The Population Genetics of Biological Noise

Daniel M. Weinreich[1,2*], Tom Sgouros[2], Yevgeniy Raynes[1], Hlib Burtsev[1], Edison Chang[1], Sanyu Rajakumar[1], Ignacio G. Bravo[3], Csenge Petak [4]

[1] Department of Ecology, Evolution and Organismal Biology, Brown University, Providence, Rhode Island 02912 USA [2] Data Science Institute, Brown University, Providence, Rhode Island 02912 USA [3] Laboratory MIVEGEC (Univ Montpellier, CNRS, IRD) Centre National de la Recherche Scientifique, Montpellier 34394 France [4] Department of Biology, University of Vermont, Burlington, Vermont 05405 USA

* Correspondence and requests for materials should be addressed to Daniel M. Weinreich: Daniel_Weinreich@Brown.edu

Abstract

Noise is intrinsic to information transmission, and information transmission is intrinsic to life. Critically, while biological noise is random, its amount is in part genetically determined. Here, we explore the population genetics of alleles that heritably influence the amount of noise in any biological mechanism. We argue that biological noise is a double-edged sword: almost always inducing deleterious phenotypes but also occasionally yielding high fitness outcomes. We further suggest that this implies the existence of an equilibrium amount of noise, at which the advantage of producing additional, rare beneficial phenotypes is balanced by the cost of producing the vastly more common deleterious phenotypes. The location of this equilibrium will reflect the rate at which its environment is changing, the persistence of association between noise-modifying alleles and the phenotypic perturbations they induce, the mode of selection, and the population size. The population genetics of alleles that heritable change mean trait values have been established for nearly a century, but to our knowledge, this is the first general treatment of the evolution of heritable changes in the statistical distribution around that mean. Our framework generalizes modifier theory in the sense used by Altenberg, Feldman and others, and resolves teleological criticisms of the hypothesis that evolvability can evolve via natural selection. We conclude with an outline of open theoretical and empirical questions posed by our framework.

Introduction

Information transmission is intrinsic to biology: during development, information contained in a newborn's inheritance informs its phenotype, and during reproduction information flows back to the next generations' newborns (Figure 1A, Maynard Smith [1999]). But biological noise is inevitable. Noise during development results in stochastic fluctuations in biochemical, cellular and organism phenotypes [Eling, Morgan, and Marioni 2019]). And during reproduction, noise causes a newborn's inheritance to differ from that of its parent(s). Critically, while biological noise is random, its magnitude can be influenced by genetics. For example, the likelihood of some proteins' misfolding can vary heritably as a function of genetic variation in chaperone protein expression level (e.g., Tokuriki and Tawfik [2009]), and DNA polymerase alleles can differ in their fidelity of replication (e.g., Matic et al. [1997]). These effects are illustrated abstractly in Figure 1B-E.

This raises the possibility that the amount of biological noise may itself be subject to natural selection [e.g. Viney and Reece 2013, Richard and Yvert 2014]. While a naive interpretation might hold that biological noise is intrinsically deleterious [Sturtevant 1937], noise can also be beneficial [Tawfik 2010, Tsimring 2014]. Here, we propose a general treatment for how natural selection regards genetic variants that heritably influence the amount of noise in any biological mechanism. We highlight three essential features of our theory: i) the critical role of indirect selection, ii) the tension between deleterious and beneficial consequences of biological noise, and iii) the importance of environmental variability. Finally, we locate our ideas in the context of previous work, and propose several avenues for future investigation.

We define *modifier alleles* as those that heritably influence the amount of noise in any biological mechanism. Modifier alleles reside at *modifier loci*, and we designate the population genetics of modifier alleles as *modifier theory*. This language generalizes previous usage [e.g. Karlin and McGregor 1974, Otto 2013, Altenberg, Liberman, and Feldman 2017], a point we develop in the Discussion. Following the logic of Figure 1A, we further partition modifier loci into modifiers of developmental noise (hereafter, *developmental modifiers*) and modifiers of reproductive noise (hereafter, *reproductive modifiers*). Tables 1 and 2 enumerate all the biological mechanisms subject to heritable control of developmental and reproductive noise that we have uncovered in the literature. For each, we also provide the timescale at which the phenotypic perturbation decays back to its expected (i.e., no-noise) value, the persistence of association between modifier allele and induced, phenotypic perturbation, and representative citations.

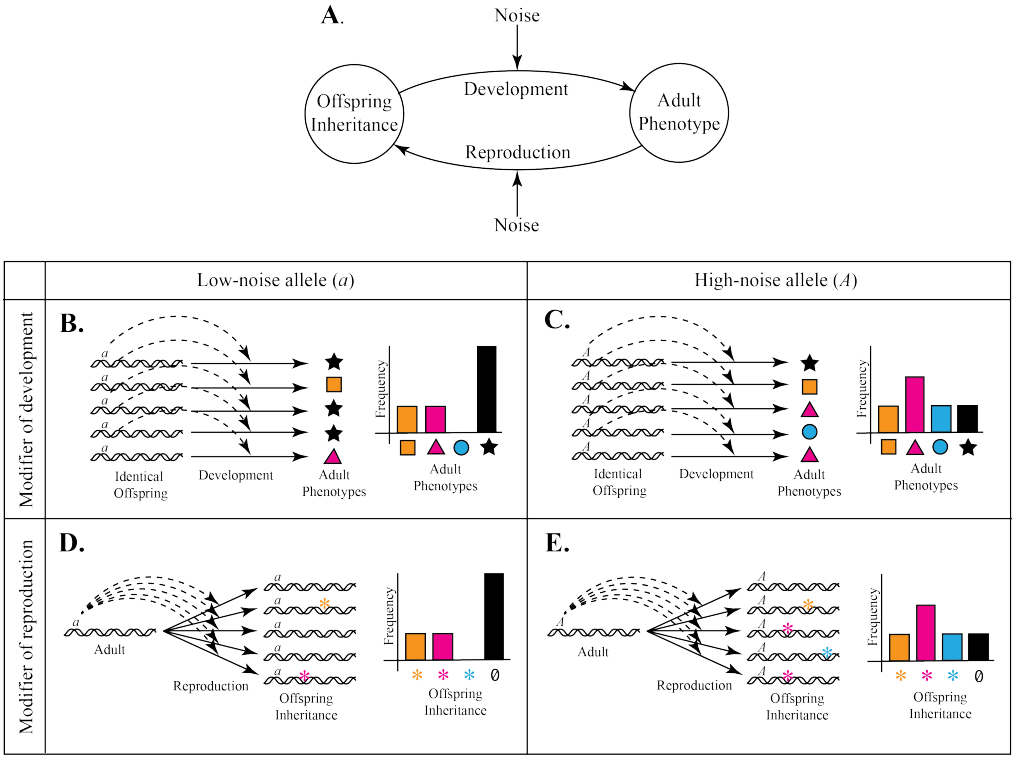

Figure 1: The bidirectional flow of information each generation suggests two classes of modifier loci. **A.** During development, information flows from the offspring's inheritance to adult phenotype, and during reproduction it flows back from adult to the next generation's inheritance. Holding environment constant, low (**B**) and high (**C**) noise developmental modifier alleles (designated *a* and *A*) differentially influence the amount of phenotypic variation among adults derived from identical offspring (represented by colored shapes). Again holding environment constant, low (**D**) and high (**E**) noise reproductive modifier alleles (designated *a* and *A*) differentially influence the amount of genetic and non-genetic variation inherited from a single parent (represented by colored asterisks).

Table 1: Developmental mechanisms subject to biological noise

Mechanism	Manifestation of noise	Timescale at which phenotypic perturbations decay back to expectations	Persistence of association between modifier allele and directly selected phenotype	Representative citations
Act via variation in intracellular molecular mechanisms				
Gene expression noise	Intracellular variation in RNA or protein number over time or space	Rate of RNA or protein synthesis and degradation, mitotic dilution	Reciprocal of promoter or ribosomal on/off rate	Hill, Vande Zande, and Wittkopp [2021]
Protein sequence variation[1]	Intracellular variation in protein sequence over time or space	Reciprocal of transcription, posttranscriptional modification, tRNA charging, translation or degradation rates[2]	One cell generation	Drummond and Wilke [2009], Evans et al. [2018]
Enzyme promiscuity[3]	Variation in substrate binding affinity and/or catalytic rate.[4]	Reciprocal of binding and/or catalysis rate	Permanent	Copley [2017]
Protein fold-switching	Heterogeneity in protein structure over time or space[5]	Reciprocal of protein structure interconversion rate[6]	Permanent	Porter [2023]
Network topology[7]	Variation in network dynamics	Reciprocal of network relaxation time	Permanent	Bruggeman and Teusink [2018]
Act via variation in environmental sensitivity				
Environmental robustness[8]	Limited phenotypic responses triggered by environmental variation	Reciprocal of rate of environmental change	Permanent	Gursky, Surkova, and Samsonova [2012]

Continued on Next Page. . .

[1]Includes mis-transcription, post-transcriptional modification, tRNA-charging, mis-translation and post-translational modification.

[2]Translation error rate can be elevated by environmental stress [Mohler and Ibba 2017], thus representing a mechanism of phenotypic plasticity (below).

[3]We follow Copley [2017] and restrict the term to enzymes that also catalyze physiologically irrelevant secondary reactions. We thus distinguish between promiscuous enzymes and those with broad, physiologically important substrate specificity.

[4]Structural proteins can also exhibit binding promiscuity [Rebeiz et al. 2011, Atkins 2015].

[5]Also called metamorphic proteins [Murzin 2008] or extant fold-switching [Porter and Looger 2018], this effect is closely related to evolved fold-switching (e.g., Chakravarty et al. [2023]), in which alternative fold structures come to be separated by only very few mutations. However, evolved fold-switching means that the local fitness landscape experienced by an evolving population has evolved. Put another way, the evolution of fold-switching influences the distribution of mutational fitness effects and is thus a modifier of reproduction (Table 2).

[6]Fold-switching can also be environmentally triggered [Kim and Porter 2021], thus representing a mechanism for phenotypic plasticity (below).

[7]Single-cell systems biology has clarified how different network motifs influence the amount of noise in a network's behavior [Çağatay et al. 2009], and these ideas may apply equally to gene regulation networks, cell signaling networks, metabolic networks, and ecological networks.

[8]Or environmental canalization. Often in opposition to diversified bet hedging.

Mechanism	Manifestation of noise	Timescale of noise decay	Timescale of association between modifier allele and directly selected phenotype	Representative citations
Phenotypic plasticity[9]	Specific phenotypic responses triggered by environmental cues	Reciprocal of rate of environmental change	Permanent	Paaby and Testa [2018]
Acts via variation in environment				
Seasonal migration[10]	Variation in environmental conditions encountered	Annual	Permanent	Winger et al. [2019]

[9]Following others (e.g., Simons [2011]), we restrict phenotypic plasticity to cases where environmental cues trigger specific phenotypic responses, sometimes also described by norms of reaction. We thus distinguish phenotypic plasticity from random environmentally triggered phenotypic perturbations such as diversified bet hedging (previous). Transgenerational phenotypic plasticity is sometimes realized via mechanisms of non-genetic inheritance outlined above [Bell and Hellmann 2019]. Thus, it is a form of Lamarkian inheritance of acquired phenotypes.

[10]Modifiers of dispersal (Table 2) increase variation in environment experienced, while modifiers of seasonal migration reduce variation in environment experienced.

Table 2: Reproductive mechanisms subject to biological noise

Mechanism	Manifestation of noise	Timescale at which phenotypic perturbations decay back to expectations	Persistence of association between modifier allele and directly selected phenotype	Representative citations
		Act via genetic variation		
Genetic mutation[1]	Genetic variation among offspring from a single parent	Reciprocal of mutation rate[2]	Recombination rate between modifier locus and directly selected locus or loci	Sniegowski et al. [2000]
Mendelian segregation[3]	Genetic variation among genotypes from a single parent	One organismal generation	Recombination rate between modifier locus and directly selected locus or loci	Chasnov [2000]
Chromosomal crossover[4]	Genetic variation among gametes from a single parent	Reciprocal of recombination rate	Recombination rate between modifier locus and directly selected locus or loci[5]	Otto and Lenormand [2002], Becks and Agrawal [2010]
Segregation distortion[6]	Allelic variation among gametes from a single parent	One organismal generation	Permanent	Hartl [1975]
Bias in genetically determined sex ratio	Variation in sex among replicate offspring from a single parent	One organismal generation	Permanent	Fisher [1930], Otto [2013]
		Acts via non-genetic variation		
DNA methylation and histone modification	Epigenetic variation among replicate offspring from a single parent	Reciprocal of forward and backward rates of methylation or histone modification	Permanent	Pujadas and Feinberg [2012]
Cytoplasmic inheritance[7]	Variation in cytoplasmic content of daughter cells produced by a single mitosis	One cell generation	Permanent	Ackermann et al. [2007]

Continued on Next Page. . .

[1]DNA polymerase alleles can also vary in mutational bias, e.g. Couce, Guelfo, and Blázquez [2013], Serero et al. [2014], Melde, Bao, and Sharp [2022], Sane et al. [2023], sometimes with evolutionary consequences [Couce and Tenaillon 2019]. See also Tenaillon and Matic [2020] for cases where mutation and recombination rates can be tuned locally. Vertebrate immunity loci and microbial contingency loci [Moxon, Bayliss, and Hood 2006] have similar properties. Finally, mitotic mutations introduce developmental noise.

[2]Mutation rate can be elevated by environmental stress (see references in Matic [2019]), thus representing a mechanism of phenotypic plasticity (Table 2).

[3]It is important to distinguish between modifiers of Mendelian segregation in diploids and modifiers of recombination rate (next). Mendelian segregation incurs many of the direct fitness costs of sex including that of finding mates, risk of predation and disease, and the 2-fold cost of sex (e.g., Otto and Lenormand [2002]).

[4]Unlike modifiers of Mendelian segregation (previous), modifiers of recombination rate influence the probability of recombination between loci, which can occur in haploids as well as in obligate sexual diploids. Direct costs here include the energetic costs of meiosis (e.g., Otto and Lenormand [2002]).

[5]This modifier allele can influence its own timescales of association with directly selected phenotype(s).

[6]Complicated by the fact that these alleles are also intrinsically directly selected.

[7]It is perhaps formally interesting to notice that with cytoplasmic segregation, increasing asymmetry increases noise, whereas in the case of genetic segregation distortion

Mechanism	Manifestation of noise	Timescale of noise decay	Timescale of association between modifier allele and directly selected phenotype	Representative citations
Other forms of non-genetic inheritance	Fidelity of vertical transmission of somatic (including prions and the microbiomes of plants and animals), nutritional, environmental and learned behavioral features of an organism	One organismal generation	Permanent	Bonduriansky and Day [2009], Henry et al. [2021]
Acts via genetic variation and also via fitness variation				
Ploidy	Genetic variation among genotypes from a single mating; also influences dominance coefficients acting on mutations.	One organismal generation	Recombination rate between modifier locus and directly selected locus or loci	Raynes and Weinreich [2021]
Act via fitness variation				
Proportion of time each generation spent in the haploid phase	Proportion of time each generation during which mutations are subject to haploid selection	One organismal generation	Permanent	Otto and Gerstein [2008]
Assortative mating	Proportion of individuals in which mutations are subject to homozygote selection	One organismal generation	Permanent	De Cara, Barton, and Kirkpatrick [2008]
Distribution of fitness effects[8]	Variation in fitness induced by mutation	Reciprocal of mutation rate	Recombination rate between modifier locus and directly selected locus or loci	Ferrare and Good [2024]
Epistasis[6]	Variation in fitness effect of an allele as a function of alleles at other loci	Reciprocal of mutation rate	Recombination rate between modifier locus and directly selected locus or loci	Liberman and Feldman [2006]
Dominance[6]	Variation in fitness effect of an allele as a function of the allele at homologous locus on the other chromosome	Reciprocal of mutation rate	Recombination rate between modifier locus and directly selected locus or loci	Fisher [1928], Otto [2013]
Acts via phenotypic variation				

Continued on Next Page…

(above), increasing asymmetry reduces noise. In the present case, transmission is zero-sum: one daughter cell inherits whatever the other does not. Whereas with genetic segregation, asymmetry means increasing the number of copies of the driver allele in the population.

[8]These includes modifiers of mutational robustness, genetic (de)canalization, and evolutionary capacitors. More abstractly, this class of modifier loci can cause populations to systematically move across the fitness landscape in response to indirect selection, i.e., to cause the local fitness landscape experienced by an evolving population to evolve (e.g., Wilke et al. [2001] but see also Archetti [2009]).

Mechanism	Manifestation of noise	Timescale of noise decay	Timescale of association between modifier allele and directly selected phenotype	Representative citations
Pleiotropy[9]	Variation in phenotypic consequence of a single mutation	Reciprocal of mutation rate	Recombination rate between modifier locus and directly selected locus or loci	Draghi and Wagner [2008], Pavlicev, Cheverud, and Wagner [2011]
Acts via variation in environment				
Dispersal[10]	Variation in environment experienced by offspring from a single parent	Reciprocal of migration rate	Rate of migration[5]	Balkau and Feldman [1973]

[9]Often in opposition to modularity.

[10]Modeled as a per-generation probability of migration m, in expectation, an environment is heritable for $1/m$ generations. In contrast, modifiers of seasonal migration act within generations and thus appear in Table 2.

Direct and indirect selection

Population genetics is the biological discipline that explores how the genetic composition of an evolving population changes over time in response to processes like mutation, natural selection and random genetic drift. Classically, models of natural selection assume that alleles heritably affect mean trait values. Thus, beneficial alleles (those whose change in mean trait value confer higher fitness) tend to spread over the course of generations, while deleterious alleles tend to be eliminated. Extending ideas of others (e.g., Karlin and McGregor [1974], Otto [2013], Sniegowski et al. [2000], we define such alleles as being *directly selected*.

By our definition, a modifier allele influences biological noise, i.e., stochastic perturbations around the mean trait value (Figure 1B-E). So natural selection will enrich for whatever modifier allele is found among organisms that have the good fortune to realize high-fitness phenotypes. The frequency of a developmental modifier allele in the next generation will be influenced by direct selection acting on whatever phenotype its carrier happens to acquire during its development. And the frequency of a reproductive modifier allele in the generation-after-next will reflect direct selection acting on the phenotypes conferred by whatever genetic or non-genetic novelties the carrier introduces in its offspring in the next generation. In all cases, natural selection acts on modifier alleles in response to the phenotypic variants they induce, and following others, we designate this as *indirect selection* [e.g. Karlin and McGregor 1974, Otto 2013, Sniegowski et al. 2000]. This is also sometimes called *second order selection* [e.g. Karlin and McGregor 1972, Tenaillon et al. 1999, Woods et al. 2011]. (Our usage of direct and indirect selection appears unconnected to that in the sexual selection literature, [e.g. Andersson 1994, Kirkpatrick and Barton 1997].)

Importantly, modifier alleles can also be directly selected. For example, generation time in some RNA viruses is limited by genome replication time [Elena and Sanjuán 2005, Furió, Moya, and Sanjuán 2005]. Thus, an RNA polymerase allele that is faster because it lacks a proofreading activity can accelerate genome replication in its carriers, thereby increasing fitness. Such direct selection is independent of any indirect selection mediated by the phenotypic consequences of whatever additional mutations the error-prone polymerase introduces. Similarly, alleles that reduce chaperone protein expression will likely be intrinsically beneficial, since all gene expression is metabolically costly. Again, this direct effect is independent of any indirect selection acting on the phenotypic consequences of an increase in the proportion of misfolded proteins.

Modeling the fitness of modifier alleles

By our definition, modifier alleles heritably influence the amount of noise in development or reproduction. In order to understand the action of indirect selection acting on a modifier allele, we thus require an explicitly stochastic approach that captures the probabilities with which the allele induces each conceivable, directly selected phenotype in a carrier. For example, Figure 1B-E schematically represent samples drawn from four modifier alleles' probability distributions. Indeed, experimental data of this sort are required to demonstrate the existence of the heritable control of biological noise. We return to this point in the Discussion.

Following others [e.g. Karlin and McGregor 1972, 1974], we here assume that modifier alleles only experience indirect selection. The interested reader is directed to Otto [2013] and to Ferrare and Good [2024], who explicitly relax this assumption. See also Sniegowski and Murphy [2006] and Wagner [2022], who argue that our simplification may sometimes be biologically untenable. We return to this point in the Discussion as well.

From biological noise to phenotypic noise to fitness

To build intuition, we introduce the toy model shown in Figure 2A. Here, we consider a one-dimensional, normally distributed phenotype (z) that can assume any real value (formally, $z \in R$). Algebraically we write

$$z_{realized} \sim N(z_0, \sigma^2_{phenotype}), \tag{1}$$

where $\sigma_{phenotype}$ is our proxy for the amount of phenotypic noise and z_0 is the organism's expected phenotype averaged over the noise (also written E[z]). This is represented at the bottom of Panel A.

Since indirect selection acts on the fitness of induced phenotypes, we also need to specify the phenotype-to-fitness mapping. Following others [e.g. Charlesworth 2013], we assume fitness drops quadratically from an optimal phenotype ($z_{optimum}$). Algebraically we write $r(z) = r_{optimum} - (z_{optimum} - z)^2$. ($r$ gives an expectation over demographic stochasticity; we will shortly address this second source of stochasticity.) Without loss of generality, we set the optimal phenotype $z_{optimum} = 0$, which means that $|z|$ is the distance to the phenotypic optimum. Finally, we read r as Malthusian fitness. Because natural selection acts on differences in Malthusian fitness, we normalize by subtraction, yielding

$$\Delta r(z) = r(z) - r_{optimum} = -z^2. \tag{2}$$

Importantly, under Equation 1, the magnitude of any phenotypic perturbation is $|z_{realized} - z_0|$, and because fitness (Equation 2) is symmetric in z, all perturbations larger than $2|z_0|$ are deleterious. These details are illustrated in the center panel of Figure 2A.

The fitness effects of phenotypic noise

The full probability density function of fitness effects induced by biological noise under our toy model is easily found (Equation A1 in the Appendix), and typical results are shown in the left panel of Figure 2A and in Figure 2B. The expected fitness effect of biological noise over all phenotypes is also easy to compute (Equation A2 in the Appendix). In expectation, phenotypic noise is deleterious (see colored solid arrows in Figure 2B and solid lines in Figure 2D). Qualitatively, this follows from two facts. First because our model assumes that noise is symmetric, small phenotypic perturbations (smaller than $2|z_0|$; see previous paragraph) are equally likely to be beneficial or deleterious. And second, because our fitness function $r(z)$ is concave

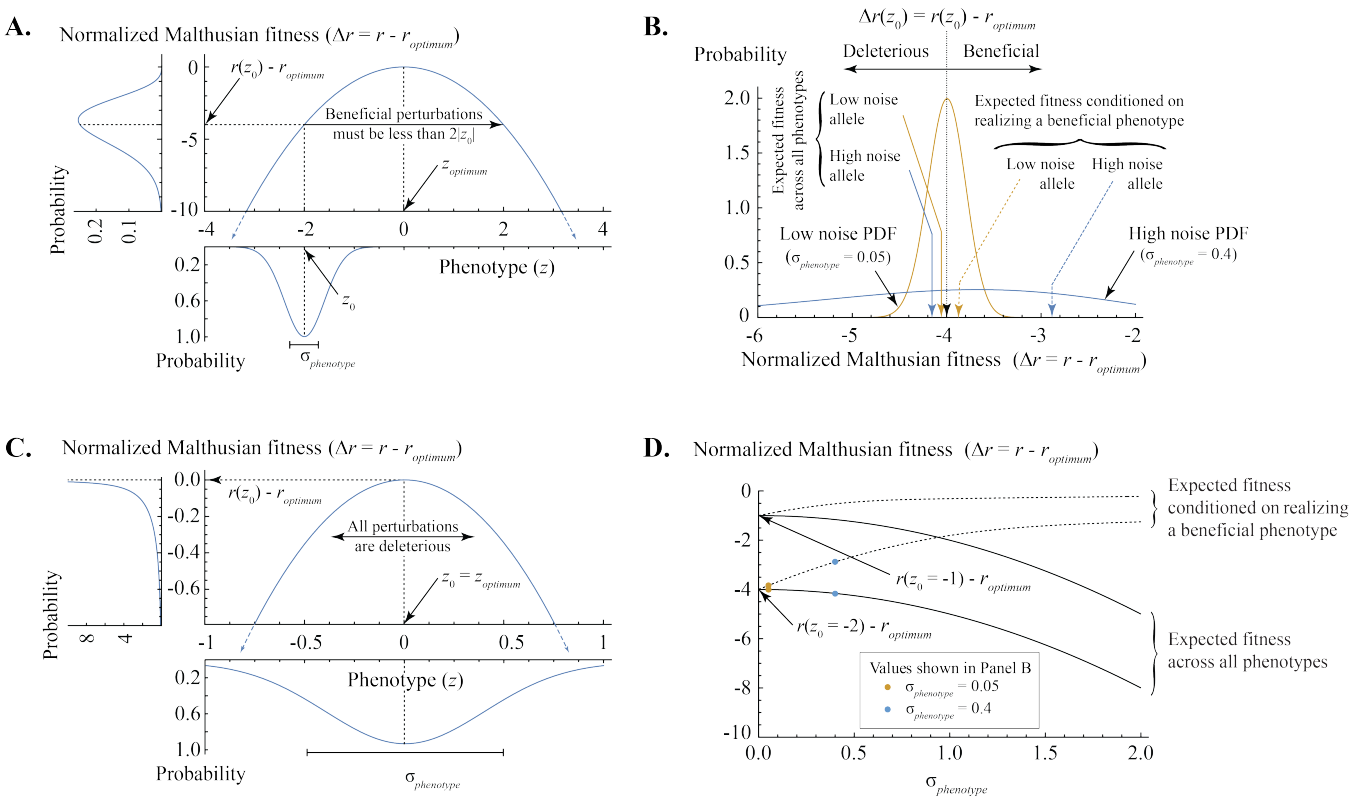

Figure 2: Fitness effects of phenotypic noise. **A.** Toy model introduced in text. Lower panel: Normally distributed phenotypic noise (Equation 1); in this figure, the organism's expected phenotype $E[z] = z_0 = -2$ and phenotypic noise $\sigma_{phenotype} = 0.4$. Main panel: Quadratic phenotype-to-fitness function, normalized by optimal fitness optimum (Equation 2). Without loss of generality we set the phenotypic optimum $z_{optimum} = 0$. Left panel: resulting probability density function of fitness values (Equation A1 in Appendix). Asymmetry induced by the fitness function's curvature is detailed in panels B and D. **B.** The probability density functions of fitness values induced by high (blue) and low (marigold) noise modifier alleles (Equation A1). High noise result the same as that shown at left in panel A, though axes have been rescaled. Low noise result shown for $\sigma_{\text{phenotype}} = 0.05$; all other parameters as in panel A. In expectation, noise is deleterious (algebraically, $E[r(z)] < r(E[z]) = r(z_0)$; see Equation A2). This can be seen by comparing the two solid, colored arrows with the black dotted arrow. But the expected fitness conditioned on inducing a beneficial phenotype (Equation A3) goes up as noise goes up. This can be seen by comparing two dashed arrows with the black dotted arrow. **C.** If an organism's expected phenotype is at the fitness optimum ($z_0 = z_{optimum} = 0$), all phenotypic noise is deleterious. All other parameters as in panel A, though note that the x- and y-axes have been rescaled to emphasize the effect. **D.** Expected fitness unconditioned on phenotype (solid, given by Equation A2) and conditioned on realizing a beneficial phenotypes (dashed, Equation A3) as a function of phenotypic noise ($\sigma_{phenotype}$) with expected phenotype (z_0) of -1 and -2.

down, the corresponding fitness consequences are not equal in magnitude. Rather, for a given (small) phenotypic effect size, deleterious perturbations will be more deleterious than beneficial perturbations are beneficial. (This effect is called Jensen's inequality [Jensen 1906].) And since all perturbations larger than $2|z_0|$ are deleterious, it follows that, averaged over phenotypic noise, the expected fitness effect of a modifier allele goes down as its noise goes up.

An important case occurs when the organism's expected phenotype is at the fitness optimum (algebraically, $E[z] = z_0 = z_{optimum}$). In this case all perturbations are deleterious, as illustrated in Figure 2C. Indeed, in populations at equilibrium between natural selection, mutation, recombination, migration and random genetic drift, any new, still-lower noise mutation, migration, or recombination modifier allele will always be selectively favored [Altenberg, Liberman, and Feldman 2017]. This result is called the *reduction principle*, and we speculate that it will generalize to all modifiers of reproductive and developmental noise.

Returning to the more general case ($E[z] = z_0 \neq z_{optimum}$), while noise is deleterious when averaged over all phenotypes (Equation A2; Jensen's inequality), high noise alleles can also sometimes realize higher fitness phenotypes than can low noise alleles (compare probability density functions in Figure 2B). We therefore also focus on a second quantity: a modifier allele's expected fitness conditioned on realizing a beneficial phenotype (Equation A3). Refining the point made at the outset, we suggest that it is the tension between these *unconditioned* and *conditioned fitness expectations* of phenotypic noise (algebraically, $E[r(z)]$ and $E[r(z)$

$| \, r(z) > r(z_0)]$, respectively) that lies at the core of modifier theory.

This tension is already evident in Figure 2B. While the unconditioned expected fitness effect of the high noise allele is lower than that of the low noise allele (compare solid arrows), the fitness effect conditioned on realizing a beneficial phenotype is higher for the high noise allele than for the low noise allele (compare dashed arrows). Figure 2D quantitatively illustrates the dependence of both expectations on $\sigma_{phenotype}$ under our toy model. As per Jensen's inequality, the unconditioned expectation (solid) goes down with noise, but the conditioned expectation (dashed) goes up before approaching a plateau.

These two features of Figure 2D imply that holding all else constant, there will exist a selective equilibrium for amount of noise in any biological mechanism. From there, higher noise modifier alleles will be disfavored because their now more modest conditioned fitness advantage is more than offset by their greater unconditioned fitness cost. And at the same time, lower noise modifier alleles will also be disfavored, because their inability to capitalize on anomalously high fitness phenotypic realization is not offset by their reduced unconditioned fitness cost.

An important determinant for the location of this selective equilibrium is phenotypic distance to the fitness optimum. The fitness of the expected organism ($r(\mathrm{E}[z]) = r(z_0)$) goes up as its phenotype z_0 approaches the optimum, so the remaining scope for conditional fitness improvement induced by noise goes down. (See Figure 2D.) Correspondingly, the equilibrium amount of noise will go down as z_0 approaches $z_{optimum} = 0$. Indeed, Figure 2C represents the limiting case: when the population is at the phenotypic optimum, indirect selection will always drive the amount of noise toward zero (see also Altenberg, Liberman, and Feldman [2017]).

As we will shortly see, three other factors also influence the location of this equilibrium: the persistence time between modifier allele and its induced, directly selected phenotypic variation, the mode of selection, and the population size.

A more general model

The assumptions underlying our toy model (Equations 1 and 2) are often implausible. Modifier alleles can have multidimensional phenotypic consequences, e.g., when they influence the pleiotropic effects of mutations [Draghi and Wagner 2008, Pavlicev, Cheverud, and Wagner 2011, Watson and Szathmáry 2016, Do O and Whitlock 2023]. Moreover, regardless of dimensionality, we have limited intuition for the distribution of fitness effects of phenotypic noise. And experimental access to these distributions is difficult for most of the mechanisms in Tables 1 and 2, though surveys of the distribution of mutational fitness effects [e.g. Eyre-Walker and Keightley 2007, Firnberg et al. 2014, Kemble, Nghe, and Tenaillon 2019] are one exception.

Nevertheless, we speculate that the existence of an equilibrium amount of biological noise may be quite robust to model specifics. For example, recent work on mutational interactions reveals that as fitness goes up, it is often the case that the advantage of beneficial mutations declines while the cost of deleterious mutations goes up (reviewed in Johnson, Reddy, and Desai [2023], Diaz-Colunga et al. [2023]). By Jensen's inequality, such negative curvature in the phenotype-to-fitness function is sufficient to cause the unconditioned fitness of modifier alleles to go down with noise. Conversely, the existence of a local optimum on the fitness landscape predicts that the conditioned, beneficial effect goes up with noise before approaching a plateau. It remains to be seen whether the fitness function induced by other forms of biological noise also exhibit negative curvature, although there are principled reasons to suppose that they may (e.g., in the case of developmental noise [Meiklejohn and Hartl 2002]). We return to this point in the Discussion

From fitness to fixation

We have argued that the crux of the problem for modeling the population genetics of modifier alleles is the tension between the deleterious and beneficial effects of biological noise. The evolutionary fate of such alleles is perhaps best understood in the case of modifiers of mutation rate.

The evolution of mutation rate in asexual organisms

High noise modifier alleles that heritably increase mutation rate (called *mutator* alleles) are commonly seen in environmental isolates of benign and pathogenic bacteria [LeClerc et al. 1996, Matic et al. 1997], of fungi [Lamb et al. 2008, Gambhir, Harris, and Everhart 2022], human viruses [Suárez, Valcárcel, and Ortín 1992, Amicone et al. 2022], and in human cancers [Bielas et al. 2006, Loeb 2011]. Similarly, laboratory-propagated microbial populations have evolved both mutator [Sniegowski, Gerrish, and Lenski 1997, Giraud et al. 2001, Raynes, Gazzara, and Sniegowski 2011, Swings et al. 2017, Bedhomme et al. 2019] and low noise (*antimutator*) alleles [Tröbner and Piechocki 1984, Giraud et al. 2001, McDonald et al. 2012, Wielgoss et al. 2013, Swings et al. 2017] (though see Johnson et al. [2021]).

In an ongoing laboratory experiment, 12 replicate asexual, bacterial populations have been serially propagated in liquid culture for over 75,000 generations in a fixed, novel environment [Lenski 2017]. After 10,000 generations, fitness had increased an average of $\sim 40\%$ across all 12 replicates [Wiser, Ribeck, and Lenski 2013], and over the same interval, mutation rate increased ~ 100-fold in three of them [Sniegowski, Gerrish, and Lenski 1997]. In all three of these populations, mutator alleles reached fixation via indirect selection mediated by beneficial mutations they had induced elsewhere in the genome [Shaver et al. 2002]. And by 40,000 generations, mutation rate had gone up in three more replicates [Tenaillon et al. 2016, Good et al. 2017].

Interestingly, many of the replicate populations that initially evolved mutator alleles subsequently fixed antimutator alleles [Wielgoss et al. 2013, Good et al. 2017]. Because the environment was held constant, the beneficial fitness effect of subsequent mutations diminished over time [Wünsche et al. 2017]. These diminishing returns then favored low noise modifier alleles via indirect selection for

their reduced deleterious mutational load (Wielgoss et al. [2013]; see also Tröbner and Piechocki [1984] for an earlier example of the same effect). Together, these results are consistent with our toy model: as fitness goes up, the amount of noise at selective equilibrium goes down (Figure 2D).

This experiment also draws our attention to a second determinant of the equilibrium amount of biological noise: the persistence time between modifier allele and induced phenotype. In the absence of recombination, mutation rate modifier alleles and their induced, directly selected mutations experience permanent genetic linkage. Because this experiment was performed with asexual bacteria, changes in frequency of mutator and antimutator alleles responded to whatever directly selected fitness effects they induced until they reached fixation or loss in the population. In contrast, recombination weakens indirect selection on mutation rate modifying alleles [Raynes, Gazzara, and Sniegowski 2011]. More abstractly, we thus predict that the amount of biological noise at selective equilibrium goes up with persistence time between a modifier allele and its induced phenotype [Otto 2013]. We develop this point more fully in the next section.

Population size also influences the equilibrium amount of biological noise. Firstly, across cellular life a very strong negative correlation exists between mutation rate and population size ($r^2 \approx 0.8$, [Lynch 2010]). The *drift-barrier hypothesis* [Lynch et al. 2016] explains this pattern by assuming that mutation rate evolution is dominated by the average, deleterious effects of high noise alleles. Since natural selection's efficiency goes up with population size [Fisher 1930, Wright 1931], indirect selection against high noise alleles should be more effective in large populations, explaining why mutation rate goes down as population size goes up in nature.

More recently, Raynes et al. [2018] refined this picture by demonstrating that the identify of the selectively favored mutation rate modifier allele depends on population size. To appreciate this finding, recall first that a new, selectively neutral mutation in a haploid population of N individuals reaches fixation with probability $1/N$ [Fisher 1930, Wright 1931]. This motivates normalizing the probability of fixation for any new mutation by this neutral benchmark (e.g., Good and Desai [2016]). Algebraically, we write $P_{fix}/(1/N) = NP_{fix}$, and for all directly selected mutations, this quantity diverges monotonically from the neutral benchmark as population size goes up (Figure 3A; the decline for deleterious mutations underlies the drift-barrier hypothesis just described).

The situation is quite different for mutation rate modifier alleles: as population size goes up, NP_{fix} for a new mutator allele drops below the neutral benchmark before reversing direction, and eventually it exceeds this value (Figure 3B; Raynes et al. [2018]). And the reverse pattern is seen for antimutator alleles. In other words, the identity of the favored mutation rate modifier allele depends on population size. Called *sign inversion* [Raynes et al. 2018], this effect is quite different from that for directly selected mutations described in the previous paragraph, and it has also been reported for modifier alleles determining whether reproduction is sexual or asexual (Figure 3C; Whitlock et al. [2016]). In both cases, NP_{fix} is concave up for the high noise allele (mutator, sexual reproduction) and concave down for the low noise allele (antimutator, asexual reproduction. We return to this point in the Discussion.

The underlying population genetics of sign inversion acting on mutation rate and sexual modifier alleles are fully developed in Raynes et al. [2018] and in Whitlock et al. [2016], respectively. Critically, sign inversion has also been described for modifier alleles that influence phenotypic noise [Gillespie 1974], evolutionary capacitance [Masel 2005], bet hedging [King and Masel 2007, Zhou and Xue 2022, Weissman, Raynes, and Weinreich 2023], ploidy [Raynes and Weinreich 2021] and for a modifier allele that influences a trait's genetic architecture [Polizzi et al. 2024]. Recalling the intrinsic double-edged consequences of phenotypic noise, it appears that sign inversion emerges because the threshold between random genetic drift and selection scales differently for the modifier allele's conditional beneficial and unconditional deleterious fitness consequences [Raynes, Burch, and Weinreich 2021]. We return to this point in the Discussion as well.

Beyond mutation rate modifiers in asexual populations

In any generation, indirect selection favors or disfavors a modifier allele via direct selection on the fitness effects of whatever beneficial or deleterious phenotypes it happens to induce in its carrier(s). At one limit, the foregoing treatment of mutation rate evolution was simplified by its assumption of an asexual population, meaning that modifier alleles and their induced, directly selected mutations remained genetically linked until the modifier allele's ultimate fixation or loss. At the other limit, imagine a modifier allele that causes phenotypes to be resampled every generation. In this case each modifier allele's induced phenotype (and thus, its directly selected fitness effect) can change in each generation. Gillespie [1974] showed that in this case, high noise modifier alleles are unconditionally deleterious in the absence of direct selection in their favor. This follows because, while still rare, the risk of stochastic loss is of much greater evolutionary consequence than is the opportunity of the corresponding stochastic increase in frequency. (This so-called *absorbing barrier* at frequency zero underlies many results in stochastic models of population biology. See Chapter 2 in Weinreich [2023] for several examples.)

In between these two limits, Carja and Plotkin [2017] showed that even a transient association between modifier allele and induced phenotype can be sufficient for indirect selection to favor high noise alleles. Thus even in sexual populations, indirect selection can act on mutation rate modifiers as well as on other modifiers of reproduction. (Indirect selection on modifiers of reproduction is also robust to recombination if their directly selected mutations are distributed across the genome (e.g., Draghi and Wagner [2008, 2009] and Whitlock et al. [2016].)

Critically for our purposes, results in Carja and Plotkin [2017] apply equally to modifiers of development, which similarly respond to indirect selection if they experience

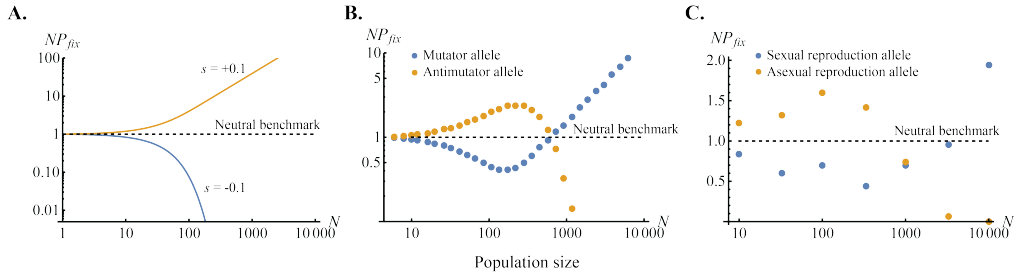

Figure 3: Probability of fixation for new allele as a function of population size, normalized by $1/N$, the neutral benchmark in a population of N haploid individuals. Thus NP_{fix} for a new neutral allele is 1 for all N. **A.** For directly selected mutations, NP_{fix} diverges monotonically from the neutral benchmark as population size goes up. (Drawn with Equation 8 in Kimura [1962].) **B** and **C.** In contrast, individual-based simulations show sign inversion [Raynes et al. 2018] at two modifier loci. When NP_{fix} crosses the neutral benchmark, the identity of the selectively favored modifier allele changes. **B.** High noise (blue) and low noise (marigold) mutation rate modifier alleles. Here, 1% of new, directly selected mutations have a fitness advantage s = +10%, the remaining 99% have a fitness deficit s = -1%, the high noise allele increases mutation rate 100-fold, and the wild type allele's deleterious mutation rate is 10^{-4} per individual per generation. (The same effect was demonstrated in the lab using hundreds of replicate populations of *Saccharomyces cerevisiae* Raynes et al. [2018].) Simulation results for the mutator allele are from Raynes et al. [2018] and for the antimutator allele are unpublished data of ours. **C.** High noise, sexual (blue) and low noise, asexual (marigold) alleles. Simulation results from Whitlock et al. [2016] kindly provided by Christina Burch. In both panels, NP_{fix} is concave up for the high noise allele and concave down for the low noise allele.

even a few generations of association with their induced phenotypes. For example, consider an allele in a single-celled organism that increases variance in the cellular concentration of some particular mRNA. Now imagine an individual in which a high-noise allele has induced a selectively favorable, anomalously high (or low) concentration of that molecule. While subsequent cell divisions will dilute such perturbations back toward the mean, that decay will be roughly exponential, meaning that indirect selection will be able to act on the high noise allele for at least a few cell generations. Another example comes from transgenerational phenotypic plasticity in multicellular organisms, which often persists for two to five generations, and persistence for even 40 generations has been described in *C. elegans* (reviewed in Bell and Hellmann [2019]).

Finally, note that the time to fixation for a beneficial mutation scales as the reciprocal of its fitness advantage (e.g., Weinreich [2023]). Thus, if the persistence time between modifier allele and an induced, directly selected beneficial phenotype lasts approximately that long, indirect selection will be as effective as it would be if the association were permanent. Timescales of persistence times of the association between modifier allele and their induced phenotypes are shown for each case in Tables 1 and 2.

Beyond fitness-proportional selection

Work summarized above assumes fitness-proportional selection. In contrast, Kolodny and Stern [2017] showed that departures from fitness-proportional selection such as truncation selection can drive high noise alleles to fixation, even if phenotypes are resampled every generation. Under truncation selection, all organisms that exceed some phenotypic threshold contribute equally to the next generation, and it emerges in many biological situations (reviewed in Kolodny and Stern [2017], Morsky and Bauch [2019]), such as when males compete for reproductive access to females. Now allowing biological noise, reproducing individuals under truncation selection will include those that happen by good luck to have experienced a phenotype above the threshold. These in turn will be enriched for individuals carrying high-noise alleles. Put another way, truncation selection reduces the fitness penalty induced by high noise alleles, which explains why high noise alleles can succeed even if phenotype is resampled every generation. Indeed, even less extreme departures from fitness proportional selection can be sufficient to favor high noise alleles in the absence of any persistence in association with induced phenotype [Kolodny and Stern 2017]. Consequently, the amount of noise expected at the selective equilibrium will also depend on the mode of selection.

Beyond constant environments

Thus far, we have largely assumed that the distribution of fitness effects of new phenotypes (e.g., the two distributions in Figure 2B) remains constant in time. The one exception involved the observation that antimutators eventually fixed in some lab-reared, asexual bacterial populations [Wielgoss et al. 2013, Good et al. 2017]. The interpretation of those results was that, as populations became better adapted to the lab environment, the opportunities for further mutational improvement declined, tipping the balance of indirect selection in favor of low noise mutation rate modifiers (Wielgoss et al. [2013]; compare lines corresponding to z_0 = -1 and -2 in Figure 2D). This line of reasoning also brought our attention to the limiting case: high noise alleles are unconditionally deleterious in a population at (Figure 2C) or very near (Altenberg, Liberman, and Feldman [2017]) the phenotypic optimum.

However, natural environments are almost always temporally variable. This means that evolving populations are

often chasing an ever-changing fitness optimum. The more quickly the environment changes, the greater the lag between the population and its phenotypic optimum, thus shifting the equilibrium in favor of high noise modifier alleles. We return to this point in the Discussion.

Another critical issue is the interplay between the timescale of environmental variation and that of organismal development. Our treatment of developmental noise has assumed that any particular individual samples its phenotype once per generation from a fixed distribution. However, development is not an instantaneous process, and stochastic phenotypic perturbations may thus be sampled repeatedly within a single generation. This is perhaps especially important in long-lived organisms, in which each individual displays a number of phenotypes during its lifespan. Moreover, the distributions from which these perturbations are sampled can themselves vary over an organism's lifetime [e.g. Bulté and Bennoun 1990, Dupont et al. 2024]. Similarly, in multicellular organisms, developmental noise can be sampled independently across cells [Bravo, Belkhir, and Paget-Bailly 2024] or cell types [Wolf et al. 2023]. It appears plausible that if the environment varies substantially within a generation, these sorts of effects may magnify the evolutionary importance of modifiers of development, but much work remains to be done in this area.

Discussion

We have argued that the population genetics of biological noise is critically dependent on indirect selection, i.e., selection mediated by statistical associations between noise modifying alleles and whatever directly selected phenotypes they induce in their carriers. Going further, we have highlighted an intrinsic tension between the occasionally beneficial phenotypes induced by noise and its much more commonly deleterious consequences. We suggest that for any source of noise, there will exist an equilibrium level at which these two consequences are balanced, determined by four factors: the rate of environmental change, the timescale of association between modifier allele and induced phenotype, the population size and the mode of selection.

Prior art

As noted at the outset, we are far from the first to explore the influence of indirect selection on modifier alleles. The use of the word "modifier" in this sense dates to R.A. Fisher's work on the evolution of dominance [Fisher 1928]. That paper first reviewed data showing that when more than two alleles (*allelomorphs* in the parlance of the time) are known at a locus, the wild type is typically dominant over all mutants, but that most pairs of mutants exhibit codominance. Fisher reasoned that

> This feature in the behavior of multiple allelomorphs appears to offer a serious difficulty to the theory that the evolutionary adaptation of specific forms has taken place by the occasional and gradual replacement by mutant genes of the allelomorphic wild type genes from which they arose. For, in their dominance, the wild type genes appear to be clearly of a different nature from the mutant genes which arise from them.

But he concluded that

> This difficulty will lose its force if it appears that there is a tendency always at work in nature which modifies the response of the organism to each mutant gene in such a way that the wild type tends to become dominant.

Fisher thus posited the existence of modifier loci, "other hereditary factors that influence the reaction of the organism to the mutant gene." The rest of his paper laid out a theory for how natural selection might act on such a factor.

Fisher was also the first to articulate the distinction between direct and indirect selection, writing [Fisher 1932] that the presentations in a session entitled "Contributions of genetics to the theory of evolution" at the 6th International Congress of Genetics in Ithaca, New York

> are of two kinds, somewhat sharply contrasted, [as] illustrated by comparing HALDANE'S subject, "Can evolution be explained in terms of present known genetical causes?" with ... [m]y own address, which might equally well have been entitled, "Can genetical phenomena be explained in terms of known evolutionary causes?"

Thus, Haldane and Fisher were concerned with direct and indirect selection, respectively. This causal bidirectionality is also nicely articulated in Do O and Whitlock [2023].

Fisher's focus on indirect selection acting on an organism's genetics (what we call modifiers of reproduction) anticipated most modifier theory to the present day. Sturtevant [1937] argued that the deleterious burden of mutation would cause indirect selection to drive mutation rate as low as biologically possible. And Nei [1967] was the first to take up the problem of indirect selection acting on modifiers of recombination rate.

An early generalization came from Karlin and McGregor [1972], who wrote, "The genotypes at the secondary (modifier) loci alter the determination of the parameter values delimiting the genetic system and mechanisms corresponding to the primary loci." Those authors then explored five modifiers of reproduction: modifiers of recombination, mutation, migration (or more strictly, dispersal – see Table 2), selfing, and dominance. Finally, Altenberg and Feldman [1987] proposed that modifier theory be defined as the population genetics of loci that influence the transmission process.

This idea appears to be fully congruent with our definition of modifiers of reproduction (Figure 1D and E). As seen in Table 2, our definition subsumes all traditionally recognized modifier alleles. But by focusing on mechanisms rather than model parameters, our definition draws attention to several modifiers of reproduction not widely recognized in this literature, e.g., modifiers of pleiotropy and of non-genetic inheritance. More broadly, defining modifiers as loci that influence biological noise also draws attention to alleles that heritably influence noise in development (Table 1). We believe this usage is original to us, though antecedents can be found in Masel and Bergman [2003], Masel [2005], King

and Masel [2007], Salathé, Van Cleve, and Feldman [2009], Carja, Liberman, and Feldman [2014]. But our perspective is closely related to established ideas in the evolution of evolvability literature, as we discuss next.

The evolution of evolvability

Darwin's model for natural selection is explicitly predicated on an evolving population's capacity to generate heritably adaptive phenotypic variants, referred to as *evolvability*, [Burch and Chao 2000, Sniegowski and Murphy 2006, Masel and Trotter 2010]. And so it was quite surprising to evolutionary biologists when computer scientists found that natural selection is often incapable of evolving improved computer programs [Wagner and Altenberg 1996]. This finding in turn focused attention on identifying those features of biological systems that do render them evolvable. Key among them are organisms' modularity [Wagner and Altenberg 1996, Pigliucci 2008, Draghi and Wagner 2008, Colegrave and Collins 2008, Clune, Mouret, and Lipson 2013], robustness [Wagner 2005, Colegrave and Collins 2008, Masel and Trotter 2010, Payne and Wagner 2019], and other features of the genotype-to-phenotype function [e.g. Wagner and Altenberg 1996, Ancel and Fontana 2000, Draghi and Wagner 2009, and citations therein].

While the features that make biological organisms evolvable are outside our scope, the population genetic theory developed here appears to provide a useful framework for addressing a closely related question: How and when will natural selection influence a population's evolvability? This follows from the fact that, like the evolution of the amount of biological noise, the evolution of evolvability is concerned with a population's capacity to generate phenotypic novelty. These two lines of inquiry apparently differ in that the former focuses on all phenotypic variation, while the latter focuses only on beneficial variation. As we now argue, this is a false dichotomy: they are really addressing the same problem.

We are not the first to see connections between modifier theory and the evolution of evolvability. For some authors, modifiers of mutation rate [Partridge and Barton 2000, Sniegowski and Murphy 2006, Díaz Arenas and Cooper 2013], recombination rate [Draghi and Wagner 2009, Masel and Trotter 2010, Díaz Arenas and Cooper 2013], or of the distribution of mutational fitness effects [Burch and Chao 2000] are canonical examples of how selection can increase evolvability. And others have explicitly noted the essential role of indirect selection in the evolution of evolvability [Sniegowski and Murphy 2006, Colegrave and Collins 2008, Draghi and Wagner 2008, Masel and Trotter 2010, Wagner 2022]. Indeed, the distinction between evolvability-as-adaptation and evolvability-as-byproduct [Sniegowski and Murphy 2006, Lynch 2007] appears to exactly mirror the distinction between direct and indirect selection that is foundational to the present work.

While several modifiers of reproduction are invoked as textbook examples of how selection can favor increased evolvability, much of the literature on the evolution of evolvability is focused on the heritable control of the statistical structure of noise in the developmental process [e.g. Wagner and Altenberg 1996, Kirschner and Gerhart 1998, Dawkins 1989, Draghi and Wagner 2008, Pigliucci 2008, Watson and Szathmáry 2016, Watson 2020]. And as we have argued, indirect selection can influence the frequency of modifiers of development, even though the resulting phenotypes are not genetically encoded. It thus appears that modifier theory as construed here can contribute to understanding how and when natural selection will influence many aspects of a population's evolvability.

Most importantly, many authors have raised teleological concerns about the hypothesis that natural selection can favor increased evolvability [e.g. Weber 1996, Sniegowski and Murphy 2006, Pigliucci 2008, Paaby and Testa 2018]. Seeing the evolution of evolvability as the population genetics of alleles that influence the amount of biological noise immediately resolves these concerns, because our treatment starts from the fact that biological noise is a double-edged sword. Thus, we have argued that indirect selection will increase biological noise only when such increases in the population's capacity to generate adaptive phenotypic variants (i.e., its evolvability) outweigh the concomitant increased cost of generating deleterious variants. Otherwise, indirect selection will reduce biological noise because this reduction in the population's capacity to generate deleterious phenotypes (what might be called its *antievolvability*) now outweighs the concomitant opportunity for generating adaptive phenotypes. Both effects are admirably illustrated by the evolution of mutation rate in the decades-long bacterial experiments described above.

Future work

We have proposed a framework for understanding the influence that indirect selection has on the amount of any form of biological noise in any species. However, much work remains before we can make confident inferences about the importance of this effect in nature. We require both quantitative models to which data can be fit, and data that isolate the consequences of indirect selection from other processes that may influence the amount of biological noise.

From fitness to fixation (and beyond) under our toy model. Our toy model (Eqns 1 and 2) yields intuitively compelling predictions for the fitness effects of modifier alleles as a function of their phenotypic noise (Figure 2D). This suggests that it may similarly be an instructive tool with which to quantitatively explore their fixation and loss. We see three opportunities.

1. The first challenge will be to quantitatively locate the amount of phenotypic noise at the selective equilibrium under our toy model as a function of the four quantities previously discussed: the population's phenotypic distance from the optimum, the timescale of association between modifier allele and induced, directly selected phenotype, the population size, and the mode of selection. Individual-based computer simulations will be useful, but we are hopeful that it will also be able to write down one-step transition probabilities as a function of modifier allele frequency in the population. From there, textbook methods (see for example Weinreich [2023] and citations therein) may yield analytic access

to fixation probabilities for new modifier alleles, and thus, to the equilibrium amount of noise.

2. That the direction of selection might vary with population size (i.e., sign inversion; see Figure 3) is outside the classical understanding of population genetics. This effect has been described for several modifier alleles [Gillespie 1974, King and Masel 2007, Whitlock et al. 2016, Raynes et al. 2018, Zhou and Xue 2022, Weissman, Raynes, and Weinreich 2023], leading us to speculate that all modifier alleles will exhibit this feature. Some progress has been made [Raynes, Burch, and Weinreich 2021] in this respect, and we suggest that our toy model may be an ideal platform with which to develop a deeper understanding of the effect. For example, is it true that NP_{fix} for all high (low) noise alleles is concave up (down)? And what does the answer to this question teach us about modifier theory?

3. Finally, modeling recurrent mutation at the modifier locus means that a population may experience periodic, stochastic perturbations around the evolutionary equilibrium described in the paragraph before last. Desai and Fisher [2011] explored this effect for modifiers of mutation rate and showed that even at equilibrium, new mutator or antimutator alleles may have the good luck to transiently rise to fixation by random genetic drift. They concluded that the mutation rate observed in any population may thus have as much to do with recent history as with the equilibrium rate predicted by theory. It will be important to incorporate all these ideas into our toy model, in order to anticipate manifestations of these effects in real experimental or observational data.

Beyond our toy model. Our toy model makes biologically unrealistic assumptions, but it can be replaced by any other distribution of fitness effects induced by biological noise. We are optimistic that closer scrutiny of the sources of biological noise enumerated in Tables 1 and 2 will yield compelling alternatives. Indeed, Cotto and Day [2023] recently provided a model inspired by data on the distribution of mutational fitness effects. Lo et al. [2024] and Wang and Zhang [2011] represent two other principled approaches. In any case, once so defined, each of the foregoing three questions can be reexamined, to assess the robustness of results to model assumptions.

Experimental opportunities. We have explored how indirect selection can influence the amount of noise in any biological mechanism. The tension between the average, deleterious fitness consequences of such noise and the possibility of rare, beneficial fitness effects is central to the population genetic framework we propose. How can one demonstrate the influence of this tension in any natural population?

Like a great many other domains of biology, recent technological advances have revolutionized empirical access to biological noise. In the case of developmental noise, cell-to-cell variability in transcription and translation rates have been measured at almost genome-wide scale with fluorescent protein constructs in *E. coli* [Silander et al. 2012], and for a library of promoters at a single locus in yeast [Duveau et al. 2018]. Similar technology was recently deployed to characterize the time course of individual bacteriophage infections [Wedd et al. 2024]. Genome-wide transcriptional fidelity [e.g. Gout et al. 2017] and rate variation [e.g. Ochiai et al. 2020] can be assessed by single-cell RNA sequencing. Translational fidelity can also be quantified [Mordret et al. 2019], though not at the single cell level (reviewed in Evans et al. [2018]). And single-cell metabolomics via mass spectrometry is now within reach [e.g. Guo, Zhang, and Le 2021]. Finally, behavioral noise in animals can also be quantified using high-throughput assays (e.g., in fruit flies Ayroles et al. [2015], Maloney et al. [2024] and in *C. elegans* Webster et al. [2023]).

Tremendous progress is also being made in the empirical assessment of reproductive noise. Whole genome sequence data from parent-offspring trios yield per-meiosis mutation rate estimates [e.g. Kong et al. 2012]. Single-sperm sequencing provides high resolution per-meiosis recombination maps [e.g. Bell et al. 2020], and epigenetic modifications can now be assessed at single-cell, multilocus scale [Meers et al. 2023]. Similarly, deep mutational scanning [Fowler and Fields 2014] gives direct access to the distribution of mutational [e..g. Firnberg et al. 2014] and epistatic Haddox et al. [e.g. 2018] effects for phenotypes of interest.

Building on work of others (Sniegowski et al. [2000], Peck and Lauring [2018], J. Masel, *pers. comm*), we propose a simple, four-step experimental design for each such experimental methodology to assess the influence that indirect selection has on the corresponding form of biological noise.

1. Perform experiments of the sort schematized in Figure 1B-E, to demonstrate the presence of modifier alleles influencing the relevant biological mechanism. One complication not explicitly contemplated here is that the amount of biological noise in an organism can be a complex genetic trait determined by allelic state at multiple loci [e.g. Ansel et al. 2008].

2. Characterize the mutational bias acting at the corresponding modifier locus. For example in bacteria, mutator alleles often correspond to loss-of-function mutations [e.g. Denamur and Matic 2006], implying a mutational bias toward high noise alleles. But conversely, one might suppose on first principles that the evolutionary transition rate from sexuals to asexuals will be higher than the reverse (inasmuch as it should be easier to disrupt sexual reproduction than to create it *de novo*). This implies a mutational bias toward the low noise allele in that case. As always, it is critical to establish the baseline mutational expectation before ascribing a selective effect [Kimura 1983].

3. Next, possible sources of direct selection acting on the modifier allele must be assessed. As for mutational bias, it appears likely that no general rules will exist. For example, whereas low noise mutation rate modifier alleles are plausibly more energetically costly than are high noise alleles, high noise sexual reproduction is certainly the more costly alternative. Moreover, metabolic cost is unlikely to be the only directly selected feature of modifier alleles; recall for example the case of accelerated genome duplication associated with mutator alleles in viruses [Elena and Sanjuán 2005, Furió, Moya, and Sanjuán 2005].

4. Only after mutational and direct fitness biases have been accounted for can we infer the action of indirect se-

lection. More specifically, given evidence of genetic control on some particular form of biological noise, deviations between the amount of noise observed and that predicted by mutational and direct fitness effects can be ascribed to the action of indirect selection. (See [Parée et al. 2024] for an exciting illustration of how this can be done.) At that point, data can be fit to the sorts of models contemplated in the previous section, in order to estimate the contribution that the rate of environmental change, the persistence of the association between modifier allele and induced phenotype, the population size and the model of selection have had on the amount of biological noise observed.

Conclusions

The population genetics of alleles that influence mean trait values has been well established for nearly a century. And while an appreciation of the evolutionary importance of genetic variation in noise around mean trait values goes back almost that long, we have lacked a unified population genetic framework [Graves and Weinreich 2017]. We suggest that the treatment developed here represents an important first step. Although a great many details remain to be developed, the very broad swath of biologists now interested in biological noise underscores the value of the sorts of ideas developed here.

Acknowledgments

This work emerged from a graduate seminar at Brown University that D.M.W. taught during the spring semester of 2020, and we thank Nigel Anderson, Alan Denadel, Gabriella Ferra, Lourdes Gomez, Jiaying Lai, Robert Lamb, Miles, David Morgan, Chibuikem Nwizu, Maya Weissman and Hannah Weller for early contributions. Guillaume Achaz and Christina Burch provided several trenchant suggestions late in the writing. E.C. and S.R. were supported by the Brown University SPRINT|UTRA program, and H.B. was supported by the Glen Scheinberg 2008 Research Fund. D.M.W. is grateful for financial support from the Brown University Faculty Research Fund during his Fall 2024 sabbatical in Montpellier, France, during which much of this work was completed. He also acknowledges space and intellectual support during that visit from the *Équipe Génétique et Écologie Évolutive* at the *Centre d'Écologie Fonctionnelle et Évolutive* in Montpellier, and by members of the *Maladies Infectieuses et Vecteurs: Écologie, Génétique, Évolution et Contrôle* laboratory in Montpellier. I.G.B. was supported by the European Union's Horizon 2020 Research and Innovation Program (ERC-CoG-647916).

Author Contributions

DMW conceived the idea for this manuscript, DMW, CP, TS, YR and IGB developed the ideas presented and contributed to the conceptual framework. DMW, CP, TS and IGB wrote the manuscript. DMW, CP, HB, EC, and SR conducted the literature review.

Competing interests

The authors declare no competing interests.

References

Ackermann, M.; Chao, L.; Bergstrom, C. T.; and Doebeli, M. 2007. On the evolutionary origin of aging. *Aging Cell*, 6(2): 235–244.

Altenberg, L.; and Feldman, M. W. 1987. Selection, generalized transmission and the evolution of modifier genes. I. The reduction principle. *Genetics*, 117(3): 559–572.

Altenberg, L.; Liberman, U.; and Feldman, M. W. 2017. Unified reduction principle for the evolution of mutation, migration, and recombination. *Proceedings of the National Academy of Sciences*, 114(12): E2392–E2400. Publisher: Proceedings of the National Academy of Sciences.

Amicone, M.; Borges, V.; Alves, M. J.; Isidro, J.; Zé-Zé, L.; Duarte, S.; Vieira, L.; Guiomar, R.; Gomes, J. P.; and Gordo, I. 2022. Mutation rate of SARS-CoV-2 and emergence of mutators during experimental evolution. *Evolution, Medicine, and Public Health*, 10(1): 142–155.

Ancel, L. W.; and Fontana, W. 2000. Plasticity, evolvability, and modularity in RNA. *Journal of Experimental Zoology*, 288(3): 242–283.

Andersson, M. B. 1994. *Sexual selection*. Monographs in behavior and ecology. Princeton, N.J: Princeton University Press. ISBN 978-0-691-20727-8.

Ansel, J.; Bottin, H.; Rodriguez-Beltran, C.; Damon, C.; Nagarajan, M.; Fehrmann, S.; François, J.; and Yvert, G. 2008. Cell-to-Cell Stochastic Variation in Gene Expression Is a Complex Genetic Trait. *PLoS Genetics*, 4(4): e1000049.

Archetti, M. 2009. Survival of the steepest: hypersensitivity to mutations as an adaptation to soft selection. *Journal of Evolutionary Biology*, 22(4): 740–750.

Atkins, W. M. 2015. Biological messiness vs. biological genius: Mechanistic aspects and roles of protein promiscuity. *The Journal of Steroid Biochemistry and Molecular Biology*, 151: 3–11.

Ayroles, J. F.; Buchanan, S. M.; O'Leary, C.; Skutt-Kakaria, K.; Grenier, J. K.; Clark, A. G.; Hartl, D. L.; and De Bivort, B. L. 2015. Behavioral idiosyncrasy reveals genetic control of phenotypic variability. *Proceedings of the National Academy of Sciences*, 112(21): 6706–6711.

Balkau, B. J.; and Feldman, M. W. 1973. Selection of Migration Modification. *Genetics*, 74(1): 171–174.

Becks, L.; and Agrawal, A. F. 2010. Higher rates of sex evolve in spatially heterogeneous environments. *Nature*, 468(7320): 89–92.

Bedhomme, S.; Amorós-Moya, D.; Valero, L. M.; Bonifaci, N.; Pujana, M.-; and Bravo, I. G. 2019. Evolutionary Changes after Translational Challenges Imposed by Horizontal Gene Transfer. *Genome Biology and Evolution*, 11(3): 814–831.

Bell, A. D.; Mello, C. J.; Nemesh, J.; Brumbaugh, S. A.; Wysoker, A.; and McCarroll, S. A. 2020. Insights into variation in meiosis from 31,228 human sperm genomes. *Nature*, 583(7815): 259–264.

Bell, A. M.; and Hellmann, J. K. 2019. An Integrative Framework for Understanding the Mechanisms and Multigenerational Consequences of Transgenerational Plastic-

ity. *Annual Review of Ecology, Evolution, and Systematics*, 50(1): 97–118.

Bielas, J. H.; Loeb, K. R.; Rubin, B. P.; True, L. D.; and Loeb, L. A. 2006. Human cancers express a mutator phenotype. *Proceedings of the National Academy of Sciences*, 103(48): 18238–18242.

Bonduriansky, R.; and Day, T. 2009. Nongenetic Inheritance and Its Evolutionary Implications. *Annual Review of Ecology, Evolution, and Systematics*, 40(1): 103–125.

Bravo, I. G.; Belkhir, S.; and Paget-Bailly, P. 2024. Why HPV16? Why, now, HPV42? How the discovery of HPV42 in rare cancers provides an opportunity to challenge our understanding about the transition between health and disease for common members of the healthy microbiota. *FEMS Microbiology Reviews*, 48(6): fuae029.

Bruggeman, F. J.; and Teusink, B. 2018. Living with noise: On the propagation of noise from molecules to phenotype and fitness. *Current Opinion in Systems Biology*, 8 : 144–150.

Bulté, L.; and Bennoun, P. 1990. Translational accuracy and sexual differentiation in Chlamydomonas reinhardtii. *Current Genetics*, 18(2): 155–160.

Burch, C. L.; and Chao, L. 2000. Evolvability of an RNA virus is determined by its mutational neighbourhood. *Nature*, 406(6796): 625–628.

Çağatay, T.; Turcotte, M.; Elowitz, M. B.; Garcia-Ojalvo, J.; and Süel, G. M. 2009. Architecture-Dependent Noise Discriminates Functionally Analogous Differentiation Circuits. *Cell*, 139(3): 512–522.

Carja, O.; Liberman, U.; and Feldman, M. W. 2014. Evolution in changing environments: Modifiers of mutation, re-combination, and migration. Proceedings of the National Academy of Sciences, 111(50): 17935–17940. Publisher: Proceedings of the National Academy of Sciences.

Carja, O.; and Plotkin, J. B. 2017. The evolutionary advantage of heritable phenotypic heterogeneity. Scientific Reports, 7(1): 5090. Number: 1 Publisher: Nature Publishing Group.

Chakravarty, D.; Sreenivasan, S.; Swint-Kruse, L.; and Porter, L. L. 2023. Identification of a covert evolutionary pathway between two protein folds. *Nature Communications*, 14(1): 3177.

Charlesworth, B. 2013. Stabilizing Selection, Purifying Selection, and Mutational Bias in Finite Populations. *Genetics*, 194(4): 955–971.

Chasnov, J. R. 2000. Mutation-Selection Balance, Dominance and the Maintenance of Sex. *Genetics*, 156(3): 1419–1425.

Clune, J.; Mouret, J.-B.; and Lipson, H. 2013. The evolutionary origins of modularity. *Proceedings of the Royal Society B: Biological Sciences*, 280(1755): 20122863.

Colegrave, N.; and Collins, S. 2008. Experimental evolution: experimental evolution and evolvability. *Heredity*, 100(5): 464–470.

Copley, S. D. 2017. Shining a light on enzyme promiscuity. *Current Opinion in Structural Biology*, 47: 167–175.

Cotto, O.; and Day, T. 2023. A null model for the distribution of fitness effects of mutations. *Proceedings of the National Academy of Sciences*, 120(23): e2218200120.

Couce, A.; Guelfo, J. R.; and Blázquez, J. 2013. Mutational Spectrum Drives the Rise of Mutator Bacteria. *PLoS Genetics*, 9(1): e1003167.

Couce, A.; and Tenaillon, O. 2019. Mutation bias and GC content shape antimutator invasions. *Nature Communications*, 10(1): 3114.

Dawkins, R. 1989. The Evolution of Evolvability. In Langdon, C., ed., *Artificial Life: Proceedings Of An Interdisciplinary Workshop On The Synthesis And Simulation Of Living Systems*, 201–220. New York: Addison-Wesley.

De Cara, M.; Barton, N.; and Kirkpatrick, M. 2008. A Model for the Evolution of Assortative Mating. *The American Naturalist*, 171(5): 580–596.

Denamur, E.; and Matic, I. 2006. Evolution of mutation rates in bacteria. *Molecular Microbiology*, 60(4): 820–827.

Desai, M. M.; and Fisher, D. S. 2011. The Balance Between Mutators and Nonmutators in Asexual Populations. *Genetics*, 188(4): 997–1014.

Diaz-Colunga, J.; Skwara, A.; Gowda, K.; Diaz-Uriarte, R.; Tikhonov, M.; Bajic, D.; and Sanchez, A. 2023. Global epistasis on fitness landscapes. *Philosophical Transactions of the Royal Society B: Biological Sciences*, 378(1877): 20220053.

Do O, I.; and Whitlock, M. C. 2023. The evolution of genetic covariance and modularity as a result of multigenerational environmental fluctuation. *Evolution Letters*, 7(6): 457–466.

Draghi, J.; and Wagner, G. P. 2008. The Evolution of Evolvability in a Developmental Model. *Evolution*, 62(2): 301–315.

Draghi, J.; and Wagner, G. P. 2009. The evolutionary dynamics of evolvability in a gene network model. *Journal of Evolutionary Biology*, 22(3): 599–611.

Drummond, D. A.; and Wilke, C. O. 2009. The evolutionary consequences of erroneous protein synthesis. *Nature Reviews Genetics*, 10(10): 715–724.

Dupont, L.; Thierry, M.; Zinger, L.; Legrand, D.; and Jacob, S. 2024. Beyond reaction norms: the temporal dynamics of phenotypic plasticity. *Trends in Ecology & Evolution*, 39(1): 41–51.

Duveau, F.; Hodgins-Davis, A.; Metzger, B. P.; Yang, B.; Tryban, S.; Walker, E. A.; Lybrook, T.; and Wittkopp, P. J. 2018. Fitness effects of altering gene expression noise in Saccharomyces cerevisiae. *eLife*, 7: e37272.

Díaz Arenas, C.; and Cooper, T. F. 2013. Mechanisms and selection of evolvability: experimental evidence. *FEMS Microbiology Reviews*, 37(4): 572–582.

Elena, S. F.; and Sanjuán, R. 2005. Adaptive Value of High Mutation Rates of RNA Viruses: Separating Causes from Consequences. *Journal of Virology*, 79(18): 11555–11558.

Eling, N.; Morgan, M. D.; and Marioni, J. C. 2019. Challenges in measuring and understanding biological noise. *Nature Reviews Genetics*, 20(9): 536–548.

Evans, C. R.; Fan, Y.; Weiss, K.; and Ling, J. 2018. Errors during Gene Expression: Single-Cell Heterogeneity, Stress Resistance, and Microbe-Host Interactions. *mBio*, 9(4): e01018–18.

Eyre-Walker, A.; and Keightley, P. D. 2007. The distribution of fitness effects of new mutations. *Nature Reviews Genetics*, 8(8): 610–618.

Ferrare, J. T.; and Good, B. H. 2024. Evolution of evolvability in rapidly adapting populations. *Nature Ecology & Evolution*.

Firnberg, E.; Labonte, J. W.; Gray, J. J.; and Ostermeier, M. 2014. A Comprehensive, High-Resolution Map of a Gene's Fitness Landscape. *Molecular Biology and Evolution*, 31(6): 1581–1592.

Fisher, R. A. 1928. The Possible Modification of the Response of the Wild Type to Recurrent Mutations. *The American Naturalist*, 62(679): 115–126.

Fisher, R. A. 1930. *The genetical theory of natural selection*. Oxford: Clarendon Press.

Fisher, R. A. 1932. The Evolutionary Modification of Genetic Phenomena. *Proceedings of the 6th International Congress of Genetics*, 1: 165–172.

Fowler, D. M.; and Fields, S. 2014. Deep mutational scanning: a new style of protein science. *Nature Methods*, 11(8): 801–807.

Furió, V.; Moya, A.; and Sanjuán, R. 2005. The cost of replication fidelity in an RNA virus. *Proceedings of the National Academy of Sciences*, 102(29): 10233–10237. Publisher: Proceedings of the National Academy of Sciences.

Gambhir, N.; Harris, S. D.; and Everhart, S. E. 2022. Evolutionary Significance of Fungal Hypermutators: Lessons Learned from Clinical Strains and Implications for Fungal Plant Pathogens. *mSphere*, 7(3): e00087–22.

Gillespie, J. H. 1974. Natural Selection for within-Generation Variance in Offspring Number. *Genetics*, 76(3): 601–606.

Giraud, A.; Matic, I.; Tenaillon, O.; Clara, A.; Radman, M.; Fons, M.; and Taddei, F. 2001. Costs and Benefits of High Mutation Rates: Adaptive Evolution of Bacteria in the Mouse Gut. *Science*, 291(5513): 2606–2608.

Good, B. H.; and Desai, M. M. 2016. Evolution of Mutation Rates in Rapidly Adapting Asexual Populations. *Genetics*, 204(3): 1249–1266.

Good, B. H.; McDonald, M. J.; Barrick, J. E.; Lenski, R. E.; and Desai, M. M. 2017. The dynamics of molecular evolution over 60,000 generations. *Nature*, 551(7678): 45–50.

Gout, J.-F.; Li, W.; Fritsch, C.; Li, A.; Haroon, S.; Singh, L.; Hua, D.; Fazelinia, H.; Smith, Z.; Seeholzer, S.; Thomas, K.; Lynch, M.; and Vermulst, M. 2017. The landscape of transcription errors in eukaryotic cells. *Science Advances*, 3(10): e1701484.

Graves, C. J.; and Weinreich, D. M. 2017. Variability in Fitness Effects Can Preclude Selection of the Fittest. *Annual Review of Ecology, Evolution, and Systematics*, 48(1): 399–417.

Guo, S.; Zhang, C.; and Le, A. 2021. The limitless applications of single-cell metabolomics. *Current Opinion in Biotechnology*, 71: 115–122.

Gursky, V. V.; Surkova, S. Y.; and Samsonova, M. G. 2012. Mechanisms of developmental robustness. *Biosystems*, 109(3): 329–335.

Haddox, H. K.; Dingens, A. S.; Hilton, S. K.; Overbaugh, J.; and Bloom, J. D. 2018. Mapping mutational effects along the evolutionary landscape of HIV envelope. *eLife*, 7: e34420.

Hartl, D. L. 1975. Modifier theory and meiotic drive. *Theoretical Population Biology*, 7(2): 168–174.

Henry, L. P.; Bruijning, M.; Forsberg, S. K. G.; and Ayroles, J. F. 2021. The microbiome extends host evolutionary potential. *Nature Communications*, 12(1): 5141.

Hill, M. S.; Vande Zande, P.; and Wittkopp, P. J. 2021. Molecular and evolutionary processes generating variation in gene expression. *Nature Reviews Genetics*, 22(4): 203–215.

Jensen, J. L. W. V. 1906. Sur les fonctions convexes et les inégalités entre les valeurs moyennes. *Acta Mathematica*, 30(0): 175–193.

Johnson, M. S.; Gopalakrishnan, S.; Goyal, J.; Dillingham, M. E.; Bakerlee, C. W.; Humphrey, P. T.; Jagdish, T.; Jerison, E. R.; Kosheleva, K.; Lawrence, K. R.; Min, J.; Moulana, A.; Phillips, A. M.; Piper, J. C.; Purkanti, R.; Rego-Costa, A.; McDonald, M. J.; Nguyen Ba, A. N.; and Desai, M. M. 2021. Phenotypic and molecular evolution across 10,000 generations in laboratory budding yeast populations. *eLife*, 10: e63910.

Johnson, M. S.; Reddy, G.; and Desai, M. M. 2023. Epistasis and evolution: recent advances and an outlook for prediction. *BMC Biology*, 21(1): 120.

Karlin, S.; and McGregor, J. 1972. The evolutionary development of modifier genes. *Proceedings of the National Academy of Sciences of the United States of America*, 69(12): 3611–3614.

Karlin, S.; and McGregor, J. 1974. Towards a theory of the evolution of modifier genes. *Theoretical Population Biology*, 5(1): 59–103.

Kemble, H.; Nghe, P.; and Tenaillon, O. 2019. Recent insights into the genotype–phenotype relationship from massively parallel genetic assays. *Evolutionary Applications*, 12(9): 1721–1742.

Kim, A. K.; and Porter, L. L. 2021. Functional and Regulatory Roles of Fold-Switching Proteins. *Structure*, 29(1): 6–14.

Kimura, M. 1962. On the Probability of Fixation of Mutant Genes in a Population. *Genetics*, 47(6): 713–719.

Kimura, M. 1983. *The Neutral Theory of Molecular Evolution*. Cambridge University Press, 1 edition. ISBN 978-0-521-23109-1 978-0-521-31793-1 978-0-511-62348-6.

King, O. D.; and Masel, J. 2007. The evolution of bet-hedging adaptations to rare scenarios. *Theoretical Population Biology*, 72(4): 560–575.

Kirkpatrick, M.; and Barton, N. 1997. The strength of indirect selection on female mating preferences. *Proceedings of the National Academy of Sciences*, 94(4): 1282–1286.

Kirschner, M.; and Gerhart, J. 1998. Evolvability. *Proceedings of the National Academy of Sciences*, 95(15): 8420–8427.

Kolodny, O.; and Stern, C. 2017. Evolution of risk preference is determined by reproduction dynamics, life history, and population size. *Scientific Reports*, 7(1): 9364. Publisher: Nature Publishing Group.

Kong, A.; Frigge, M. L.; Masson, G.; Besenbacher, S.; Sulem, P.; Magnusson, G.; Gudjonsson, S. A.; Sigurdsson, A.; Jonasdottir, A.; Jonasdottir, A.; Wong, W. S. W.; Sigurdsson, G.; Walters, G. B.; Steinberg, S.; Helgason, H.; Thorleifsson, G.; Gudbjartsson, D. F.; Helgason, A.; Magnusson, O. T.; Thorsteinsdottir, U.; and Stefansson, K. 2012. Rate of de novo mutations and the importance of father's age to disease risk. *Nature*, 488(7412): 471–475.

Lamb, B. C.; Mandaokar, S.; Bahsoun, B.; Grishkan, I.; and Nevo, E. 2008. Differences in spontaneous mutation frequencies as a function of environmental stress in soil fungi at "Evolution Canyon," Israel. *Proceedings of the National Academy of Sciences*, 105(15): 5792–5796.

LeClerc, J. E.; Li, B.; Payne, W. L.; and Cebula, T. A. 1996. High Mutation Frequencies Among Escherichia coli and Salmonella Pathogens. *Science*, 274(5290): 1208–1211. Publisher: American Association for the Advancement of Science.

Lenski, R. E. 2017. Experimental evolution and the dynamics of adaptation and genome evolution in microbial populations. *The ISME Journal*, 11(10): 2181–2194.

Liberman, U.; and Feldman, M. W. 2006. Evolutionary theory for modifiers of epistasis using a general symmetric model. *Proceedings of the National Academy of Sciences*, 103(51): 19402–19406.

Lo, T. W.; Choi, H. J.; Huang, D.; and Wiggins, P. A. 2024. Noise robustness and metabolic load determine the principles of central dogma regulation. *Science Advances*, 10(34): eado3095.

Loeb, L. A. 2011. Human cancers express mutator phenotypes: origin, consequences and targeting. *Nature Reviews Cancer*, 11(6): 450–457. Publisher: Nature Publishing Group.

Lynch, M. 2007. The frailty of adaptive hypotheses for the origins of organismal complexity. *Proceedings of the National Academy of Sciences*, 104(suppl_1): 8597–8604.

Lynch, M. 2010. Evolution of the mutation rate. *Trends in Genetics*, 26(8): 345–352.

Lynch, M.; Ackerman, M. S.; Gout, J.-F.; Long, H.; Sung, W.; Thomas, W. K.; and Foster, P. L. 2016. Genetic drift, selection and the evolution of the mutation rate. *Nature Reviews Genetics*, 17(11): 704–714.

Maloney, R.; Ye, A.; Saint-Pre, S.-K.; Alisch, T.; Zimmerman, D.; Pittoors, N.; and De Bivort, B. L. 2024. Drift in Individual Behavioral Phenotype as a Strategy for Unpredictable Worlds.

Masel, J. 2005. Evolutionary Capacitance May Be Favored by Natural Selection. *Genetics*, 170(3): 1359–1371.

Masel, J.; and Bergman, A. 2003. The Evolution of the Evolvability Properties of the Yeast Prion [PSI $^{+}$]. *Evolution*, 57(7): 1498–1512.

Masel, J.; and Trotter, M. V. 2010. Robustness and Evolvability. *Trends in Genetics*, 26(9): 406–414.

Matic, I. 2019. Mutation Rate Heterogeneity Increases Odds of Survival in Unpredictable Environments. *Molecular Cell*, 75(3): 421–425.

Matic, I.; Radman, M.; Taddei, F.; Picard, B.; Doit, C.; Bingen, E.; Denamur, E.; and Elion, J. 1997. Highly Variable Mutation Rates in Commensal and Pathogenic *Escherichia coli*. *Science*, 277(5333): 1833–1834.

Maynard Smith, J. 1999. The Idea of Information in Biology. *The Quarterly Review of Biology*, 74(4): 395–400.

McDonald, M.; Hsieh, Y.-Y.; Yu, Y.-H.; Chang, S.-L.; and Leu, J.-Y. 2012. The Evolution of Low Mutation Rates in Experimental Mutator Populations of Saccharomyces cerevisiae. *Current Biology*, 22(13): 1235–1240.

Meers, M. P.; Llagas, G.; Janssens, D. H.; Codomo, C. A.; and Henikoff, S. 2023. Multifactorial profiling of epigenetic landscapes at single-cell resolution using MulTI-Tag. *Nature Biotechnology*, 41(5): 708–716.

Meiklejohn, C. D.; and Hartl, D. L. 2002. A single mode of canalization. *Trends in Ecology & Evolution*, 17(10): 468–473.

Melde, R. H.; Bao, K.; and Sharp, N. P. 2022. Recent insights into the evolution of mutation rates in yeast. *Current Opinion in Genetics & Development*, 76: 101953.

Mohler, K.; and Ibba, M. 2017. Translational fidelity and mistranslation in the cellular response to stress. *Nature Microbiology*, 2(9): 17117.

Mordret, E.; Dahan, O.; Asraf, O.; Rak, R.; Yehonadav, A.; Barnabas, G. D.; Cox, J.; Geiger, T.; Lindner, A. B.; and Pilpel, Y. 2019. Systematic Detection of Amino Acid Substitutions in Proteomes Reveals Mechanistic Basis of Ribosome Errors and Selection for Translation Fidelity. *Molecular Cell*, 75(3): 427–441.e5.

Morsky, B.; and Bauch, C. T. 2019. The impact of truncation selection and diffusion on cooperation in spatial games. *Journal of Theoretical Biology*, 466: 64–83.

Moxon, R.; Bayliss, C.; and Hood, D. 2006. Bacterial Contingency Loci: The Role of Simple Sequence DNA Repeats in Bacterial Adaptation. *Annual Review of Genetics*, 40(1): 307–333.

Murzin, A. G. 2008. Metamorphic Proteins. *Science*, 320(5884): 1725–1726.

Nei, M. 1967. Modification of Linkage Intensity by Natural Selection. *Genetics*, 57(3): 625–641.

Ochiai, H.; Hayashi, T.; Umeda, M.; Yoshimura, M.; Harada, A.; Shimizu, Y.; Nakano, K.; Saitoh, N.; Liu, Z.; Yamamoto, T.; Okamura, T.; Ohkawa, Y.; Kimura, H.; and Nikaido, I. 2020. Genome-wide kinetic properties of transcriptional bursting in mouse embryonic stem cells. *Science Advances*, 6(25): eaaz6699.

Otto, S. 2013. Evolution of Modifier Genes and Biological Systems. In Baum, D. A.; and Futuyma, D. J., eds., *Princeton Guide to Evolution*, 872. Princeton, NJ: Princeton University Press. ISBN 978-0-691-14977-6.

Otto, S. P.; and Gerstein, A. C. 2008. The evolution of haploidy and diploidy. *Current Biology*, 18(24): R1121–R1124.

Otto, S. P.; and Lenormand, T. 2002. Resolving the paradox of sex and recombination. *Nature Reviews Genetics*, 3(4): 252–261.

Paaby, A. B.; and Testa, N. D. 2018. Developmental Plasticity and Evolution. In Nuno De La Rosa, L.; and Müller, G., eds., *Evolutionary Developmental Biology*, 1–14. Cham: Springer International Publishing. ISBN 978-3-319-33038-9.

Partridge, L.; and Barton, N. H. 2000. Evolving evolvability. *Nature*, 407(6803): 457–458.

Parée, T.; Noble, L.; Roze, D.; and Teotónio, H. 2024. Selection can favor a recombination landscape that limits polygenic adaptation.

Pavlicev, M.; Cheverud, J. M.; and Wagner, G. P. 2011. Evolution of adaptive phenotypic variation patterns by direct selection for evolvability. *Proceedings of the Royal Society B: Biological Sciences*, 278(1713): 1903–1912.

Payne, J. L.; and Wagner, A. 2019. The causes of evolvability and their evolution. *Nature Reviews Genetics*, 20(1): 24–38.

Peck, K. M.; and Lauring, A. S. 2018. Complexities of Viral Mutation Rates. *Journal of Virology*, 92(14): e01031–17.

Pigliucci, M. 2008. Is evolvability evolvable? *Nature Reviews Genetics*, 9(1): 75–82.

Polizzi, B.; Calvez, V.; Charlat, S.; and Rajon, E. 2024. (Epi)mutation Rates and the Evolution of Composite Trait Architectures. *The American Naturalist*, 204(3): E42–E56.

Porter, L. L. 2023. Fluid protein fold space and its implications. *BioEssays*, 45(9): 2300057.

Porter, L. L.; and Looger, L. L. 2018. Extant fold-switching proteins are widespread. *Proceedings of the National Academy of Sciences*, 115(23): 5968–5973.

Pujadas, E.; and Feinberg, A. 2012. Regulated Noise in the Epigenetic Landscape of Development and Disease. *Cell*, 148(6): 1123–1131.

Raynes, Y.; Burch, C. L.; and Weinreich, D. M. 2021. When good mutations go bad: how population size can change the direction of natural selection.

Raynes, Y.; Gazzara, M. R.; and Sniegowski, P. D. 2011. Mutator dynamics in sexual and asexual experimental populations of yeast. *BMC Evolutionary Biology*, 11(1): 158.

Raynes, Y.; and Weinreich, D. M. 2021. Sign inversion in selection on ploidy.

Raynes, Y.; Wylie, C. S.; Sniegowski, P. D.; and Weinreich, D. M. 2018. Sign of selection on mutation rate modifiers depends on population size. *Proceedings of the National Academy of Sciences*, 115(13): 3422–3427. Publisher: Proceedings of the National Academy of Sciences.

Rebeiz, M.; Jikomes, N.; Kassner, V. A.; and Carroll, S. B. 2011. Evolutionary origin of a novel gene expression pattern through co-option of the latent activities of existing regulatory sequences. *Proceedings of the National Academy of Sciences*, 108(25): 10036–10043.

Richard, M.; and Yvert, G. 2014. How does evolution tune biological noise? *Frontiers in Genetics*, 5: 374.

Salathé, M.; Van Cleve, J.; and Feldman, M. W. 2009. Evolution of stochastic switching rates in asymmetric fitness landscapes. *Genetics*, 182(4): 1159–1164.

Sane, M.; Diwan, G. D.; Bhat, B. A.; Wahl, L. M.; and Agashe, D. 2023. Shifts in mutation spectra enhance access to beneficial mutations. *Proceedings of the National Academy of Sciences*, 120(22): e2207355120.

Serero, A.; Jubin, C.; Loeillet, S.; Legoix-Né, P.; and Nicolas, A. G. 2014. Mutational landscape of yeast mutator strains. *Proceedings of the National Academy of Sciences*, 111(5): 1897–1902.

Shaver, A. C.; Dombrowski, P. G.; Sweeney, J. Y.; Treis, T.; Zappala, R. M.; and Sniegowski, P. D. 2002. Fitness Evolution and the Rise of Mutator Alleles in Experimental *Escherichia coli* Populations. *Genetics*, 162(2): 557–566.

Silander, O. K.; Nikolic, N.; Zaslaver, A.; Bren, A.; Kikoin, I.; Alon, U.; and Ackermann, M. 2012. A Genome-Wide Analysis of Promoter-Mediated Phenotypic Noise in Escherichia coli. *PLoS Genetics*, 8(1): e1002443.

Simons, A. M. 2011. Modes of response to environmental change and the elusive empirical evidence for bet hedging. *Proceedings of the Royal Society B: Biological Sciences*, 278(1712): 1601–1609.

Sniegowski, P. D.; Gerrish, P. J.; Johnson, T.; and Shaver, A. 2000. The evolution of mutation rates: separating causes from consequences. *BioEssays*, 22(12): 1057–1066.

Sniegowski, P. D.; Gerrish, P. J.; and Lenski, R. E. 1997. Evolution of high mutation rates in experimental populations of E. coli. *Nature*, 387(6634): 703–705.

Sniegowski, P. D.; and Murphy, H. A. 2006. Evolvability. *Current Biology*, 16(19): R831–R834.

Sturtevant, A. H. 1937. Essays on Evolution. I. On the Effects of Selection on Mutation Rate. *The Quarterly Review of Biology*, 12(4): 464–467.

Suárez, P.; Valcárcel, J.; and Ortín, J. 1992. Heterogeneity of the mutation rates of influenza A viruses: isolation of mutator mutants. *Journal of Virology*, 66(4): 2491–2494.

Swings, T.; Van Den Bergh, B.; Wuyts, S.; Oeyen, E.; Voordeckers, K.; Verstrepen, K. J.; Fauvart, M.; Verstraeten, N.; and Michiels, J. 2017. Adaptive tuning of mutation rates allows fast response to lethal stress in Escherichia coli. *eLife*, 6: e22939.

Tawfik, D. S. 2010. Messy biology and the origins of evolutionary innovations. *Nature Chemical Biology*, 6(10): 692–696.

Tenaillon, O.; Barrick, J. E.; Ribeck, N.; Deatherage, D. E.; Blanchard, J. L.; Dasgupta, A.; Wu, G. C.; Wielgoss, S.; Cruveiller, S.; Médigue, C.; Schneider, D.; and Lenski, R. E.

2016. Tempo and mode of genome evolution in a 50,000-generation experiment. *Nature*, 536(7615): 165–170.

Tenaillon, O.; and Matic, I. 2020. The Impact of Neutral Mutations on Genome Evolvability. *Current Biology*, 30(10): R527–R534.

Tenaillon, O.; Toupance, B.; Le Nagard, H.; Taddei, F.; and Godelle, B. 1999. Mutators, Population Size, Adaptive Landscape and the Adaptation of Asexual Populations of Bacteria. *Genetics*, 152(2): 485–493.

Tokuriki, N.; and Tawfik, D. S. 2009. Chaperonin overexpression promotes genetic variation and enzyme evolution. *Nature*, 459(7247): 668–673.

Tröbner, W.; and Piechocki, R. 1984. Selection against hypermutability in Escherichia coli during long term evolution. *Molecular and General Genetics MGG*, 198(1): 177–178.

Tsimring, L. S. 2014. Noise in biology. *Reports on Progress in Physics. Physical Society (Great Britain)*, 77(2): 026601.

Viney, M.; and Reece, S. E. 2013. Adaptive noise. *Proceedings of the Royal Society B: Biological Sciences*, 280(1767): 20131104.

Wagner, A. 2005. Robustness, evolvability, and neutrality. *FEBS Letters*, 579(8): 1772–1778.

Wagner, A. 2022. Adaptive evolvability through direct selection instead of indirect, second-order selection. *Journal of Experimental Zoology Part B: Molecular and Developmental Evolution*, 338(7): 395–404.

Wagner, G. P.; and Altenberg, L. 1996. Perspective: Complex Adaptations and the Evolution of Evolvability. *Evolution*, 50(3): 967.

Wang, Z.; and Zhang, J. 2011. Impact of gene expression noise on organismal fitness and the efficacy of natural selection. *Proceedings of the National Academy of Sciences*, 108(16).

Watson, R. A. 2020. Evolvability. In Nuno De La Rosa, L.; and Müller, G., eds., *Evolutionary Developmental Biology*, 1–16. Cham: Springer International Publishing. ISBN 978-3-319-33038-9.

Watson, R. A.; and Szathmáry, E. 2016. How Can Evolution Learn? *Trends in Ecology & Evolution*, 31(2): 147–157.

Weber, M. 1996. Evolutionary plasticity in prokaryotes: A panglossian view: Evolutionary plasticity in prokaryotes. *Biology & Philosophy*, 11(1): 67–88.

Webster, A. K.; Willis, J. H.; Johnson, E.; Sarkies, P.; and Phillips, P. C. 2023. Epigenetic context predicts gene expression variation and reproductive traits across genetically identical individuals.

Wedd, C.; Yunusov, T.; Smith, A.; Li, R.; Hardo, G.; Hunter, M.; Majed, R.; Fusco, D.; and Bakshi, S. 2024. Single-cell imaging of the lytic phage life cycle in bacteria.

Weinreich, D. M. 2023. *The foundations of population genetics*. Cambridge, Massachusetts: The MIT Press. ISBN 978-0-262-37256-5 978-0-262-37257-2.

Weissman, M.; Raynes, Y.; and Weinreich, D. 2023. Beyond the (geometric) mean: stochastic models undermine deterministic predictions of bet hedger evolution.

Whitlock, A. O. B.; Peck, K. M.; Azevedo, R. B. R.; and Burch, C. L. 2016. An Evolving Genetic Architecture Interacts with Hill–Robertson Interference to Determine the Benefit of Sex. *Genetics*, 203(2): 923–936.

Wielgoss, S.; Barrick, J. E.; Tenaillon, O.; Wiser, M. J.; Dittmar, W. J.; Cruveiller, S.; Chane-Woon-Ming, B.; Médigue, C.; Lenski, R. E.; and Schneider, D. 2013. Mutation rate dynamics in a bacterial population reflect tension between adaptation and genetic load. *Proceedings of the National Academy of Sciences*, 110(1): 222–227.

Wilke, C. O.; Wang, J. L.; Ofria, C.; Lenski, R. E.; and Adami, C. 2001. Evolution of digital organisms at high mutation rates leads to survival of the flattest. *Nature*, 412(6844): 331–333.

Winger, B. M.; Auteri, G. G.; Pegan, T. M.; and Weeks, B. C. 2019. A long winter for the Red Queen: rethinking the evolution of seasonal migration. *Biological Reviews*, 94(3): 737–752.

Wiser, M. J.; Ribeck, N.; and Lenski, R. E. 2013. Long-Term Dynamics of Adaptation in Asexual Populations. *Science*, 342(6164): 1364–1367.

Wolf, S.; Melo, D.; Garske, K. M.; Pallares, L. F.; Lea, A. J.; and Ayroles, J. F. 2023. Characterizing the landscape of gene expression variance in humans. *PLOS Genetics*, 19(7): e1010833.

Woods, R. J.; Barrick, J. E.; Cooper, T. F.; Shrestha, U.; Kauth, M. R.; and Lenski, R. E. 2011. Second-Order Selection for Evolvability in a Large *Escherichia coli* Population. *Science*, 331(6023): 1433–1436.

Wright, S. 1931. Evolution in Mendelian Populations. *Genetics*, 16(2): 97–159.

Wünsche, A.; Dinh, D. M.; Satterwhite, R. S.; Arenas, C. D.; Stoebel, D. M.; and Cooper, T. F. 2017. Diminishing-returns epistasis decreases adaptability along an evolutionary trajectory. *Nature Ecology & Evolution*, 1(4): 0061.

Zhou, X.; and Xue, B. 2022. Effect of compositional fluctuation on the survival of bet-hedging species. *Journal of Theoretical Biology*, 553: 111270.