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GENETICS OF FLUCTUATING ASYMMETRY: A DEVELOPMENTAL MODEL OF DEVELOPMENTAL INSTABILITY

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Abstract.—Although numerous studies have found that fluctuating asymmetry (FA) can have a heritable component, the genetic and developmental basis of FA is poorly understood. We used a developmental model of a trait, according to a diffusion–threshold process, whose parameters are under genetic control. We added a small amount of random variation to the parameter values of this model to simulate developmental noise. As a result of the nonlinearity of the model, different genotypes differed in their sensitivity to developmental noise, even though the noise is completely random and independent of the genotype. The heritable component of FA can thus be understood as genetically modulated expression of variation that is itself entirely nongenetic. The loci responsible for this genetic variation of FA are the same that affect the left/right mean of the trait, showing that genetic variation for FA does not require genes that specifically control FA. Furthermore, the model offers alternative explanations for phenomena widely discussed in the literature on FA, for instance, the correlations between FA and heterozygosity and between FA and trait size. The model underscores the importance of dominance and epistasis, and therefore unites the study of FA with the classical theory of quantitative genetics.

Key words.—Development, diffusion–threshold model, dominance, epistasis, fluctuating asymmetry, heritability, quantitative genetics.

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Fluctuating asymmetry (FA) has received much recent interest as a measure of individual quality and as a possible target for natural and sexual selection (reviewed by Møller and Swaddle 1997). This approach is based on the premise that FA is the measurable expression of developmental instability. Developmental instability stands for the tendency of the phenotypic value of a trait to deviate from the value expected for an individual with a given genotype and environment. Developmental instability is measurable as FA because the two sides of a bilaterally symmetric organism share the same genome (barring somatic mutation) and environmental differences between sides, integrated over the time of development, are likely to be small (though this may not be true for sessile organisms). Therefore, differences between the body sides are mostly attributable to random "errors" that accrue during development. Despite the simplicity of this logic, the developmental basis of developmental instability is not well understood (Markow 1995), and even the degree to which FA is genetically based has been contentious (Møller and Swaddle 1997; Møller and Thornhill 1997; but see also the criticism, e.g., by Leamy 1997; Markow and Clarke 1997; Whitlock and Fowler 1997). Detailed case studies of the developmental mechanisms and genetic architecture that underlie FA have only been carried out recently, either through the study of candidate genes (Batterham et al. 1996) or through the search for quantitative trait loci (Leamy et al. 1997, 1998).

Theoretical treatments of FA have used either of two approaches: (1) statistical analysis of asymmetry measured at the phenotypic level; or (2) modeling of the dynamics of a developmental process assumed to generate asymmetry. The statistical approach is more widespread and is the basis of the method for analyzing asymmetry by ANOVA (Leamy 1984; Palmer and Strobeck 1986; Palmer 1994; Møller and Swaddle 1997). Theoretical models of this kind are usually based on the assumption that phenotypic values for left and right body sides are drawn independently from a distribution whose dispersion is an expression of developmental instability (e.g., Palmer and Strobeck 1992; Whitlock 1996; Houle 1997). This kind of linear statistical modeling usually assumes, implicitly or explicitly, that asymmetries result from small developmental imprecisions that are "random, independent, and cumulative" (Palmer and Strobeck 1992, p. 59), and therefore will approximate a normal distribution. The normal distribution has played a prominent role in studies of asymmetry, for example, as the criterion that distinguishes FA (an expression of "pure" developmental noise) from antisymmetry (where individuals are expected to be asymmetric, but handedness is random), which is usually assumed to have a genetic component (Van Valen 1962; Palmer and Strobeck 1986, 1992; Palmer et al. 1993; Palmer 1994; Rowe et al. 1997). This linear statistical approach is akin to the additive models routinely used in quantitative genetics (Falconer and Mackay 1996; Roff 1997; Lynch and Walsh 1998). Hence it is possible to use relatively simple genetic models (Gavrilets and Hastings 1994) and the powerful statistical machinery of normal theory for the study of FA (Whitlock 1996). This simplification, however, comes at the price of making the strong assumption that developmental effects are additive, which may often not be the case.

The second approach is based on explicit models of the developmental processes responsible for building the structures under study. These models almost invariably are nonlinear (e.g., Oster 1988; Murray 1993), and therefore the processes involved are not additive. Nonlinearity is near-ubiquitous in development, for example, in the multiplicative nature of growth that underlies allometry and that leads to change in proportions with increasing size (Huxley 1932).

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Nonlinearity is also the hallmark of the diffusion–threshold, reaction–diffusion, and signaling systems that are involved in patterning processes (Oster 1988; Held 1992; Murray 1993). For example, in the egg of *Drosophila*, diffusion–threshold systems turn smooth gradients of proteins like Bicoid and Nanos into discrete stripes of gene expression by the blastoderm stage (e.g., Lawrence 1992, ch. 2; Gilbert 1997, ch. 14), and in later stages, gradients of Wingless protein affect the patterning and growth of the wing discs (Neumann and Cohen 1997a). Nonlinear models of development have been applied to the study of asymmetry by Emlen et al. (1993) and Graham et al. (1993). The primary focus of these models, which were designed primarily in the context of fractals and chaotic dynamics, was the temporal dynamics of systems in which feedback interactions lead to oscillations in the concentrations of activator and inhibitor substances. It is not entirely clear, however, whether similar results also may apply to developmental processes where there is no evidence of oscillations.

In this paper, we use a different developmental model, extending previous work by Nijhout and Paulsen (1997). In this model, a point source releases a morphogen that creates a concentration gradient as it diffuses into the surrounding tissue where it gradually decays. After a set time, morphogen concentrations in the tissue are compared to a threshold, and the distance within which the concentration exceeds the threshold determines the phenotypic value of the trait. Nijhout and Paulsen (1997) simulated genetic variation by assuming that each parameter of the model is controlled by a "gene" with two "alleles" of different values. Here we extend this model to the study of FA by adding a small random component of "developmental noise" to each of the developmental parameters. The resulting phenotypic values can be analyzed with the statistical methods routinely used for analyzing FA and with those of quantitative genetics. Thus, we integrate the developmental and statistical models of fluctuating asymmetry into a single, unified approach.

The principal result of this study is that the developmental system alone is sufficient to produce a heritable component of asymmetry, even though the actual left-right differences are based entirely on random variation. Genetic variation of FA does not require modifier loci separate from the loci that affect trait size or special mechanisms such as a buffering system compensating for genetic or environmental stress, which appear, at least implicitly, in many discussions of this topic (e.g., Palmer and Strobeck 1992; Møller and Pomiankowski 1993a,b; Palmer 1994; Leamy et al. 1997, 1998; Møller and Swaddle 1997; Møller and Thornhill 1997; Swaddle 1997). Our analyses suggest an alternative explanation for the association of FA with heterozygosity and with trait size that is substantially different from explanations discussed in the recent literature (e.g., Clarke 1993; Mitton 1993; Møller and Pomiankowski 1993a,b; Gavrilets and Hastings 1994; Møller and Swaddle 1997). This alternative explanation emphasizes the physiological origins of dominance and epistasis and their relation to the corresponding variance components in a population (Kacser and Burns 1981; Moreno 1994; Cheverud and Routman 1995, 1996; Routman and Cheverud 1997; Lynch and Walsh 1998). Rather than making a priori assumptions about a particular mode of inheritance for FA, the new explanation ties the findings of earlier FA studies firmly to the classical theory of quantitative genetics.

The Developmental Model

Basic Model and Genetic Control

We used the model proposed by Nijhout and Paulsen (1997), which simulates the development of a simple trait based on morphogen diffusion and a threshold response. Models of morphogen gradients have been widely used in theoretical studies of patterning processes (Held 1992; Murray 1993), and have been confirmed by experimental studies in vertebrate and invertebrate model systems (reviewed by Lawrence 1992; Neumann and Cohen 1997b).

The model of the diffusion-threshold process has six parameters, each of which is controlled by a single locus with two alleles (Nijhout and Paulsen 1997). In this model, a point source produces a morphogen that can diffuse into the surrounding tissue, which is represented by a linear array, as in a section through an epithelium. The rate of release of the morphogen from the source (*Source*), and the rate of diffusion (*Diffusion*) are both parameters of the model. In the tissue, the morphogen decays at a rate proportional to its concentration (a constant percentage per unit of time; *Decay*), but a background level of morphogen is also produced throughout the tissue at a constant rate (a constant amount per unit of time; *Background*). When the morphogen source has been active for a certain period (*Time*), morphogen concentrations throughout the tissue are compared to a threshold value (*Threshold*). The phenotypic value of the trait is determined by the distance from the morphogen source to the point where the morphogen concentration equals the threshold value.

The alleles at all six loci controlling the developmental parameters act in a strictly additive manner (i.e., the parameter value of the heterozygote is the average of the two allelic values). The alleles at each locus are named "Small" and "Large" according to their effect on the phenotypic value (e.g., the Large allele for *Decay* has a lower parameter value than the Small allele, because a low decay rate results in a high phenotypic value). We used the same allelic values for developmental parameters (Table 1) as Nijhout and Paulsen (1997); these values were chosen to produce an adequate amount of morphological variation between genotypes. We also conducted simulations with different parameter values, which yielded qualitatively similar results.

Modeling Developmental Noise

Developmental noise is usually defined as small, random variation of developmental processes resulting, for example, from the stochastic nature of processes at the molecular scale (Palmer 1996; Møller and Swaddle 1997). We modeled developmental noise as small, random deviations from the genetically determined values of developmental parameters. It can thus be envisioned, for example, as stochastic variation in the activity of the metabolic pathway that produces the morphogen, random fluctuations in the rate of morphogen release or movement through the intercellular matrix, or accidental variability in the availability of receptor molecules in target cells.

TABLE 1. Values of the developmental parameters used in the simulations. The genotypic value of each parameter is governed by a single locus with two alleles, which are denoted "Small" or "Large" according to their effect on the value of the phenotypic trait. The allelic values are the genotypic values for the homozygotes, whereas genotypic values for the heterozygotes are exactly intermediate (additive gene action). In the simulations that include developmental noise, a small random deviation is added to the genotypic value on each body side separately. These random deviations are normally distributed with means of zero and standard deviations according to the Low, Medium, or High levels of developmental noise.

	Allelic values		Developmental noise (standard deviation)		
Locus	Small	Large	Low	Medium	High
Source	800	3000	8	16	40
Background	0.1	1.0	0.001	0.002	0.005
Decay	0.008	0.001	0.00001	0.00002	0.00005
Diffusion[1]	0.02	0.2	0.0002	0.0004	0.001
Time	50	200	0.5	1.0	2.5
Threshold	400	250	0.75	1.5	3.75

[1] Allelic values for *Diffusion* differ from those given by Nijhout and Paulsen (1997) by a factor of 0.2 that was included in their program. In our model, the diffusion parameter cannot exceed 0.5, which indicates complete permeability for the morphogen.

For each individual, we added the genotypic value and a random component of developmental noise specific to each body side, and used the resulting parameter values to compute trait values for left and right sides separately. The random deviations for the six developmental parameters were simulated as independent and normally distributed variables with means of zero. The standard deviations, which determined the amount of developmental noise, were set to one of three levels: Low, Medium, or High (Table 1; these values were chosen so that the random component of each of the six parameters would have a roughly similar effect on the resulting asymmetry and to ensure that no parameter values were zero or negative). Developmental noise was thus random and entirely independent of the genotype.

Our model differs in a number of respects from the one used by Graham et al. (1993). Their model considers a dynamic system of two morphogens that interact as activator and repressor, but it does not have an explicit spatial component. Instead of a morphological trait determined by the morphogens, Graham et al. (1993) analyze asymmetry in the concentration of one of the morphogens. Their model simulates developmental noise as small random deviations added directly to the concentrations at each time step. In the system of Graham et al. (1993), morphogen concentrations can oscillate and follow cyclic or even chaotic dynamics. In contrast, given sufficient time, morphogen concentrations in our system would approach an equilibrium between diffusion from the source and local decay. While oscillating systems can occur in metabolic and developmental systems (Segel 1984; Murray 1993), most developmental processes do not show oscillations. Studies of metabolism have shown that multienzyme systems can be remarkably well buffered even against large perturbations (e.g., Kacser and Burns 1981).

Our model of asymmetry also differs from the model by Graham et al. (1993), in that it does not include feedback between body sides. Although there is experimental evidence for such interactions, either by differential use of structures on the two body sides (Smith and Palmer 1994; Trinkaus et al. 1994) or by competition for a limiting resource (Klingenberg and Nijhout 1998), the underlying developmental mechanisms are not well understood. Moreover, the study of Graham et al. (1993) showed that left-right interactions are more important for antisymmetry and directional asymmetry than for FA, which is the focus of this paper (see also Van Valen 1962; Palmer and Strobeck 1992).

SIMULATION OF GENOTYPIC VARIATION

A first round of simulations concentrated on variation among genotypes without considering population parameters such as allele frequencies. For each of the 729 ($=3^6$) genotypes that are possible with the six diallelic loci, we calculated the exact phenotypic values without developmental noise. To assess genetic variation of FA, we ran simulations for 1000 individuals of each genotype and repeated these simulations separately for the Low, Medium, and High levels of developmental noise.

For each genotype, we calculated the mean of individual left-right averages of the trait values ([R + L]/2). Because these averages were very similar to the trait values computed without developmental noise, we only present the analysis for the exact trait values. We also calculated the following measures of FA: the mean of unsigned asymmetry |R − L| (the index FA1 of Palmer 1994), and the mean of its size-corrected equivalent |R − L|/([R + L]/2) (FA2 of Palmer 1994), the variance of signed asymmetry var(R − L) (FA4 of Palmer 1994) and its size-corrected version var[(R − L)/([R + L]/2)] (FA6 of Palmer 1994). The results were qualitatively similar for all indices, with some parallel differences between uncorrected and size-corrected indices (i.e., FA1 vs. FA2 and FA4 vs. FA6). Here we only present detailed data for FA1, because it is the index used most frequently, but we note where other indices produced different results.

Analysis of Individual Genotypes

There was extensive variation of both trait size and FA among genotypes, with evidence of both dominance and epistasis. Because conventional quantitative genetic estimates involve averaging over multiple genetic backgrounds, which may obscure some of these features, we first used an approach related to sensitivity analysis in dynamic modeling, focusing on the variation of single parameters and individual genotypes.

First, we examined the developmental system by considering the developmental parameters (loci) one by one. To ensure that the genetic background for these analyses was

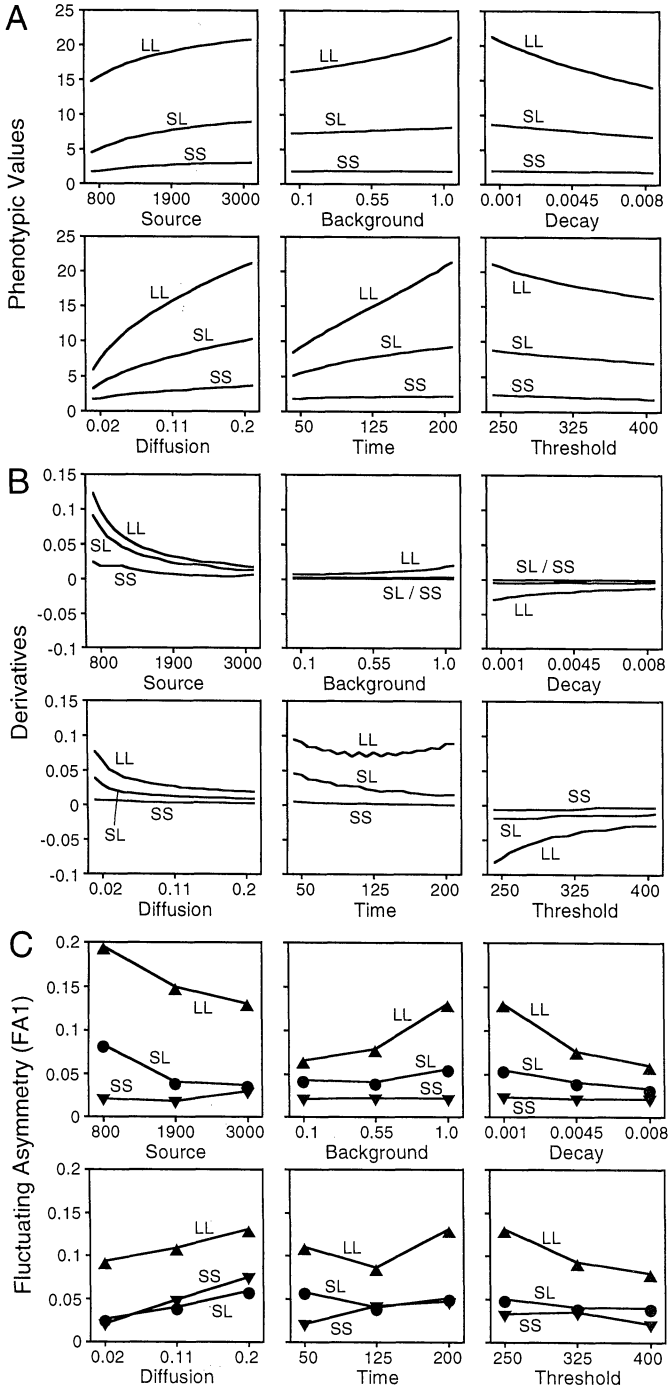

FIG. 1. Analysis of individual genotypes. (A) Exact phenotypic trait values without developmental noise, plotted as continuous functions of single developmental parameters (after Nijhout and Paulsen 1997, fig. 2). In each panel, the *x* axis shows continuous variation over the full range of parameter values, even though the corresponding loci of our genetic model each have only two alleles (the values at the tick marks indicate the genotypic values for the two homozygotes and heterozygote). The functions are graphed for three genetic backgrounds: SS, all five other loci homozygous for the Small allele; SL, all five other loci heterozygous; LL, all five other loci homozygous for the Large allele. (B) Derivatives of the functions depicted in (A), scaled by the amounts of variation used in the simulations of fluctuating asymmetry. Although calculated in an entirely deterministic model, these derivatives are an indication of the sensitivity of the system to variation in a single parameter (and therefore sensitivity to developmental noise, below). (C) Fluctuating asymmetry (FA1) of individual genotypes, simulated at the Medium level of developmental noise. These simulations used the discrete genotypes for all six loci (therefore, the plots are not continuous functions of the developmental parameters as in A and B).

comparable, we set the remaining five loci either homozygous for the Small alleles (SS genetic background), heterozygous (SL genetic background), or homozygous for the Large alleles (LL genetic background). Moreover, we calculated the phenotypic values not just for the three genotypes at the locus of interest, but as a continuous function spanning the range between the two allelic values of the corresponding developmental parameter.

Most of the lines depicting phenotypic values as functions of the developmental parameters are slightly curved (Fig. 1A). This curvature indicates partial dominance of one allele at the respective locus, because the phenotypic value of the heterozygote is closer to one of the homozygotes than to the other and not exactly intermediate. Moreover, the curvature and slope of the functions strongly depend on the genetic background, which indicates that epistasis is an important factor.

Developmental noise adds small random deviations to the genotypic values of developmental parameters (i.e., a small shift to the left or to the right from the genotypic values marked on the horizontal axis of the graphs in Fig. 1A). The extent to which a given perturbation produces a phenotypic response (in the direction of the vertical axis) can be estimated from the slope of the respective curve. A comparison of these slopes should thus give an indication of how sensitive the different genotypes are to developmental noise in the respective parameter. For this purpose, we computed the derivatives of these functions (Fig. 1B). Because the direction in which developmental noise acts in a particular instance is random, and because analyses of FA most often use a measure of absolute left-right differences, the sign of the derivatives is unimportant and we can concentrate primarily on their absolute magnitude.

For four of the six parameters (*Source, Decay, Diffusion*, and *Threshold*), the derivatives are of relatively large magnitude at low parameter values and go toward zero at higher parameter values. Sensitivity to developmental noise is thus expected to decline with increasing parameter values. Moreover, the magnitude of the derivatives decreases more in the lower half than in the upper half of the range of parameter values considered here, which reflects the fact that the curves in Figure 1A are steeper at low than at intermediate or high parameter values. Therefore, for the corresponding four loci, the homozygote for the allele with a low developmental parameter value is expected to have more FA than the heterozgote or the homozygote with a high parameter value. Overall, in these four loci, the allele with the higher parameter value (which is the Large allele for *Source* and *Diffusion*, and the Small allele for *Decay* and *Threshold*) is expected to have lower FA and to exhibit partial dominance in this respect. It is clear, however, that this interpretation is somewhat over-

simplified because the allelic effects of all four of these loci show some dependence on the genetic background.

The two remaining loci, however, behave quite differently. For the *Background* parameter, when all other loci are homozygous for the Large allele (LL), the curve becomes steeper with increasing parameter values (Fig. 1A), and thus the derivative shows an increase, not a decrease, with increasing parameter values (Fig. 1B). But still, as for the four loci discussed above, the *Background* allele with the lower level of FA is expected to be partially dominant. With SL and LL genetic backgrounds, the curves for *Background* are essentially horizontal (Fig. 1A), the derivatives zero (Fig. 1B), and no genetic effects on FA are expected. For *Time*, there is an even stronger dependence on the genetic background. On a genetic background homozygous for the Small allele at all other loci (SS), the derivative is very close to zero for all values of *Time* and it is positive and gradually declining with increasing *Time* when all other loci are heterozygous (SL). In contrast, when the other loci are homozygous LL, the derivative is high with only a small decrease at intermediate *Time* values (the roughness of the curves in Fig. 1B is due to the discrete increments of *Time* by integers). Such dependence of the behavior of one developmental parameter on the genetic background indicates that there is ample epistasis.

The levels of FA obtained by simulation correspond to these expectations in part (Fig. 1C). These simulations were only run with the discrete genotypes, but not for intermediate values of the developmental parameters. As expected, FA tends to decrease with increasing parameter values for *Source, Decay*, and *Threshold* (except for the SS genetic background), and for *Background*, FA increases with increasing parameter value. The simulations do not confirm the predictions for *Diffusion*, however, because there is a nearly linear increase of FA with increasing parameter values instead of the expected decrease.

The prediction of partial dominance for the allele with the lower level of FA also holds for several loci (except for the SS genetic background): *Source, Background, Decay*, and *Threshold*. In Figure 1C, this is visible where the lines connecting the genotypes for a particular genotype are concave upward. For *Time*, FA even exhibits underdominance (again, except for the SS genetic background), that is, the heterozygote has less FA than either of the homozygotes.

For each of the developmental parameters, the genetic background has a substantial influence on FA. For all six loci, the three genotypes with a homozygous LL genetic background are substantially more asymmetric than corresponding genotypes with SS or SL backgrounds. In Figure 1C, the lines connecting the genotypes for the SL background are consistently closer to the lines representing the SS backgrounds than to the lines for the LL background. Thus, the aggregate of all the loci making up the genetic background has a dominance effect on FA. This effect is strongest for *Diffusion* (Fig. 1C). The genetic background also has a substantial influence on the dominance effects of individual loci. For example, the lines connecting the genotypes for *Time* and *Threshold* with a LL genetic background are concave upward, whereas the corresponding lines for the SS backgrounds are convex upward—this represents a change in sign of the dominance effects for the two loci.

The data presented in Figure 1 highlight that even this fairly simple developmental model can show complex behavior. The nonlinearity of the diffusion–threshold process manifests itself at the genetic level as dominance and epistasis. In the following section we quantify these effects in a decomposition of genotypic values with the classical approach of quantitative genetics.

Quantitative Genetic Analysis

To analyze the variation among genotypes and to establish a basis for interpreting the results of selection models (below), we used the standard decomposition of genotypic values into physiological components of additive, dominance, and epistatic variation (Cheverud and Routman 1995). Physiological effects, in contrast to the average effects and components of variance in a population (see next section), refer exclusively to genotypic values and are therefore independent of population characteristics like allele frequencies or genotype frequencies. Here we limit the analysis to additive, dominance, and two-locus epistatic effects (Cheverud and Routman 1995, 1996; Routman and Cheverud 1997; Lynch and Walsh 1998). The strategy to extract the coefficients of interest is to compute unweighted averages for the effects of interest over all possible genetic backgrounds (for details of the calculation, see the Appendix; this extends the two-locus analyses described by Cheverud and Routman 1995; Routman and Cheverud 1997). Some of the variation shown in the preceding analyses (Fig. 1A,C) may thus be obscured by the averaging process (but in principle, it could be recovered from higher-level components of epistasis). In return, however, averaging across genetic backgrounds permits a simplified presentation of the overall contributions of particular loci and their mode of action.

Additive and Dominance Effects.—Physiological effects of single loci on trait size and FA for all three levels of developmental noise are presented in Table 2. For trait size, *Diffusion, Source*, and *Time* had the largest additive values (the positive signs of additive values result from the calculation via differences of Large minus Small alleles). All dominance values were of substantially smaller magnitude than the corresponding additive values, indicating that there was only partial dominance, as is consistent with the moderate nonlinearities of the curves in Figure 1A. Three loci had positive dominance values for trait size and three had negative dominance values.

The single-locus additive and dominance values for FA increased in nearly proportional manner over the three levels of developmental noise (Table 2). This suggests that, for each genotype, FA scales more or less linearly with developmental noise, at least at the levels used in our analyses (but some nonlinearity is evident from the findings on epistasis, see below). Most additive values were positive, indicating that the Large allele for most loci was associated with higher FA. The additive value of *Source*, however, was relatively large and negative.

Five of the six loci had negative dominance values, indicating that heterozygotes tend to have less FA than one would expect from the average of the two homozygotes at the same locus (the exception is *Diffusion*, which had a very small

TABLE 2. Single-locus additive and dominance effects on the trait value and fluctuating asymmetry at three levels of developmental noise (dev. noise). The measure FA1 (Palmer 1994) is the mean |R − L|, calculated for 1000 individuals per genotype. Tabulated values are the additive (*a*) and dominance (*d*) genotypic values (Falconer and Mackay 1996). Values for each locus are averaged over all genotypes at all other loci.

	Trait value		FA1, Low dev. noise		FA1, Medium dev. noise		FA1, High dev. noise	
Locus	*a*	*d*	*a*	*d*	*a*	*d*	*a*	*d*
Source	1.733	0.646	−0.0059	−0.0067	−0.0130	−0.0143	−0.0337	−0.0369
Background	0.442	−0.059	0.0029	−0.0010	0.0053	−0.0017	0.0131	−0.0036
Decay	0.786	−0.109	0.0049	−0.0006	0.0087	−0.0011	0.0216	−0.0033
Diffusion	2.758	0.658	0.0109	0.0006	0.0205	0.0010	0.0509	0.0023
Time	1.685	0.328	0.0019	−0.0019	0.0052	−0.0038	0.0146	−0.0095
Threshold	0.763	−0.096	0.0021	−0.0007	0.0039	−0.0011	0.0098	−0.0040

positive dominance value, consistent with the nearly additive effects of this locus suggested by Fig. 1C). The locus *Source* was also unusual for its dominance value, which was of slightly greater magnitude than the additive value, thus suggesting underdominance (i.e., on average the heterozygote is less asymmetric than either homozygote). *Time* also showed substantial dominance for FA (complete dominance with Low developmental noise).

The additive and dominance values presented in Table 2 are for |L − R| (the index FA1), a measure of asymmetry that is not corrected for size. The results for another size-uncorrected index (FA4) correspond closely. In contrast, the size-corrected measure FA2 shows some differences (the same applies to FA6). Most notably, the additive values for FA2 were negative not only for *Source*, but also for *Diffusion, Time*, and *Threshold* (at all levels of developmental noise). *Source* had by far the largest additive effects, followed by *Time*. Dominance values for FA2 were all negative except for *Decay*, which showed very weak positive dominance. Dominance was very marked for FA2: *Diffusion* showed strong underdominance (the absolute value of *d* exceeded *a* by a factor of more than 1.6 at all three levels of developmental noise), *Time* showed nearly complete dominance (underdominance at the High level), and although *Source* did not show underdominance as for FA1, it showed nearly complete dominance for reduced FA (i.e., for the Large allele). In summary, the Large alleles of most loci were associated with decreased relative asymmetry (FA2), but increased absolute asymmetry (FA1), and dominance was stronger overall for the size-corrected index FA2 than for FA1.

Two-Locus Epistasis.—To analyze the role of epistasis, we used the decomposition of two-locus epistasis into the following four components for each combination of loci: additive × additive, additive × dominance, dominance × additive, and dominance × dominance (Cheverud and Routman 1996; Routman and Cheverud 1997). These four components are named after their mode of action. For instance, additive × dominance epistasis characterizes how additive variation at the first locus and dominance variation at the second locus mutually influence one another.

For the trait values, additive × additive epistasis appeared to be more important than the other components, as the *aa* coefficients were larger than the coefficients for the other epistasis components in all but two of the 15 pairwise combinations of loci (Table 3). Most of the *aa* coefficients for trait size were positive, which reflects the strong tendency of the Large alleles at different loci to reinforce each other's effects (Fig. 1A). In contrast, there appeared to be no preferred direction for additive × dominance and dominance × additive epistasis on trait size. The coefficients for dominance × dominance epistasis were small for both trait size and FA, which suggests that these effects are relatively weak.

The relative importance of additive × additive epistasis for FA depended on the level of developmental noise. With increasing developmental noise, the *aa* coefficients tended to become larger relative to the other components of epistasis; at the Low level, the *aa* coefficient was larger than the three other epistasis coefficients for only one of the 15 pairs of loci, but with High developmental noise the *aa* coefficient was largest for six pairs of loci (Table 3). Two *aa* coefficients for FA were consistently negative and large (similar in magnitude to the additive and dominance values): those of *Source* and *Diffusion* and of *Source* and *Time*. Both of these, despite the negative sign, manifest themselves as mutual reinforcement of the effects of the loci involved (the sign is negative because for *Source*, unlike other loci, the Small allele tends to be associated with greater asymmetry; Table 2).

FA appeared to be affected by additive × dominance and dominance × additive epistasis to a greater extent than trait size. Some of these coefficients were similar in magnitude to both additive and dominance values, and the majority of *ad* and *da* coefficients were negative (Table 3). As for the *aa* coefficients, the locus combinations of *Source* with *Diffusion* and *Source* with *Time* had unusually large *ad* and *da* coefficients as well.

The results on two-locus epistasis for other measures of FA were similar to the ones for FA1 presented above. As for the single-locus effects, however, additive effects had reversed signs for the size-corrected indices (FA2 and FA6), and accordingly the majority of the additive × additive values were negative for these indices, rather than positive (FA1, FA4). Although there were many differences in the magnitude or sign of specific values, there also were many shared features, like the large magnitude of epistatic effects involving the locus *Source*.

In summary, while many of the epistatic effects can be interpreted by reference to the functions relating developmental parameters to phenotypes (Fig. 1), other effects cannot be explained in this manner, such as the differences in epistasis coefficients between the levels of developmental noise. These effects are a reminder of the complexity of this non-

TABLE 3. Two-locus epistasis values for trait size and FA. The values are labeled *aa* for additive × additive epistasis, *ad* for additive × dominance epistasis, *da* for dominance × additive epistasis, and *dd* for dominance × dominance epistasis (Routman and Cheverud 1997). The measure of FA is the index FA1 (mean |R − L|; Palmer 1994), calculated for 1000 individuals per genotype.

		Trait value				FA1, Low developmental noise			
		aa	*ad*	*da*	*dd*	*aa*	*ad*	*da*	*dd*
Source	*Backgr.*	−0.0191	0.0186	0.0029	0.0011	0.00047	−0.00104	0.00114	−0.00006
Source	*Decay*	0.0448	0.0721	0.0770	0.0064	0.00023	−0.00082	0.00054	−0.00044
Source	*Diffusion*	0.8358	0.3955	0.6463	0.0354	−0.00560	−0.00580	−0.00737	−0.00043
Source	*Time*	0.4972	0.2363	0.3622	0.0240	−0.00609	−0.00483	−0.00740	−0.00064
Source	*Threshold*	−0.1051	0.0158	−0.1111	−0.0017	0.00138	0.00041	0.00474	−0.00040
Backgr.	*Decay*	0.1739	−0.0935	−0.0764	0.0077	0.00148	−0.00113	−0.00123	0.00013
Backgr.	*Diffusion*	0.2141	0.1037	−0.0569	−0.0034	0.00113	0.00144	−0.00109	−0.00000
Backgr.	*Time*	0.3510	−0.1065	−0.1308	0.0136	0.00132	−0.00062	−0.00161	0.00009
Backgr.	*Threshold*	0.1362	−0.0794	−0.0647	0.0062	0.00115	−0.00122	−0.00163	0.00016
Decay	*Diffusion*	0.3942	0.1948	−0.1061	−0.0059	0.00208	0.00115	−0.00133	0.00011
Decay	*Time*	0.6483	−0.2162	−0.2570	0.0305	0.00215	0.00003	−0.00090	0.00008
Decay	*Threshold*	0.0816	−0.0300	−0.0153	0.0018	0.00034	0.00037	0.00002	−0.00026
Diffusion	*Time*	0.8513	0.3457	0.4256	0.0203	−0.00116	−0.00302	−0.00178	−0.00033
Diffusion	*Threshold*	0.3646	−0.0901	0.1713	−0.0025	0.00040	−0.00043	−0.00068	0.00013
Time	*Threshold*	0.2973	−0.1078	0.0190	0.0051	0.00005	−0.00186	−0.00167	0.00018

linear and multidimensional system, even though it results from a relatively simple developmental model.

Simulation of Genetic Variation in Populations under Selection

Selection Model

The population model and selection procedure for our simulations were the same as those described by Nijhout and Paulsen (1997), but we ran separate simulations in which the target variable was either trait size or FA. Each simulation started with an initial generation of 2000 individuals, to which genotypes were randomly assigned according to specified allele frequencies. In the simulations presented here, we set the initial allele frequencies at all loci to either 0.1 or 0.9 for the Large allele, but we also conducted simulations with other initial allele frequencies, which produced qualitatively similar results (not presented here).

The individuals of the starting generation were randomly assigned to pairs, of which each produced 10 offspring. Offspring genotypes were produced by randomly and independently taking one allele per locus from each parent (i.e., without linkage among loci). For each of the 10,000 resulting offspring, the phenotypic values for both body sides were computed with a constant Medium level of developmental noise (Table 1). This setup can thus be viewed as a completely panmictic population in a completely controlled environment.

The known genotypes and phenotypes of all offspring before selection were used to compute population statistics for each generation. The statistics included the allele frequencies for the six loci, means and phenotypic variances of trait size (average of left and right sides) and of FA (|L − R|, FA1 of Palmer 1994), additive effects (the average effect of allelic substitution, α; Lynch and Walsh 1998) and dominance values for trait size and FA, the additive genetic and dominance variances for trait size and FA, the phenotypic and additive genetic correlations between trait size and fluctuating asymmetry, and the correlation between FA and the number of heterozygous loci of each individual (the calculations for these statistics are described in the Appendix). Because all calculations were based on the complete enumeration of the genotypes and phenotypic values throughout the entire population, we treated the resulting statistics as population parameters rather than as estimates. Accordingly, we did not perform significance tests or compute standard errors. Replicates of all four simulation runs described below produced results that were similar even for rather intricate details of the temporal changes in trait size, FA, allele frequencies, and other population statistics. Thus, we conclude that the population size was sufficiently large to ensure that genetic drift had only a minor influence on the results.

Following Nijhout and Paulsen (1997), we implemented selection for either target variable as truncation selection with a threshold value set at 0.5 standard deviations below the population mean for upward selection (and 0.5 sd above the mean for downward selection). For a normally distributed population, this procedure would eliminate about 31% of the offspring (note, however, that under our model the population can deviate from the normal distribution, depending on its genetic composition). From the selected offspring, 2000 individuals were randomly drawn to form breeding pairs to produce the next generation. This procedure was repeated for 50 generations or until selection had exhausted genetic variation completely because all six loci were fixed for one of the alleles.

Selection for Increased Trait Size

Upward selection on trait size, starting from initial frequencies of 0.1 for the Large alleles at all loci, produced a strong and nearly linear response from the start of selection to generation 22, when the Large alleles at all loci were fixed or nearly fixed (Fig. 2A). In contrast, the correlated response of FA was nonlinear. After an initial increase until generation 6, asymmetry decreased slightly (to generation 12) before it increased at an accelerating rate as long as there was genetic variation.

The frequencies of the Large alleles at the six loci increased sequentially, rather than simultaneously, with the peak rates of change occurring at different times for different loci (Fig.

TABLE 3. Extended.

FA1, Medium developmental noise				FA1, High developmental noise			
aa	*ad*	*da*	*dd*	*aa*	*ad*	*da*	*dd*
0.00088	−0.00123	0.00221	−0.00008	0.00172	−0.00387	0.00348	−0.00094
0.00035	−0.00158	0.00051	−0.00053	0.00036	−0.00301	−0.00009	−0.00083
−0.01184	−0.01174	−0.01642	−0.00091	−0.03067	−0.02717	−0.04250	−0.00254
−0.01156	−0.01071	−0.01293	−0.00125	−0.02843	−0.03072	−0.03072	−0.00302
0.00281	0.00054	0.00936	−0.00063	−0.00718	0.00318	0.02205	−0.00170
0.00267	−0.00203	−0.00231	0.00010	0.00597	−0.00518	−0.00594	0.00036
0.00209	0.00164	−0.00153	−0.00026	0.00519	0.00198	−0.00335	−0.00063
0.00237	−0.00206	−0.00282	0.00023	0.00668	−0.00374	−0.00621	0.00030
0.00204	−0.00175	−0.00254	0.00031	0.00510	−0.00482	−0.00436	0.00109
0.00344	0.00275	−0.00218	0.00004	0.00822	0.00505	−0.00506	−0.00005
0.00408	−0.00110	−0.00088	0.00035	0.00976	−0.00353	−0.00513	0.00096
0.00051	0.00020	−0.00044	−0.00055	0.00126	−0.00205	−0.00128	0.00003
−0.00163	−0.00540	−0.00348	−0.00043	−0.00320	−0.01441	−0.00954	−0.00077
0.00063	−0.00109	−0.00132	0.00044	0.00135	−0.00445	−0.00307	0.00125
0.00022	−0.00369	−0.00363	0.00035	0.00115	−0.00809	−0.00929	0.00059

2B). This closely parallels the dynamics of allele frequencies in the model of Nijhout and Paulsen (1997, fig. 3), except for the slightly longer duration to fixation (23 instead of 19 generations), which is due to the small component of environmental variation introduced by developmental noise. The sequence in which the loci went to fixation corresponds to the relative magnitudes of their additive values (Table 2).

The additive effects of the loci on trait size (as measured by the average effect of allelic substitution, α) tended to increase over the course of the simulation (Fig. 2C). However, there was substantial variation around this overall trend. For instance, the effect of *Diffusion* first decreased for four generations and only increased later. These are consequences of both changes in allele frequencies at the loci concerned and of epistatic effects from other loci. The role of epistatic effects in the increase of additive effects becomes intuitively clear by reference to Figure 1A. As the simulation progressed, the genetic background changed from mostly Small alleles to mostly Large alleles and, thus, from relatively flat (SS) to steeper curves (LL). The same interpretation can also be derived from the mostly positive coefficients for additive × additive epistasis for the trait value (Table 3). The additive effects of *Source* showed large changes late in the simulation (after generation 15), when the Large allele was very close to fixation (allele frequency > 99%; accordingly, genetic effects had to be estimated from very few individuals with the rare SL and SS genotypes). These fluctuations had only a minor effect on the additive genetic variance.

The additive effects for FA (Fig. 2D) changed to a lesser degree than those for trait size (except for the erratic behavior of *Source* at extreme allele frequencies), and no consistent overall trend was present in these changes. Still, the observed changes point out that genetic effects were clearly dependent on the allele frequencies in the population.

Additive genetic variance for trait size showed two peaks (Fig. 2E). The first peak was in generations 6–11, when for several loci with the largest single-locus additive values (*Diffusion, Source*, and *Time*; Table 2) both alleles were simultaneously present in the population at intermediate frequencies. The second peak occurred shortly before the end of the simulation, when epistatic effects had increased the additive effects of *Background*, the only locus that still had intermediate allele frequencies. Two similar peaks also were present for the additive variance of FA. However, the first of these was earlier than the peak for trait size, and substantially smaller than the second one. Whereas the additive genetic variance for trait size was always substantially larger than the dominance variance, these variance components were of similar magnitude for FA during most of the simulation (generations 7–19). The dominance variances of both trait size and asymmetry did not change as much as the additive genetic variances and were roughly proportional to each other (their trajectories are nearly superimposed in Fig. 2E, but notice the different scaling of *y* axes).

The phenotypic correlation between trait size and FA was first positive, then negative (generations 5–11), then positive again, and finally reached zero in generation 22, as nearly all genetic variation had been eliminated (Fig. 2F). Throughout the simulation, these correlations were small or moderate in magnitude. The genetic correlation between trait size and asymmetry followed a similar positive–negative–positive trajectory, but these correlations were higher than the phenotypic ones at the beginning and became extreme toward the end of the simulation. The correlations between FA and heterozygosity (the number of heterozygous loci for each individual) were of moderate magnitude and varied around zero.

Selection for Decreased Trait Size

In a population with initial frequencies of 0.9 for the Large alleles of all loci, downward selection on trait size produced a decline in the target variable that was rapid initially, but then tapered off (Fig. 3A). The correlated response of FA followed the same pattern, although the decline was not as smooth as for trait size. The response of trait size to selection and the trajectories of allele frequencies (Fig. 3B) were very similar to those obtained by Nijhout and Paulsen (1997, fig. 5), except for the longer duration of the simulation (all loci were fixed after 30 instead of 21 generations).

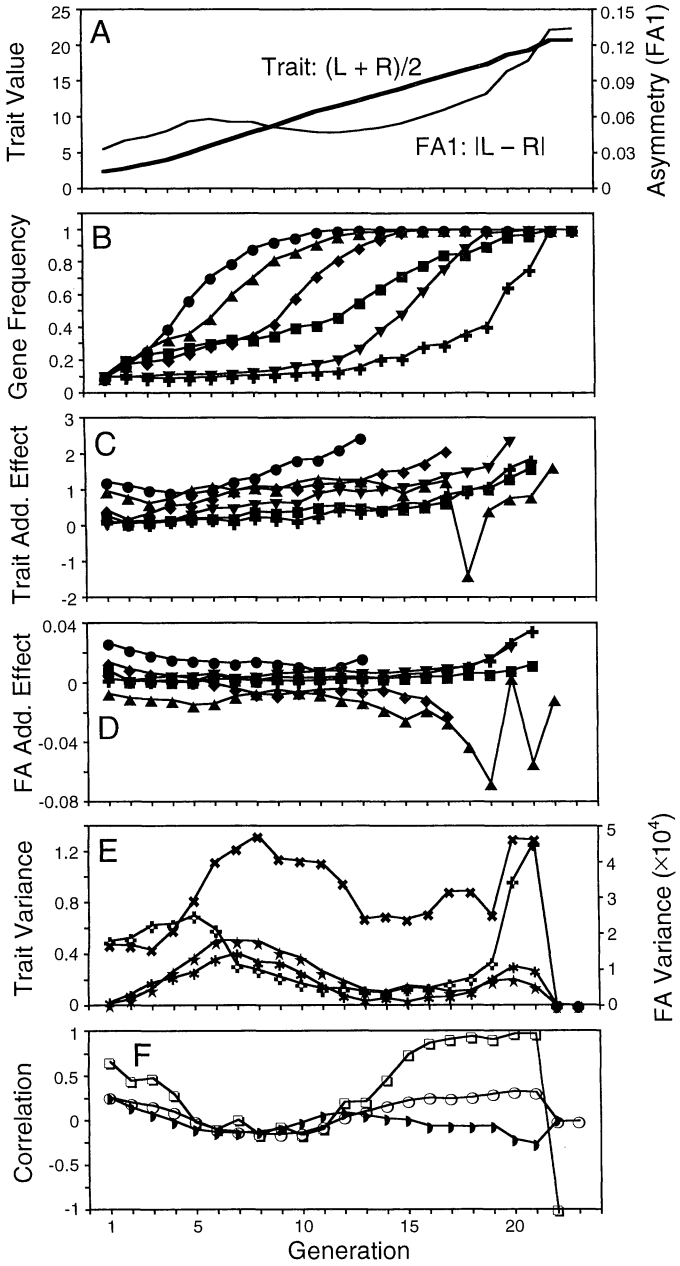

FIG. 2. Selection for increased trait value, (L + R)/2. (A) Population means for the trait value (bold line, left axis) and FA (FA1, thin line, right axis). (B) Frequencies of the Large alleles at the six loci. Symbols: ▲, *Source*; ✚, *Background*; ▼, *Decay*; ●, *Diffusion*; ♦, *Time*; ■, *Threshold*. (C) Additive effects of the loci on the trait value (L + R)/2. Plotted values are the average effect of allelic substitution, α (e.g., Lynch and Walsh 1998). Symbols are the same as for (B). (D) Additive effects of the loci on FA (FA1, Palmer 1994). Plotted values are the average effect of allelic substitution, α. Symbols are the same as for (B). (E) Additive and dominance variances for trait size and FA (FA1). Symbols: ✖, additive genetic variance of trait size; ★, dominance variance of trait size; ✜, additive genetic variance of FA1; ✱, dominance variance of FA1. (F) Correlations between FA and heterozygosity (◗), phenotypic correlations between fluctuating asymmetry and mean trait values (○), and additive genetic correlations between FA and mean trait values (□) in each generation.

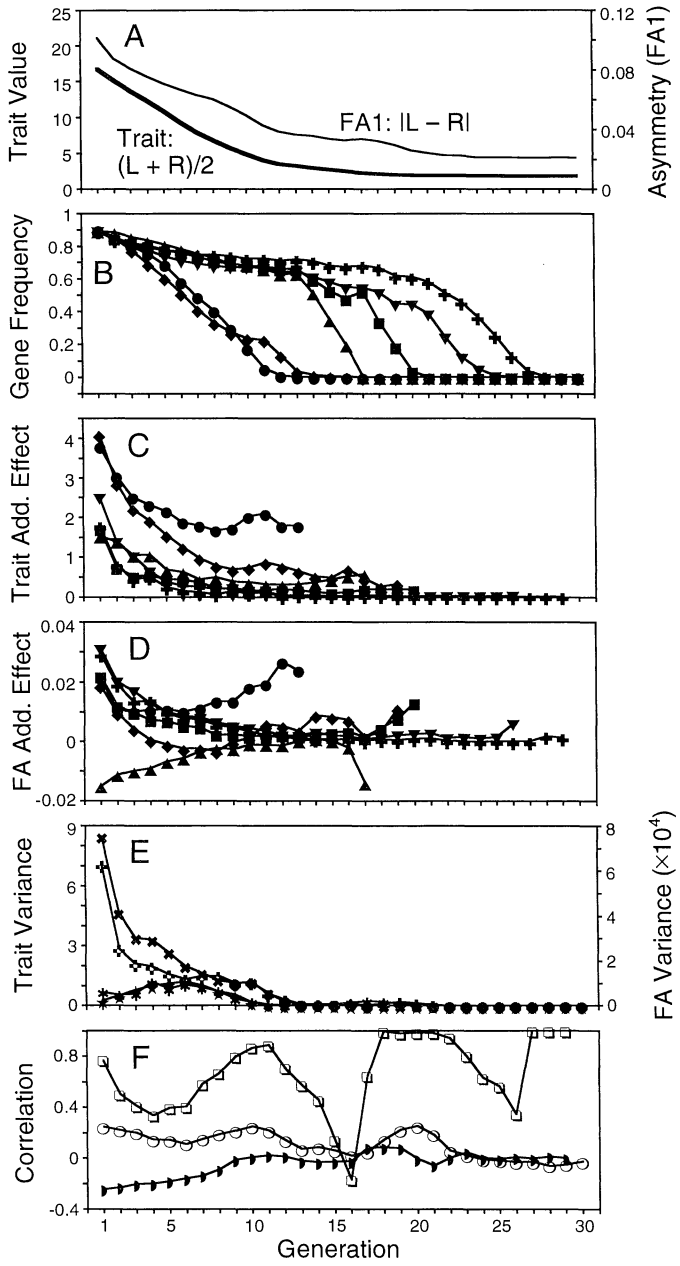

FIG. 3. Selection for decreased trait value, (L + R)/2. (A) Population means for the trait value (bold line, left axis) and FA (FA1, thin line, right axis). (B) Frequencies for the Large alleles at the six loci. Symbols: ▲, *Source*; ✚, *Background*; ▼, *Decay*; ●, *Diffusion*; ♦, *Time*; ■, *Threshold*. (C) Additive effects of the loci on the trait value (L + R)/2. Plotted values are the average effect of allelic substitution, α (e.g., Lynch and Walsh 1998). Symbols are the same as for (B). (D) Additive effects of the loci on FA (FA1, Palmer 1994). Plotted values are the average effect of allelic substitution, α. Symbols are the same as for (B). (E) Additive and dominance variances for trait size and FA (FA1). Symbols: ✖, additive genetic variance of trait size; ★, dominance variance of trait size; ✜, additive genetic variance of FA1; ✱, dominance variance of FA1. (F) Correlations between FA and heterozygosity (◗), phenotypic correlations between FA and mean trait values (○), and additive genetic correlations between FA and mean trait values (□) in each generation.

The additive effects of most loci on both trait size and FA diminished sharply during the first few generations of selection and converged toward zero (Fig. 3C,D). There were increases in the magnitude of additive effects for FA, which occurred mostly as the respective loci were approaching fixation (except for *Diffusion*, whose additive effect mostly increased during this simulation). The reason for the overall decrease of additive effects is epistasis: as the genetic background changed from a condition dominated by Large alleles to one where Small alleles were more abundant, the additive effects of the loci decreased for both trait size and FA (Fig. 1A,C). The overall decrease in the magnitude of additive effects was accompanied by a corresponding decrease in the additive genetic variances for both trait size and FA (Fig. 3E).

The phenotypic correlation between trait size and FA was positive for most of the simulation, and moderate in magnitude. In contrast, the genetic correlation between these two variables was fairly high during the first part of the simulation, but later showed sudden jumps and extreme values under the influence of rare alleles with relatively large effects on FA. Given the small amount of genetic variation for both trait size and asymmetry due to the decrease in additive effects, these genetic correlations must be interpreted with caution. The correlation between FA and heterozygosity was negative initially, and approached zero as the simulation progressed.

Selection for Increased Fluctuating Asymmetry

Selection on FA produced a substantially slower response than selection on trait size. The direct response to selection for increased FA was fairly fast during the first 15–20 generations, then gradually slowed, but still continued at generation 50, when the simulation was stopped (Fig. 4A). The correlated response of trait size followed a similar trajectory.

Changes of gene frequencies varied substantially among loci (Fig. 4B), as for selection on trait size. Initially, there was a steep increase of the Large allele of *Diffusion* and to a lesser degree of *Time* and a decrease of the Large allele of *Source*. There was also a marked increase of the Large allele for *Decay*, but only after a delay of about 20 generations. This is consistent with the additive effects of these loci on FA (Fig. 4D): *Diffusion* had a high and fairly constant positive effect, *Time* and *Decay* had a somewhat weaker positive effect, but *Source* had an increasingly strong negative effect. Except for *Source*, these additive effects changed in a fairly subtle manner during the course of the simulation, slightly increasing in their magnitude. The additive effects on trait size also increased gradually, thus providing the basis for a sustained correlated response of trait size to selection on FA (Fig. 4C; exceptions are the strong increase for *Source* and the initial decline for *Diffusion*).

The additive genetic variance for FA showed a brief initial increase (generations 1–9) and then gradually declined for the rest of the simulation (Fig. 4E). In contrast, the additive genetic variance of trait size stayed roughly constant for the first 30 generations of the simulation and later slowly increased. The dominance variances of both variables were substantially smaller than the additive variances for the respective variable and stayed fairly constant throughout most of the simulation.

Selection for Decreased Fluctuating Asymmetry

Downward selection on FA initially produced a smooth and rather slow decline in FA, but this response gradually diminished, so that by generation 40, the decrease in FA had virtually ceased (Fig. 5A). Trait size gradually declined as a correlated response to selection on FA. Notice that the level at which FA had stabilized when the simulation was stopped after 50 generations (0.036; Fig. 5A) was still *higher* than the level of FA after 30 generations of downward selection on trait size (0.021; Fig. 3A).

The changes in allele frequencies were more gradual than under the other selection regimes, and changes tended to occur at several loci simultaneously, rather than sequentially (Fig. 5B; cf. Figs. 3B, 4B). The allele frequencies at some loci stabilized at intermediate levels after initial change (e.g., *Time* after generation 11, *Diffusion* after generation 30).

The magnitude of additive effects on FA decreased markedly during the course of the simulation, and had stabilized to nearly constant levels close to zero by generation 35 (Fig. 5D). The only loci that maintained some of their additive effects were *Decay* and *Source*, but these were near fixation by that time. This reduction of additive effects on FA is similar to the one observed for downward selection on trait size (Fig. 3D), but the decrease was slower and affected all loci without exception. In contrast, the additive effects on trait size decreased less strongly, but stabilized at various levels above zero (Fig. 5C). This decrease was only minor for *Diffusion*, whereas the effect of *Source* varied around a fairly constant average level throughout the simulation.

The decrease of additive effects on FA coincided with the loss of additive genetic variance, which dropped steadily until generation 15 and subsequently continued to diminish, although at a slower rate, until the simulation was discontinued (Fig. 5E). From generation 31 on, the dominance variance of FA, which declined more slowly, exceeded the additive genetic variance. The additive genetic variance of trait size showed a modest decline initially and remained stable afterward, whereas the dominance variance remained at a fairly constant level for most of the simulation.

The phenotypic correlation between FA and trait size decreased from moderate positive values toward zero (Fig. 5F). The additive genetic correlation was initially very high, gradually decreased, and became rather unstable in the last few generations before the simulation was stopped. The correlation between heterozygosity and FA, which had moderate negative values initially, gradually became weaker, and had essentially vanished by generation 25.

Discussion

In this paper, we have presented a simple model that focuses on the developmental basis of a morphological trait and its consequences on trait variation, FA, and their inheritance. The genetic architecture of the trait and that of FA are viewed as consequences of the developmental system. Dominance, epistasis, and the correlations between FA and trait size or heterozygosity are outcomes of the model, rather

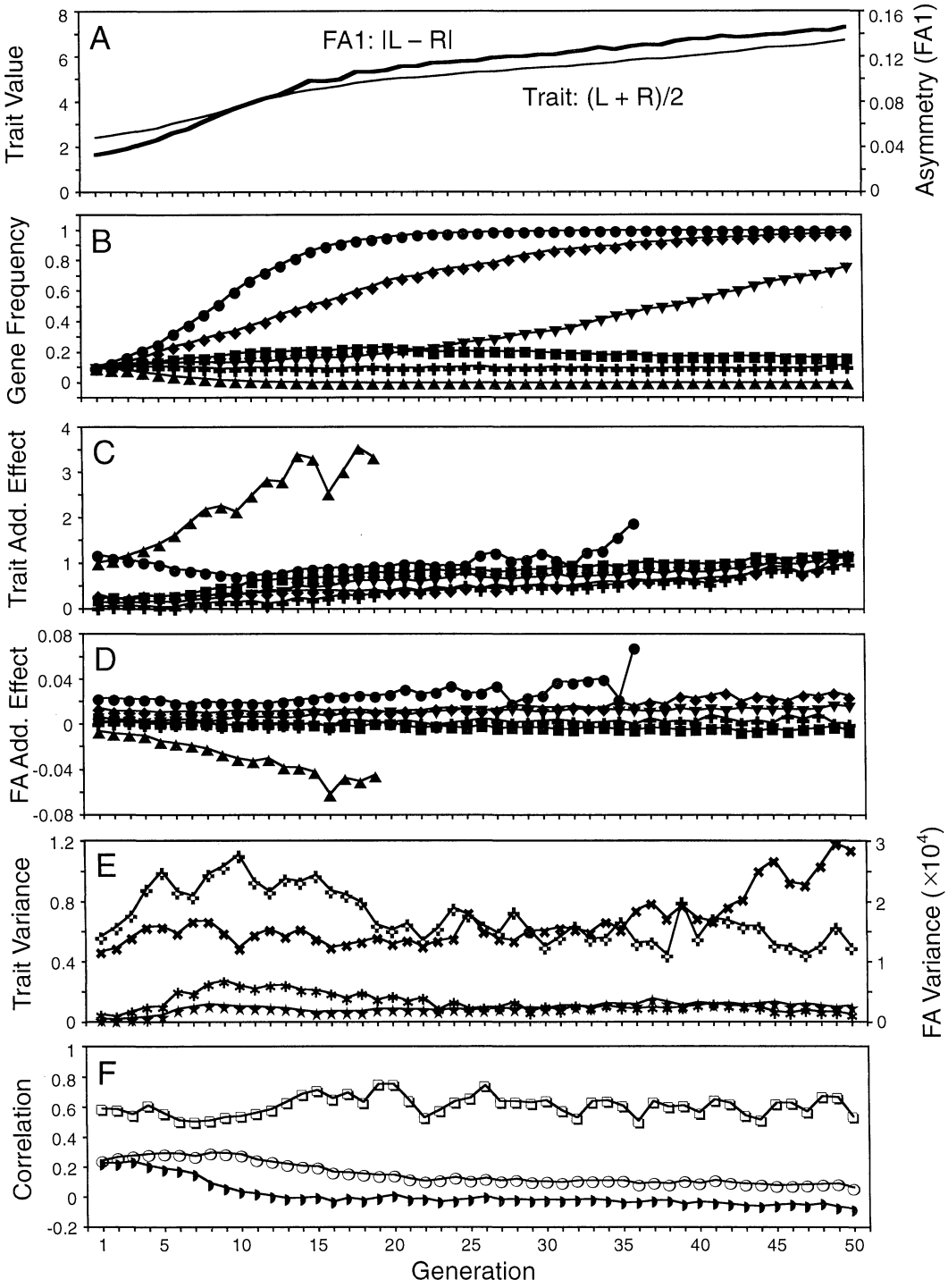

FIG. 4. Selection for increased fluctuating asymmetry, |L – R|. (A) Population means for the trait value (thin line, left axis) and FA (FA1, bold line, right axis). (B) Frequencies for the Large alleles at the six loci. Symbols: ▲, *Source*; ✚, *Background*; ▼, *Decay*; ●, *Diffusion*; ♦, *Time*; ■, *Threshold*. (C) Additive effects of the loci on the trait value (L + R)/2. Plotted values are the average effect of allelic substitution, α (e.g., Lynch and Walsh 1998). Symbols are the same as for (B). (D) Additive effects of the loci on FA (FA1, Palmer 1994). Plotted values are the average effect of allelic substitution, α. Symbols are the same as for (B). (E) Additive and dominance variances for trait size and FA (FA1). Symbols: ✖, additive genetic variance of trait size; ★, dominance variance of trait size; ✣, additive genetic variance of FA1; ✱, dominance variance of FA1. (F) Correlations between FA and heterozygosity (◑), phenotypic correlations between FA and mean trait values (○), and additive genetic correlations between FA and mean trait values (□) in each generation.

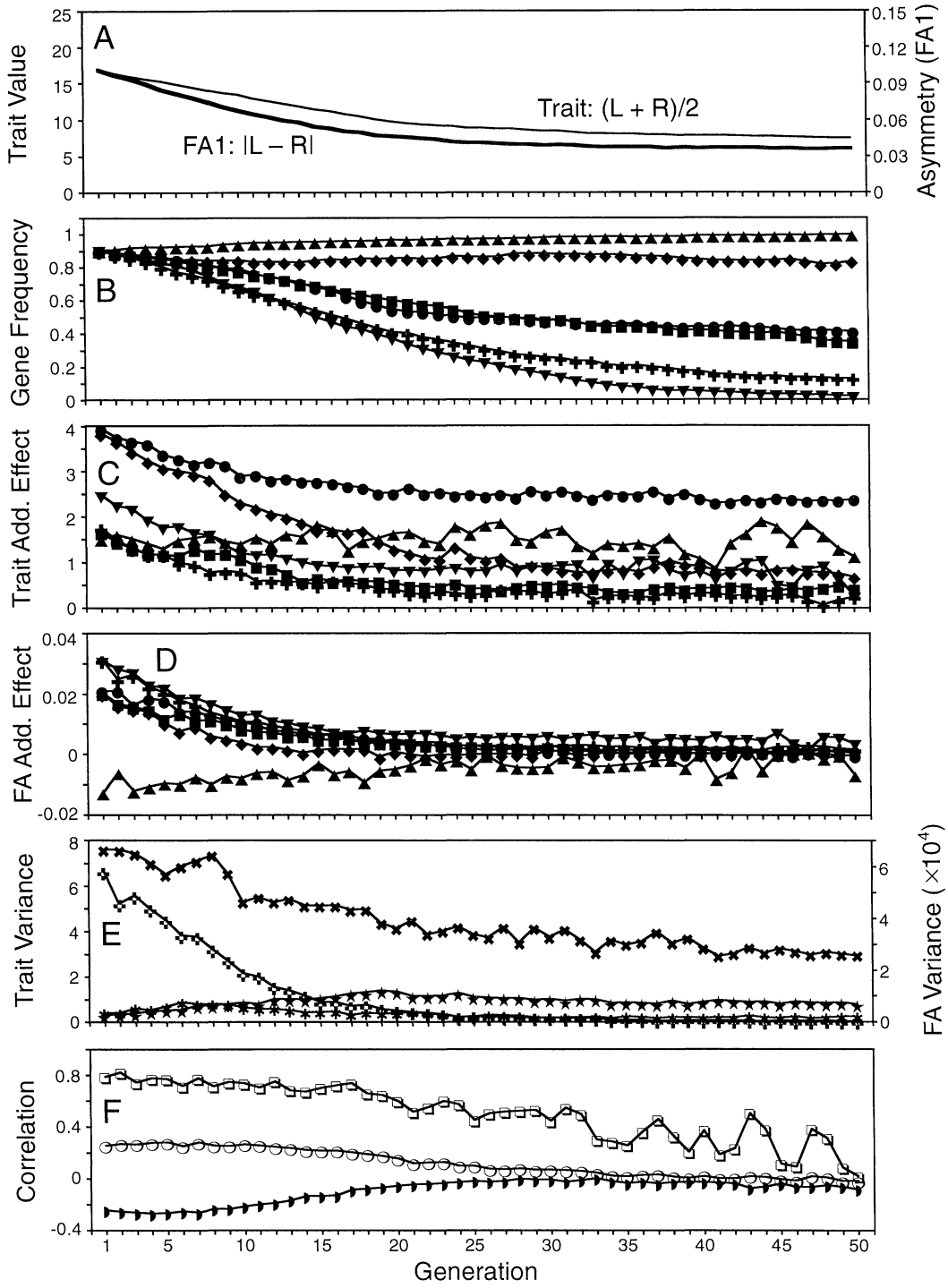

FIG. 5. Selection for decreased fluctuating asymmetry, |L – R|. (A) Population means for the trait value (thin line, left axis) and FA (FA1, bold line, right axis). (B) Frequencies for Large alleles at the six loci. Symbols: ▲, *Source*; ✚, *Background*; ▼, *Decay*; ●, *Diffusion*; ◆, *Time*; ■, *Threshold*. (C) Additive effects of the loci on the trait value (L + R)/2. Plotted values are the average effect of allelic substitution, α (e.g., Lynch and Walsh 1998). Symbols are the same as for (B). (D) Additive effects of the loci on FA (FA1, Palmer 1994). Plotted values are the average effect of allelic substitution, α. Symbols are the same as for (B). (E) Additive and dominance variances for trait size and FA (FA1). Symbols: ✖, additive genetic variance of trait size; ★, dominance variance of trait size; ✜, additive genetic variance of FA1; ✱, dominance variance of FA1. (F) Correlations between FA and heterozygosity (◗), phenotypic correlations between FA and mean trait values (○), and additive genetic correlations between FA and mean trait values (□) in each generation.

than features built into it a priori (the "built-in" component is the purely additive inheritance of developmental parameters). Therefore, this developmental model can generate many of the characteristics observed in empirical studies of FA as emergent properties. This sets our model apart from previous models that have addressed similar questions, but that have specified the genetic architecture of FA in advance (e.g., Palmer and Strobeck 1992; Gavrilets and Hastings 1994).

The principal result of our study is that genetic variation in the development of the trait is sufficient to generate an association between the genotype and the *expression* of random variation that is itself of purely nongenetic origin. Heritability of FA as well as some other phenomena observed in many studies on FA can therefore be explained without invoking "genes for FA." This means that no special mechanisms regulating asymmetry are required to account for the observed patterns (of course, the model cannot rule out that such mechanisms exist in nature, it merely demonstrates that none are necessary). Below, we discuss the broad principles underlying the results of our model, the theoretical implications for the study of FA, and the existing empirical evidence.

Genetic Architecture of Developmental Instability

Origin of Genetic Variation for FA.—Developmental instability refers to the sensitivity of developmental systems to perturbations from the environment or from within the organism itself (e.g., random variation of cellular processes). It is thus a tendency, or ability, to generate morphological variation in response to some perturbation (variability rather than variation, in the terminology of Wagner and Altenberg 1996). Developmental stability, the reverse of developmental instability, is the ability of an organism to resist perturbations and to produce a predictable target morphology regardless of developmental noise. Developmental stability is thus closely related to the concept of canalization (Waddington 1942; Møller and Swaddle 1997; Wagner et al. 1997). FA or various morphological abnormalities (phenodeviants; see Møller and Swaddle 1997) are the observable joint manifestations of developmental instability and the random perturbations that occurred during an organism's development.

We have modeled the perturbations, or developmental noise, as small random deviations from the average values of parameters in a developmental model. The sensitivity of the system to these perturbations, developmental instability, can then be characterized as the slope of a graph of the morphological trait against the value of the developmental parameter or as a partial derivative in a system with several parameters. If the curves relating a developmental parameter to the resulting phenotypes are nonlinear (e.g., Fig. 1A), the slopes change with the parameter values (Fig. 1B). Consequently, organisms with different parameter values differ in their developmental instability and their levels of FA (Fig. 1C). A nonlinear relation between the developmental parameter and the phenotype, when combined with genetic variation of the parameter, is sufficient to produce genetic variation of developmental stability.

We expect that this principle is applicable in a wide variety of biological systems, but above all, it ties the study of FA to physiological genetics. Our argument is closely related to the logic by which Kacser and Burns (1981) explained dominance on the basis of the classical Michaelis-Menten theory of enzyme kinetics (e.g., Segel 1976; Murray 1993). The basic model underlying this explanation is a series of several enzymes that are the catalysts in an unbranched biochemical pathway, in which the product of one enzymatic step is the substrate for the next one. Consider a situation in which one of the enzymes in the pathway varies in its activity, for example, because of a mutation, while the other enzymes have constant activities. As a result, the flux through the pathway will be a nonlinear function of the activity of the one variable enzyme (Fig. 6A). In such a system it is plausible that the activity of the variable enzyme will vary in an additive manner, that is, that the activity of the heterozygote (*aA*) will be midway between the activity levels of the "wild type" (*AA*) and "mutant" (*aa*) homozygotes. The resulting metabolic flux in the heterozygote, however, will be much closer to the *AA* than to the *aa* homozygote, and thus the *A* allele will appear dominant over the *a* allele. If the metabolic flux through this pathway affects the phenotype, corresponding dominance will also be expressed at the morphological level.

Figure 6B shows the derivative of metabolic flux with regard to the activity of the variable enzyme. The heterozygote is more similar to the *AA* homozygote than to the *aa* homozygote, and the dominance of the *A* allele over the *a* allele therefore applies to the derivative as well. Kacser and Burns (1981) used a scaled version of this derivative, called sensitivity coefficient, in the context of metabolic control. This derivative directly expresses the sensitivity of the system to small perturbations, which, in the context of morphological traits, is developmental instability. This line of reasoning firmly links dominance to the genetic basis of developmental stability. Both rely on the nonlinearity of the function that relates the phenotype to an underlying developmental or biochemical value or, in the context of discrete genotypes, to the dosage of one of the alleles (e.g., zero, one, or two copies of the *A* allele in Fig. 6).

Because the crucial factor is not dominance per se, but variation among genotypes in the slope of the relationship between a developmental parameter and the phenotype, genetic variation for developmental instability can also be caused by other nonadditive genetic effects that influence this slope. For instance, with additive × additive epistasis, one locus can influence the additive effect of another locus (Cheverud and Routman 1996) and therefore can cause genetic variation for developmental stability through the developmental parameter controlled by the second locus. An example of this is the collective influence of the genetic background on the additive effects of *Time* (Fig. 1A).

Epistasis is rampant in our system because gene effects depend strongly on the genetic background (Fig. 1, Table 3). Therefore, considerations of the effects of single loci, including the arguments made above, are conditional on a given genetic background. Averaging over multiple genetic backgrounds (e.g., Tables 2, 3) may not fully capture the intricate interactions among loci (Fig. 1). Epistasis is also the reason why predictions based on derivatives with respect to single developmental parameters were often inaccurate or only in

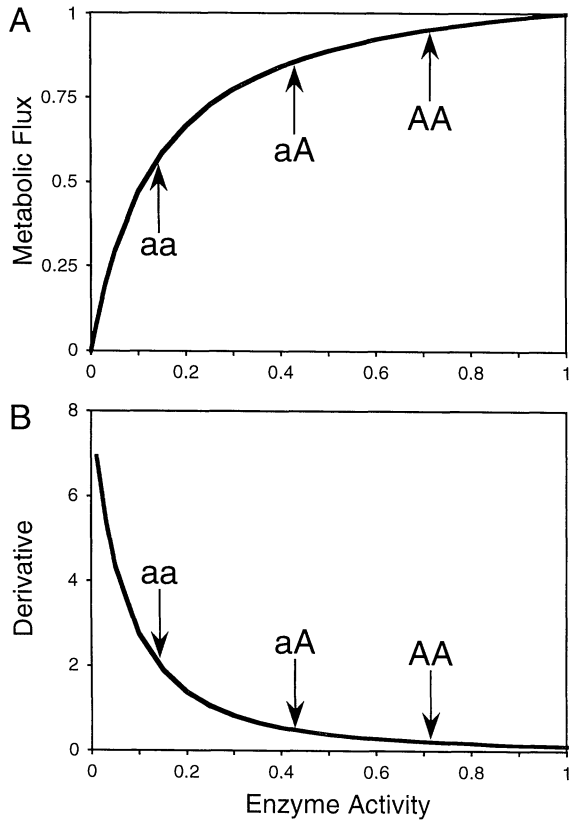

FIG. 6. The biochemical basis of dominance and its relation to developmental stability. (A) A model of metabolic flux in a linear chain of eight enzymes (Kacser and Burns 1981). Seven of these enzymes have a constant activity of 1.0, and the activity of only one enzyme is variable (abscissa). Arrows show the three genotypes for a locus. Although the enzyme activity for the heterozygote (*aA*) is exactly intermediate between the two homozygotes, its metabolic flux is much closer to the *AA* homozygote than to the *aa* homozygote, which implies that the *A* allele is dominant. (B) The derivative of the curve in (A). The value of the derivative for the *aa* homozygote is substantially higher than for the heterozygote and *AA* homozygote, rendering the *aa* homozygote more sensitive to perturbations in activity than the other two genotypes. If the metabolic flux affects a morphological trait, variations in enzyme activity can be interpreted as developmental noise. Consequently, the *A* allele is dominant with regard to FA. As the derivative decreases with increasing enzyme activity, sensitivity to developmental noise will also decrease. Moreover, unless the *a* allele is rare, the FA of the heterozygote will be less than the average asymmetry of both homozygotes, thereby establishing a negative relation between heterozygosity and FA.

qualitative agreement with the simulation results (Fig. 1C). These predictions consider only perturbations of a single developmental parameter and implicitly assume that the effects of perturbations of the other five parameters will average out, but this may not be the case in reality. If the gene under consideration has an epistatic effect on other loci, the sensitivity to perturbations of the respective developmental parameters may change as well. As a result, FA in response to perturbations of all six parameters will not exactly match the expectations derived from considering a single parameter alone. In these cases, improved predictions should be possible by considering partial derivatives with respect to multiple parameters.

Epistasis has been shown empirically to be important for FA. The clearest case is that of the Australian sheep blowfly, where a gene (*Rop-1*) that confers resistance to the insecticide diazinon also has a dominant effect that substantially increases the level of asymmetry in bristle numbers. A second gene (*Modifier*, also dominant) can interact with *Rop-1* and restore asymmetry to the levels of susceptible flies (McKenzie and Clarke 1988; see also Batterham et al. 1996). Epistasis is also inherent in the argument that inbreeding or hybridization can increase FA because of the breakup of coadapted gene complexes (Clarke 1993; see also Møller and Swaddle 1997).

Gene Effects on FA.—In Figure 6A, the function relating enzyme activity to metabolic flux consists of a steep part to the left, in which the homozygote for the recessive *a* allele is situated, and a more level part in which the heterozygote and *AA* homozygote are found. Thus the allele that is partially dominant with respect to the phenotypic value is associated with a smaller (absolute) slope than the recessive allele.

Two hypotheses follow from this reasoning. First, an allele that is partially or completely dominant with regard to trait size should have a negative additive effect on developmental instability and thus on FA. Moreover, because the slope of the heterozygote is closer to the *AA* than to the *aa* homozygote, the slope (and developmental instability) of the heterozygote is less than the average of the two homozygotes. Therefore, second, the allele associated with the lower level of FA should be dominant with respect to FA, which means that the dominance value for FA should be negative. The strength of both these effects will depend on the strength of the dominance for trait size (relative to the additive effect). Both hypotheses are derived from the curvature of the function relating the developmental parameter and the phenotypic value; to the extent that the developmental processes can be characterized by considering one parameter at a time, this should therefore be applicable to a wide variety of traits.

In our developmental model, the partially dominant allele for trait size has reduced FA and, thus, satisfies the first hypothesis for the loci *Source, Background, Decay*, and *Threshold* (Fig. 1A, C, Table 2; notice that there is partial dominance for the Large allele at *Source*, whereas there is partial dominance for the Small allele at *Background, Decay*, and *Threshold*). Furthermore, for all but one of the loci (*Diffusion*) in our model, the dominance values for FA are negative (Table 2) and, thus, are in accordance with the second hypothesis.

Unfortunately, it is difficult to evaluate this hypothesis for empirical data because of the paucity of case studies in which the necessary information is available. The best-documented example concerns two genes conferring insecticide resistance in the Australian sheep blowfly. Resistant alleles at two loci (*Rop-1* and *Rdl*) also greatly increase levels of asymmetry in bristle counts, and are dominant in this respect (McKenzie and Clarke 1988; McKenzie and Yen 1995). In the presence of the *Rop-1* resistant allele, a dominant allele at the *Modifier* locus can reduce asymmetry to normal levels (McKenzie and Clarke 1988). However, it does not appear that the mechanism of our model applies in this case, because neither *Rop-1* nor *Modifier* have a sufficiently strong effect on the mean bristle counts (J. A. McKenzie, pers. comm.). Yet, the extremely

strong effects of these three genes on FA suggest that they may not be representative of the genes affecting FA in other species.

Leamy et al. (1997, 1998) conducted two analyses of quantitative trait loci (QTLs) for FA and directional asymmetry in mice, focusing on mandible measurements and qualitative characters of the skull, respectively. The results of these studies (and of an extra analysis provided by L. J. Leamy, pers. comm.) are partly consistent with the two hypotheses, but also produced some patterns that did not conform to the expectations. Although these results do not allow a firm evaluation of the hypothesis, they point out that QTL analyses, and especially a directed search for QTLs with joint effects on both trait size and FA, are a promising approach for a more thorough assessment.

Heterozygosity and FA.—The arguments above have led to the prediction that the allele with lower FA should be dominant, that the dominance values for FA should be negative, and thus that the heterozygote should display less FA than the *average* of the two homozygotes. This is a new explanation for the association between heterozygosity and FA, which has been observed in many empirical studies and has been discussed extensively in the literature (reviewed by Clarke, 1993; Mitton 1993, 1997, p. 98 ff.; Møller and Swaddle 1997). Previously, Mitton (1993, p. 58 ff.) suggested that slower turnover of enzymes in heterozygotes may lead to lower metabolic costs, and thereby reduce developmental instability (see also Møller and Swaddle 1997). This explanation presumes that the half-life of a given allelic form of an enzyme is longer when two forms of the enzyme co-occur than when there is a double dosage of the same allelic form. Our alternative explanation does not require that cellular processes depend on the co-occurrence of allelic forms, because differences in the catalytic activities of the allelic forms of an enzyme are sufficient to account for the observed pattern.

To evaluate our explanation, data are needed that distinguish not only between homozygous and heterozygous individuals, but also between the two homozygotes (see also Lynch and Walsh 1998, pp. 118–120). The hypothesis of a true heterozygote advantage predicts underdominance of FA, that is, that heterozygotes have higher developmental stability than both homozygotes. In contrast, under our alternative hypothesis the patterns of dominance for FA should be a function of the patterns of dominance for trait size and also should be related to the additive effect on FA. Leary et al. (1993) reported that rainbow trout heterozygous or homozygous for null alleles of lactate dehydrogenase showed increased FA. However, low sample sizes and the heterogeneous genetic background make it difficult to interpret the results with regard to dominance for trait size. Unfortunately, most published studies of the relation between FA and heterozygosity have lumped together the homozygotes of all alleles and often have not indicated their relative frequencies.

Overdominance and Underdominance.—A true heterozygote advantage with respect to FA is expected if there is overdominance or underdominance of trait size, that is, where the function linking the developmental parameter to the phenotype is $\cap$- or $\cup$-shaped, respectively. In both cases, the absolute values of the derivatives for both homozygotes are higher than for the heterozygote, and developmental instability will therefore show underdominance. Notice, however, that over- or underdominance of trait size is not a necessary condition for underdominance of FA (e.g., see *Time*, Fig. 1; *Source*, Table 2).

Underdominance of FA has been found for several QTLs in a cross of two mouse strains (Leamy et al. 1997, 1998), but these studies also revealed several QTLs with overdominance for FA, which cannot be explained by this model. To generate overdominance of developmental instability following the same logic, the derivative of the function relating trait size to a developmental parameter should be steeper for the heterozygotes than for both homozygotes, as in a sigmoid curve. Simple dominance is unable to generate such a curve, but sigmoid curves are typical for the dynamics of allosteric enzymes, where substrates interact cooperatively with the enzyme (e.g., Segel 1976).

Trait Size and FA.—Because both trait size and developmental instability depend upon the developmental parameter, variation in the parameter value can generate an association between FA and trait size. The form of this relationship, for instance, the sign and magnitude of the correlation, will vary with the properties of each locus and with the genetic background (cf. Fig. 1). For loci like *Source* (Fig. 1) or in a situation as Figure 6, where the allele with the higher parameter value is partially dominant, this will result in a negative correlation among individuals between trait size and FA. Such negative correlations between trait size and asymmetry have been reported from various organisms (Møller and Pomiankowski 1993b; Rowe et al. 1997) and have been explained with models of reliable signaling in the context of models for sexual selection (Møller and Pomiankowski 1993b; Møller and Swaddle 1997). Under the appropriate conditions, however, variation in developmental parameters alone is sufficient to generate the same pattern.

Moreover, for most of the time in all our simulations of selection, both the phenotypic and additive genetic correlations between FA and trait size were positive. There was only one instance in which these correlations changed sign and were negative for a few generations during upward selection on trait size (Fig. 2F). Because the single-locus additive values (a) for FA had the same sign as those for trait size for all loci except *Source* (Table 2), positive correlations are likely to occur under a broad range of conditions. Assuming a model in which genetic variation of FA is due to modifier loci affecting the canalization (variance) of a trait, Møller and Pomiankowski (1993a,b) predicted that positive correlations between FA and trait size would occur under directional selection for increased trait size and suggested that such correlations may be used as diagnostic tools to identify selective regimes. Because our model suggested alternative explanations for both positive and negative correlations between FA and trait size and the simulations of selection yielded positive correlations no matter which selective regime was applied, we conclude that inferences about selection based on correlations between FA and trait size should only be made with utmost caution, if at all.

Response to Selection

The simulations of selection produced remarkably varied results because the dynamics of the response to selection

depend strongly on the context. Both the target variable (trait size vs. FA) and the direction of selection, in conjunction with the initial genetic composition of the population, are important determinants of the response.

Direction of Selection.—Upward and downward selection differed substantially in their dynamics. Under upward selection, the magnitude of additive effects on both FA and trait size mostly remained constant or increased as the loci went toward fixation. In contrast, under downward selection, the additive effects tended to diminish. This is a consequence of epistasis in the genetic system of our model. The values of additive × additive epistasis were positive for most pairs of loci, both for trait size and for FA (Table 3), which indicates that the additive effect of a given gene tended to be enhanced by Large alleles at other loci (see also Fig. 1). Therefore, as the proportion of Large alleles in the genetic background increased under upward selection, the additive effects of the loci that were still polymorphic were enhanced. This epistasis sustained relatively constant levels of additive genetic variance even as the loci with the largest single-locus effects were going toward fixation (Figs. 2E, 4E). Therefore, the overall response to upward selection on trait size was nearly linear until all loci were fixed (Fig. 2A), and for upward selection on FA, a sustained response was present for the entire duration of the simulation (Fig. 4A). In contrast, the initial response to downward selection reduced the proportion of Large alleles and therefore also reduced the average magnitude of additive effects in the population by epistasis, and eroded the additive genetic variance much faster than by merely reducing the frequencies of Large alleles (Figs. 3E, 5E). As a result, the response to selection reached very low levels long before all loci were fixed (under downward selection for trait size, Fig. 3) or before the simulation was ended (selection for FA, Fig. 5). This effect on the dynamics of selection response is an important consequence of epistasis and its contribution to additive genetic variance (Cheverud and Routman 1996).

The predominance of positive values for additive × additive epistasis results from the model and the parameter values assigned to the alleles (Table 1). The functions relating parameter values to phenotypes (Fig. 1A) are relatively steep near the position of the homozygotes for the Large alleles (at least on a LL genetic background), which indicates that the size of the trait does not approach some plateau where mutual enhancement of gene effects would become less effective. Higher allelic values of the loci *Background, Decay*, and *Time* may produce such a plateau.

Target of Selection.—The second consistent finding of our simulations was that the response to selection was much slower in simulations where selection was on FA rather than on trait size. This is most evident for downward selection; after selecting against FA for 50 generations, the FA level was still substantially higher than after 30 generations of selection for small trait size (cf. Figs. 3A, 5A). The correlated response of FA to selection on trait values was stronger than the direct response to selection on FA, and to achieve lower FA, it therefore would be more effective to select on trait size rather than on FA directly. While the conditions necessary for this phenomenon are rather restrictive (Falconer and Mackay 1996, p. 319 ff.), and this specific finding thus appears to be a consequence of the special circumstances of our simulation, it still illustrates the point that selection on FA can generally be expected to be less effective than selection on trait size.

This lower effectiveness of selection for FA is due to the fact that the random component of asymmetry weakens the association between and individual's FA and its genotype. This has been emphasized by recent studies that pointed out that using FA to infer an individual's developmental stability is equivalent to estimating a variance from two data points (Whitlock 1996, 1998; Van Dongen 1998). Accordingly, the estimate of developmental instability has a high sampling error and there is an upper limit that the heritability of observed FA cannot exceed (Whitlock 1996, 1998; Van Dongen 1998). Therefore, the observed FA is a rather poor predictor of an individual's genotype, which must limit the effectiveness of selection on FA. That the system is fairly sensitive to this influence of nongenetic variation from developmental noise is underscored by the fact that time to fixation was slightly longer in our simulations of selection on trait size than in those of Nijhout and Paulsen (1997), which did not include any nongenetic variation.

Conclusions

Our study showed that a developmental model is sufficient to generate heritable variation in FA as well as many of the patterns observed in empirical FA studies, for instance, the associations between FA and heterozygosity or trait size. The model therefore suggests new explanations for these patterns. While different developmental models may achieve the same result, the decisive property is that the functions relating the developmental parameters to the resulting phenotype are nonlinear. When combined with genetic variation for the developmental parameters, this nonlinearity translates into dominance and epistasis, which are central concepts for integrating the study of FA into the theory of quantitative genetics.

The discussion about the genetic basis of FA has traditionally focused on models in which the control of developmental instability is separate from that of trait size, for instance, through specific modifier loci that are distinct from the loci affecting trait size. Here we have explored a model in which trait size and FA result jointly from the same developmental system. Heritable variation of FA is due to the genetically modulated expression of random noise that is itself independent of the genotype. There is therefore no clear-cut dichotomy between genetic and environmental variation. In addition, the model also underscores the fact that heritable variation observed for a trait does not automatically imply that it is generated by genes that specifically control that trait. Genetic architecture and external appearance of variation need not correspond in a one-to-one manner.

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Literature Cited

Batterham, P., A. G. Davies, A. Y. Game, and J. A. McKenzie. 1996. Asymmetry—where evolutionary and developmental genetics meet. BioEssays 18:841–845.

Cheverud, J. M., and E. J. Routman. 1995. Epistasis and its contribution to genetic variance components. Genetics 139: 1455–1461.

———. 1996. Epistasis as a source of increased additive genetic variance at population bottlenecks. Evolution 50:1042–1051.

Clarke, G. M. 1993. The genetic basis of developmental stability. I. Relationships between stability, heterozygosity and genomic coadaptation. Genetica 89:15–23.

Emlen, J. M., D. C. Freeman, and J. H. Graham. 1993. Nonlinear growth dynamics and the origin of fluctuating asymmetry. Genetica 89:77–96.

Falconer, D. S., and T. F. C. Mackay. 1996. Introduction to quantitative genetics. 4th ed. Longman, Essex, U.K.

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

Gilbert, S. F. 1997. Developmental biology. 5th ed. Sinauer, Sunderland, MA.

Graham, J. H., D. C. Freeman, and J. M. Emlen. 1993. Antisymmetry, directional asymmetry, and dynamic morphogenesis. Genetica 89:121–137.

Held, L. I., Jr. 1992. Models for embryonic periodicity. Karger, Basel, Switzerland.

Houle, D. 1997. Comment on "A meta-analysis of the heritability of developmental stability" by Møller and Thornhill. J. Evol. Biol. 10:17–20.

Huxley, J. S. 1932. Problems of relative growth. Dial Press. Reprinted 1993, Johns Hopkins Univ. Press, Baltimore, MD.

Kacser, H., and J. A. Burns. 1981. The molecular basis of dominance. Genetics 97:639–666.

Klingenberg, C. P., and H. F. Nijhout. 1998. Competition among growing organs and developmental control of morphological asymmetry. Proc. R. Soc. Lond. B Biol. Sci. 265:1135–1139.

Lawrence, P. A. 1992. The making of a fly: the genetics of animal design. Blackwell Scientific, Oxford, U.K.

Leamy, L. 1984. Morphometric studies in inbred and hybrid house mice. V. Directional and fluctuating asymmetry. Am. Nat. 123: 579–593.

———. 1997. Is developmental stability heritable? J. Evol. Biol. 10:21–29.

Leamy, L. J., E. J. Routman, and J. M. Cheverud. 1997. A search for quantitative trait loci affecting asymmetry of mandibular characters in mice. Evolution 51:957–969.

———. 1998. Quantitative trait loci for fluctuating asymmetry of discrete skeletal characters in mice. Heredity 80:509–518.

Leary, R. F., F. W. Allendorf, and K. L. Knudsen. 1993. Null alleles at two lactate dehydrogenase loci in rainbow trout are associated with decreased developmental stability. Genetica 89: 3–13.

Lynch, M., and B. Walsh. 1998. Genetics and analysis of quantitative traits. Sinauer, Sunderland, MA.

Markow, T. A. 1995. Evolutionary ecology and developmental instability. Annu. Rev. Entomol. 40:105–120.

Markow, T. A., and G. M. Clarke. 1997. Meta-analysis of the heritability of developmental stability: a giant step backward. J. Evol. Biol. 10:31–37.

McKenzie, J. A., and G. M. Clarke. 1988. Diazinon resistance, fluctuating asymmetry and fitness in the Australian sheep blowfly, *Lucilia cuprina*. Genetics 120:213–220.

McKenzie, J. A., and J. L. Yen. 1995. Genotype, environment and the asymmetry phenotype. Dieldrin-resistance in *Lucilia cuprina* (the Australian sheep blowfly). Heredity 75:181–187.

Mitton, J. B. 1993. Enzyme heterozygosity, metabolism, and developmental stability. Genetica 89:47–65.

———. 1997. Selection in natural populations. Oxford Univ. Press, New York.

Møller, A. P., and A. Pomiankowski. 1993a. Punctuated equilibria or gradual evolution: fluctuating asymmetry and variation in the rate of evolution. J. Theor. Biol. 161:359–367.

———. 1993b. Fluctuating asymmetry and sexual selection. Genetica 89:267–279.

Møller, A. P., and J. P. Swaddle. 1997. Asymmetry, developmental stability, and evolution. Oxford Univ. Press, Oxford, U.K.

Møller, A. P., and R. Thornhill. 1997. A meta-analysis of the heritability of developmental stability. J. Evol. Biol. 10:1–16.

Moreno, G. 1994. Genetic architecture, genetic behavior, and character evolution. Annu. Rev. Ecol. Syst. 25:31–44.

Murray, J. D. 1993. Mathematical biology. 2d ed. Springer, New York.

Neumann, C. J., and S. M. Cohen. 1997a. Long-range action of Wingless organizes the dorsal-ventral axis of the *Drosophila* wing. Development 124:871–880.

———. 1997b. Morphogens and pattern formation. BioEssays 19: 721–729.

Nijhout, H. F., and S. M. Paulsen. 1997. Developmental models and polygenic characters. Am. Nat. 149:394–405.

Oster, G. F. 1988. Lateral inhibition models of developmental processes. Math. Biosci. 90:265–286.

Palmer, A. R. 1994. Fluctuating asymmetry analyses: a primer. Pp. 335–364 *in* T. A. Markow, ed. Developmental instability: its origins and evolutionary implications. Kluwer, Dordrecht, The Netherlands.

———. 1996. Waltzing with asymmetry. BioScience 46:518–532.

Palmer, A. R., and C. Strobeck. 1986. Fluctuating asymmetry: measurement, analysis, patterns. Annu. Rev. Ecol. Syst. 17:391–421.

———. 1992. Fluctuating asymmetry as a measure of developmental stability: implications of non-normal distributions and power of statistical tests. Acta Zool. Fennica 191:57–72.

Palmer, A. R., C. Strobeck, and A. K. Chippindale. 1993. Bilateral variation and the evolutionary origin of macroscopic asymmetries. Genetica 89:201–218.

Roff, D. A. 1997. Evolutionary quantitative genetics. Chapman & Hall, New York.

Routman, E. J., and J. M. Cheverud. 1997. Gene effects on a quantitative trait: two-locus epistatic effects measured at microsatellite markers and at estimated QTL. Evolution 51:1654–1662.

Rowe, L., R. R. Repasky, and A. R. Palmer. 1997. Size-dependent asymmetry: fluctuating asymmetry versus antisymmetry and its relevance to condition-dependent signaling. Evolution 51:1401–1408.

Segel, I. H. 1976. Biochemical calculations. 2d ed. Wiley, New York.

Segel, L. A. 1984. Modeling dynamic phenomena in molecular and cellular biology. Cambridge Univ. Press, Cambridge, U.K.

Smith, L. D., and A. R. Palmer. 1994. Effects of manipulated diet on size and performance of brachyuran crab claws. Science 264:710–712.

Swaddle, J. P. 1997. On the heritability of developmental stability. J. Evol. Biol. 10:57–61.

Trinkaus, E., S. E. Churchill, and C. B. Ruff. 1994. Postcranial robusticity in *Homo*. II. Humeral bilateral asymmetry and bone plasticity. Am. J. Phys. Anthropol. 93:1–34.

Van Dongen, S. 1998. How repeatable is the estimation of developmental stability by fluctuating asymmetry? Proc. R. Soc. Lond. B Biol. Sci. 265:1423–1427.

Van Valen, L. 1962. A study of fluctuating asymmetry. Evolution 16:125–142.

Waddington, C. H. 1942. Canalization of development and the inheritance of acquired characters. Nature 150:563–565.

Wagner, G. P., and L. Altenberg. 1996. Complex adaptations and the evolution of evolvability. Evolution 50:967–976.

WAGNER, G. P., G. BOOTH, AND H. BAGHERI-CHAICHIAN. 1997. A population genetic theory of canalization. Evolution 51:329–347.

WHITLOCK, M. 1996. The heritability of fluctuating asymmetry and the genetic control of developmental stability. Proc. R. Soc. Lond. B Biol. Sci. 263:849–854.

———. 1998. The repeatability of fluctuating asymmetry: a revision and extension. Proc. R. Soc. Lond. B Biol. Sci. 265:1429–1431.

WHITLOCK, M. C., AND K. FOWLER. 1997. The instability of studies of instability. J. Evol. Biol. 10:63–67.

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APPENDIX: CALCULATION OF QUANTITATIVE GENETIC PARAMETERS

Computation of Physiological Effects from Genotypic Values

Physiological gene effects were analyzed using genotypic values: either trait values computed without developmental noise or measures of FA from simulations with 1000 individuals per genotype (Tables 2, 3). Our analysis extends the methods of Cheverud and Routman (1995) and Routman and Cheverud (1997) from the two-locus case to our simulation with six loci.

Single-locus additive and dominance effects are averaged over all possible genetic backgrounds at the remaining five loci. Consider one of the loci in the simulation, and let G_{SS} ..... G_{SL} ....., and G_{LL} ..... be the genotypic values of the SS, SL, and LL genotypes at this locus averaged over all combinations of genotypes at the other five loci (i.e., each of these values is the average of $3^5 = 243$ genotypic values). The additive (a) and dominance (d) effects for this locus can then be calculated as follows:

$$a = (G_{LL} ..... - G_{SS} .....)/2 \quad \text{(A1a)}$$

$$d = G_{SL} ..... - (G_{SS} ..... + G_{LL} .....)/2 \quad \text{(A1b)}$$

Two-locus epistatic effects are calculated by averaging over all possible genetic backgrounds for the four remaining loci. Consider two loci, and let G_{ij} .... be the genotypic value for the genotype i at the first locus and j at the second locus (where i and j can each take the values SS, SL, and LL), averaged over the $3^4 = 81$ possible genetic backgrounds at the four other loci. For instance, G_{SSSL} .... is the average genotypic value over all genotypes homozygous for the Small allele at the first locus and heterozygous at the second locus (i.e., averaged over all genotypes at the remaining four loci). Then the two-locus epistatic effects can be calculated as follows:

$$aa = (G_{SSSS} .... - G_{SSLL} .... - G_{LLSS} .... + G_{LLLL} ....)/4 \quad \text{(A2a)}$$

$$ad = (G_{SSLL} .... + G_{SSSS} .... - 2G_{SSSL} .... - G_{LLSS} .... - G_{LLLL} .... + 2G_{LLSL} ....)/2 \quad \text{(A2b)}$$

$$da = (G_{LLSS} .... + G_{SSSS} .... - 2G_{SLSS} .... - G_{SSLL} .... - G_{LLLL} .... + 2G_{SLLL} ....)/2 \quad \text{(A2c)}$$

$$dd = (G_{SSSS} .... - 2G_{SSSL} .... + G_{SSLL} .... - 2G_{SLSS} .... + 4G_{SLSL} .... - 2G_{SLLL} .... + G_{LLSS} .... - 2G_{LLSL} .... + G_{LLLL} ....)/9 \quad \text{(A2d)}$$

These parameters were calculated for all 15 pairwise combinations of loci.

Quantitative Genetic Parameters in Populations under Selection

In addition to the mean trait value and FA and the allele frequencies at each locus, we computed a number of quantitative genetic parameters in each generation of our population simulations. These were computed from the known genotypes and phenotypes of the offspring generation before each round of selection.

As a measure of additive effect of each locus, we computed the average effect of allelic substitution, α, for each locus (Falconer and Mackay 1996; Lynch and Walsh 1998). First, we calculated the average excess for each allele, $\alpha_S{}^*$ and $\alpha_L{}^*$, as the difference between the mean phenotype of all individuals that carry at least one copy of the respective allele from the mean phenotype in the population. In a random-mating population, as in our simulations, this is equivalent to the additive effects of the alleles, α_S and α_L as they could be obtained from regression analysis (Lynch and Walsh 1998). The average effect of allelic substitution was then obtained as $\alpha = \alpha_L{}^* - \alpha_S{}^*$.

Additive genetic variance was computed by full enumeration. First, the breeding value was computed for every individual by adding the average excess values for both alleles and over all six loci. The genetic variance was then calculated as the average of the squared breeding values (this is possible because the breeding values have a mean of zero). Additive genetic correlations between trait size and FA were computed as the product-moment correlation between the breeding values of the two variables in the population.

To compute dominance variance, we first obtained the dominance deviations δ_{ij} (where i and j can take the values S and L) for the three genotypes at each locus as

$$\delta_{ij} = \mu_{ij} - (\mu + \alpha_i{}^* + \alpha_j{}^*), \quad \text{(A3)}$$

where μ_{ij} is the mean phenotype of the individuals that carry alleles i and j at the locus of interest, and μ is the mean phenotype of the entire population. To compute the dominance variance, the dominance deviations were added over all loci for each individual, and the resulting sums were squared and averaged across the population.