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# Abstract

# Introduction

Quantitative genetics aims to quantify genetic diversity; this diversity has broad implications for adaptation, it is well described how diversity enhances efficiency of adaptation; adaptation with more Va = faster, more efficient; particularly seen in the case of quantitative genetics, where stabilizing selection is often assumed; different story with the maintenance of variation around a fitness optimum, i.e. after the adaptive walk what happens?; several models have appeared over the last 50 years to explain the maintenance of variation; continuum of alleles vs diallelic; within continuum of alleles, the approximate distribution of allelic effects depends on the relative mutation rate to selection strength; in other words, the strength of new mutations to standing genetic variation; models over the last 50 years have failed to explain natural diversity observed in populations;

The ubiquity of adaptation in evolutionary studies is telling of the impact of Darwin’s seminal work. Over 4600 papers featuring the keyword ‘adaptation’ were published in Nature research journals in 2019. The allure of adaptation comes from the power of Darwin’s theory to explain natural diversity both within and between populations (Brady *et al.* 2019). Yet explanations of diversity via Darwin’s theory have been misinterpreted before – prior to Williams publishing his thesis (1966), the theory of ‘group adaptation’, whereby adaptation is driven by altruistic mutations that benefit populations at the cost of the individual gene, was commonplace and well-regarded. The focus on adaptive traits, and the ability of populations to adapt to new situations is wonderfully intuitive, however, populations are never perfectly adapted: trait values are rarely optimal, populations decline, and extinctions are commonplace (Brady *et al.* 2019). The extent of maladaptation, where populations are stable at some distance from a fitness optimum, seems wide, however the extent of maladaptation is rarely discussed (Nesse 2005). The keyword ‘maladaptation’ was mentioned in just 45 papers in Nature research journals published in 2019, yet it is expected to .

**Evidence for stab sel: in gene regulation, Hodgins-Davis et al – this paper, also m/s/d balance for quant gen models;**

The extent of maladaptation in natural populations has been debated for some time – are populations likely to be restricted in their fit to a phenotypic optimum? Is such an optimum attainable for populations, or is there more likely to be local, stable optima that are difficult to escape?

Much of the variation between and within populations is the result of continuous variability in traits individuals in those populations possess. Differences in such quantitative traits lead to adaptation and speciation. Underpinning adaptation by quantitative traits is additive genetic variance (VA), the heritable component of variation; the amount of which has mystified quantitative geneticists for close to 100 years. Predicting levels VA is reliant on mutation rate and selection strength: both of which are notoriously difficult to estimate in natural populations. Many quantitative traits are subject to stabilizing selection, where intermediate trait values have maximum fitness. Theory suggests that adaptation via stabilizing selection should be more efficient with higher standing VA, and that the selective fixation of this standing variation should decrease VA as adaptation takes place (Fisher 1930; Lande 1975). However, these expectations have not always coincided with observed data.

Depletion of VA with stabilizing selection has been shown both experimentally , and analytically, however increasing amounts of more modern work show no effect of selection strength on VA. Sztepancz and Blows (2017) showed that there was no relationship between genetic variation and the strength of stabilizing selection in *Drosophila serrata­­­.* More modern analysis of Fisher’s (1930) geometric model (upon which stabilizing selection is built) has shown that when the individual effect of selection in alleles is weak, stabilizing selection has a minimal effect on VA, as drift at any individual locus may compete with selection to adjust allele frequencies (Barton 2017). Discrepancies such as these can perhaps be explained by the relative effect of stabilizing selection, depending on where a population is relative to the optimum. When populations are far from the optimum, mutations act under a directional selection model, where larger mutations that bring an individual closer to the optimum are more beneficial (Zhang 2012). However, as populations approach that optimum, large effect mutations become costly as they are more likely to drag populations further away from the optimum.

Barton 2017: selection negligible when individual allles is weak and comparable to drift

Thornton 2019: when phenotypes approach optimum, strength of selection on indivual muts decreases effect on Va under infinitesimal model, selection gets more info when selection on

Zhang 2012: when phenotypes near optimum, selection is stab, while far away, closer to dir sel

Sztep: dir sel more common in nature? Populations more commonly maladapted?

At the heart of the evolutionary sciences is the need to understand the natural world’s diversity. Darwin’s (1863; SOURCE) introduction of natural selection some 140 years ago led to increasingly accurate glimpses into the units of evolution, genes, and their movement through a population in response to selection (SOURCE). However, these movements, particularly in a multivariate trait space, become a challenging realm to predict (SOURCE; Lande 1979, 80 etc.). To navigate this space, it is necessary to reduce the predictors of trait trajectories to their principles: how they affect additive genetic variance, the heritable component of trait variability.

Additive genetic variability, VA, is regarded as the most important predictor of a population’s adaptability (Lynch and Lande 1998; Aguirre *et al.* 2014; Careau *et al.* 2015), and hence it’s trajectory through time towards a phenotypic optimum. Although a multitude of stochastic and deterministic processes also contribute to the population’s total trait variability, VA is heritable, and therefore predictive of a population’s trajectory over micro-evolutionary time. The VA of a population determines the phenotypic space that population can explore. Hence, it is predicted that populations with large amounts of VA are best suited to adapting to novel environments (Barton and Charlesworth 1998). Such an example is X. However this is not always the case, standing genetic variation is characterized by a variety of architectural and population-level constraints such as rates of pleiotropy, selection strength, additive effect size, linkage, and deleterious mutation/background selection (SOURCE). For example, under infinitesimal models, selection has a trivial impact on standing variation (Barton 2017).

including genetic drift, selective pressures, additive effect sizes, between- and within-gene interactions, and heritability (SOURCES).

* Natural diversity, population movements in trait space
* Heritable variation
* Stabilising selection, effect on variation/need for variation vs drift
  + Expected to remove variation, mutation alone can’t explain why in natural populations we see so much variation: why?
* Additive effect sizes, effects on variation
* Background selection, effect on variation
* Population genetics expectations of variation under bkg sel, additive effects

# Methods

Using the forward-genetics modelling package SLiM 3.4 (Haller and Messer 2019), we constructed two models to explore a portion of the multivariate parameter space that explains genetic variability in natural populations. These parameters included genome wide recombination rate, the additive effect size distribution, the rate of universal pleiotropy, mutational correlation between traits, and the selection strength multiplier, (Table 1). The relative rate of non-QTL, deleterious mutation compared to trait mutations was also varied across models. This parameter led to two alternate outcomes that could influence variation and adaptation: either the reduction in QTL mutation rate due to increasing deleterious mutation rate could cause observed differences, or the effect of the deleterious mutations on fitness could be attributed to the differences. Preliminary analyses indicated that the ratio of QTL mutations to deleterious mutations remained constant across increasing levels of this parameter (Figure S3). This suggests that a similar deleterious load was experienced across populations, and that the effects of increasing this rate were attributable to changes in QTL mutation rate rather than the deleterious effects of non-QTL mutations. The highest QTL mutation rates were experienced by models with low rates of deleterious mutation, and vice versa. Thus, models with high mutation rate and low selection strength (deleterious mutation rate < 0.33; > 660) approximated the Kimura-Fleming-Lande Gaussian approximation of allelic effects (Kimura 1965; Lande 1975; Fleming 1979), while models with low mutation rates and high selection strength approximated Turelli’s (1984) House-of-Cards model. Among all parameter combinations, multiple conditions and assumptions were shared.

## Common model elements

Both of my experimental models consisted of a SLiM 3.4 model simulating a Wright-Fisher population of 8000 diploid individuals evolving over 100,000 generations. Populations were assumed to be completely allopatric. Populations first were subject to 50,000 generations of burn-in to build standing variation to mutation-drift balance (figure S1). Individuals were characterized by 8 traits, controlled by 100 loci each. Each trait had an identical effect on fitness, forming a ‘mega-trait’ with varying variance-covariance structures depending on pleiotropy rates. Each locus was assumed to have identical length, and each base pair within it mutationally independent – hence mutations occurred at an arbitrary position within the locus. This assumption is supported by a study by Thornton (2019), which found that within-locus differences in linkage had no average effect on either genetic variance or the mean trait value, indicating within-locus independence. In addition, the average number of base pairs per locus is highly conserved within eukaryotes (Xu *et al.* 2006), lending credence to the assumption of equal gene length. Mutations were assumed to be completely additive in effect, with no dominance or epistatic interactions, aside from additive epistasis occurring as a result of the fitness function. Mutational effects were in phenotypic units, an arbitrary unit denoting relative differences in phenotype. All loci were assumed to be on the same chromosome, with genetic distance being determined by the recombination rate parameter, r (Table 1). Both models had a genome-wide germline mutation rate of 8.045x10-6 per locus per generation, based on an average of five groups of eukaryotes (Aston *et al.* 2017).

The effective population size, Ne = 8000, was chosen to compromise between computational performance and the effect of genetic drift on populations under stabilizing selection. This value results in weak genetic drift in comparison with the strength of selection (under strong selection pressures), and appropriate standing genetic variation following burn-in to allow for adaptation (Lynch and Lande 1998).

Mutational effects on trait values were sampled from a normal distribution,

where λ is the additive effect size (Table 1). In the case of pleiotropy, a multivariate normal distribution was used, where n = 8, and

where **Σ** is a covariance matrix with diagonal values equal to λ and non-diagonals pulled from a normal distribution:

where is the parameter value of mutation correlation. **Σ** was ensured to be positive definite by multiplication with its transpose,

Non-trait deleterious mutations had fitness effects sampled from a gamma distribution:

Where and (SLiM Manual). This describes a distribution of weak deleterious mutations on average.

All models were subject to 50,000 generations of burn-in, where mutations accumulate until the population reaches mutation-drift equilibrium. This is tracked as heterozygosity through the simulation, where mutation-drift equilibrium occurs when:

where µ represents the per-locus mutation rate per generation (Kimura and Crow 1964). A population at equilibrium was assumed sufficiently burnt in. Trials indicated that 50,000 generations of burn-in was sufficient for our population size (Figure S1). Deleterious mutation/mutation rate lowered the value of away from expectation in initial burn-in tests, however an alternative equilibrium was reached, satisfying the requirements of burn-in regardless of the parameter (Figure S1). During the simulation run, trait variances, covariances, and trait means were collected every 500 generations to track distances from the optimum and trait variability over time. At the end of the simulation, the allelic effects of segregating mutations in all populations were collected.

## Model specific characteristics

After reaching equilibrium, populations evolved for 100,000 generations of neutral drift or stabilizing selection, depending on the treatment. Neutral drift entailed no change from the properties of the burn-in, whereas stabilizing selection imposed a fitness function on phenotypes, invoking a multivariate optimum a fixed distance from the population mean phenotype post-burn-in. The position of the optimum is defined as:

Where is the vector of phenotype means, is the per-locus, per-generation mutation rate; , is the number of mutational steps to reach the optimum, and is the number of generations of burn-in. For our purposes, 8.045x10-6, 100, and . This distance was close to the original phenotypes, meaning most of the simulation (approximately 98000 generations of the simulation) investigated the maintenance of variation at a fitness optimum.

The fitness of an individual in the population was defined as:

Where s is the selection coefficient, represents the gradient of the selection curve, n is the number of traits, and xn is the phenotype for trait n. To ensure a theoretical minimum and maximum fitness, s was fixed at 0.9, ensuring minimum fitness was , and maximum fitness was 1. This results in individuals at the optimum being at most ten times as fit as those infinitely far from the optimum. The model-specific maximum fitness difference depends on, which adjusts the realized fitness gradient via the curvature of the fitness function.

## Model Parameterization

Five parameters were shared between models, with a sixth for testing selection (Table 1). These were sampled using a Latin hypercube sampling design, with 1024 parameter combinations testing the null model, and 256 for the selection model (Figure S2). The hypercube sampling was necessary to explore the entire parameter space, as simple factorial designs would have been impractical to achieve. Each hypercube sample represents a combination of parameters, with the total set of samples designed to maximize the distance between samples (sampling more of the total space), and minimize correlations between them (Helton and Davis 2003). Hypercube samples were generated using the R packages ‘DoE.Wrapper’ and ‘LHS’, using the maximin algorithm (Melo *et al.* 2015; R Developmental Core Team 2019). Each sample/model was repeated 100 times, using 100 seed values fed to SLiM. These seeds were randomly sampled from a uniform distribution of the total range of unsigned 32 bit integers (1 to 232 – 1) (R Developmental Core Team 2019). The array of parameter combinations and replicates was processed across 1152 cores on the University of Queensland’s Tinaroo high performance computing (HPC) system, using embedded Nimrod scripts to feed parameter/seed combinations to individual SLiM processes.

## Analysis

Despite not all data conforming to normality, no data was transformed owing to the large sample sizes. Previous work into the robustness of t-tests, and F-tests have shown that departures from normality can usually still provide reliable estimates, provided the number of observations is large enough that coefficient estimates are approximately normally distributed due to the central limit theorem (Lumley *et al.* 2002). This was verified with diagnostic tools in the R package “jtools” (Long 2020). In terms of regression analysis, heteroscedasticity can still remain a problem, even with large sample sizes. To account for this, we used Eicker-Huber-White (EHW) HC2 or HC3 robust standard errors in our linear regression models via the ‘estimatr’ package in R (Eicker 1967; Huber 1967; White 1980; Hayes and Cai 2007; Blair 2020) . Due to the large sample sizes (128000 total models), we were able to find significant differences between groups with extremely small effect sizes. To ensure we focused only on biologically meaningful differences, we calculated the relative contributions of factors to the appropriate regression, using the Lindeman, Merenda and Gold method (Lindeman 1980), explaining only the factors that contributed meaningfully to variation.

For analysis, the interaction between and mutation rate was treated as a ‘model’ parameter, indicating whether the hypercube sample approximated House-of-Cards allelic effects, or Gaussian effects. An additional model type, ‘Null’, summarized the models with no selection and any mutation rate. Remaining models with intermediate selection strengths and deleterious mutation rates were not considered for analysis, although that remains an exciting prospect for the future. Additive effect size, recombination rate, pleiotropy rate, and mutational correlation hypercube values were binned into low, medium, and high categories for simpler analysis.

We compared responses at the final generation of the simulation (100,000) across all analyses. Trait variances and covariances were pooled and averaged to form a ‘mega-trait’ average variance and covariance, since traits were functionally identical. In addition, we computed the population mean Euclidean distance from the optimum for each replicate and model:

Where pi and qi are the population mean and optimum value, respectively, for trait *i*.

To determine the effects of CoA model on adaptation, we explored the distribution of final distances from the optimum, finding a distinct ‘dead zone’ where distances were not represented. We used this dead zone to classify models into two categories: adapted, or maladapted. Adapted models had distances from the optimum less than 16 units, and maladapted with distances greater than 16 units. We used a Chi-square test followed by an odds-ratio post-hoc to determine the differences in representation among CoA models in adapted and maladapted categories. Following this, we discarded maladapted populations, choosing to focus on investigating the genetic architectures underlying adapted populations.

To evaluate the effects of genetic architecture on adaptation under the CoA models, we used EHW-error multiple regression models to determine the effects of CoA model type, additive effect size, recombination rate, pleiotropy rate, and mutational correlations between traits on distance from the optimum, additive variance, and trait covariances.

We also collected the mutational effects of segregating alleles at the end of the simulation. With this, we compared mean distributions of allelic effect sizes in adapted populations according to additive effect size with multivariate multiple regression. Responses included mean allelic effect, variance, and kurtosis of the distribution, as well as the mutation counts contributing to VA within each model. We adjusted for heteroskedasticity with EHW robust standard errors. Multiple regressions were calculated across 50 replicates owing to RAM limitations.

# Results

## Tracking population dynamics over time

To determine whether populations were under mutation-selection-drift balance by the end of the simulation, we plotted additive variance and covariance over time across selection strengths. We reasoned that the joint effects of mutation, which creates variance, and drift and selection, which remove variance, would lead to stable levels of genetic variability over long periods of time. We found that after 100,000 generations (2 days of run-time per model), variance increased asymptotically in all models (Figure 3) suggesting that levels of genetic variability were unlikely to change significantly in longer model runs. Mean additive variance was consistently higher under a Gaussian model, whereas it remained low and almost constant in the House-of-Cards models (Figure 3A). Both selection models clearly behaved differently from a null model where genetic drift was expected to dominate. Covariance between traits acted similarly across models (Figure 3B). Knowing that by generation 100,000 we are at mutation-selection-drift equilibrium, we can now investigate whether populations are well-adapted under different selection and genetic models.

## General patterns of adaptation with Continuum of Alleles models

We explored the distribution of Euclidean distances around a phenotypic optimum under House-of-Cards and Gaussian models of allelic effects and compared them to a null model without selection (Fig. 4A). Both Gaussian and House-of-Cards models showed a small proportion of populations that came within 16 phenotypic units from the optimum, with a visible division between adapted and maladapted populations (Fig. 4). The ‘dead space’ that separated these populations did not exist in the null model. To further explore this bimodality, we examined the differences between models in their ability to reach the adapted space. Populations were more likely to be found in the adapted zone if they belonged to either selection model over the null model (χ2 = 9602.1, df = 2, p < 0.0001). 15.23% of Gaussian populations reached the adapted space, while House-of-Cards populations reached this 16.1% of the time. By contrast, 0.53% of null populations reached the adapted space. A post-hoc odds ratio test found significant differences between null and Gaussian (OR = 33.566, 95% C.I. = 29.5, 38.2, p < 0.0001) and null and House-of-Cards (OR = 35.872, 95% C.I. = 31.5, 40.85, p < 0.0001), but not between Gaussian and House-of-Cards (OR = 1.069, 95% C.I. = 0.93, 1.23, p = 0.35). To understand the underlying genetic architectures of populations that were able to come close to the phenotypic optimum, we compared the effects of genetic architecture on distance to the optimum (Fig. 5; Table 2), mean trait variance (Fig. 6), and mean trait covariance (Fig. 7) across the two selection models.

## Genetic architecture effects on adaptation with Continuum of Alleles models

We compared the effects of varying additive effect sizes, recombination rates, pleiotropy rates, and mutational correlations on Euclidean distances of populations close to the phenotypic optimum under Gaussian or House-of-Cards mutational models. Table 2 shows the mean effects of these variables on how close populations get to the phenotypic optimum, as well as the effects on trait variance and covariance. Although all genetic architecture parameters had significant effects on distance, variance and/or covariance, most of these effects were small in magnitude. For brevity, we discuss only the parameters that explain the most variation in distance, variance, and covariance. Variation in distance was explained mostly by pleiotropy (explaining 8.6% of total variation among models), model type (explaining 5.4% of variation, and additive effect size (explaining 2.8% of variation). Mean distance from the optimum was lowest when additive effect sizes were low (0.841 ± 0.181; Fig. 4); this did not change between CoA models (t921 = -0.422, p = 0.998). However, House-of-Cards models were more robust to changes in additive effect size than Gaussian models (t921 = -2.583, p = 0.01). When increasing effect size from low to high under a Gaussian mutation model, adapted populations’ mean distance from the optimum increased by 2.203 ± 0.232 phenotypic units (t921 = -9.504, p < 0.0001). The same change in effect size under a House-of-Cards model resulted in no significant change to mean distance (t921 = -0.587, p = 0.827). Figure 5 shows how patterns of adaptation varied between Continuum of Alleles models when increasing the variance of allelic effect sizes. Pleiotropy rate increased distance, however there was no interaction between pleiotropy and model type (t921 = 0.843, p = 0.399; Fig. 4C). Increasing pleiotropy rate from low to high led to an average 1.261 ± 0.178 unit decrease in distance from the optimum (t921 = 7.099, p < 0.0001). These effects on distance were not necessarily mirrored with the effects of genetic architecture on trait variance, which was explained by additive effect size (45.1% of variation in trait variance), and its interaction with model type (explaining 14.4% of this variation).

On average, House-of-Cards models near the optimum had considerably more additive variance than Gaussian models (40.4 ± 18.72 units vs 2.6 ± 0.07 units; t921 = -2.019, p = 0.044). Under a Gaussian model, increasing the additive effect size of populations in the adapted zone marginally increased trait variance (t921 = -14.386, p < 0.0001; Fig. 5A), however this was not the case under a House-of-Cards model (t921 = -1.958, p = 0.123). Figure 6 shows how additive effect size interacts with Gaussian and House-of-Cards models to drive differences in variance in adapted populations. Note that several outliers were removed from Figures 6 and 7 owing to their distortion of the figures. These outliers had variance greater than 50 and covariance less than -5 (Fig. S3, S4). Similarly to variance, differences in covariance could be explained mainly be differences in additive effect size (explaining 46.4% of variation), and the interaction between effect size and the Continuum of Alleles model type (explaining 15.6% of variation).

Average trait covariance differed between models (t921 = 2.147, p = 0.032; Fig. 6), with Gaussian models carrying very little genetic covariance amongst traits (0.014 ± 0.005), and House-of-Cards models carrying slightly more (-3.616 ± 1.691). Increasing additive effect size from low to high in Gaussian models led to slight declines in covariance (a decrease of 0.039 ± 0.005; t921 = 7.526, p < 0.0001; Fig. 6A). No significant effect of increasing additive effect size on covariance was seen in House-of-Cards models (t921 = 1.937, p = 0.129). The difference in response to additive effect size between models was marginally insignificant (t921 = -1.929, p = 0.054). Figure 7 shows the effects of increasing additive effect variance and Continuum of Alleles model type on covariance.

These analyses therefore suggest that additive variance and covariance are rather robust under House-of-Cards models, and less so under Gaussian models. Additive effect size in particular seems important to understanding the interplay between adaptation and additive variance. To analyze the underlying cause of these variances, covariances, and by extension, distance to the phenotypic optimum, we need to study the underlying allelic effect size distributions of the models. We compared the means, variances, kurtosis, and count of mutations contributing to these distributions across models to understand the mutational limitations imposed by genetic architectures under the two CoA models.

## Allelic effect size distributions with Continuum of Alleles models

The distributions of allelic effects are dependent on several parameters: the mean of the effects, which may be biased in some direction by genetic architectures, the variance of the distribution, indicating the variability in size-effects that are sampled, and the kurtosis of the distribution, indicating the rarity of large-effect alleles. To assess the mutational bias of models, we first compared the means of distributions across models and genetic architectures. The resulting regression was insignificant (F17, 411 = 1.127, p = 0.325, Adjusted R2 = 0.189), indicating a lack of directional mutational bias. We then turned our attention to the variance of distributions to understand the constraints that genetic architectures may apply to mutational models (F17, 411 = 55.04, p < 0.0001, Adjusted R2 = 0.851). Additive effect size explained 66.2% of total variability between models. Under a Gaussian model, increasing additive effect size from low to medium significantly increased allelic effect variance by 6.02 ± 0.372 phenotypic units (t411 = -16.188, p < 0.0001; Fig. 8), however no significant difference occurred for increasing variance from low to high or from medium to high. No significant changes to variance with increasing effect size were seen under House-of-Cards models. Figure 8 shows the distributions of allelic effects with changing additive effect size variability under Gaussian and House-of-Cards models. Leading on from the variance of allelic effects is the kurtosis, which describes the rarity of large-effect alleles.

Kurtosis differed significantly across models and genetic architectures with additive effect size variance explaining 31.9% of variability between models, and the interaction between additive effect size and model type contributing another 15.5%(F17, 411 = 12.36, p < 0.0001, Adjusted R2 = 0.6). Under the Gaussian model, increasing additive effect size from low to medium increased kurtosis by 0.985 ± 0.159 (t411 = -6.206, p < 0.0001). No analogous effect was seen under the House-of-Cards model (t411 = -0.944, p = 0.6130). As well as the distributions of allelic effects, the absolute counts of mutations contributing to each distribution gives an indicator of the genetic diversity of populations.

To assess the effects of genetic architecture and models on promoting many or few alleles, we compared mutation counts between models, finding significant differences among models (F17, 411 = 580.2, p < 0.0001, Adjusted R2 = 0.94). Model type contributed the most to explaining mutation count variation among models, describing 58.4% of among-model variation. Pleiotropy rate explained 10% of variation, however this is explicated by each pleiotropic mutation contributing multiple effects with a single mutation. The mean number of mutations in Gaussian models was considerably higher than that of House-of-Cards models, but over a large range of values: 1516 ± 6608 mutations for Gaussian models versus 374 ± 114 for House-of-Cards (t411 = 0.173, p = 0.863).

# Discussion

Weird outliers in variance and covariance: could be that these populations did have high variance and covariance due to wildly different individual phenotypes, but when you took the mean distance of the population, that mean was somewhere in the middle of all that variation, which happened to be close to the optimum.

Truly adapted ones – at distance = 0

Really rare to be close under null

At the optimum (Po = 1), the Gaus and HoC are not like Null

This is the power of pop gen models – get to see these allele frequencies, the whole spectrum

Surprising result: deleterious mutation reduces distance to optimum under maintenance, particularly under large size effects; still anchored in quant gen theory, even though pop gen predicts the opposite; pop gen – Ne reduced with BS, decreased variation expected, worse selection, more drift etc.

We found that increasing rates of deleterious mutation resulted in populations being more able to maintain their position around the optimum, overcoming some of the difficulties of fending with large-effect alleles that may pull populations away from the optimum (Figures 3, 5). Although it may at first seem counter-intuitive that stronger background selection increases the ability of populations to maintain their position at an optimum (Figure 5, 6), the effect can be explained with existing quantitative genetics expectations. To understand this, we must first explore the population genetics expectations of the effects of background selection on adaptation, and distinguish the difference in expectations between a population approaching the optimum and maintaining its position once it has arrived there.

It is well understood in population genetics that background selection reduces effective population size, reducing the effectiveness of selection and increasing the strength of genetic drift (Charlesworth *et al.* 1997; Houle 1998). As deleterious mutations are removed from the population, close-by linked QTLs are also removed (Charlesworth and Charlesworth 2010). The effect of this is decreased genetic diversity. In population genetics studies this is usually expressed in terms of FST or , whereas in quantitative genetics the analog is additive genetic variance (Falconer 1996; Charlesworth *et al.* 1997). Reductions in VA with increasing background selection were observed in this study, supporting this expectation (Figure 3A, 4). The expected effect of this on adaptation is quite clear when considering the initial approach towards the optimum: in quantitative genetics models, genetic variability is expected to increase the trait space that populations are able to explore, improving their ability to travel towards an optimum (Fisher 1930; Charlesworth and Charlesworth 2010; Aguirre *et al.* 2014). Indeed, these theoretical expectations have been found in natural populations: for example, Pujol and Pannell (2008) showed that populations of annual mercury, *Mercualis annua,* were able to respond to selection for pollen production when standing genetic variation was higher. Similarly, studies into the adaptation of red flour beetle (*Tribolium castaneum*) populations to new niches found high standing variation decreased the likelihood of extinction, and increased rates of niche expansion (Agashe and Bolnick 2010; Agashe *et al.* 2011). However, these expectations do not describe what we found in the current study: the most well-adapted populations consistently have higher rates of deleterious mutation, and hence lower standing genetic variance. The key to this lies in the expectations of the *maintenance* of variation and fitness around an optimum rather than the *approach* towards said optimum. The expectations surrounding this temporal space is considerably less extensive than that of the adaptive walk.

While reduced standing variation is expected to increase the time a population takes to reach an optimum (or perhaps prevent populations from reaching it at all), once a population has reached its optimum or stabilizes around its ‘local optimum’, the closest position it can maintain given the selected traits’ genetic architectures, mutation rates, and the population size – where does the population go?

In theoretical quantitative genetics, much debate is had over which particular models best describe the maintenance of variation in the presence of stabilizing selection over time. Selection is able to retain variation, particularly in large populations where drift is weak, and scenarios where balancing selection creates a non-linear fitness landscape, however the extent of this differs depending on many factors, including selection strength, genetic architectures, epistatic and dominant interactions, and the strength of selection relative to mutation (Walsh and Lynch 2018). Understanding the relative strength of selection to mutation has led to two distinct approximations of expected distributions of allelic effects. When mutation is much stronger than selection, Kimura (**1965a**) and Fleming’s (**1979**) Gaussian approximation holds true, whereas when the opposite occurs, Turelli’s (**1984**) house of cards approximation is more accurate. This distinction between models is arbitrarily granular, mostly for analytical viability. Computational methods allow for a continuous exploration of this space of models.

Figure 3: decrease in var with deleterious mutation is analogous to effects of lower Ne, but on a per locus level rather than genome wide. Hence, gives a proxy of the assumptions of CoA models with N -> Inf

Loss of fitness due to variation around optimum: expected to be 1/4Ne without any background selection (will vary with Ne due to effect on local Ne ) – Lande 1976

# Snippets

Underpinning this model is the continuum of alleles model of allelic effects, suggesting large numbers of alleles at many loci forming a continuous distribution of effect sizes, usually Normal in shape (Lande).

Pleiotropy fundamentally alters the signatures of HCA vs Gaussian approximation in COA so they approach each other - other parameters may as well?

Most effort in understanding stabilizing selection has focused on assuming either a Gaussian (as in this paper) or quadratic fitness function

Pleiotropy also had strong effects, due to contributing more than one trait value per mutation. Increasing pleiotropy rate by 10% increased RAF by 59.366 ± 2.531 alleles under no selection (t63937 = 23.458, p < 0.0001). Increasing deleterious mutation with pleiotropy rate significantly reduced this effect, with a 10% increase in pleiotropy rate and deleterious mutation rate simultaneously leading to a total loss of 22.555 ± 5.994 alleles (t63937 = -17.795, p < 0.0001). Under stabilising selection, a simultaneous 10% increase in pleiotropy rate and deleterious mutation rate led to an increase of 23.553 ± 8.567 alleles (t63937 = 19.193, p < 0.0001).

# Tables

**Table 1:** Model parameters for both null and stabilizing selection models. The range of values is based on literature, but values are adjusted to be practical for the time of the experiment.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Parameter** | **Symbol** | **Range** | **Description** | **Source(s)** |
| Genome wide recombination rate | r | 0 to 1.241x10-4 per locus | The singular recombination rate used across the entire simulated genome. | Stapley et al. 2017 |
| Background selection rate | δ | 0 to 1 | The number of non-trait deleterious mutations that occur relative to trait mutations. |  |
| Rate of universal pleiotropy | ϖ | 0 to 0.5 | The proportion of trait mutations that affect all traits rather than a single trait. While 100 loci control a trait independently by default, this may be changed by this parameter. However, ratios of loci affecting each trait will remain constant, especially across multiple replicates. | Chesmore et al. 2017; |
| Mutational pleiotropic correlation | m | 0 to 0.5 | The mutational correlation between additive effects of pleiotropic mutations determines the similarity of trait effects between traits for the same pleiotropic mutation. |  |
| Additive effect size | λ | 0.1 to 10 | Additive effect size controls the variance of trait effect size around mean 0, so that N(0, λ). | Albert et al. 2008; |
| Selection strength (selection model only) |  | 10 to 10000 | The parameter that controls the curve of the fitness function (eq. 3), with higher values resulting in a smaller difference in fitness between trait-differing individuals. |  |

**Table 2:** Means of distance from the optimum (, variance (VA), and covariance among traits for levels of additive effect size, recombination rate, pleiotropy rate, and mutational correlations for Gaussian and House-of-Cards models. Values in bold are mentioned in the main text and featured in Figures 5, 6, or 7. Values in italics indicate means that include outliers that were excluded from figures 6 and 7 for better readability. \* denotes values of interest.

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | | | δ | | | VA | | | Covariance | | |
| Null | House-of-Cards | Gaussian | Null | House-of-Cards | Gaussian | Null | House-of-Cards | Gaussian |
| Additive effect size (α) | |  |  |  |  |  |  |  |  |  |  |
|  | Low |  | **11.151** | **0.715** | **1.384** | **1.035** | **0.241** | **1.747** | **0.069** | **0.002** | **0.015** |
| S.E. | 0.133 | 0.033 | 0.065 | 0.067 | 0.013 | 0.025 | 0.008 | 0.0002 | 0.002 |
| n | 545 | 458 | 445 | 545 | 458 | 445 | 545 | 458 | 445 |
| Medium |  | -\* | **3.109** | **2.085** | - | **9.733** | **3.120** | - | **-0.699** | **0.034** |
| S.E. | - | 1.110 | 0.213 | - | 8.590 | 0.163 | - | 0.709 | 0.010 |
| n | 0 | 6 | 26 | 0 | 6 | 26 | 0 | 6 | 26 |
| High |  | -\* | **1.202** | **2.699** | - | ***110.240*** | **2.943** | - | ***-9.932*** | **-0.022** |
| S.E. | - | 0.258 | - | - | 57.536 | - | - | 5.077 | - |
| n | 0 | 3 | 1 | 0 | 3 | 1 | 0 | 3 | 1 |
| Recombination rate | |  |  |  |  |  |  |  |  |  |  |
|  | Low |  | 10.349 | 0.664 | 1.543 | 0.822 | 4.151 | 1.595 | 0.066 | -0.452 | 0.006 |
| S.E. | 0.197 | 0.066 | 0.101 | 0.063 | 3.080 | 0.026 | 0.013 | 0.371 | 0.003 |
| n | 267 | 46 | 234 | 267 | 46 | 234 | 267 | 46 | 234 |
| Medium |  | 11.452 | 1.372 | 1.467 | 0.898 | 5.941 | 3.412 | 0.068 | -0.355 | 0.043 |
| S.E. | 0.191 | 0.289 | 0.219 | 0.079 | 5.297 | 0.408 | 0.009 | 0.357 | 0.011 |
| n | 219 | 37 | 2 | 219 | 37 | 2 | 219 | 37 | 2 |
| High |  | 13.658 | 0.699 | 1.310 | 2.507\* | 0.232 | 2.048 | 0.089 | 0.002 | 0.024 |
| S.E. | 0.241 | 0.033 | 0.076 | 0.418 | 0.012 | 0.048 | 0.022 | 0.0002 | 0.002 |
| n | 59 | 37 | 236 | 59 | 384 | 236 | 59 | 384 | 236 |
|  | | | δ | | | VA | | | Covariance | | |
| Null | House-of-Cards | Gaussian | Null | House-of-Cards | Gaussian | Null | House-of-Cards | Gaussian |
| Pleiotropy rate | |  |  |  |  |  |  |  |  |  |  |
|  | Low |  | 10.965 | 1.073 | 1.736 | 0.846 | 2.011 | 1.912 | 0.056 | -0.194 | 0.003 |
| S.E. | 0.137 | 0.127 | 0.100 | 0.037 | 1.559 | 0.046 | 0.007 | 0.196 | 0.002 |
| n | 512 | 85 | 270 | 512 | 85 | 270 | 512 | 85 | 270 |
| Medium |  | 14.139 | 1.024 | 0.966 | 4.472 | 3.458 | 1.786 | 0.465 | -0.207 | 0.042 |
| S.E. | 0.288 | 0.114 | 0.077 | 0.623 | 2.438 | 0.040 | 0.098 | 0.167 | 0.003 |
| n | 15 | 83 | 109 | 15 | 83 | 109 | 15 | 83 | 109 |
| High |  | 13.952 | 0.581 | 1.063 | 3.539 | 0.140 | 1.639 | 0.113 | 0.002 | 0.019 |
| S.E. | 0.308 | 0.028 | 0.067 | 1.362 | 0.006 | 0.047 | 0.060 | 0.0002 | 0.003 |
| n | 18 | 299 | 93 | 18 | 299 | 93 | 18 | 299 | 93 |
| Mutational correlation | |  |  |  |  |  |  |  |  |  |  |
|  | Low |  | 13.722 | 0.629 | 1.499 | 1.559 | 0.414 | 1.981 | 0.005 | -0.016 | 0.0003 |
| S.E. | 0.161 | 0.039 | 0.076 | 0.098 | 0.208 | 0.051 | 0.009 | 0.017 | 0.002 |
| n | 117 | 253 | 238 | 117 | 253 | 238 | 117 | 253 | 238 |
| Medium |  | 13.885 | 2.521 | 1.118 | 3.378 | 22.616 | 1.681 | 0.196 | -1.461 | 0.016 |
| S.E. | 0.240 | 0.778 | 0.100 | 0.509 | 21.752 | 0.054 | 0.036 | 1.468 | 0.002 |
| n | 51 | 9 | 83 | 51 | 9 | 83 | 51 | 9 | 83 |
| High |  | 9.983\* | 0.820 | 1.479 | 0.555 | 0.934 | 1.672 | 0.072 | -0.077 | 0.039 |
| S.E. | 0.148 | 0.054 | 0.145 | 0.031 | 0.647 | 0.030 | 0.009 | 0.081 | 0.004 |
| n | 377 | 205 | 151 | 377 | 205 | 151 | 377 | 205 | 151 |

# Figure legends

**Figure 3:** Mean additive variance (VA; panel A) and mean between-trait covariance (B) over 100,000 generations of stabilizing selection of different strengths (). 256 total models were sampled across the spectrum of selection strengths () with an additional 1024 models sampling the null space of parameters ().

**Figure 4:** Euclidean distances from the optimum over models. (A): total distributions of all models. (B): distributions of adapted models with small distance to the optimum.

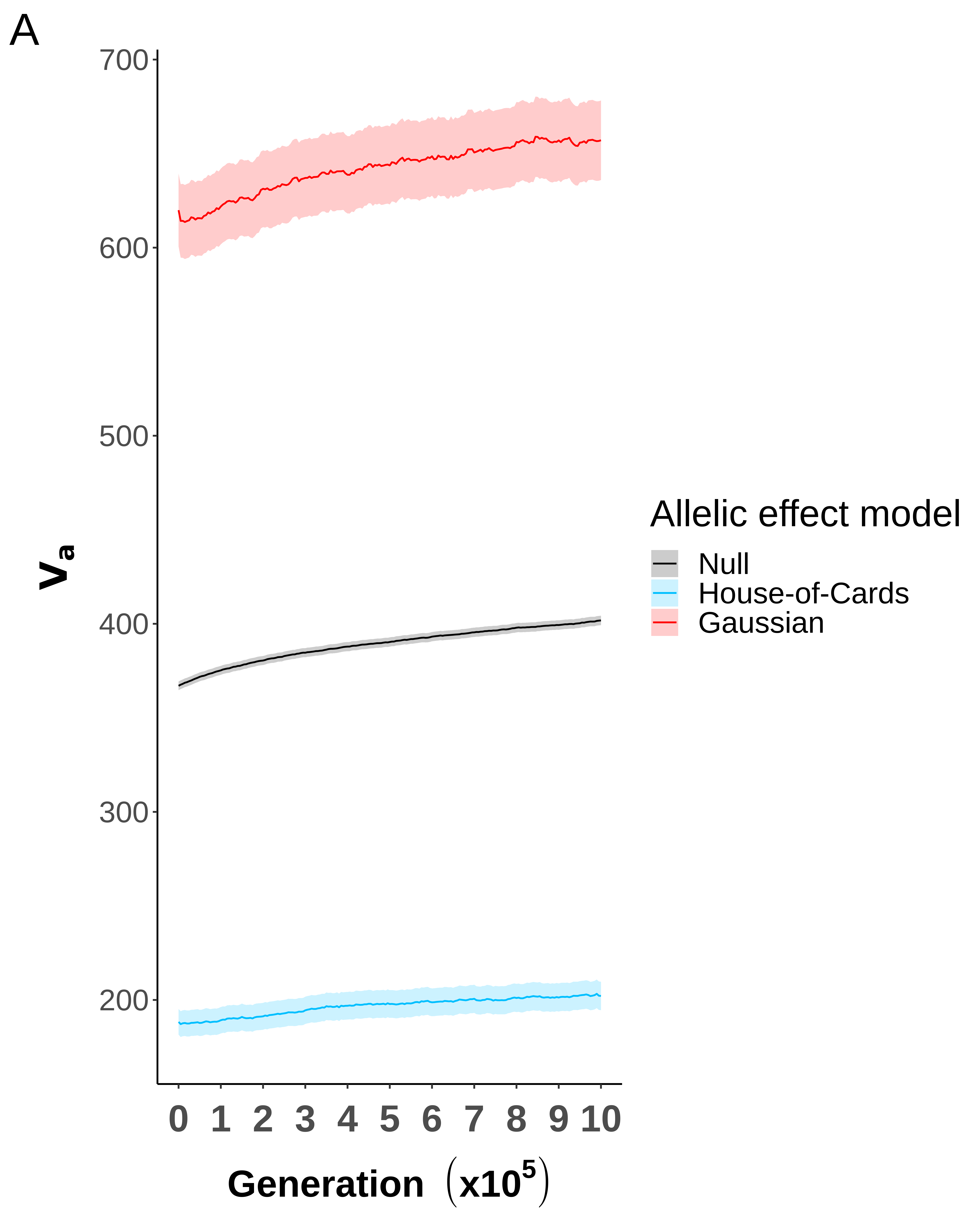
**Figure 5:** Euclidean distances from the optimum () among adapted populations with increasing additive effect size Note that there was only one adapted Gaussian population with high additive effect size.

**Figure 6:** Mean additive variance (VA) among adapted populations with increasing additive effect size Note that there was only one adapted Gaussian population with high additive effect size. Several outliers were removed for improved readability.

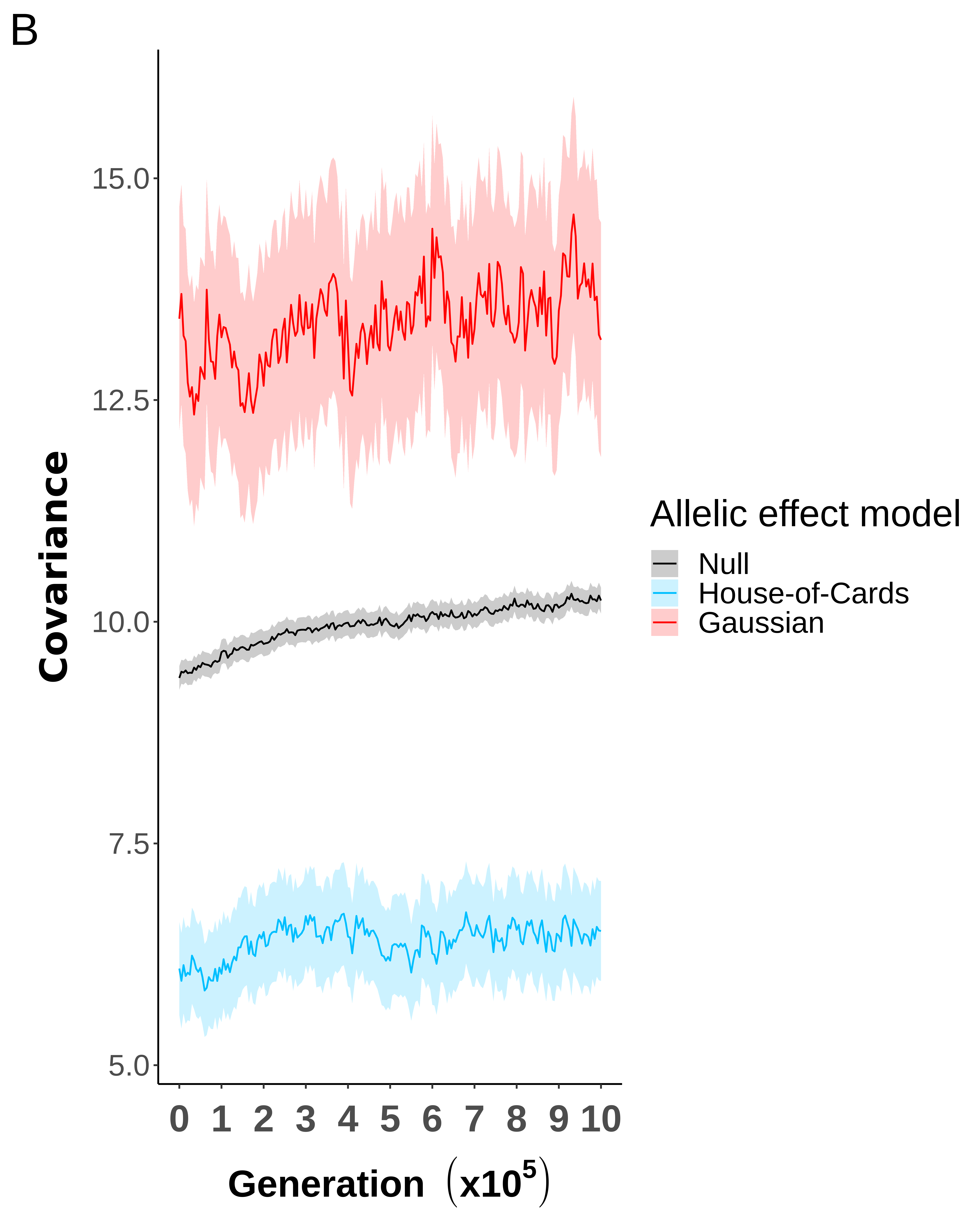
**Figure 7:** Mean trait covariance among adapted populations with increasing additive effect size. Note that there was only one adapted Gaussian population with high additive effect size. Several outliers were removed for improved readability.

**Figure 8:** Density estimates of mutational effect sizes for adapted populations at generation 100,000 under different Continuum of Alleles models, with additive effect size distribution.

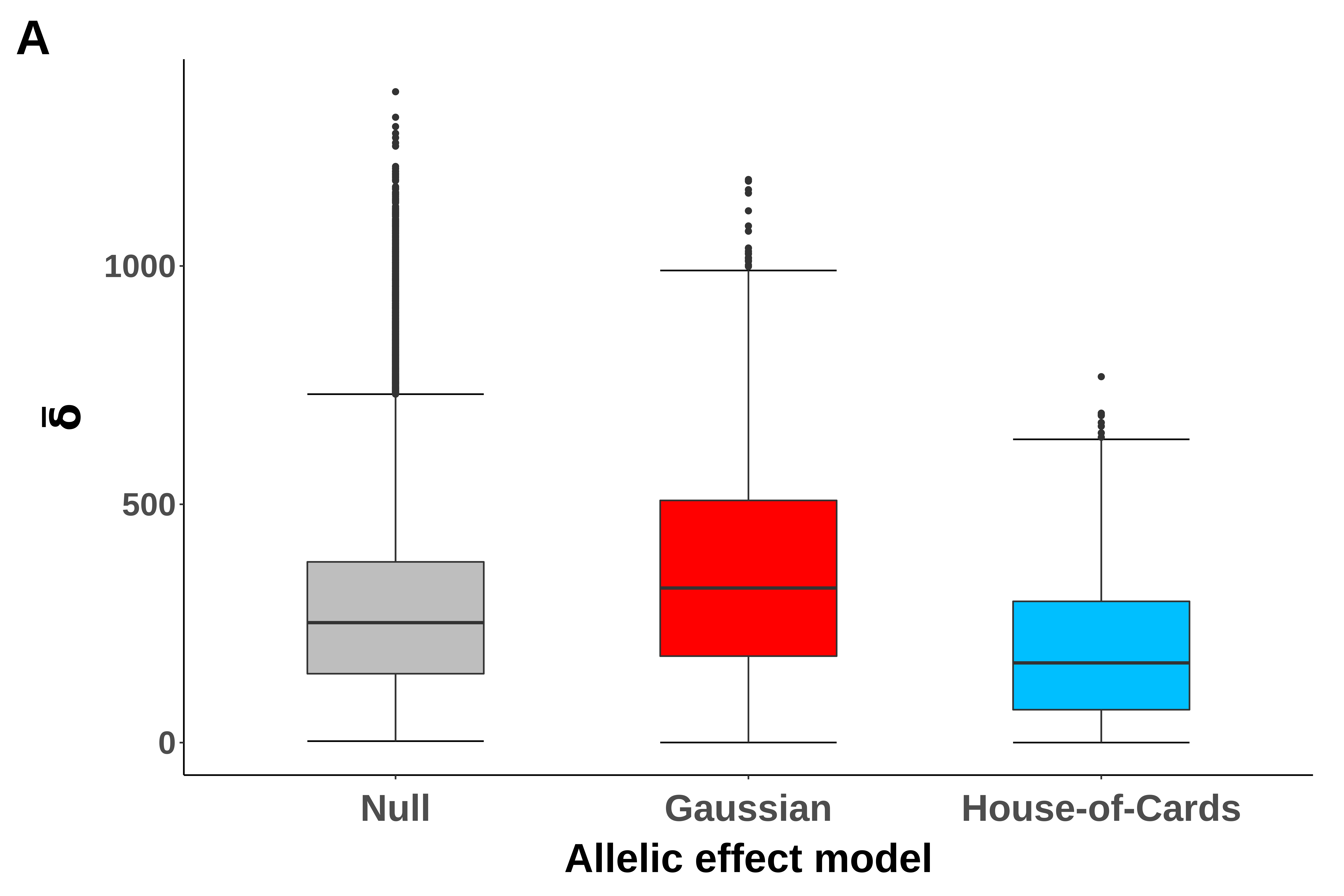
# Figure 3A



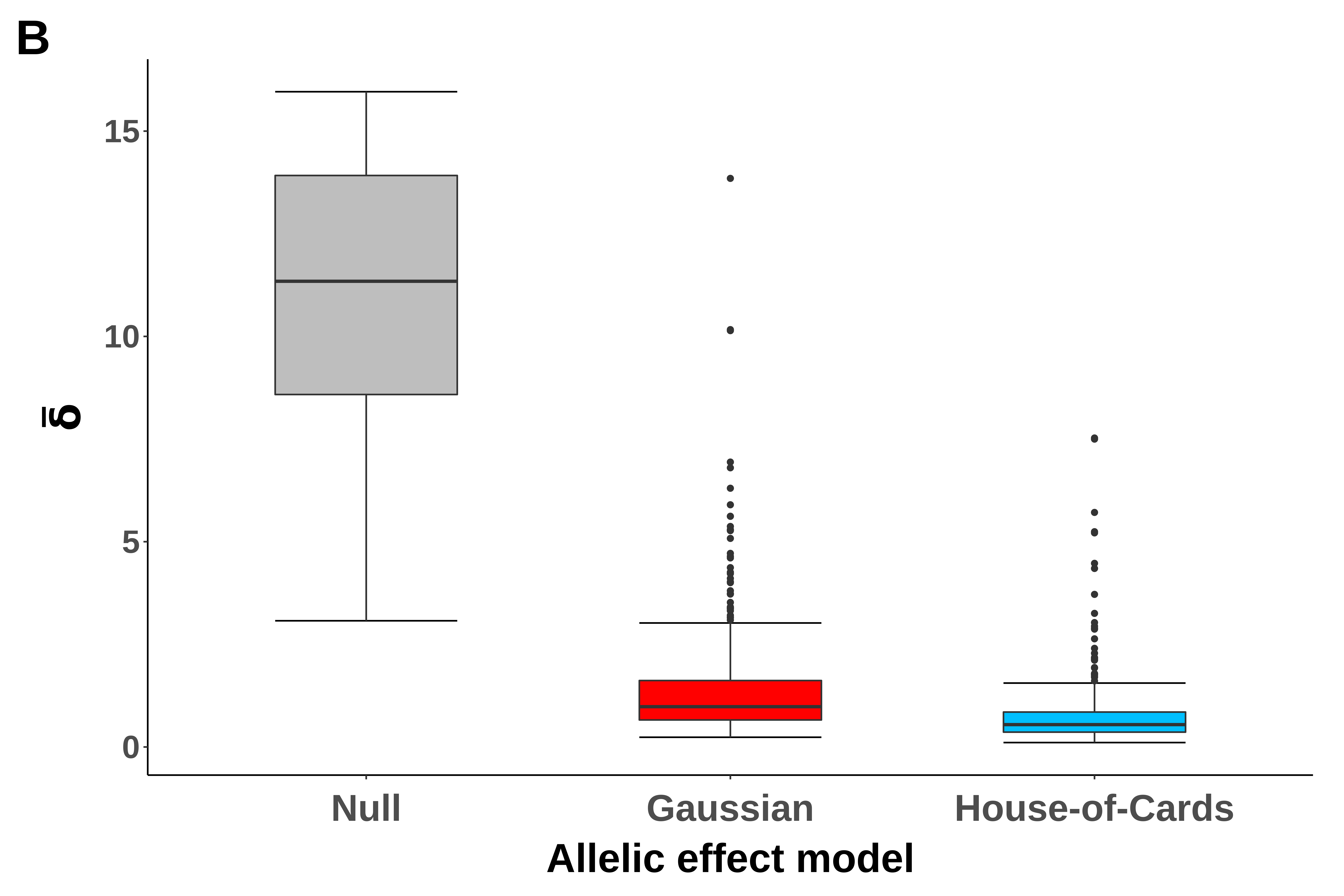
# Figure 3B



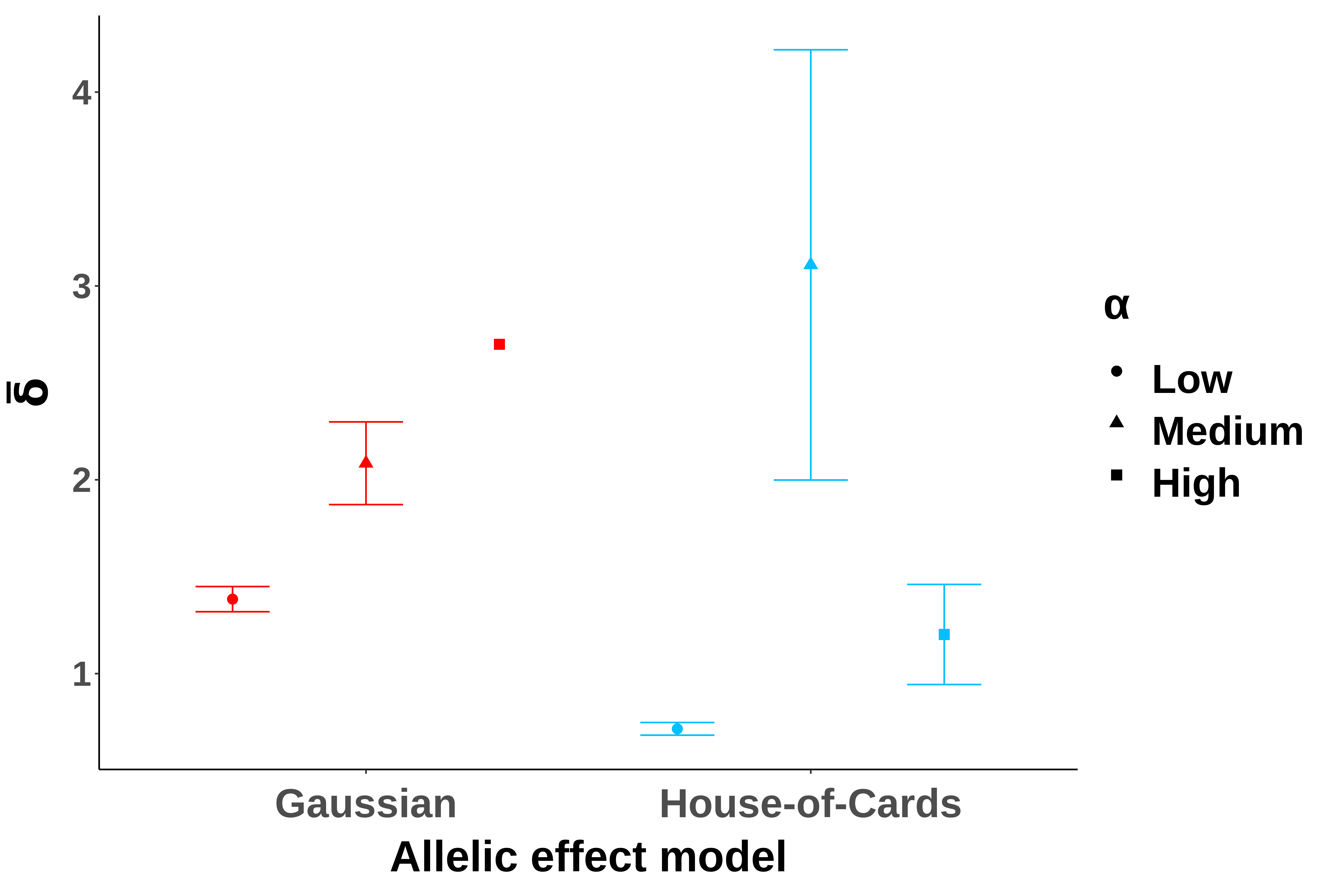
# Figure 4A



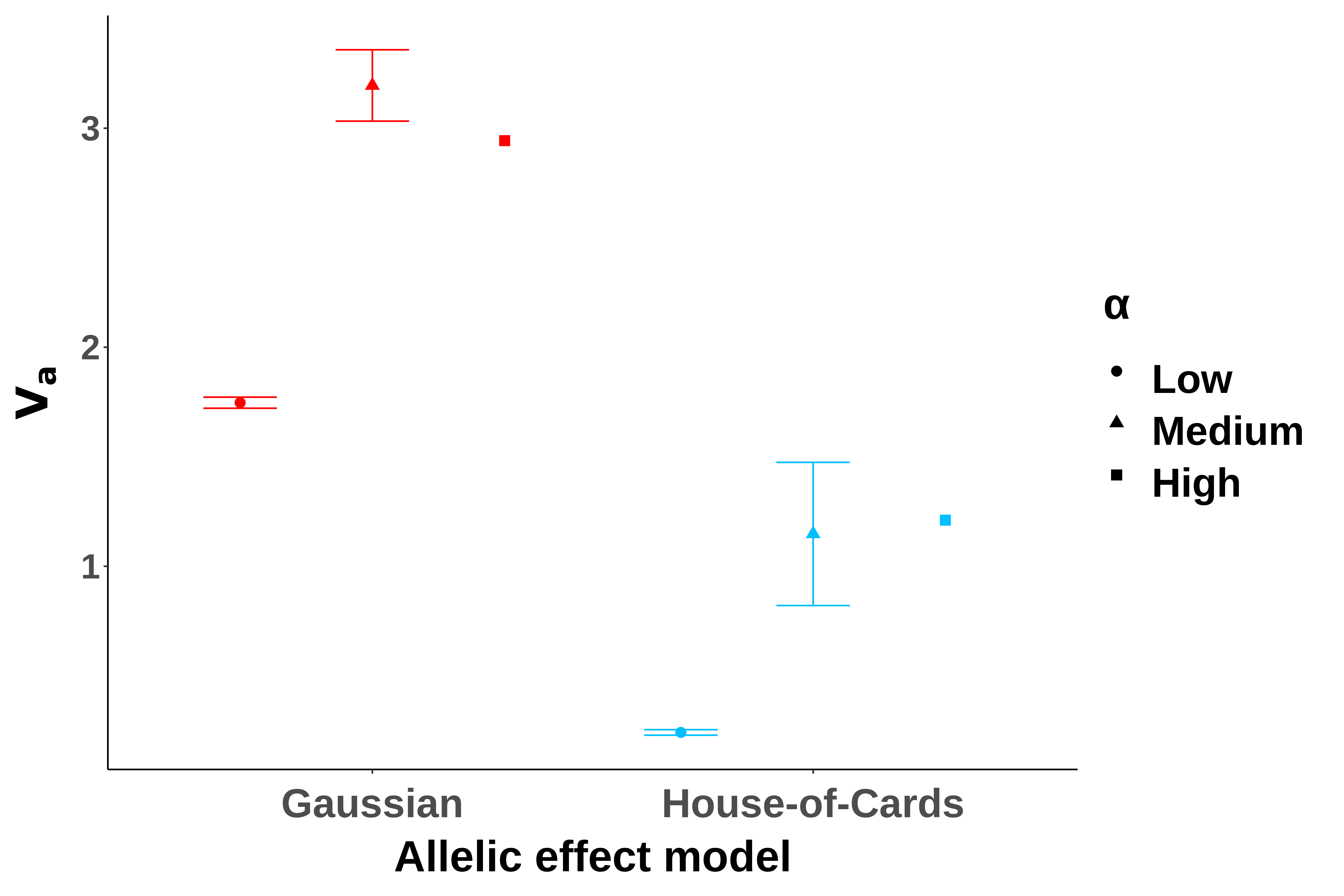
# Figure 4B



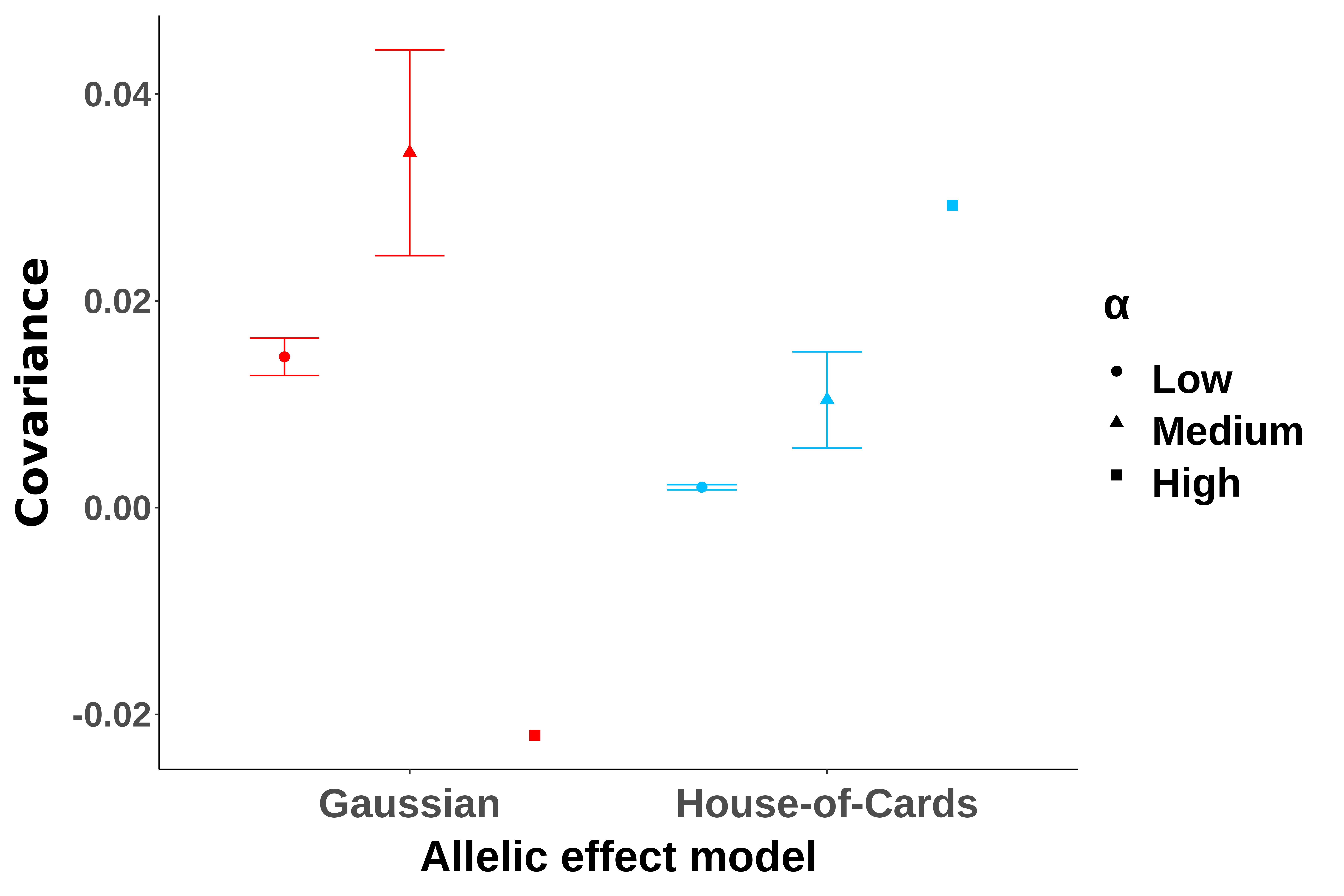
# Figure 5



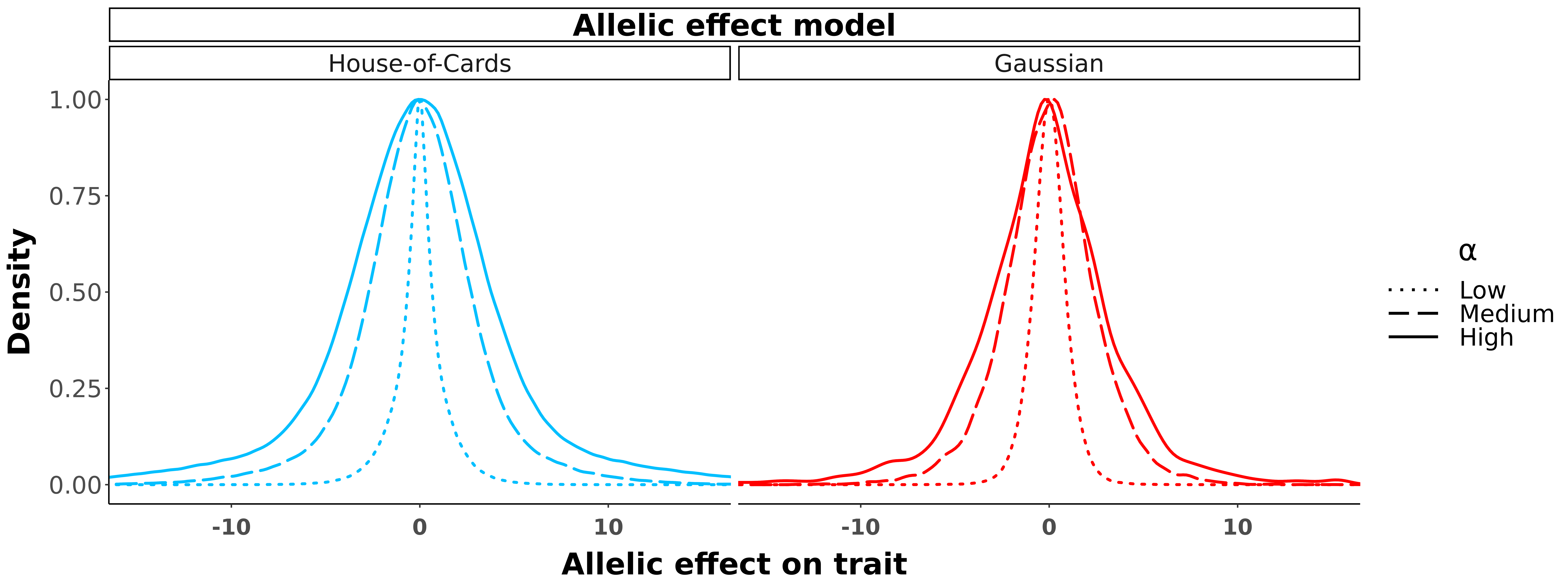
# Figure 6



# Figure 7



# Figure 8



# Supplementary material

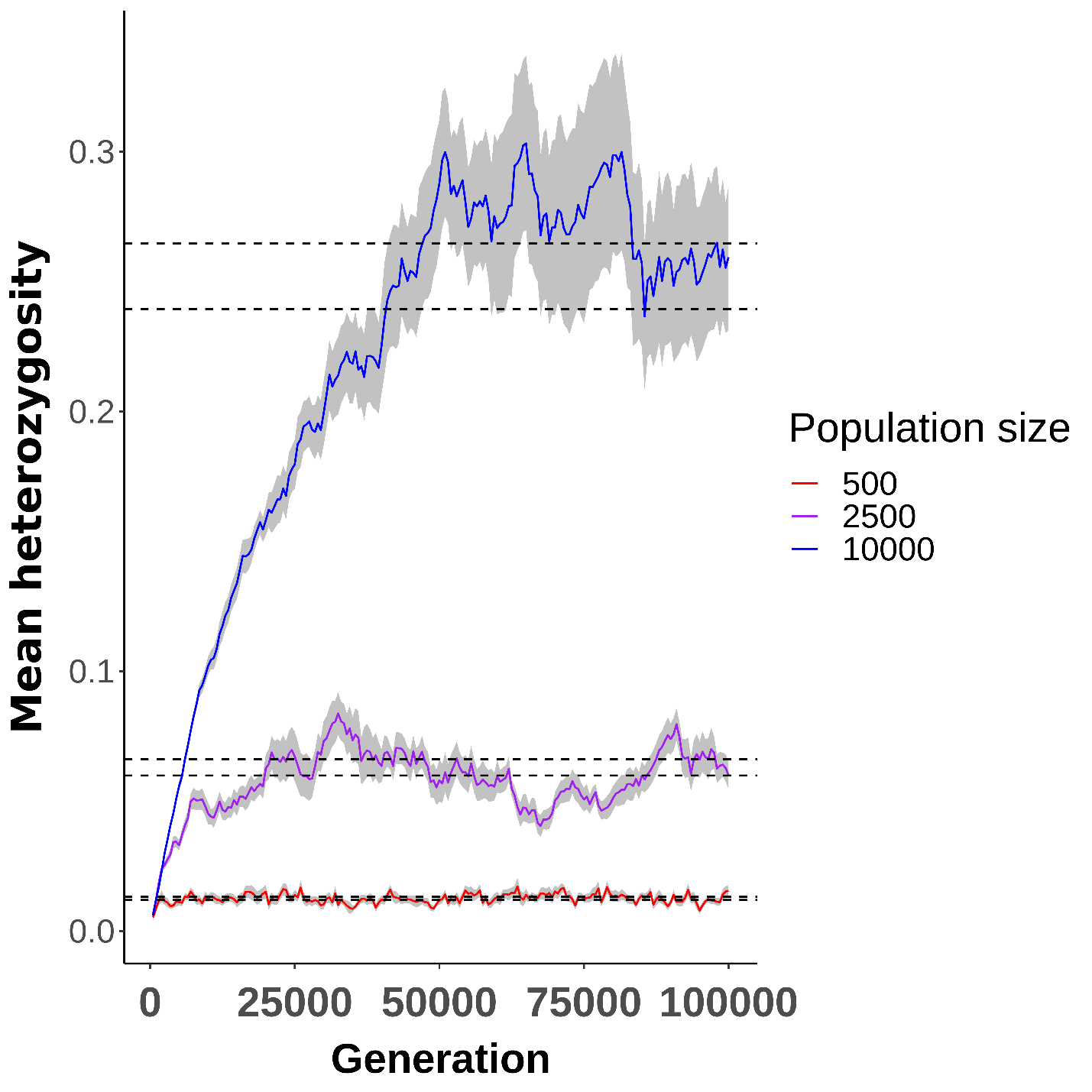


Figure S1: Mean population heterozygosity over time. Lines represent mean trajectories of 20 replicates, with ribbons representing standard errors. Dotted lines represent expected heterozygosities ± 5%, given by .

Figure S2: A) Null model LHC B) Sel model LHC

Figure S3) Data cut from figure 6 - outliers

Figure S4) Data cut from figure 7 - outliers

# References

Agashe, D., and D. I. Bolnick, 2010 Intraspecific genetic variation and competition interact to influence niche expansion. Proc Biol Sci 277**:** 2915-2924.

Agashe, D., J. J. Falk and D. I. Bolnick, 2011 Effects of founding genetic variation on adaptation to a novel resource. Evolution 65**:** 2481-2491.

Aguirre, J. D., E. Hine, K. McGuigan and M. W. Blows, 2014 Comparing G: multivariate analysis of genetic variation in multiple populations. Heredity 112**:** 21-29.

Aston, E., A. Channon, R. V. Belavkin, D. R. Gifford, R. Krasovec *et al.*, 2017 Critical Mutation Rate has an Exponential Dependence on Population Size for Eukaryotic-length Genomes with Crossover. Sci Rep 7**:** 15519.

Barton, N. H., 2017 How does epistasis influence the response to selection? Heredity (Edinb) 118**:** 96-109.

Barton, N. H., and B. Charlesworth, 1998 Why sex and recombination? Science 281**:** 1986-1990.

Blair, G. C., J. Coppock, A. Humphreys, M. Sonnet, L., 2020 estimatr: Fast Estimators for Design-Based Inference, pp.

Brady, S. P., D. I. Bolnick, A. L. Angert, A. Gonzalez, R. D. H. Barrett *et al.*, 2019 Causes of maladaptation. Evol Appl 12**:** 1229-1242.

Careau, V., M. E. Wolak, P. A. Carter and T. Garland, Jr., 2015 Evolution of the additive genetic variance-covariance matrix under continuous directional selection on a complex behavioural phenotype. Proc Biol Sci 282.

Charlesworth, B., and D. Charlesworth, 2010 *Elements of Evolutionary Genetics*. Roberts and Company, Greenwoord Village, Colorado, USA.

Charlesworth, B., M. Nordborg and D. Charlesworth, 1997 The effects of local selection, balanced polymorphism and background selection on equilibrium patterns of genetic diversity in subdivided populations. Genet Res 70**:** 155-174.

Eicker, F., 1967 Limit theorems for regressions with unequal and dependent errors, pp. 59-82 in *Proceedings of the Fifth Berkeley Symposium on Mathematical Statistics and Probability, Volume 1: Statistics*. University of California Press, Berkeley, Calif.

Falconer, D. S. M., T. F. C., 1996 *Introduction to Quantitative Genetics*. Pearson Education Limited, Longmans Green, Harlow, Essex, UK.

Fisher, R. A., 1930 *The genetical theory of natural selection*. The Clarendon press, Oxford, UK.

Fleming, W. H., 1979 Equilibrium Distributions of Continuous Polygenic Traits. Siam Journal on Applied Mathematics 36**:** 148-168.

Haller, B. C., and P. W. Messer, 2019 SLiM 3: Forward Genetic Simulations Beyond the Wright-Fisher Model. Molecular Biology and Evolution 36**:** 632-637.

Hayes, A. F., and L. Cai, 2007 Using heteroskedasticity-consistent standard error estimators in OLS regression: An introduction and software implementation. Behavior Research Methods 39**:** 709-722.

Helton, J. C., and F. J. Davis, 2003 Latin hypercube sampling and the propagation of uncertainty in analyses of complex systems. Reliability Engineering & System Safety 81**:** 23-69.

Houle, D., 1998 How should we explain variation in the genetic variance of traits? Genetica 102-103**:** 241-253.

Huber, P. J., 1967 The behavior of maximum likelihood estimates under nonstandard conditions, pp. 221-233 in *Proceedings of the Fifth Berkeley Symposium on Mathematical Statistics and Probability, Volume 1: Statistics*. University of California Press, Berkeley, Calif.

Kimura, M., 1965 A stochastic model concerning the maintenance of genetic variability in quantitative characters. Proc Natl Acad Sci U S A 54**:** 731-736.

Kimura, M., and J. F. Crow, 1964 The Number of Alleles That Can Be Maintained in a Finite Population. Genetics 49**:** 725-738.

Lande, R., 1975 The maintenance of genetic variability by mutation in a polygenic character with linked loci. Genet Res 26**:** 221-235.

Lindeman, R. H. M., P.F. Gold, R.Z., 1980 *Introduction to Bivariate and Multivariate Analysis*. Scott, Foresman, Glenview, IL.

Long, J. A., 2020 jtools: Analysis and Presentation of Social Scientific Data

Lumley, T., P. Diehr, S. Emerson and L. Chen, 2002 The importance of the normality assumption in large public health data sets. Annu Rev Public Health 23**:** 151-169.

Lynch, M., and R. Lande, 1998 The critical effective size for a genetically secure population. Animal Conservation 1**:** 70-72.

Melo, D., G. Garcia, A. Hubbe, A. P. Assis and G. Marroig, 2015 EvolQG - An R package for evolutionary quantitative genetics. F1000Research 4**:** 925.

Nesse, R. M., 2005 Maladaptation and natural selection. Q Rev Biol 80**:** 62-70.

Pujol, B., and J. R. Pannell, 2008 Reduced responses to selection after species range expansion. Science 321**:** 96.

R Developmental Core Team, 2019 R: A language and environment for statistical computing, pp. R Foundation for Statistical Computing, Vienna, Austria.

Sztepanacz, J. L., and M. W. Blows, 2017 Artificial Selection to Increase the Phenotypic Variance in gmax Fails. Am Nat 190**:** 707-723.

Thornton, K. R., 2019 Polygenic Adaptation to an Environmental Shift: Temporal Dynamics of Variation Under Gaussian Stabilizing Selection and Additive Effects on a Single Trait. Genetics 213**:** 1513-1530.

Turelli, M., 1984 Heritable Genetic-Variation Via Mutation Selection Balance - Lerch Zeta Meets the Abdominal Bristle. Theoretical Population Biology 25**:** 138-193.

Walsh, B., and M. Lynch, 2018 *Evolution and selection of quantitative traits*. Oxford University Press, New York, NY.

White, H., 1980 A Heteroskedasticity-Consistent Covariance-Matrix Estimator and a Direct Test for Heteroskedasticity. Econometrica 48**:** 817-838.

Williams, G. C., 1966 *Adaptation and natural selection; a critique of some current evolutionary thought*. Princeton University Press, Princeton, N.J.,.

Xu, L., H. Chen, X. Hu, R. Zhang, Z. Zhang *et al.*, 2006 Average gene length is highly conserved in prokaryotes and eukaryotes and diverges only between the two kingdoms. Mol Biol Evol 23**:** 1107-1108.

Zhang, X. S., 2012 Fisher's geometrical model of fitness landscape and variance in fitness within a changing environment. Evolution 66**:** 2350-2368.