Online Supplemental Information

WITHIN-LOCUS INCOMPATIBILITIES: MODEL

In the simplest possible scenario, the two new mutations arise at the same locus at the same time, but in different patches. In this model, the ancestral allele is *b*, and the two new alleles are *a* and *A*. The patch in which *a* arises is arbitrarily designated Patch 1, and the patch in which *A* arises is designated Patch 2. Since we are interested in the scenario in which both alleles are advantageous compared to ancestral allele *b*, but incompatible with each other, we have $w_{AA} = w_{Ab} > w_{aa} = w_{ab} > w_{bb} = 1 \geq w_{Aa}$ (where w_{xy} is the fitness of an individual with alleles *x* and *y* at the locus of interest; see Table 1 in main text).

Allele frequencies were tracked as follows. If q_i is the frequency of allele *a* in patch *i* and p_i is the frequency of allele *A* in patch *i*, then the frequency of allele *b* in patch *i* is $r_i = 1 - q_i - p_i$, and the frequency of *A* in patch *i* in the next generation will be its proportional weighted fitness (e.g., Graur & Li 2000, p. 42):

$$\frac{p_i q_i w_{Aa} + p_i r_i w_{Ab} + p_i^2 w_{AA}}{2p_i q_i w_{Aa} + 2p_i r_i w_{Ab} + 2q_i r_i w_{ab} + p_i^2 w_{AA} + r_i^2 w_{bb} + q_i^2 w_{aa}}. \quad (1)$$

Similar equations were constructed for updating the frequencies of *a* and *b*. Other parameters, including variation in q_{thresh} and t_{contact}, were as for the epistasis model (see main text). Also as described for the epistasis model, two sets of simulation runs were conducted with the same parameter value ranges for fitnesses, *m*, q_{thresh}, and t_{contact}.

WITHIN-LOCUS INCOMPATIBILITIES: RESULTS

Using the same classification scheme as for the epistasis model discussed in the main text, we found that some mutation-order divergence occurred under a range of conditions (online figures S1-S3). However, as in the epistasis model, strong population divergence was unlikely unless gene flow was absent or extremely low. Extreme M.O. and Moderate M.O. were never observed at migration rates above 0.01 and consistently occurred only when $m < 0.001$.

Three factors other than gene flow strongly affected the probability of mutation-order divergence. First, as the difference in selective advantage of the two new mutations decreased

(i.e., as the ratio w_{AA}/w_{aa} decreased toward unity), mutation-order divergence became more likely (online figures S1-S3). This was because large differences in relative fitness caused the most fit allele, *A*, to fix in both patches. Second, staggering the timing of origination of mutations affected the outcome, but its effect depended upon other parameters in the model. When the first mutation (allele *a*) originated slightly before the second, mutation-order divergence was promoted, for example being observed when differences in the selective advantage of new mutations were relatively large (and larger than possible for extreme M.O. in the case of simultaneous emergence of both mutations, compare online figure S3a to S3b and online figure S3d to S3e). In contrast, when the first mutation, *a*, originated long before the second mutation (i.e. was given a large 'leg-up'), the less advantageous allele could actually spread between and even fix in both patches, sometimes preventing mutation-order divergence (online figures S2 and S3). In part, this was due to the second mutation, *A*, which is most advantageous on its own, often finding itself in a low fitness heterozygote (*Aa*). The latter effect could only be ameliorated by extremely low migration rates that prevented allele *a* from rising in frequency in Patch 2. Third, an initial period of allopatric divergence tended to promote mutation-order divergence (online figure S3). Fourth, hybrid fitness has some effects, but they were weak: as w_{Aa} decreased from 0.9 (moderate incompatibility) to 0.001 (nearly complete incompatibility), the size of the '*A* fixed' region was slightly reduced (compare top and bottom rows of online figure S2) as it became harder for allele *A* to spread to fixation in both patches.

DIFFERENCES BETWEEN WITHIN- AND BETWEEN-LOCUS MODELS

These results of the two models were qualitatively, and largely quantitatively, very similar. The main difference between the single locus and epistasis models was that in the epistasis model Moderate M.O. was often observed at parameter values where Extreme M.O. was seen in the single locus model (e.g. figure 3 versus online figure S2). This difference is easily understood by noting the fact that in the epistasis model, the high fitness genotypes are those that include ancestral alleles (*AAbb* and *aaBB*). By contrast, in the single-locus model, each derived allele directly competes with the one ancestral allele, since all are at the same locus. Thus in the epistasis model, the preservation of ancestral alleles combined with migration makes our 'Extreme M.O.' criterion harder to achieve.

Figure legends for supplementary online figures

online figure S1. The effect of migration rate and relative fitnesses on mutation-order divergence in the single-locus model. The circles with dashed line show the frequency of allele *A* in Patch 2 (in which allele *A* initially arose). Squares with solid line show the same for Patch 1 (in which allele *a* initially arose). Relative fitnesses, migration rate, and q_{thresh} all interact to determine which allele fixes and whether mutation-order divergence occurs. Extreme mutation-order divergence occurs in this figure when $p_1 \approx 0$ ($q_1 \approx 1$) and $p_2 \approx 1$ ($q_2 \approx 0$).

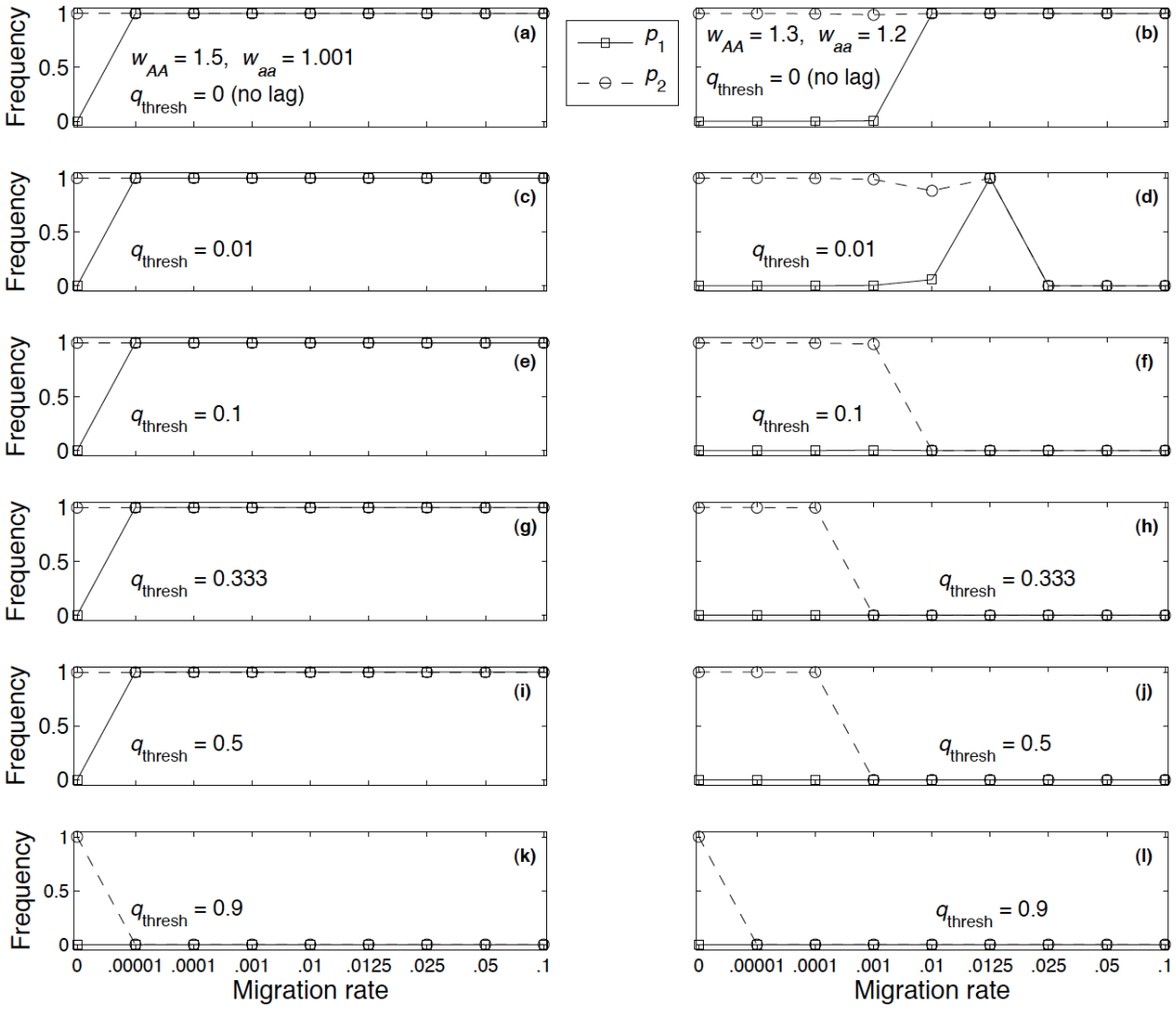
online figure S2. Outcomes of simulations in the single-locus model as influenced by migration rate, the ratio (w_{AA}/w_{aa}) of fitnesses of the two favored alleles, the lag between when the two different mutations arise (q_{thresh}), and the fitness of the genotype with the two incompatible alleles (w_{Aa}). Each panel represents outcomes from 10,000 simulations for varying combinations of the fitness ratio (w_{AA}/ w_{aa}) and the migration rate. Panel (e) shows the meaning of the different degrees of shading for all six panels. See main text for definitions of outcomes and abbreviations.

online figure S3. The role of a period of t_{contact} generations of allopatric divergence on the outcome of mutation-order divergence in the single-locus model. Shading is as for online figure S2. Top row: parameter values as in online figure S2a; bottom row: parameters values as for online figure 2f.

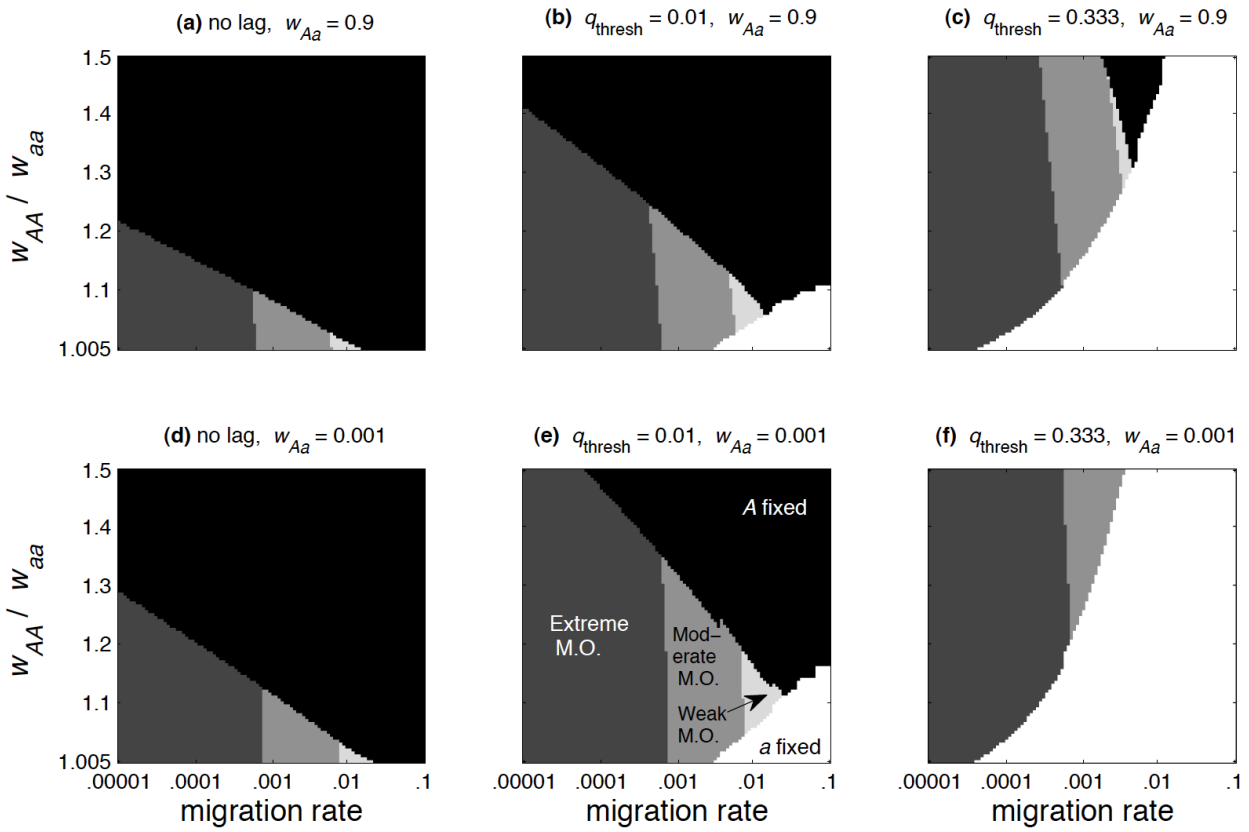
online figure S4. The role of extended periods of allopatric divergence in the epistasis model. Top row: parameter values from figure 3a (main text); bottom row: parameters values as figure 3f (main text). Details are as for figure 4 in the main text.

online figure S5. The effects of a range of parameter values for hybrid fitness and q_{thresh} for 30 generations of allopatric divergence in the epistasis model. For comparison, q_{thresh} and t_{contact} take the same values here as in the six panels of figure 3 of main text.

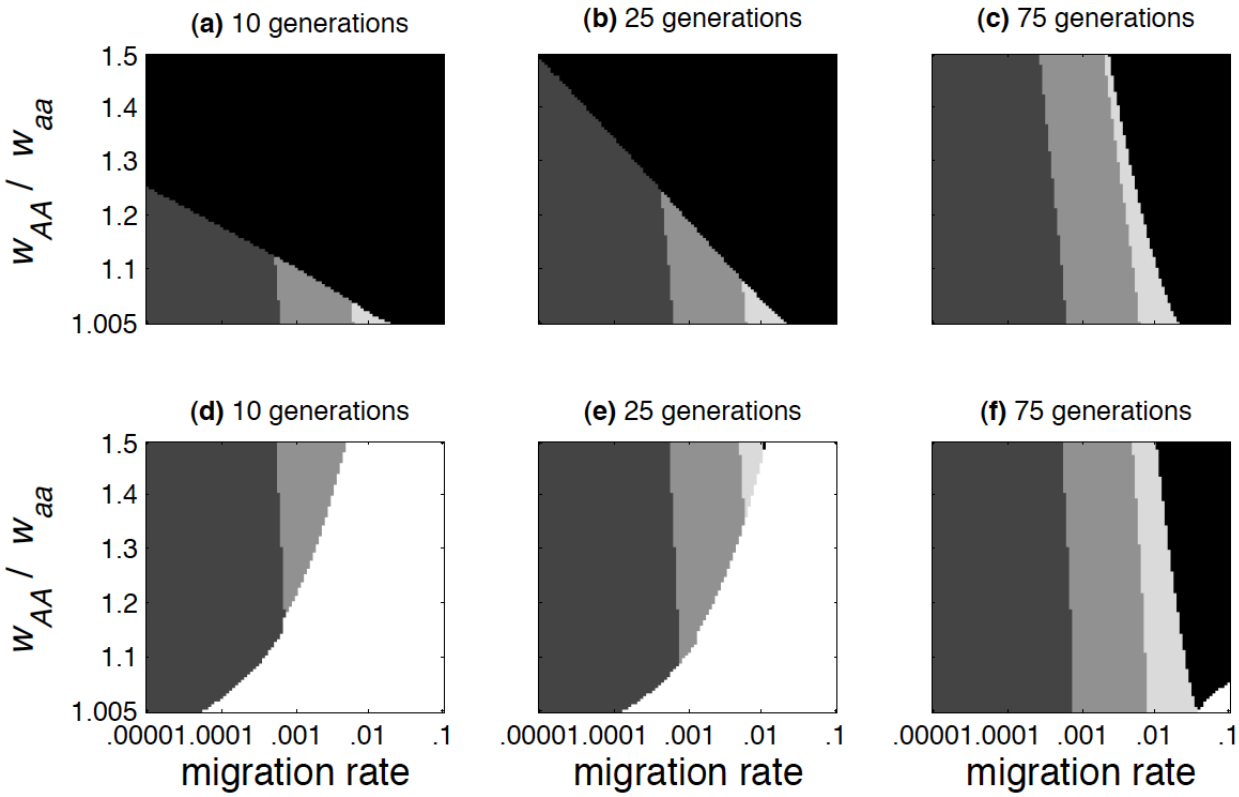
online figure S1



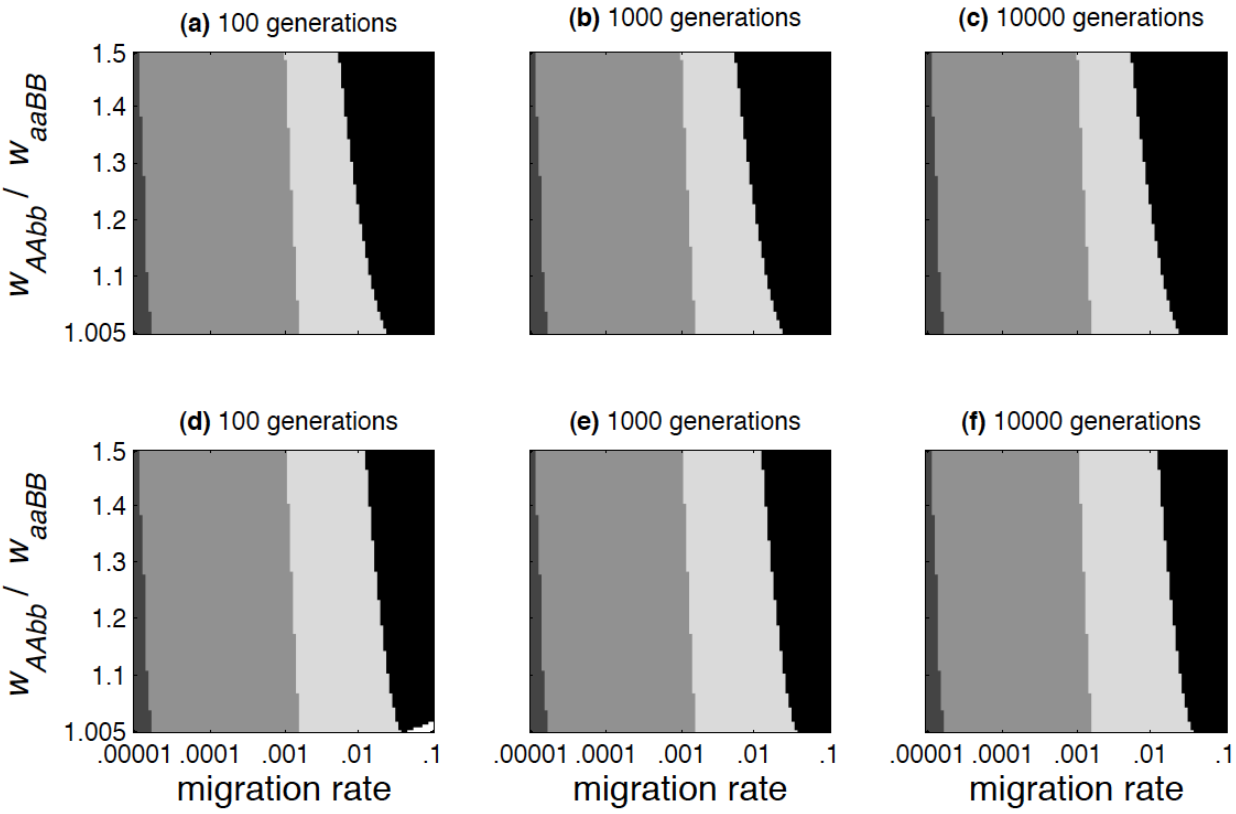
online figure S2



online figure S3



online figure S4



online figure S5