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Molecular mechanisms of dominance evolution in Müllerian mimicry

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Natural selection acting on dominance between adaptive alleles at polymorphic loci can be sufficiently strong for dominance to evolve. However, the molecular mechanisms underlying such evolution are generally unknown. Here, using Müllerian mimicry as a case-study for adaptive morphological variation, we present a theoretical analysis of the invasion of dominance modifiers altering gene expression through different molecular mechanisms. Toxic species involved in Müllerian mimicry exhibit warning coloration, and converge morphologically with other toxic species of the local community, due to positive frequency-dependent selection acting on these colorations. Polymorphism in warning coloration may be maintained by migration–selection balance with fine scale spatial heterogeneity. We modeled a dominance modifier locus altering the expression of the warning coloration locus, targeting one or several alleles, acting in *cis* or *trans*, and either enhancing or repressing expression. We confirmed that dominance could evolve when balanced polymorphism was maintained at the color locus. Dominance evolution could result from modifiers enhancing one allele specifically, irrespective of their linkage with the targeted locus. Nonspecific enhancers could also persist in populations, at frequencies tightly depending on their linkage with the targeted locus. Altogether, our results identify which mechanisms of expression alteration could lead to dominance evolution in polymorphic mimicry.

KEY WORDS: **Balancing selection, enhancers, gene expression, modifier, polymorphism, spatial heterogeneity.**

Understanding the mechanisms driving the emergence and persistence of adaptive variation is a central question for evolutionary biologists. The level of expression of new alleles is a crucial factor both for their invasions and their maintenance in natural populations. Most mutations are more likely to cause a loss or impairment of function, rather than to lead to a new adaptive phenotype. Deleterious mutations are thus generally recessive (Orr 1991), because heterozygotes usually carries a functional alternative allele. For instance, for genes encoding enzymes, new mutations easily reduce enzyme activity and, because of a nonlinear relationship between enzyme activity and biochemical reaction, heterozygotes for such mutations often show an enzymatic flux similar to wild-type homozygotes (Kacser and Burns 1981). Generally, deleterious alleles are predicted to be recessive due to the intrinsic properties of the encoded proteins (Wright 1934). On the contrary, adaptive variants are predicted to be generally dominant to ancestral ones: because new variants mainly occur as heterozygotes, and dominant mutations are more likely to invade by positive selection than recessive mutations easily lost by drift, a phenomenon known as "Haldane's sieve" (Haldane 1956). However, adaptive mutants recruited from standing variation after an environmental change should be less prone to Haldane's sieve (Orr and Betancourt 2001), stressing the importance of the evolutionary history of alleles. Dominance can indeed vary between

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allelic combinations (a given allele can be recessive to some alleles and dominant to others), between tissues (Hawkins et al. 2014), and between populations or species (Wittkopp et al. 2008). This suggest that dominance levels are a property emerging from the action of natural selection on allelic combinations at a locus, but also that dominance levels may evolve (Fisher 1930). However, the evolutionary mechanisms underlying such evolution of dominance are currently underexplored.

Loci maintaining adaptive polymorphism represent an ideal framework to investigate the evolution of dominance. Indeed heterozygous genotypes are frequently formed and may represent a sizeable fraction of the population exposed to selection, favoring the evolution of dominance between alleles (Otto and Bourguet 1999). One of the major drivers of balanced polymorphism is negative frequency-dependent selection (NFDS), which may stem from various ecological mechanisms and has been detected in various species. For instance, NFDS caused by intraspecific competition, acting on the gene *foraging* in *Drosophila melanogaster* larvae (Fitzpatrick et al. 2007) provides a favorable framework for the evolution of dominance (Peischl and Burger 2008). In flowering plants of the family Brassicaceae, the polymorphic self-incompatibility locus is under NFDS through the effect of allelic variation on mate availability, and undergoes strong selection on dominance (Llaurens et al. 2009; Schoen and Busch 2009). Modification of dominance is also documented in butterflies involved in polymorphic Batesian mimicry, where distinct wing-pattern morphs in edible species are under NFDS for the mimicry of their respective toxic models. Clarke and Sheppard (1960) described dominance variations among alleles of the locus *H* controlling color pattern variation in the Batesian mimic *Papilio dardanus,* and found strong dominance when crossing morphs living in sympatry (heterozygote wing phenotype similar to one of the corresponding homozygotes), whereas crosses between morphs sampled from distant populations produced heterozygotes with intermediate patterns. Variations of dominance were therefore consistent with an evolution of dominance in polymorphic populations in response to selection against nonmimetic intermediate phenotypes. Further results by Nijhout (2003) suggested that dominance between alleles found in sympatry may be shaped by the joint evolution of alleles at the major wing-patterning gene and their genetic background. These results are consistent with theoretical predictions by Charlesworth and Charlesworth (1975) who showed that dominance of Batesian mimicry alleles can evolve through the effect of independent modifier loci. When polymorphism is maintained by selection–migration equilibrium, however, the evolution of dominance modifiers may be hampered by gene flow limiting the fixation of modifier alleles.

Unlike Batesian mimicry, the adaptive resemblance of defended species, or Müllerian mimicry, is thought to evolve through the benefits of spreading the mortality incurred by predator education across species, which translates into strong positive frequency-dependent selection on warning signals at the local scale. Most species involved in Müllerian mimicry are indeed monomorphic at the local scale, but in certain cases stable polymorphism can be maintained through selection–migration equilibrium, such as in the Amazonian butterfly *Heliconius numata* (Joron et al. 1999). In this species, mimicry polymorphism is controlled by a single supergene locus (Joron et al. 2006) with multiple adaptive alleles associated with distinct genomic rearrangements (Joron et al. 2011). Dominance is strict between alleles found in sympatry, but is relaxed between alleles from different populations, highlighting the effect of selection acting on dominance (Le Poul et al. 2014). Furthermore, Le Poul et al. (2014) detected two distinct control of dominance, depending on the genomic arrangements of the alleles, which may suggest a link between the evolution of dominance and the diversification of alleles in the natural populations of *H. numata*. In contrast to the significant effect of the genomic background on the dominance variations found in *P. dardanus*, dominance in *H. numata* seems more directly linked to the different mimetic alleles. NFDS promoting local polymorphism in the Batesian mimic *P. dardanus* might indeed favor the fixation of dominance modifiers at local scale irrespective of their linkage with the targeted locus. However, balancing selection arising from selection–migration balance in *H. numata* limits local adaptation and might therefore favor the evolution of dominance by linked modifiers traveling across populations in association with the targeted color pattern alleles. This hypothesis needs to be tested by investigating the interaction between selection acting on dominance and allele migration, to better understand the evolution of dominance in heterogeneous environment. Evidence for the concurrent action of distinct controls of dominance in *H. numata* (Le Poul et al. 2014) also stresses the importance of understanding the genetic mechanisms underlying changes in dominance. Dominance variation can arise via many distinct mechanisms, including changes of expression levels by *cis*- or *trans*-acting factors, which might lead to different evolutionary outcomes (Lemos et al. 2008).

In a previous model describing a polymorphic species involved in Müllerian mimicry, the conditions for the persistence of a balanced polymorphism of mimetic alleles were investigated (Joron and Iwasa 2005). By modeling two populations with contrasted mimetic assemblages (see Fig. 1), strong selection due to spatial heterogeneity of mimetic communities allowed balanced polymorphism for a medium range of migration rates. When migration increased, polymorphism tended to be lost, because migration prevented local adaptation of mimetic alleles (Joron and Iwasa 2005). Similarly, when the toxicity of the focal species increased, balanced polymorphism was lost because individuals were highly defended against predators regardless of their resemblance to the local communities. Here, to study whether

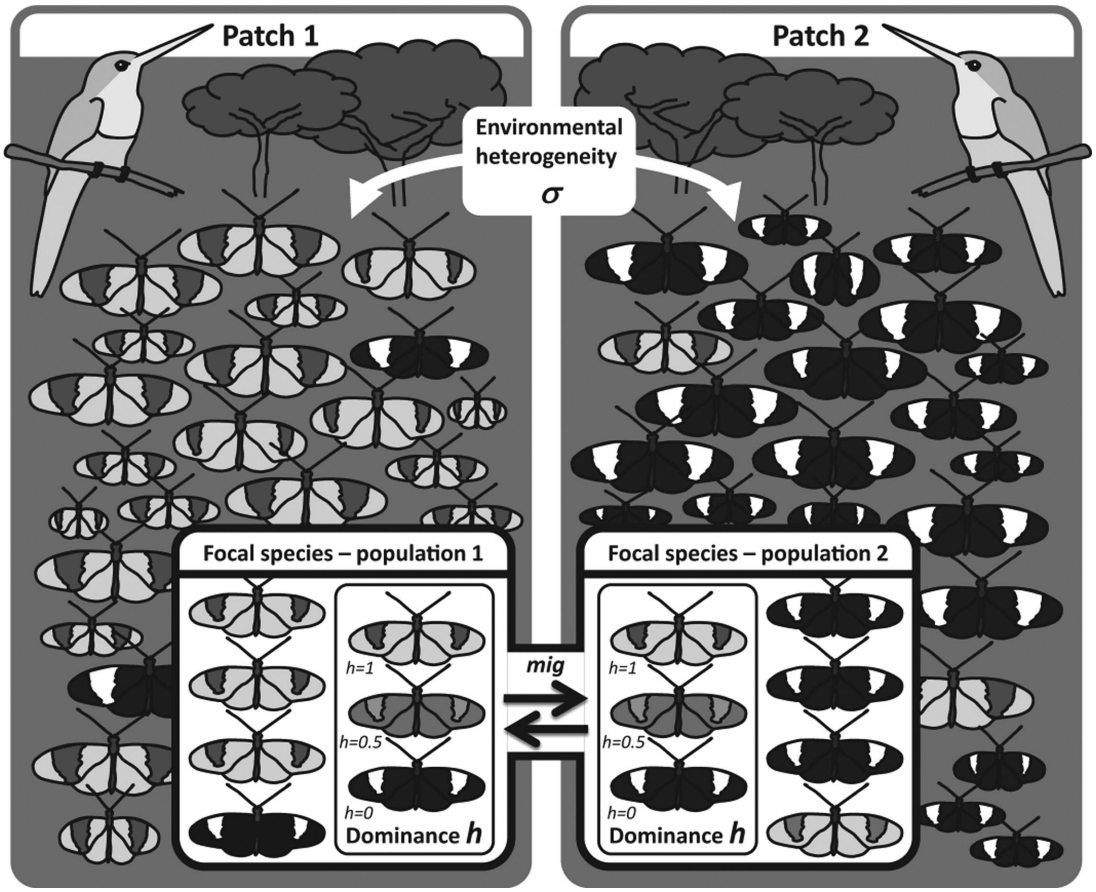

Figure 1. **Schematic view of spatially heterogeneous selection on mimicry in different communities of defended butterflies. In patch 1, the light gray morph (phenotype A) is more frequent than the black morph (phenotype B) in the community of defended butterflies and thus benefit from greater protection against predators, whereas the reverse is true in patch 2. This spatial heterogeneity in mimetic community is described by the parameter σ that represents the decrease (−σ) or increase (+σ) in predation risk for individuals being, respectively, mimetic or nonmimetic to the local community of butterflies in the patch. Individuals from the focal species can move from one patch to the other with a migration rate *mig*. Polymorphism in color patterns could be maintained through an equilibrium between (1) local positive frequency-dependent selection acting in opposite directions in patches 1 and 2 and (2) migration between the two patches. Homozygotes from the focal species display either the light gray morph (mimetic in patch 1) or the black morph (mimetic in patch 2). The phenotype of heterozygotes depends on the dominance coefficient *h*, ranging from 0 (black morph) to 1 (light gray morph). Codominant phenotypes display an intermediate nonmimetic phenotype when $h = 0.5$ (gray morph). The persistence of adaptive polymorphism through migration–selection balance generates a large proportion of heterozygotes, thus favoring the evolution of dominance between mimetic alleles.**

dominance among mimetic alleles can evolve, we focused on a parameter range where balanced polymorphism is maintained, generating a high frequency of heterozygotes subject to natural selection on dominance. Besides local adaptation in a spatially heterogeneous landscape, selection against heterozygotes displaying an intermediate, nonmimetic phenotype may promote stronger dominance levels (Llaurens et al. 2013). We investigate the evolution of dominance between alleles of a mimicry locus via the action of a separate modifier locus, with varying levels of linkage. We also explicitly tested different types of modifier action on dominance at the molecular level, changing the expression levels of the mimetic alleles, to draw predictions on the mechanisms likely to favor the evolution of dominance at a locus under balancing selection.

Materials and Methods

MODEL

As in Llaurens et al. (2013), we assumed two populations of a focal species involved in Müllerian mimicry with other local species. The environment, represented by the community of local species involved in mimicry, was considered spatially heterogeneous for the focal species: population 1 was situated in a patch where the local community is dominated by species displaying phenotype

A as a warning color, and population 2 in a patch where mimetic species displayed phenotype *B* as a warning color (see Fig. 1 for illustration). The focal species was polymorphic for those two phenotypes, corresponding to the mimicry optima *A* or *B*. The focal species was considered diploid and color pattern variation controlled by a single locus, referred as the color pattern locus *P* hereafter.

The phenotype of each genotype in the polymorphic mimetic species was described by a phenotypic value *p* set to 1 or 0 when individuals displayed phenotypes A or B, respectively. At initial stage, heterozygotes *ab* had the phenotype $p_{het} = h$, with *h* as the dominance coefficient.

Resemblance R_{ij} between pairs of individuals *i* and *j* exhibiting phenotypes p_i and p_j, respectively, was computed using the following generalization function describing the ability of bird predators to distinguish phenotypes. $R_{ij} = 1$ when the two phenotypes were identical and $R_{ij} = 0$ when phenotypes were perceived as different by predators.

$$R_{ij} = g\left(1 - \left(p_i - p_j\right)\right), \tag{1}$$

and

$$g(x) = e^{-x^2/2\gamma^2}, \tag{2}$$

where γ determines the width of the Gaussian function.

EFFECT OF THE DOMINANCE MODIFIER

Dominance at the color pattern locus *P* was assumed to be controlled by a modifier locus *D* with a wild-type allele *m*, and a mutant allele *M*. Ten genotypes segregate in the model populations: *aamm*, *aaMM*, *aamM*, *bbmm*, *bbMM*, *bbmM*, *abmm*, *abMM*, *abmM*, and *abMm*. Because we aimed at investigating the fate of *cis*- and *trans*-acting mutations explicitly, we considered genotypes *abmM* and *abMm* as distinct (assuming that in genotype *abmM*, allele *a* is linked to allele *m* and allele *b* is linked to allele *M*) and therefore allowing them to display different phenotypes depending on the mode of action of the modifier *M*. For simplicity, we assumed that the mutant modifier allele *M* was dominant, so that both *mM* and *MM* genotypes had the same effect on dominance at the color pattern locus. The recombination rate between the color pattern locus *P* and the dominance modifier locus *D* was noted as ρ.

We assumed that the modification of dominance was due to a modification of expression of alleles at the color pattern locus *P*, either by enhanced or repressed expression, modifying the involved phenotypes by a fixed factor *e*. The phenotypes associated with the different combinations of gametes can be summarized as followed:

	am	bm	aM	bM
am	1	h	$1 + e_1$	$h + e_5$
bm		0	$h + e_4$	e_2
aM			$1 + e_1$	$h + e_3$
bM				e_2

with e_1 describing the effect of the modifier on homozygote *aa* (i.e., genotypes *aamM* or *aaMM*), e_2 on homozygote *bb* (i.e., genotypes *bbmM* or *bbMM*), e_3 on genotype *abMM*, and e_4 and e_5 on genotypes *abMm* and *abmM*, respectively.

The modification of expression could target either (1) specifically only one allele of the color pattern locus or (2) target any allele of the color pattern locus depending on their linkage. To encompass this diversity of modes of action for the dominance modifier, we considered three models (I, II, and II) with modifiers triggering different effects on the different genotypic combinations at the color pattern locus (see Table 1 for details). Model I: the modifier allele *M* is specific and targets the expression of one allele only, regardless of linkage between the modifier and the targeted allele (i.e., genotypes *abMm* and *abmM* express the same phenotype). Model II: the modifier allele *M* is nonspecific and can modify the expression of either allele *a* or *b* depending on the *cis*- or *trans*-mode of action of the modifier. Model III: the modifier allele *M* is specific but modifies the expression of the targeted allele depending on the *cis*- or *trans*-mode of action of the modifier. In models II and III, genotypes *abMm* and *abmM* display different phenotypes: a *cis*-acting modifier targeting the allele *a* modifies the expression of *a* in the genotypes *abMm* but not in the genotype *abmM*, whereas a *trans*-acting modifier modifies the expression of allele *a* in the genotype *abmM* and not in the genotype *abMm*.

In the case of a repressing effect of the modifier, certain genotypic combinations could exist where both alleles of the color pattern locus are repressed, such as homozygotes *aaMM* and a nonspecific *cis*-acting repression. In such cases, we considered that the phenotype would become either (1) strictly nonmimetic (and would suffer the same predation risk as any novel or exotic phenotype in the local patch) or (2) modified with factor $-e$, so that the phenotype of *aaMM* genotypes would vary from 1 to $1 - e$. In the numerical analyses, however, both hypotheses (1) and (2) resulted in similar outcomes, so only results based on hypothesis (2) are presented here. Such effect of repressors of expression on the phenotype of homozygotes represents the major difference with enhancers, and might lead to contrasted evolutionary outcomes.

PREDATION

Every individual of the focal (polymorphic) species suffered a predation risk modulated by its resemblance to the local mimetic

Table 1. Description of the effect of dominance modifiers on phenotypes for the 16 models implemented, which differed by the specificity of the modifier (allele-specific or nonspecific), its effect (enhancer or repressor), and the mode of action (*cis*- or *trans*-acting).

Model type	Model number	Specificity	Effect	Mode of action	e_1	e_2	e_3	e_4	e_5
I	I.1	*a*-Specific	Enhancer	Any	0	0	e	e	e
	I.2		Repressor	Any	$-e$	0	$-e$	$-e$	$-e$
	I.3	*b*-Specific	Enhancer	Any	0	0	$-e$	$-e$	$-e$
	I.4		Repressor	Any	0	e	e	e	e
II	II.1	*Nonspecific*	Enhancer	*cis*	0	0	0	e	$-e$
	II.2		Enhancer	*trans*	0	0	0	$-e$	$+e$
	II.3		Repressor	*cis*	$-e$	e	0	$-e$	e
	II.4		Repressor	*trans*	$-e$	e	0	e	$-e$
III	III.1	*a*-Specific	Enhancer	*cis*	0	0	e	e	0
	III.2		Enhancer	*trans*	0	0	e	0	e
	III.3		Repressor	*cis*	$-e$	0	$-e$	$-e$	0
	III.4		Repressor	*trans*	$-e$	0	$-e$	0	$-e$
	III.5	*b*-Specific	Enhancer	*cis*	0	0	$-e$	0	$-e$
	III.6		Enhancer	*trans*	0	0	$-e$	$-e$	0
	III.7		Repressor	*cis*	0	e	e	0	e
	III.8		Repressor	*trans*	0	e	e	e	0

For each model, the effect of the modifier on phenotypes is displayed, with e_1 describing the effect of the modifier on homozygote *aa* (i.e., genotypes *aamM* or *aaMM*), e_2 on homozygote *bb* (i.e., genotypes *bbmM* or *bbMM*), e_3 on genotype *abMM*, and e_4 and e_5 on genotypes *abMm* and *abmM*, respectively.

community of butterflies. We assumed a symmetrical condition where the mortality coefficient was $d\ (1 - \sigma)$ for phenotypes matching the local mimicry ring and $d\ (1 + \sigma)$ otherwise, where d represented the baseline predation risk and σ the spatial heterogeneity in the distribution of the two mimicry communities (see Fig. 1 for illustration): $+\sigma$ measures the additional risk of being attacked when individual are nonmimetic in the patch (i.e., black in patch 1 or light gray in patch 2 in Fig. 1) and $-\sigma$ measures protection gained by individuals displaying mimetic phenotype in the patch (i.e., light gray in patch 1 and black in patch 2 in Fig. 1).

Survival of a given phenotype depended on its match to the local mimicry environment, but also on its own abundance in the patch. Number-dependent predator avoidance in the focal species was therefore assumed to depend on its toxicity, l, and the density of each phenotype, using a function similar to the one used in the haploid model of Joron and Iwasa (2005).

The change in the number of each genotype i in population k due to predation N_{ik}^P was described by:

$$\frac{\mathrm{d}N_{ik}^P}{\mathrm{d}t} = -\frac{d\,(1+\sigma)\,(1-R_{imimic}) + d\,(1-\sigma)\,R_{imimic}}{1+1\left(\sum_{\mathrm{j}=1}^{\mathrm{j}=\mathrm{n}} R_{ij}N_{jk}\right)} N_{ik}, \quad (3)$$

with N_{ik} representing the total number of individuals with genotype i in population k, R_{imimic} representing the resemblance of the phenotype expressed by genotype i to the local mimetic community. This resemblance is equal, in population 1, to the resemblance of the genotype i to the wild-type homozygote *aamm* (displaying phenotype A), and to homozygote *bbmm* in population 2 (displaying phenotype B).

MIGRATION

The change in number of each genotype i in population k due to migration N_{ik}^M between populations k and c was given by:

$$\frac{\mathrm{d}N_{ik}^M}{\mathrm{d}t} = mig\ (N_{ic} - N_{ik}), \quad (4)$$

with $k \neq c$ and where mig is the migration coefficient.

REPRODUCTION

We assumed a balanced sex ratio and that reproduction was performed by half of the population (i.e., by females). We assumed density-dependent reproduction and with a per capita growth rate r and a carrying capacity K in each population.

Because we aimed to model sexual reproduction explicitly, the variations of the number of individuals carrying a given genotype in the populations depended on the frequencies of each genotype, referred to as f_{ik} in the following equation (5). We assumed Mendelian segregation at both the color pattern locus and the modifier locus, recombining at rate ρ. The change in the number of genotype i in population k due to reproduction N_{ik}^R was then described as follows:

$$\frac{\mathrm{d}N_{ik}^R}{\mathrm{d}t} = \frac{r}{2}\left(1 - \frac{N_k}{K}\right) f_{ik}, \quad (5)$$

with N_k representing the total number of individuals in population k.

The frequency of genotype i in population k (referred as f_{ik}) was computed by considering the frequencies of each genotype from the population and taking into account recombination between the modifier locus and the color pattern locus (see details in Supporting Information).

NUMERICAL ANALYSIS

Overall, the change in the density of genotype i in population k is given by:

$$\frac{\mathrm{d}N_{ik}}{\mathrm{d}t} = \frac{\mathrm{d}N_{ik}^{P}}{\mathrm{d}t} + \frac{\mathrm{d}N_{ik}^{M}}{\mathrm{d}t} + \frac{\mathrm{d}N_{ik}^{P}}{\mathrm{d}t}. \quad (6)$$

The dynamical system described by equation (6) above was too complex to be analyzed explicitly: we found no tractable equilibrium of the systems and thus we were unable to perform a classical stability analysis. The only tractable equilibria were obtained in cases without migration or in infinite migration rate (see Llaurens et al. 2013 for more details); however, in both cases, polymorphism at the color pattern locus was not maintained, preventing any evolution of dominance. We thus performed a numerical analysis focusing in parameter range where polymorphism was maintained. We introduced a mutant allele M at the modifier locus D at low frequency, and then tracked its frequency using deterministic simulations following the procedure described in Nuismer and Otto (2005). Note that in our model, all events can occur simultaneously.

INITIAL STATE WITHOUT MODIFIER

To set the initial conditions before the introduction of mutant M, each model was first run for 10,000 steps (i.e., burn-in period insuring reaching stable equilibrium), assuming exact codominance between the two alleles a and b at the color pattern locus P ($h = 0.5$) and using default parameter values (described in Table 2).

INTRODUCTION OF THE MODIFIER

After computing equilibrium frequencies at the color pattern locus, we introduced the mutant allele M at an initial frequency of order $1/K$ (with *abmm* individuals replaced by *abMM* individuals in both populations), with K the carrying capacity of the population. This ensured that initial conditions were symmetrical regarding habitat heterogeneity (the modifier allele is initially present in both patches and at the same frequency) and that mutant allele M was introduced in linkage equilibrium with both alleles a and b of the color pattern locus. Introducing mutant M in heterozygous genotypes *abmM* or *abMm* did not change the outcome of simulations, suggesting that the results of our model are robust to initial conditions regarding how the mutant is introduced.

Table 2. Summary of parameters used in simulations with their default values.

Parameter names	Default values
Dominance coefficient associated with the wild-type allele m	$h = 0.5$
Mutation size	$e = 0.5$
Toxicity	$l = 0.0025$
Migration rate	$mig = 0.2$
General predation risk	$d = 0.5$
Spatial heterogeneity of mimetic communities	$\sigma = 0.9$
Width of predators' generalization function	$\gamma = 0.01$
Growth rate	$r = 2$
Carrying capacity of each population	$K = 1000$
Recombination rate	$\rho = 0.5$

ESTIMATION OF SELECTION COEFFICIENT

To quantify selection acting on the dominance modifier, we computed the coefficient of selection s associated with the mutant allele M during the first 100 time steps after introduction of the mutant. We assumed the default parameters values (Table 2), except for migration rate (mig) and toxicity (l) where we explored a parameter range, that is, $mig \in [0;\ 0.2]$ and $l \in [0;\ 0.0025]$, which corresponded to the range where polymorphism in color pattern could be maintained in a previous study (Llaurens et al. 2013). We assumed that $\frac{\mathrm{d}f_M}{\mathrm{d}t} = s f_M(1 - f_M)$ (eq. 7), with s as the selection coefficient and f_M representing the frequency of the modifier across both populations. We thus estimated the coefficient of selection as the slope $s = \frac{\frac{\mathrm{d}f_M}{\mathrm{d}t}}{f_M(1-f_M)}$ by extracting the $f_M(t)$ values in the time interval $t \in [0; 100]$ from our simulations.

COMPUTATION OF EQUILIBRIUM FREQUENCIES

Simulations were run for 100,000 time steps, and genotype frequencies were recorded at the end of the iterations. Linkage disequilibrium LD between the color pattern locus P and the modifier locus D was computed using the equilibrium frequencies in overall both populations. $LD = f_{aM} - f_a f_M$ with f_{aM} being the frequency of chromosomes carrying both alleles a and M, f_a and f_M being the frequency of allele a and allele M, respectively. Computations assumed the default parameters resulting in strict symmetry of local selection favoring allele a in population 1 and allele b in population 2. We used a high spatial heterogeneity together with intermediate migration rate, which are suitable conditions for the persistence of balanced polymorphism, as explored in a previous model without dominance modifier (Llaurens et al. 2013).

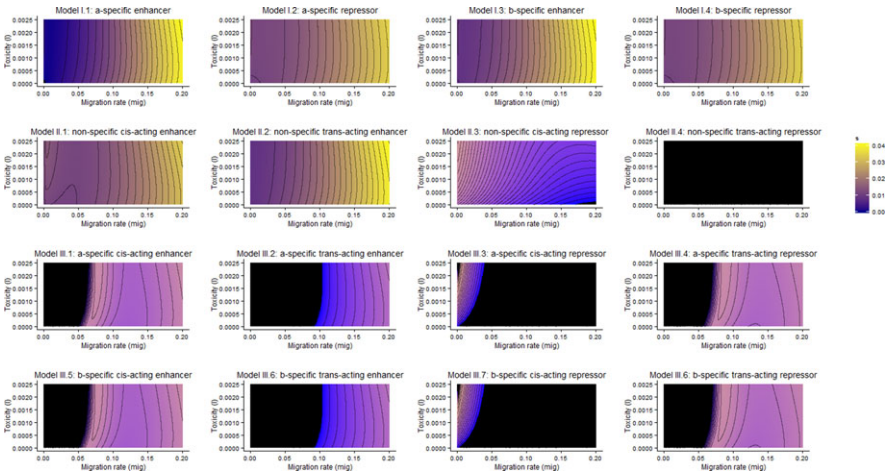

Figure 2. **Effect of migration rate (*mig*) and toxicity (*l*) on the coefficient of selection (*s*) associated with the dominance modifier *M* in the 16 different models. Black areas represent counterselection acting on the modifier ($s < 0$), and colored areas represent positive values of the coefficient of selection, that is, parameter ranges where dominance is likely to evolve. Simulations were run for 100 generations, for a range of migration rates $mig \in [0;\ 0.2]$ and a range of toxicity $l \in [0;\ 0.0025]$ and the default parameter values.**

Results

STRENGTH OF POSITIVE SELECTION ON DOMINANCE MODIFIERS

By estimating the coefficient of selection associated with the modifier *M*, we quantified the strength of selection acting on dominance modifiers within the parameter range where color pattern polymorphism is maintained, that is, for intermediate levels of migration and toxicity (Fig. 2). For models where modifiers were under positive selection (i.e., coefficient of selection $s > 0$), selection was generally higher when migration increased. Increased migration indeed leads to greater frequency of heterozygotes (Llaurens et al. 2013), enhancing the efficiency of positive selection acting on dominance. In few cases (model III.3 and III.7), lower levels of migration allows increased selection coefficient on the modifier *M*. This effect is due to opposite direction of selection acting on the modifier and targeted alleles in the two populations, so that those modifiers are under positive selection only when migration between populations is limited. Toxicity also influenced the strength of selection acting on the dominance modifier with slightly stronger positive selection for lower toxicity. As highlighted in Joron et al. (2005), when toxicity increases in the focal species, positive density-dependent selection within that species overcomes local adaptation (i.e., mimicry of alternative models in the two patches), leading to a decrease in the strength of balancing selection acting on color pattern polymorphism. Altogether, we detected positive selection on most dominance modifiers because they allow improving the fitness of heterozygotes: after the invasion of the modifiers, heterozygous phenotypes become mimetic in one of the two patches (Fig. 1). This positive selection observed in the specific case of polymorphic Müllerian mimicry studied here is in accordance with previous theoretical results (Otto and Bourguet 1999), showing that by generating a high frequency of heterozygotes at the locus targeted by the modifier, balancing selection favors the evolution of dominance.

IMPACT OF THE MODIFIER LOCUS ON POLYMORPHISM AT THE TARGETED LOCUS

In all models where dominance modifiers were maintained, frequencies at the color pattern locus *P* were altered (Table 3). Depending on their mode of action, modifiers could make the targeted *P*-allele either recessive or dominant, which influenced their equilibrium frequencies. Recessive alleles at the locus *P* increased in overall frequency because they benefited from mimicry in both populations. In the population where the recessive allele encodes for a nonmimetic allele, it can be protected from predators when occurring at heterozygote state because the alternative allele is mimetic. In contrast, color pattern alleles becoming dominant decreased in equilibrium frequency because any phenotype carrying the dominant allele is mimetic in only one out of the two populations. The dynamics of mimicry alleles at local and global scales might therefore be influenced by the dominance modifier locus.

DOMINANCE MECHANISMS

Three possible outcomes were observed for the modifier mutant (Table 3), stressing the importance of the molecular mechanism involved. (1) When the dominance modifier was a specific *a*- or *b*-enhancer acting in *cis*-, *trans*-, or both, then the mutant allele would go to fixation. (2) When the dominance modifier was a specific repressor acting in *cis*-, or a nonspecific repressor acting in *trans*-, then the mutant allele would go extinct. (3) In all other cases, polymorphism would persist at modifier locus *D*.

Table 3. Equilibrium frequencies at the color pattern locus *P* and the dominance modifier locus *D* in population 1, population 2, and over the two-patch system.

Model number	Specificity	Effect	Mode of action	Initial frequency of *a* Pop 1	Initial frequency of *a* Pop 2	Final frequency of *a* Pop 1	Final frequency of *a* Pop 2	Final frequency of *a* Average	Final frequency of *M* Pop 1	Final frequency of *M* Pop 2	Final frequency of *M* Average	Population size variation (%)
I.1	a-Specific	Enhancer	Any	0.78	0.22	0.53	0.17	0.35	1.00	1.00	1.00	+5.65
I.3	b-Specific	Enhancer	Any	0.78	0.22	0.83	0.47	0.65	1.00	1.00	1.00	+5.65
III.1	a-Specific	Enhancer	*cis*	0.78	0.22	0.53	0.17	0.35	1.00	1.00	1.00	+5.65
III.2	a-Specific	Enhancer	*trans*	0.78	0.22	0.53	0.17	0.35	1.00	1.00	1.00	+5.65
III.5	b-Specific	Enhancer	*cis*	0.78	0.22	0.83	0.47	0.65	1.00	1.00	1.00	+5.65
III.6	b-Specific	Enhancer	*trans*	0.78	0.22	0.83	0.47	0.65	1.00	1.00	1.00	+5.65
II.1	Nonspecific	Enhancer	*cis*	0.78	0.22	0.73	0.27	0.50	0.50	0.50	0.50	+1.86
II.2	Nonspecific	Enhancer	*trans*	0.78	0.22	0.73	0.27	0.50	0.50	0.50	0.50	+1.86
I.2	a-Specific	Repressor	Any	0.78	0.22	0.79	0.38	0.58	0.18	0.51	0.35	+1.60
I.4	b-Specific	Repressor	Any	0.78	0.22	0.62	0.21	0.42	0.51	0.18	0.35	+1.60
III.4	a-Specific	Repressor	*trans*	0.78	0.22	0.78	0.36	0.56	0.17	0.51	0.35	+0.98
III.8	b-Specific	Repressor	*trans*	0.78	0.22	0.64	0.22	0.44	0.51	0.17	0.35	+0.98
II.3	Nonspecific	Repressor	*cis*	0.78	0.22	0.78	0.22	0.50	0.00	0.00	0.00	0
II.4	Nonspecific	Repressor	*trans*	0.78	0.22	0.78	0.22	0.50	0.00	0.00	0.00	0
III.3	a-Specific	Repressor	*cis*	0.78	0.22	0.78	0.22	0.50	0.00	0.01	0.00	0
III.7	b-Specific	Repressor	*cis*	0.78	0.22	0.78	0.22	0.50	0.01	0.00	0.00	0

The variation in population size compares population size at the initial equilibrium (before the introduction of the mutant allele *M* at locus *D*) and at the final equilibrium in each model. Simulations were run for 100,000 generations, assuming the default parameter values (Table 2).

MODIFICATION OF EXPRESSION

All modifiers enhancing expression were under positive selection in the intermediate range of migration (Fig. 2). In contrast, repressor mutants were generally selected against because of the negative effect of making homozygotes nonmimetic (see Table 1). Molecular mechanisms leading to the repression of expression of mimetic phenotypes are thus less prone to allow adaptive dominance because of their negative impact on homozygote phenotypes. However, some modifiers described as enhancers, for example, enhancing allele *b* (I.3, III.5, and III.6) could be equally considered repressors acting against allele *a* in heterozygotes only (see Table 1). Such modifiers would become fixed, suggesting that both enhancers and repressors can be positively selected as long as the molecular mechanism involved does not negatively affect the phenotypes of homozygotes.

TARGETS SPECIFICITY

Positive selection acting on enhancers (Fig. 2) can either lead to their fixation or to their persistence at intermediate frequency depending on their affinity toward the different mimetic alleles (Table 3). Modifiers specifically targeting the expression of a single allele at locus *P* (either *a* or *b*) became fixed. In contrast, nonspecific modifiers enhancing the expression of any *P* allele remained polymorphic. By computing selection coefficient associated with allele *m* invading populations where allele *M* is fixed, we confirmed positive selection acting on both alleles and the persistence of polymorphism for nonspecific enhancers (see Fig. 2).

CIS- VERSUS *TRANS*-ACTION

Cis- and *trans*-acting repressors had different evolutionary fates (Fig. 2): *trans*-acting repressors persist at intermediate frequencies, whereas *cis*-acting were eliminated (Table 3). In model III.3, for instance, the *cis*-acting repression of the *a* allele leads to strong negative selection on the association between *a* and *M* in population 1 because of the nonmimetic homozygotes produced, and in population 2 because of the nonmimetic heterozygotes *abMm.* On the contrary, in model III.4, the specific *trans*-repression of the *a* allele leads to a negative selection of the association between *a* and *M* in population 1, because of the *aaMM* and *aamM* homozygotes, but positive selection in population 2 because of the mimicry of *abmM* genotypes (see Table 1 for details). Selection on modifiers in opposing direction in the two populations results in the persistence of polymorphism at the modifier due to spatial heterogeneity in mimicry.

INFLUENCE OF RECOMBINATION ON THE FREQUENCY OF THE MODIFIER

Positive selection acting on specific enhancers is so high that they would always become fixed as long as there was some recombination ($\rho > 0$; see Fig. 3). However, the equilibrium frequency of nonspecific enhancer tightly depended on recombination rate.

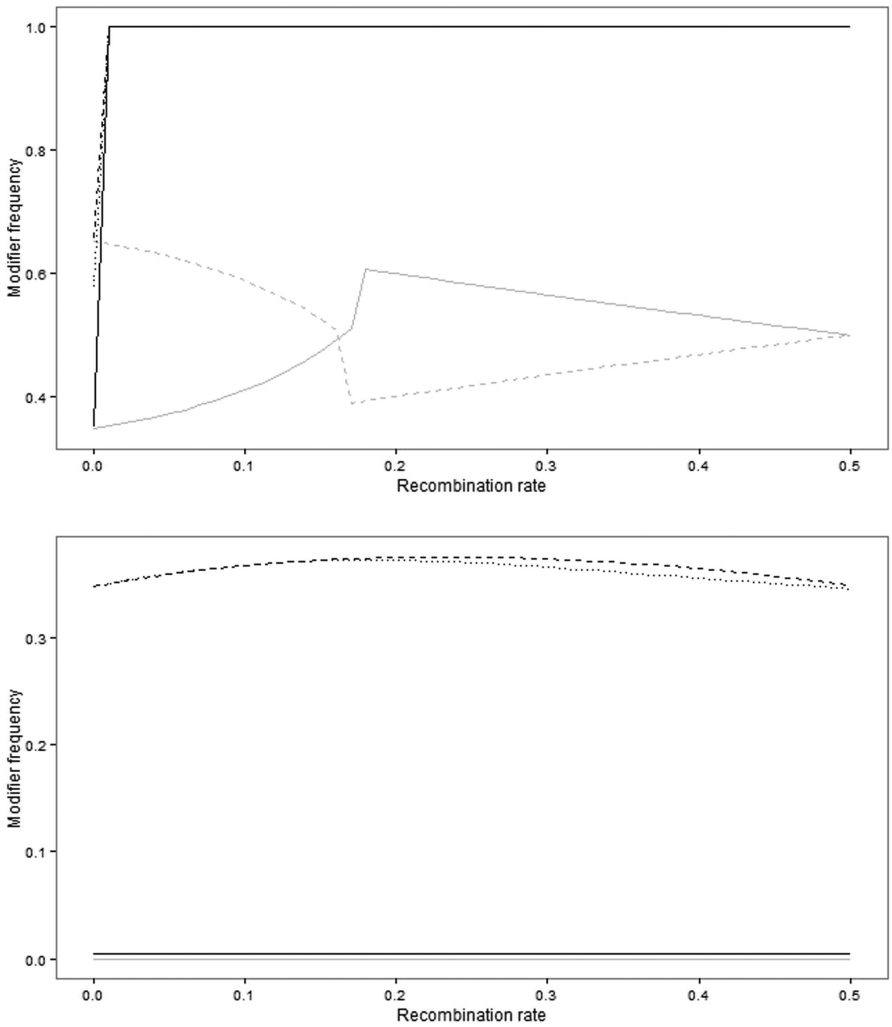

Figure 3. **Effect of recombination on the frequency of the dominance modifier in the two-patch system for enhancers (top graph) and repressors (bottom graph). Black lines are for specific modifiers targeting *a* (models I and III), gray lines for nonspecific modifiers (model II), dotted lines for specific modifiers acting irrespective of their association (models I.1 and I.2), dashed lines for *trans*-acting modifiers (models II.2, II.4, III.2, and III.4), and solid lines for *cis*-acting modifiers (models II.1, II.3, III.1, and III.3). Simulations were run for 100,000 generations, for a range of a recombination rates [0; 0.5] with an increment of 0.01 and using the default parameter values. Mutant allele *M* was introduced simultaneously in both populations, with 10 individuals carrying the genotype *abmm* switching to genotype *abMM* in each population.**

Nonspecific *cis*-enhancers reached high frequencies for low recombination rates and limited migration, as they could be in linkage disequilibrium with different alleles in the two populations (see Fig. 3 for more details on the interaction between recombination and migration). *Trans*-acting enhancer frequencies also depended on the recombination rate in a symmetrical manner because of the reduced frequency of the associated dominant alleles (see above). Contrastingly, recombination had only a marginal effect on the frequency of a *trans*-acting specific repressor whatever the migration rate assumed. For any other repressor type, dominance modifiers were not able to invade irrespective of the recombination rate (Fig. 3).

Overall, this suggests that the evolution of dominance can happen either through unlinked enhancers targeting the expression of one allele specifically or through linked enhancers targeting the expression of a nearby locus.

Discussion

EVOLUTION OF DOMINANCE MEDIATED BY MODIFIERS

Our model shows that genes underlying adaptive change in dominance at the color pattern locus are likely to be under strong positive selection when the color pattern locus is polymorphic. Because intermediate phenotypes are likely to be nonmimetic

and therefore suffer from reduced fitness, any mechanisms lowering their prevalence in an individual's progeny is likely to be promoted by selection. Dominance or recombination modifiers, but also assortative mating may indeed be promoted when traits are under disruptive selection (Rueffler et al. 2006). Previous general models describing dominance modifiers have indeed demonstrated that dominance can respond to natural selection at loci under balancing selection (Otto and Bourguet 1999; Peischl and Burger 2008; Yanchukov and Proulx 2014). The efficiency of natural selection on a dominance modifier increases with the frequency of heterozygotes, and so is high for polymorphic loci (Bourguet 1999; Bagheri 2006). Modifiers of dominance have been described for Müllerian mimicry loci in the butterfly *Heliconius cydno*, where locus *J* modified the relative expression of alleles in heterozygotes of the locus *Sb* (Naisbit et al. 2003). Such epistasis between genes could provide a molecular basis for dominance to evolve. Changes in the regulatory regions of genes might also contribute to the *cis*-regulation of expression and therefore modify dominance. *Cis*-regulatory changes seem widespread among species (e.g., among *Drosophila* species, see Wittkopp et al. 2008) and might play a substantial adaptive role. For instance, in several genes responsible for switching between mimetic wing patterns in *Heliconius*, the functional mutations controlling phenotypic variation are located outside the coding region (e.g., for the optix gene, see Reed et al. 2011). These opportunities for the shaping of dominance by selection suggest that as soon as heterozygotes are frequent, dominance is likely to evolve. By focusing on mimicry polymorphism where the direction of selection may be easily inferred, our model highlights (1) the positive selection acting on dominance modifiers and (2) the contrasted evolutionary fates among the different molecular pathway driving dominance variations.

MECHANISMS OF DOMINANCE MODIFICATION

By investigating different modes of action of dominance modifiers, our model shows that the evolution of dominance is more likely to happen via enhancers. This is due to our assumption that repressors caused nonmimetic phenotypes in homozygotes and had therefore a negative impact on their fitness. Repressors having an impact on the phenotypes of heterozygotes only are more prone to invade. The specificity and mode of action of modifiers is also important, in particular *trans*-acting specific repressors being more prone to be positively selected than *cis*-acting ones. This is in accordance with the recent discovery of dominance modifiers at the self-incompatibility locus in *Brassica* where recessive alleles are repressed by specific *trans*-acting siRNA linked to dominant alleles (Tarutani et al. 2010; Durand et al. 2014).

Although the modification of dominance described in the model could derive from many different mechanisms and at different steps during development, one might hypothesize that specific dominance modification could rely on allele-specific transcription factors. In contrast, nonspecific modifications could derive from variations in the promoting region, leading to variation of expression independently from the alleles themselves. In butterflies and many other animals with complex colorations, color pattern development may involve diffusion–reaction signals encoded by morphogens (Kondo and Miura 2010). Gilchrist and Nijhout (2001) showed that dominance at these morphogens could be an emergent property of the developmental pathway parameters. Indeed changes in the level of expression of morphogens, diffusion rates, or sensitivity threshold of receptors produce drastic change in dominance. In such genes, dominance could thus be modified through selection on other loci, for example, receptors of the signaling molecule, controlling the sensitivity threshold. Our model shows that in case of adaptive polymorphism, such selection on dominance is strong and might target different genes in the wing color pattern developmental pathway.

EVOLUTION OF DOMINANCE IN SPATIALLY HETEROGENEOUS ENVIRONMENT

The model presented here reveals the major role played by the level of linkage between the dominance modifier locus and the targeted locus in the evolution of dominance in structured populations. In the butterfly *H. numata* where polymorphism is maintained by a selection–migration balance, dominance among alleles of the wing color pattern supergene appears to have evolved (Le Poul et al. 2014). In *Heliconius*, dominance is generally thought to be determined by a hierarchy in the expression of the different colors throughout the wing (Gilbert et al. 1988). However, an ancestral and a more derived class of allele, differing in the presence of segmental chromosomal inversions, coexist in polymorphic populations of *H. numata*, and their heterozygotes do not show this general rule of dominance based on color hierarchy. Instead, ancestral alleles, lacking the inversions, are fully recessive to alleles carrying the inversions, independently of their colors. This strict dominance seems to be tightly associated with the inversions, suggesting a tightly linked control of dominance, preventing to conclude whether dominant alleles were simply captured by the inversion or dominance increased in response to selection through the effect of linked modifier. However, different alleles carried by the inverted gene order, displaying different phenotypes seemed all dominant over the ancestral alleles, suggesting a nonspecific control of dominance independent from the encoded phenotype. In accordance with our theoretical predictions, modification of dominance by nonspecific linked modifier could then explain the strict dominance of derived alleles over the ancestral ones in this case of balanced polymorphism maintained by the spatially heterogeneous selection. In contrast, in polymorphic populations of the African swallowtail *P. dardanus,* variation of dominance is observed at larger geographical scale and seems to rely on

differences in the genetic background (Nijhout 2003), suggesting the existence of unlinked modifier specifically targeting local alleles.

Here, we focused on a particular case of spatial heterogeneity due to variation of mimetic communities and showed that the requested level of linkage between the modifier and the targeted locus for dominance to evolve depends on the spatial structure of populations. Müllerian mimicry is a text-book example of positive selection acting at local scale, however, cases of polymorphism kept by selection–migration equilibrium can often be encountered in species living in spatially heterogeneous environments. The predictions of this model allow the identifications of the necessary conditions of evolution of dominance modifiers for adaptive polymorphism driven by heterogeneous environments.

Conclusions

By focusing on polymorphisms driven by selection–migration balance, this model demonstrates that the level of linkage between modifier and targeted locus will depend both on the mechanism responsible for dominance modification and on the amount of gene flow among heterogeneous environments. This highlights the need to investigate the spatial variation at both the polymorphic locus and the genomic background to identify potential dominance modifiers.

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Supporting Information

Additional Supporting Information may be found in the online version of this article at the publisher's website:

Figure S1. Frequency of heterozygotes at the color pattern locus *P* (red line) and of the modifier *M* (dotted blue line) for the generations following the interdiction of the mutant.

Figure S2. Effect of migration rate (*mig*) and toxicity (*l*) on the coefficient of selection (*s*) associated with the wild-type allele *m* in the 16 different models.

Figure S3. Effect of recombination on the frequency of *cis*-acting nonspecific enhancer and its linkage disequilibrium with the supergene controlling wing color pattern, based on 100 simulations per conditions.