Heterozygote advantage as a natural consequence of adaptation in diploids

Diamantis Sellis*, Benjamin J. Callahanb, Dmitri A. Petrova,1 and Philipp W. Messera,1

Departments of *Biology and bApplied Physics, Stanford University, Stanford, CA 94305

Edited by Boris I. Shraiman, University of California, Santa Barbara, CA, and approved November 2, 2011 (received for review September 7, 2011)

Molecular adaptation is typically assumed to proceed by sequential fixation of beneficial mutations. In diploids, this picture presupposes that for most adaptive mutations, the homozygotes have a higher fitness than the heterozygotes. Here, we show that contrary to this expectation, a substantial proportion of adaptive mutations should display heterozygote advantage. This feature of adaptation in diploids emerges naturally from the primary importance of the fitness of heterozygotes for the invasion of new adaptive mutations. We formalize this result in the framework of Fisher's influential geometric model of adaptation. We find that in diploids, adaptation should often proceed through a succession of short-lived balanced states that maintain substantially higher levels of phenotypic and fitness variation in the population compared with classic adaptive walks. In fast-changing environments, this variation produces a diversity advantage that allows diploids to remain better adapted compared with haploids despite the disadvantage associated with the presence of unfit homozygotes. The short-lived balanced states arising during adaptive walks should be mostly invisible to current scans for long-term balancing selection. Instead, they should leave signatures of incomplete selective sweeps, which do appear to be common in many species. Our results also raise the possibility that balancing selection, as a natural consequence of the fitness of heterozygotes for the invasion of new adaptive mutations, might play a more prominent role among the forces maintaining genetic variation than is commonly recognized.

Adaptation by natural selection is the key process responsible for the fit between organisms and their environments. The invasion of new adaptive mutations is an essential component of this process that fundamentally differs between haploid and diploid populations. In diploids, while a new mutation is still rare, natural selection acts primarily on the mutant heterozygote (1). As a result, only those adaptive mutations that confer a fitness advantage as heterozygotes have an appreciable chance of invading the population (“Haldane’s sieve”). However, if the heterozygote of an invading adaptive mutation is fitter than the mutant homozygote, the mutation will not be driven to fixation but, instead, maintained at an intermediate, balanced frequency.

We argue that heterozygote advantage should be very common during adaptation in diploids if selection is stabilizing and at least some mutations are large enough to overshoot the optimum. Consider, for example, adaptation through changes in gene expression (Fig. 1A), which is an important, if not the dominant, mechanism of adaptation (2). Here, mutations of small effect (Fig. 1B) will be adaptive when they modify expression in the adaptive direction whether the organism is haploid or diploid. However, mutations of large effect (Fig. 1C) can be nonadaptive in haploids, because they overshoot the optimum, yet be adaptive in diploids, because their phenotypic effect is moderated when heterozygous. These mutations will have heterozygote advantage and lead to balancing selection in diploids.

The components of the simple model in Fig. 1, stabilizing selection and availability of mutations of various sizes, are well established by empirical data in the case of gene expression. Pervasive stabilizing selection is indicated by the lack of large gene expression differences between and within species despite the abundance of mutations that change gene expression (3–7). In addition, expression-altering mutations are known to come in a variety of sizes ranging from subtle changes to dramatic ones of tens of fold or even hundreds of fold (4, 5, 8).

The model in Fig. 1, albeit instructive, may lack generality because it incorporates only a single trait. However, adaptation might often involve mutations that have complex pleiotropic effects in an effectively multidimensional phenotypic space. The classic model that incorporates this key feature of adaptation is Fisher’s geometric model (9–13). In this single-locus model, phenotypes are vectors in an abstract geometric space (Fig. 2A), with the orthogonal axes representing different, independent traits, such as color and height. Mutations move phenotypes in a random direction, with the size of the step corresponding to the effect size of the mutation. The fitness landscape is peaked around a single optimal phenotype, capturing our intuition of stabilizing selection. Below, we extend Fisher’s geometric model to diploidy and confirm the intuition from Fig. 1 that adaptation should generically lead to heterozygote advantage.

Results

Consider an allele a with homoyzgous phenotype r_{aa} in Fisher’s model. Mutations are modeled by adding a mutation vector m to the phenotype of the mutated allele (Fig. 2A). The direction of the mutation vector is chosen uniformly, and its size (m) is drawn from a specified distribution P(m). In diploids, the phenotype is determined by its two constituent alleles. The geometric nature of Fisher’s model offers a straightforward mapping between the phenotype of the mutant heterozygote (r_{ab}) and the homozygote phenotypes (r_{aa} and r_{bb} = r_{aa} + m) in terms of the weighted average: r_{ab} = r_{aa} + h m, where the weight h specifies the phenotypic dominance of the new mutation. For convenience, we assign the phenotype of a diploid homozygote to be equal to that of a haploid carrying the same allele (i.e., r_{bb} = r_b).

Fitness w(r) decreases with the distance between the organismal phenotype and the phenotypic optimum. In haploids, adaptive mutations, w(r_{ab}) > w(r_{aa}), are those that bring the mutant allele closer to the optimum (i.e., the mutant falls inside the sphere r_{opt} of radius r_c centered at the optimum) (Fig. 2A). In diploids, it is the fitness of the mutant heterozygote r_{ab} that primarily determines the probability of successful invasion; therefore, we define the area in which mutations are adaptive in diploids (r_{opt}) by the condition w(r_{ab}) > w(r_{aa}).

Let us first focus on the instructive case of strict phenotypic codominance, h = 1/2. Here, mutations in diploids have half the initial phenotypic effect that they would have in a haploid.

*Author contributions: D.A.P. and P.W.M. designed research; D.S., B.J.C., and P.W.M. performed research; D.S., B.J.C., and P.W.M. contributed new reagents/analytic tools; D.S., B.J.C., D.A.P., and P.W.M. analyzed data; and D.S., B.J.C., D.A.P., and P.W.M. wrote the paper.

The authors declare no conflict of interest.

1To whom correspondence may be addressed. E-mail: dpetrov@stanford.edu or messer@stanford.edu.

This article contains supporting information online at www.pnas.org/lookup/suppl/doi:10.1073/pnas.1114573108/-/DCSupplemental.
Mutations of small effect thus have reduced initial selective effects in diploids compared with in haploids, but the area in which mutations are adaptive in diploids (\(\alpha_{\text{dip}}\)) is significantly enlarged (Fig. 2A). If the supply of mutations is restricted to those of small size compared with the distance to the optimum, adaptive mutations will invade a diploid population at a slower rate than they would a haploid population because of the reduced initial selection (SI Text and Figs. S1 and S2). In contrast, once mutations of large enough size are available, diploids start reaping the benefits of the larger space of adaptive mutants available to them. In this case, adaptive mutations will invade at a higher rate in diploids than in haploids, even after controlling for the difference in mutation rate that results from a diploid population having twice as many chromosomes as a haploid population of equal size (14).

The larger range of adaptive mutations available to diploids comes with a catch, however; many of these adaptive mutations display heterozygote advantage, and thus will not simply go to fixation. The fraction \(\delta_{\text{a}}\) of adaptive mutations with heterozygote advantage \(\left[ w(r_{\text{ab}}) = w(r_{\text{aa}}) < w(r_{\text{bb}}) \right]\) in Fisher’s model is the fraction of mutations that fall into the sphere \(\alpha_{\text{dip}}\left(w(r_{\text{ab}}) < w(r_{\text{aa}}) < w(r_{\text{bb}})\right)\) but not the sphere \(\left[ w(r_{\text{ab}}) > w(r_{\text{bb}}) > w(r_{\text{aa}})\right]\) (Fig. 2A). In the small-mutation limit, \(\delta_{\text{a}}\) goes to zero because the nonoverlapping space between \(\alpha_{\text{dip}}\) and \(\gamma\) is not reachable by mutations that fall in the immediate neighborhood of \(r_{\gamma}\). In the limit in which mutations of all sizes are equally likely, however, most adaptive mutations show heterozygote advantage \(\left(\delta_{\text{a}} \gtrsim 1/2\right)\), except for the special cases of perfect dominance or perfect recessiveness (SI Text).

The crucial criterion determining the probability of heterozygote advantage during adaptation in diploids is the availability of “large” mutations. In Fisher’s model, for heterozygote advantage to be common, the average size of mutations \(\langle m \rangle\) has to be at least of order \(r_{\text{max}}/d\), the distance to the optimum divided by the square root of the number of phenotypic dimensions (SI Text). However, even when mutations are initially small compared with this distance, during an adaptive walk, a population gradually approaches a fitness optimum via successive adaptations (11). Thus, at some point on the walk, this condition will be met; thereafter, heterozygote advantage will be likely.

We performed simulations of adaptive walks in Fisher’s model to test these theoretical predictions and further investigate the consequences of frequent heterozygote advantage in such walks. Specifically, we simulated a single locus in a 2D phenotypic space under phenotypic codominance in a Wright–Fisher framework using an exponential distribution of mutation sizes and a symmetrical Gaussian fitness landscape (Materials and Methods).

Fig. 1. Adaptation to a change in the optimal level of gene expression. (A) In both haploids (hap) and diploids (dip), the wild type (wt) is perfectly adapted to the original fitness function (dashed black curve). After an external change, the optimal expression level becomes twice the original level (solid red curve). Note that we are assuming phenotypic codominance; thus, the two individual gene copies in a diploid each contribute expression level 0.5, such that overall expression is 1. (B) Fitness effects of a small mutation that increases expression level by 1.5-fold. The mutant heterozygote (het) is less fit than both the haploid mutant (mut) and the mutant homozygote (hom). (C) Effects of a large mutation that increases expression by threefold. In this case, the mutant heterozygote effectively has only twofold increased expression, and thus lands right at the new fitness optimum. In contrast, both the haploid mutant and the mutant homozygote “overshoot” the optimum.

We find that adaptive walks in diploids typically involve the succession of many intermediate balanced polymorphisms that tend to be ephemeral; they are quickly displaced by new adaptive alleles, themselves often displaying heterozygote advantage (Fig. 2 B and C). These dynamics contrast sharply to those in haploid populations, where adaptive walks proceed by successive sweeps of adaptive mutations and populations are generally monomorphic between sweeps (Fig. 2C).

We confirm that balanced states are likely during adaptive walks (Fig. S3), provided that mutation sizes meet our theoretical conditions and selection is strong enough to maintain balanced states despite the stochastic fluctuations arising from genetic drift (SI Text). Fig. 2D shows the probability of observing balanced states during adaptive walks under various parameter settings (Table S1). Note that the probability of observing balanced states correlates very strongly with the overall probability of observing successful adaptation. For example, in our scenarios where the population eventually traversed at least 90% of the initial fitness distance to the optimum, balanced states were observed in ≥90% of the runs and were present ≥40% of the time during walks.

Simulated haploid populations approach the optimum faster, on average, than simulated diploid populations despite diploid populations generally containing the most fit individual (Fig. S4). This is because diploid populations suffer from a high genetic load, which can be identified as the segregation load attributable to pervasive heterozygote advantage. The balanced polymorphisms that result from the invasion of adaptive mutations with heterozygote advantage also cause high levels of standing variance in both phenotype and fitness to persist during the adaptive walk (Fig. S4).

The maintenance of genetic variation via heterozygote advantage during adaptation is a striking difference between the adaptive walks in haploids and diploids. Although the load resulting from the maintained variation is costly during adaptation to a constant environment, could this effect be advantageous in a changing environment? Environmental changes that displace the phenotypic optimum can convert dominance variance of fitness into additive variance, which would then fuel adaptation. To investigate this possibility, we incorporated a randomly

Sellis et al.  December 20, 2011  vol. 108  no. 51  PNAS  20667
Fig. 2. Fisher’s geometric model of adaptation in two dimensions. (A) Two orthogonal axes represent independent character traits. Fitness is determined by a symmetrical Gaussian function centered at the origin. Consider a population initially monomorphic for the wild-type allele \( r_{\text{hap}} = (2,0) \). A mutation \( m \) gives rise to a mutant phenotype vector \( r_{\text{mut}} = r_{\text{hap}} + m \). The phenotype of the mutant heterozygote assuming phenotypic codominance (\( h = 1/2 \)) is \( r_{\text{mut}} = r_{\text{hap}} + m/2 \). The different circles specify the areas in which mutations are adaptive in haploids (\( r_{\text{hap}} \)), adaptive in diploids (\( r_{\text{dip}} \)), and replacing in diploids (\( r_{\text{dip}}^\ast \)). (B) Frequency trajectories of all alleles present during a representative adaptive walk in a diploid population with \( N = 5 \times 10^6, r_{\text{hap}} = (2,0) \), and \( <m> = r_{\text{mut}} = 1 \). Different colors represent different alleles. The black bars over the graph indicate the periods during which a balanced polymorphism was present. (C) Representative adaptive walks in a haploid population and a diploid population. Vectors depict the successive mutations that led to the prevalent allele at the end of the walk. The haploid walk consists of a single lineage of successive mutations, each conferring a selective advantage over the previous one. In the diploid walk, the first mutation overhats the fitness optimum, generating a sequence of intermediate balanced states. Note that the areas \( r_{\text{hap}} \) and \( r_{\text{dip}} \) from panel A apply only to the first mutation in the walk when the population is still monomorphic for \( r_{\text{hap}} \). (D) Probability of observing balanced polymorphism during adaptive walks toward a fixed fitness optimum as a function of mutation sizes scaled by effective drift radius \( r_0 \) (SI Text) for the various settings of \( N, \sigma \), and \( <m> \) specified in Table S1. Circles show the probability of at least one balanced state arising over the course of a walk, and squares show the fraction of time during which balanced states were present. Coloration indicates the average “adaptedness” achieved during a walk, defined by the improvement in mean population fitness over the walk (\( <w_{\text{end}}> - <w_{\text{start}}><w_{\text{start}}> \)) relative to the maximally possible improvement (\( 1 - <w_{\text{start}}><w_{\text{start}}> \)).

Moving optimum into our simulations (Materials and Methods). The optimum moves every generation in a random direction with step sizes sampled from a Gaussian distribution, the variance of which (\( \sigma_{\text{env}}^{-2} \)) determines the speed of optimum movement.

Pervasive balanced polymorphism remains a feature of adaptation in diploid populations in the moving optimum scenario (typical trajectories are shown in Fig. S5). In simulations with a fast-moving optimum (\( \sigma_{\text{env}}^{-2} = 10^{-2} \)), polymorphisms at frequencies 0.05 < \( x < 0.95 \) are present around 30% of the time when averaged over replicate runs. More than 80% of these polymorphisms are balanced. Polymorphisms are observed less frequently (\( \sim 5\% \)) when environmental change becomes very slow (\( \sigma_{\text{env}}^{-2} = 10^{-3} \)), because populations are well adapted most of the time. The fraction of polymorphisms that are balanced, however, remains substantial (\( \sim 58\% \)) in this scenario. Table S2 shows the percentages of time during which polymorphisms were observed in our simulations and the fractions of those polymorphisms that were balanced for a wide range of values of \( \sigma_{\text{env}}^{-2} \). We also demonstrate that many of the balanced polymorphisms eventually fix such that a substantial proportion of substitutions (60–70%) pass through a balanced state (Fig. S6 and Table S2). These substitutions tend to be generated by larger phenotypic effect mutations than the substitutions that go to fixation without an intermediate balanced state (Fig. S6).

To evaluate how effectively populations followed the moving fitness optimum, we measured the mean population fitness averaged over a walk (\( <w> \)). The difference, \( \lambda = 1 - <w> \), is then the average lag in fitness between the population and the optimum. Fig. 3A shows the ratio \( \lambda_{\text{hap}}/\lambda_{\text{dip}} \) between haploid and diploid populations for different values of \( \sigma_{\text{env}}/\sigma_w \). In slow-changing environments (\( \sigma_{\text{env}}/\sigma_w < 10^{-2} \)), haploids follow the moving optimum more closely than diploids, replicating the constant environment result. However, in fast-changing environments (\( \sigma_{\text{env}}/\sigma_w > 10^{-4} \)), it is diploids that prevail.

The diversity advantage of diploid populations derives from the greater variation maintained during adaptation. Specifically, this greater variation should lead to an increased range of mutations starting from multiple balanced alleles as well as the ability of diploids to adapt by fast adjustment of the frequencies.
follow the moving optimum more closely than haploids (hap) and diploids (dip) as a function of the speed of environmental change. In fast-changing environments, diploid populations generate only limited variation in fitness (Fig. 3B) and phenotype (Fig. 3C). In contrast, in fast-changing environments, balanced alleles are much younger than comparably frequent neutral alleles. These young balanced alleles are often very distinct phenotypically, and thus produce high levels of standing variation in fitness (Fig. 3B) and phenotype (Fig. 3C).

**Discussion**

Our finding that heterozygote advantage emerges naturally in Fisher’s geometric model if mutations are sufficiently large is very robust to the details of the model, such as the number of dimensions; the choice of phenotypic dominance rules, including under-, over-, and incomplete dominance; mutation rate and size distribution; population size; and flatness of the fitness function (SI Text, Figs. S1–S3, S7, and Table S1).

The features that underlie pervasive heterozygote advantage in Fisher’s model are also likely to apply very generally in nature: (i) mutations in diploids should initially segregate as heterozygotes, (ii) selection should be stabilizing for some traits, and (iii) some invading mutations should be large enough to overshoot the local optimum. Indeed, rare mutations are generally heterozygous, except for cases of exceptionally strong inbreeding. It is also well established that stabilizing selection acts on many phenotypic traits (17, 18). Even if “more is always better” holds for certain traits, as long as adaptive mutations generally influence at least one trait under stabilizing selection, we still expect heterozygote advantage to be frequent. Finally, mutations of large phenotypic effect have been observed in many organisms, and it is becoming increasingly apparent that such large mutations do contribute to adaptation. For example, adaptive mutations of large effect have occurred in the domestication of maize (19) and dogs (20), the evolution of sticklebacks to fresh water habitats (21, 22), pesticide resistance in insects (8), coat color in mammals (23, 24), and several studies of experimental evolution (25, 26). Furthermore, analyses of mutation accumulation lines have demonstrated the availability of mutations causing multiple-fold changes in gene expression (4, 5). Because the expression of most genes is known to be constrained by stabilizing selection over much narrower ranges (3–7), we must conclude that gene expression mutations that overshoot the optimum are likely abundant.

The classic model of adaptation holds that adaption is driven by adaptive mutations that sweep quickly to fixation. Although our model also predicts such fast fixation events, it additionally predicts that many adaptive mutations will initially only sweep to intermediate frequencies, where they are then maintained for a period of time by balancing selection, before continuing on to either fixation or loss. Both models thus predict an elevated rate of fixation at functional sites compared with the neutral expectation and a local reduction of genetic diversity around adaptive sites, as have been observed in a range of organisms (27–30). However, in contrast to the classic model, we also expect the presence of many incomplete selective sweeps. Indeed, the genomic signatures of such incomplete sweeps appear to be plentiful in a number of organisms (31–33). Incomplete sweeps also appear to be common in experimental evolution in *Drosophila*, where virtually no classic sweeps have been detected after 600 generations of evolution despite abundant evidence of phenotypic adaptation over this period (34).

The abundance of incomplete sweeps in natural and experimental populations is consistent with but, unfortunately, not uniquely predictive of adaptation-driven balancing selection. Other scenarios, such as frequency-dependent selection, adaptation to specific subhabitats, and polygenic adaptation, also predict incomplete sweeps. The only way to test the hypothesis of heterozygote advantage explicitly is to measure fitness of the homozygotes and heterozygotes for the putatively balanced alleles directly. Such measurements are difficult but not impossible and can now be carried out systematically in laboratory systems of artificial selection, such as yeast or *Drosophila* (26, 34, 35).

---

Fig. 3. Statistics of adaptive walks under a moving fitness optimum. (A) Ratio of the average lag in fitness \( \lambda_{hap}/\lambda_{dip} \) between the population and the optimum in haploids (hap) and diploids (dip) as a function of the speed of environmental change. In fast-changing environments, diploid populations follow the moving optimum more closely than haploids \( \lambda_{hap}/\lambda_{dip} > 1 \). (B) Fitness variance attributable to balanced polymorphisms (frequency 0.05 < \( x < 0.95 \)) and the age of the balanced polymorphism for different values of \( \sigma_{env} \). Both quantities are estimated from the balanced polymorphisms that were present at the end of simulation runs. The age of a balanced polymorphism is defined as the time since the most recent common ancestor of its constituent alleles. Data points are medians over 10\(^3\) runs, and error bars specify the 10\% and 90\% quantiles. The gray-shaded area (0.04N < \( x < 4N \)) indicates the expected age range of common neutral polymorphisms at frequencies between 0.05 < \( x < 0.95 \). (C) Same as in B but phenotypic variance is shown instead of fitness variance.
However, in many other systems, direct fitness measurements are not feasible and balanced polymorphisms have to be identified by alternative approaches. The standard scans for balanced polymorphisms are inappropriate for our purposes because they typically search for very ancient balanced alleles (36–38), whereas the balanced alleles predicted by our model are often short-lived. To identify young balanced alleles specifically, one can search for polymorphisms that maintain their frequencies in the face of strong bottlenecks (39). A particularly powerful system in this context is provided by human genetics because of the multiple recent bottlenecks associated with human migrations. Other opportune systems are island species recovering from natural disasters or man-made relocations of species to new but environmentally similar locations, such as the relocation of *Euphydryas gillettii* from Wyoming to Colorado (40).

Adaptive evolution is generally thought to be antithetical to the maintenance of genetic variation. In contrast, in our model, pervasive adaptation systematically generates genetic variation by promoting balanced polymorphisms. These balanced polymorphisms are expected to segregate at high frequencies yet can affect both phenotype and fitness substantially. This is very different from the common view that frequent polymorphisms should be neutral or subject to only weak selective forces.

We argue that adaptation-driven balanced polymorphisms can be an important source of consequential genetic variation. In particular, we believe that the balanced polymorphisms predicted by our model can be associated with human disease. Some of the common disease variants could be mutations that are maintained at high population frequencies because of strong heterozygote advantage, although they are very harmful as homozygotes.

Balancing selection and, specifically, the prevalence of heterozygous advantage was once considered the dominant force maintaining variation in natural populations (41–43). This view fell out of favor with the rise of the neutral theory in the 1960s (10). The neutral theory postulates that only a small proportion of substitutions are adaptive and that these substitutions are fixed very quickly; this, in turn, implies that practically all polymorphisms should be either neutral or slightly deleterious. However, recent genomic evidence has suggested that the rate of adaptation is substantial in some organisms, with, for example, ~50% of all amino acid substitutions in *Drosophila* driven by positive selection (28). Here, we argue that such a high rate of adaptation in diploids should also lead to a high rate at which balanced polymorphisms are driven into the population. We thus localize the evolutionary theory of genetic variation should be given new life and reassessed using all the modern genomic tools at our disposal.

**Materials and Methods**

**Monte Carlo Simulations of a Single Adaptive Mutation in Fisher’s Model.** We investigate the evolution of a single locus in Fisher’s geometric model (9). Alleles are represented by vectors \( \mathbf{r} \) in an abstract, d-dimensional Euclidean phenotype space. Mutant alleles are obtained by adding a mutation vector \( \mathbf{m} \) to the parental allele: \( \mathbf{r}_u = \mathbf{r} + \mathbf{m} \). The directions of mutation vectors are distributed uniformly; mutation sizes \( \mathbf{m} \) are distributed according to a probability distribution \( P(\mathbf{m}) \) with an average mutation size \( \mathbf{c} \). We consider two such distributions in detail, the uniform distribution \( P(\mathbf{m}) = \frac{1}{2d} \mathbf{c} \) and the exponential distribution \( P(\mathbf{m}) \propto \exp(-\mathbf{c} \cdot \mathbf{m}) \). Haplod organisal phenotypes are equal to the allelic phenotype. In diploids, the phenotype is a weighted average of its two constituent alleles. In case of the heterozygote advantage, the phenotype: \( \mathbf{r}_u = \mathbf{r}_u + \mathbf{r}_u \).

For convenience, we set the scale of the space such that \( \mathbf{c} = 1 \). We begin by considering mutations arising in a population that is monomorphic for the wild-type allele: \( \mathbf{r}_u = \mathbf{r} \). The rates of at which adaptive mutations occur in diploids vs. haploids were estimated by counting the overall numbers of successfully invading mutations, the expectation values, \( \mathbf{r}_u \) and \( \mathbf{r}_u \), and \( \mathbf{r}_u \) and \( \mathbf{r}_u \), respectively. For the ratios \( \mathbf{r}_u \) and \( \mathbf{r}_u \), the expectations were then obtained by counting the fraction of adaptive mutations with heterozygote advantage in the diploid scenario. For \( \mathbf{r}_u \), each adaptive mutation was thereby additionally weighted by its respective invasion probability.

**Simulation of Adaptive Walks Toward a Fixed Fitness Optimum.** To investigate adaptive walks toward a fixed fitness optimum, we simulated the full stochastic population dynamics in the above scenario under an infinite alleles assumption. We focused on the instructive case of a 2D Fisher’s model with complete phenotypic codominance (\( h = 1 \)). The phenotype of a heterozygous diploid is then always the coordinate-wise average of its two alleles: \( \mathbf{r}_u = (\mathbf{r} + \mathbf{r}_u)/2 \).

Mutations are modeled by a Poisson process with rate \( \mu = 2.5 \times 10^{-4} \) per individual and generation. Mutation directions are drawn uniformly, and mutation sizes are sampled from an exponential distribution with mean \( \mathbf{c} = 2.5 \times 10^{-4} \). Population sizes are \( \mathbf{N}_{\text{hap}} = 10^5 \) for haploids and \( \mathbf{N}_{\text{hap}} = 5 \times 10^5 \) for diploids, ensuring that new mutations arise at equal overall rates in the two populations (\( \mathbf{N}_{\text{hap}} = 0.05 \), where \( c \) is ploidy).

For the state of the population at any given time point is specified by the set of alleles \( \mathbf{r} \) present in the population and their associated population frequencies \( f(x) \). Allele frequency dynamics are modeled in a Wright–Fisher framework with selection (44). For haploids, we use the standard Wright–Fisher sampling procedure in which an allele frequency \( x(t + 1) \) in the next generation are drawn from a multinomial distribution \( P(x(t + 1)) \) with selection-adjusted probabilities: \( P(x(t + 1)) = x(t) \cdot \mathbf{r}_u \cdot \mathbf{r}_u \). Allele frequencies \( x(t + 1) \) are then drawn from \( P(x(t + 1)) \). In both cases, \( x \) and \( \mathbf{r} \) are normalized such that \( \sum x = 1 \).

As specified above, simulations start from a population that is monomorphic for the wild type \( \mathbf{r}_u = (0,0) \) with the optimal phenotype located at the origin, yielding an initial population average fitness of \( \mathbf{r}_u \). Populations are then evolved for \( 10^5 \) generations, which typically suffices to approach the fitness optimum closely (\( \mathbf{r}_u > 0.96 \)) at end of a run (Fig. S4A).

**Simulations Under a Moving Fitness Optimum.** For the analysis of the moving optimum scenario, we adjust our simulation as follows. At the start of the simulation, the population is initialized to be monomorphic for the optimal phenotypes \( \mathbf{r}_u (t = 0) = \mathbf{r}_u \) and, at each generation, the optimum \( \mathbf{r}_u (t + 1) \) moves one step in a random direction and the size of the mutation is sampled from the positive half of a Gaussian distribution with variance \( \mathbf{r}_u^2 \). In a single simulation run, the population is evolved for \( 10^5 \) generations (~100N). We exclude the first \( 10^5 \) generations of each run from our analysis as a “burn-in” period so as to remove the influence of the initial state of the population.

**Ascertainment of Balanced Polymorphisms During Adaptive Walks.** Balanced polymorphisms can consist of several alleles (45, 46). We determine the presence of a balanced polymorphism at a given time point in our simulation runs using Kimura’s analytic conditions (47). Assume that \( n \) alleles \( r_1, r_2, \ldots \) are present in the population with diploid fitness values given by \( w(r) \). Let \( T \) be the matrix defined by \( T_{ij} = w(r_i) - w(r_j) \). The eigenvalues \( \lambda_i \) of \( T \) are the determinant obtained when substituting all elements in the ith column of the fitness matrix \( w(\mathbf{r}) \) (\( \mathbf{r} = 1 \ldots n \)) with 1. The necessary and sufficient conditions for the existence of a stable equilibrium \( \mathbf{r} \) with all individual population frequencies \( x_i \) of the alleles being nonzero are then that \( T \) is negative definite and that \( 1-\lambda_i > 0 \) for all \( i = 1 \ldots n \). Geometrically, these first two conditions specify a peak in n-dimensional fitness space, and only one such peak is allowed for all alleles to coexist (47).

For heterozygote advantage to be consequential (i.e., to be capable of effectively stabilizing a balanced polymorphism against the stochastic fluctuations arising from random genetic drift), the fitness advantages of a heterozygote over one of its two homozygotes have to be at least of order \( 1/N \). Because we are only interested in such consequential cases of heterozygote advantage, we thus require, as a third condition, that for at least one pair of alleles in a balanced polymorphism, it holds that \( w(r_i) > \max\{w(r_{ij}), w(r_{ji})\} \) and \( 1/N \).
In our simulations, we evaluate these three conditions for the fitness matrix of all alleles with frequencies 0.05 < \(i\) < 0.95. Negative definiteness of \(T\) is tested by numerically calculating eigenvalues using symmetrical bidia-
gonalization with the QR reduction method (48) and checking for the
negativity of all eigenvalues. Signs of determinants \(\Delta\) are estimated using
numerical LU decompositions (48).

All source code is openly available online at: http://sourceforge.net/projects/
fgm. Simulations were run on the Bio-X\(^3\) cluster at Stanford University.

1. Haldane JBS (1927) A mathematical theory of natural and artificial selection, part V:
expression profiles of pri mates, mice, and flies: Stabilizing selection and variability
5. Rifkin SA, Houlé D, Kim J, White KP (2005) A mutation accumulation assay re-
veals a broad capacity for rapid evolution of gene expression. Nature 438:
220–223.
Cambridge, UK).
102–
103:525–533.
119–127.
R1121–R1124.
15. Goldstein DB (1992) Heterozygote advantage and the evolution of a dominant dip-
16. Orr HA, Otto SP (1994) Does diplody increase the rate of adaptation? Genetcs 136:
1475–1480.
17. Haldane JBS (1959) Natural selection. Darwin's Biological Work, ed Bell PR (Cam-
18. Wright S (1977) Evolution and the Genetics of Populations (Univ of Chicago Press,
Chicago).
20. Sutter NB, et al. (2007) A single IGFI allele is a major determinant of small size in
melanism in pocket mice. Proc Natl Acad Sci USA 100:5268–5273.
acid mutation contributes to adaptive beach mouse color pattern. Science 313:
101–104.

ACKNOWLEDGMENTS. We thank members of the D.A.P. laboratory, Daniel
Watersworth, Adam Eyre-Walker, Hugh Fraser, David Kingsley, Molly Prze-
worski, Matthew Stephens, Warren Ewens, Nick Barton, Kevin Bullaughay,
Marc Feldman, Daniel Fisher, Daniel Hartl, Ward Watt, and six anonymous
reviewers for helpful feedback. This research was supported by grants from
the National Institutes of Health Grant GM 077368 and National Science
Foundation Grant 0317171, CNS-0619926 (to D.A.P.). D.S. is a Stanford
Graduate Fellow. P.W.M. was a Human Frontier Science Program post-
doctoral fellow.

adaptive evolution in asexual populations of Saccharomyces cerevisiae. Nat Genet 40:
1499–1504.
568–575.
29. Cai JJ, Macpherson JM, Sella G, Petrov DA (2009) Pervasive hitchhiking at coding and
32. Gonzalez J, Lenkov K, Lipatov M, Macpherson JM, Petrov DA (2008) High rate of
recent transposable element-induced adaptation in Drosophila melanogaster. PLoS
Biol 6:e251.
e1000700.
increased NaCl concentration due to dominant beneficial mutations. Genetcs 139:
177–186.
Genet 21:30–32.
37. Bubb KL, et al. (2006) Scan of human genome reveals no new loci under ancient
39. Moragues BS, Cano JM, Merila J (2008) Identifying footprints of directional and bal-
ancing selection in marine and freshwater three-spined stickleback (Gasterosteus
42. Dobzhansky T (1952) Nature and the origin of heterosis. Heterosis: A Record of Re-
searches Directed Toward Explaining and Utilizing the Vigor of Hybrids (Iowa State
45. Hastings A, Hom CL (1989) Pleiotropic stabilizing selection limits the number of
polyorphic loci to at most the number of characters. Genetics 122:459–463.
UK).
Supporting Information

Sellis et al. 10.1073/pnas.1114573108

SI Text

Definitions. In Fisher’s geometric model (1–5), phenotypes are represented by vectors \( r \) that exist in a \( d \)-dimensional Euclidean phenotype-space \( \mathbb{P} = \mathbb{R}^d \). In haploids, the genotype space \( \mathbb{G}_{hap} \) is isomorphic to the phenotype space, and it is therefore convenient simply to label alleles by their corresponding phenotype \( r \) and work directly with phenotypes. Mutations are modeled by adding a mutation vector \( m \) to the mutated allele (i.e., \( r \rightarrow r + m \)). We assume that mutational direction is sampled from a uniform distribution and that mutational magnitudes are sampled from a specified probability distribution \( P(m) \).

The phenotype of a diploid organism \( r_{dip} \) is the product of two constituent alleles \( r_d \) and \( r_h \); thus, to incorporate diploidy, we must define the mapping from the diploid genotype to the organismal phenotype. The organismal phenotype space for diploids is identical to that for haploids, \( \mathbb{P} = \mathbb{R}^d \), whereas the diploid genotype space is the direct product of two allelic genotype spaces, each of which is isomorphic to the organismal phenotype space (i.e., \( \mathbb{G}_{dip} = \mathbb{R}^{2d} \otimes \mathbb{R}^{2d} \)). We are free to label individual alleles by the organismal phenotype they produce when homozygous (i.e., \( r_d = r_h \)). We then define the diploid genotype-phenotype mapping as the weighted average of the two constituent alleles:

\[
 r_{dip} = (c_r r_d + c_r r_h) / (c_d + c_h).
\]

The weighting represents the phenotypic dominance relationship between the alleles. Because the direct relation between allelic genotypes and organismal phenotypes in diploids, it is again convenient to elide the genotype-phenotype map and speak simply of allelic “phenotypes” analogous to haploid phenotypes, which mutate analogously to those haploid phenotypes as well.

In our theoretical analysis, we focus on the specific situation of a mutant allele \( r_d + m \) arising in a population initially monomorphic for the wild-type allele \( r_d \). It is then convenient to express the phenotype of the mutant heterozygote as \( r_{dip} = r_d + h m \), where \( h \) specifies the phenotypic dominance relation of the mutation \( m \) with respect to the wild type.

To map organismal phenotype to fitness, we define a fitness function \( w(r) \). We restrict our consideration to fitness functions that depend only on the distance from an optimal phenotype and are monotonically decreasing in distance from that optimum. For convenience, we set the origin to be at the fitness optimum; hence, \( w(r) \rightarrow w(r) \).

Range of Adaptive Mutant Alleles. Adaptive mutations are those that increase the fitness of organisms carrying the mutant allele \( r_d = r_h + m \). Given our assumptions about the form of the fitness function, this requires that \( |r_d| < r_h \) in haploids. In diploids, under the Hardy–Weinberg assumptions, it is the fitness of the mutant heterozygote that primarily determines the probability of a new mutation invading the population. This makes it convenient to define adaptive mutations in diploids as those that are adaptive immediately on origination as a heterozygote (i.e., \( |r_{dip}| < r_h \)). Thus, the range of adaptive mutant alleles in haploids (\( \alpha_{hap} \)) and diploids (\( \alpha_{dip} \)) is described by:

\[
\alpha_{hap} = \{ r_d : |r_d| < r_h \} \quad \text{[S1]} \]

\[
\alpha_{dip} = \left\{ r_d : \left| r_d - \frac{(1 - h)}{h} r_h \right| < r_h / h \right\}. \quad \text{[S2]}
\]

Both are spheres: \( \alpha_{hap} \) has radius \( r_h \) and is centered at the origin, and \( \alpha_{dip} \) has radius \( r_h / h \) and is centered at \(-((1 - h)/h) r_h \).

In diploids, we also wish to distinguish between those adaptive mutations that are expected to replace the wild type [i.e., \( w(r_h) > w(r_{dip}) \geq w(r_{hap}) \)] and those that are not expected to do so because they have heterozygote advantage [i.e., \( w(r_h) < w(r_{dip}) \geq w(r_{hap}) \)]. The range of replacing mutant alleles \( \gamma \) is given by:

\[
\gamma = \left\{ \begin{array}{ll}
\varnothing & h < 0 \\
\{ r_d : \left| r_d - \frac{h}{1 + h} r_h \right| < r_h / (1 + h) \} & 0 < h < 1 \\
\{ r_d : \left| r_d - \frac{h}{1 + h} r_h \right| > r_h / (1 + h) \} & h > 1.
\end{array} \right. \quad \text{[S3]}
\]

For incomplete phenotypic dominance \( 0 < h < 1 \), \( \gamma \) is a sphere of radius \( r_h / (1 + h) \) centered at \( h(1 + h) r_h \). For phenotypic overdominance \( h > 1 \), the direction of the inequality is switched and \( \gamma \) is all points that are excluded from this sphere (but that fall in \( \alpha_{dip} \)). The range of adaptive mutants that have heterozygote advantage is then \( \alpha_{hap} \) without \( \gamma \). Note that \( \alpha_{hap} \), \( \alpha_{dip} \), and \( \gamma \) are collinear spheres and \( r_h \) lies on the surface of each (Fig. 2A).

Probability of Adaptive Mutations. The probability that a mutation in this model is adaptive can be expressed as a purely geometric question: What is the probability that the end point of a vector originating on the surface of a sphere (the mutation) will lie within that sphere (the range of adaptive mutants), given that the vector’s direction is sampled uniformly? This probability, \( P < (m/R,d) \), depends on the ratio of the magnitude of the vector \( m \) to the radius of the sphere \( R \) and the dimensionality \( d \). For convenience, let the origin of our space be at the center of the sphere. We choose \( R \) to be the point on the sphere’s surface from which the vector originates. The condition for the end point to lie within the sphere is:

\[
|R + m| < R \Leftrightarrow \cos(\phi) > \frac{m}{2R}. \quad \text{[S4]}
\]

Here, \( \phi \) is \( \pi \) minus the angle between \( m \) and \( R \). Note that this inequality can only be satisfied if \( m \) is less than \( 2R \), because \( \cos(\phi) \leq 1 \). We can determine \( P < (m/R,d) \) by enforcing the condition on \( \phi \), while integrating over all orientations of \( m \):

\[
p < (m/R,d) = \int_0^\infty \int_0^{\pi/2} \sin^2(\phi) \left[ \cos^{-1} \left( \frac{m}{2R} \right) \right] \left( \frac{m}{2R} \sin(\phi) \right)^{d-2} d\phi d\theta, \quad \text{[S5]}
\]

\[
= \frac{1}{\sqrt{\pi}} \frac{m}{2R} \left( \frac{d}{2} + 1 \right) \left( \frac{d}{2} + 2 \right) \left( \frac{m}{2R} \right)^{d-2}. \quad \text{[S6]}
\]

The probability function \( P < (m/R,d) \) is monotonically decreasing over the relevant range of \( 0 < m \leq 2R \) for all dimensionalities. Two special values deserve mention: \( P < (0,d) = 1/2 \) and \( P < (2,d) = 0 \) independent of dimension. The shape of \( p < (m/R,d) \) for several dimensionalities is shown in Fig. S7. We compare this exact result with a long-standing approximation used in the study of Fisher’s geometric model (1–3), that 

\[
p < (m/R,d) \approx \int_0^\infty \frac{1}{\sqrt{2\pi}} e^{-\frac{m^2}{2}} dt.
\]

This approximation is very good at high dimensions, which pre-
Adaptive Mutations in Haploids and Diploids. Let $\alpha$ be the range of adaptive mutations, and assume that the mutation supply is identical for haploids and diploids. The rate at which adaptive mutations occur in a monomorphic population ($\alpha$) is then:

$$u = \Theta \int_{\alpha} P(m) \, dm = \Theta \int_{\alpha}^\infty P(m) \, p < (m/R_d; d) \, dm. \quad [S7]$$

Here, $\Theta/2 = cN\mu$ ($c$, ploidy level; $N$, population size; $\mu$, mutation rate per individual) is the overall rate at which mutations occur in the population. We have used the fact that $\alpha$ is a sphere with the wild-type allele on its boundary in this model. $R_d$ is the radius of the adaptive region; $R_d = r_d$ for haploids and $R_d = r_d/h$ for diploids.

We further define the rate at which adaptive mutations invade the population ($\alpha$) and the average initial selective effect of adaptive mutations ($\Delta \omega$):

$$\nu = \Theta \int_{\alpha} P(m) \pi(m) \, dm. \quad [S8]$$

$$\langle \Delta \omega \rangle = \Theta \int_{\alpha} P(m) \pi(m) \Delta \omega(m) \, dm. \quad [S9]$$

The invasion probability $\pi(m)$ is proportional to $\Delta \omega(m)$, the fitness difference between the mutant individual and the wild-type fitness ($R$). In haploids, the mutant individual is heterozygous in the mutant allele, so $\pi_{hap}(m) \propto \Delta \omega_{hap}(m) = w(r_c + h) - w(r_c)$, whereas in haploids, $\pi_{hap}(m) \propto \Delta \omega_{hap}(m) = w(r_c + m) - w(r_c)$.

We are particularly interested in the relative rates of adaptation in haploids and diploids. For that reason, we consider the ratios of $\nu$, $u$, and $\langle \Delta \omega \rangle$ between haploids and diploids. We calculated these ratios numerically for different values of $\alpha$ and over a range of the parameter ($m$) using the exponential (Fig. S1) and uniform (Fig. S2) distributions of mutation sizes. Here, we focus on two limiting cases: (i) the "large-mutation" limit, where $P(m)$ is uniform over the entire range of adaptive mutations (which is the case in both the uniform mutation model and the exponential mutation model if $(m) = r_c$), and (ii) the "small-mutation" limit, where $P(m)$ is nonzero only for infinitesimal mutations.

Large-Mutation Limit. In the large-mutation limit, $P(m)$ is constant over the entire range of adaptive mutations. The ratio of the rates of occurrence of new mutations $u_{hap}/u_{hap}$ is then:

$$\frac{u_{hap}}{u_{hap}} = \left[ \frac{\int_{\alpha} P(m) \, dm}{\int_{\alpha}^\infty P(m) \, p < (m/R_d; d) \, dm} \right] \frac{1}{|h|} \quad [S10]$$

This result is achieved by a change of variables $m' = |h|m$ in the numerator. Note that this result is the ratio of the radii of the respective adaptive ranges and is independent of dimension. For the ratio of the invasion rates $\nu$, we have:

$$\frac{\nu_{hap}}{\nu_{hap}} = \left[ \frac{\int_{\alpha} P(m) \pi_{hap}(m) \, dm}{\int_{\alpha}^\infty P(m) \pi_{hap}(m) \, dm} \right] \frac{1}{|h|} \quad [S11]$$

Once again, this result is obtained by a change of variables, $m' = |h|m$. We made use of the fact that $h\pi_{hap} \equiv \pi_{hap}$ (i.e., "multiplying" a sphere by a constant results in another sphere). Finally, the ratio of the average initial selective increments ($\Delta \omega$) is as follows:

$$\langle \Delta \omega \rangle_{hap} = v_{hap} \int_{\alpha} P(m) \pi_{hap}(m) \Delta \omega_{hap}(m) \, dm$$

$$\langle \Delta \omega \rangle_{hap} / u_{hap} \int_{\alpha} P(m) \pi_{hap}(m) \Delta \omega_{hap}(m) \, dm = 1. \quad [S12]$$

Applicability of the Large-Mutation Limit. The large-mutation limit, as we have defined it, requires that the entire range of adaptations be accessible to mutation. In diploids, this means that mutations must reach a size of $2r_c/|h|$. However, our qualitative conclusions, such as the higher rate at which adaptive mutations arise in diploids under incomplete phenotypic dominance ([0 < $h < 1$]), will apply as long as the mutation supply reaches most of the range of adaptive mutations (i.e., $(m) \sim r_c$). This condition can be relaxed even further at high dimension. In Fig. S7, we see that as dimensionality increases the range over which there are an appreciable number of adaptive mutations contracts (i.e., as dimension goes up), the characteristic size of potential adaptive mutations goes down. We can use the approximation $p < (m/R, d) \approx \int_{\alpha}^\infty \sqrt{z^2 - d^2} \, dz$ to get a heuristic understanding of how this affects the applicability of the large-mutation limit. In this high-dimension approximation, $m$ enters only in the combination $mn/\alpha$; $R$ is effectively modified by $1/\sqrt{d}$. Therefore, the mutational supply reaches most adaptive mutations if:

$$\langle m \rangle \approx \frac{r_c}{\sqrt{d}} \quad [S13]$$

The higher the dimensionality, the weaker is the condition on $m$, and the more applicable the large-mutation limit becomes.

Small-Mutation Limit. In the small-mutation limit, all mutations are infinitesimal, as enforced by $P(m)$. Therefore, $P(m) < (m/R, d) \sim P(0, d) = 1/2$. Clearly then, $u_{hap} = u_{hap}$ in this limit. Also, because the mutations are infinitesimal, a linear approximation to $\Delta \omega$ is appropriate and $\Delta \omega_{hap}(m) = w(r_c + h) - w(r_c) \sim h \Delta \omega_{hap}(m)$. So, $\nu_{hap} \sim |\hbar|\nu_{hap}$ and $\langle \Delta \omega \rangle_{hap} \sim |\hbar|\Delta \omega_{hap}$.

Heterozygote Advantage in Diploids. We now characterize the fraction of adaptive mutations in diploids that display heterozygote advantage ($\delta$). We differentiate between $\delta_a$, the fraction among all adaptive mutations, and $\delta_w$, the fraction among those that invade:

$$\delta_a = 1 - \frac{\int_{\alpha} P(m) \, dm}{\int_{\alpha} P(m) \pi_{hap}(m) \, dm}$$

$$\delta_w = \frac{1 - \int_{\alpha} P(m) \pi_{hap}(m) \, dm}{\int_{\alpha} P(m) \pi_{hap}(m) \, dm} \frac{1}{|h|} \quad [S14]$$

The small-mutation limit is trivial, $\delta_{hap}$ and $\gamma$ completely overlap for infinitesimal mutations, and homozygote mutants will never have lower fitness than heterozygotes for $0 < h < 1$, whereas all adaptive mutations will have heterozygote advantage if $h > 1$. Recall also that all adaptive mutations have heterozygote advantage when $h < 0$. The large-mutation limit derives immediately from our previous results concerning the rates of ratios between diploids and haploids. The only difference is that we are comparing $\delta_{hap}$ with $\gamma$, which has a radius of $r_c/(1 + h)$. So, in the large-mutation limit:

$$\delta_a = \begin{cases} 0 & h < 0 \\ \frac{r_c/(1 + h)}{r_c/h} & 0 < h < 1 \\ \frac{r_c/(1 + h)}{r_c/h} & h > 1 \end{cases} \quad [S15]$$

There are also two special cases: perfect phenotypic recessiveness $h = 0$ and perfect phenotypic dominance $h = 1$. In the case $h = 0$, mutations cannot be adaptive by our definition because their fitness as a heterozygote is not better than that of the wild type. In the case $h = 1$, the fitness of the mutant heterozygote is equal
to that of the mutant homozygote; thus, strictly speaking, there is never heterozygote advantage. We also note that in the special case of phenotypic codominance \((h = 1/2)\), \(\delta_h = 2/3\). Additionally, \(\delta_h > 1/2\) for all values of \(h\) (except \(h = 0, 1\)). \(\delta_h\) cannot be obtained by our previous methods; in this case, the \(X^2\) terms in the numerator and denominator differ after the usual change of variables. In the case of \(h = 1/2\), we made numerical calculations of \(\delta_h\) and \(\delta_v\) as a function of \((m)\) for exponential and uniform distributions of mutation length, as shown in Figs. S1 and S2. Conditioning on invasion is always seen to increase the frequency of heterozygote advantage in the large-mutation limit.

**Effectiveness of Heterozygote Advantage in the Presence of Drift.**

Selection is only consequential when it is strong enough to outcompete the stochastic fluctuations that arise from random genetic drift. In the context of balancing selection, this requires that the fitness advantage of the heterozygote over each homozygote has to be stronger than drift for balanced states to be effectively maintained.

Formally, the condition for selection to outcompete drift is that fitness differences have to be at least of order \(1/N\) the inverse of the population size \((2)\). In our scenario of a symmetrical Gaussian fitness function, \(w(r) = \exp(-r^2/(2\sigma_m^2))\), this defines a sphere around the optimum of radius:

\[
\rho_0 = \sqrt{-2\sigma_m^2 \log \left(\frac{1}{N}\right)} \approx \sigma_m \sqrt{\frac{2}{N}} \quad [S16]
\]

inside of which fitness will become effectively indiscernible from the optimal fitness. Once the population is located within this effectively neutral sphere, adaptation will cease. Similarly, if both the heterozygote and homozygote of a mutation with \(w_{ah} < w_{bb}\) (i.e., an adaptive mutation with heterozygote advantage) are located inside the sphere, selection will not be sufficient to stabilize the balanced state. The radius \(\rho_0\) is proportional to the SD of the fitness function and decreases with the inverse square root of the population size.

The radius of the effectively neutral sphere sets a limit on how closely adaptive walks will approach the optimum. It also imposes a condition on the possibility of adaptation-driven balanced polymorphisms. It is required that \((m) > \rho_0\) for the selection to be able to outcompete drift and maintain a balanced polymorphism. Heuristically, this can be understood as the requirement that mutations be able to span the neutral sphere, thereby allowing selection to distinguish between the homozygotes outside the sphere and the heterozygote within it. Note that this condition is different in kind from our previous condition for heterozygote advantage to be common, \((m) \gtrsim r/N\). Because \(\rho_0\) decreases over the course of an adaptive walk, we expect that although heterozygote advantage might not initially be frequent, it will eventually become so. The drift condition, however, does not change over the course of a walk; if it is not met, we expect drift to preclude the possibility of adaptation-driven balanced polymorphisms entirely.

Our heuristic explanation of the drift condition is appropriate at small dimensions; however, the consideration of large dimensions requires more care. In high-dimensional phenotypic spaces, most adaptive mutations are not primarily directed toward the optimum but, instead, are largely lateral displacements with just a small component in the direction of the optimum. Heterozygote advantage arises when the intermediate heterozygote is closer to the optimum than either homozygote (just as the midpoint of a chord is closer to the center of the circle than either end point). We can evaluate the possibility of heterozygote advantage in this situation, at least in the limit that \(m << r_0\) (note that this is not inconsistent with our condition for frequent heterozygote advantage \((m) \gtrsim r_0/\sqrt{d}\) when \(d\) is large). In this limit, the maximum decrease in the distance to the phenotypic optimum when comparing the mutant heterozygote with the fittest of the two homozygotes is approximately \(\delta r = (m/2)^2/(2r_0)\). The selective advantage of the heterozygote can then be approximated as \(w'(r)/w(r)\delta r\). Therefore, for the heterozygote advantage to exceed drift, given our Gaussian fitness function, it is necessary that \((m) > \sigma_m \sqrt{2/N} = r_0\). Conveniently, this results in the same condition that we found previously (when we assumed that \(m \approx r_0\)). Thus, this serves as a universal condition for the possibility of adaptation-driven balanced polymorphisms in our model when accounting for drift. It should be noted that the identity between these low- and high-dimension conditions is a peculiarity of our choice of a Gaussian fitness function rather than a general feature of all single-peaked landscapes.

To verify these theoretical predictions, we simulated adaptive walks toward a fixed fitness optimum for various settings of \(N\) and \(\sigma_m\) and \((m)\). As previously, a 2D codominant Fisher’s model was used, starting from a monomorphic population at \(r_0 = (2, 0)\); mutations were modeled by an exponential mutation-size distribution. We kept \(\Theta = 0.1\) constant over all scenarios. Adaptive walks were simulated until either the population successfully adapted [as defined by at least 90% of individuals being located inside the effectively neutral sphere (i.e., \(\rho_0 < r_0\))] or after 10N generations, whichever came first.

The results of these simulations are shown in Table S1 and summarized in Fig. 2D. As predicted by our theoretical arguments, consequential balanced states (e.g., where the fitness differences between heterozygotes and homozygotes are at least of size \(1/N\)) (as described in the section on ascertainment of balanced polymorphisms during adaptive walks in Materials and Methods) are always common in adaptive walks unless average mutation sizes \((m)\) become on the order of \(\rho_0\) or smaller. Interestingly, in our simulations, the presence of consequential balanced polymorphisms corresponds well with whether successful adaptation, as defined by the population traversing at least 90% of the initial fitness distance to the optimum, is observed at all. This results from the fact that when \(m \approx r_0\) in our scenario, it is typical that \(r_0 \approx r_0\) also. Thus, in these cases, adaptive walks start already very close to the effectively neutral sphere; consequently, further fitness improvements can no longer outcompete the stochastic fluctuations attributable to drift.

Fig. S1. Characteristics of single adaptive mutations in the uniform mutation model as a function of \( \langle m \rangle / r_a \). The five columns span the range from phenotypic underdominance \( (h = -0.25) \) to overdominance \( (h = 1.25) \). Different colors show results for \( d = 2 \) (black), \( d = 10 \) (blue), and \( d = 50 \) (red) phenotypic dimensions. (A) Ratio of the rates at which new adaptive mutations occur. (B) Ratio of the rates of invading mutations. (C) Ratio of the average fitness advantage conferred by invading mutations. (D) Fraction of adaptive mutations in diploids that have heterozygote advantage. (E) Fraction of invading mutations in diploids that have heterozygote advantage.
Fig. S2. Same as in Fig. S1, but data are shown for the exponential mutation model.
Fig. S3. Probability of observing a balanced polymorphism (frequency 0.05 < x < 0.95) in a diploid population during adaptive walks toward a fixed optimum for different values of Θ. Solid lines are the absolute probabilities of observing a balanced polymorphism, and dotted lines are the probabilities conditional on the presence of a polymorphism. The mutation rate was always μ = 2.5 × 10⁻⁷. Different values of Θ = 4Nμ correspond to different population sizes. Statistics were obtained by averaging over 10⁴ walks for each value of Θ.

Fig. S4. Statistics of adaptive walks toward a fixed fitness optimum in haploid and diploid populations. (A) Mean population fitness (w) in haploids is greater than that of the diploid population at all times when averaged over replicate walks. (B) Fitness of the most fit individual (w_max) is higher in diploids than it is in haploids at all time points when averaged over replicate walks. (C) Fitness variance over all individuals. Diploid populations suffer from a high genetic load, which can be identified as the segregation load attributable to pervasive fitness overdominance. (D) Genotypic variance over all individuals (calculated separately for each dimension and then summed over all dimensions). Data points are medians over 10³ runs, and error bars specify the first and third quartiles.
Fig. S5. Typical allele frequency trajectories in the moving optimum scenario. The four graphs show the trajectories of all alleles present during the first $5 \times 10^5$ generations of four simulation runs with different speeds of environmental change $\sigma_{env}$. Simulation parameters were $N = 5 \times 10^4$, $r_{aa} = (2, 0)$, and $\langle m \rangle = \sigma_w = 1$ (Materials and Methods). Different colors represent different alleles. Under a fast-moving optimum (A), polymorphisms are shorter lived and adaptive alleles fix at a higher rate compared with a slow-moving optimum (D). The trajectories of balanced alleles in the moving optimum scenario differ from those in a fixed optimum scenario (Fig. 2B) in the frequency at which two alleles’ balance may shift in response to the movement of the optimum, as can be seen in B and even more clearly in C.
Fig. S6. Statistics of mutation effects in the moving optimum model. The two upper plots show the distributions of mutation sizes under a fast-moving optimum (A) and a slow-moving optimum (B) for different categories of mutations. Dotted lines indicate all newly occurring mutations specified by our standard exponential mutational model with $P(m) \propto \exp(-m)$, red indicates mutations that eventually became fixed in the population but were never part of a balanced polymorphism, blue indicates mutations that eventually became fixed but had previously been part of a balanced polymorphism, and green indicates mutations that were part of a balanced polymorphism but never became fixed. Average mutation sizes decrease, in order, from newly occurring mutations, balanced but never fixed mutations, balanced and eventually fixed mutations, to fixed but never balanced mutations. The fast- and slow-moving optimum scenarios show qualitatively similar behavior, with distributions being shifted toward larger sizes in the faster changing environment. The two lower plots show the distributions of fitness effects (mutant homozygote vs. wild-type homozygote) of the different mutation categories under fast-changing (C) and slow-changing (D) environments. Most newly occurring mutations in our model are deleterious as homozygotes. The fixed but never balanced mutations, as expected, confer the highest average homozygote fitness advantage. The fixed mutations that were intermediately balanced confer slightly lower average fitness advantage. Note that a substantial amount of these mutations initially have a negative fitness effect as homozygotes, as indicated by the below-zero tail of the distributions. Finally, the balanced mutations that never became fixed have the lowest fitness advantage as homozygotes. The fast- and slow-moving optimum scenarios again show qualitatively similar behavior, with distributions being shifted toward larger fitness differences in the faster changing environment. The distributions of mutation sizes and fitness effects in each scenario were estimated from $10^4$ simulation runs over $10^7$ generations each (Materials and Methods).

Fig. S7. Probability that a vector originating on the surface of a sphere has its end point within that sphere as a function of the vector size $m$ (scaled to the radius of the sphere $R$). Several numbers of dimensions are shown. The plots labeled “Exact” show the exact result for $P < (m/R; d)$ from Eq. S6. The plots labeled “Approx” show the high-dimension approximation to this function originally used by Fisher, $p < (m/R; d) \approx \frac{1}{2}\sqrt{\pi/R} \exp(-m^2/2R^2)$.$^4$ The approximation is effective at high dimension but is inappropriate at low dimension.
Table S1. Probability of observing balanced polymorphisms during adaptive walks toward a fixed fitness optimum as a function of relative mutation sizes for various settings of population size $N$ and SD $\sigma_w$ of the Gaussian fitness function

<table>
<thead>
<tr>
<th>$N$</th>
<th>$\sigma_w$</th>
<th>$\langle m \rangle$</th>
<th>$\langle m \rangle/\sigma_w$</th>
<th>Adaptedness</th>
<th>$P_{bal}$ (run)</th>
<th>$P_{bal}$ (time)</th>
</tr>
</thead>
<tbody>
<tr>
<td>$10^3$</td>
<td>1</td>
<td>22</td>
<td>0.90</td>
<td>0.91</td>
<td>0.45</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>11</td>
<td>0.86</td>
<td>0.86</td>
<td>0.38</td>
<td></td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>4.5</td>
<td>0.61</td>
<td>0.70</td>
<td>0.22</td>
<td></td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>2.2</td>
<td>0.26</td>
<td>0.49</td>
<td>0.11</td>
<td></td>
</tr>
<tr>
<td></td>
<td>20</td>
<td>1.1</td>
<td>0.06</td>
<td>0.34</td>
<td>0.04</td>
<td></td>
</tr>
<tr>
<td>$10^4$</td>
<td>1</td>
<td>71</td>
<td>0.98</td>
<td>0.96</td>
<td>0.50</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>35</td>
<td>0.97</td>
<td>0.95</td>
<td>0.48</td>
<td></td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>14</td>
<td>0.92</td>
<td>0.89</td>
<td>0.39</td>
<td></td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>7.1</td>
<td>0.78</td>
<td>0.79</td>
<td>0.31</td>
<td></td>
</tr>
<tr>
<td></td>
<td>20</td>
<td>3.5</td>
<td>0.50</td>
<td>0.63</td>
<td>0.17</td>
<td></td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>1.4</td>
<td>0.10</td>
<td>0.35</td>
<td>0.04</td>
<td></td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>0.71</td>
<td>0.00</td>
<td>0.30</td>
<td>0.02</td>
<td></td>
</tr>
<tr>
<td>$10^5$</td>
<td>1</td>
<td>224</td>
<td>1.00</td>
<td>0.98</td>
<td>0.52</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>112</td>
<td>0.99</td>
<td>0.97</td>
<td>0.51</td>
<td></td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>45</td>
<td>0.99</td>
<td>0.95</td>
<td>0.47</td>
<td></td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>22</td>
<td>0.96</td>
<td>0.91</td>
<td>0.45</td>
<td></td>
</tr>
<tr>
<td></td>
<td>20</td>
<td>11</td>
<td>0.88</td>
<td>0.86</td>
<td>0.38</td>
<td></td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>4.5</td>
<td>0.61</td>
<td>0.69</td>
<td>0.23</td>
<td></td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>2.2</td>
<td>0.25</td>
<td>0.48</td>
<td>0.11</td>
<td></td>
</tr>
<tr>
<td></td>
<td>200</td>
<td>1.1</td>
<td>0.06</td>
<td>0.36</td>
<td>0.04</td>
<td></td>
</tr>
<tr>
<td></td>
<td>400</td>
<td>0.56</td>
<td>0.00</td>
<td>0.32</td>
<td>0.02</td>
<td></td>
</tr>
<tr>
<td>$10^4$</td>
<td>10</td>
<td>0.5</td>
<td>0.72</td>
<td>0.65</td>
<td>0.17</td>
<td></td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>0.2</td>
<td>1.4</td>
<td>0.43</td>
<td>0.15</td>
<td>0.01</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>0.1</td>
<td>0.71</td>
<td>0.16</td>
<td>0.04</td>
<td>0.00</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>0.05</td>
<td>0.35</td>
<td>0.05</td>
<td>0.02</td>
<td>0.00</td>
</tr>
</tbody>
</table>

*Adaptedness* measures the average improvement in mean population fitness achieved over a walk relative to the maximally possible improvement: $(\langle w_{end} \rangle - \langle w_{start} \rangle)/(1 - \langle w_{start} \rangle)$. $P_{bal}$ (run) is the probability of at least one balanced polymorphism arising over the course of the walk. $P_{bal}$ (time) is the average time during which balanced polymorphisms are present over the walk. In the last four rows, $N$ and $\sigma_w$ were kept constant and the average mutation size $\langle m \rangle$ was varied instead. Statistics were obtained by averaging over $10^3$ walks for each parameter setting. In accordance with our theoretically predicted limit, consequential balanced polymorphisms are common until the ratio $\langle m \rangle/\sigma_w$ becomes on the order of 1 or less.

Table S2. Probability of observing balanced polymorphisms in adaptive walks toward a moving fitness optimum under different speeds of environmental change

<table>
<thead>
<tr>
<th>$\sigma_{env}$</th>
<th>Runs extinct, %*</th>
<th>Time polymorphic, %†</th>
<th>Polymorphisms balanced, %‡</th>
<th>Substitutions balanced, %§</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>100</td>
<td>4</td>
<td>1</td>
<td>—</td>
</tr>
<tr>
<td>$10^{-1}$</td>
<td>25</td>
<td>38</td>
<td>44</td>
<td>68</td>
</tr>
<tr>
<td>$10^{-2}$</td>
<td>0</td>
<td>31</td>
<td>74</td>
<td>67</td>
</tr>
<tr>
<td>$10^{-3}$</td>
<td>0</td>
<td>29</td>
<td>85</td>
<td>67</td>
</tr>
<tr>
<td>$10^{-4}$</td>
<td>0</td>
<td>23</td>
<td>93</td>
<td>61</td>
</tr>
<tr>
<td>$10^{-5}$</td>
<td>0</td>
<td>5</td>
<td>58</td>
<td>—</td>
</tr>
</tbody>
</table>

For each speed $\sigma_{env}$ of the optimum, $10^4$ simulation runs of $10^7$ generations each were simulated (Materials and Methods). Under the very fast-moving optimum ($\sigma_{env} = 1$), populations typically became extinct quickly and only a few polymorphisms were observed in the short time window before extinction. Those polymorphisms were mostly unbalanced. However, once the optimum moved slower, such that the population could effectively follow it, polymorphisms became common; typically, more than 50% of those polymorphisms were balanced. Under a very slowly moving optimum ($\sigma_{env} = 10^{-5}$), populations were well adapted most of the time, and thus again less frequently polymorphic. The fraction of balanced polymorphisms among all polymorphisms, however, remained substantial in this scenario. In all scenarios, a substantial fraction (60–70%) of the adaptive mutations that eventually became fixed in the population did go through an intermediate balanced state (these data have not been collected for the extremely fast-moving, $\sigma_{env} = 1$, and extremely slow-moving, $\sigma_{env} = 10^{-5}$, optima).

*Percentage of runs in which the population could not successfully follow the fitness optimum (extinction defined by the mean population fitness approaching zero).
†Percentage of time during which a polymorphism (frequency $0.05 < x < 0.95$) was present in the population averaged over runs (only the time before extinction was considered).
‡Percentage of polymorphisms that were balanced.
§Percentage of all observed substitutions that had ever been part of a balanced polymorphism.