

# Epigenetic processes and genetic architecture in character origination and evolution

Alexander V. Badyaev and J. Bruce Walsh

## 11.1 Origination of the relationship between genetic, developmental, and functional dimensions of a phenotype

How do genes get associated with a particular developmental or functional roles and contexts in evolution? How and why do they lose it? Why is there a general lack of determinism in the genotype–phenotype map and how does this map evolve? Few questions are more important to evolutionary theory, and few have been more elusive. The field of epigenetics studies properties of emergent, self-regulatory, and compensatory interactions that arise above the level of the gene, but are not directly predictable from the intrinsic properties of either phenotype or genotype (e.g. Hallgrímsson & Hall 2011). Here we suggest that explicit consideration of these mechanisms in the evolution of integration between genomic and phenotypic dimensions of organisms provides an important insight into these problems.

The evolution of genetic architecture reflects historical correspondence among genomic, genetic and phenotypic dimensions of an organism (Turelli 1988; Falconer & MacKay 1996; Hansen 2006; Arnold *et al.* 2008). Within each of these dimensions, evolutionary dynamics are dictated by patterns of connectivity among its elements (e.g. among genes,

alleles, proteins, or traits) and the rate and time at which a population can sample various combinations of such elements (Fisher 1930; Lynch & Walsh 1998). Once a trait combination with significant fitness consequences is found, natural selection can either stabilise or delete it, and genetic architecture (summarized by the *G*-matrix) then evolves to approach the shape of the fitness landscape (Cheverud 1996; Wagner & Altenberg 1996; Lynch & Walsh 1998)—that is to mimic the most consistently beneficial functional and developmental relationships of the organism (Cheverud 1988). Genetic architecture is further modified by random drift and by non-additive effects introduced by epistatic interactions of alleles across changing genetic backgrounds (Wright 1931; Rice 2001; Hansen & Houle 2008).

Because of its relationship to the fitness landscape, the *G*-matrix, by definition, is considered to be agnostic to specific proximate processes that generate phenotypes and integration of its components; instead it is shaped by the effects of individual alleles/genotypes on the traits of interest. Part of this view reflects an approach where character identity is either given outright or is defined by independent contributions to fitness (e.g. Wagner 2001). For instance, the theory of morphological integration (Olson & Miller 1958; Berg 1960; Cheverud 1982; Riska 1989) posits that

stabilising selection leads to the evolution of strong genetic correlations among functionally compatible traits, ultimately resulting in the modularization of the phenotype into relatively independent, functionally interacting groups of characters (see also Badyaev & Foresman 2004)—in other words, the formation of developmental *maps* that partly canalises the *input* values (that may range from timing and levels of transcripts to intermediate developmental stages of emerging traits). It is assumed that developmental processes can produce and transmit the required genotype–phenotype modularity for such outcomes; but direct evidence is lacking and some empirical and conceptual results suggest that evolution of such developmental modularity is unlikely from both population–genetic and developmental perspectives (Bulmer 1971; Slatkin & Frank 1990; Rice 2001; Beldade *et al.* 2002; Welch & Waxman 2003; Wagner *et al.* 2007; Conner 2012; Salazar-Ciudad & Marin-Riera 2013).

A central element of development is the divergent goals of canalisation (buffering genetic and environmental perturbations) on one hand and phenotypic plasticity on the other. Both of these processes can have genetic components, but the extent to which these processes are captured by a *G*-matrix is unclear (see Section 11.5 for an empirical illustration). In general, the *G*-matrix can be strongly shaped by modification of the underlying developmental processes (i.e. *maps*) on which segregating variation is translated into traits. A one-dimensional example of this is the mapping of some underlying score (a liability) into a trait. In a simple threshold model (where a trait only appears when the liability score of an individual exceeds a critical value), either evolution to increase the mean liability score, or evolution to decrease the threshold value (or potentially both), results in an increase in the population frequency of the trait (see also Wright 1934; Lande 1977). On such a one-dimensional scale, the difference between changes in the developmental map by moving the threshold vs changes in the liability given a set threshold is rather trivial. In a more complex morphospace, however, this distinction is of fundamental importance, especially if one envisions a set of loosely interconnected modular developmental maps acting on different suites of

traits (see Section 11.5). We can think of this distinction as variation/evolution in the inputs (e.g. the state of early developmental structures or molecular features that interact with the entire developmental sequence to produce the final phenotype) that developmental maps transform into trait values vs variation/evolution in the shape/form of the maps and the connectivity between the various maps that together form the suite of traits under consideration. Small underlying genetic changes in one or more of these maps could result in significant morphological changes that are then further canalised from genetic variance in the input variables (liability in the simple case). This allows genetic variation to be shielded from selection whilst at the same time generating covariance structures that appear to be held under stabilising selection.

Proximally, the consistency and coordination of genomic and functional elements—which are captured by the *G*-matrix—are at least partially produced by epigenetic processes of development (that is, emergent interactions that arise above (i.e. *epi*-) the level of the gene (in the sense of DNA sequence) and those properties are not predictable from intrinsic properties of either phenotype or genotype). Therefore, the patterns and prevalence of such processes within a population have significant consequences for the distribution of variances among characters that form integrated complexes (Rice 2002, 2004ab; Hansen 2011). Explicit consideration of the correspondence between the genomic and phenotypic dimensions of an organism (its developmental dynamics) can resolve apparent discrepancies between predicted and observed evolutionary patterns. For example, when microevolutionary response in the direction of abundant genetic variation is inhibited by developmental interactions (Maynard Smith *et al.* 1985) or when significant response to selection is produced in the direction orthogonal to absolute genetic correlation among traits, or, more generally, when strong selection coexists with abundant genetic variation, and both coexist with a preponderance of evolutionary stasis (Arnold 1992; Hansen 2006, Walsh and Blows 2009). Overall, however, these discrepancies show that whatever the *G*-matrix indicates, it does not fully cover the mechanisms that actually produce

an evolutionary response. Ignorance of the mechanisms by which the phenotype is produced translates, by necessity, into ignorance of the mechanisms by which it changes.

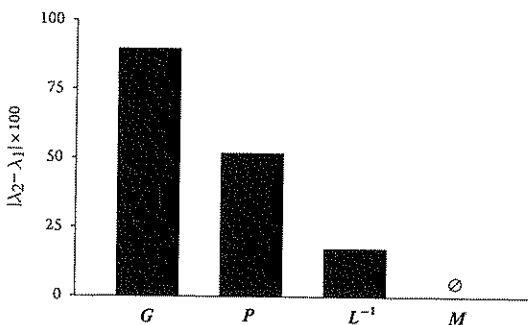
## 11.2 A developmental view of the $G$ -matrix

Whilst most individual traits harbour significant additive genetic variation, most of the additive genetic variance in multivariate space is often concentrated on just a few axes of variation (the first few eigenvalues of  $G$  account for the vast majority of its total variation, e.g. Figure 11.1; Björklund 1996; Schluter 1996; Arnold *et al.* 2008; Kirkpatrick 2009; Walsh & Blows 2009). Thus, the presence of additive genetic variation (i.e. non-zero heritability) for each element in a suite of traits under selection is no guarantee that the population will evolve, as the direction favoured by directional selection may be largely orthogonal to these major axes of variation (reviewed by Teplitsky *et al.* in Chapter 12). The population-genetic explanation for the observation of low effective dimensionality for the  $G$ -matrices is that persistent selection has eroded genetic variation in that direction (Falconer & MacKay 1996). This leaves significant variation in

other directions, and the projection of this variation onto single traits yields significant amounts of additive genetic variance. This argument is independent of any underlying developmental processes.

An alternative explanation is developmental, as in our model of variation in input variables and variation in the structure of developmental maps transforming these input variables into an integrated multivariate phenotype (see Section 11.1). As is the case for a threshold trait, there is significant variation in a population in the underlying liability score, much of which may be additive genetic. When mapped into the observed presence/absence trait, the resulting variation is much lower. In multivariate morphospace, one can imagine a set of input variables showing considerable variation that is transformed by development maps into a more limited space. The average tendency for these maps over a large number of offspring is a measure of their genetic value, whilst specific individuals may show deviations from this average trend due to environmental and developmental noise. Hence, the distribution of phenotypes can be rather different from the underlying set of distributions of average developmental trajectories (the genetic values of the maps averaged over input variables). Therefore, the structure of the  $G$ -matrix may be more reflective of these development maps, and hence show canalised multivariate directions. Conversely, if an underlying single trait is more strongly influenced by input variables than is the entire integrated multivariate phenotype, then the trait would show more variation; but this is constrained by the map into a more limited range. Importantly, as with the input variables, these developmental maps can also respond to selection, potentially selecting for more favourable maps (i.e. more variation in constraints along the directions favoured by selection).

Obviously, the current structure of the  $G$ -matrix could be a result of both of these features. However, if some  $G$ -matrices are indeed more strongly shaped by changes in developmental maps, then the potential exists for rapid transformation of one covariance structure into another, whilst still keeping most of the variation in the underlying traits.



**Figure 11.1** Relative difference between the first ( $\lambda_1$ ) and the second ( $\lambda_2$ ) eigenvalues of additive genetic covariance matrix ( $G$ ), phenotypic covariance matrix ( $P$ ), matrix of ontogenetic variation ( $L^{-1}$ ), and matrix describing adaptive landscape ( $M$ ) in Montana house finches over 19 generations after population establishment. Smaller values indicate greater dimensionality of variability. The third eigenvalues were not significant in any of the matrices. From Badyaev 2010.

### 11.3 Does interchangeability of genotype–phenotype relationships matter for stability of the G-matrix, and what role does development play in this process?

Discoveries from genomics bring additional considerations to our understanding of the relationship between genotypes and phenotypes and to the overall realisation that the evolutionary forces that shape the evolution of genomes and their architecture are likely to be distinct from those that shape phenotypic diversity (Müller & Newman 2003; Badyaev 2011b; Koonin 2011). Five insights are particularly relevant here. The first is the tremendous evolutionary conservation of most genes (i.e. their orthologous lineages) that persist unchanged over billions of years of exceptional phenotypic diversifications (Tatusov *et al.* 2003; Shubin *et al.* 2009; Wolf *et al.* 2009). The second is the relative fluidity of genomes, with gene orders and genome architecture often showing little conservation even among closely related species (reviewed in Koonin 2011). The third is the interchangeability of the relationship between genes and their functions at all levels of organismal organisation, from non-orthologous gene replacements, where unrelated sets of genes fulfil identical sets of cell functions (Wolf *et al.* 2006) to the ‘many to many’ mapping of genes on developmental and functional aspects of phenotypes (Wilkins 2001; Wainwright 2007). The fourth, and related to it, is a new appreciation of the high dimensionality of genotypes in relation to phenotypic diversity and therefore of exceptional redundancy of the genotype-to-fitness relationship (van Nimwegen & Crutchfield 2000; Gavrillets 2004; Wagner 2011). The fifth is the empirical finding that, despite such dimensionality and redundancy, only a small portion of theoretically possible pathways are assessable to evolution, a constraint that emphasises the overwhelming importance of epistasis in maintaining the evolutionary cohesiveness of evolving genomes despite their high dimensionality and ubiquitous redundancy of relationships with phenotypes (Weinreich *et al.* 2005; Gravner *et al.* 2007; Poelwijk *et al.* 2007; Breen *et al.* 2012). This echoes a persistent finding of quantitative genetics that genetic variance tends to be concentrated along

only a few axes of morphospace (Kirkpatrick 2009; Walsh & Blows 2009), such that most empirically derived G-matrices often have far fewer dimensions than their phenotypic equivalents (although the issue might be due to limited statistical power in some cases), whilst patterns of pleiotropy tend to be highly restrictive, reflecting variational modularity of phenotypes (Hansen 2011; Wagner & Zhang 2011). Such channelling of variation to a few dimensions has significant implications for evolution because it makes it more likely that random mutational input will affect beneficial combination of traits. The effect is further amplified if these combinations of traits can actually be produced by developmental processes (Alberch 1991; West-Eberhard 2003).

These insights bring a new urgency to understanding the rules by which ubiquitous interchangeability of genotype–phenotype relationships is translated into the evolutionary stability of the G-matrix and the role developmental processes play in this process. Can non-linear and emergent developmental interactions be realistically taken into account when phenotypic evolution is modelled? A recent shift in focus from concerns about availability of genetic variance (that is rarely limited by mutational input) to the patterns of its distribution among trait combinations (Larsen 2005; Blows 2007; Blows & Walsh 2009) needs to be augmented by the realisation that evolution of complex structures depends on coordinated variability, which often arises through epigenetic developmental processes (e.g. Section 11.2; see also Riedl 1978; Hallgrímsson & Hall 2011).

### 11.4 How do emergent processes influence genetic dimensionality of the phenotype?

What is the relationship between basic elements of morphogenesis and its genetic architecture? At what point in organic evolution do the genes (‘building blocks of basic processes’) get associated with the consistent developmental programs that the G-matrix describes?

One suggestion is that the origination and evolution of structures are based on different

mechanisms: emergent processes of morphogenesis form a template (developmental maps) for the subsequent accumulation of genetic networks that generate variation in the developed phenotype (Newman & Müller 2000; Newman 2005; Newman & Bhat 2008). Under this framework, the initial prevalence of plastic and environmentally contingent developmental processes is gradually replaced, over evolutionary time, by genetic networks that assure reliability of developmental outcomes in particular environmental conditions (Reid 2007; Newman 2011). In terms of our model, this amounts to genetic assimilation of a new developmental map (Waddington 1961). Thus, more recurrent organism–environment associations are expected to accumulate greater redundancy and determinism in their genetic architecture—these associations are then most likely to form additive genotype–phenotype associations (Nowak *et al.* 1997; Badyaev 2007; 2011b). One striking observation on this point comes from isogenic inbred lines in mice and *Drosophila* (Lemos *et al.* 2005; Flint & Mackay 2009). When these lines are used to introgress specific combinations of alleles from different loci into an otherwise identical genetic background, they tend to show strong epistasis. However, when these alleles are scored in outbred populations, they are largely additive. The strong epistatic effects that appear when scored over a single genetic background are randomised into additive effects when scored over a diverse background of random genotypes. Overall, according to this scenario, correspondence of one genotype to one phenotype is a highly derived condition in which an over-determining genetic circuitry ensures that changes in external and internal environments have less impact on phenotypic outcomes (Salazar-Ciudad, Newman & Sole 2001; Newman 2005; Badyaev 2011b). In less recurrent associations, there is a greater role of environmental plasticity and contingent developmental interactions (West-Eberhard 2003).

Such a scenario can play out not just over evolutionary time, but also at different levels of organismal organisation where reciprocal interactions between traits and tissue types provide each other ‘environments’ during development; that is form linkages and associations that ensure the

development of a structure, but which themselves do not experience selection acting on the final structures (and thus do not deplete variance among their elements; Kirschner & Gerhart 2005; Gerhart & Kirschner 2007). Such hierarchically arranged variability harbours genetic variance and can contribute to variational pleiotropy of complex structures (Hansen 2011; Wagner & Zhang 2011; Salazar-Ciudad & Marin-Riera 2013).

Further, selection efficacy negatively correlates with the complexity of organisms, including the complexity of genetic architecture (Lynch 2010), although this model assumes weak or absent indirect selection on the elements of genetic architecture. If the model is correct, then in complex organisms occurring in small populations, the selection ability to streamline and stabilise the most fit configurations of a structure is increasingly determined by homeostatic, entropy-reducing mechanisms (Badyaev 2013) because such elements of selection are most consistent across environmental contexts (Schmalhausen 1938). Accumulations of complexity and associated channelling effects of development on genetic and phenotypic variance further emphasise geometric considerations in studies of genetic variance; developmental modularity becomes an increasingly powerful force in shaping possible functional and, ultimately, additive genetic associations among traits.

### 11.5 Reconciling precise adaptation and evolutionary change: lessons from the long-term study of avian beak evolution

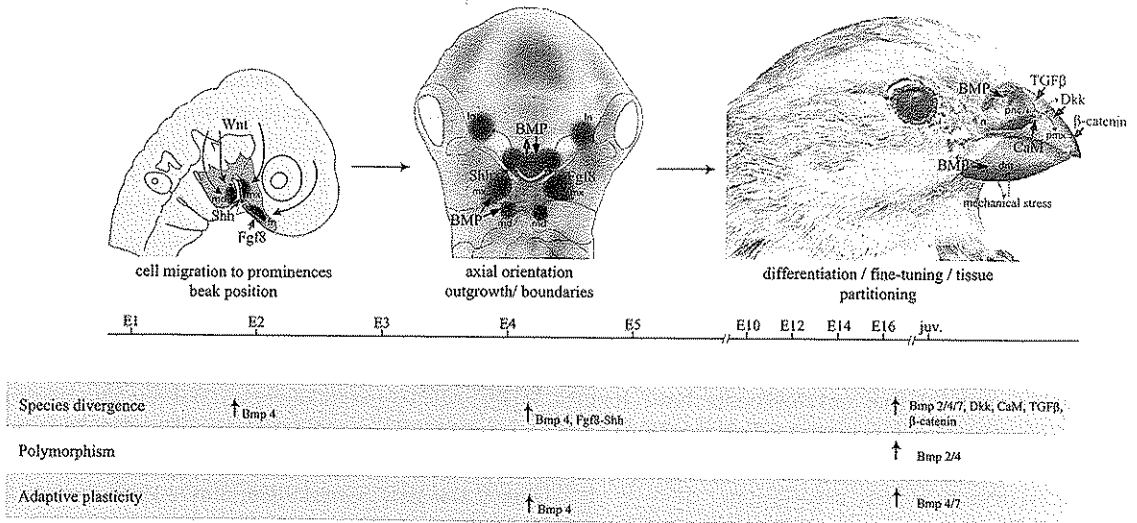
Avian beaks are some of the best examples of precise adaptation and extreme evolutionary diversification (Lack 1947; Grant 1986; Smith 1990; Benkman 1993). These traits typically experience strong selection on precise functional integration needed for food handling and bite force, and such functional integration is accomplished by coordinated changes of many developmental components that are often under distinct genetic control (Eames & Helm 2004; Grant *et al.* 2006; Mallarino *et al.* 2011, 2012). Thus, it is a particularly suitable structure in which to study the evolutionary changes in integration of genetic,

developmental, and functional dimensions of the phenotype. A particular paradox is how to reconcile adaptation and diversification—evolution of local adaptations requires close genetic integration of beak components and high heritability of their development for incremental fine-tuning of beak morphology. However, such consistent reduction of developmental variability should, at the same time, prevent the evolutionary diversifications in beaks routinely observed in birds.

The proximate resolution of this paradox could come from significant redundancy of the developmental pathways that produce beaks and the ubiquitous reuse of conserved regulatory elements in developmental and functional integration of beak components (Badyaev 2011a)—in other words, changes in the structure of the development *map* rather than in the *input* variables. The evolutionary significance of such modular organisation and conserved signalling (Figure 11.2) is that only a few genetic changes in regulatory elements are

needed for rapid evolution of local adaptation and extensive evolutionary diversification without depletion of genetic variance in beak morphology (see Section 11.1). Under this scenario, the role of natural selection is limited to eliminating developmental abnormalities and to stabilising developmental configurations most adaptive under prevalent conditions. Genetic fixation of mutations in regulatory elements can enable the evolutionary persistence of the most favoured configurations, but redundancy of the regulatory network, compensatory interactions among its elements, and the overall highly modular organisation assures short-term evolutionary retention of many functional configurations of beaks (Badyaev 2011a).

What consequences does such developmental organisation have on the evolution of the *G*-matrix? To answer this question empirically, we undertook a long-term study of multivariate coevolution of genetic, developmental, and functional integration in beaks of house finches (*Carpodacus mexicanus*),



**Figure 11.2** Modular core processes are regulated by conserved growth factors in beak development. Main growth factors are wingless type (*Wnt*), fibroblast growth factor 8 (*Fgf8*), sonic hedgehog (*Shh*), bone morphogenetic proteins (BMP), transforming growth factor beta (*TGFβ*), calmodulin (*CaM*), Dickkopf (*Dkk*), and  $\beta$ -catenin. Facial prominences (shown in dark), formed by proliferation of neural crest cells, are frontonasal (fn), lateral nasal (ln), mandibular (md), and maxillary (mx). Cartilage and bone areas arising in late development are prenasal cartilage (pnc), premaxillary bone (pmx), nasal bone (n), and dentary bone (dnt). Double-headed arrows show interactions between neighbouring prominences during growth and expansion (left and middle figure), tissue partitioning between pnc and pmx and effects of mechanical stress conductance in late developmental stages (right figure). Table shows developmental stage and growth factors that were shown to regulate species divergence, polymorphism, and adaptive plasticity in beak size and shape. Based on references in Badyaev 2011a and Mallarino *et al.* 2012.

tracing the evolution of novel beak configurations across 20 consecutive generations during the origin of local adaptation (Badyaev 2010). We were particularly interested in how abundant genetic variance in beak ontogeny is modulated to produce both precise local adaptations (that require significant reduction of variance; Badyaev *et al.* 2000; Badyaev & Martin 2000) and continuing diversification of beak morphology among different populations as this species expands its ecological and geographic range (Badyaev, Belloni & Hill 2012).

We derived an overall genetic variance-covariance matrix and genetic correlations among three beak components (width, length, and depth) from a fully resolved pedigree consisting of full- and half-sib groups across 11 generations (Badyaev 2005, 2010). Additive genetic ( $G$ ) and common environmental ( $E$ ; e.g. nest effects) variance-covariance matrices were derived by fitting phenotypic data to a multivariate animal model of the general form:  $y = Xb + Za + Ec + e$ , where  $y$  is a vector of trait values,  $b$  is a vector of fixed effects,  $a$  is a vector of additive genetic effects,  $c$  is a vector of common environmental effects,  $e$  is a vector of residual variation, and  $X$ ,  $Z$ , and  $E$  are incidence matrices for the fixed, additive genetic, and common environmental effects, respectively (Lynch & Walsh 1998). Analysis was carried out using restricted maximum likelihood implemented in ASReml (2.0) software. We used univariate general linear models (PROC GLM, SAS Inc.) to identify significant fixed effects, and any fixed-effect term that was significant in at least one trait was included in the final multivariate model. This resulted in the inclusion of year, offspring sex and age. Nest identity (nested within dam identity) was included as a random effect representing variance due to common environment (e.g. nest environment, parental effects).

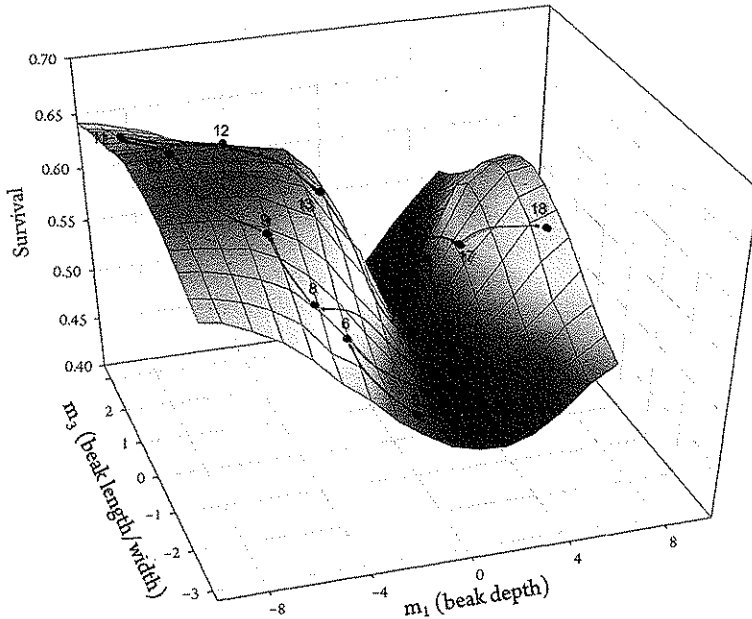
In each generation, we measured multivariate selection on beak morphology by fitting the full second-order polynomial equation (Lande & Arnold 1983)  $w = \alpha + z^T\beta + z^T\gamma z$ , where  $z$  are the three original traits,  $w$  is juvenile survival associated with the onset of independent foraging (from 40 to 80 days post-fledging),  $\beta$  is the vector of standardised directional selection gradients, and  $\gamma$  is the

matrix of quadratic and cross-product terms among the traits. We also performed canonical rotation of the  $\gamma$  matrix (Box & Draper 1987; Blows & Brooks 2003) to create the  $M$ -matrix, in which the eigenvectors  $m_i$  describe the shape of the response surface and the direction of its principal orientation. The largest eigenvalues,  $\lambda_i$ , are associated with the greatest curvatures in the response surface; positive eigenvalues indicate upward curvature, and negative eigenvalues indicate downward curvature (Phillips & Arnold 1989).

For each generation, we also measured developmental variability by constructing age-specific correlation matrices for the growth sequence of all beak components from age 1–16 days and calculated the overall correlational matrix  $L$  and associated eigenvalues and eigenvectors for the entire growth sequence (e.g. Badyaev & Martin 2000). To assess multivariate directions of greatest independent developmental variation (i.e. lowest ontogenetic integration) of the three beak components, we calculated  $L^{-1}$  for each generation, whose vectors are projections of traits with greatest independent variation during growth.

Although we measured 'end products'—beak dimensions during different times of ontogeny—our measure of the  $G$ -matrix and multivariate selection nevertheless accurately described patterns of microevolutionary change (Figure 11.3). The direction of long-term stabilising selection on beak depth was indistinguishable from the direction of maximum variance of the  $G$ -matrix whereas fluctuating selection on relative expression of beak length–beak width dimensions was strongly concordant with the second eigenvalue of the  $G$ -matrix. The role of directional selection was largely confined to elimination of phenotypic extremes formed by compensatory developmental interactions (Figure 11.4) and these diverse developmental interactions during initial stages of population establishment did not have a significant imprint in either genetic variance-covariance structure or patterns of long-term stabilising selection (Figure 11.4).

We also found high genetic correlations between beak components that experienced variable and frequently antagonistic developmental interactions,



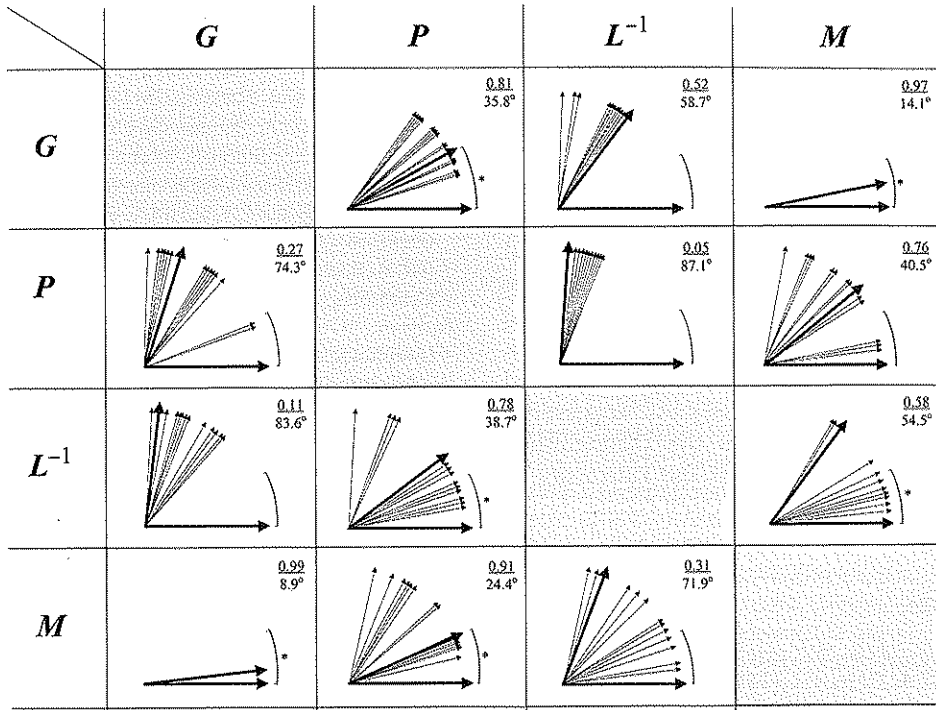
**Figure 11.3** Overall fitness surface (juvenile post-fledging survival) defined by variation in beak depth ( $m_1$ ) and relative expression of beak length and width ( $m_3$ ). The  $G$ -matrix orients congruently with the  $M$ -matrix (85% along  $m_1$  and 8.7% along  $m_3$ ). Dots are average coordinates of  $m_1$ ,  $m_3$ , and survival for each generation (shown by consecutive numbers), starting with generation 5. Arrows connect subsequent generations. Modified from Badyaev (2010).

had low additive genetic variance, and were likely regulated by non-overlapping gene cascades (Abzhanov *et al.* 2006; Badyaev 2010). This further emphasises that genetic and developmental integration are rarely congruent, even for frequently co-selected traits (Badyaev 2004; Frankino *et al.* 2007). At the same time, this finding confirmed the expectation that when variation in partitioning of developmental precursors among traits is consistently greater than variation in fitness consequences of their end products—as is expected in adaptively equivalent combinations of traits—the developmentally interacting traits will evolve genetic correlations even when their relative expression is antagonistic (Bulmer 1971; Houle 1991).

Compensatory adjustments of beak components during development and the adaptive equivalence of distinct configurations can have important consequences for the structure of the  $G$ -matrix that can reconcile adaptation and evolutionary change. First, such developmental interactions extended the phenotypic range of locally adaptive morphologies converting overall disruptive selection during colonisation of a novel environment to overall stabilising selection where several adaptive phenotypes could be maintained—a condition that

favours long-term stability in the  $G$ -matrix (Lande 1976; Kopp & Hermisson 2006). Second, compensatory adjustments among beak components shielded genetic variance in individual traits, such that a small number of conserved developmental modules produced both locally adaptive morphology and evolutionary diversifications. Third, compensatory development and functional equivalence (when distinct configurations of traits produce the same functional outcome) might explain the puzzling result whereby dimensions of the strongest and most persistent stabilising selection coincided with dimensions of greatest additive genetic variance, whilst traits under fluctuating selection had lower additive genetic variance. Thus, diverse developmental adjustments among beak components can either replenish genetic variance along the axis of most consistent selection or shield variance in the trait that is most consistently subject to selection (Walsh & Blows 2009). The finding that only the most recurrent developmental and functional interactions were represented in  $G$ -matrix structure provides an important insight into the hierarchical arrangement of epigenetic processes in development in relation to their genetic stabilisation.





**Figure 11.4** Concordance of  $G$  (additive genetic covariance matrix),  $P$  (phenotypic covariance matrix),  $L^{-1}$  (negative inverse covariance matrix of ontogenetic integration), and  $M$  (matrix of multivariate selection) of beak morphology in Montana house finches during 19 generations following population establishment. Shown are the first (above diagonal) and the second (below diagonal) eigenvectors and the corresponding vector correlations and angles (in degrees) between them. Thick lines show overall vectors calculated across all generations, thin lines show within-generation vectors. For  $L^{-1}$  vs  $G$  and  $P$  vs  $G$  comparisons, within-generation vectors of  $L^{-1}$  and  $P$  are compared with the vectors of overall  $G$ . For all other comparisons ( $L^{-1}$  vs  $P$ ,  $L^{-1}$  vs  $M$ , and  $P$  vs  $M$ ) vectors are compared within each generation. Brackets show bootstrapped limits ( $n = 100$ ) within which the vectors are not distinct. Asterisks indicate similarity of overall vector correlations (if thick vectors are within the bracket) or similarity of within-generation vectors (if most of the within-generation vectors are within the bracket). From Badyaev (2010).

## 11.6 Conclusions

One of the central questions in evolutionary quantitative genetics is, what forces shape the current  $G$ -matrix for a given population? Are these largely population-genetic, namely the removal of additive variation along persistent directions of natural selection or, as we suggest here, could emergent processes of development also play a major role? Either of these forces can generate a  $G$ -matrix that is highly constrained (the first few eigenvalues accounting for the vast majority of variation). In such cases, evolution along the lines of genetic least resistance (in directions of maximal variation in the  $G$ -matrix) is inevitable. If trait evolution is largely governed by drift (potentially in the face

of weak stabilising selection), then the maximal divergence occurs along axes with maximal variation. Conversely, under directional selection, with a very constrained  $G$ -matrix the projection of any random selection gradient onto the first eigenvector likely dominates the projection of that vector onto the whole of the  $G$ -matrix, and hence the response closely aligns to the first eigenvalue, i.e. along the line of least resistance. In our empirical example, it seems that the lines of least genetic resistance are, proximately speaking, the lines of greatest recurrence of organism-environment associations that reflect the evolved correspondence between epigenetic processes of developmental and functional integrations stabilised by increasingly redundant additive genetic linkages. This and other empirical

studies show that the genetic dimensionality of a structure is often far smaller than is expected from dimensionality of its phenotype.

## Acknowledgments

Empirical work reported here was supported by the National Science Foundation (DEB-0075388, DEB-0077804, IBN-0218313) and the David and Lucille Packard Foundation Fellowship. We are grateful to Anne Charmantier, Loeske Kruuk, and Dany Garant for invitation to contribute. We thank the editors, Renee Duckworth, Badyaev Lab, and anonymous reviewers for insightful suggestions and comments. AVB thanks the Distinguished Visiting Professor program of the University of Miami for their hospitality in Coral Gables during writing of this chapter.

## References

- Abzhanov, A., Kuo, W.P., Hartmann, C., Grant, B.R., Grant, P.R. & Tabin, C.J. (2006) The calmodulin pathway and evolution of elongated beak morphology in Darwin's finches. *Nature*, **442**, 563–567.
- Alberch, P. (1991) From genes to phenotype: dynamical systems and evolvability. *Genetica*, **84**, 5–11.
- Arnold, S.J. (1992) Constraints on phenotypic evolution. *American Naturalist*, **140**, S85–S107.
- Arnold, S.J., Bürger, R., Hohenlohe, P.A., Ajie, B.C. & Jones, A.G. (2008) Understanding the evolution and stability of the G-matrix. *Evolution*, **62**, 2451–2461.
- Badyaev, A.V. (2004) Integration and modularity in the evolution of sexual ornaments: an overlooked perspective. In: *Phenotypic integration: the evolutionary biology of complex phenotypes*. (ed. M. Pigliucci & K. Preston), pp. 50–79. Oxford University Press, Oxford.
- Badyaev, A.V. (2005) Maternal inheritance and rapid evolution of sexual size dimorphism: passive effects or active strategies? *American Naturalist*, **166**, S17–S30.
- Badyaev, A.V. (2007) Evolvability and robustness in color displays: bridging the gap between theory and data. *Evolutionary Biology*, **34**, 61–71.
- Badyaev, A.V. (2010) The beak of the other finch: coevolution of genetic covariance structure and developmental modularity during adaptive evolution. *Philosophical Transactions of The Royal Society of London Series B, Biological Sciences*, **365**, 1111–1126.
- Badyaev, A.V. (2011a) How do precise adaptive features arise in development? Examples with evolution of context-specific sex-ratios and perfect beaks. *Auk*, **128**, 467–474.
- Badyaev, A.V. (2011b) Origin of the fittest: link between emergent variation and evolutionary change as a critical question in evolutionary biology. *Proceedings of the Royal Society B-Biological Sciences*, **278**, 1921–1929.
- Badyaev, A.V. (2013) "Homeostatic hitchhiking": a mechanism for the evolutionary retention of complex adaptations. *Integrative and Comparative Biology*, **53**, 913–922.
- Badyaev, A.V., Belloni, V. & Hill, G.E. (2012) House Finch (*Haemorhous mexicanus*). *The birds of North America online* (ed. A. Poole), pp. doi:10.2173/bna.2146. Cornell Lab of Ornithology, Ithaca.
- Badyaev, A.V. & Foresman, K.R. (2004) Evolution of morphological integration: I. Functional units channel stress-induced variation in shrew mandibles. *American Naturalist*, **163**, 868–879.
- Badyaev, A.V., Hill, G.E., Stoehr, A.M., Nolan, P.M. & McGraw, K.J. (2000) The evolution of sexual dimorphism in the house finch: II. Population divergence in relation to local selection. *Evolution*, **54**, 2134–2144.
- Badyaev, A.V. & Martin, T.E. (2000) Individual variation in growth trajectories: phenotypic and genetic correlations in ontogeny of the house finch (*Carpodacus mexicanus*). *Journal of Evolutionary Biology*, **13**, 290–301.
- Beldade, P., Koops, K. & Brakefield, P.M. (2002) Developmental constraints versus flexibility in morphological evolution. *Nature*, **416**, 844–846.
- Benkman, C.W. (1993) Adaptation to single resources and the evolution of crossbill (*Loxia*) diversity. *Ecological Monographs*, **63**, 305–325.
- Berg, R.L. (1960) The ecological significance of correlation pleiades. *Evolution*, **14**, 171–180.
- Björklund, M. (1996) The importance of evolutionary constraints in ecological time scales. *Evolutionary Ecology*, **10**, 423–431.
- Blows, M.W. (2007) A tale of two matrices: multivariate approaches in evolutionary biology. *Journal of Evolutionary Biology*, **20**, 1–8.
- Blows, M.W. & Brooks, R. (2003) Measuring nonlinear selection. *American Naturalist*, **162**, 815–820.
- Blows, M.W. & Walsh, B. (2009) Spherical cows grazing in Flatland: constraints to selection and adaptation. In: *Adaptation and fitness in animal populations: evolutionary and breeding perspectives on genetic resource management* (ed. J. van der Werf, H.-U. Graser, R. Frankham & C. Gondro), pp. 83–101. Springer Netherlands, Dordrecht.
- Box, G.E.P. & Draper, N.R. (1987) *Empirical model-building and response surfaces*. Wiley Series in Probability and Mathematical Statistics, New York.
- Breen, M.S., Kemena, C., Vlasov, P.K., Notredame, C. & Kondrashov, F.A. (2012) Epistasis as the primary factor in molecular evolution. *Nature*, **490**, 535–538.

- Bulmer, M.G. (1971) The effect of selection on genetic variability. *American Naturalist*, **105**, 201–211.
- Cheverud, J.M. (1982) Relationships among ontogenetic, static, and evolutionary allometry. *American Journal of Physical Anthropology*, **59**, 139–149.
- Cheverud, J.M. (1988) A comparison of genetic and phenotypic correlations. *Evolution*, **42**, 958–968.
- Cheverud, J.M. (1996) Developmental integration and the evolution of pleiotropy. *American Zoologist*, **36**, 44–50.
- Conner, J.K. (2012) Quantitative genetic approaches to evolutionary constraint: how useful? *Evolution*, **66**, 3313–3320.
- Eames, B.F. & Helm, J.W. (2004) Conserved molecular program regulating cranial and appendicular skeletogenesis. *Developmental Dynamics*, **231**, 4–13.
- Falconer, D.S. & MacKay, T.D. (1996) *Quantitative Genetics*. Longman, Essex, UK.
- Fisher, R.A. (1930) *The genetical theory of natural selection*. Clarendon Press, Oxford.
- Flint, J. & Mackay, T.F.C. (2009) Genetic architecture of quantitative traits in mice, flies, and humans. *Genome Research*, **19**, 723–733.
- Frankino, W.A., Zwaan, B.J., Stern, D.L. & Brakefield, P.M. (2007) Internal and external constraints in the evolution of morphological allometries in a butterfly. *Evolution*, **61**, 2958–2970.
- Gavrilets, S. (2004) *Fitness landscapes and the origin of species*. Princeton University Press, Princeton, NJ.
- Gerhart, J. & Kirschner, M. (2007) The theory of facilitated variation. *Proceedings of the National Academy of Sciences of the United States of America*, **104**, 8582–8589.
- Grant, P.R. (1986) *Ecology and evolution of Darwin's finches*. Princeton University Press, Princeton.
- Grant, P.R., Grant, B.R. & Abzhanov, A. (2006) A developing paradigm for the development of bird beaks. *Biological Journal of Linnean Society*, **88**, 17–22.
- Gravner, J., Pitman, D. & Gavrilets, S. (2007) Percolation on fitness landscapes: Effect of correlation, phenotype, and incompatibilities. *Journal of Theoretical Biology*, **248**, 627–645.
- Hallgrímsson, B. & Hall, B.K. (2011) Epigenetics: linking genotype and phenotype in development and evolution. University of California Press, Berkeley.
- Hansen, T.F. (2006) The evolution of genetic architecture. *Annual Review of Ecology, Evolution and Systematics*, **37**, 123–157.
- Hansen, T.F. (2011) Epigenetics: adaptation or contingency? In: *Epigenetics: linking phenotype and genotype in development and evolution* (ed. B. Hallgrímsson & B.K. Hall), pp. 357–376. University of California Press, Berkeley.
- Hansen, T.F. & Houle, D. (2008) Measuring and comparing evolvability and constraint in multivariate characters. *Journal of Evolutionary Biology*, **21**, 1201–1219.
- Houle, D. (1991) Genetic covariance of fitness correlates: what genetic correlations are made of and why it matters. *Evolution*, **45**, 630–648.
- Kirkpatrick, M. (2009) Patterns of quantitative genetic variation in multiple dimensions. *Genetica*, **136**, 271–284.
- Kirschner, M. & Gerhart, J.C. (2005) *The plausibility of life: resolving Darwin's Dilemma*. Yale University Press, New Haven.
- Koonin, E.V. (2011) *The logic of chance: the nature and origin of biological evolution*. FT Press Science, Upper Saddle River.
- Kopp, M. & Hermisson, J. (2006) The evolution of genetic architecture under frequency-dependent disruptive selection. *Evolution*, **60**, 1537–1550.
- Lack, A.J. (1947) *Darwin's finches*. Cambridge University Press, Cambridge.
- Lande, R. (1976) Natural selection and random genetic drift in phenotypic evolution. *Evolution*, **30**, 314–334.
- Lande, R. (1977) Evolutionary mechanisms of limb loss in tetrapods. *Evolution*, **32**, 73–92.
- Lande, R. & Arnold, S.J. (1983) The measurement of selection on correlated characters. *Evolution*, **37**, 1210–1226.
- Larsen, E. (2005) Developmental origins of variation. In: *Variation* (ed. B. Hallgrímsson & B.K. Hall), pp. 113–129. Elsevier Academic Press, San Diego.
- Lemos, B., Meiklejohn, C.D., Cáceres, M. & Hartl, D.L. (2005) Rates of divergence in gene expression profiles of primates, mice, and flies: stabilizing selection and variability among functional categories. *Evolution*, **59**, 126–137.
- Lynch, M. (2010) Scaling expectations for the time to establishment of complex adaptations. *Proceedings of the National Academy of Sciences of the United States of America*, **107**, 16577–16582.
- Lynch, M. & Walsh, B. (1998) *Genetics and analysis of quantitative traits*. Sinauer Associates, Sunderland.
- Mallarino, R., Campàs, O., Fritz, J.A., Burns, K.J., Weeks, O.G., Brenner, M.I. & Abzhanov, A. (2012) Closely related bird species demonstrate flexibility between beak morphology and underlying developmental programs. *Proceedings of the National Academy of Sciences of the United States of America*, **109**, 16222–16227.
- Mallarino, R., Grant, P.R., Grant, B.R., Herrel, A., Kuoa, W.P. & Abzhanov, A. (2011) Two developmental modules establish 3D beak-shape variation in Darwin's finches. *Proceedings of the National Academy of Sciences of the United States of America*, **108**, 4057–4062.
- Maynard Smith, J., Burian, R., Kauffman, S., Alberch, P., Campbell, J., Goodwin, B., Lande, R., Raup, D. & Wolpert, L. (1985) Developmental constraints and evolution. *The Quarterly Review of Biology*, **60**, 266–287.
- Müller, G.B. & Newman, S. (2003) *Origination of organismal form: beyond the gene in developmental and evolutionary biology*. The MIT Press, Cambridge.

- Newman, S.A. (2005) The pre-Mendelian, pre-Darwinian world: shifting relations between genetic and epigenetic mechanisms in early multicellular evolution. *Journal of Biosciences*, **30**, 75–85.
- Newman, S.A. (2011) The evolution of evolutionary mechanisms: a new perspective. In: *Biological evolution: facts and theories* (ed. G. Auletta, M. Leclerc & R.A. Martinez), pp. 169–191. Gregorian & Biblical Press, Rome.
- Newman, S.A. & Bhat, R. (2008) Dynamical patterning modules: physico-genetic determinants of morphological development and evolution. *Physical Biology*, **5**, 1–14.
- Newman, S.A. & Müller, G.B. (2000) Epigenetic mechanisms of character origination. *Journal of Experimental Zoology*, **288**, 304–314.
- Nowak, M.A., Boerlijst, M.C., Cooke, J. & Smith, J.M. (1997) Evolution of genetic redundancy. *Nature*, **388**, 167–171.
- Olson, E.C. & Miller, R.L. (1958) *Morphological integration*. University of Chicago Press, Chicago.
- Phillips, P.C. & Arnold, S.J. (1989) Visualizing multivariate selection. *Evolution*, **43**, 1209–1222.
- Poelwijk, F.J., Kiviet, D.J., Weinreich, D.M. & Tans, S.J. (2007) Empirical fitness landscapes reveal accessible evolutionary paths. *Nature*, **445**, 383–386.
- Reid, R.G.B. (2007) *Biological emergences: evolution by natural experiment*. MIT Press, Cambridge.
- Rice, S.H. (2001) The evolution of developmental interactions; epistasis, canalization, and integration. In: *Epistasis and the evolutionary process* (ed. J.B. Wolf, I. Brodie Edmund D & M.J. Wade), pp. 82–98. Oxford University Press, New York.
- Rice, S.H. (2002) A general population genetic theory for the evolution of developmental interactions. *Proceedings of the National Academy of Sciences of the United States of America*, **99**, 15518–15523.
- Rice, S.H. (2004a) Developmental associations between traits: covariance and beyond. *Genetics*, **166**, 513–526.
- Rice, S.H. (2004b) *Evolutionary theory: mathematical and conceptual foundations*. Sinauer Associates, Sunderland.
- Riedl, R. (1978) *Order in living organisms: a systems analysis of evolution*. Wiley, New York.
- Riska, B. (1989) Composite traits, selection response, and evolution. *Evolution*, **43**, 1172–1191.
- Salazar-Ciudad, I. & Marin-Riera, M. (2013) Adaptive dynamics under development-based genotype-phenotype maps. *Nature*, **497**, 361–364.
- Salazar-Ciudad, I., Newman, S.A. & Sole, R.V. (2001) Phenotypic and dynamical transitions in model genetic networks I. Emergence of patterns and genotype-phenotype relationships. *Evolution and Development*, **3**, 84–94.
- Schluter, D. (1996) Adaptive radiation along genetic lines of least resistance. *Evolution*, **50**, 1766–1774.
- Schmalhausen, I.I. (1938) *Organism as a whole in individual development and history*. Academy of Sciences, Leningrad.
- Shubin, N., Tabin, C.J. & Carroll, S. (2009) Deep homology and the origins of evolutionary novelty. *Nature*, **457**, 818–823.
- Slatkin, M. & Frank, S.A. (1990) The quantitative genetic consequences of pleiotropy under stabilizing and directional selection. *Genetics*, **125**, 207–213.
- Smith, T.B. (1990) Natural selection on bill characters in the two bill morphs of the African finch *Pyrenestes ostrinus*. *Evolution*, **44**, 832–842.
- Tatusov, R.L., Fedorova, N.D., Jackson, J.D., Jacobs, A.R., Kiryutin, B., Koonin, E.V., Krylov, D.M., Mazumder, R., Mekhedov, S.L., Nikolskaya, A.N., Rao, B.S., Smirnov, S., Sverdlov, A.V., Vasudevan, S., Wolf, Y.I., Yin, J.J. & Natale, D.A. (2003) The COG database: an updated version includes eukaryotes. *BMC Bioinformatics*, **4**, 41.
- Turelli, M. (1988) Phenotypic evolution, constant covariances, and the maintenance of additive genetic variance. *Evolution*, **42**, 1342–1347.
- van Nimwegen, E. & Crutchfield, J.P. (2000) Metastable evolutionary dynamics: crossing fitness barriers or escaping via neutral paths. *Bulletin of Mathematical Biology*, **62**, 799–848.
- Waddington, C.H. (1961) Genetic assimilation. *Advances in Genetics*, **10**, 257–290.
- Wagner, A. (2011) *The origins of evolutionary innovations: a theory of transformative change in living systems*. Oxford University Press, Oxford.
- Wagner, G.P. (2001) *The character concept in evolutionary biology*. Academic Press, San Diego.
- Wagner, G.P. & Altenberg, L. (1996) Complex adaptation and the evolution of evolvability. *Evolution*, **50**, 967–976.
- Wagner, G.P., Pavlicev, M. & Cheverud, J.M. (2007) The road to modularity. *Nature Reviews Genetics*, **8**, 921–931.
- Wagner, G.P. & Zhang, J. (2011) The pleiotropic structure of the genotype-phenotype map: the evolvability of complex organisms. *Nature Reviews Genetics*, **12**, 204–213.
- Wainwright, P.C. (2007) Functional versus morphological diversity in macroevolution. *Annual Review of Ecology, Evolution and Systematics*, **38**, 381–401.
- Walsh, B. & Blows, M.W. (2009) Abundant genetic variation + strong selection = multivariate genetic constraints: a geometric view of adaptation. *Annual Review of Ecology, Evolution, and Systematics*, **40**, 41–59.
- Weinreich, D.M., Watson, R.A. & Chao, L. (2005) Perspective: sign epistasis and genetic constraint on evolutionary trajectories. *Evolution*, **59**, 1165–1174.
- Welch, J.J. & Waxman, D. (2003) Modularity and the cost of complexity. *Evolution*, **57**, 1723–1734.

- West-Eberhard, M.J. (2003) *Developmental plasticity and evolution*. Oxford University Press, Oxford.
- Wilkins, A.S. (2001) *The evolution of developmental pathways*. Sinauer Associates, Sunderland.
- Wolf, Y.I., Carmel, L. & Koonin, E.V. (2006) Unifying measures of gene function and evolution. *Proceedings of the Royal Society B—Biological Sciences*, **273**, 1507–1515.
- Wolf, Y.I., Novichkov, P.S., Karev, G.P., Koonin, E.V. & Lipman, D.J. (2009) The universal distribution of evolutionary rates of genes and distinct characteristics of eukaryotic genes of different apparent ages. *Proceedings of the National Academy of Sciences of the United States of America*, **106**, 7273–7280.
- Wright, S. (1931) Evolution in Mendelian populations. *Genetics*, **16**, 97–159.
- Wright, S. (1934) An analysis of variability in number of digits in an inbred strain of guinea pigs. *Genetics*, **19**, 506–536.