultimately originates from mutations).

Genetic drift

Random genetic drift refers to the change in allele frequencies that is due to randomness in the transmission of genetic material from one generation to the next (Hartl and Clark, 1997). For neutral genetic markers, i.e., markers that are not influenced by natural selection, drift is the only process that can cause significant change in the short term. Because of random drift, even populations that are initially identical and live under identical conditions will become more and more different over time. Environment plays no role here, apart from influencing the population size. This is the basis for differentiating populations using neutral markers—they are not 'contaminated' by the environment.

The main challenge in using neutral markers to measure population separateness is that the rate of change in allele frequency under drift is inversely proportional to effective population size (e.g., Hartl and Clark, 1997). In practice this means that in small populations with few effective breeders, drift is quite significant, but its speed quickly declines with increasing effective population size. This does not render drift totally unimportant in moderately large populations, but it means that significant differences due to drift alone require many generations to develop (Hauser and Carvalho, 2008).

Selection

Selection is the phenomenon whereby certain phenotypes have higher net reproductive rates than others. Selection is based on phenotypic differences. When these differences have some genetic basis, selection also results in genetic differences as reflected in non-neutral markers. Selection can be separated into viability selection whereby fitness differences are due to differences in survival between phenotypes, and to fertility selection whereby differences arise from differential reproductive rates among individuals that have reached maturity. Genetic change caused by viability selection can accrue fast and become visible within the life-time of a single cohort, even in organisms with long generation time. Thus, groups of individuals originating from the same spawning (thereby representing the same parental gene pool) can become genetically different as they grow older if they are exposed to different selective environments (e.g., temperature, predation regime). However, they would still be similar with respect to neutral genetic markers, provided that the individual groups are large enough for drift not to be significant.

Gene flow

Gene flow refers to transfer of genes from one subpopulation or population to another. This is mediated through dispersal that can occur at different life stages. Many fish have pelagic eggs and larvae that can be passively transported over long distances by currents. Juvenile and adult fish can have active dispersal or migratory behaviours. In the absence of perfect natal homing and geographic barriers and assuming that successful mating will follow, dispersal will lead to transfer of genetic material from one population to another. The consequence of such gene flow is to reduce genetic differences between source and recipient populations.

Rates of dispersal are difficult to estimate, especially for types of dispersal that are rare (e.g., long-distance dispersal). Rare long-distance dispersal may be ecologically insignificant, yet it may have important genetic consequences (Kinlan et al., 2005): if genetic drift is weak, as it is in large populations, even rare dispersal events suffice to offset its effects.

Because drift is the only driver of differentiation in neutral markers, neutral markers are insensitive to stock structure whenever populations are large, as they tend to be in the marine realm (Hauser and Carvalho, 2008). On the other hand, because ongoing selection is a potent source of genetic differentiation also in large populations, non-neutral markers can show substantial differentiation even in presence of significant levels of gene flow (Nielsen et al., 2009).

3 Disentangling sources of phenotypic variation

We have now discussed different sources of variation in quantitative traits. It is clear that it would be desirable to be able to disentangle the contributions of different components. There are a number of ways of doing this. Below is a simple categorization of approaches, with some overlap between them.

Sample standardization

The simplest way of reducing the confounding effects of demography and the environment when comparing contingents (here defined as groups of fish that co-occur in space and time, and that are candidates for being separate populations) is to compare individuals that are as similar as possible. To account for demographic effects, only individuals with similar age or size should be compared. To account for environmental effects, samples should be collected from as similar habitats as possible. For example, in a species with benthic and limnetic morphs such as Arctic charr Salvelinus alpinus (Gíslason et al., 1999), inter-lake comparisons

will be more meaningfully if fish were collected from the same habitat. However, it is obvious that such sample standardization has quite limited power to control for confounding effects.

Statistical methods

Statistical methods enable us to describe the dependence of a quantitative trait on explanatory variables. Typically, these explanatory variables are either individual variables (e.g., size) or environmental variables (e.g., temperature). Including individual variables allows accounting for demographic differences (as described in the example in Section 0), and environmental variables allows accounting for environmental effects (i.e., phenotypic plasticity or selection).

When information on pedigrees is available, it is possible to partition variability to environmental and genetic components using quantitative genetics methods (see section 0). These methods tend to be data-hungry and work best in experiments where parentage can be controlled. However, even with incomplete parentage data it is still possible to include this information as an explanatory variable in a multiple regression model. For example, when individuals can be assigned to families defined by mothers, but paternity is unknown, family can be used as an explanatory variable. This allows reducing unaccounted variability, but not fully disentangling the genetic component. The variability assigned to families reflects not only their shared genetic material but also shared environmental effects, including trans-generational plasticity.

Without pedigree data—the typical situation when studying populations in the wild—classic multiple regression models can be used. Here, the goal is to test whether contingent-specific differences exist that cannot be explained by other explanatory variables. When the explanatory variables include the key drivers of variability in the quantitative trait(s) in question but significant contingent-specific residual differences remain, the result suggests genetically-determined differences, possibly reflecting local adaptation. However, this cannot be strictly proven as one can never be sure that all salient explanatory variables have been included. Thus, contingent-specific residual differences are attributed to a mixture of environmental and genetic effects.

Interpretation of statistical results requires care as models tend to highlight a single answer even when data allow for alternative interpretations. If observations of quantitative traits include little variability in explanatory variables, such as temperature, within contingents, then the temperature and contingent terms in the model are statistically confounded (Figure 1, top row). A statistical model will provide an answer, attributing some

proportion of variability to the temperature effect and the rest to contingent-specific deviations; the latter could represent either local adaptation or unaccounted for environmental effects. However, without additional information, the interpretation of the temperature effect remains ambiguous. It could be fully coincidental (spurious; Figure 1B), fully biologically founded (Figure 1C), or anything in between. This is unfortunate because a spurious temperature effect would conceal variability that is in fact contingent-specific (possibly genetic), possibly leading to type II error (population structure exists but is not detected). In contrast, if the temperature effect is biologically founded, then the contingent-specific residual component is correctly estimated. Type I error (a structure is declared to exist when in fact it is absent) can still occur if the residual variability is entirely due to an accounted environmental factor.

Variability in the explanatory variable within contingents can help in the interpretation of between-contingent patterns (Figure 1, bottom row). A within-contingent pattern can be interpreted as a reaction norm (albeit observed at group rather than genotype level). If the within-contingent reaction norm shows a similar pattern as the global pattern, then it is likely a manifestation of the same reaction norm, and the statistical relationship has meaningful biological interpretation (Figure 1D). If, however, the reaction norms observed for the contingents differ from the global pattern, then the global pattern is driven by contingent-specific effects (Figure 1E). As before, these could be either genetic or due to unaccounted environmental factors. Finally, within-contingent patterns can reveal counter-gradient variation (Conover and Schultz, 1995) where contingent-specific genetic influences on a trait oppose between-contingent environmental influences, leading to reduced phenotypic variation across the gradient (Figure 1F).

Reaction norms for age and size at life-history transitions are a particular class of statistical, regression-based methods. The appeal of reaction norms is that they do not require knowing the environment beyond what is already captured by the individual-level knowledge on age and size (though additional information can be used) (Dieckmann and Heino, 2007; Heino and Dieckmann, 2008). As with other regression-based methods, care is needed in assigning residual patterns to genetic differences: unaccounted environmental factors can introduce spurious differences, but they could also hide real differences.

Genetic methods

Using molecular genetic methods, it is possible to directly probe a single component of phenotypic variability. Showing a difference in a gene known to be related to variation in a

quantitative trait is proof that genetic differences are contributing to phenotypic differences in the trait. However, this does not indicate how large the genetic effect is on the trait. Proving the opposite (that there is no genetic effect) is more difficult, though not fundamentally impossible. Any single quantitative trait is likely influenced by many genes and regulatory factors, only some of which are likely covered in genotyping of the sampled individuals. Thus, failing to find significant genetic differences could be a result of not looking in the right places. Mapping from genotype to phenotype is a complex task; however, with the advancement of molecular genetics and the associated declines in costs this will become easier.

By definition, neutral genetic markers do not tell anything about phenotypic traits themselves, but they can help interpreting the variability in them. If neutral markers show significant differentiation, then it is likely (but not certain) that genetic differences in phenotypic traits also exist.

Quantification of the genetic contribution is possible through quantitative genetics methods that do not require knowing the genetic basis of traits (Falconer and Mackay, 1996; Hartl and Clark, 1997; Wilson et al., 2010). Because the more individuals share genetic material, the more alike they are likely to be, it is possible to estimate the relative contributions of genetic and environmental factors using knowledge about the relationships among individuals in a sample and data on quantitative traits. These methods require pedigree data, which are easiest to achieve in experiments (see section 0). If parentage can be determined with genotyping, the methods are in principle also available for wild animals (Blouin, 2003), although in practice this will only be possible when populations are small.

Experimental approaches

In experimental settings, the experimenter can either standardize the environment or characterize the environment in ways that are impossible in the wild. Thus, the confounding effects of the environment can be either eliminated or statistically controlled. Moreover, the experimenter can induce variation in certain aspects of the environment so that reaction norms with respect to that environmental dimension can be estimated (See Section 0).

In common-garden experiments, individuals are raised under standardized conditions. Thus, the confounding effects of the environment are excluded. The age of experimental animals is usually known, their initial numbers are standardized, and mortality at later stages is either largely eliminated or kept at some ‘standard’ level. Therefore,, the confounding

effect of demography can also, for the most part, be eliminated. The only remaining complication is that environmental effects may persist from one generation to the next, reflecting trans-generational plasticity (Sultan and Stearns, 2005; Jablonka and Raz, 2009), which include the well-known maternal effects. To avoid this, it is usual that common garden experiments are run for two (or more) generations in captivity. Thus, offspring (F1) are collected from wild-born individuals and raised under standard conditions until they produce offspring (F2). These are again raised under standard conditions, and their traits are then measured under these same conditions. Phenotypic differences that persist after two generations in captivity are very likely of genetic origin.

Common-garden experiments are a very powerful tool as they can provide evidence of genetic differences without requiring knowledge about the genetic basis of quantitative traits. The downside is that they are laborious and can take a long time, especially when run for two captive generations. For larger species or for species with longer generation times, the scale and costs of the necessary experimental facilities may be prohibitive. Furthermore, experience for carrying out full life-cycle experiments does not exist for most species.

Suitably designed quantitative genetic experiments also allow partitioning of phenotypic variability into genetic and environmental components (Hartl and Clark, 1997). Compared with common-garden experiments, this requires additionally that parentage is known; this is best achieved with a suitable mating design but also less controlled structures can be used as long as pedigrees can be constructed (Wilson et al., 2010). Again, constrains of animal husbandry are often far more restrictive than the fundamental power of these methods.

Quantifying differences between contingents

In the end, it would be desirable to be able to quantify the degree of genetic differentiation among populations displayed by quantitative traits. For neutral genetic markers, it is usual to describe differentiation with the fixation index, F_{ST}. An analogous metric can be defined for quantitative traits and is known as Q_{ST} (Merilä and Crnokrak, 2001; McKay and Latta, 2002). In principle, this index allows using information on quantitative traits to highlight situations with relatively high levels of differentiation between contingents, similarly as F_{ST} is used with neutral genetic markers. Importantly, experience cross many different systems shows that divergence in quantitative traits is usually larger than in neutral markers (Leinonen et al., 2008), suggesting that neutral markers tend to underestimate differentiation in non-neutral loci.

In practice, utility of Q_{ST} in stock identification is probably limited. First, Q_{ST} estimates are likely to have poor precision in situations where the number of contingents is <20 (O'Hara and Merilä, 2005), which is typical for stock identification problems. Second, estimation of Q_{ST} requires quantitative genetic experiments, which are not feasible for many marine species. This requirement can be circumvented if Q_{ST} is approximated by the phenotypic divergence in a trait across populations, sometimes referred to as P_{ST}. Although comparative studies do not suggest systematic differences between Q_{ST} and P_{ST} (Leinonen et al., 2008), it is very difficult to assess how good (or bad) this approximation is (Brommer, 2011). Nevertheless, while not offering a solution to stock identification based on quantitative traits, Q_{ST} and P_{ST} can be useful metrics to describe degree of quantitative trait differentiation among populations.

4 Conclusions

Groups of fish can exhibit differences in quantitative traits for a number of reasons: 1) differences in demographic structure, 2) differences in their environment, or 3) genetic differences. Genetic differences in neutral loci are often considered as the 'true' indicators of stock structure because such differences indicate that gene flow between stock units is negligible, and consequently, that the units can be considered reproductively isolated. Thus, finding such differences is indeed an unambiguous sign of population separateness. Yet it can be argued that while genetic differences are a sufficient condition for considering contingents as separate populations, genetic differences in neutral markers are not a necessary condition for biologically significant structures to exist (Waples, 1998; Waldman, 2005; Hauser and Carvalho, 2008). The argument here is two-fold.

First, in large populations, the process responsible for creating differences in neutral loci, genetic drift, is weak, often practically absent. Low rates of gene flow suffice to counter the heterogeneity caused by drift. The power of neutral genetic markers to demonstrate population structure depends on population sizes in question, becoming extremely weak for large populations. While effective population sizes can be far smaller than census population sizes (Hauser and Carvalho, 2008; Palstra and Ruzzante, 2008), marine species of commercial importance typically have population sizes such that neutral genetic markers are insensitive to population structure. Thus, populations can in practical terms be reproductively isolated without neutral genetic markers showing it.

Second, biologically-relevant population structures may exist even when the contingents are not completely reproductively isolated. Within-population structure can be important in fisheries management if the response of subpopulations to exploitation or other

external drivers shows a large degree of independence. While dispersal has an overall effect of synchronizing and stabilizing dynamics of sub-populations (reviewed by Abbott, 2011), dynamics can remain virtually independent at levels of dispersal that are high enough to be practically observable in genetics and tagging studies (i.e., exchange rates in excess of few percent per generation). There is a large range of intermediate dispersal rates that are too low to have much impact on population dynamics, yet they are too high for significant differentiation in neutral genetic markers in all but small populations. This range of dispersal rates also allows for local adaptation. Indeed, adaptive local evolution is common in marine fishes, despite low levels of population differentiation in neutral markers (Nielsen et al., 2009). Thus, neutral markers are often too conservative in stock identification, resulting in a rate of type II errors that is not precautionary.

The arguments presented above suggest a greater role for use of quantitative traits and genetic markers under selection in stock identification. Variation in quantitative traits can be used as a surrogate of genetic differences, provided that care is taken to remove the effects of demography and major environmental drivers. This involves a possibility that 'stocks' that are defined do not correspond to biological populations, but rather to subpopulations. Sometimes, this also means that too much stock structure is identified relative to what sustainable fisheries management really requires. While such redundant structure complicates management and may lead to inefficiency, the possible consequences of ignoring important stock structure are graver still, including exhaustion of less productive stock components.

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5 References

Abbott, K. C. (2011). A dispersal-induced paradox: synchrony and stability in stochastic metapopulations. Ecol. Lett., 14, 1158–1169.

Alm, G. (1959). Connection between maturity, size, and age in fishes. Rep. Inst. Freshw. Res. Drottningholm, 40, 5–145.

Barot, S., Heino, M., O'Brien, L., and Dieckmann, U. (2004). Long-term trend in the maturation reaction norm of two cod stocks. Evol. Appl., 14, 1257–1271.

Bekkevold, D., Clausen, L. A. W., Mariani, S., André, C., Hatfield, E. M. C., Torstensen, E., Ryman, N., Carvalho, G. R., and Ruzzante, D. E. (2011). Genetic mixed-stock analysis of Atlantic herring populations in a mixed feeding area. Mar. Ecol. Prog. Ser., 442, 187–199.

Blouin, M. S. (2003). DNA-based methods for pedigree reconstruction and kinship analysis in natural populations. Trends Ecol. Evol., 18, 503–511.

Bouton, N., Witte, F., and Van Alphen, J. J. M. (2002). Experimental evidence for adaptive phenotypic plasticity in a rock-dwelling cichlid fish from Lake Victoria. Biol. J. Linn. Soc., 77, 185–192.

Brommer, J. E. (2011). Whither P_{ST}? The approximation of Q_{ST} by P_{ST} in evolutionary and conservation biology. J. Evol. Biol., 24, 1160–1168.

Brönmark, C., and Miner, J. G. (1992). Predator-induced phenotypical change in body morphology in crucian carp. Science, 258, 1348–1350.

Chambers, R. C., and Trippel, E. A. (1997). Early Life History and Recruitment in Fish Populations. Chapman and Hall, London. 638 pp.

Chivers, D., Zhao, X., Brown, G., Marchant, T., and Ferrari, M. (2008). Predator-induced changes in morphology of a prey fish: the effects of food level and temporal frequency of predation risk. Evol. Ecol., 22, 561–574.

Conover, D. O., and Schultz, E. T. (1995). Phenotypic similarity and the evolutionary significance of countergradient selection. Trends Ecol. Evol., 10, 248–252.

Dannevig, A. (1953). The littoral cod of the Norwegian Skagerak coast. ICES Document CM 1953/8. 15 pp.

Dieckmann, U., and Heino, M. (2007). Probabilistic maturation reaction norms: their history, strengths, and limitations. Mar. Ecol. Prog. Ser., 335, 253–269.

Endler, J. A. (1986). Natural Selection in the Wild. Princeton University Press, Princeton.

Falconer, D. S., and Mackay, T. F. C. (1996). Introduction to Quantitative Genetics. Longman, Essex.

Gavrilets, S., and Hastings, A. (1994). A quantitative-genetic model for selection on developmental noise. Evolution, 48, 1478–1486.

Gíslason, D., Ferguson, M. M., Skúlason, S., and Snorrason, S. S. (1999). Rapid and coupled phenotypic and genetic divergence in Icelandic Arctic char (Salvelinus alpinus). Can. J. Fish. Aquat. Sci., 56, 2229–2234.

Gross, M. R. (1984). Sunfish, salmon, and the evolution of alternative reproductive strategies and tactics in fishes. In "Fish Reproduction: Strategies and Tactics" (G.W. Potts and R.J. Wootton, Eds.), pp. 55–75. Academic Press, London.

Hartl, D. L., and Clark, A. G. (1997). Principles of Population Genetics. 3rd edn. Sinauer Associates. 542 pp.

Haugen, T. O., and Vøllestad, L. A. (2000). Population differences in early life-history traits in grayling. J. Evol. Biol., 13, 897–905.

Hauser, L., and Carvalho, G. R. (2008). Paradigm shifts in marine fisheries genetics: ugly hypotheses slain by beautiful facts. Fish Fish., 9, 333–362.

Hazel, W. N., Smock, R., and Johnson, M. D. (1990). A polygenic model for the evolution and maintenance of conditional strategies. Proc. R. Soc. Lond. B, 242, 181–187.

Heino, M., and Dieckmann, U. (2008). Detecting fisheries-induced life-history evolution: an overview of the reaction norm approach. Bull. Mar. Sci., 83, 69–93.

Holopainen, I. J., Aho, J., Vornanen, M., and Huuskonen, H. (1997). Phenotypic plasticity and predator effects on morphology and physiology of crucian carp in nature and in the laboratory. J. Fish Biol., 50, 781–798.

Jablonka, E., and Raz, G. (2009). Transgenerational epigenetic inheritance: prevalence, mechanisms, and implications for the study of heredity and evolution. Q. Rev. Biol., 84, 131–176.

Johnson, G. D., Paxton, J. R., Sutton, T. T., Satoh, T. P., Sado, T., Nishida, M., and Miya, M. (2009). Deep-sea mystery solved: astonishing larval transformations and extreme sexual dimorphism unite three fish families. Biol. Lett., 5, 235–239.

Jonsson, B., and Jonsson, N. (2011). Ecology of Atlantic Salmon and Brown Trout. Springer Netherlands, Dordrecht. 708 pp.

Jorde, P. E., Knutsen, H., Espeland, S. H., and Stenseth, N. C. (2007). Spatial scale of genetic structuring in coastal cod Gadus morhua and geographic extent of local populations. Mar. Ecol. Prog. Ser., 343, 229–237.

Kinlan, B. P., Gaines, S. D., and Lester, S. E. (2005). Propagule dispersal and the scales of marine community process. Divers. Distrib., 11, 139–148.

Knutsen, H., Jorde, P. E., André, C., and Stenseth, N. C. (2003). Fine-scaled geographical population structuring in a highly mobile marine species: the Atlantic cod. Mol. Ecol., 12, 385–394.

Lavin, P. A., and McPhail, J. D. (1986). Adaptive divergence of trophic phenotype among freshwater populations of the threespine stickleback (Gasterosteus aculeatus). Can. J. Fish. Aquat. Sci., 43, 2455–2463.

Leinonen, T., O'hara, R. B., Cano, J. M., and Merilä, J. (2008). Comparative studies of quantitative trait and neutral marker divergence: a meta-analysis. J. Evol. Biol., 21, 1–17.

McKay, J. K., and Latta, R. G. (2002). Adaptive population divergence: markers, QTL and traits. Trends Ecol. Evol., 17, 285–291.

Merilä, J., and Crnokrak, P. (2001). Comparison of genetic differentiation at marker loci and quantitative traits. J. Evol. Biol., 14, 892–903.

Miller, M. J. (2009). Ecology of anguilliform leptocephali: remarkable transparent fish larvae of the ocean surface layer. Aqua-BioScience Monographs, 2, 1–94.

Morita, K., Tsuboi, J., and Nagasawa, T. (2009). Plasticity in probabilistic reaction norms for maturation in a salmonid fish. Biol. Lett., 5, 628–631.

Muschick, M., Barluenga, M., Salzburger, W., and Meyer, A. (2011). Adaptive phenotypic plasticity in the Midas cichlid fish pharyngeal jaw and its relevance in adaptive radiation. BMC Evol. Biol., 11, 116.

Nielsen, E. E., Hemmer-Hansen, J., Larsen, P. F., and Bekkevold, D. (2009). Population genomics of marine fishes: identifying adaptive variation in space and time. Mol. Ecol., 18, 3128–3150.

O'Hara, R. B., and Merilä, J. (2005). Bias and precision in Q_{ST} estimates: problems and some solutions. Genetics, 171, 1331–1339.

Olsen, E. M., Lilly, G. R., Heino, M., Morgan, M. J., Brattey, J., and Dieckmann, U. (2005). Assessing changes in age and size at maturation in collapsing populations of Atlantic cod (Gadus morhua). Can. J. Fish. Aquat. Sci., 62, 811–823.

Olsson, J., Svanbäck, R., and Eklöv, P. (2006). Growth rate constrain morphological divergence when driven by competition. Oikos, 115, 15–22.

Palstra, F. P., and 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? Mol. Ecol., 17, 3428–3447.

Parker, G. A. (1992). The evolution of sexual dimorphism in fish. J. Fish Biol., 41, 1–20.

Pauly, D. (2010). Gasping Fish and Panting Squids: Oxygen, Temperature and the Growth of Water-Breathing Animals (O. Kinne, Ed.). Excellence in Ecology 22. International Ecology Institute, Oldendorf/Luhe. 216 pp.

Poulin, R. (2000). Variation in the intraspecific relationship between fish length and intensity of parasitic infection: biological and statistical causes. J. Fish Biol., 56, 123–137.

Proulx, R., and Magnan, P. (2004). Contribution of phenotypic plasticity and heredity to the trophic polymorphism of lacustrine brook charr (Salvelinus fontinalis M.). Evol. Ecol. Res., 6, 503–522.

Robinson, B. W., and Parsons, K. J. (2002). Changing times, spaces, and faces: tests and implications of adaptive morphological plasticity in the fishes of northern postglacial lakes. Can. J. Fish. Aquat. Sci., 59, 1819–1833.

Roff, D. A. (1996). The evolution of threshold traits in animals. Q. Rev. Biol., 71, 3–35.

Ruzzante, D. E., Mariani, S., Bekkevold, D., André, C., Mosegaard, H., Clausen, L. A. W., Dahlgren, T. G., Hutchinson, W. F., Hatfield, E. M. C., Torstensen, E., Brigham, J., Simmonds, E. J., Laikre, L., Larsson, L. C., Stet, R. J. M., Ryman, N., and Carvalho, G. R. (2006). Biocomplexity in a highly migratory pelagic marine fish, Atlantic herring. Proc. R. Soc. B, 273, 1459–1464.

Salinas, S., and Munch, S. B. (2012). Thermal legacies: transgenerational effects of temperature on growth in a vertebrate. Ecol. Lett., 15, 159–163.

Smith-Gill, S. J. (1983). Developmental plasticity: developmental conversion versus phenotypic modulation. Am. Zool., 23, 47–55.

Sollid, J., Weber, R. E., and Nilsson, G. E. (2005). Temperature alters the respiratory surface area of crucian carp Carassius carassius and goldfish Carassius auratus. J. Exp. Biol., 208, 1109–1116.

Stearns, S. C. (1989). The evolutionary significance of phenotypic plasticity. BioScience, 39, 436–445.

Stearns, S. C. (1992). The Evolution of Life Histories. Oxford University Press, Oxford, UK. 249 pp.

Sultan, S. E., and Stearns, S. C. (2005). Environmentally contingent variation: phenotypic plasticity and norms of reaction. In "Variation" (B. Hallgrimsson and B. Hall, Eds.), pp. 303–332. Elsevier Academic Press, Boston.

Svanbäck, R., and Schluter, D. (2012). Niche specialization influences adaptive phenotypic plasticity in the threespine stickleback. Am. Nat., 180, 50–59.

Swain, D. P., and Foote, C. J. (1999). Stocks and chameleons: the use of phenotypic variation in stock identification. Fish. Res., 43, 113–128.

Swain, D. P., Hutchings, J. A., and Foote, C. J. (2005). Environmental and genetic influences on stock identification characters. In "Stock Identification Methods. Applications in Fishery Science" (S.X. Cadrin, K. Friedland, and J. Waldman, Eds.), pp. 45–85. Elsevier.

Tonn, W. M., Holopainen, I. J., and Paszkowski, C. A. (1994). Density-dependent effects and the regulation of crucian carp populations in single-species ponds. Ecology, 75, 824–834.

Trexler, J. C., Travis, J., and Trexler, M. (1990). Phenotypic plasticity in the sailfin molly, Poecilia latipinna (Pisces: Poeciliidae). II. laboratory experiment. Evolution, 44, 157–167.

Vainikka, A., Mollet, F. M., Casini, M., and Gårdmark, A. (2009). Spatial variation in growth, condition and maturation reaction norms of the Baltic herring Clupea harengus membras. Mar. Ecol. Prog. Ser., 383, 285–294.

Via, S., Gomulkiewicz, R., De Jong, G., Scheiner, S. M., Schlichting, C. D., and Van Tienderen, P. H. (1995). Adaptive phenotypic plasticity: consensus and controversy. Trends Ecol. Evol., 10, 212–217.

Waldman, J. R. (2005). Definition of stocks: an evolving concept. In "Stock Identification Methods. Applications in Fishery Science" (S.X. Cadrin, K. Friedland, and J. Waldman, Eds.), pp. 7–16. Elsevier.

Wang, H.-Y., Höök, T. O., Ebener, M. P., Mohr, L. C., and Schneeberger, P. J. (2008). Spatial and temporal variation of maturation schedules of lake whitefish (Coregonus clupeaformis) in the Great Lakes. Can. J. Fish. Aquat. Sci., 65, 2157–2169.

Waples, R. S. (1998). Separating the wheat from the chaff: patterns of genetic differentiation in high gene flow species. J. Hered., 89, 438–450.

Wilson, A. J., Réale, D., Clements, M. N., Morrissey, M. M., Postma, E., Walling, C. A., Kruuk, L. E. B., and Nussey, D. H. (2010). An ecologist's guide to the animal model. J. Anim. Ecol., 79, 13–26.

Wimberger, P. H. (1992). Plasticity of fish body shape. The effects of diet, development, family and age in two species of Geophagus (Pisces: Cichlidae). Biol. J. Linn. Soc., 45, 197–218.

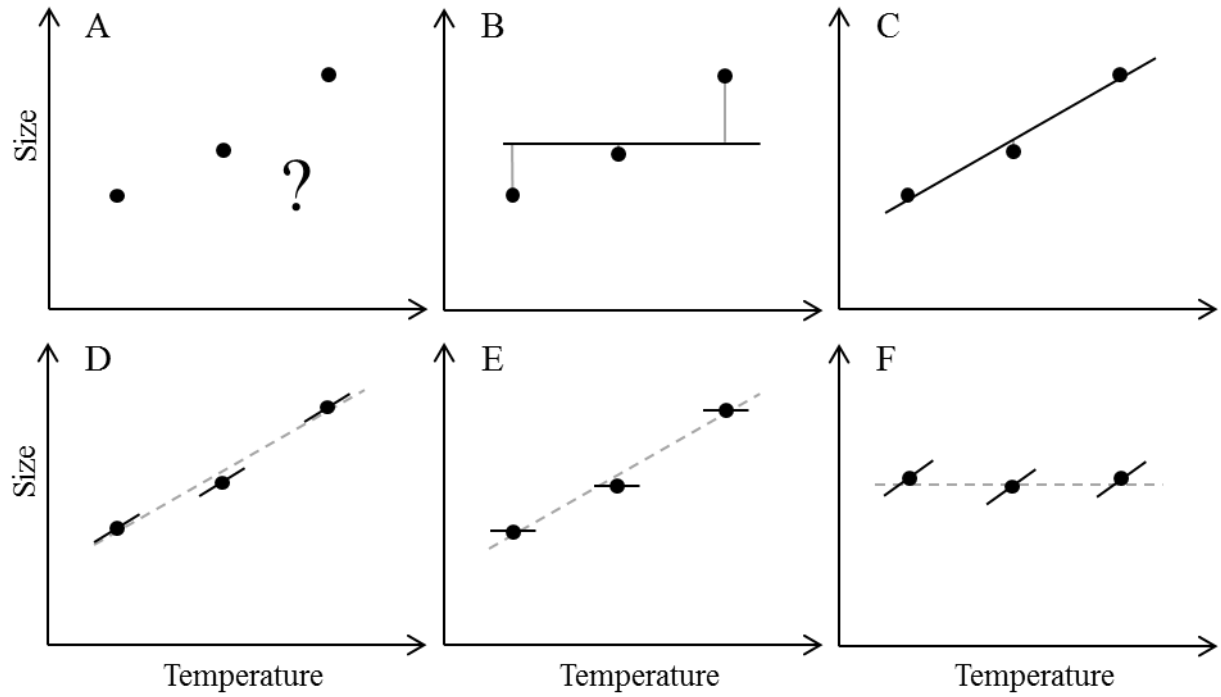

Figure 1. Graphs illustrate the interpretation of variability in a quantitative trait (size) across contingents and environments (temperature regimes). In A, a positive relationship between temperature and size is observed. Without additional information, the interpretation of this situation is ambiguous. One interpretation (B) is that the temperature-size relationship is spurious, and that the contingent-specific deviations (grey vertical bars) indicate genetic differences, or unaccounted environmental influences. Alternatively (C), it possible that the relationship is real, in which case no significant contingent-specific deviations remain. It is possible to disentangle interpretations B and C if the relationship between size and temperature is known from earlier studies, or if the relationship can be observed within contingents. In D, temperature-size reaction norm is observed within each contingent; they show a pattern that closely resembles the global temperature-size relationship. This strengthens the conclusion that contingents are genetically similar. However, in E, temperature-size reaction norms for contingents do not match the global pattern. This suggests that the global pattern is determined by genetic differences between the contingents, or other unaccounted factors. In F, temperature-size reaction norms reveal counter-gradient variation that is hiding the dependence of size on temperature.