Genome Growth and the Evolution of the Genotype-Phenotype Map

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The evolution of new genes is distinct from evolution through allelic substitution in that new genes bring with them new degrees of freedom for genetic variability. Selection in the evolution of new genes can therefore act to sculpt the dimensions of variability in the genome. This "constructional" selection effect is an evolutionary mechanism, in addition to genetic modification, that can affect the variational properties of the genome and its evolvability. One consequence of selective genome growth is a form of genic selection: genes with large potential for generating new useful genes when duplicated ought to proliferate in the genome, rendering it ever more capable of generating adaptive variants. A second consequence is that alleles of new genes whose creation produced a selective advantage may be more likely to also produce a selective advantage, provided that gene creation and allelic variation have correlated phenotypic effects. A fitness distribution model is analyzed which demonstrates these two effects quantitatively. These are effects that select on the nature of the genotype-phenotype map. New genes that perturb numerous functions under stabilizing selection, i.e. with high pleiotropy, are unlikely to be advantageous. Therefore, genes coming into the genome ought to exhibit low pleiotropy during their creation. If subsequent offspring genes also have low pleiotropy, then genic selection can occur. If subsequent allelic variation also has low pleiotropy, then that too should have a higher chance of not being deleterious. The effects on pleiotropy are illustrated with two model genotype-phenotype maps: Wagner's linear quantitative-genetic model with Gaussian selection, and Kauffman's "NK" adaptive landscape model. Constructional selection is compared with other processes and ideas about the evolution of constraints, evolvability, and the genotype-phenotype map. Empirical phenomena such as dissociability in development, morphological integration, and exon shuffling are discussed in the context of this evolutionary process.

1 Introduction

In this chapter I discuss an evolutionary mechanism whose target is specifically the ability of genomes to generate adaptive variants. It is about the evolution

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of evolvability. The main focus of action for this process is the genotype-phenotype map (Wagner 1984, 1989), i.e. the way genetic variation maps to phenotypic variation. The genotype-phenotype map is the concept underpinning the classical concepts of pleiotropy, polygeny, epistasis, constraints, and gradualness.

The way that genetic variation maps to phenotypic variation is fundamental to whether or not that variation has the possibility of producing adaptive change. Even when strong opportunity exists for new adaptations in an organism, many of its previously evolved functions will remain under stabilizing selection. Adaptation requires variation that is able to move the organismal phenotype toward traits under directional selection without greatly disturbing traits remaining under stabilizing selection. Variation that disturbs existing adaptations as it produces new adaptations — i.e. variation which is *pleiotropic* — will have difficulty producing an overall fitness advantage.

Other aspects of the genotype-phenotype map that affect evolvability include:

- Gradualness: genetic changes with extreme effects are less likely to be advantageous;
- Rugged landscapes: adaptive changes that require the simultaneous altering of several genes are less likely to evolve; and
- Constraints: adaptations for which no genetic variability exists are unable to evolve.

The question of whether the genotype-phenotype map has evolved so as to systematically affect evolvability has been dealt with in a variety of ways in the literature. Approaches include the following:

The genome as fluid: Evolvability is not limited; genetic variation exists within populations for any trait one wishes to select on.

The internalist view: The degree of evolvability is a byproduct of the physics of development. It is fortunate that physics permitted evolvability.

Lineage selection: Different developmental systems may have different evolvabilities; those which happen to have high evolvability will proliferate as species lineages.

Genetic modification: Selection for adaptedness happens to systematically produce high evolvability.

This paper adds an additional hypothesis to this list:

Constructional selection: Selection during the origin of genes provides a filter on the construction of the genotype-phenotype map that naturally produces evolvability.

The internalist viewpoint is what this paper will take issue with most. The internalist viewpoint holds that the variational properties of the genotype-phenotype map are the result of the physics of development (Goodwin 1989). The process of morphogenesis is proposed as a complex dynamical system toward which genes contribute, but which has internal macroscopic properties that determine what kinds of phenotypic variability exist.

One can ask, however, whether morphogenetic dynamics could have been shaped by evolutionary forces that systematically affect the nature of developmental constraints, or the smoothness of the adaptive landscape, or its evolvability. Here I discuss an evolutionary mechanism by which selection can come to act indirectly on evolutionary potential, as a consequence of how genes come into being in the first place.

The main idea, in a nutshell, is this: the genes that stably exist in a genome share the common feature that, when they were created, they produced a selective advantage for the organism. But when a new gene is created, it not only produces its current phenotypic effect, but carries with it a new "neighborhood" in "sequence space" — the kinds of variants that it can in turn give rise to. The phenotypic character of this neighborhood depends on the gene's mode of action. Different modes of gene action can be expected to have different overall likelihood of producing adaptive variants. The fact that a gene's existence is predicated on it having originally produced a selective advantage means that the accumulation of new genes in the genome should be biased toward modes of action whose variants are more likely to be fruitful in adaptation.

Since there is a diversity of modes of gene action, the question remains as to why there are the kinds there are, in the frequencies they are found, within the genomes of organisms. This chapter presents a theory about the statistical properties of genotype-phenotype maps, and how these statistics would be expected to change in the course of the evolutionary construction of the genome toward ways that facilitate the generation of adaptive variants.

There are two basic aspects to the idea of a genotype-phenotype map. One can think of the genotype as a "representation" or description of the phenotype. Representation has two aspects: *generative* and *variational*. The generative aspect of a representation is how the representation is actually used to produce the object, which in genetics would be the process of gene expression and its integration in development. It is not the mechanisms of how this map is accomplished that is relevant to evolvability; rather, what matters is the variational aspect of a representation — how changes in the representation map to changes in the object. Variational aspects can be described by their statistical properties without having to deal with the generative mechanisms. The principal variational aspect I will be concerned with is pleiotropy — the constellation of phenotypic effects from single mutations.

1.1 Bonner's Low Pleiotropy Principle

Bonner (1974) has articulated a basic "design principle" for the genotype-phenotype map necessary to allow the generation of adaptive variants through random genetic variation, a principle of low pleiotropy:

> We presume that it is of a distinct advantage to keep a number of the units of gene action of the organism quite independent of one another. The reason for this seems straightforward: mutations that affect a number of construction units are more likely to be lethal than those that

> affect only one. Or to put it another way, the fewer the interconnections of gene action (the less the pleiotropy), the greater the chances of its being a viable mutant. A viable mutant may be one that appears late in development, such as the pigmentation of hair, eyes, or feathers, or one that acts in a small developmental unit that is independent of the others. (1974, p. 61)

Lewontin (1978) proposed the low pleiotropy principle in a somewhat different manner, as a principle of "quasi-independence", i.e. that there must be "a great variety of alternative paths by which a given characteristic may change, so that some of them will allow selection to act on the characteristic without altering other characteristics of the organism in a countervailing fashion: pleiotropic and allometric relations must be changeable."

However, this design principle suffers from the "for the good of the species" problem. Even though a property might be "good for the species", it can only evolve if organisms bearing it (or "replicators" to be more general (Brandon 1990)) have higher fitness. Although it would be a marvelous design for the organism to have a genome organized for its future adaptive potential, this future advantage does not give an organism the present advantage it needs in order to pass on such a trait.

2 Constructional Selection

All variational aspects of the genotype-phenotype map face the "good of the species" problem, because variation is not the phenotype of an organism, but a property of genetic transmission between organisms. How, therefore, can organismal selection get a "handle" on the processes that produce variation? The general answer to this question is that there must be correlations between variational properties and properties affecting organismal fitness. These correlations can come about through diverse means.

In the case of variational properties like recombination and mutation rates, correlations can be induced by the evolutionary dynamics of modifier genes — genes that control recombination, mutation, and so forth. Genes modifying recombination rates, for example, can evolve linkage associations to genes under selection whose transmission they affect. In this case, it is the modifier gene that provides natural selection with the "handle" to change recombination rates (Liberman and Feldman 1986, Altenberg and Feldman 1987).

Modifier genes are rather specialized mechanisms. But here I consider a means by which selection can gain a handle on the variational properties of any gene, through the selective forces operating during the origin of the gene. All genes face the problem of selection during their creation, and those genes that produce a selective disadvantage never become stably incorporated in the genome. Therefore, existing genes share the common history of having once produced a selective advantage to the organism. But new genes bring with them new degrees of freedom for variability in the genome. These new degrees of freedom are of two types:

Type I: new genes serve as new templates for further genome growth, and
Type II: new genes afford new sites at which allelic variation can occur.

The phenotypic effects of either of these new degrees of freedom depend on the physical nature of the gene's action. And the gene's mechanisms of action is unlikely to change radically between its creation and subsequent gene duplications and allelic variations. Therefore it is reasonable to expect a correlation to exist between the phenotypic or fitness effects of a newly created gene and subsequent duplications and allelic changes. This then is a means by which variational properties of the genome can become correlated with organismal selection.

Therefore, without the postulation of additional modifier genes, selection during the creation of new degrees of freedom for genetic variability can gain a handle on the quality of those degrees of freedom. The strength of this handle depends on the strength of the correlations. When referring to this process, I will summarize it with the term "constructional selection", since it is tied to the construction of new genes (Altenberg 1985).

2.1 Riedl's Theory

Riedl's (1977) theory for the evolution of "genome systemization" is the main earlier example of a constructional selection theory for the genotype-phenotype map. He considers the situation where functional interactions arise in the organism that require the coordinated change of several phenotypic characters in order to produce adaptive variants. When this would require simultaneous mutations at several genes, he argues that the evolution of a new gene that produces the needed coordinated variability — a "superimposed genetic unit" — is a far more likely possibility. Thus Riedl is proposing that the genotype-phenotype map can evolve in directions that facilitate adaptation through selective genome growth.

2.2 Fine Points

It is important at this point to be clear that this is not an argument that most adaptive evolution happens through the origin of new genes, as opposed to allelic substitution. Rather, I am proposing that the events surrounding the creation of new genes may play a special role in the evolution of the genotype-phenotype map because of their distinct property of adding new degrees of freedom to the genome.

Also, it should be understood that "new genes" can refer equally to new parts of genes or new clusters of genes, i.e. new sections of DNA sequence that are of functional use to the organism. Therefore, the arguments here apply to such elements as exons, promoters, enhancers, operators, other regulatory elements, etc..

Throughout this chapter, pleiotropy must be understood to refer not to multiple effects on arbitrary "characters" of the organism, since these are artifacts of measurement and description, but to organismal functions that are components of adaptation, what Nemeschkal *et al.* (1992) refer to as a "unit of characters

working together to accomplish a common biological role". Moreover, in the case of new genes, the definition of "multiple" effects that is germane as a definition of pleiotropy is when the gene not only produces variability for functions under directional selection, but also disturbs functions under stabilizing selection. "Low pleiotropy" will refer to genes that affect mainly functions under directional selection and leave functions under stabilizing selection unaffected.

2.3 Pleiotropy and Constructional Selection

Let us examine Bonner's low pleiotropy principle in the context of the genome growth process. New genes which have fewer pleiotropic effects when added to the genome, whose action causes the phenotype to change mainly in dimensions that are under directional selection, stand a better chance, by Bonner's principle, of providing a selective advantage. This is would hold even if that chance is still slight. Genes which disturb many adapted functions of the phenotype are unlikely to be advantageous, and thus would not be incorporated in the genome.

Therefore, selection can filter the pleiotropy of genes as they are added to the genome. If there is any correlation between the pleiotropic effects during the gene's addition and the pleiotropy of subsequent additions or allelic changes in the gene, then the genome shall have expanded its degrees of freedom in directions with lower pleiotropy.

The effects of constructional selection on the two forms of genetic variation, Type I and II above, are distinct, so each is taken up in turn.

2.4 Type I Effect: The Genome as Population.

If there are correlations between the phenotypic effects of duplicated genes and the effects of their subsequent duplications during macroevolutionary time scales, then a novel form of intragenomic "genic" selection process becomes possible. This selection process is based on looking at the genome as a "population" of genes, as in the case of genic selection in the evolution of transposable elements. The idea that transposable elements are genetic parasites propagating within the genome (Cavalier-Smith 1977, Doolittle and Sapienza 1980, Orgel and Crick 1980) lead to the idea that the genome could be considered a population of genes, within which a new level of selection can operate when certain sequences can proliferate *within* the genome. Such "genic" selection is usually associated with transposable elements, whose activity is generally in conflict with organismal selection. The type I effect, however, is a form of genic selection in harmony with organismal selection, which, moreover, has organismal selection as a *subprocess*.

Where do new genes come from? Although there is a certain amount of *de novo* synthesis of DNA in the genome, most genes originate from template based duplication of existing sequences. And while the vast majority of gene duplications may go to extinction, the genes currently functioning in an organism will possess an unbroken backward genealogy to earlier, ancestral genes (complicated perhaps by the occasional reactivation or insertion of pseudogene sequences). So

there exists an "intra-genomic phylogeny", which is actually beginning to be taken as an object of study as the accumulation of DNA sequences allows the construction of "gene-trees" (Dorit and Gilbert 1990, Dorit *et al.* 1991, Strong and Gutman 1992, Burt and Paton 1992, Klenova, *et al.* 1992, Streydio *et al.* 1992, Haefliger *et al.* 1989).

If one picks any functioning gene in the genome, what would a typical story for its origin be? One could generally list:

1. Sequence duplication;
2. Fixation in the population, through selection or drift;
3. Maintenance of function by selection;
4. Sequence evolution under mutation and selection.

Differences in gene properties that systematically bias the chances of the above events can produce a Darwinian process on the level of genome-as-population. Darwinian process have three basic elements: viability, fecundity, and heritability. If there exist properties which show heritable variation in viability or fecundity, those properties can evolve over time. Viability, fecundity, and heritability each have their analogs on the level of genome-as-population:

Viability:

The viability of a genetic sequence is simply its survival in the genome. This will depend on whether selection establishes it in the population, and maintains it against mutational degradation or replacement by other genes. This in turn depends on[1]:

1. there being adaptive opportunity for properties of the sequence;
2. the sequence having functional properties which are not disrupted by new functional contexts; and
3. the sequence having properties that allow its duplication without disrupting existing functions of genes with which it interacts.

Fecundity:

The fecundity of a genetic sequence is the rate at which copies of it appear in the genome. This depends on:

1. the rate of illegitimate recombination events involving that sequence; and
2. whether the sequence codes for its own duplicative transposition.

[1] Note added in revision: Originally these three items were placed under it Fecundity, below, because I conceived of taking the "census" of offspring genes after they had been established as a functional genes. Censusing immediately after gene duplication produces a cleaner distinction between intragenomic viability and fecundity, and is adopted here. The problem of when to census also arises in standard population genetics when trying to distinguish viability fitness components from fecundity fitness components.

Heritability:

Heritability here refers to ancestral and offspring genes having correlated properties, and depends on:

1. Conservation of the property of a gene over the time scale on which gene duplications occur; and
2. Carry-over of the property from ancestral to offspring genes.

In each case above, one could just as well substitute "genetic element" for "gene", since the principles apply equally well to exons, promoters, regulatory sequences, and so forth.

If there are systematic differences between sequences in the likelihood that duplications of them give rise to useful new genes (viability in the genome), and these different likelihoods are conserved between gene origins, and carried from ancestral to offspring genes (heritability), then the genome will become populated with genes that are better able to give rise to other genes. The type I, or "genic selection" effect of constructional selection, therefore, is to increase the genome's ability to evolve new genes. This is an effect on the variational properties of the genome.

The genome-as-population analogs of viability, fecundity and heritability in the type I effect can be contrasted with these analogs in the case of transposable elements. For such "selfish" DNA, *viability* as genes is low: on a macroevolutionary time scale, individual copies of transposons are transient, since they exist either as transient allelic polymorphisms or, if they ever go to fixation, are deleted or silenced rapidly because as alleles they are usually neutral or deleterious, and genetically unstable. The *fecundity* of transposons in the genome, however, is unsurpassed, and overcomes their sub-viability in the genome as individual copies. Furthermore, their heritability as genes is extremely high, since offspring genes mostly have the same ability to transpose. Thus the type I effect of constructional selection and "selfish DNA" are two kinds of intragenomic genic selection, and are in a sense opposite points within a continuum defined by the genome-as-population analogs of the Darwinian elements, viability, fecundity, and heritability. [2]

2.5 Type II Effect: Correlated Allelic Variation.

If there are correlations between the phenotypic effects of duplicated genes and the effects of their subsequent allelic variants, then selection during the creation of new genes can come to affect the nature of allelic variants produced in the genome. This is what I call the type II or correlated allelic variation effect of selective genome growth. In particular, if a new gene was advantageous because

[2] Note added in revision: Transposable elements and normal genes represent the intragenomic analogues of "r-" versus "K-selection": transposable elements have high "r", reproducing rapidly but with low genomic viability; conversely, functioning organismal genes have low "r", being duplicated infrequently, but these duplications may come to exhibit a "K" strategy of higher intragenomic viability—i.e. a higher chance of being established and preserved by organismal selection.

it moved the phenotype along dimensions under directional selection, then subsequent alleles of that new gene may also be likely to be advantageous if they vary the phenotype along the same "lines" as occurred during the gene's original incorporation in the genome. By "likely", I mean relative to the effects of allelic variation at all the genes that were generated by duplication processes, but never fixed in the population and maintained by selection. If the pleiotropy of a gene is a relatively fixed result of its mode of action, then there will be a correlation between the phenotypic effects of the gene's origin and its subsequent allelic variation. If low pleiotropy helped the gene become established in the first place, then the subsequent low pleiotropy of its allelic variants would enhance their likelihood of being adaptive rather than universally deleterious.

An important case of the correlated allelic variation effect is "function splitting", where a gene that has been selected as a compromise for carrying out several organismal functions is duplicated and the separate copies can evolve to specialize in some subset of functions. An example is the duplication of the hemoglobin gene and its specialization for fetal or postnatal oxygen transport conditions. In this case, the duplication causes changes in the genotype-phenotype maps of both resulting genes, with the net result of lowering the pleiotropy of allelic variation at these genes, and better optimization of the adaptive functions. This is an area which has already received a good deal of empirical and theoretical study (Ohta 1991, 1988, Kappen *et al.* 1989, Li 1985).

The type II effect is entirely dependent on there being correlations between the phenotypic effects of a new gene and the effects of allelic variation at that locus. For genes of recent origin, correlations would be expected. However, over time these correlations would be expected to weaken due to several factors. First, substantial sequence changes may occur as the gene diverges in function from that of its ancestral state. Second, whatever novel advantage the gene may have offered when it first arose will tend to change from being a "luxury" to being a necessity, as other functions evolve conditioned on the current state of that gene. This is what Riedl (1977) calls "burden" (and what Wimsatt and Schank call "generative entrenchment" (Schank and Wimsatt 1987, Wimsatt and Schank 1988). Histones, polymerases, snRPs, etc, are extreme examples of burdened genes, since effectively all characters of the organism depend on them; their mutations are of necessity highly pleiotropic, and they are extremely well conserved. So over macroevolutionary time scales, the correlated allelic variation effect may become "stale" once a gene is in place. The low pleiotropy might be kept "fresh", however, if changing selection or polymorphism produces a history of variation in the gene to which other genes coadapt.

2.6 An Overall Picture of Genome Growth.

These considerations lead to the following picture of the intra-genomic phylogeny: There should be a static core of genes which have ceased to give rise to new genes in the genome; these may be extremely ancient and functionally burdened, or so highly specialized as to have little adaptive potential for duplications. Once genes enter this core, they should tend to remain there (though

they may continue sequence evolution). There should in addition be a "growth front" in the genome consisting of genes that are prolific in generating offspring genes. The growth front would gradually lose genes to the static core once they were created, but would be renewed by the influx of newly created genes, which would be the most likely to give rise to the next set of new genes. On occasion, static genes would be revived into the growth front by new adaptive opportunities conferred by changes in organismal selection. In addition, there would be the various "exceptional" families of genes, including transposable elements, highly repetitive genes selected for quantity production, "junk" and structural DNA, and so forth.

2.7 Constraints and Latent Directional Selection.

An examination of the situations discussed in the literature in which the genotype-phenotype map constrains evolution shows them to be of two basic kinds: kinetic and range constraints. A range constraint is simply where no genetic variation exists for phenotype or specific combination of phenotypic changes. Kinetic constraints emerge from the population genetic dynamics when the probability of creating given phenotypic variants is vanishingly low. A softer version of this is a kinetic bias, in which the most probable variant that responds to a selective pressure has specific phenotypic forms. The problem of adaptation on "rugged fitness landscapes" (Kauffman 1989a) is an example of kinetic constraints, in that what keeps a population at a local fitness peak is the improbability of generating fitter variants (in fact it is transmission probabilities that define what a neighborhood is in the sequence space). This includes the situation considered by Riedl (1977), where mutations are needed at several loci to produce a given phenotype.

The general consequence of either range or kinetic constraints is that to varying extents, organisms will be suboptimally adapted. There may be phenotypes that would be more adapted if only the genome could produce them. The population may have reached a mutation-selection balance, in which new variants are all deleterious, and so appear to be at an adaptive peak, when the lack of fitter variants is due to kinetic or range constraints. In such cases one could say that there exists a "latent" directional selection, which would become visible if genetic variation existed in this direction.

Riedl's idea is that much of the adaptive opportunity for the evolution of new genes may come from latent directional selection. But constructional selection effects would apply to conditions of normal directional selection as well. There would be adaptive opportunity for any new gene whose effects on the phenotype were in the direction of the current directional selection on the organism. Therefore, genes may to some extent reflect the historical sequence of directional selection experienced by the organism's lineage. Even ancient and highly functionally burdened genes may reveal the functions they conferred in their origin. For example, homeotic mutations which change insect segment identity are universally deleterious. But if an alteration of segment identity was what

the gene did when it was created (and thereby presupposed to have been selectively advantageous), then the gene's current function may be a reflection of the directional selection that existed at the time of its origin.

2.8 Models Illustrating Constructional Selection

To give explicit mathematical form to the ideas sketched so far about genome growth, several models will be developed. The first is a simple model showing both type I and II effects, which uses probability distributions of fitness effects for gene additions and subsequent allelic variation. The analysis shows the exponential quality of the genic selection effect, and the dependence on correlations in the correlated allelic variation effect. The second and third models are further illustrations of the correlated allelic variation effect, using as concrete examples of genotype-phenotype map functions:

1. Wagner's linear quantitative-genetic model with Gaussian stabilizing selection (Wagner 1989); and
2. Kauffman's (1989a) epistatic "NK" adaptive landscape model.

The linear model illustrates latent directional selection arising from constraints on the range of phenotypic variation produced by the genotype, and exhibits selection for new genes that overcome these range constraints. The NK model illustrates latent directional selection arising from kinetic constraints due to the ruggedness of the adaptive landscape, and exhibits selection for genes that overcome the kinetic constraints and produce smoother adaptive landscapes.

The Discussion follows, with an overview of the results, an examination of relevant empirical phenomena, and a discussion of the relation of constructional selection to current thinking about the evolution of evolvability.

3 A Fitness Distribution Model

The effects of constructional selection can be described directly in terms of the fitness distributions of new mutations, without having to specify the genotype-phenotype maps that give rise to these distributions. In the case of the genic selection effect, the mutation is a gene duplication; in the case of the correlated allelic variation effect, the mutation is an allelic change.

In this model, a new gene is randomly created from the existing genes in the genome. Selection then determines whether the gene is kept in the genome. The model considers what happens when either allelic mutations or subsequent gene duplications occur. The genes in the population come in different types that determine the fitness distribution of their mutations. The main elements in the model are as follows. Let:

$\mathcal{G}$ be the space of different types of genetic sequences;
p_i be the probability that a newly created gene is of type $i \in \mathcal{G}$;
w be the fitness of the genome with the new gene, relative to its value before the addition;

$g_i(w)$ be the probability that a new gene of type i has relative fitness effect w;
x_i be the probability that a new gene of type i is kept in the genome by selection.

The probability density $g_i(w)$ would be the result of the phenotypic properties of the gene, as described under *Viability* in Sect. 2.4, including its pleiotropy, modularity, and adaptive opportunity. A concrete illustration is developed in Sect. 5, on Kauffman's NK adaptive landscapes.

In a simple-minded approach, a gene would be kept by selection if it increased fitness, i.e. if $w > 1$. Then the probability that the gene is kept is

$$x_i = \int_{w=1}^{\infty} g_i(w)\, dw \ .$$

But in finite populations, or in any population dynamics where there is a chance that a gene will not be passed down to any offspring, even a gene increasing fitness can sometimes be lost from the population. The probability that a new gene is successfully incorporated in the genome will be some increasing function $\phi(w)$ of its fitness w. Classical results using branching process models or diffusion approximations give a success probability of 0 if $w < 1$, and $\phi(w) \approx 2(w-1)$ for $w \approx 1$ (Haldane 1927, Crow and Kimura 1970). So a more general formula for the likelihood that a new gene of type i is fixed is:

$$x_i = \int_{0}^{\infty} \phi(w)\, g_i(w)\, dw \ . \tag{1}$$

The fixation probability over all random newly created genes is:

$$\overline{x} = \sum_{i \in \mathcal{G}} x_i\, p_i \ .$$

With these definitions, results for both the genic selection effect and the correlated allelic variation effect will be derived.

3.1 The Correlated Allelic Variation Effect

Here we will see how selection on the creation of new genes can cause subsequent allelic variation of the genes to be more likely to be adaptive. We will look at the fitness distributions of alleles from all new genes and from only those genes that selection stably incorporates into the genome.

Suppose that a newly created gene of type i gives rise to allelic variants. Let the allelic fitnesses, w', be distributed with probability density $f_i(w')$. No assumptions need to be made about this density, so it would certainly include the biologically plausible case in which most of the alleles are deleterious. For a gene or type i, we see that the proportion

$$F_i(w) = \int_{w}^{\infty} f_i(y)\, dy \ ,$$

of its alleles are fitter than w.

Result 1 (Correlated allelic variation)
Let

$\overline{F}(w)$ *be the proportion of new alleles of randomly created genes that are fitter than* w*, and*
$F^*(w)$ *be the proportion of new alleles of stably incorporated genes that are fitter than* w*.*

Then

$$F^*(w) = \overline{F}(w) + \mathrm{Cov}[F_i(w),\ x_i/\overline{x}] \ . \tag{2}$$

Proof. The proportion of alleles that are fitter than w, among randomly created gene, is

$$\overline{F}(w) = \sum_{i \in \mathcal{G}} F_i(w)\, p_i \ ,$$

while among genes that are stably incorporated in the genome it is

$$\begin{aligned} F^*(w) &= \Pr[w' > w \mid \text{the gene was fixed}] \\ &= \Pr[w' > w, \text{ and the gene was fixed}] \ / \ \Pr[\text{the gene was fixed}] \\ &= \sum_{i \in \mathcal{G}} F_i(w)\, x_i\, p_i \ / \ \overline{x} = \overline{F}(w) + \mathrm{Cov}[F(w_i),\ x_i/\overline{x}] \ . \end{aligned}$$

■

If there is a positive correlation between the fixation probability

$$x_i = \int_0^\infty \phi(w)\, g_i(w)\, dw$$

of a new gene, and the fitness distribution

$$F_i(w) = \int_w^\infty f_i(y)\, dy$$

of its alleles, then $F^*(w)$ is greater than $\overline{F}(w)$. Similarity between the functions $g_i(w)$ and $f_i(w)$ would produce a positive covariance. The biological foundation for a positive covariance would include:

1. there continuing to be adaptive opportunity for variation in the phenotype controlled by the gene, and
2. the same suite of phenotypic characters being affected by the alleles of the gene as were affected during the gene's origin.

With these plausible and general provisions, we see how selection on new genes can also select on the fitness distributions of the alleles that these genes generate.

3.2 The Genic Selection Effect

Now we will see how selection on new genes can increase the chance that new genes are adaptive when created. We will examine how genes with a higher chance of producing adaptive variants tend to proliferate as the genome grows, as reflected in the evolution of p_i. The model I am considering is this: genes are randomly picked from the genome and copied. Their fitness effect determines whether they are stably incorporated in the genome. If they are, then the pool of genes subject to duplication is increased by one, and the process repeated. In this way genes of different types come to proliferate at different rates within the genome.

Consider the process of sequence duplication that is the starting point for the history of every gene (or part of a gene). One can think of the rate that a gene gives rise to new, successfully incorporated genes as its "constructional fitness". This will be the product of

1. the rate that copies of the gene are produced (intragenomic fecundity), and
2. the likelihood that they are fixed in the genome by having provided a selective advantage to the organism (intragenomic viability).

While genetic elements such as transposons or highly repetitive sequences may proliferate because of factor 1, here I wish consider only factor 2, and assume no systematic differences among sequences in the rate that gene copies are produced.

Perfect Transmission of the Gene's Type. Suppose for now that copies of genes of type i are also of type i. Because the gene's type is transmitted from a gene to its offspring genes, this provides a correlation between the fitness effects of a new gene and its subsequent duplications. As in equation (1), a new gene of type i will have probability x_i of fixation due to its yielding a selective advantage.

Let

$n_i(t)$ be the number of genes in the genome of type i at time t,

$N(t) = \sum_{i \in \mathcal{G}} n_i(t)$ be total number of genes in the genome at time t, so that the frequency of genes of type i is $p_i(t) = n_i(t) \,/\, N(t)$, and

α be the rate each gene is duplicated per unit time.

One then obtains this differential equation for the change in the composition of the genome (approximating the number of genes with a continuum), using the fixation probability, x_i, for new genes of type i:

$$\frac{d}{dt} n_i(t) = \alpha \, x_i \, n_i(t) \ ,$$

which has solution:

$$n_i(t) = e^{\alpha x_i t} \, n_i(0) \ .$$

The ratio between the frequencies in the genome of sequences with different constructional fitnesses grows exponentially with the degree of difference between them:

$$\frac{n_i(t)}{n_j(t)} = e^{(x_i - x_j)\alpha t} \, \frac{n_i(0)}{n_j(0)} \ .$$

Result 2 (Fisher's Theorem applied to genome growth)
The average constructional fitness of the genome,

$$\overline{x}(t) = \sum_{i \in \mathcal{G}} x_i \, p_i(t) \ ,$$

which is the portion of new duplicated genes that go to fixation, increases at rate

$$\frac{d}{dt}\overline{x}(t) = \alpha \, \mathrm{Var}(x) > 0 \ .$$

Proof.

$$\begin{aligned}
\frac{d}{dt}\overline{x}(t) &= \sum_{i \in \mathcal{G}} x_i \frac{d}{dt} p_i(t) \\
&= \sum_{i \in \mathcal{G}} x_i [\frac{d}{dt} n_i(t) \,/\, N(t) - n_i(t) \frac{d}{dt} N(t) \,/\, N(t)^2] \\
&= \frac{\alpha}{N(t)} \sum_{i \in \mathcal{G}} x_i^2 \, n_i(t) - \frac{\alpha}{N(t)^2} \left(\sum_{i \in \mathcal{G}} x_i \, n_i(t) \right)^2 \\
&= \alpha \left[\sum_{i \in \mathcal{G}} x_i^2 p_i(t) - \overline{x}(t)^2 \right] \\
&= \alpha \, \mathrm{Var}(x) > 0 \ .
\end{aligned}$$

■

This result is Fisher's fundamental theorem of Natural Selection (Fisher 1930), but here, what is evolving is the probability of gene duplications giving rise to new useful genes.

Imperfect Transmission of the Gene's Type. The model can be extended to less-than-perfect heritability of constructional fitness by defining a transmission function, $T(i \leftarrow j)$, which is the probability that a gene of type j gives rise to a copy of type i (Slatkin 1970, Altenberg and Feldman 1987). It satisfies conditions

$$\sum_{i \in \mathcal{G}} T(i \leftarrow j) = 1 \text{ for all } j \in \mathcal{G}, \text{ and } T(i \leftarrow j) \geq 0 \text{ for all } i, j \in \mathcal{G} \ .$$

Here, the fraction of the new genes that are of type i is

$$p_i(t) = \sum_{i,j \in \mathcal{G}} T(i \leftarrow j)\, n_j(t) \,/\, N(t) \ .$$

The dynamics now become:

$$\frac{d}{dt} n_i(t) = \alpha\, x_i \sum_{j \in \mathcal{G}} T(i \leftarrow j)\, n_j(t) \ .$$

Price's Covariance and Selection theorem (Price 1970, 1972) emerges when we consider selection in the presence of arbitrary transmission:

Result 3 (Price's Theorem applied to genome growth)
For a gene of type j, let

$$\xi_j = \sum_{i \in \mathcal{G}} x_i\, T(i \leftarrow j) \ .$$

be the fraction of its duplicate offspring genes that are stably incorporated in the genome. Then rate of change in the average constructional fitness of the genome evaluates to

$$\frac{d}{dt} \overline{x}(t) = \alpha \left\{ \mathrm{Cov}(\xi, x) + [\overline{\xi}(t) - \overline{x}(t)]\, \overline{x}(t) \right\} \ ,$$

where

$$\overline{\xi}(t) = \sum_{i \in \mathcal{G}} \xi_i\, p_i(t), \quad \textit{and} \quad \mathrm{Cov}(\xi, x) = \sum_{i \in \mathcal{G}} \xi_i\, x_i\, p_i(t) - \overline{\xi}(t)\, \overline{x}(t) \ .$$

Proof. The portion of gene duplications that go to fixation is

$$\overline{x}(t) = \sum_{i \in \mathcal{G}} x_i\, p_i(t) = \sum_{i \in \mathcal{G}} x_i \sum_{j \in \mathcal{G}} T(i \leftarrow j)\, n_j(t) \,/\, N(t) = \sum_{j \in \mathcal{G}} \xi_j n_j(t)/N(t) \ .$$

This changes at the rate:

$$\begin{aligned}
\frac{d}{dt} \overline{x}(t) &= \sum_{i,j \in \mathcal{G}} x_i\, T(i \leftarrow j) \left[\frac{dn_j(t)}{dt}/N(t) - \frac{dN(t)}{dt} n_j(t)/N(t)^2 \right] \\
&= \alpha \sum_{i,j \in \mathcal{G}} x_i\, T(i \leftarrow j) \left[x_j \sum_{k \in \mathcal{G}} T(j \leftarrow k)\, n_k(t) \,/\, N(t) \right. \\
&\qquad \left. - n_j(t) \sum_{k,h \in \mathcal{G}} x_k\, T(k \leftarrow h)\, n_h(t) \,/\, N(t)^2 \right] \\
&= \alpha \sum_{j \in \mathcal{G}} \xi_j \left[x_j \sum_{k \in \mathcal{G}} T(j \leftarrow k)\, n_k(t) \,/\, N(t) \right. \\
&\qquad \left. - n_j(t) \sum_{h \in \mathcal{G}} \xi_h\, n_h(t) \,/\, N(t)^2 \right]
\end{aligned}$$

$$= \alpha \sum_{j \in \mathcal{G}} \xi_j x_j p_j(t) - \alpha \left[\sum_{j \in \mathcal{G}} x_j p_j(t) \right]^2$$
$$= \alpha \left\{ \mathrm{Cov}(\xi, x) + [\overline{\xi}(t) - \overline{x}(t)]\, \overline{x}(t) \right\} \ .$$

■

The covariance term is between a gene's probability of fixation and its offspring genes' average probability of fixation. Note that the frequencies used in the covariance are the frequencies of different types among gene duplications, not the current genes in the genome.

A positive correlation between ξ_i and x_i is to be expected if a gene and its offspring genes affect the same sort of phenotypic characters, and the adaptive opportunity that existed for these characters still exists. Genes (or gene parts, e.g. exons) that code for generally useful products, such as promoters, transmembrane linkers, catalytic sites, developmental controls, etc., would have such continuing adaptive opportunity, and they would contribute to making $\mathrm{Cov}(\xi, x) > 0$.

The term $\overline{\xi}(t) - \overline{x}(t)$ is the net bias in the transmission of constructional fitness between a gene and its offspring genes. A conservative assumption is that the transmission bias is negative — i.e. the chance that gene duplications are adaptive is less for a gene's grand-offspring than it is for the gene's offspring. This is a reasonable assumption since duplications of a gene (or gene part) would diverge to various extents from the ancestral gene's effects, selection may change, or the adaptive opportunity for new copies of the gene may get saturated.

But even with a negative transmission bias, the average constructional fitness, $\overline{x}(t)$, increases as long as

$$\overline{\xi}(t) - \overline{x}(t) > -\mathrm{Cov}(\xi, x) \,/\, \overline{x}(t) \ . \tag{3}$$

As an illustrative example, we can set $\xi_i = \beta x_i$ with $\beta < 1$, a downward transmission bias. Still, $\overline{x}(t)$ increases as long as

$$\beta > \frac{1}{1 + \mathrm{Var}(x_i / \overline{x}(t))} \ . \tag{4}$$

Evaluation of (4) requires evaluating the magnitude of $\mathrm{Var}(x_i/\overline{x}(t))$, which depends on the distribution of constructional fitness values in the genome. Let $q(x)$ be the portion of gene duplications with constructional fitness x. The conditions for (4) under a variety of distributions are:

- A uniform distribution, $q(x) = 1$: $\overline{x}$ increases if $\beta > 3/4$;
- An exponential distribution, $q(x) = \nu\, e^{-\lambda x}$ (ν is the normalizer): for large λ, $\overline{x}$ increases if $\beta > 1/2$;
- A Gaussian initial distribution, $q(x) = \nu\, e^{-\lambda x^2}$: for large λ, $\overline{x}$ increases if $\beta > 2/\pi$;

– A Gamma distribution,

$$q(x) = \Gamma(x;\gamma,\lambda) = \begin{cases} \lambda^{\gamma}\, x^{\gamma-1}\, e^{-\lambda x}/\Gamma(\gamma), & x > 0, \\ 0, & x \le 0, \end{cases} \quad :$$

for large λ, $\overline{x}$ increases if $\beta > \frac{\gamma}{\gamma+1}$. Since one can choose $\gamma > 0$ close to 0, distributions can be found for any arbitrarily small β in which the average constructional fitness of the genome grows.

Thus, even for arbitrarily strong downward transmission bias, where the probability of a gene giving rise to a useful offspring gene decreases by a factor β each gene duplication, the average probability in the genome that a gene duplication produces a selective advantage may still increase in time, depending on the initial distribution of these probabilities in the genome.

As $n_i(t)$ evolves, both $\mathrm{Cov}(\xi, x)$ and the net transmission bias will change. Under a wide variety of well-behaved transmission functions, where the net transmission bias initially satisfies equation (3), the distribution of constructional fitness values will shift upward until the net bias balances the covariance or the covariance is exhausted.

Results 1 and 3 are extensions of a line of theorems in quantitative genetics based on the covariance of different traits with fitness, including Fisher's fundamental theorem, Robertson's "secondary theorem of Natural Selection" (Robertson 1966), and a result by Price (1970) on gene frequency change, which were elaborated upon by Crow and Nagylaki (1976) and Lande and Arnold (1983). Price's theorem has been applied in a number of different contexts in evolutionary genetics, including kin selection (Grafen 1985, Taylor 1988), group selection (Wade 1985), the evolution of mating systems (Uyenoyama 1988), and quantitative genetics (Frank and Slatkin 1990). I have applied it to performance analysis of genetic algorithms in Altenberg (1994, 1995).

4 Wagner's Linear Quantitative-Genetic Model with Gaussian Selection

Wagner (1984, 1989) has investigated evolutionary aspects of the genotype-phenotype map through analysis of linear maps combined with a number of different fitness surfaces, including "corridor" and Gaussian fitness functions. In this section I investigate the correlated allelic variation effect of genome growth using a variant of Wagner's (1989) model of "constrained pleiotropy". The model here is a multilayered linear map from the genotype to the organismal phenotype, and from the phenotype to the adaptive functions they carry out. Figure 1 illustrates this model.

What I want to capture with this model is the following idea: genes don't "know" *a priori* what they are doing, what functions they are carrying out; i.e. there is "universal pleiotropy". Pleiotropic constraints may limit the genotype's ability to optimize simultaneously all the functions it controls, so that the best phenotype achievable, given the genetic variability available, may be a

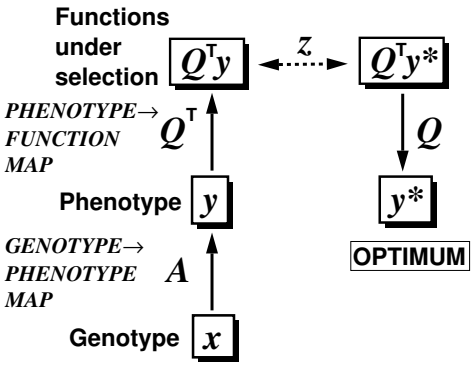

Fig. 1. Wagner's linear model of the genotype-phenotype map with a Gaussian fitness function on the departure, $\boldsymbol{z}$, from optimality.

compromise between tradeoffs that represents a departure from the global selective optimum. The genotype may appear to be at a selective peak, but if new dimensions of genetic variability were opened up, this peak would be revealed to be on the slope of a larger selective peak.

Therefore, at these constrained peaks there exists a "latent" directional selection to which the population could respond if the proper dimension of genetic variation existed. In such situations, events which makes the proper variation possible can be major factors in evolution. Genetic changes that alter the nature of the pleiotropic constraints can therefore come under selection. In this model, I will show how, when there exists variability in the pleiotropic effects of genes coming into existence, genes which are most aligned with the latent directional selection will have the best chance of being incorporated into the genome, and the genomes that result will be able to simultaneously optimize all the adaptive functions much better than would be expected from the underlying distribution of pleiotropic effects. Moreover, the pattern of phenotypic effects of each gene will tend to reflect the directional selection that existed when the gene came into being. The phenotypic variability present in the genomes will therefore indicate the history of directional selection that the genomes experienced during their evolutionary construction.

4.1 The Adaptive Landscape

The organismal phenotype is defined as a k-element long vector, $\boldsymbol{y} \in \Re^k$. The organism carries out f different adaptive functions. The optimal organismal phenotype is $\boldsymbol{y}^*$, which would perform each of these functions maximally. For each of the f organismal functions there will be a vector $\boldsymbol{q}_i \in \Re^k$ such that when the phenotype $\boldsymbol{y}$ departs from $\boldsymbol{y}^*$ in the direction $\boldsymbol{q}_i$, only the performance of adaptive function i is altered. Thus the set of $\{\boldsymbol{q}_i\}$ must be orthogonal. The amount, z_i, of this departure of adaptive function i from its optimum is simply

the component of $\boldsymbol{q}_i$ present in $\boldsymbol{y} - \boldsymbol{y}^*$, i.e., the projection of $\boldsymbol{y} - \boldsymbol{y}^*$ onto $\boldsymbol{q}_i$:

$$z_i = \boldsymbol{q}_i^{\mathrm{T}}(\boldsymbol{y} - \boldsymbol{y}^*) \ .$$

Let the departures from optimality in each adaptive function interact multiplicatively in reducing the fitness of the organism, with the relative importance of function i measured by a value $\lambda_i > 0$. A Gaussian selection scheme satisfies these specifications, giving

$$w(\boldsymbol{y}) = \exp\left[-(\boldsymbol{y} - \boldsymbol{y}^*)^{\mathrm{T}}\boldsymbol{Q}\boldsymbol{\Lambda}\boldsymbol{Q}^{\mathrm{T}}(\boldsymbol{y} - \boldsymbol{y}^*)\right] = \exp\left[-\sum_{i=1}^{f}\lambda_i z_i^2\right] \ , \qquad (5)$$

where

$$\boldsymbol{Q} = \left\|\boldsymbol{q}_1, \ldots, \boldsymbol{q}_f\right\|$$

is the matrix whose columns are $\boldsymbol{q}_i$, and $\boldsymbol{\Lambda}$ is the diagonal matrix

$$\boldsymbol{\Lambda} = \mathbf{diag}\left\|\lambda_i\right\|_{i=1}^{f} \ .$$

Assume that $\{\boldsymbol{q}_i\}$ are linearly independent, which requires $f \leq k$. Let them also be normalized, so that $\boldsymbol{Q}^{\mathrm{T}}\boldsymbol{Q} = \boldsymbol{I}$ (if $f = k$ then $\boldsymbol{Q}$ is an orthogonal matrix, hence $\boldsymbol{Q}^{\mathrm{T}} = \boldsymbol{Q}^{-1}$).

Together, $\boldsymbol{y}^*$, $\boldsymbol{Q}$, and $\boldsymbol{\Lambda}$ determine the structure of the "adaptive landscape" in terms of the organismal phenotype, $\boldsymbol{y}$.

4.2 Genetic Control of the Phenotype

Suppose there are n genes, and the allelic state at each gene i determines a genotype $x_i \in \Re$. The organismal phenotype, $\boldsymbol{y}$, is the sum of a set of normalized vectors $\boldsymbol{a}_i \in \mathbb{S}^k$ on the unit k-sphere, weighted by the values x_i. Hence

$$\boldsymbol{y} = \boldsymbol{A}\boldsymbol{x} \ , \qquad (6)$$

where

$$\boldsymbol{A} = \left\|\boldsymbol{a}_1, \ldots, \boldsymbol{a}_n\right\|$$

is the matrix whose columns are the vectors $\boldsymbol{a}_j$. The gene effects on the phenotype are additive, by the linearity of equation (6). The magnitude is partitioned from the direction of the gene's effects by normalizing $\boldsymbol{a}_j$, so that

$$\boldsymbol{a}_j^{\mathrm{T}}\boldsymbol{a}_j = \sum_i a_{ij}^2 = 1$$

for all j. The allelic value x_j controls the magnitude of the gene's effects.

The fitness function for the genotype is:

$$w(\boldsymbol{x}) = \exp\left[-(\boldsymbol{A}\boldsymbol{x} - \boldsymbol{y}^*)^{\mathrm{T}}\boldsymbol{Q}\boldsymbol{\Lambda}\boldsymbol{Q}^{\mathrm{T}}(\boldsymbol{A}\boldsymbol{x} - \boldsymbol{y}^*)\right] \ .$$

A note on epistasis: Although the loci interact additively in this model, they are also epistatic in terms of fitness, since the contribution of each allelic value to fitness depends on the value of the alleles at the other loci:

$$\partial w(\boldsymbol{x})/\partial x_i = -2w(\boldsymbol{x})\ (\boldsymbol{A}\boldsymbol{x} - \boldsymbol{y}^*)^{\mathrm{T}}\boldsymbol{Q}\boldsymbol{\Lambda}\boldsymbol{Q}^{\mathrm{T}}\boldsymbol{a}_i \ . \qquad (7)$$

4.3 "Latent" Directional Selection at Fitness Peaks under Pleiotropic Constraints

I assume that each of the elements of $\boldsymbol{x}$ are free to evolve, and that the population will eventually become fixed, through allelic substitution, on the genotype vector $\hat{\boldsymbol{x}}$ that produces the maximum fitness, i.e. which minimizes

$$\delta(\boldsymbol{x}) = (\boldsymbol{A}\boldsymbol{x} - \boldsymbol{y}^*)^{\mathrm{T}} \boldsymbol{Q}\boldsymbol{\Lambda}\boldsymbol{Q}^{\mathrm{T}} (\boldsymbol{A}\boldsymbol{x} - \boldsymbol{y}^*) \ . \tag{8}$$

This is illustrated in Fig. 2. The dynamics of the evolution toward this optimum

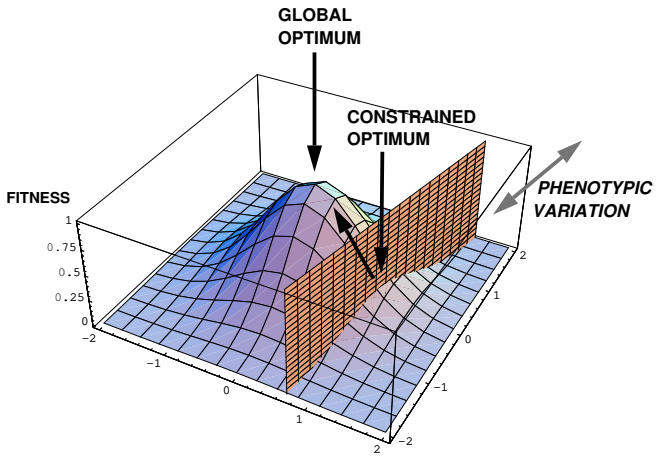

Fig. 2. Illustration of the "latent" directional selection remaining when adaptation is constrained by phenotypic variability to be suboptimal. The global optimum phenotype is $\boldsymbol{y}^*$ and the constrained optimum is $\hat{\boldsymbol{y}}$.

are not critical to what follows, but the gradient ascent model of Via and Lande (1985), extended to arbitrary dimensions, would be applicable. The constraints in this model are therefore entirely range constraints, and not kinetic constraints, on the attainable optima.

To find the minimum of $\delta(\boldsymbol{x})$ in equation (8) one differentiates. Let

$$\boldsymbol{M} = \boldsymbol{Q}\boldsymbol{\Lambda}\boldsymbol{Q}^{\mathrm{T}} \ .$$

Then $\boldsymbol{M}$ is positive definite (if $f = k$) or semi-definite (if $f < k$). The system

$$\boldsymbol{A}^{\mathrm{T}}\boldsymbol{M}(\boldsymbol{A}\hat{\boldsymbol{x}} - \boldsymbol{y}^*) = \frac{1}{2}\partial\delta(\boldsymbol{x})/\partial\boldsymbol{x} = \boldsymbol{0} \tag{9}$$

represents the "normal equations" for the minimization problem (Luenberger 1968). The closed-form solution is

$$\hat{\boldsymbol{x}} = (\boldsymbol{A}^{\mathrm{T}}\boldsymbol{M}\boldsymbol{A})^{-1}\boldsymbol{A}^{\mathrm{T}}\boldsymbol{M}\boldsymbol{y}^* \ , \tag{10}$$

and requires that the matrix $\boldsymbol{A}^{\mathrm{T}}\boldsymbol{M}\boldsymbol{A}$, known as the Gram matrix of $\boldsymbol{A}$, be positive definite. This is assured if: $\boldsymbol{A}$ is full rank, i.e. $\boldsymbol{a}_i$ are linearly independent, $\boldsymbol{M}$ is positive semi-definite, and no $\boldsymbol{a}_i$ is in the null space of $\boldsymbol{M}$, i.e. for all i, $\boldsymbol{Q}^{\mathrm{T}}\boldsymbol{a}_i \neq 0$ and $\lambda_i \neq 0$. Note that numerical computation of $\hat{\boldsymbol{x}}$ uses LU decomposition, not the matrix inversion in equation (10).

In his analysis of variability maintained by a mutation-selection balance in this model, Wagner (1989) changes coordinates so that $\boldsymbol{y}^* = \boldsymbol{0}$. But then by equation (10), $\hat{\boldsymbol{y}} = \boldsymbol{y}^*$, so the system evolves to the global fitness peak, and is not constrained by variation to be suboptimal. Although this is of no consequence for the nature of a mutation-selection balance, it eliminates the evolutionary potential afforded by the "latent" directional selection that exists when the population is constrained to be suboptimal, which is what I consider here.

Quantitative genetic models with the kind of constrained optima described here present a number of important features. Adding allelic polymorphism to the current model, as in Wagner (1989), would reveal that there can be additive genetic variance for a trait under directional selection and yet no evolution of that trait. Moreover, if selection is increased on any trait, the population will respond to it and move in the direction of the increase of selection until a new balance is found; upon relaxation of the selection to the former level, the population would return to the previous value.

4.4 Constructional Selection

The presence of latent directional selection at a constrained optimum creates adaptive opportunity for new genes that give different directions of phenotypic variability, and so until evolution reaches the global maximum, there is always the opportunity for genome growth. The process of adding new genes to the genome then is modeled as increasing the matrix $\boldsymbol{A}$ column by column. Here this process is examined under very simple evolutionary dynamics, where the population is fixed on its best attainable genotype at the time a new gene is tested in the genome. If the new gene increases fitness, it is added to the genome, and before any new genes are tested, the genotype evolves through allelic substitution to the new optimum that the new gene allows it to attain. This process is then repeated and the genome thus built up.

A new gene is added to the genome according to some random sampling process, producing a random vector, $\boldsymbol{a}_{n+1}$ — its vector of effects on the organismal phenotype — which expands $\boldsymbol{A}$ by one column to yield $\boldsymbol{A}'$. Addition of a new gene increases the length of $\hat{\boldsymbol{x}}$ by one element, x_{n+1}, a random variable, to yield $\boldsymbol{x}'$. The number of phenotypic characters, k, remains unchanged. Once the new gene is added to the genome, mutations in its allelic value x_{n+1} will change the phenotype along the same vector of variation, $\boldsymbol{a}_{n+1}$, as produced by the gene's creation. Thus there is complete correlation in this model between the phenotypic effects from the creation of the gene and the effects of its subsequent allelic variation, which is what provides the basis of the correlated allelic variation effect of constructional selection.

The departure of the fitness components from the optimum before the addition of the new gene is:

$$\delta(\boldsymbol{x}) = \boldsymbol{z}^{\mathrm{T}}\boldsymbol{\Lambda}\boldsymbol{z} = \sum_i \lambda_i {z_i}^2 \ .$$

where $\boldsymbol{z} = \boldsymbol{Q}^{\mathrm{T}}(\boldsymbol{A}\hat{\boldsymbol{x}} - \boldsymbol{y}^*)$, and each z_i is the departure of phenotype from perfect realization of adaptive function i. The fitness of the organism after addition of the new gene is

$$w(\boldsymbol{x}') = e^{-\delta(\boldsymbol{x}')}$$

where

$$\delta(\boldsymbol{x}') = (\boldsymbol{A}\hat{\boldsymbol{x}} + x_{n+1}\boldsymbol{a}_{n+1} - \boldsymbol{y}^*)^{\mathrm{T}}\boldsymbol{Q}\boldsymbol{\Lambda}\boldsymbol{Q}^{\mathrm{T}}(\boldsymbol{A}\hat{\boldsymbol{x}} + x_{n+1}\boldsymbol{a}_{n+1} - \boldsymbol{y}^*) \ . \tag{11}$$

Define:

$$\boldsymbol{\epsilon} = x_{n+1}\boldsymbol{Q}^{\mathrm{T}}\boldsymbol{a}_{n+1} \ .$$

Then

$$\delta(\boldsymbol{x}') = (\boldsymbol{z} + \boldsymbol{\epsilon})^{\mathrm{T}}\boldsymbol{\Lambda}(\boldsymbol{z} + \boldsymbol{\epsilon}) \ . \tag{12}$$

So fitness increases if and only if

$$\begin{aligned} \delta(\boldsymbol{x}') - \delta(\boldsymbol{x}) &= 2x_{n+1}\,(\boldsymbol{A}\hat{\boldsymbol{x}} - \boldsymbol{y}^*)^{\mathrm{T}}\boldsymbol{M}\boldsymbol{a}_{n+1} + x_{n+1}^2\,\boldsymbol{a}_{n+1}^{\mathrm{T}}\boldsymbol{M}\boldsymbol{a}_{n+1} \\ &= (2\boldsymbol{z} + \boldsymbol{\epsilon})^{\mathrm{T}}\boldsymbol{\Lambda}\boldsymbol{\epsilon} = \sum_i \lambda_i(2z_i + \epsilon_i)\epsilon_i < 0 \ . \end{aligned} \tag{13}$$

The effect of the new gene on fitness depends on both its magnitude x_{n+1} and its direction $\boldsymbol{a}_{n+1}$. In order for changes in function i to contribute toward increased fitness, z_i and ϵ_i must be of opposite sign (i.e. the new gene changes the genotype in the opposite direction from its error), and

$$|\epsilon_i| < 2|z_i| \ . \tag{14}$$

If x_{n+1} is very small, then

$$(2z_i + \epsilon_i)\epsilon_i \approx 2z_i\epsilon_i \ ,$$

and under a wide variety of assumptions about the distributions of x_{n+1}, the probability that a new gene will produce a fitness increase would be 1/2, independent of the new gene's pleiotropy vector, $\boldsymbol{a}_{n+1}$. Thus there would be no constructional selection on $\boldsymbol{a}_{n+1}$.

If x_{n+1} is distributed with larger values, however, the condition in equation (14) corresponds to the new gene not causing the phenotype to overshoot the maximum for function i and produce a fitness contribution lower than before. If any z_i has evolved to be very small, i.e., the organismal phenotype has realized adaptive function i very well, then a large perturbation ϵ_i from any new gene reduces the chance that it increases fitness. This selection against large ϵ_i is greater with larger λ_i. Thus there will be selection against the addition of new genes that alter existing highly adapted functions. Under this model, new genes that are incorporated in the growing genome will therefore tend to have lower pleiotropy for existing organismal functions than randomly added genes.

A Measure of Pleiotropy. A measure $p_{\boldsymbol{A}}(\boldsymbol{a}_{n+1})$ of the pleiotropy of the new gene can be defined to display the extent to which the new gene disturbs the existing constrained optimum:

$$p_{\boldsymbol{A}}(\boldsymbol{a}_{n+1}) = \frac{\hat{\boldsymbol{x}}^{\mathrm{T}} \boldsymbol{A}^{\mathrm{T}} \boldsymbol{M} \boldsymbol{a}_{n+1}}{\boldsymbol{y^*}^{\mathrm{T}} \boldsymbol{M} \boldsymbol{a}_{n+1}} \ .$$

We see from equation (9) that pleiotropy is large for a new gene that moves the phenotype in a direction within the space of variability that it is already optimized for:

$$p_{\boldsymbol{A}}(\boldsymbol{a}_i) = 1 \quad \text{for } i = 1 \ldots n \ .$$

Whereas pleiotropy is small when the new gene moves the phenotype in the exact direction of the global optimum, $\boldsymbol{A}\hat{\boldsymbol{x}} - \boldsymbol{y^*}$:

$$p_{\boldsymbol{A}}(\boldsymbol{A}\hat{\boldsymbol{x}} - \boldsymbol{y^*}) = 0 \ .$$

Then condition equation (13) for a fitness increase can be written:

$$\delta(\boldsymbol{x}') - \delta(\boldsymbol{x}) = x_{n+1}^2 \, \boldsymbol{a}_{n+1}^{\mathrm{T}} \, \boldsymbol{M} \, \boldsymbol{a}_{n+1} - 2x_{n+1} \, \boldsymbol{y^*}^{\mathrm{T}} \, \boldsymbol{M} \, \boldsymbol{a}_{n+1} \, [1 - p_{\boldsymbol{A}}(\boldsymbol{a}_{n+1})] < 0 \ .$$

Since the first term is always positive, a fitness increase requires that:

1. $\boldsymbol{x}_{n+1}$ be the same sign as $\boldsymbol{y^*}^{\mathrm{T}} \, \boldsymbol{M} \, \boldsymbol{a}_{n+1}$, i.e. the change is toward rather than away from the optimum; and that
2. the term $1 - p_{\boldsymbol{A}}(\boldsymbol{a}_{n+1})$ be as large as possible, i.e. that the pleiotropy value is small.

Genetic Modifiers of Pleiotropy. It should be mentioned that the same analysis applies to selection on a modifier gene that changes the $\boldsymbol{A}$ matrix. Suppose an allele at a modifier locus changes matrix $\boldsymbol{A}$ to $\boldsymbol{A} + \boldsymbol{C}$. Then with the substitution $x_{n+1}\boldsymbol{a}_{n+1} = \boldsymbol{C}\hat{\boldsymbol{x}}$ in equation (11) the subsequent analysis (through equation (14)) applies. The selective advantage of the modifier relative to the unmodified genotype is

$$\frac{w'(\boldsymbol{x})}{w(\boldsymbol{x})} - 1 = e^{\delta(\boldsymbol{x}) - \delta'(\boldsymbol{x})} - 1 \ .$$

Here, w' and δ' indicate values using $\boldsymbol{A} + \boldsymbol{C}$ for $\boldsymbol{A}$. So any modifier locus which is able to change the genotype-phenotype map, $\boldsymbol{A}$, has a potential selective advantage of as much as the "latent" directional selection, $e^{\delta(\boldsymbol{x})} - 1$.

4.5 Numerical Simulation

A numerical simulation of this model illustrates the constructional selection process. The genome is grown gene by gene according to the algorithm illustrated in Fig. 3:

1. Randomly create the adaptive landscape matrices matrices $\boldsymbol{Q}$, $\boldsymbol{\Lambda}$, and optimal phenotype vector $\boldsymbol{y}^*$:
   (a) pick the elements of $\boldsymbol{Q}$ uniformly on $[-1, 1]$ and then orthogonalize the columns (the Modified Gram-Schmidt algorithm was used (Golub and Van Loan 1983));
   (b) generate the diagonal elements of $\boldsymbol{\Lambda}$ uniformly on $[0, 1]$;
   (c) generate elements y_i^* uniformly on $[-1, 1]$.
2. Add a new gene to the genome:
   (a) create a new pleiotropy vector $\boldsymbol{a}_{n+1}$ by picking elements a_i uniformly on $[-1, 1]$ and then normalizing so that $\sum_i a_i^2 = 1$;
   (b) let the allelic value, x_{n+1}, for the new gene equal a scale value which exponentially decreases until the new gene is kept.
3. In a run when constructional selection is acting: if the new gene decreases fitness, reject it and repeat step 2. Otherwise, keep it.
4. Adapt $\boldsymbol{x}$ to the new optimum $\hat{\boldsymbol{x}}$.
5. Repeat step 2 until the genome has 32 genes.

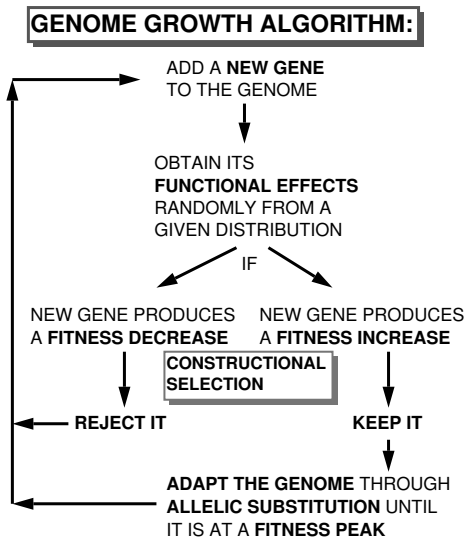

Fig. 3. The genome growth algorithm used in the simulation.

In this simulation, the pleiotropy vectors, $\boldsymbol{a}_{n+1}$, are chosen from the same distribution throughout the run. Therefore, there is no heritability on the level of genome-as-population, and thus no opportunity for the genic selection effect. The obvious scheme of heredity for gene-to-gene duplications will not produce meaningful results given the way the model is set up. Consider a simple form of heredity, where new vectors $\boldsymbol{a}_{n+1}$ are resampled from $\{\boldsymbol{a}_1, \ldots, \boldsymbol{a}_n\}$, the columns

of $\boldsymbol{A}$. The new gene would have maximal pleiotropy and always be deleterious since it could only move the phenotype off its constrained peak; the new matrix $\boldsymbol{A}'$ would be less than full rank, moreover, giving a continuum of constrained optima. So with the linear genotype-phenotype map, the genic selection effect would not occur under this model of heredity.

The procedure for choosing x_{n+1} in step 2b was taken instead of choosing x_{n+1} from some random distribution in order to lessen the variance in the stringency of constructional selection on $\boldsymbol{a}$ (as discussed in Sect. 4.4) and to maintain a roughly constant stringency of constructional selection as the genome grows.

Simulation were run both with and without constructional selection (where each new gene is accepted in the genome regardless of its immediate effect on fitness), to allow comparison between genomes resulting from constructional selection and genomes sampled from the underlying random distribution of gene effects. In these simulations, there are 64 organismal functions under Gaussian stabilizing selection, and the genomes evolve from one gene to 32.

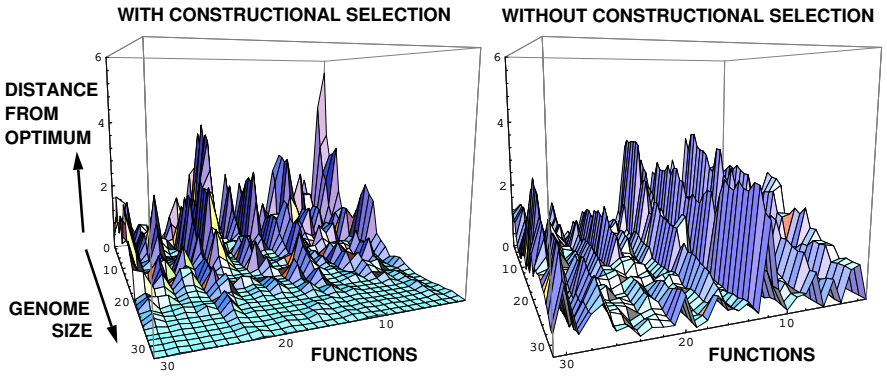

Fig. 4. Fitness components for multiple organismal functions during genome growth: in a genome evolved with (left) and without (right) constructional selection. The height, $\lambda_i z_i^2$, measures departure of each organismal function from optimality. For clarity, only 32 of the 64 different adaptive functions are plotted, in arbitrary order.

Figure 4 shows the evolution of the fitness components for each organismal function as the genome grows. The height, $\lambda_i z_i^2$, plotted for each function i, represents the departure of each component from optimality as the genome is increased from 1 to 32 genes. The bumps in the landscape indicate where gene addition decreases the adaptation for certain components, while raising it for other. Comparison between the genomes grown with and without constructional selection shows that adaptation simultaneously at many organismal functions can be achieved with a much smaller genome when constructional selection acts during the evolution of the genotype-phenotype map.

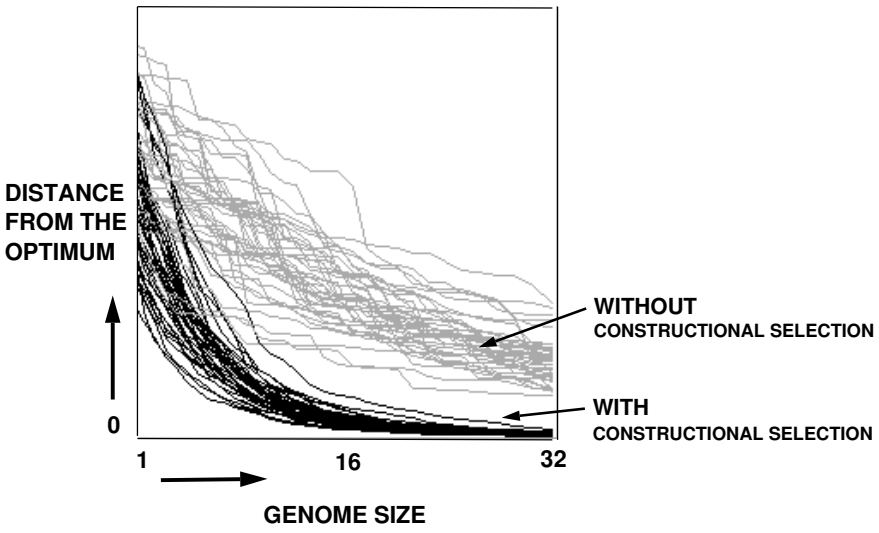

Fig. 5. Organismal fitness as a function of genome size for several runs of the genome growth algorithm, with (dark lines) and without (light lines) constructional selection.

Figure 5 shows the trajectories of organismal fitness as new genes are added to the genome. The phenotype $\boldsymbol{y}$ always moves closer to $\boldsymbol{y}^*$ whether or not constructional selection is acting, because any generic new gene increases the phenotype subspace spanned by the genetic variation regardless of its immediate effect on fitness. With constructional selection, however, rapid approach to the global optimum in the adaptive landscape occurs with much smaller genome size. Genomes with the random distribution of phenotypic effects had to grow to a size of 32 genes to reach the same fitnesses attained by genomes of only around 5 genes when these underwent constructional selection. In these simulations, most of the adaptation occurs not from the addition of the new genes, but from the climb to the constrained fitness peaks that occurs between gene additions, the part attributable to allelic substitution.

5 The "NK" Adaptive Landscape Model

Kauffman's "NK" adaptive landscape model (1989) will be used to illustrate the effects of constructional selection because it explicitly shows the epistatic structure of the genotype-phenotype map. A separate presentation of this material can be found in Altenberg (1994). First I will describe the NK model and review existing analytical work on its evolutionary behavior. Then I will examine the properties of genomes evolved under constructional selection including their adaptive performance and the nature of the emergent genotype-phenotype maps.

Kauffman's NK model has the following components:

- A genome consists of n genes;
- Each gene contributes a fitness component to the organism, and these are summed to give the total organismal fitness;
- The fitness component contributed by a given gene i depends on the allelic state at k other genes.

Although Kauffman ascribes each fitness component to a particular gene, in his model control over each fitness component is, in fact, symmetric with respect to all the genes that affect it. So in the development to follow, I recast the NK model in terms of a map between a set of genes and a set of fitness components. This allows the number of fitness components to differ from the number of genes, and allows genes to be added to the genome while keeping the set of fitness components fixed. This is illustrated in Fig. 6. The elements of the model are recast as follows:

1. The haploid genome consists of n binary-valued genes, that exert control over f phenotypic functions, each of which contributes a component to the total fitness.
2. Each gene controls a subset of the f fitness components, and in turn, each fitness component is controlled by a subset of the n genes. This genotype-phenotype map can be represented by a matrix,
$$\boldsymbol{M} = \|m_{ij}\|,\ i = 1 \ldots n,\ j = 1 \ldots f,$$
of indices $m_{ij} \in \{0, 1\}$, where $m_{ij} = 1$ indicates that gene i affects fitness component j;
3. The columns of $\boldsymbol{M}$, called the *polygeny vectors*, $\boldsymbol{g}_j = \|m_{ij}\|$, $i = 1 \ldots n$, give the genes controlling each fitness component j;
4. The rows of $\boldsymbol{M}$, called the *pleiotropy vectors*, $\boldsymbol{p}_i = \|m_{ij}\|$, $j = 1 \ldots f$, give the fitness components controlled by each gene i;
5. If any of the genes controlling a given fitness component mutates, the new value of the fitness component will be uncorrelated with the old. Each fitness component ϕ_i is a uniform pseudo-random function[3] of the genotype, $\boldsymbol{x} \in \{0, 1\}^n$:
$$\phi_i(\boldsymbol{x}) = \Phi(\boldsymbol{x} \circ \boldsymbol{g}_i, i, \boldsymbol{g}_i) \sim \text{uniform on } [0, 1] \ ,$$
where $\Phi : \{0, 1\}^n \times \{1, \ldots, n\} \times \{0, 1\}^n \mapsto [0, 1]$, $\circ$ is the Schur product ($\boldsymbol{x} \circ \boldsymbol{g}_j = \|x_i m_{ij}\|$, $i = 1 \ldots n$). Any change in i, $\boldsymbol{g}_i$, or $\boldsymbol{x} \circ \boldsymbol{g}_i$ gives a new value for $\Phi(\boldsymbol{x} \circ \boldsymbol{g}_i, i, \boldsymbol{g}_i)$ that is uncorrelated with the old;
6. If a fitness component is affected by no genes, it is assumed to be zero:
$$\Phi(\boldsymbol{x} \circ \boldsymbol{g}_i, i, \boldsymbol{g}_i) = 0 \text{ for all } \boldsymbol{x}, \text{ if } \boldsymbol{g}_i = \left\| 0 \ldots 0 \right\| \ ;$$
7. The total fitness is the normalized sum of the fitness components:
$$w(\boldsymbol{x}) = \frac{1}{f} \sum_{i=1}^{f} \phi_i(\boldsymbol{x}) \ . \tag{15}$$

[3] The popular Park-Miller algorithm generates non-random bits, so the encryption-like algorithm *ran4* described in Press, *et al.* (1992) was used.

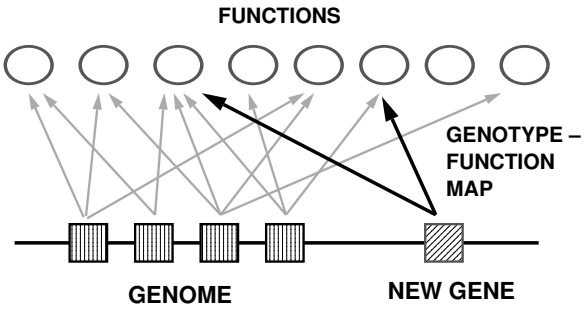

Fig. 6. Kauffman's NK model recast as a map between the genotype and a set of fitness components. Arrows indicate that the gene affects the fitness component. A new gene with effects on two fitness components is shown being introduced to the genome.

5.1 Pleiotropy and Evolvability

With the random fitness function $w(\boldsymbol{x})$ now defined, the relationship between the genotype-phenotype map and the model's adaptive behavior can be investigated. The random fitness function $w(\boldsymbol{x})$ causes genotypes that are one mutational event away from one another to be more or less correlated, depending on the genotype-phenotype map. The statistical property that affects adaptation is the likelihood that a genotype is fitter than all the genotypes that are one mutation different from it. The set of genotypes that are one mutation away from a given genotype can be called its "neighborhood", and if it is the fittest genotype in its neighborhood, then it is a fitness "peak", to use the metaphor of the adaptive landscape (Wright 1932). The NK fitness function thus produces a tunably rugged landscape (Kauffman 1989).

Mutation is not the only variation-producing mechanism involved in evolution. Recombination is also very important. However, in the case of sequence evolution on rugged adaptive landscapes, it has been argued that single mutations are the main mechanism of change. Maynard Smith (1970) proposed that molecular evolution must be limited mainly to moves from a genotype to one of its fitter single- mutation neighbors. Gillespie (1984) provided a theoretical population genetic analysis corroborating that evolution on "mutational landscapes" would consist mainly of "adaptive walks", in which the population moves from fixation of one genotype to fixation of a neighboring genotype of greater fitness. So such adaptive walks will be used here.

Adaptive walks have been used to study the statistics of adaptation on NK fitness landscapes (Kauffman and Levin 1987, Kauffman 1989, Macken and Perelson 1989, Weinberger 1991). Beginning with a chosen genotype, the fitness of each of its 1-mutant neighbors is evaluated. If there are no fitter genotypes, the genotype is at a fitness peak and the adaptive walk stops. Otherwise, one moves to the fittest genotype and begins the process again.

In the NK model, the chance that a mutation produces a fitness increase will depend on the pleiotropy of the genotype-phenotype map. This effect can be analyzed as follows. Define the *pleiotropy value*,

$$k_i = \sum_{j=1}^{f} m_{ij}$$

to be the number of fitness components affected by gene i (the K in Kauffman's usage is $k_i - 1$ here). Define the *marginal fitness* of gene i as the sum of the fitness components it affects:

$$w_i(\boldsymbol{x}) = \sum_{j=1}^{f} m_{ij}\phi_j(\boldsymbol{x}) \ .$$

When gene i mutates, each fitness component it affects is resampled uniformly from [0,1] independently. The probability that its new marginal fitness will be less than y is

$$\begin{aligned} F_k(y) &= \Pr[S_k < y] \\ &= \frac{1}{k!}\sum_{i=0}^{k}(-1)^i \binom{k}{i}\left(\frac{y - i + |y-i|}{2}\right)^k \ , \end{aligned} \tag{16}$$

where S_k is the sum of k independent uniform random variables on [0,1] (Feller 1971). The probability distribution $F_k(y)$ is plotted against y/k for different values of k in Fig. 7. One can see how as k increases, the probability density concentrates around the expected value $E(\phi_i) = 1/2$, an illustration of the Central Limit Theorem. Thus in genes with higher pleiotropy k, mutations have a stronger regression toward the fitness $y/k = 1/2$, eith diminishing upper tails of the fitness density. Therefore, lower fitnesses are likely to be fitness peaks (i.e. all its 1-mutant neighbors are less fit) for genotype-phenotype maps with high pleiotropy.

5.2 Statistics of Fitness Peaks on Generic Landscapes

We would like to take our knowledge of the neighborhood properties of the fitness function in equation (16) and see how evolution proceeds. A principle question is how fit the peaks are that are arrived at from random starting points. Analysis of the probability distribution of endpoints of such adaptive walks have been made for $k = 1$ and $k = n$ (Kauffman and Levin 1989, Macken and Perelson 1989).

Intermediate values of k present analytical difficulties, so Weinberger (1991) took an indirect approach to solving the distribution of fitness peaks. Instead of looking at the distribution of fitness peaks arrived at from random initial genotypes, he looked at the fitness distributions of adaptive peaks from among the unweighted set of fitness peaks. The results are reviewed as follows.

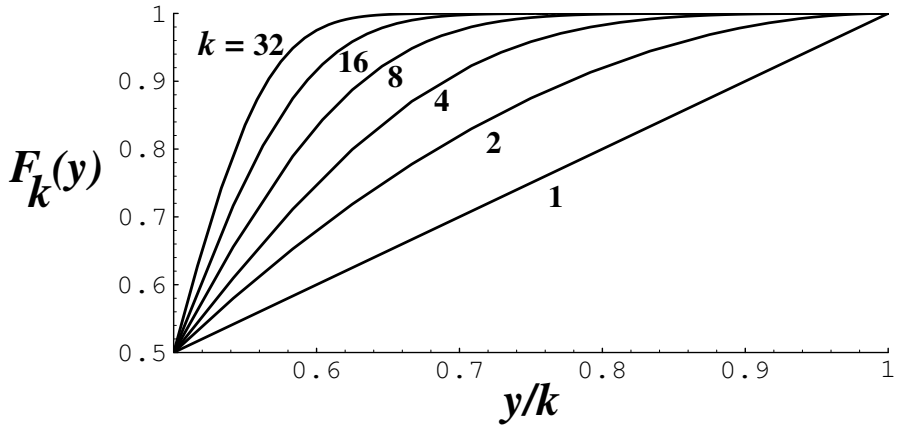

Fig. 7. The probability, $F_k(y)$, that the marginal fitness of a mutation affecting k fitness components will be greater than y. Plotted with the abscissa normalized by k, for $k = 1 \ldots 32$.

Given that a genotype $\boldsymbol{x}$ has marginal fitnesses, $w_j(\boldsymbol{x})$, the probability that it is a fitness peak is

$$\Pr[\boldsymbol{x} \text{ is a local fitness peak} \mid \boldsymbol{\phi}] = \prod_{i=1}^{n} F_{k_i}\left(w_i(\boldsymbol{x})\right) \quad , \tag{17}$$

where $\boldsymbol{\phi} = \|\phi_i\|$, $i = 1 \ldots f$, and $\{S_{k_i}\}$ are independent random variables with distributions F_{k_i}. Letting ϕ_i and S_{k_i} be random variables, the probability that a random point is a fitness peak is

$$\begin{aligned} \hat{p} &= \Pr[\sum_{j=1}^{f} m_{ij}\phi_j > S_{k_i} \forall i] \\ &= \int_{\boldsymbol{\phi} \in [0,1]^f} \prod_{i=1}^{n} F_{k_i}(\sum_{j=1}^{f} m_{ij}\phi_j) d\boldsymbol{\phi} \ . \end{aligned} \tag{18}$$

The probability, $G(y)$, that a genotype, given it is a fitness peak, has fitness less than y is

$$G(y) = \frac{1}{\hat{p}} \Pr[\sum_{i=1}^{f} \phi_i < y \text{ and } \sum_{j} m_{ij}\phi_j > S_{k_i} \forall i] \ . \tag{19}$$

Weinberger (1991) obtained normal approximations of $G(y)$ for intermediate large values of k, with $f = n$. Assuming $k_i = k$ for all i, and denoting $\mu = \mathrm{E}[\phi_i(\boldsymbol{x})]$ and $\sigma^2 = \mathrm{Var}[\phi_i(\boldsymbol{x})]$, Weinberger's (1991) approximation for the distribution of fitnesses among fitness peaks is

$$G(y) = \mathcal{N}\left(\frac{y/f - \mu_G}{\sigma_G}\right) \quad , \tag{20}$$

where $\mathcal{N}()$ is the normal distribution with mean 0 and variance 1, and

$$\mu_G = \mu + \sigma\sqrt{\frac{2\ln(k)}{k}} \text{ and } \sigma_G^2 = \frac{\sigma^2}{n\left[1+(1+\frac{1}{k})2\ln(k)\right]} \ .$$

For the uniform fitness functions, $\mu = 1/2$ and $\sigma^2 = 1/12$. Figure 8 shows Weinberger's approximation of $G(y)$ plotted for $n = 31$ and several values of k. As k increases, the distribution of fitness peaks gets lower (when normalized by f), approaching the expectation for random genotypes, $f/2$. In other words, genotype-phenotype maps with large amounts of pleiotropy do not allow 1-mutant adaptive walks to get very near their global optima ($\approx f$).

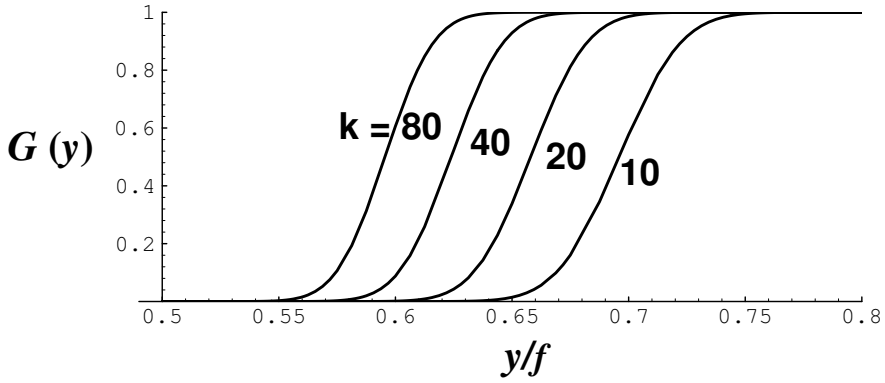

Fig. 8. Weinberger's (1991) normal approximation for the distribution of fitnesses among fitness peaks in the NK model. Plotted for $n = 31$ and $k = 10$, 20, 40, and 80.

5.3 Constructional Selection for Low Pleiotropy

The effect of selective genome growth on the degree of pleiotropy in the evolved genotype-phenotype map can be analyzed, as follows.

Suppose a gene newly added to the genome has pleiotropy vector $\boldsymbol{p}_{n+1}$, and affects $k_{n+1} = \sum_{j=1}^{f} m_{n+1\,j}$ fitness components, which become resampled uniformly from the interval [0,1]. If a fitness component is not yet affected by any gene, then its preexisting value is 0.

Let y be the sum, before the new gene is added, of the fitness components the new gene is going to alter. The probability that the new sum will be less than y is $F_{k_{n+1}}(y)$ from equation (16). Then, from equation (16), the probability that the new gene will produce a fitness increase is $1 - F_{k_{n+1}}(y)$. As can be seen from Fig. 7, when the average of the fitness components to be altered by the new gene is above 1/2, the greater k_{n+1} is, the less the chance that the new gene

will produce a fitness increase, precipitously less so for highly adapted fitness components. Since the new gene is kept only if it produces a fitness increase, constructional selection will filter out genes with high k.

Suppose that there is an underlying probability density $s(k)$ of pleiotropy values k for genes newly added to the genome. Then the density $s^*(k)$ of pleiotropy values among genes that are kept by the genome (i.e. which improve fitness) will be

$$s^*(k) = s(k) \sum_{\boldsymbol{p} \in \{0,1\}^f} \Pr[\boldsymbol{p}|k] \left[1 - F_k(\boldsymbol{p}^{\mathrm{T}} \boldsymbol{\phi})\right] / N \ , \tag{21}$$

where $\boldsymbol{\phi}$ is the vector of fitness components before the gene was added, $\Pr[\boldsymbol{p}|k]$ is the probability of sampling pleiotropy vector $\boldsymbol{p}$ given that the new gene's pleiotropy value is k, and N is the normalizer so that $\sum_k s^*(k) = 1$.

The way constructional selection filters out high pleiotropy as the adaptedness of the genome increases is illustrated in Fig. 9. It plots equation (21) with the assumption that all fitness components are the same, i.e. $\phi_i = \overline{\phi}$ for all i. The underlying density of pleiotropy values before selection is taken to be uniform on $1 \ldots f$. The figure shows that the more highly adapted the genome is, the more severe is the selection against high pleiotropy.

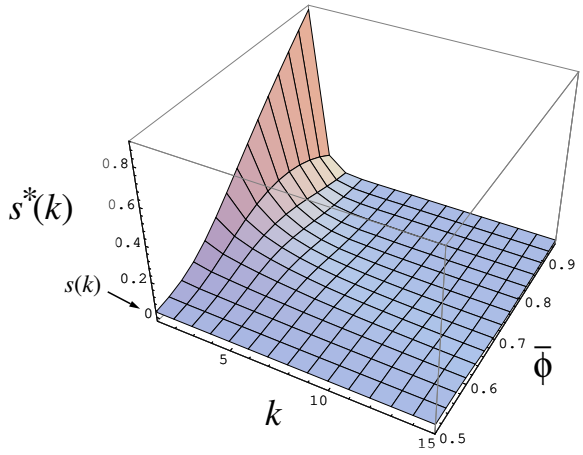

Fig. 9. The density, $s^*(k)$, of pleiotropy values k, among genes successfully incorporated in the genome, plotted as a function of the fitness component average, $\overline{\phi}$, prior to the gene's addition. The arrow points out the plot of the prior density $s(k)$, of pleiotropy values from which the genes are sampled. $s(k)$ is uniform on $\{1, \ldots, f\}$, and here, $f = 31$.

5.4 Numerical Results

A numerical simulation of constructional selection in the NK model was performed using the same genome growth algorithm as was used in Sect. illustrated in Fig. 3:

1. Add a new gene to the genome:
   (a) create a new pleiotropy vector $\boldsymbol{p}_{n+1}$, choosing uniformly (from $\{1, \ldots, 31\}$) the number, k_{n+1}, of fitness components to be affected by the new gene, and then selecting randomly which fitness components these are, from a set of $f = 31$ possible;
   (b) pick the allelic value, x_{n+1}, of the new gene with probability 1/2 being either 0 or 1.
2. If the new gene decreases fitness, reject it and repeat step 1. Otherwise, keep it.
3. Adapt $\boldsymbol{x}$ to the new (local) optimum $\hat{\boldsymbol{x}}$ by allelic substitution through a "greedy" 1-mutant adaptive walk.
4. Repeat step 1 until the genome has 31 genes.

The pleiotropy vectors, $\boldsymbol{p}_{n+1}$, are chosen from the same uniform distribution throughout the run. As a basis for comparison, the genome growth algorithm is also run without step 2, giving the result of choosing representations *a priori*.

Evolved Genotype-Phenotype Maps. Figure 10 shows typical genotype-phenotype maps produced during runs with and without constructional selection. The run without constructional selection reflects the underlying distribution of pleiotropy vectors sampled for each new gene. In the run with constructional selection, during the evolution of the first few genes, the discovery of new fitness components selects for high pleiotropy, but as these fitness components evolve toward their optima, selection becomes strong against new genes affecting them.

This increasing selection for low pleiotropy can be seen in Fig. 11, which shows the distribution of pleiotropies k_n as the genome grows, over repeated runs of genome growth. It can be seen to resemble the predicted distribution in Fig. 9. The mode for k_n is always 1 after the first few genes, but as shown in Fig. 12, the mean k_n tends toward 1 from initial values of around 16, or half of the maximum possible, $f = 31$.

The progress in adaptation can be compared between runs with and without constructional selection. Figure 13 shows plots for a number of runs. Without constructional selection, disruptive new genes are not filtered out, and adaptation shows little progress once the fitness components are saturated with genes that affect them. With constructional selection, however, fitness continues to increase with each new gene throughout the genome growth.

As the genome grows, the trajectories of individual fitness components can be seen in Fig. 14. With constructional selection, once a fitness component has reached a high value (low points in graph), only new genes that leave it alone are likely to be incorporated in the genome. Occasionally, however, one component is

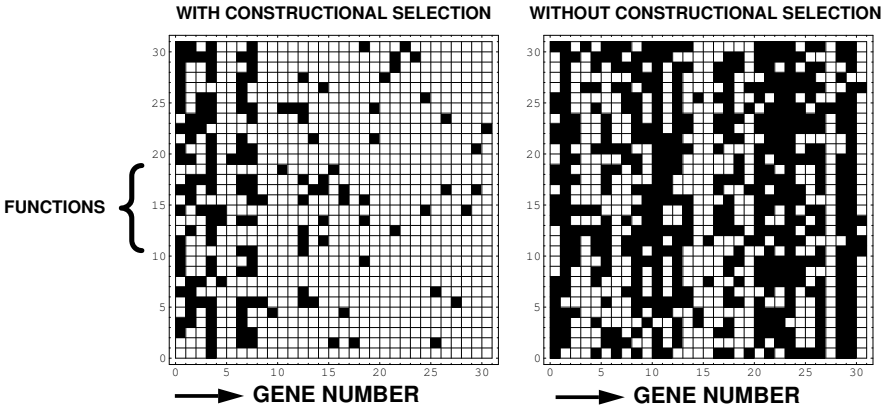

Fig. 10. Two genotype-phenotype maps evolved through genome growth, with (left) and without (right) constructional selection. Dark squares indicate that fitness component j depends on gene i. The columns in the right map reflect the sampling distribution of the pleiotropy vectors, in which the number of fitness components affected is uniform on $[1, f]$. The left map shows how under constructional selection, later genes have lower pleiotropy as the genome grows and becomes more adapted.

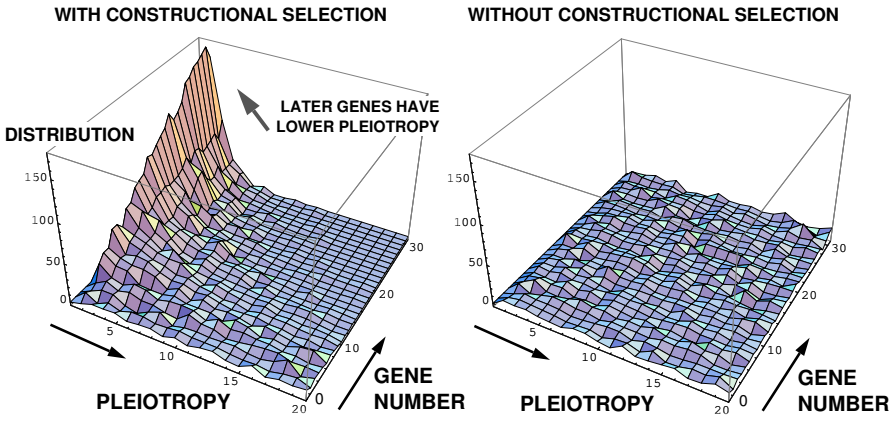

Fig. 11. The distribution, from repeated runs of the genome growth algorithm, of pleiotropy values k_n, from each gene's pleiotropy vector $\boldsymbol{p}_n$, as the genome grows; with (left) and without (right) constructional selection.

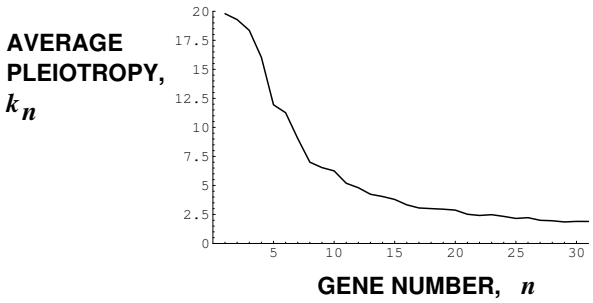

Fig. 12. The average pleiotropy values k_n for each gene as the genome grows, from the runs in Figure 11, with constructional selection.

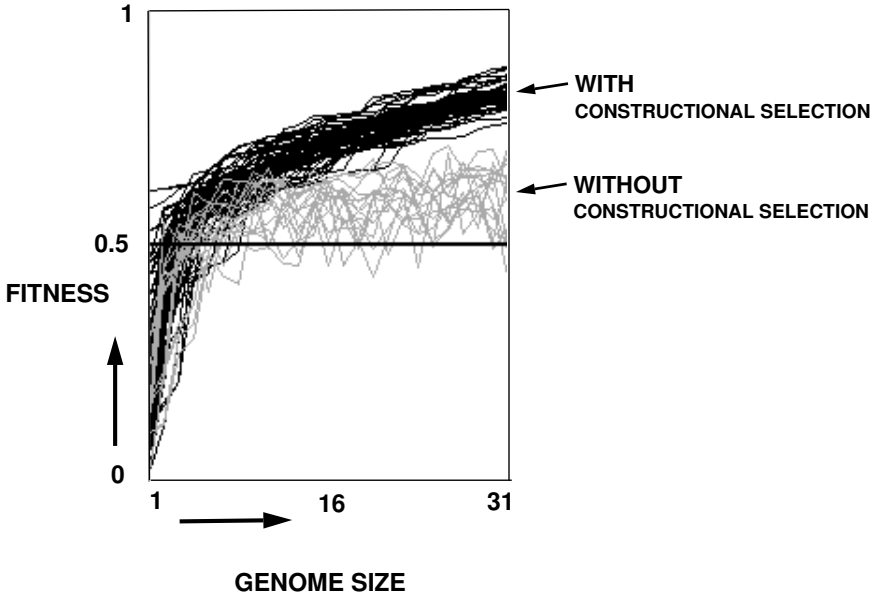

Fig. 13. Fitness as a function of genome size for several runs of the genome growth algorithm. Dark lines are with, and light lines without constructional selection.

sacrificed for the improvement in another, which show up as spikes in the graph. By the time the genome has reached a size of 31 genes, most of the components have reached values well above their expected value of 1/2. Without constructional selection, the jumble of spikes represents the continuing randomization of the fitness components as genes with random pleiotropy are incorporated into the genome.

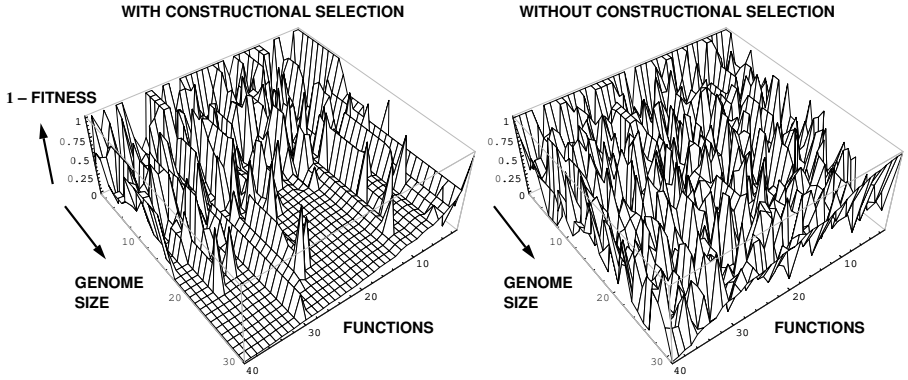

Fig. 14. Fitness components during genome growth, for one genome evolved with (left) and one without (right) constructional selection. Fitness components are sorted according to their value at the end of the run.

Here, most of the adaptation occurs during the incorporation of new genes, rather than during the adaptive walks (through allelic substitution) between gene additions. This is because there is a much larger pool of new pleiotropy vectors to sample from than the pool of genotypes in the 1-mutant neighborhood of an existing genotype (2^f vs. n). The evolutionary process under constructional selection is figuratively the "building" of a fitness peak, gene by gene, rather than the climbing of a fitness peak.

The correlated allelic variation effect as discussed in Sect. 3.1 is illustrated here by the fact that low pleiotropy evolves. Compared to average genes being tested, the genes kept by the genome will have a higher correlation between the fitness when the gene was added and its alternate allele because of its low pleiotropy. However, by the nature of the NK model, the single alternate allele of an advantageous new gene is unlikely to be fitter still. The ability of the correlated allelic variation effect to enhance evolvability, through the low pleiotropy of genotype-phenotype maps produced under constructional selection, would be evident in the event of shifts in the adaptive peak. Should any of the fitness components change due to a changed environment, the low pleiotropy of the genes that affect these fitness components would enhance their chance of producing alleles that respond to the change, without causing a prohibitive disruption of functions for which selection has not changed.

Non-Generic Properties of Evolved Landscapes. Existing theory for adaptive walks on NK landscapes, as in Sect. 5.2, has been derived for generic landscapes, i.e. landscapes that one would typically obtain from a random sampling of landscapes with given values of n and k (Kauffman and Levin 1987, Kauffman 1989, Weinberger 1991) . The applicability of these results to organic evolution assumes that evolutionary processes produce such generic adaptive landscapes. However, the distribution of fitness peaks in the NK landscapes grown here under constructional selection are nowhere near the distributions for generic NK landscapes with identical genotype-phenotype maps.

Constructional selection produces genotype-phenotype maps that are much more finely tuned to the fitness function under which they evolved. To illustrate this, the distribution of fitness peaks for several landscapes evolved under constructional selection are plotted in Fig. 15. For comparison, distributions are plotted for landscapes using the same genotype-phenotype map, but with fitness functions, Φ, chosen *a priori*. Each point represents the fitness peak obtained by starting an adaptive walk from a randomly sampled genotype. The distributions are plotted by sorting the fitness peaks by size (the transpose of the figure therefore represents the cumulative probability distribution for fitness peaks). The width of horizontal plateaus represents the size of the domain of attraction for a particular fitness peak.

The plateaus, and discontinuities between them, indicate fewer and larger domains of attraction for the evolved landscapes, i.e. they are smoother than the generic landscapes. The distributions for the generic landscapes follow roughly the Gaussian approximation derived by Weinberger (1991) as seen in Fig. 8 (fitting close to the generic $k = 10$ landscapes). While the least-fit peaks are approximately the same for both evolved and generic landscapes, at various points in the ranking, the fitness of the evolved landscapes grows much higher. Interestingly, the jumps in the distribution are highly variable.

An additional beneficial outcome of constructional selection is that the genotypes resulting at the end of the run are usually the apparent global fitness peak. In 77% of adaptive landscapes evolved under constructional selection (304 sampled), the genotypes attained at the end of genome growth were fitter than any other adaptive peak found (from 250 other starting genotypes). Of the remaining landscapes, only an average of 19% of random initial genotypes evolved to peaks fitter than the genotype attained at the end of genome growth.

5.5 Lineage Selection vs. Constructional Selection

The NK model can be used to compare the effectiveness of lineage selection (see Sect. 6.3) with that of constructional selection in producing evolvable genotype-phenotype maps. The idea of lineage selection is that the organisms whose developmental mechanisms happen to be most evolvable will found the most successful phyletic lineages, so that evolvable species will proliferate at the greatest rate (Dawkins 1988). It leaves evolvability within lineages as a byproduct of their genotype-phenotype map not subject to secular evolutionary pressure.

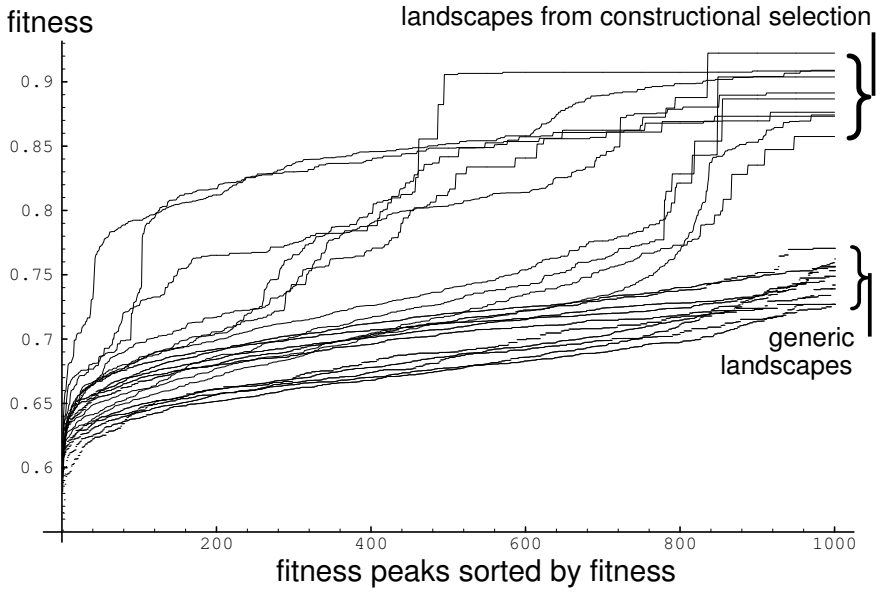

Fig. 15. Distributions of fitness peaks of NK landscapes: upper 10 plots are for adaptive landscapes evolved under constructional selection; lower 10 plots are with the same genotype-phenotype maps but randomized fitness functions. In each plot, the peaks attained from 1000 random starting genotypes are sorted by fitness. Plateaus indicate large domains of attraction for the peak.

Lineage selection in the NK model could be implemented by generating random NK landscapes, selecting those with the greatest evolvability, and evaluating the height of the fitness peaks on these landscapes. This can be compared to the height of fitness peaks of genomes evolved through constructional selection. The basis for comparison will be the fitness of the fittest individual obtained after a set number of genotypes have been generated. The payoff in the level of optimization obtained through constructional selection shows it to be a much more powerful than lineage selection.

To give lineage selection the best possible advantage, I will consider the class of NK landscapes with the highest expected fitness peaks, the $k = 1$ landscapes (which is $K = 0$ in Kauffman's original definition). In the $k = 1$ landscapes, there is a one-to-one map from each gene to each fitness component, and $f = n$.

Each gene can be optimized individually, so it takes the evaluation of $2n$ genotypes to find the global peak for the landscape. Each fitness component is i.i.d., where the optimal $\hat{\phi}_i$ is distributed as the maximum of two independent uniform random variables on [0,1]. The probability density of each maximum $\hat{\phi}_i$ is $f(\hat{\phi}) = 2\hat{\phi}$. So $\mathrm{E}(\hat{\phi}) = 2/3$ and $\mathrm{Var}(\hat{\phi}) = 1/18$. With $n = f = 31$ genes and fitness components, one obtains:

$$\mathrm{E}[w(\hat{\boldsymbol{x}})] = 2/3, \text{ and } \mathrm{Var}[w(\hat{\boldsymbol{x}})] = \frac{1}{18f} \approx 0.00179 \ .$$

The average fitness attained under constructional selection in the numerical simulations (where the average k is much larger than 1) is about 0.89, which is therefore some 5 standard deviations (5×0.0423) above the expected value of peaks obtained from generic $k = 1$ landscapes. The fraction of randomly generated $k = 1$ landscape having a peak with fitness at least 0.89, under a normal approximation, is 3×10^{-7}. So over 2×10^6 different $k = 1$ genotype-phenotype maps would need to be sampled to be likely to obtain fitness peaks as high as those obtained through constructional selection, which in the runs here took some 3000 sampled genotypes. The ability to select on the genotype-phenotype map as it is constructed is the key to finding higher fitness values.

For a more even comparison with lineage selection, we can see what kind of $k = 1$ landscapes constructional selection can produce. A simple way to implement this would be to add one gene to the genome at a time, map the gene to each of the remaining unmapped fitness components, evaluate the fitnesses of both alleles with each map, and keep the allele and map that give the fittest value.

The first gene would be evaluated for the f possible genotype-phenotype maps, for a total of $2f$ evaluations. The second gene would be sampled with the remaining $f - 1$ unmapped fitness components, and so forth, giving a total of $f(f + 1)$ genotypes sampled. Each resulting fitness component would be the maximum of $2(f - i + 1)$ uniform i.i.d. values, for the ith gene/gene map pair. So the ith fitness component would be distributed as $F_i(\hat{\phi}) = \hat{\phi}^{2(f-i+1)}$. The

fitness components would have expectation

$$\mathrm{E}(\hat{\phi}_i) = \frac{2(f+1-i)}{2(f+1-i)+1} \ .$$

The expected value for the fitness peaks obtained through this constructional selection process would be

$$\mathrm{E}[w(\hat{\boldsymbol{x}})] = \frac{1}{f}\sum_{i=1}^{f}\frac{2i}{2i+1} \ .$$

For $n = f = 31$, the expected fitness peak would be $\mathrm{E}[w(\hat{\boldsymbol{x}})] \approx 0.945$, and would take $31 \times 32 = 992$ genotypes to find. So when lineage selection and constructional selection are both compared with $k = 1$ landscapes, the levels of adaptation achieved with constructional selection are seen to be vastly greater.

6 Discussion

6.1 Overview of Results

The goal of this chapter is to introduce the idea of "constructional" selection as a description of how the evolutionary acquisition of new genes can produce a genome better able to generate adaptive variants. In the Introduction I sketched out the basic conceptual framework of the idea, the two main parts being the genic selection effect (type I) and the correlated allelic variation effect (type II). The genic selection effect was based on the idea of defining viability, fecundity, and heritability differences at the level of the genome-as-population. I described how characteristics of the genotype-phenotype map, in particular Bonner's low pleiotropy principle, would lead to predicted differences in gene fecundity for giving rise to new, useful genes, and how this in turn would filter genome growth in the direction of lower pleiotropy.

Three models were provided to give concrete illustrations of constructional selection effects. The genic selection effect was illustrated with a simple model in which genes are ascribed "constructional" fitnesses — the probability that duplications of them are useful genes. The result is exponential growth in the genome of genes better able to spawn new genes. The correlated allelic variation effect was illustrated with some concrete examples of genotype-phenotype map functions, Wagner's linear quantitative-genetic map with Gaussian selection, and the epistatic NK fitness landscape of Kauffman. The NK model, because of the discreteness of the different organismal functions, provided a good example of how the level of pleiotropy of a new gene's effects affects the probability that it is selectively beneficial.

One general implication of constructional selection is that the genotype-phenotype map ought to be less complex than one might suppose. In other words, to some degree "bean bag genetics" might not be entirely wrong, at least for more recently evolved genes. One can also expect that there would be an

attunement (Barwise and Perry 1983) between the dimensions of recurrent environmental variation and the dimensions of genetic variation, in that recurrent, environmentally caused shifts of optimal phenotypes along certain phenotypic dimensions would expose the lineage to repeated directional selection along the same phenotypic axes; this would create the potential for the evolution of genes with phenotypic effects along these dimensions.

In the simulation models here, because the selection functions do not change during evolution, once a gene is incorporated in the genome, it is always deleterious to delete it. Examples are accumulating of genes that have been lost in the course of evolution (Brakenhoff, *et al.* 1990, Nishikimi *et al.* 1992, Wu *et al.* 1992), so clearly gene loss is a possibility, but the degree to which genes turn over in the genome is not known yet. The addition of gene loss to the models here would not prevent the constructional selection process; in fact, systematic differences in the rate that different genes are lost would also contribute to constructional selection as viability differences on the level of genome-as-population.

6.2 Empirical Phenomena

The main empirical predictions that come out of the processes discussed here are that:

1. There ought to be signatures of differential gene fecundity within the genome, and these should relate to way the gene duplications and/or allelic variation maps to the phenotype;
2. There ought to be dimensions of variation within the genome with low pleiotropy, affecting a relatively small suite of organismal functions; and
3. More recently evolved functions ought to have the least pleiotropic genetic control.

The advent of intra-genomic gene trees makes it foreseeable that some of these predictions could be tested. Observations on allelic variation by itself, without knowledge of the lineage of the gene, could be manifestations of other evolutionary processes besides constructional selection, and present methodological difficulties. Also, the prediction of low pleiotropy requires a basis for comparison in order to be tested, i.e. null hypotheses about levels of pleiotropy. And pleiotropy itself is a slippery concept, because the discretization of organismal traits is primarily an observer artifact; what is required is a quantification of functional relationships.

Genic Selection Effect. Numerous examples can be pointed to of genetic elements with specific functions that appear to have proliferated in the genome. Promoter sequences provide one example. A comparison can be made between transcription promoters that are external to the transcribed sequence, and internal promoters, whose sequence is part of the transcribed gene. Whereas internal promoters (which use RNA polymerase III) cannot be recombined with other

genes without large pleiotropic effects (Shi and Tyler 1991), external promoters can routinely be recombined with other peptide coding sequences, with the promoter retaining its regulatory properties and the peptide retaining its functional properties. External promoters are ubiquitous (i.e. they evidently have high constructional fitness), while internal promoters are restricted mainly to rRNA, tRNA, and snRNA genes and appear to be of ancient origin.

Signal peptides may be another example of a low-pleiotropy module that has proliferated. But because the constraints on the amino acids of signal sequences are rather broad, accurate intragenomic phylogenies are difficult to be certain of. Multi-gene families are examples of sequences that proliferate because of their ability to produce new useful variants, and their parts and subparts, as in the case of serine proteases, tend to have very specific functions that are retained in their different combinations (Doolittle 1985). The immunoglobulins are a spectacular example of genes of specific function which when duplicated produce offspring genes with a very high likelihood of being selectively advantageous.

There are several ways that the genic selection effect may have left a mark on multi-gene families, which I give with some suggestive anecdotal examples. These predictions apply to those gene families that are highly diversified (rather than multiple-copy gene families which have a very different selection and transmission dynamic due to unequal crossing over and gene conversion):

1. Gene families should show periods of exponential growth, possibly followed by logistic-like stasis as the genes saturate the available adaptive opportunities, depart from their original effects on organismal function, or become functionally burdened (e.g. the vertebrate Wnt developmental gene family (Sidow 1992); antennapedia-class vertebrate homeobox genes (Kappen *et al.* 1989));
2. During periods of exponential gene family growth, adjacent branches in the gene tree should show correlations in the time intervals between gene origins, producing acceleration in the branching rates in fecund branches; i.e. most new genes should come from genes that are themselves new;
3. Gene families that are in the process of expanding should continue to do so in independent lineages after taxon branching (e.g. neurofilament proteins in fish (Mencarelli *et al.* 1991)).

Exon Shuffling. The genic selection effect offers some explanatory clarity to questions about exon shuffling. Gilbert (1978) proposed that the characteristic exon/intron mosaic structure eukaryotic genomes existed so as to "speed evolution" by the creation of new genes through exon shuffling. But Crick (1979) criticized this evolutionary reasoning as being non-Darwinian, because it appeared to rely on "evolutionary foresight" in the genome — structures existing for their future evolutionary potential. Blake (1978) instead proposed that exons were descendents of the original "proto-genes", later assembled into complex, multi-exon proteins. Others, however, have proposed that introns were inserted later into contiguous genes through a transposition process. Subsequently, the debate on the exon-shuffling hypothesis has focused on a number of issues:

- Whether introns arose "early" or "late" — i.e. were present at the origin of eukaryotic genes or were inserted later into contiguous genes, possibly through a transposition process;
- Whether exons correspond to units of peptide structure or function, i.e. whether exons are "modular";
- Whether protein evolution through exon shuffling has indeed occurred;
- Whether selection could create a correspondence between exons and protein structures or whether exons would need to be descendents of the original "proto-genes".

Proving a correspondence between intron position and peptide structure has been seen as crucial for answering whether gene evolution through exon shuffling has occurred, and whether introns arose "early" or "late". Evidence has been marshalled both in favor (e.g. Gilbert 1993) and against (Stoltzfus 1994) there being a significant correspondence between exon structure and protein structure in eukaryotic genes.

Once it is understood that constructional selection would enter into exon shuffling dynamics, however, the question of when exons arose early or late becomes decoupled from the phenomenon of exon modularity. The genic selection effect provides a Darwinian mechanism for the evolution of modular exons through exon shuffling (Altenberg 1985). Exon modularity is the equivalent of low pleiotropy on the molecular level, so more modular exons would be expected to have a better chance of producing useful variation when recombined with other genes. Even a genome composed of randomly partitioned exons would come to be populated by modular exons if enough genome growth had occurred to allow the differential proliferation of exons, i.e. the genic selection effect (Altenberg and Brutlag 1986). Moreover, the evolutionary increase in modular exons needn't be relegated to the distant past, but would be occurring presently in any evolution of new genes through exon shuffling. This and other hypotheses for the evolution of split genes are reviewed in Doolittle (1987).

Second, the genic selection effect clarifies the feature of exons that would help them proliferate in the genome and speed evolution. What matters fundamentally is not that introns fall between structural elements of the peptide, but that each exon be able to maintain, within a new peptide environment, the properties that it was selected for. It has been assumed that the latter would require the former, but this assumption needs to be justified. Therefore, the negative statistical results carried out by Stoltzfus (1994), based on where introns fall in the peptide structure, might not be measuring whether exons have modular properties under exon shuffling. The more definitive test is to perform experimental manipulation of gene or peptide structure showing functional autonomy of the product of the exon or set of exons (e.g Craik 1980, Sanctis 1986, Zonneveld 1986, de Vries 1988, and Casorati 1993). Modularity of exon function has been found in many but not all cases.

A testable prediction I proposed as to whether exons with modular properties may have proliferated in the genome was to examine the reading frame statistics of exons (Altenberg 1983). Exons or group of exons that were a multiple of 3

nucleotides long would have greater modularity, because insertion of such exons into a protein-coding gene does not shift the reading frame down-stream. Such exons would have a constructional fitness advantage under exon shuffling. Data confirming this prediction were presented (Altenberg 1985, Altenberg and Brutlag 1986) showing statistical excesses of exons and pairs of exons with lengths a multiple of three. Subsequently, Patthy (1987) proposed what is also a constructional selection theory for exon shuffling, and also made similar predictions about exon reading frame properties. Smith (1988) and Gelfand (1992) have both corroborated these statistical findings on exon reading frame lengths.

Selection has been proposed as a possible cause of modular exons, but without sufficient attention to the level at which selection would have to act. The genic selection effect clarifies the means by which selection could produce modular exons. Doolittle (1985) had proposed that modular exons should be prevalent because "introns that occur between potentially useful domains will have added survival value". But one must ask, survival value for whom? It cannot be the survival of the organism carrying the intron, because intron position generally does not affect organismal viability. It could be perhaps the long term survival of the intron within the gene, if rates of introns loss were found to correlate with protein structure. In terms of the genic selection effect, intron survival within a gene corresponds to *viability* on the level of genome-as-population. If there were differences in intron longevity based on peptide position, this could produce a correspondence between intron position and peptide structure. But the differential proliferation of modular exons within the genome is a matter of differential survival, but differential *fecundity* on the level of genome-as-population.

Dissociability in Development and Morphological Integration. Dissociability in development and morphological integration are two aspects of the correlation structure of phenotypic variability. Morphological integration is said to be present where morphological characters which are functionally interdependent are also genetically correlated. Dissociability is where one such suite of functionally related characters has variability independent from another such suite. It is a form of low pleiotropy in developmental processes (defined for phenotypic as well as genetic perturbations), in that the development of certain structures can be changed without altering the development of other structures.

Both phenomena are predicted outcomes of constructional selection. But both can come about from modifier evolution as well, in which evolution under organismal selection at one locus systematically changes the genotype-phenotype maps at other loci, which is a form of epistasis (see Sect. 6.3, below). So evidence of their existence cannot alone be taken as support for an impact of constructional selection processes.

Cheverud (1984) has hypothesized modifier evolution under stabilizing selection as a primary mechanism producing morphological integration. The argument is principally one of genetic load: modifiers should evolve to reduce the genetic variance for the phenotypic dimensions under the most severe stabilizing selection. Wagner (1988) notes, however, that for populations in mutation-

selection balance around a fitness peak, the strength of selection on modifiers of the genotype-phenotype map can be only on the order of the mutation rate. Wagner has consequently looked to situations in which the population is not near equilibrium, but is at an earlier stage of directional selection, where there can be stronger selection on modifiers. He notes that rather special ecological conditions may be needed, however, to keep a population continually far from equilibrium, limiting the ubiquity of this mechanism for generating morphological integration.

This difficulty is overcome by recalling, from Sect. 4.4, that pleiotropic constraints can be expected to leave the population in a state of "latent" directional selection that can provide strong selective advantage to modifiers, or newly created genes, that break or shift the constraints in the right way. This is the context for Riedl's (1977) constructional selection mechanism. Therefore, morphological integration could be the result not of ongoing stabilizing selection about a fitness peak, but the legacy of changes in the genotype-phenotype map driven by the presence of latent directional selection for the morphological function. If the origin of the morphological adaptation involved the creation of new genes, then morphological integration could reflect the suite of phenotypic correlations that originally gave these genes their selective advantage (the correlated allelic variation effect). This would require a certain degree of stability in the correlation structures over evolutionary time.

One of the means to test for morphological integration is to examine the eigenvalues of the genetic correlation matrices for quantitative traits. Wagner (1984) pointed out that conclusions about the significance of the eigenvalues requires a null hypothesis. Wagner investigated of the eigenvalue distributions for random genotype-phenotype maps as a null hypothesis, and this was extended to other statistics on genetic correlation matrices (Cheverud *et al.* 1989). Significant departures in real data from the random expectation were found. The genotype-phenotype maps evolved here in simulations of genome growth can be used to derive genetic correlation matrices and thus yield expectations from the action of constructional selection that can be compared with quantitative genetic data.

Patterns of dissociability are ubiquitous in development, but little quantitative theory for the origins or maintenance of dissociability has been developed. Rieppel (1991) proposes that phylogenetically successful taxa are those which have been able to achieve greater dissociability in their ontogenetic systems, using snakes as his example. This is exactly what is expected from constructional selection acting during genome growth, and what may also result from the evolution of modifiers of the genotype-phenotype map.

Much of the thinking about dissociability places it as an aspect of von Baer's Laws (1828), that dissociable developmental pathways are those added recently in evolution to later stages of ontogeny. A view of developmental mechanisms as causal cascades gives rise to this view (Riedl 1977, Schank and Wimsatt 1987, Wimsatt and Schank 1988), but it should not be forgotten that there is variation for developmental mechanisms and that what emerges is filtered by selection. Terminal additions in ontogeny may be prevalent because on the average they

are less likely to be pleiotropic. But if pleiotropic cascading effects constrain the evolutionary malleability of earlier ontogeny, then there may exist large degrees of latent directional selection which would drive the evolution of new dimensions of genetic variability that produced dissociability. An example is the evolution of imaginal disks in Drosophila, a highly derived trait which effectively decouples larval and adult morphology. Variation in larval morphology, as long as it doesn't impinge on the imaginal disks, has little "generative" consequence for adult functioning, thereby unburdening the larval form. Imaginal disks thus were an invention that reduced pleiotropic constraints within Drosophila development.

Raff and coworkers (Raff 1992, Raff *et al.* 1992) have demonstrated especially well with sea urchins that divergent early development does not necessitate divergent adult forms, so that cascading effects of perturbations to early development must be seen as contingent results of evolution. Other examples include the frog *Gastrotheca* and the clam *Unio* (Levinton 1988, del Pino and Elinson 1983). This supports a view of the evolution of the genotype-phenotype map in which constraints on early development are not a mechanistic necessity, but are always unstable to new dimensions of variation that compartmentalize the genetic underpinnings for different adaptive functions.

Allelic Polymorphisms. If constructional selection has indeed produced genomes with a prevalence of genes with low pleiotropy, this might be expected to be evident in the phenotypic nature of allelic polymorphisms. Koehn *et al.* (1983) surveyed several cases of enzyme polymorphisms to get a sense of the levels of pleiotropy that typically exist. Their evaluation was that natural polymorphisms have low levels of pleiotropy when compared with the possibilities that exist for wide functional effects. The genes studied may not be reflective of genes in general because the nature of their pleiotropy may influence whether the genes maintain polymorphisms. Gimelfarb (1986, 1992) and Hastings and Hom (1989, 1990) have shown in the case of linear genotype-phenotype maps and stabilizing selection (as in Sect. 4), how the degree of pleiotropy can be critical to the number of loci at which polymorphisms can be maintained. In these quantitative genetic models, higher pleiotropy allows for greater polymorphism, which would strengthen the significance of Koehn *et al.*'s observations. Further theoretical study is needed in this area before natural genetic polymorphisms can be interpreted as evidence with respect to the genotype-phenotype map, and therefore constructional selection.

Macroevolutionary Dynamics. The creation of new genes may represent only a tiny fraction of the genetic events that contribute to adaptation, yet they may play a significant role in the sculpting of the genotype-phenotype map. All that is required is that the mode of action of the gene on the phenotype be somewhat conserved over macroevolutionary time scales. Still, however, one should not overlook the possibility that the evolution of new genes may often have a profound effect on the rates and direction of evolution. Several cases have been found where gene duplication was followed by accelerated rates of allelic

substitution (Li 1985). Because changed selection regimes would be expected to increase adaptive opportunities for the evolution of new genes, and because new genes can open up new dimensions of adaptive variation, one might expect to find associations between the origin of new genes and the origin of new taxa. This should be statistical testable once sufficient quantities of gene tree data accumulate.

In discussions of macroevolutionary dynamics, Eldredge (1989) points out that since Wright (1932) introduced the notion of the "adaptive landscape", adaptive change has been seen as either the tracking of moving adaptive peaks, or shifts from one adaptive peak to another. The dynamics considered here are another kind of adaptive change, in which new dimensions of variability in the phenotype are created, and what appeared to be an adaptive peak is now revealed to be a "slice" through the side of a peak of higher phenotypic dimensions. The organismal change afforded by new dimensions of variability may be incremental or profound, depending on how well the new variability allows decoupling of conflicting pleiotropic constraints and progress toward new adaptive optima. But the potential at least exists that certain "punctuations" or "saltational" changes during phylogeny reflect rapid climbing of pre-existing adaptive peaks through the introduction of new degrees of genetic freedom.

6.3 The Evolution of Evolvability

The main significance of constructional selection is that it is a mechanism that can apply more or less to all genes, with the effect of enhancing the ability of the genome to generate adaptive variants, and the effect of extending the genotype-phenotype map in the direction of lower pleiotropy. As such, it is an anagenetic mechanism that can enhance the genome's evolvability. Several other mechanisms that have been proposed for the evolution of evolvability are reviewed below.

Lineage Selection. Dawkins (1989) has discussed a mechanism for the evolution of evolvability that has perhaps made inroads into making the "evolution of evolvability" more discussible in evolutionary research (Arnold, *et al.* 1989, Alberch 1991). Dawkins's mechanism is lineage selection. In lineage selection, organisms whose genotype-phenotype map by happenstance makes them evolvable — i.e. better able to generate adaptive variants — are the ones whose lineages would have most proliferated and endured. Thus, even though there would never be selection for evolvability *within* a lineage (consonant with Concept 3), most of the species we see would have high evolvability.

Dawkins proposes lineage selection for the prevalence of species with evolvable developmental mechanisms; Doolittle proposed lineage selection for the evolvability-enhancing property of introns in protein evolution (Doolittle 1987); lineage selection has also been proposed for the evolution of sex (Stanley 1976, Aboitiz 1991). However, lineage selection must still turn to chance as an explanation of why certain genomes came to be more evolvable; it cannot produce an

increase in the evolvability of the genome *within* lineages. Genetic modification and constructional selection are mechanisms that can change evolvability within lineages.

Genetic Modification. Modification, as mentioned earlier, is a form of epistasis, in which the nature of the phenotypic variability determined by one locus is affected by the allelic state at another locus, the modifier.

The most widely discussed idea for the evolution of evolvability through genetic modification is "regulation" in development, otherwise called canalization (Waddington 1957), developmental homeostasis (Lerner 1954), morphogenetic correlations (Schmalhausen 1949), or morphological integration (Olson and Miller 1958, Cheverud 1984). In this mechanism, genes are selected on for their organismal fitness effects but modify the variational properties of the genome as a systematic side effect. Most discussions of this mechanism do not explicitly describe it as a modifier effect. Selection to stabilize morphological functions against environmental and genetic variability can systematically lead to the reduction of pleiotropic effects from genetic background variability. Therefore, this may endow the developmental system with "extrapolation" capabilities if it can produce the morphological function in the face of evolutionary changes in other parts of the organism (Frazzetta 1975). Kauffman argues that the properties of dynamical systems would produce such a systematic correlation between phenotypic stability, which is selected for, and smooth adaptive landscapes, which are not directly selected for (Kauffman 1989b).

Riedl, in his theory of 'genome systemization", adds a modifier effect to the evolution of "superimposed" regulatory genes, through an unnecessary assumption about the nature of these genes. The process Riedl proposes is constructional selection, because new genes evolve that produce coordinated variation in the right direction for adaptation. Existing genes are unable to produce the adaptation because it would require the simultaneous mutation of several of them. This creates the situation of latent directional selection. But Riedl emphasizes that the regulatory genes will also eliminate the previously existing uncoordinated dimensions of variability. This is the basis of his argument for hierarchies of constraint in the phenotype. Yet it is not the suppression of uncoordinated variability that allows a new superimposed gene to survive. It is its ability to produce variation in the direction of adaptive opportunity. The former effect would produce a reduction in genetic load, which is another example of the genetic modification described above. Although the evolution of new genes could involve both constructional selection and genetic modification effects, these are not inherently linked.

One additional modifier mechanism is the idea of hitchhiking, which Conrad (1979 1982) proposes can evolve smoother adaptive landscapes, and Wagner(1981) proposes can accelerate responses to selection. A near-neutral modifier allele that increases the chance that mutations at another locus are adaptive can hitchhike along with these mutations. Conrad provides no population genetic analysis of the hitchhiking model, but it is plausible based on the model of Eshel

(1973) for modification of mutation rates. Modifiers that alter the adaptive landscape, however, would in general be expected to have direct effects on fitness, either advantageous or deleterious, which would swamp any hitchhiking effects on the modifier evolution. Wagner also proposed what is effectively a hitchhiking mechanism for the evolution of increased rates of adaptation in his idea of "feedback" selection (Wagner 1981). Wagner considers the general situation where a neutral modifier can evolve if it increases the rate at which selection increases the fitness of the modifier's carriers.

Evolvability and the Randomness of Mutation. There is often a Darwinian hesitation in discussing the evolution of evolvability, because it seems to step outside of what natural selection can act upon, or invoke non-random mutational processes. To claim that the evolution of new genes should enhance the genome's ability to produce adaptive variants is, on the surface, contrary to the idea that mutation is random with respect to adaptation. But if one looks more closely at these notions of randomness, one finds three different concepts:

Concept 1: Mutation pressure by itself will not produce adaptive evolution.
Concept 2: Current selective pressures do not affect the direction of mutations, with respect to those selective pressures.
Concept 3: The ability of the genome to generate adaptive variants is not molded in any systematic way by its evolutionary history.

Neither Concepts 1 nor 2 are at issue with constructional selection. Concept 2, I should note, has been the center of the controversial claim of "directed mutation" in bacteria (Cairns *et al.* 1988, Lenski *et al.* 1989, Hall *et al.* 1990, Cairns and Foster 1991, Foster and Cairns 1992, and Mittler and Lenski 1992). The mechanism of lineage selection that Dawkins (1988) proposed for the evolution of evolvability is in keeping with Concept 3. It is Concept 3, however, with which I have taken issue here. Concept 3 is well exemplified by Maynard Smith, *et al.* (1985):

> Furthermore, there is usually no reason to suppose that the developmental mechanisms in question evolved because of the particular phenotypes that they make readily accessible. In general, therefore, the direction of the resulting constraints (biases on the production of variant phenotypes) is "accidental" or "random" with respect to the demands of adaptive evolution. (1985, p. 269)

The argument of this chapter could be boiled down simply to this: the proposition in the first sentence of this quote does not logically entail the assertion in the second sentence. In other words, while it is reasonable and Darwinian to claim that developmental mechanisms do not evolve *because of* the phenotypes they make readily accessible, the resulting accessible phenotypes will not be "accidental" with respect to the demands of adaptive evolution if they are correlated with the phenotypes originally made accessible by the developmental mechanisms.

6.4 Future Directions

The models described here have been designed to be simple and illustrative of the constructional selection effects. There are numerous refinements and elaborations one could make to the genome growth models, including allelic polymorphism, changing selection and coevolution, stochastic mixing of gene duplication and allelic mutation events, and finite population size. Other models for genotype-phenotype maps can be analyzed for their evolution under constructional selection. I suspect that the basic findings about the evolution of pleiotropy will be robust under these elaborations, and further phenomena may emerge as well.

Models specific to some of the predictions made here need to be developed, such as statistics of gene-tree topologies under the genic selection effect. Riedl's idea of burden, while not directly dealt with in the models here, may be incorporated with minor modifications.

Moreover, the model genotype-phenotype maps evolved under constructional selection can be utilized in providing underlying models for what the effects of genetic constraints on evolution should look like. Theories about how constraints affect morphological evolution and cladistics can be concretely simulated with such model maps.

Levinton (1988) writes,

> Evolutionary biologists have been mainly concerned with the *fate of variability* in populations, not the *generation of variability*. ... Whatever the reason, the time has come to reemphasize the study of the origin of variation.

Levinton's call is certainly heeded in this chapter, which has attempted to provide a framework for thinking about the evolutionary forces acting on the generation of variability, and to describe new mechanisms which enable the evolvability of the genome to evolve.

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