A Century of Evolution: Ernst Mayr (1904–2005)

Ernst Mayr and the integration of geographic and ecological factors in speciation

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Mayr's best recognized scientific contributions include the biological species concept and the theory of geographic speciation. In the latter, reproductive isolation evolves as an incidental by-product of genetic divergence between allopatric populations. Mayr noted that divergent natural selection could accelerate speciation, but also argued that gene flow so strongly retards divergence that, even with selection, non-allopatric speciation is unlikely. However, current theory and data demonstrate that substantial divergence, and even speciation, in the face of gene flow is possible. Here, I attempt to connect some opposing views about speciation by integrating Mayr's ideas about the roles of ecology and geography in speciation with current data and theory. My central premise is that the speciation process (i.e. divergence) is often continuous, and that the opposing processes of selection and gene flow interact to determine the degree of divergence (i.e. the degree of progress towards the completion of speciation). I first establish that, in the absence of gene flow, divergent selection often promotes speciation. I then discuss how population differentiation in the face of gene flow is common when divergent selection occurs. However, such population differentiation does not always lead to the evolution of discontinuities, strong reproductive isolation, and thus speciation *per se*. I therefore explore the genetic and ecological circumstances that facilitate speciation in the face of gene flow. For example, particular genetic architectures or ecological niches may tip the balance between selection and gene flow strongly in favour of selection. The circumstances allowing selection to overcome gene flow to the extent that a discontinuity develops, and how often these circumstances occur, are major remaining questions in speciation research. © 2008 The Linnean Society of London, *Biological Journal of the Linnean Society*, 2008, **95**, 26–46.

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INTRODUCTION

Ernst Mayr made numerous contributions to speciation research from conceptual, systematic, genetic, and ecological perspectives. His greatest conceptual influence comes from the popularization of the biological species concept (BSC), which considers species as groups of actually or potentially interbreeding natural populations that are reproductively isolated from other such groups (Mayr, 1942). Elements of the BSC had been noted before Mayr (Wallace, 1864; Dobzhansky, 1935, 1937; Haffer, 2006), but Mayr

spread and crystallized the idea that the evolution of reproductive isolation is an essential component of speciation. Another well-recognized contribution is the theory of geographic speciation, whereby genetic divergence between geographically-separated (allopatric) populations results in speciation. Under this scenario, gene flow between populations prevents the evolution of reproductive isolation, most speciation is allopatric, and speciation in the face of gene flow is considered rare, if it occurs at all. Although this is probably still the dominant view among biologists, current theory and data demonstrate that substantial divergence, and even speciation, in the face of gene flow is possible (Turelli, Barton & Coyne, 2001; Via,

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2001; Berlocher & Feder, 2002; Drès & Mallet, 2002; Kirkpatrick & Ravigné, 2002; Bush & Butlin, 2004; Coyne & Orr, 2004; Dieckmann *et al.*, 2004; Hey, 2006; Nosil, 2008).

A thorough discussion of the above contributions by Mayr, and his theory of founder effect speciation, can be found in Coyne (1994). I instead focus my attention on Mayr's ideas concerning ecological factors in speciation (Mayr, 1942, 1947, 1963). Although Mayr's contributions to this area are often overshadowed by those outlined above, he did discuss the role of ecological factors in speciation. However, Mayr generally considered ecological factors to play a secondary role to geographic ones (Mayr, 1947, 1963). Here, I integrate ideas about ecological and geographic modes of speciation in an attempt to unify some opposing views about the mechanisms of speciation. I restrict my attention to 'ecological speciation', where divergent selection arises due to differences between environments, and results in the evolution of reproductive isolation (Schluter, 2000; Rundle & Nosil, 2005; reinforcement is reviewed elsewhere, Butlin, 1995; Servedio & Noor, 2003). I stress that ecological speciation can occur under any geographic scenario, including strict allopatry (Gavrilets, 2004; Rundle & Nosil, 2005).

The central premise underlying my arguments is that divergence during the speciation process is often continuous (even if the end point of this continuous process is the development of a discontinuity, such as strong bimodality in morphological traits or genetic markers). Although there certainly are exceptions, (e.g. polyploidy speciation; Ramsey & Schemske, 1998; Coyne & Orr, 2004), speciation may often be a continuous process. For example, the magnitude of reproductive isolation can vary from none, to weak, to intermediate, to strong, to complete (Coyne & Orr, 1989, 2004; Ramsey, Bradshaw & Schemske, 2003; Morjan & Rieseberg, 2004; Nosil, Vines & Funk, 2005; Funk, Nosil & Etges, 2006). Likewise, the degree of genotypic clustering can vary quantitatively (Mallet, 1995; Jiggins & Mallet, 2001), as can the extent of lineage sorting in gene genealogies (Beltrán *et al.*, 2002; Funk & Omland, 2003; Dopman *et al.*, 2005; Geraldes, Ferrand & Nachman, 2006; Putnam, Scriber & Andolfatto, 2007). These different degrees of divergence can be thought of as arbitrary 'stages' of the continuous process of speciation, with greater divergence equating to greater progress towards the completion of speciation (Wu, 2001; Drès & Mallet, 2002; Hey *et al.*, 2003; De Queiroz, 2005; Rueffler *et al.*, 2006; Gray & McKinnon, 2007; Mallet *et al.*, 2007; Svensson *et al.*, 2008, Mallet, 2008, this volume). This argument about the continuous nature of divergence can be applied to many species concepts (e.g. biological, genotypic, and phylogenetic, respectively, in the examples above), and it holds irrespec-

tive of the degree of divergence that different workers decide is necessary for speciation to be complete. In general though, speciation is considered complete when reproductive isolation is strong or near complete, or when genotypic clusters become largely non-overlapping (i.e. a discontinuity has developed).

The present paper is broken down into three main sections. Within each section, I highlight particularly relevant theoretical and empirical studies. However, the research of collaborators and myself on speciation in *Timema* walking-stick insects is used throughout to help bridge the three sections. First, I consider the evolution of reproductive isolation between allopatric pairs of populations. In this section, there is no variation in geographic scenario, and speciation in the absence of gene flow is the main focus. With respect to the role of ecology, the question is whether divergent natural selection promotes the process of allopatric speciation? Second, I introduce gene flow. I discuss trait divergence under a balance between selection and gene flow. It is clear that, when selection is strong, or gene flow is moderate or weak, divergence in the face of gene flow can occur. However, trait divergence does not always equate to speciation (i.e. the evolution of a discontinuity). In the third and final section, I consider the genetic and ecological circumstances that facilitate speciation in the face of gene flow.

The main conclusion is that speciation, like ordinary phenotypic evolution within species, tends to reflect a balance between selection and gene flow. Although this balance may often be tipped in favour of gene flow when it comes to the evolution of discontinuities (i.e. speciation), there are scenarios that facilitate speciation in the face of gene flow. Allopatric and sympatric speciation lie at opposite extremes with respect to how much gene flow occurs during divergence (Gavrilets, 2004: 11–14; Bolnick & Fitzpatrick, 2007; Emerson, 2008, this volume). Although Mayr's ideas helped generate polarized views concerning the geographic mode and mechanisms of speciation, his ideas can also be used to connect disparate views of the speciation process.

NATURAL SELECTION AND BY-PRODUCT SPECIATION IN ALLOPATRY

General theory and explicit tests

'The . . . evolution of isolating mechanisms as a by-product of the steady genetic divergence is inevitable' (Mayr, 1963: 581)

'Many isolating mechanisms have ecological components. The ecological shifts in incipient species are bound to have an effect on their isolating mechanisms' (Mayr, 1963: 551)

The preceding quotations underlie the two major components of 'by-product' speciation via natural

selection (see also Muller, 1942). First, reproductive isolation will evolve as an incidental by-product of genetic divergence. Mayr argued that such genetic divergence occurs between allopatric populations and is eroded by gene flow. Second, ecological shifts that expose populations to divergent natural selection will accelerate genetic divergence, and thus the process of by-product speciation. Speciation is accelerated because ecological traits under divergent selection, or traits that are genetically-correlated with such traits, can incidentally affect reproductive isolation (Funk, 1998; Schluter, 2000; Rundle & Nosil, 2005).

Soon after its conception, the theory of by-product speciation, with or without selection, was generally well-accepted. New species were thought to arise simply as a by-product of changes that occurred in allopatry, by selection, by random genetic drift, or by a combination of these factors. Although Mayr did discuss how selection would accelerate the process, explicit empirical investigations of the ecology of speciation during the time of the modern synthesis were few (but see Blair, 1950). Perhaps this was due, at least in part, to the emphasis on the importance of allopatry; given geographic isolation and enough time, speciation was seen as inevitable (Mallet, 2008, this volume).

The 1990s saw renewed interest in the role of ecology in speciation. The term 'ecological speciation' became widely-used to describe the process of speciation via divergent, ecologically-based natural selection (Schluter & Nagel, 1995; Schluter, 1996; Schluter, 1998, 2001; Rundle & Nosil, 2005; Nosil & Rundle, 2008). Explicit predictions of ecological speciation were made. For example, ecologically-divergent pairs of populations were predicted to exhibit higher levels of reproductive isolation than ecologically-similar pairs of populations of similar age (Schluter & Nagel, 1995; Funk, 1998; Funk, Filchak & Feder, 2002). Another prediction is that traits under divergent selection often incidentally affect reproductive isolation (e.g. habitat or mate preference, hybrid fitness; Jiggins et al. 2001). These predictions apply across geographic scenarios. All else being equal, divergent selection is predicted to promote the evolution of reproductive isolation, as now shown empirically in some taxa (Funk, 1998; Via, 1999; Rundle et al., 2000; Jiggins et al., 2001; Bradshaw & Schemske, 2003; Rundle & Nosil, 2005). For example, greater reproductive isolation between ecologically-divergent versus ecologically-similar pairs of populations has been documented using allopatric populations that likely undergo little or no gene flow (Funk, 1998; Nosil, Crespi & Sandoval, 2002; Vines & Schluter, 2006). These results show how ecological divergence can promote allopatric speciation.

A systematic evaluation of whether ecology plays a general role in driving the evolution of reproductive isolation was conducted by Funk et al. (2006). The authors used an approach that adds an ecological dimension to comparative studies investigating the relationship between reproductive isolation and divergence time (Coyne & Orr, 1989). Specifically, Funk et al. (2006) quantified ecological divergence for > 500 species pairs from eight plant, invertebrate, and vertebrate taxa and statistically isolated the association of ecological divergence with reproductive isolation (i.e. independent from time). They found a significant and positive association between ecological divergence and reproductive isolation across the eight taxa (Fig. 1). The findings are consistent with the hypothesis that ecological adaptation plays a widespread role in promoting speciation. A remaining question is what types of reproductive barriers are particularly likely to be involved.

CASE STUDIES OF ECOLOGICAL SPECIATION: HOST-ASSOCIATED *TIMEMA* WALKING-STICK INSECTS

A number of systems have been developed to examine ecological speciation over recent years, including *Gasterosteus* sticklebacks (Schluter, 1996; Rundle et al., 2000), *Heliconius* butterflies (Jiggins et al., 2001), *Rhagoletis* flies (Feder et al., 1994), *Littorina* snails (Rolan-Alvarez et al., 1999), *Acyrthosiphon* pea aphids (Via, 1999; Hawthorne & Via, 2001), *Neochlamisus* leaf beetles (Funk, 1998; Egan, Nosil & Funk, 2008), *Coregonus* whitefish (Lu & Bernatchez, 1999; Campbell & Bernatchez, 2004), *Gambusia* fishes (Langerhans, Gifford & Joseph, 2007), *Mimulus* monkeyflowers (Macnair & Christie, 1983; Bradshaw & Schemske, 2003; Ramsey et al., 2003), *Loxia* crossbills (Smith & Benkman, 2007), and *Pundamilia* cichlids (Maan et al., 2006; Seehausen, 2008), amongst others (Rundle & Nosil, 2005). Here, I focus on the work of my collaborators and myself on speciation in *Timema* walking-stick insects (Nosil, 2007 for complete review). I will refer to these insects in subsequent sections, and so provide some background here.

Timema are wingless insects inhabiting the chaparral of Southwestern North America (Vickery, 1993; Crespi & Sandoval, 2000). Individuals feed and mate exclusively on the host plants upon which they live. Most work to date has focused on *Timema cristinae*, which uses two host plant species (*Ceanothus spinosus*: Rhamnaceae and *Adenostoma fasciculatum*: Rosaceae). *T. cristinae* is composed of two ecotypes (the '*Ceanothus* ecotype' and the '*Adenostoma* ecotype'), which are defined by the host species they are found upon (Fig. 2; for details, see Nosil, 2007). A 'population' is all of the walking-sticks col-
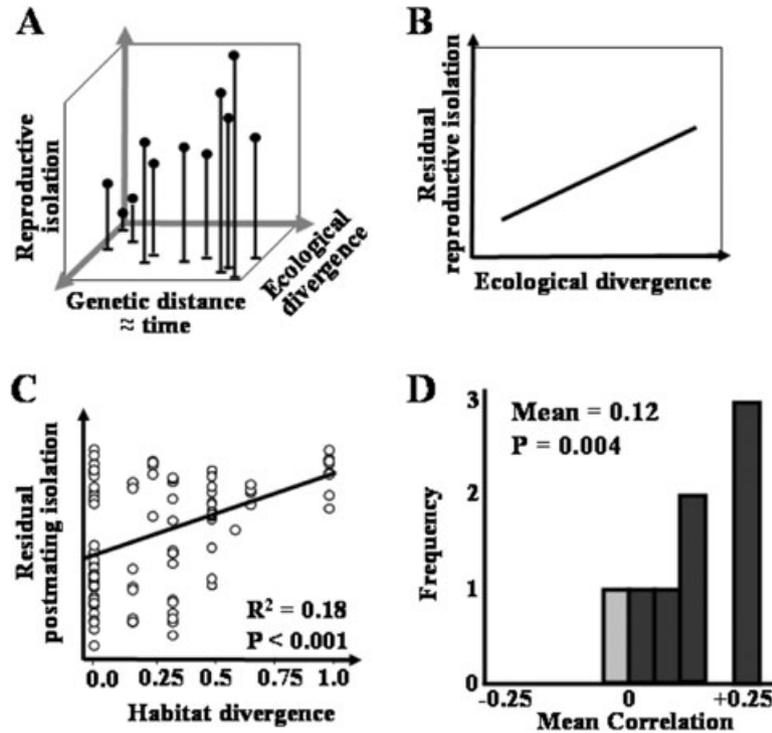
Figure 1. Isolating the relationship between ecological divergence and reproductive isolation, independent from time (see also Funk *et al.*, 2002). A, hypothetical scenario where reproductive isolation increases with both genetic distance (a proxy for time) and ecological divergence. B, hypothetical best-fit line illustrating the predicted positive association between ecological divergence and residual reproductive isolation upon statistical removal of the contributions of time. C, individual analysis showing the predicted pattern, in this case a positive association between habitat divergence and postmating isolation between angiosperm taxa. D, distribution of correlation coefficients from analyses of residual reproductive isolation (effects of time removed via regression of reproductive isolation and genetic distance) against ecological divergence across the eight taxa examined by Funk *et al.* (2006). Each data point is the mean correlation coefficient from a particular taxon. The mean of the distribution was significantly greater than zero using a one-sample *t*-test. Modified from Funk *et al.* (2006) and reprinted with permission of the National Academy of Sciences of the United States of America.

lected within a homogenous patch of a single host species (Nosil *et al.*, 2002; Nosil, Crespi & Sandoval, 2003; Nosil, 2007); many such patches exist in a geographic mosaic such that multiple populations on a given host comprise each ecotype. Patches of the two host species used by *T. cristinae* are often distributed in adjacent patches of varying size that are in geographic contact with one another. Insect populations associated with such patches are referred to as 'parapatric' (Nosil *et al.*, 2003). Other host patches are separated from all other patches of the alternative host by distances > 50 times the *Timema* 12 m per generation gene flow distance (Sandoval, 2000). Insect populations in such geographically-separated patches are termed 'allopatric'. Molecular and morphological data indicate that such allopatric populations incur little or no gene flow from populations on the alternative

host, and certainly incur less such gene flow than parapatric populations (Sandoval, 1994a; Bolnick & Nosil, 2007; Nosil, 2007).

The *T. cristinae* ecotypes are divergently adapted to their different hosts, at least when it comes to morphological traits. For example, they differ in a suite of adaptive morphological traits and population divergence in these traits has evolved via host-specific selection for crypsis from visual predators (Sandoval, 1994a, b; Nosil *et al.*, 2002; Nosil, 2004; Nosil & Crespi, 2004, 2006a). Pairs of populations on the same host are considered 'ecologically-similar' and pairs of populations on different hosts are considered 'ecologically divergent'. These two types of population pairs (referred to as same-host pairs and different-host pairs hereafter) can isolate a role for host-associated selection in divergence (Funk, 1998; Funk *et al.*, 2002; Funk & Nosil, 2007).
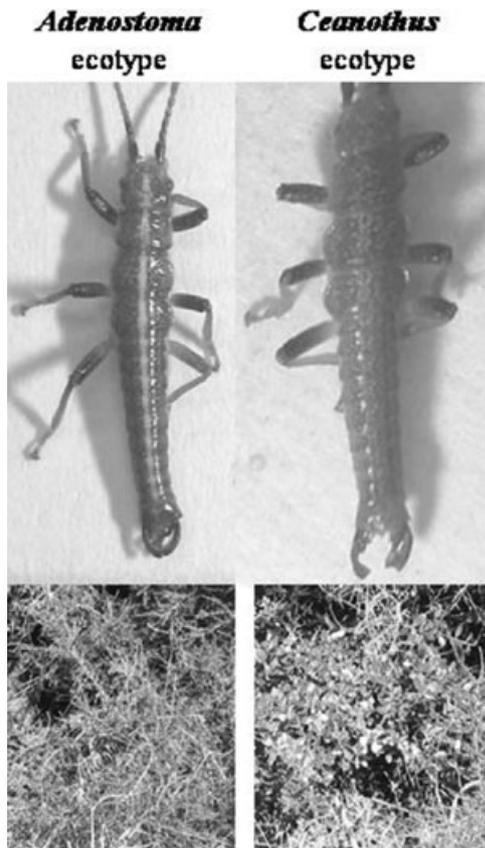
Figure 2. Host-plant ecotypes of *Timema cristinae* depicted above their respective hosts. The most conspicuous difference is the frequency of individuals with the presence versus absence of a dorsal stripe. Populations rarely show fixed differences, but the unstriped morph is more common on *Ceanothus* (mean frequency = 81%) and the striped morph is more common on *Adenostoma* (mean frequency = 72%). Thus most, but not all, individuals of the *Adenostoma* ecotype are striped. The ecotypes also differ in many other traits. For example, individuals from the *Ceanothus* ecotype tend to exhibit bright bodies, lack a dorsal stripe, and have relatively narrow heads with long legs. By contrast, individuals from the *Adenostoma* ecotype tend to have smaller and duller bodies, large and bright dorsal stripe patterns, and relatively wide heads with short legs. Modified from Nosil (2007) and reprinted with the permission of the American Society of Naturalists.

TYPES OF REPRODUCTIVE BARRIERS

For the vast majority of animals, it is still not known which particular isolating mechanisms prevent closely-related species from interbreeding (Mayr, 1963: 91–92)

Numerous types of reproductive barriers can isolate species. Mayr's observation regarding our ignorance of the types of barriers important for isolating recently-formed species is still relatively valid today (Turelli *et al.*, 2001; Coyne & Orr, 2004); but see

(Howard *et al.*, 1998; Ramsey *et al.*, 2003). We understood even less about which barriers are most important in ecological speciation specifically (Rundle & Nosil, 2005). Some barriers, such as habitat preference and ecological selection against hybrids, are inherently ecological. However, divergent selection might also affect barriers that are not inherently ecological. For example, natural selection might result in the fixation of alleles within populations that cause genetic incompatibilities in between-population hybrids (Gavrilets, 2004; Dettman *et al.*, 2007). This process would accelerate the evolution of intrinsic postmating isolation. The prediction that ecologically-divergent pairs of populations exhibit greater levels of reproductive isolation than ecologically-similar pairs of populations of similar age can be tested using any type of reproductive barrier, thereby allowing comparison of the role of ecology in the evolution of different barriers.

The ecotypes of *T. cristinae* discussed above were used to test which reproductive barriers are affected by divergent selection during allopatric divergence. The results indicate that multiple forms of reproductive isolation are stronger between allopatric different-host pairs than between allopatric same-host pairs, including habitat isolation (i.e. divergent host-plant preferences; Nosil & Crespi, 2006a,b), immigrant inviability (Nosil, 2004; Nosil *et al.*, 2005), sexual isolation (Nosil *et al.*, 2002, 2003, 2007), and cryptic postmating isolation (i.e. reduced survival and fecundity of females following between-population mating; Nosil & Crespi (2006b). Thus, ecological divergence promoted progress towards allopatric speciation and affected numerous reproductive barriers, including barriers that are not inherently ecological, such as sexual isolation and cryptic postmating isolation (Fig. 3; Nosil, 2007). Studies examining multiple barriers in other systems are required to test the generality of these findings from *Timema*.

GENERAL ACCEPTANCE OF ECOLOGICAL SPECIATION IN ALLOPATRY

'. . . the rate of speciation is apparently determined by ecological factors. When the range of a species is dissected by geographic and ecological barriers and there is very restricted gene flow in this species, speciation will be rapid and frequent' (Mayr, 2001: 186)

Interestingly, very little formal mathematical theory initially focused on how divergent selection could promote allopatric speciation (Turelli *et al.*, 2001; Kirkpatrick & Ravigné, 2002; Gavrilets, 2004), perhaps because the idea was so intuitive it was presumed to be correct. Instead, mathematical theory focused on the controversial theory of sympatric spe-
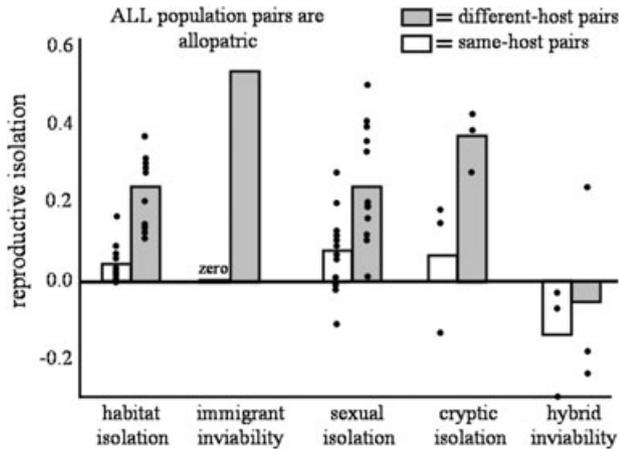
Figure 3. The effects of ecological divergence on the evolution of a diverse array of reproductive barriers between allopatric populations of *Timema cristinae* walking-sticks. Bars represent mean reproductive isolation and each black dot depicts results from an individual population pair. In general, different-host population pairs exhibit significantly greater reproductive isolation than do same-host pairs, across reproductive barriers ($P < 0.05$ in all cases except intrinsic hybrid inviability). Thus, ecological divergence promotes progress towards allopatric speciation. Modified from Nosil (2007) and reprinted with the permission of the American Society of Naturalists.

ciation, but different studies used different assumptions making it difficult to make comparisons among them (Turelli *et al.*, 2001; Bolnick & Fitzpatrick, 2007). However, in addition to the empirical studies outlined above, recent years have seen theoretical studies demonstrating that divergent selection can promote allopatric speciation (Gavrilets, 2004). Thus, the statement that divergent natural selection can sometimes, and to some extent, promote allopatric speciation is relatively uncontroversial. But what happens if some gene flow is introduced to the scenario of speciation via natural selection?

TRAIT DIVERGENCE UNDER A BALANCE BETWEEN SELECTION AND GENE FLOW

'It is very unlikely that the purely genetic processes of mutation pressure and random fixation cause changes of a sufficiently high order . . . to hold any sizable gene flow in check. It may be different with selection pressure . . .' (Mayr, 1947: 268)

Although Mayr notes that strong genetic divergence in the face of high gene flow is unlikely via drift alone, he implies that divergence with gene flow is possible if selection occurs. While avoiding explicit treatment of the subject, Mayr nonetheless acknowledges that population differentiation could reflect a

balance between the diversifying effects of divergent selection and the homogenizing effects of gene flow. Mayr's quotation is rather vague, implying that selection/gene flow balance was not well worked out at the time. However, studies explicitly tackling the issue predated his 1947 article (and more emerged before Mayr, 1963).

SELECTION/GENE FLOW BALANCE: THEORY

Divergence under a balance between selection and gene flow was considered for Mendelian traits during the 1930s and 40s (Fisher, 1930; Wright, 1931, 1940, 1943). For example, Haldane (1930, 1932) considered an island that receives immigrants at rate, m, from the mainland, where different alleles are favored on the island versus on the mainland. These two populations differ at a single biallelic locus. The frequency (p) of allele A is initially assumed to be close to 1.0 on the island. The frequency (q) of the alternative allele a is assumed to be fixed on the mainland. Haldane showed that when selection (s) acts against maladapted alleles (i.e. selection favors genotypes that are adapted to the island environment, thereby selecting against both immigrants aa and hybrids aA), the allele conferring adaptation to the island can be maintained provided $s > m$. The maintenance of the island allele was also possible if selection acted only against hybrids, but required stronger selection relative to migration. Wright (1931, 1940, 1943) considered similar scenarios and likewise found that selection can overcome gene flow to cause divergence (for further discussion of work on selection/gene flow balance by Haldane and Wright, see also Bulmer, 1972; Slatkin, 1981; Vines *et al.*, 2003; Nosil *et al.*, 2005; Mallet, 2006; Bolnick & Nosil, 2007).

The emergence of cline theory, starting with Haldane (1948) and Fisher (1950), showed how ecological selection and gene flow would balance in a spatial context. This initial cline theory was extended by others (Endler, 1973, 1977; Slatkin, 1973; May, Endler & McMurtie, 1975; Roughgarden, 1979), and was developed in detail by Barton and colleagues (who introduced heterozygote disadvantage, a form of postmating isolation; Barton, 1979, 1983; Barton & Hewitt, 1985, 1989). Over the 75 years following the seminal work on the topic, theory concerning selection/gene flow balance has been extended to quantitative traits and applied to a wide range of demographic scenarios (Felsenstein, 1976; Slatkin, 1985; Garcia-Ramos & Kirkpatrick, 1997; Hendry, Day & Taylor, 2001; Lenormand, 2002; Kawecki & Ebert, 2004). The general conclusion emerging from this theoretical work is that strong selection can overcome moderate gene flow to result in population divergence in trait means. Judging from the timing of

Table 1. Statistical analyses examining divergence under a balance between selection and gene flow in *Timema cristinae* walking-sticks

| | Differences between hosts (*t*-tests) | | | Selection/gene flow balance | | | |
| | | | | *Ceanothus* populations | | *Adenostoma* populations | |
Trait	*t*	d.f.	*P*	rho	*P*	rho	*P*
Body brightness	**3.59**	**24**	**0.001**	**0.51**	**0.032**	**0.71**	**0.005**
Stripe brightness	1.55	24	0.133	**0.50**	**0.034**	−0.03	0.465
% striped	**3.03**	**28**	**0.005**	**0.77**	**< 0.001**	**0.70**	**0.004**
Male size	**2.35**	**15**	**0.033**	**0.71**	**0.017**	**0.81**	**0.007**
Male shape	**2.65**	**15**	**0.018**	0.31	0.206	**0.81**	**0.007**
Female size	**2.27**	**15**	**0.039**	**0.71**	**0.014**	0.41	0.160
Female shape	**3.54**	**15**	**0.003**	−0.07	0.429	**0.88**	**0.002**
Host preference	**3.79**	**25**	**0.001**	**0.91**	**< 0.001**	0.31	0.175

Differences between hosts in each trait were examined using *t*-tests. Within each host, whether mean traits values were correlated with the proportion of the patch occupied by *Ceanothus* (i.e. an index of selection/gene flow balance) was tested using Spearman rank correlation. Significant results are shown in bold.
d.f., degrees of freedom.
For graphical results, see Fig. 4.

even the initial theoretical developments, contemporaries of Mayr were likely aware of this conclusion, as are current workers.
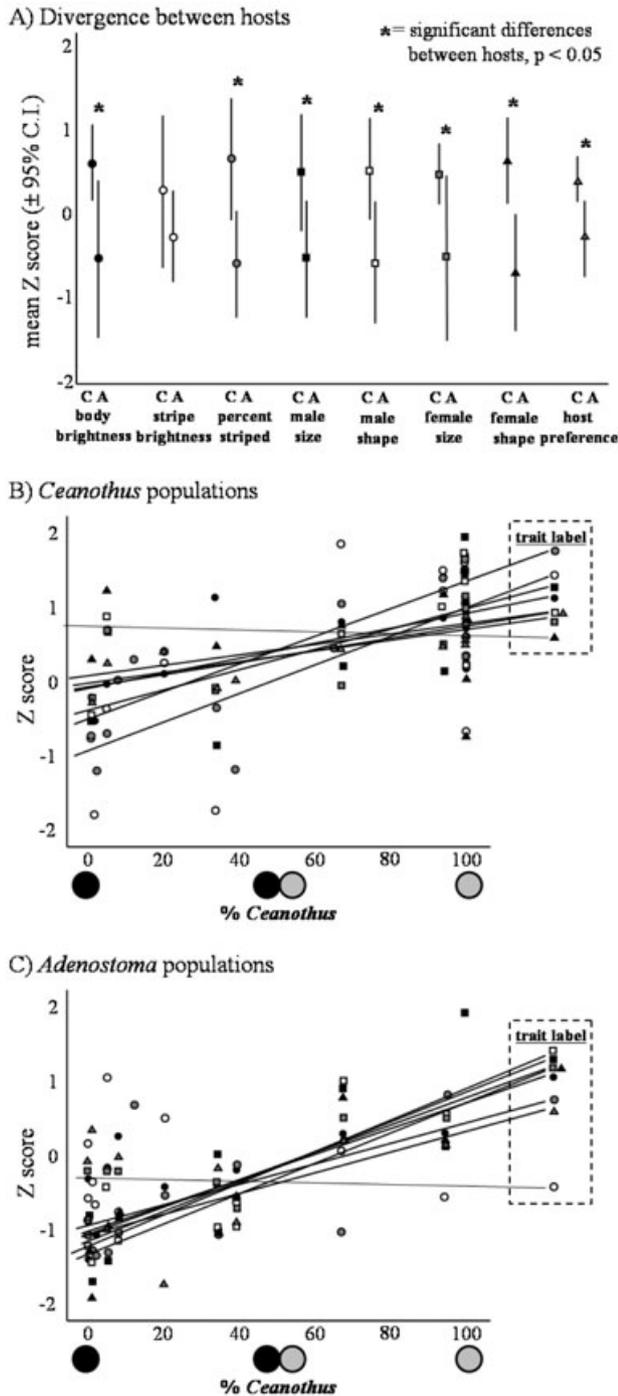
SELECTION/GENE FLOW BALANCE: EMPIRICAL EXAMPLES

A number of empirical studies have documented divergence under a balance between selection and gene flow, thereby providing general support for theory. A common form of evidence for a balance between selection and gene flow is an inverse association between levels of population divergence in adaptive phenotypic traits and levels of gene flow (fish: Lu & Bernatchez, 1999; Hendry, Taylor & McPhail, 2002; Moore, Taylor & Hendry, 2007, amphibians: Storfer & Sih, 1998, Storfer *et al.*, 1999, birds: Dhondt *et al.*, 1990; Smith *et al.*, 1997, reptiles: King & Lawson, 1995, insects: Sandoval, 1994a; Ross & Keller, 1995; Arachnids: Riechert, 1993; Riechert & Hall, 2000; Riechert, Singer & Jones, 2001; for a discussion of reverse causality, see Hendry & Taylor, 2004; Nosil & Crespi, 2004). Likewise, clines in genotype or phenotype frequency, for example in hybrid zone studies, also often represent divergence under a balance between selection and gene flow (Barton & Hewitt, 1985; Mallet *et al.*, 1990; Johannesson, Johannesson & Rolan-Alvarez, 1993).

Ecotypes of *T. cristinae* provide an example of divergence under a balance between selection and gene flow. Here, I present novel analyses of published data

to illustrate this balance. I consider how traits differ both between populations using different hosts (due to divergent selection) and among populations within hosts (due to variation in levels of gene flow). Multiple populations of each host were examined such that populations, rather than individuals, comprise the unit of replication in all statistical analyses. The eight traits considered are body brightness, stripe brightness, percent of individuals within a population exhibiting the absence of a striped pattern, male body size, male body shape, female body size, female body shape, and host preference. Body and 'stripe' brightness are quantitative measures of the brightness of the exterior and central parts of the body respectively, irrespective of the presence/absence of a stripe pattern itself (Nosil & Crespi, 2006a). The data on brightness stem from Nosil & Crespi (2006a), all size and shape data are from Nosil & Crespi (2004), frequency of striped individuals is from Nosil (2004), and host preference was estimated as the percent of individuals picking *Ceanothus* over *Adenostoma* in choice experiments from data presented in Nosil & Crespi (2006a). Each trait was standardized to mean zero and standard deviation one (*Z*-scores). All traits exhibited substantial differences in mean values between populations using different hosts, and *t*-test analyses demonstrate significant differences for seven of eight traits (Fig. 4, Table 1).

Whether trait values for individual populations reflect the effects of a balance between selection and gene flow was then assessed using a quantitative

A) Divergence between hosts

★= significant differences between hosts, p < 0.05

B) Ceanothus populations

C) Adenostoma populations

Figure 4. Divergence under a balance between selection and gene flow in *Timema cristinae* walking-sticks. A, divergence between hosts for eight traits, where populations are the unit of replication. Shown are mean (± 95% confidence interval) trait values for each host. Traits that differed significantly between populations on different hosts in *t*-tests are denoted by asterisks (C, *Ceanothus*; A, *Adenostoma*). B, C, for *Ceanothus* and *Adenostoma* populations respectively, the correlation between the proportion of a host patch occupied by *Ceanothus* and mean trait values. The trait that each line corresponds to is labeled within a box on the extreme right of the graph (black circles, body brightness; white circles, stripe brightness; grey circles, percent striped; black squares, male size; white squares, male shape; grey squares, female size; black triangles, female shape; grey triangles, host preference). In all panels, stripe brightness was converted to one minus stripe brightness before standardization to keep all means larger on *Ceanothus*. Data are from Nosil (2004), Nosil & Crespi (2004), Nosil & Crespi (2006a, b). For statistical analyses, see Table 1.

◄————————————————————

index of this balance. This index is simply the proportion of the total area of a site (area of the study population plus the area of the adjacent population using the alternate host) occupied by *Ceanothus*, and is referred to as the 'index of selection/gene flow' hereafter. Allopatric populations (which do not have an adjacent population on the alternative host) apparently incur little or no gene flow (Nosil *et al.*, 2003;

Bolnick & Nosil, 2007), and are assigned values of zero (for *Adenostoma* populations) or 100 (for *Ceanothus* populations). Parapatric populations are assigned values between zero and 100, based upon the relative abundance of *Ceanothus*. Previous work indicates that this index accurately estimates the geographic potential for gene flow as: (1) host patch size and *T. cristinae* population size are strongly, positively correlated (Sandoval, 1994a) and (2) for parapatric populations, the relative size of the population using the alternative host that is adjacent to a focal population is strongly correlated with the migration rate from the adjacent population into the focal population (Nosil *et al.*, 2003; Nosil & Crespi, 2004); migration estimated from mitochondrial DNA sequence data and the coalescent-based methods of (Beerli & Felsenstein, 2001). Nosil & Crespi (2004) provide a more detailed validation of this index.

As values of the index increase, populations are expected to be more '*Ceanothus*-like', irrespective of what actual host they are on (e.g. as index values increase, *Adenostoma* populations are incurring more alleles from *Ceanothus* via gene flow). Spearman rank correlation was used to test whether trait values were correlated with the index of selection/gene flow. *Ceanothus* populations exhibit greater mean values than *Adenostoma* populations for all traits (Fig. 4A). Thus, due to the a priori expectation of a positive correlation, I report one-tailed probabilities. To avoid conflating differences between hosts with differences among populations within hosts, analyses were run separately for populations using each host species (a conservative approach). On both hosts, positive

correlations between mean trait value and the index of gene flow were observed for seven of eight traits. Significantly positive correlations were observed for six of eight traits for *Ceanothus* populations and five of eight traits for *Adenostoma* populations. Even if two-tailed probabilities are used, many correlations remain significant (and pronounced positive associations are evident irrespective of significance testing; Fig. 4). Thus, variation in trait values among populations within hosts is reliably predicted by differences in levels of between-host gene flow. Collectively, trait values for multiple traits are largely explained by a balance between divergent selection (host plant use) and gene flow. Likewise, a recent study employing population genomic methods in a genome scan indicates that molecular genetic divergence also proceeds, in part, via a balance between selection and gene flow (Nosil, Egan & Funk, 2008). The results from *T. cristinae*, coupled with the theoretical and empirical work reviewed above, indicate that divergence under a balance between selection and gene flow may be a general feature of evolutionary diversification.

TRAIT DIVERGENCE DOES NOT ALWAYS EQUATE TO SPECIATION

'without an extrinsic reduction of gene flow, the ecological variability cannot become a primary source of discontinuity' (Mayr, 1947: 281)

There is considerable theoretical and empirical evidence for trait divergence under a balance between selection and gene flow. However, trait divergence does not always equate to speciation, under either a biological (Mayr, 1942, 1963) or clustering (Mallet, 1995) species concept. For example, populations may differ in mean trait values, yet exhibit only weak reproductive isolation and largely overlapping genotypic clusters. As noted in the Introduction, what is considered the 'completion' of speciation may differ among evolutionary biologists. Irrespective of this, the point is that the degree of divergence can vary, and the degree may often not be large enough to equate to the completion of speciation. (For further consideration of such 'intermediate' stages of divergence see Knapp, 2008; Mallet, 2008, this volume.) Was Mayr correct in his assertion that speciation in the face of gene flow is highly unlikely? Some insight stems from the ecotypes of *T. cristinae*, which exhibit partial, but incomplete, progress towards ecological speciation. Specifically, although partial reproductive isolation has evolved between the ecotypes, experimental, molecular, and morphological lines of evidence suggest that progress towards speciation is incomplete, perhaps due to the constraining effects of gene flow (Nosil, 2007).

The arguments for incomplete speciation between *T. cristinae* ecotypes focus on the observation that divergence tends to be weaker in parapatry than in allopatry, a classic signature of gene flow, and thus incomplete reproductive isolation, between parapatric populations (Coyne & Orr, 2004; Grant, Grant & Petren, 2005). First, experimental data from the ecotypes indicates that even for the most extreme cases of allopatric divergence, there is only an 80% barrier to gene flow (Fig. 5A). Total barriers to gene flow are somewhat weaker in parapatry. Second, molecular data support the claim of incomplete reproductive isolation: adjacent pairs of populations using different hosts exhibit consistently and significantly weaker levels of mitochondrial DNA differentiation than do geographically-separated populations (Fig. 5B; Nosil *et al.*, 2003). The pattern of weaker population divergence with increasing gene flow is also seen for morphological traits (Sandoval, 1994a; Nosil & Crespi, 2004; Bolnick & Nosil, 2007; present study, Fig. 4), and a genome scan using hundreds of amplified fragment length polymorphisms further supports gene flow between ecotypes (Nosil *et al.*, 2008). Third, despite differentiation in trait means, bimodality in morphological traits is weak in *T. cristinae* (Fig. 5C; Nosil, 2007). Moreover, any bimodality that does exist often collapses in parapatry, and many intermediate phenotypes are evident (Nosil, 2007). In general, such a lack of strong bimodality, and its collapse in non-allopatry, is evidence for incomplete speciation/reproductive isolation (Mallet, 1995; Jiggins & Mallet, 2001).

In summary, although population differentiation in trait means in the face of gene flow might be common, it is not clear how often the process of divergence with gene flow can lead to complete speciation. Mayr is likely correct that speciation in the face of high gene flow is difficult. However, compelling examples of substantial divergence, and even speciation, in the face of some gene flow do exist (Feder *et al.*, 1994; Via, 2001; Emelianov, Marec & Mallet, 2004; Coyne & Orr, 2004; Barluenga *et al.*, 2006; Hey, 2006; Savolainen *et al.*, 2006; Bolnick & Fitzpatrick, 2007; Niemiller, Fitzpatrick & Miller, 2008; Nosil, 2008), raising questions concerning the factors that facilitate speciation in the face of gene flow.

SPECIATION UNDER A BALANCE BETWEEN SELECTION AND GENE FLOW

'. . . the less gene flow there is between populations, the more rapidly speciation will occur, all else being equal' (Mayr, 2001: 186)

Mayr's quotation is likely true, but what about cases where 'all else is not equal'? Are there genetic

A) Experimental evidence

B) Molecular evidence (mtDNA)

C) Morphological evidence
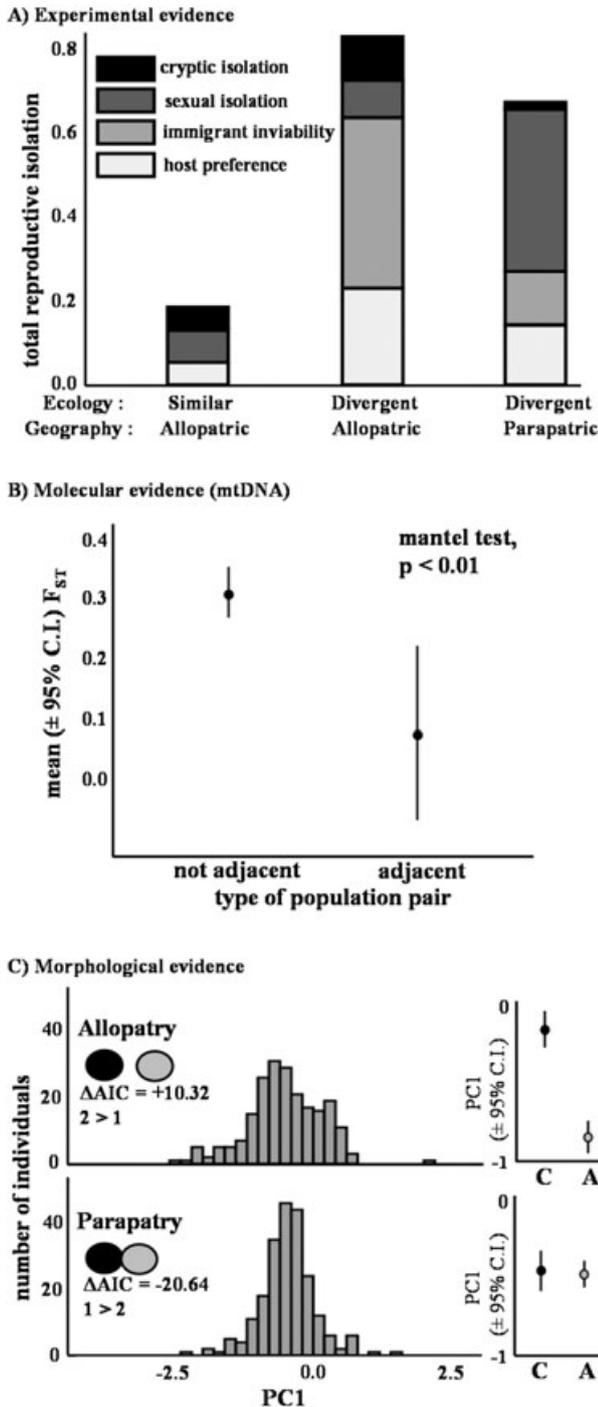
Figure 5. Evidence for gene flow and incomplete speciation between host ecotypes of *Timema cristinae*. Weaker divergence in parapatry relative to allopatry is consistent with gene flow between parapatric populations, and a constraining role for gene flow in divergence. A, experimental evidence. Total reproductive isolation never achieves values near one, and for different-host pairs is weaker in parapatry than in allopatry. B, molecular evidence (data from Nosil *et al.*, 2003). Genetic differentiation in mtDNA, estimated as F_{ST}, was significantly lower for seven adjacent, parapatric population pairs than for 129 population pairs that were not adjacent to one another. C, morphological evidence. PC1 is a multivariate, principal components axis that describes male body size. There was statistical support for bimodality in allopatry (positive Akaike information criterion values), but bimodality collapsed in parapatry (negative Akaike information criterion values). Moreover, despite statistical support for bimodality, many intermediates were evident. Thus there was much overlap between the two 'distributions', distinct clusters are not evident, and trait means differ significantly between hosts only in allopatry. Similar results were observed for other traits. Modified from Nosil (2007) and reprinted with the permission of the American Society of Naturalists.

It is obvious that divergent selection is important, but what ecological scenarios are particularly conducive to speciation with gene flow? The genetic and ecological circumstances listed below are not mutually exclusive, and could interact to promote speciation (Grant & Grant, 1997). This is obvious given that evolutionary response is a function of selection (i.e. ecology) and genetics (Lande, 1979). Figure 6 illustrates how the genetic and ecological factors considered here might affect speciation.

GENETIC CIRCUMSTANCES

Speciation via divergent selection requires a genetic mechanism by which divergent selection on ecological traits is transmitted to the genes causing reproductive isolation. The two ways this can occur are distinguished by the relationship between the genes under divergent selection (i.e. those affecting ecological traits) and the genes causing reproductive isolation (Kirkpatrick & Ravigné, 2002; Coyne & Orr, 2004). When these genes are one in the same, reproductive isolation evolves by direct selection because it is the pleiotropic effect of the genes under selection ('single-variation' models of Rice & Hostert, 1993).

However, the genes under divergent selection may often be different from the genes causing reproductive isolation. In this case, reproductive isolation evolves by indirect selection arising from the nonrandom association (linkage disequilibrium) of the genes for

or ecological circumstances where speciation in the face of gene flow is possible, or even likely? The genetic circumstances facilitating speciation in the face of gene flow have received considerable attention (Coyne & Orr, 2004), and I begin by reviewing them. The specific ecological circumstances facilitating speciation in the face of gene flow have received less attention (but see Bolnick & Fitzpatrick, 2007).
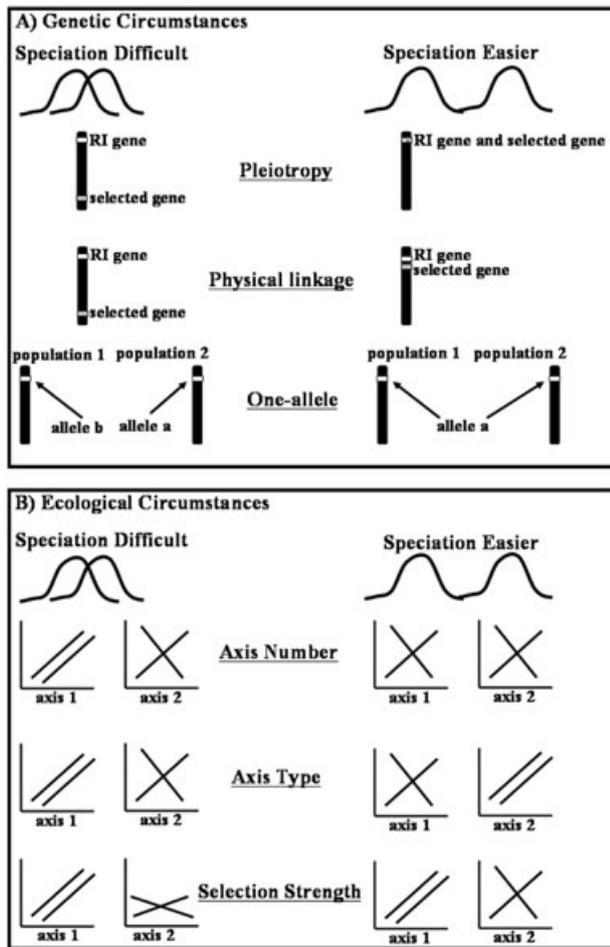
Figure 6. Genetic and ecological circumstances facilitating speciation, particularly in the face of gene flow. A, Genetic circumstances. Each bar is a chromosome (RI, reproductive isolation). Unfilled boxes within chromosomes represent genes conferring reproductive isolation. Grey boxes within chromosomes represent genes under divergent selection. Speciation is facilitated by pleiotropic effects of genes affecting traits under selection on reproductive isolation, by physical linkage between such genes, and by the fixation of the same allele conferring reproductive isolation in both diverging populations. B, ecological circumstances. Each graph depicts fitness (*y*-axis) against trait value (*x*-axis). Two ecological axes are depicted (e.g. morphology and physiology). Selection is divergent whenever lines cross within a graph, and stronger selection is depicted by steeper slopes of the lines. In the top scenario, divergent selection on both axes is required for speciation. In the center scenario, divergent selection on axis 1, rather than axis 2, is required for speciation. In the bottom scenario, stronger selection along a particular axis promotes speciation. In the top and bottom scenarios, the total strength of divergent selection, summed across the two dimensions, is greater in the right-hand column (i.e. speciation easier) than in the left-hand column (i.e. speciation difficult). For details, see text.

reproductive isolation and those for traits under selection ('double-variation' models of Rice & Hostert, 1993). Indirect selection is thought to be less effective than direct selection in the evolution of reproductive isolation because the genetic association between the genes under selection and those causing reproductive isolation is imperfect, thereby weakening selection on the latter (Kirkpatrick & Ryan, 1991; Kirkpatrick & Barton, 1997; Kirkpatrick & Ravigné, 2002). This point raises a further, fundamental problem with speciation in the face of gene flow: recombination destroys associations between alleles at loci under divergent selection and alleles at loci affecting reproductive isolation. When gene flow between populations, and subsequent recombination, occurs in nature, associations between selected and reproductive isolation loci are often expected to be weak. When such associations are weak, selection on loci affecting ecological traits will not cause a strong correlated evolutionary response in levels of reproductive isolation, and thus speciation is difficult. This point was driven home in a classic paper that still represents one of the major motivations for the field of speciation genetics (Felsenstein, 1981).

As a consequence of the logic outlined above, speciation in the face of gene flow is often considered unlikely. However, there are a number of genetic circumstances that facilitate this process. Three commonly discussed ones are pleiotropy, physical linkage, and one-allele assortative mating mechanisms (Servedio & Noor, 2003; Coyne & Orr, 2004; Rundle & Nosil, 2005; Bolnick & Fitzpatrick, 2007). The role of each has received theoretical treatment, but empirical evidence is still restricted to just one or a few key examples. Thus, although it is often stated that particular genetic architectures promote speciation, a systematic evaluation of how important genetic architecture is for speciation in nature is lacking.

Pleiotropy
There are numerous ways that pleiotropy (i.e. direct selection) can facilitate speciation. In some scenarios, divergent selection is essentially one and the same as reproductive isolation (and thus selection is direct). For example, prezygotic isolation can arise when immigrants into foreign habitats suffer reduced survival because they are poorly adapted to the non-native habitat. Although not normally considered a form of reproductive isolation, such 'immigrant inviability' can directly reduce gene flow between populations by lowering the rate of heterospecific mating encounters (Mallet & Barton, 1989; Funk, 1998; Via, Bouck & Skillman, 2000; Nosil, 2004; Nosil *et al.*, 2005). By reducing interbreeding between populations, natural selection against immigrants consti-

tutes a legitimate reproductive barrier, even though it is the direct consequence of ecologically-based divergent selection. In a more explicit genetic sense, alleles favored by selection in one environment directly reduce fitness in the other, thereby reducing gene flow (Via & Hawthorne, 2002). Similar arguments can be made for ecologically-based reductions in hybrid fitness (i.e. ecologically-dependent postzygotic isolation; Rundle & Whitlock, 2001; Rundle, 2002). In this case, the genes that adapt parental species to divergent environments also reduce the fitness of hybrids, whose intermediate phenotypes are unfit in the parental environments.

Other forms of reproductive isolation might also evolve via direct selection. For example, habitat isolation can evolve as a consequence of direct selection on habitat preference genes. This is the route by which sympatric speciation is thought to be most likely (Coyne & Orr, 2004: 130–131) and has been demonstrated in a laboratory experiment (Rice & Salt, 1990). Direct selection can also act on mate preferences, with sexual isolation evolving as a pleiotropic consequence of the adaptive divergence of mating/communication systems (Kirkpatrick, 1996; Servedio, 2001). For example, divergent environments may differ in their signal transmission properties such that different signals are most detectable in different habitats (i.e. 'sensory drive'; Morton, 1975; Ryan, Cocroft & Wilczynski, 1990; Endler, 1992; Ryan & Rand, 1993; Boughman, 2002; Slabbekoorn & Smith, 2002; Patten, Rotenberry & Zuk, 2004; Fuller, Houle & Travis, 2005). This could generate direct and habitat-specific selection to prefer the most detectable signal, resulting in adaptive divergence in mate preferences as a consequence. Likewise, genes under divergent selection might incidentally affect traits used in mate choice, resulting in sexual isolation (Jiggins et al., 2001; Jiggins, Estrada & Rodrigues, 2004). In plants, pollinator isolation can evolve as a direct consequence of adaptation to different pollinators (i.e. direct selection acts on genes affecting pollination success, such as flower colour genes; Schemske & Bradshaw, 1999; Bradshaw & Schemske, 2003). Likewise, temporal isolation, caused by differences in flowering time, could arise as the pleiotropic effect of adaptation to different environments (Macnair & Gardner, 1998). Finally, intrinsic postzygotic isolation can arise pleiotropically if alleles favoured by selection within each population contribute to incompatibilities between them (Dettman et al., 2007). How common any of these scenarios are in nature warrants investigation.

Physical linkage

The maintenance of linkage disequilibrium is greatly facilitated by the physical linkage of genes on a chromosome, which reduces recombination between them (Lynch & Walsh, 1998; Ortiz-Barrientos et al., 2002). In practice, separating pleiotropy from close physical linkage will be a difficult task because linkage disequilibrium caused by tight physical linkage has a similar effect to pleiotropy (Via & Hawthorne, 2002). Quantitative trait locus (QTL) mapping in pea aphids showed that a number of loci affecting performance and habitat preference appear to reside in similar regions of the genome, suggesting tight physical linkage of genes under selection and those conferring reproductive isolation (Hawthorne & Via, 2001; Via & Hawthorne, 2002). In monkeyflowers, hybrid sterility is either a pleiotropic effect of an allele for resistance to copper contaminated soils, or is caused by something tightly linked to it (Macnair & Christie, 1983). Chromosomal inversions may play a role here; by protecting large regions of the genome from recombination, they can facilitate speciation in the face of gene flow (Noor et al., 2001, 2007; Rieseberg, 2001; Ortiz-Barrientos et al., 2002; Feder et al., 2003a, b; Navarro & Barton, 2003; Kirkpatrick & Barton, 2006).

One-allele mechanism

The genetic basis of reproductive isolation can also affect the likelihood of speciation in the face of gene flow. Felsenstein (1981) pointed out that there are two distinct possibilities, which he termed one-allele and two-allele mechanisms. In a one-allele mechanism, reproductive isolation is caused by the same allele fixing in both populations (e.g. an allele causing individuals to prefer mates phenotypically similar to themselves). In a two-allele mechanism, different alleles fix in each population (e.g. a preference allele for large individuals in one population and a different preference allele for small individuals in the other). This distinction is important when considering the effects of recombination. Recombination in a two-allele mechanism breaks down linkage disequilibrium, weakening the genetic association between genes under selection and those causing reproductive isolation. By contrast, this problem is alleviated for a one-allele mechanism. The frequency of one-allele mechanisms in nature is unknown. The strongest evidence for a one-allele mechanism is provided by a study of *Drosophila pseudoobscura* (Ortiz-Barrientos & Noor, 2005). Females from populations of *D. pseudoobscura* that co-occur with its sibling species, *Drosophila persimilis*, exhibit high reluctance to mate with *D. persimilis* males (i.e. reinforcement; Noor, 1995). By contrast, females from allopatric populations of *D. pseudoobscura* are more inclined to hybridize. Ortiz-Barrientos & Noor (2005) show that alleles conferring high or low assortative mating in *D. pseudoobscura* produce the same effects when inserted

into *D. persimilis*, suggesting that the same assortative mating allele occurs in both species.

<div align="center">ECOLOGICAL CIRCUMSTANCES</div>

'the stronger their need for local adaptation . . . the greater the probability of changes in the components of isolating mechanisms' (Mayr, 1963: 495)

The magnitude, targets, and causes of divergent selection are affected by ecological conditions (Endler, 1977, 1986; Reimchen, 1979; Schluter, 2000; Kingsolver *et al.*, 2001; Grant & Grant, 2002; Reimchen & Nosil, 2002, 2004). Thus, there are not only genetic, but also ecological, scenarios that might allow speciation in the face of gene flow. As exemplified by Mayr's quotation, it is generally believed that as local adaptation increases, so does the probability of speciation (Gavrilets, 2004). However, what is meant by 'stronger need for local adaptation'? Obviously, stronger total divergent selection is likely to be important. But do the types and number of characters that divergent selection acts on matter? For example, strong divergent selection on traits that do not confer reproductive isolation (and are not genetically-correlated with such traits) might result in a striking polymorphism, but not speciation (Rueffler *et al.*, 2006; Gray & McKinnon, 2007; Svensson *et al.*, 2008). Another question is how does stronger total divergent selection generally arise in nature; via stronger selection on a trait already under selection or via selection on additional traits? Selection on more traits (i.e. more 'ecological dimensions'; see below) may be important simply because it is a common mechanism by which the total strength of divergent selection that population pairs are exposed to is increased in nature (Nosil & Harmon, 2008).

Here, I focus on selection acting on general 'ecological dimensions', where the idea is that different dimensions are uncorrelated (and any single dimension may involve suites of correlated traits). The main concepts are the strength of divergent selection within a dimension, the types of dimensions upon which divergent selection acts, and the number of such dimensions. However, I stress that these factors are not mutually exclusive, and might interact to affect speciation. For example, divergent selection might act on morphology, behavior, physiology, life-history, or any combination of these 'dimensions', and the strength of selection within any of these dimensions can vary. I acknowledge that 'ecological dimensions' can be subdivided and defined in different ways. Ideally, ecologically relevant and statistically independent dimensions can be identified, for example using multivariate statistics (Green, 1971; Harmon

et al., 2005), but genetic and functional independence could also be considered (Nosil & Harmon, 2008).

Selection strength within a dimension
In theory, stronger selection on a trait/gene increases the probability of speciation, all else being equal (Gavrilets, 2004). There is almost no direct data from nature. Most tests of ecological speciation use ecological divergence as a proxy for divergent selection, without measuring selection itself (Rundle & Nosil, 2005). Moreover, only two discrete categories of ecological divergence, corresponding to the presence versus absence of divergent selection, are generally considered (i.e. ecologically-divergent versus ecologically-similar population pairs; Feder *et al.*, 1994; Funk, 1998; Rundle *et al.*, 2000; Jiggins *et al.*, 2001; Nosil *et al.*, 2002; Rundle & Nosil, 2005). Two methods could be used to link the strength of selection to progress towards speciation. First, quantitative indices of ecological divergence along a single dimension could be used to indirectly estimate different strengths of divergent natural selection along a particular axis, and then relate these indices to levels of reproductive isolation (Funk *et al.*, 2006). Second, actual measurements of the strength of divergent selection in nature could be related to progress towards speciation. The first approach is less direct, but easier to implement.

Types of ecological dimensions
The types of ecological dimensions upon which selection acts can vary (Emerson, 2008, this volume). There is very little direct information on how the probability of speciation varies according to the types of traits that selection acts on (but see Price, 2008 for an exception). For example, is selection on morphology or on behavior more likely to result in speciation? Because a pleiotropic effect of genes affecting selected traits on reproductive isolation is a powerful mechanism of speciation, one prediction is that the types of dimensions most important for speciation are those most likely to have such a pleiotropic effect. Notably, which particular dimensions have such pleiotropic effects might vary among taxa. For example, divergence in morphology might be most likely to pleiotropically affect sexual isolation in visually-oriented taxa (mimetic colour patterns; Jiggins *et al.*, 2001), whereas physiology might be more likely to do so in olfactory-based taxa (i.e. via its effects on pheromones; Nosil *et al.*, 2007).

Number of ecological dimensions: dimensionality of niche divergence
The nature of the ecological niche has received continued study over the last century (Grinnell, 1917; Elton, 1927; Hutchinson, 1957, 1959; Pianka, 1978;

Schoener, 1989; Wiens & Graham, 2005). A major aspect of the niche is its dimensionality (Hutchinson, 1957; Harmon *et al.*, 2005). According to this concept, an organism's niche is defined by numerous biotic and abiotic variables, each of which can be considered an axis in multidimensional space. The dimensionality of the ecological niche can be assessed using measurements of ecological variables (Maguire, 1967; Green, 1971) or phenotypic variables that interact with the environment to determine fitness (Vandermeer, 1972; Harmon *et al.*, 2005). A distinction between considering niche dimensionality for a species versus for the process of speciation is that *differences in the niche among sets of taxa* must be examined for the latter (rather than niche properties of a single taxon) (Nosil & Harmon, 2008). Here, the focus is on dimensionality of niche *divergence*, rather than niche dimensionality *per se*. This point is critical. For example, sets of populations that each individually exhibit highly dimensional niches would not exhibit highly dimensional niche divergence if their niches were similar to one another in all axes.

Divergent selection between niches is a central component of ecological speciation (Schluter, 2000), but little is known about how, or whether, dimensionality of niche divergence affects speciation (but see Rice & Hostert, 1993). Does greater dimensionality of niche divergence increase the speed or probability of speciation? Interestingly, ideas about niche dimensionality and diversity stem back to a classic paper by Hutchinson (1959), which tackles the same issues as Felsenstein (1981), but from an ecological rather than a genetic perspective (the two papers have very similar titles).

Perhaps the best evidence that dimensionality of niche divergence might affect speciation comes from laboratory selection experiments using *Drosophila*. Such experiments have demonstrated that the evolution of reproductive isolation in the face of gene flow, via divergent selection, can occur (Rice & Hostert, 1993). Importantly, both genetic and 'ecological' (i.e. selective regime in the laboratory) factors affected the success of different experiments. For example, reproductive isolation rarely evolved when reproductive isolation was only indirectly selected (i.e. when selection was applied on a fitness trait, such as body size, rather than on reproductive isolation directly). By contrast, the evolution of reproductive isolation in the face of gene flow was more common when direct selection was applied (e.g. selection on habitat preference). However, even among studies applying direct selection, the magnitude of reproductive isolation that evolved varied among experiments according to the nature of the selective regime applied. Rice & Hostert (1993) suggest that this variation may be explained, at least in part, by the number of traits (i.e. dimen-

sions) subjected to divergent selection (i.e. 'multifarious' versus 'single-factor' divergent selection). For example, Hurd & Eisenburg (1975) selected divergently on a single trait (geotaxis) and saw only partial premating isolation evolve. A similar experiment by Soans, Pimentel & Soans (1974) achieved a similar result. By contrast, Rice and co-workers (Rice, 1985; Rice & Salt, 1988, 1990) selected on habitat preference using multiple selective regimes (phototaxis, geotaxis, chemotaxis, and development time) and observed near or complete premating isolation even under conditions of maximal potential gene flow. To the extent that the number of traits subject to selection in a lab experiment is analogous to the number of ecological dimensions subject to divergent selection in nature, this provides evidence that dimensionality of niche divergence affects speciation.

In nature, herbivorous insects provide opportunities to examine the relationship between dimensionality of niche divergence and speciation, because many axes of differentiation can be identified and tractably explored. For example, divergent host-adaptation, via divergent selection, might result in physiological trade-offs in the ability to digest different host species (Rausher, 1982; Craig, Horner & Itami, 1997; Funk *et al.*, 2002; Forister, 2004). However, host-related divergent selection may also act on morphology, to facilitate camouflage on different plants (Sandoval, 1994a, b; Nosil, 2004; Nosil & Crespi, 2006a), or for more biomechanical reasons linked to maneuvering or ovipositing on different types of host substrates (Moran, 1986; Bernays, 1991; Carroll & Boyd, 1992). Other dimensions potentially subject to divergent selection between hosts include parasite resistance and trophic morphology. Thus, herbivorous insects offer promise for examining the effects of dimensionality of niche divergence on speciation.

Some very preliminary insights come from *Timema* walking-sticks. In this system, divergent selection may act on morphology (to evade visual predation), physiology (to detoxify plant chemicals), or both of these niche dimensions. Consider two species, *Timema podura* and *T. cristinae*, who have both formed ecotypes on the same host genera, *Ceanothus* and *Adenostoma* (Law & Crespi, 2002; Sandoval & Nosil, 2005), providing a replicated test of how divergent selection affects speciation. Between ecotypes of both species, incomplete progress towards ecological speciation has occurred (data for *T. cristinae* reviewed above; for *T. podura*, see Vickery, 1993; Law & Crespi, 2002; Sandoval & Nosil, 2005). In ecotypes of both species, there is also evidence for strong divergent selection, but only along the single axis of cryptic morphology. Thus, there are strong trade-offs between hosts in survival in the face of predation, resulting in divergent selection on morphology (Sandoval, 1994b;

Nosil, 2004; Sandoval & Nosil, 2005; Nosil & Crespi, 2006a). By contrast, divergent selection on physiology does not occur in either species; irrespective of ecotype or species, fecundity in the absence of predation is always higher on *Ceanothus* (Sandoval & Nosil, 2005). Recent data indicate that dimensionality of niche divergence may explain why only partial progress towards speciation has occurred. The species pair *Timema chumash / T. podura* exhibits greater phenotypic divergence, reproductive isolation, and neutral genetic differentiation than the ecotype pairs (Nosil & Sandoval, 2008). The species pair is also subject to selection on a greater number of niche dimensions (i.e. both crypsis and physiology), suggesting that selection on more dimensions results in greater evolutionary divergence. Selection on crypsis alone may initiate speciation, resulting in ecotype formation, but selection on additional niche dimensions may be required to complete speciation. Although the available data are scarce, existing information illustrates how variability in selection might be related to speciation using insect herbivores, and *Timema* in particular.

CONCLUSIONS

'The rapidity with which an isolate is converted into a separate species depends upon . . . the selection pressure to which the isolated population is exposed, and the effectiveness of the isolation' (Mayr, 1963: 585)

Speciation may often reflect a balance between the opposing processes of selection and gene flow. Mayr's quotation above essentially acknowledges this point. Thus, differences between proponents of allopatric and sympatric speciation may simply reflect differences in opinion concerning when, and how often, selection overcomes gene flow to the extent that a discontinuity develops. When this does occur, it is of interest to determine the genetic or ecological circumstances that facilitated divergence. Ultimately, agreement on the roles of geography and ecology in speciation, and amongst proponents of allopatric versus sympatric speciation, awaits further empirical and theoretical studies examining these circumstances. It may be that progress towards speciation in the face of gene flow can be predicted using standard concepts in genetics (recombination; Felsenstein, 1981), population genetics (strength of selection), and ecology (the ecological niche; Hutchinson, 1957). Finally, the timing of the onset of gene flow during the process of divergence can vary (Feder *et al.*, 2003b; Rundle & Schluter, 2004; Rundle & Nosil, 2005; Won & Hey, 2005; Xie *et al.*, 2007; Niemiller *et al.*, 2008; Nosil, 2008; Emerson, 2008, this volume), and might affect how these ecological and genetic factors affect speciation, with greater differentiation before the onset of gene flow promoting speciation (Liou & Price, 1994; Kirkpatrick & Ravigné, 2002). Thus, studies that integrate ecology, genetics, and geography at different stages of evolutionary divergence are likely to yield the most comprehensive understanding of the speciation process, an understanding that is not restricted to polarized and dichotomized views of the process.

REFERENCES

Barluenga M, Stölting KN, Salzburger W, Muschick M, Meyer A. 2006. Sympatric speciation in Nicaraguan crater lake cichlid fish. *Nature* **439:** 719–723.

Barton NH. 1979. The dynamics of hybrid zones. *Heredity* **43:** 341–359.

Barton NH. 1983. Multilocus clines. *Evolution* **37:** 454–471.

Barton NH, Hewitt GM. 1985. Analysis of hybrid zones. *Annual Review of Ecology and Systematics* **16:** 113–148.

Barton NH, Hewitt GM. 1989. Adaptation, speciation and hybrid zones. *Nature* **341:** 497–503.

Beerli P, Felsenstein J. 2001. Maximum-likelihood estimation of a migration matrix and effective population sizes in n subpopulations by using a coalescent approach. *Proceedings of the National Academy of Sciences of the United States of America* **98:** 4563–4568.

Beltrán M, Jiggins CD, Bull V, Linares M, Mallet J, McMillan WO, Bermingham E. 2002. Phylogenetic discordance at the species boundary: comparative gene genealogies among rapidly radiating *Heliconius* butterflies. *Molecular Biology and Evolution* **19:** 2176–2190.

Berlocher SH, Feder JL. 2002. Sympatric speciation in phytophagous insects: moving beyond controversy? *Annual Review of Entomology* **47:** 773–815.

Bernays EA. 1991. Evolution of insect morphology in relation to plants. *Philosophical Transactions of the Royal Society of London Series B, Biological Sciences* **333:** 257–264.

Blair WF. 1950. Ecological factors in speciation of *Peromyscus*. *Evolution* **4:** 253–275.

Bolnick DA, Nosil P. 2007. Natural selection in populations subject to a migration load. *Evolution* **61:** 2229–2243.

Bolnick DI, Fitzpatrick BM. 2007. Sympatric speciation: models and empirical evidence. *Annual Review of Ecology, Evolution, and Systematics* **38:** 459–487.

Boughman JW. 2002. How sensory drive can promote speciation. *Trends in Ecology & Evolution* **10:** 1–7.

Bradshaw HD, Schemske DW. 2003. Allele substitution at a flower colour locus produces a pollinator shift in monkeyflowers. *Nature* **426:** 176–178.

Bulmer MG. 1972. Multiple niche polymorphism. *The American Naturalist* **106:** 254–257.

Bush GL, Butlin RK. 2004. Sympatric speciation in insects: an overview. In: Dieckmann U, Doebeli M, Metz H, Tautz D, eds. *Adaptive speciation*, Cambridge: Cambridge University Press, 229–248.

Butlin RK. 1995. Reinforcement: an idea evolving. *Trends in Ecology & Evolution* **10:** 432–433.

Campbell D, Bernatchez L. 2004. Generic scan using AFLP markers as a means to assess the role of directional selection in the divergence of sympatric whitefish ecotypes. *Molecular Biology and Evolution* **21:** 945–956.

Carroll SP, Boyd C. 1992. Host race radiation in the soapberry bug: natural history with the history. *Evolution* **46:** 1052–1069.

Coyne JA. 1994. Ernst Mayr and the origin of species. *Evolution* **48:** 19–30.

Coyne JA, Orr HA. 1989. Patterns of speciation in *Drosophila*. *Evolution* **43:** 362–381.

Coyne JA, Orr HA. 2004. *Speciation*. Sunderland, MA: Sinauer Associates, Inc.

Craig TP, Horner JD, Itami JK. 1997. Hybridization studies on the host races of *Eurosta solidaginis*: implications for sympatric speciation. *Evolution* **51:** 1552–1560.

Crespi BJ, Sandoval CP. 2000. Phylogenetic evidence for the evolution of ecological specialization in *Timema* walking-sticks. *Journal of Evolutionary Biology* **13:** 249–262.

De Queiroz K. 2005. Ernst Mayr and the modern concept of species. *Proceedings of the National Academy of Sciences of the United States of America* **102:** 6600–6607.

Dettman JR, Sirjusingh C, Kohn LM, Anderson JB. 2007. Incipient speciation by divergent adaptation and antagonistic epistasis in yeast. *Nature* **447:** 585–588.

Dhondt AA, Adriaensen F, Matthysen E, Kempenaers B. 1990. Nonadaptive clutch size in tits. *Nature* **348:** 723–725.

Dieckmann U, Doebeli M, Metz JAJ, Tautz D. 2004. *Adaptive speciation*. Cambridge: Cambridge University Press.

Dobzhansky T. 1935. A critique of the species concept in biology. *Philosophy and Science* **2:** 344–355.

Dobzhansky T. 1937. *Genetics and the origin of species*. New York, NY: Columbia University Press.

Dopman ED, Perez L, Bogdanowicz SM, Harrison RG. 2005. Consequences of reproductive barriers for genealogical discordance in the European corn borer. *Proceedings of the National Academy of Sciences of the United States of America* **102:** 14706–14711.

Drès M, Mallet J. 2002. Host races in plant-feeding insects and their importance in sympatric speciation. *Philosophical Transactions of the Royal Society of London Series B, Biological Sciences* **357:** 471–492.

Egan SP, Nosil P, Funk DJ. 2008. Selection and genomic differentiation during ecological speciation: isolating the contributions of host-association via a comparative genome scan of *Neochlamisus bebbianae* leaf beetles. *Evolution* **62:** 1162–1181.

Elton CS. 1927. *Animal ecology*. London: Sidwich & Jackson.

Emelianov I, Marec F, Mallet J. 2004. Genomic evidence for divergence with gene flow in host races of the larch budmoth. *Proceedings of the Royal Society of London Series B, Biological Sciences* **271:** 97–105.

Emerson BA. 2008. Speciation on islands – what are we learning? *Biological Journal of the Linnean Society* **95:** 47–52.

Endler JA. 1973. Gene flow and population differentiation. *Science* **179:** 243–250.

Endler JA. 1977. *Geographic variation, speciation and clines*. Princeton, NJ: Princeton University Press.

Endler JA. 1986. *Natural selection in the wild*. Princeton, NJ: Princeton University Press.

Endler JA. 1992. Signals, signal conditions, and the direction of evolution. *The American Naturalist* **139:** 125–153.

Feder JL, Berlocher SH, Roethele JB, Dambroski H, Smith JJ, Perry WL, Gavrilovic V, Filchak KE, Rull J, Aluja M. 2003a. Allopatric genetic origins for sympatric host-plant shifts and race formation in *Rhagoletis*. *Proceedings of the National Academy of Sciences of the United States of America* **100:** 10314–10319.

Feder JL, Opp SB, Wlazlo B, Reynolds K, Go W, Spsisak S. 1994. Host fidelity is an effective premating barrier between sympatric races of the apple maggot fly. *Proceedings of the National Academy of Sciences of the United States of America* **91:** 7990–7994.

Feder JL, Roethele FB, Filchak K, Niedbalski J, Romero-Severson J. 2003b. Evidence for inversion polymorphism related to sympatric host race formation in the apple maggot fly, *Rhagoletis pomonella*. *Genetics* **163:** 939–953.

Felsenstein J. 1976. The theoretical population genetics of variable selection and migration. *Annual Review of Genetics* **10:** 253–280.

Felsenstein J. 1981. Skepticism towards Santa Rosalia, or why are there so few kinds of animals? *Evolution* **35:** 124–138.

Fisher RA. 1930. *The genetical theory of natural selection*. Oxford: Claredon Press.

Fisher RA. 1950. Gene frequencies in a cline determined by selection and diffusion. *Biometrics* **6:** 353–361.

Forister ML. 2004. Oviposition preference and larval performance within a diverging lineage of lycaenid butterflies. *Ecological Entomology* **29:** 264–272.

Fuller RC, Houle D, Travis J. 2005. Sensory bias as an explanation for the evolution of mate preferences. *The American Naturalist* **166:** 437–446.

Funk DJ. 1998. Isolating a role for natural selection in

speciation: host adaptation and sexual isolation in *Neochlamisus bebbianae* leaf beetles. *Evolution* **52:** 1744–1759.

Funk DJ, Filchak KE, Feder JL. 2002. Herbivorous insects: model systems for the comparative study of speciation ecology. *Genetica* **116:** 251–267.

Funk DJ, Nosil P. 2007. Comparative analyses and ecological speciation in herbivorous insects. In: Tilmon K, ed. *Ecology and evolution of herbivorous insects*. Berkeley, CA: California University Press, 117–135.

Funk DJ, Nosil P, Etges W. 2006. Ecological divergence exhibits consistently positive associations with reproductive isolation across disparate taxa. *Proceedings of the National Academy of Sciences of the United States of America* **103:** 3209–3213.

Funk DJ, Omland KE. 2003. Species-level paraphyly and polyphyly: frequency, causes, and consequences, with insights from animal mitochondrial DNA. *Annual Review of Ecology, Evolution, and Systematics* **34:** 397–423.

Garcia-Ramos G, Kirkpatrick M. 1997. Genetic models of adaptation and gene flow in peripheral populations. *Evolution* **51:** 21–28.

Gavrilets S. 2004. *Fitness landscapes and the origin of species*. Princeton, NJ: Princeton University Press.

Geraldes A, Ferrand N, Nachman NW. 2006. Contrasting patterns of introgression at X-linked loci across the hybrid zone between subspecies of the European rabbit (*Oryctolagus cuniculus*). *Genetics* **173:** 919–933.

Grant PR, Grant BR. **1997.** Genetics and the origin of bird species. *Proceedings of the National Academy of Sciences of the United States of America* **94:** 7768–7775.

Grant PR, Grant BR. 2002. Unpredictable evolution in a 30-year study of Darwin's finches. *Science* **296:** 707–711.

Grant PR, Grant BR, Petren K. 2005. Hybridization in the recent past. *The American Naturalist* **166:** 56–67.

Gray SM, McKinnon JS. 2007. Linking color polymorphism maintenance and speciation. *Trends in Ecology & Evolution* **22:** 71–79.

Green RH. 1971. A multivariate statistical approach to the Hutchinsonian niche: bivalve mollusks of central Canada. *Ecology* **52:** 543–556.

Grinnell J. 1917. The niche relationships of the California thrasher. *Auk* **34:** 427–433.

Haffer J. 2006. The history of the biological species concept. *Acta Zoologica Sinica* **52:** 415–420.

Haldane JBS. 1930. A mathematical theory of natural and artificial selection. Part IV. Isolation. *Proceedings of the Cambridge Philosophical Society* **26:** 220–230.

Haldane JBS. 1932. *The causes of evolution*. New York, NY: Longmans.

Haldane JBS. 1948. The theory of a cline. *Journal of Genetics* **48:** 277–284.

Harmon LJ, Kolbe JJ, Cheverud JM, Losos JB. 2005. Convergence and the multidimensional niche. *Evolution* **59:** 409–421.

Hawthorne DJ, Via S. 2001. Genetic linkage of ecological specialization and reproductive isolation in pea aphids. *Nature* **412:** 904–907.

Hendry AP, Day T, Taylor EB. 2001. Population mixing and the adaptive divergence of quantitative traits in discrete populations: a theoretical framework for empirical tests. *Evolution* **55:** 459–466.

Hendry AP, Taylor EB. 2004. How much of the variation in adaptive divergence can be explained by gene flow? An evaluation using lake-stream stickleback pairs. *Evolution* **58:** 2319–2331.

Hendry AP, Taylor EB, McPhail JD. 2002. Adaptive divergence and the balance between selection and gene flow: lake and stream stickleback in the Misty system. *Evolution* **56:** 1199–1216.

Hey J. 2006. Recent advances in assessing gene flow between diverging populations and species. *Current Opinion in Genetics & Development* **16:** 592–596.

Hey J, Waples RS, Arnold ML, Butlin RK, Harrison RG. 2003. Understanding and confronting species uncertainty in biology and conservation. *Trends in Ecology & Evolution* **18:** 597–603.

Howard DJ, Gregory PG, Chu JM, Cain ML. 1998. Conspecific sperm precedence is an effective barrier to hybridization between closely related species. *Evolution* **52:** 511–516.

Hurd LE, Eisenburg RM. 1975. Divergent selection for geotactic response and the evolution of reproductive isolation in sympatric and allopatric populations of houseflies. *The American Naturalist* **109:** 353–358.

Hutchinson GE. 1957. Concluding remarks. *Cold Spring Harbor Symposia on Quantitative Biology* **22:** 415–427.

Hutchinson GE. 1959. Homage to Santa Rosalia or Why are there so many kinds of animals? *The American Naturalist* **93:** 145–159.

Jiggins CD, Estrada C, Rodrigues A. 2004. Mimicry and the evolution of premating isolation in *Heliconius melpomene* Linnaeus. *Journal of Evolutionary Biology* **17:** 680–691.

Jiggins CD, Mallet J. 2001. Bimodal hybrid zones and speciation. *Trends in Ecology & Evolution* **15:** 250–255.

Jiggins CD, Naisbit RE, Coe RL, Mallet J. 2001. Reproductive isolation caused by colour pattern mimicry. *Nature* **411:** 302–305.

Johannesson K, Johannesson B, Rolan-Alvarez E. 1993. Morphological differentiation and genetic cohesiveness over a microenvironmental gradient in the marine snail *Littorina saxatilis*. *Evolution* **47:** 1770–1787.

Kawecki TJ, Ebert D. 2004. Conceptual issues in local adaptation. *Ecology Letters* **7:** 1225–1241.

King RB, Lawson R. 1995. Color-pattern variation in Lake Erie water snakes: the role of gene flow. *Evolution* **49:** 885–896.

Kingsolver JG, Hoekstra HE, Hoekstra JM, Berrigan D, Vignieri SN, Hill CE, Hoang A, Gibert P, Beerli P. 2001. The strength of phenotypic selection in natural populations. *The American Naturalist* **157:** 245–261.

Kirkpatrick M. 1996. Good genes and direct selection in evolution of mating preferences. *Evolution* **50:** 2125–2140.

Kirkpatrick M, Barton N. 2006. Chromosome inversions, local adaptation and speciation. *Genetics* **173:** 419–434.

Kirkpatrick M, Barton NH. 1997. The strength of indirect

selection on female mating preferences. *Proceedings of the National Academy of Sciences of the United States of America* **94:** 1282–1286.

Kirkpatrick M, Ravigné V. 2002. Speciation by natural and sexual selection: models and experiments. *The American Naturalist* **159:** S22–S35.

Kirkpatrick M, Ryan MJ. 1991. The evolution of mating preferences and the paradox of the lek. *Nature* **350:** 33–38.

Knapp S. 2008. Species concepts and floras – what are species for? *Biological Journal of the Linnean Society* **95:** 17–25.

Lande R. 1979. Quantitative genetic-analysis of multivariate evolution, applied to brain – body size allometry. *Evolution* **33:** 402–416.

Langerhans RB, Gifford ME, Joseph EO. 2007. Ecological speciation in *Gambusia* fishes. *Evolution* **61:** 2056–2074.

Law JH, Crespi BJ. 2002. The evolution of geographic parthenogenesis in *Timema* walking sticks. *Molecular Ecology* **11:** 1471–1489.

Lenormand T. 2002. Gene flow and the limits to natural selection. *Trends in Ecology and Evolution* **17:** 183–189.

Liou LW, Price TD. 1994. Speciation by reinforcement of premating isolation. *Evolution* **48:** 1451–1459.

Lu G, Bernatchez L. 1999. Correlated trophic specialization and genetic divergence in sympatric lake whitefish ecotypes (*Coregonus clupeaformis*): support for the ecological speciation hypothesis. *Evolution* **53:** 1491–1505.

Lynch M, Walsh B. 1998. *Genetics and analysis of quantitative traits*. Sunderland, MA: Sinauer Associates.

Maan ME, Hofker KD, Van Aphen JJM, Seehausen O. 2006. Sensory drive in cichlid speciation. *The American Naturalist* **167:** 947–954.

Macnair MR, Christie P. 1983. Reproductive isolation as a pleiotropic effect of copper tolerance in *Mimulus guttatus*? *Heredity* **50:** 295–302.

Macnair MR, Gardner M. 1998. The evolution of edaphic endemics. In: Howard DJ, Berlocher SH, eds. *Endless forms: species and speciation*. Oxford: Oxford University Press, 157–171.

Maguire B Jr. 1967. A partial analysis of the niche. *The American Naturalist* **101:** 515–523.

Mallet J. 1995. A species definition for the modern synthesis. *Trends in Ecology & Evolution* **10:** 294–299.

Mallet J. 2006. Mayr's view of Darwin: was Darwin wrong about speciation? *Biological Journal of the Linnean Society* **95:** 3–16.

Mallet J. 2008. What does *Drosophila* genetics tell us about speciation? *Trends in Ecology & Evolution* **21:** 186–193.

Mallet J, Barton NH. 1989. Strong natural selection in a warning-color hybrid zone. *Evolution* **43:** 421–431.

Mallet J, Barton NH, Jose Santisteban C, Manuel Muedas M, Eeley H. 1990. Estimates of selection and gene flow from measures of cline width and linkage disequilibrium in *Heliconius* hybrid zones. *Genetics* **124:** 921–936.

Mallet J, Beltran M, Neukirchen W, Linares M. 2007. Natural hybridization in heliconiine butterflies: the species boundary as a continuum. *BMC Evolutionary Biology* **7:** 28.

May RM, Endler JA, McMurtie RE. 1975. Gene frequency clines in the presence of selection opposed by gene flow. *The American Naturalist* **109:** 650–676.

Mayr E. 1942. *Systematics and the origin of species*. New York, NY: Columbia University Press.

Mayr E. 1947. Ecological factors in speciation. *Evolution* **1:** 263–288.

Mayr E. 1963. *Animal species and evolution*. Cambridge, MA: Harvard University Press.

Mayr E. 2001. *What evolution is*. New York, NY: Basic Books.

Moore J-S, Gow JL, Taylor EB, Hendry AP. 2007. Quantifying the constraining influence of gene flow on adaptive divergence in the lake-stream threespine stickleback system. *Evolution* **61:** 2015–2026.

Moran NA. 1986. Morphological adaptation to host plants in *Uroleucon* (Homoptera: Aphididae). *Evolution* **40:** 1044–1050.

Morjan CL, Rieseberg LH. 2004. How species evolve collectively: implications of gene flow and selection for the spread of advantageous alleles. *Molecular Ecology* **13:** 1341–1356.

Morton ES. 1975. Ecological sources of selection on avian sounds. *The American Naturalist* **109:** 17–34.

Muller HJ. 1942. Isolating mechanisms, evolution and temperature. *Biological Symposia* **6:** 71–125.

Navarro A, Barton NH. 2003. Accumulating postzygotic isolation genes in parapatry: a new twist on chromosomal speciation. *Evolution* **57:** 447–459.

Niemiller ML, Fitzpatrick BM, Miller BT. 2008. Recent divergence-with-gene-flow in Tennessee Cave Salamanders (Plethodontidae: *Gyrinophilus*) inferred from gene genealogies. *Molecular Ecology* **17:** 2258–2275.

Noor MAF. 1995. Speciation driven by natural selection in Drosophila. *Nature* **375:** 674–675.

Noor MAF, Garfield DA, Schaeffer SW, Machado CA. 2007. Divergent between the *Drosophila pseudoobscura* and *D. persimilis* genome sequences in relation to chromosomal inversions. *Genetics* **177:** 1417–1428.

Noor MAF, Grams KK, Bertucci LA, Reiland J. 2001. Chromosomal inversions and the reproductive isolation of species. *Proceedings of the National Academy of Sciences of the United States of America* **98:** 12084–12088.

Nosil P. 2004. Reproductive isolation caused by visual predation on migrants between divergent environments. *Proceedings of the Royal Society of London Series B, Biological Sciences* **271:** 1521–1528.

Nosil P. 2007. Divergent host-plant adaptation and reproductive isolation between ecotypes of *Timema cristinae* walking-sticks. *The American Naturalist* **169:** 151–162.

Nosil P. 2008. Speciation with gene flow could be common. *Molecular Ecology* **17:** 2103–2106.

Nosil P, Crespi BJ. 2004. Does gene flow constrain trait divergence or vice-versa? A test using ecomorphology and sexual isolation in *Timema cristinae* walking-sticks. *Evolution* **58:** 101–112.

Nosil P, Crespi BJ. 2006a. Experimental evidence that predation promotes divergence during adaptive radiation. *Proceedings of the National Academy of Sciences of the United States of America* **103:** 9090–9095.

Nosil P, Crespi BJ. 2006b. Ecological divergence promotes the evolution of cryptic reproductive isolation. *Proceedings of the Royal Society of London Series B, Biological Sciences* **273:** 991–997.

Nosil P, Crespi BJ, Gries R, Gries G. 2007. Natural selection and divergence in mate preference during speciation. *Genetica* **128:** 309–327.

Nosil P, Crespi BJ, Sandoval CP. 2002. Host-plant adaptation drives the parallel evolution of reproductive isolation. *Nature* **417:** 441–443.

Nosil P, Crespi BJ, Sandoval CP. 2003. Reproductive isolation driven by the combined effects of ecological adaptation and reinforcement. *Proceedings of the Royal Society of London Series B, Biological Sciences* **270:** 1911–1918.

Nosil P, Egan SP, Funk DJ. 2008. Heterogeneous genomic differentiation between walking-stick ecotypes: 'isolation-by-adaptation' and multiple roles for divergent selection. *Evolution* **62:** 313–336.

Nosil P, Harmon LJ. 2008. Niche dimensionality and ecological speciation. In: Butlin R, Bridle J, Schluter D, eds. *Ecology and speciation*. Cambridge: Cambridge University Press.

Nosil P, Rundle HD. 2008. Ecological speciation: natural selection and the formation of new species. In: Losos J, ed. *Autecology*. Princeton, NJ: Princeton University Press.

Nosil P, Sandoval CP. 2008. Ecological niche dimensionality and the evolutionary diversification of stick insects. *PLoS ONE* **3:** E1907.

Nosil P, Vines TH, Funk DJ. 2005. Perspective: reproductive isolation caused by natural selection against immigrants from divergent habitats. *Evolution* **59:** 705–719.

Ortiz-Barrientos D, Noor MAF. 2005. Evidence for a one-allele assortative mating locus. *Science* **310:** 1467–1467.

Ortiz-Barrientos D, Reiland J, Hey J, Noor MAF. 2002. Recombination and the divergence of hybridizing species. *Genetica* **116:** 167–178.

Patten MA, Rotenberry JT, Zuk M. 2004. Habitat selection, acoustic adaptation, and the evolution of reproductive isolation. *Evolution* **58:** 2144–2155.

Pianka E. 1978. *Evolutionary ecology*, 3rd edn. New York, NY: Harper & Row.

Price T. 2008. *Speciation in birds*. Greenwood Village, CO: Roberts and Company.

Putnam AS, Scriber JM, Andolfatto P. 2007. Discordant divergence times among Z chromosome regions between two ecologically distinct swallowtail butterfly species. *Evolution* **61:** 912–927.

Ramsey J, Bradshaw HD Jr, Schemske DW. 2003. Components of reproductive isolation between the monkeyflowers *Mimulus lewissii* and *M. cardinalis* (Phrymaceae). *Evolution* **57:** 1520–1534.

Ramsey J, Schemske DW. 1998. Pathways mechanisms and rates of polyploid formation in flowering plants. *Annual Review of Ecology and Systematics* **29:** 467–501.

Rausher MD. 1982. Population differentiation in *Euphydryas editha* butterflies: larval adaptation to different hosts. *Evolution* **36:** 581–590.

Reimchen TE. 1979. Substratum heterogeneity, crypsis, and colour polymorphism in an intertidal snail. *Canadian Journal of Zoology* **57:** 1070–1085.

Reimchen TE, Nosil P. 2002. Temporal variation in divergent selection on spine number in threespine stickleback. *Evolution* **56:** 2472–2483.

Reimchen TE, Nosil P. 2004. Variable predation regimes predict the evolution of sexual dimorphism in a population of threespine stickleback. *Evolution* **58:** 1274–1281.

Rice WR. 1985. Disruptive selection on habitat preference and the evolution of reproductive isolation: an exploratory experiment. *Evolution* **39:** 645–656.

Rice WR, Hostert EE. 1993. Laboratory experiments on speciation: what have we learned in 40 years? *Evolution* **47:** 1637–1653.

Rice WR, Salt GW. 1988. Speciation via disruptive selection on habitat preference: experimental evidence. *The American Naturalist* **131:** 911–917.

Rice WR, Salt GW. 1990. The evolution of reproductive isolation as a correlated character under sympatric conditions: experimental evidence. *Evolution* **44:** 1140–1152.

Riechert SE. 1993. Investigation of potential gene flow limitation of behavioral adaptation in an aridlands spider. *Behavioral Ecology and Sociobiology* **32:** 355–363.

Riechert SE, Hall RF. 2000. Local population success in heterogeneous habitats: reciprocal transplant experiments completed on a desert spider. *Journal of Evolutionary Biology* **13:** 541–550.

Riechert SE, Singer FD, Jones TC. 2001. High gene flow levels lead to gamete wastage in a desert spider system. *Genetica* **112–113:** 297–319.

Rieseberg LH. 2001. Chromosomal rearrangements and speciation. *Trends in Ecology & Evolution* **16:** 351–358.

Rolan-Alvarez E, Elrandsson J, Johannesson K, Cruz R. 1999. Mechanisms of incomplete prezygotic reproductive isolation in an intertidal snail: testing behavioural models in wild populations. *Journal of Evolutionary Biology* **12:** 879–890.

Ross KG, Keller L. 1995. Joint influence of gene flow and selection on a reproductively important genetic polymorphism in the fire ant *Solenopsis invicta*. *The American Naturalist* **146:** 325–348.

Roughgarden J. 1979. *Theory of population genetics and evolutionary ecology: an introduction*. New York, NY: MacMillan.

Rueffler C, Van Dooren TJM, Leimar O, Abrams PA. 2006. Disruptive selection and then what? *Trends in Ecology & Evolution* **21:** 238–245.

Rundle HD. 2002. A test of ecologically dependent postmating isolation between sympatric sticklebacks. *Evolution* **56:** 322–329.

Rundle HD, Nagel L, Boughman JW, Schluter D. 2000. Natural selection and parallel speciation in sympatric sticklebacks. *Science* **287:** 306–308.

Rundle HD, Nosil P. 2005. Ecological speciation. *Ecology Letters* **8:** 336–352.

Rundle HD, Schluter D. 2004. Natural selection and ecological speciation in sticklebacks. In: Dieckmann U, Doebeli

M, Metz JAJ, Tautz D, eds. *Adaptive speciation*. Cambridge: Cambridge University Press, 192–209.

Rundle HD, Whitlock M. 2001. A genetic interpretation of ecologically dependent isolation. *Evolution* **55:** 198–201.

Ryan MJ, Cocroft RB, Wilczynski W. 1990. The role of environmental selection in intraspecific divergence of mate recognition signals in the cricket frog, *Acris crepitans*. *Evolution* **44:** 1869–1872.

Ryan MJ, Rand AS. 1993. Species recognition and sexual selection as a unitary problem in animal communication. *Evolution* **47:** 647–657.

Sandoval CP. 1994a. The effects of relative geographic scales of gene flow and selection on morph frequencies in the walking stick *Timema cristinae*. *Evolution* **48:** 1866–1879.

Sandoval CP. 1994b. Differential visual predation on morphs of *Timema cristinae* (Phasmatodeae: Timemidae) and its consequences for host range. *Biological Journal of the Linnean Society* **52:** 341–356.

Sandoval CP. 2000. Presence of a walking-stick population (Phasmatoptera: Timematodea) after a wildfire. *Southwestern Naturalist* **45:** 19–25.

Sandoval CP, Nosil P. 2005. Counteracting selective regimes and host preference evolution in ecotypes of two species of walking-sticks. *Evolution* **59:** 2405–2413.

Savolainen V, Anstett MC, Lexer C, Hutton I, Clarkson JJ, Norup MV, Powell MP, Springate D, Salamin N, Baker WJ. 2006. Sympatric speciation in palms on an oceanic island. *Nature* **441:** 210–213.

Schemske DW, Bradshaw HD. 1999. Pollinator preference and the evolution of floral traits in monkeyflowers (Mimulus). *Proceedings of the National Academy of Sciences of the United States of America* **96:** 11910–11915.

Schluter D. 1996. Ecological speciation in postglacial fishes. *Philosophical Transactions of the Royal Society of London Series B, Biological Sciences* **351:** 807–814.

Schluter D. 1998. Ecological causes of speciation. In: Howard DJ, Berlocher SH, eds. *Endless forms: species and speciation*. Oxford: Oxford University Press, 114–129.

Schluter D. 2000. *The ecology of adaptive radiation*. Oxford: Oxford University Press.

Schluter D. 2001. Ecology and the origin of species. *Trends in Ecology & Evolution* **16:** 372–380.

Schluter D, Nagel LM. 1995. Parallel speciation by natural selection. *The American Naturalist* **146:** 292–301.

Schoener TW. 1989. The ecological niche. In: Cherrett JM, ed. *Ecological concepts*. Oxford: Blackwell Scientific, 79–113.

Seehausen O. 2008. The sequence of events along a 'speciation transect' in the Lake Victoria cichlid fish *Pundamilia*. In: Butlin R, Bridle J, Schluter D, eds. *Ecology and speciation*. Cambridge: Cambridge University Press.

Servedio M. 2001. Beyond reinforcement: the evolution of premating isolation by direct selection on preferences and postmating, prezygotic incompatibilities. *Evolution* **55:** 1909–1920.

Servedio MR, Noor MAF. 2003. The role of reinforcement in speciation: theory and data. *Annual Review of Ecology, Evolution, and Systematics* **34:** 339–364.

Slabbekoorn H, Smith TB. 2002. Habitat-dependent song divergence in the little greenbul: an analysis of selection pressures on acoustic signals. *Evolution* **56:** 1849–1858.

Slatkin M. 1973. Gene flow and selection in a cline. *Genetics* **75:** 733–756.

Slatkin M. 1981. Pleiotropy and parapatric selection. *Evolution* **36:** 263–270.

Slatkin M. 1985. Gene flow in natural populations. *Annual Review of Ecology and Systematics* **16:** 393–430.

Smith JW, Benkman CW. 2007. A coevolutionary arms race causes ecological speciation in crossbills. *The American Naturalist* **169:** 455–465.

Smith TB, Wayne RK, Girman DJ, Bruford MW. 1997. A role for ecotones in generating rainforest biodiversity. *Science* **276:** 1855–1857.

Soans AB, Pimentel D, Soans JS. 1974. Evolution of reproductive isolation in allopatric and sympatric populations. *The American Naturalist* **108:** 117–124.

Storfer A, Cross J, Rush R, Caruso J. 1999. Adaptive coloration and gene flow as a constraint to local adaptation in the streamside salamander, *Ambystoma barbouri*. *Evolution* **53:** 889–898.

Storfer A, Sih A. 1998. Gene flow and ineffective antipredator behavior in a stream-breeding salamander. *Evolution* **52:** 558–565.

Svensson EI, Abbot JK, Gosden TP, Coreau A. 2008. Female polymorphisms, sexual conflict and limits to speciation processes in animals. *Evolutionary Ecology* doi: 10.1007/s10682-007-9208-2.

Turelli M, Barton NH, Coyne JA. 2001. Theory and speciation. *Trends in Ecology & Evolution* **16:** 330–343.

Vandermeer JH. 1972. Niche theory. *Annual Review of Ecology and Systematics* **3:** 107–132.

Via S. 1999. Reproductive isolation between sympatric races of pea aphids. I. Gene flow restriction and habitat choice. *Evolution* **53:** 1446–1457.

Via S. 2001. Sympatric speciation in animals: the ugly duckling grows up. *Trends in Ecology & Evolution* **16:** 381–390.

Via S, Bouck AC, Skillman S. 2000. Reproductive isolation between divergent races of pea aphids on two hosts. II. Selection against migrants and hybrids in the parental environments. *Evolution* **54:** 1626–1637.

Via S, Hawthorne DJ. 2002. The genetic architecture of ecological specialization: correlated gene effects on host use and habitat choice in pea aphids. *The American Naturalist* **159:** S76–S88.

Vickery VR. 1993. Revision of *Timema* Scudder (Phasmatoptera: Timematodea) including three new species. *The Canadian Entomologist* **125:** 657–692.

Vines TH, Köhler SC, Thiel M, Ghira I, Sands IR, MacCallum CJ, Barton NH, Nürnberger B. 2003. The maintenance of reproductive isolation in a mosaic hybrid zone between the fire-bellied toads *Bombina bombina* and *B. ariegate*. *Evolution* **57:** 1876–1888.

Vines TH, Schluter D. 2006. Strong assortative mating between allopatric sticklebacks as a by-product of adaptation to different environments. *Proceedings of the Royal Society of London. Series B, Biological Sciences* **273:** 911–916.

Wallace AR. 1864. The Malayan Papilionidae or swallow-tailed butterflies, as illustrative of the theory of natural selection. In: Wallace AR, ed. *Contributions to the theory of natural selection*. London, 130–200.

Wiens JJ, Graham CH. 2005. Niche conservatism: integrating evolution, ecology, and conservation biology. *Annual Review of Ecology, Evolution, and Systematics* **36:** 519–539.

Won YJ, Hey J. 2005. Divergence population genetics of chimpanzees. *Molecular Biology and Evolution* **22:** 297–307.

Wright S. 1931. Evolution in Mendelian populations. *Genetics* **16:** 97–159.

Wright S. 1940. Breeding structure of populations in relation to speciation. *The American Naturalist* **74:** 232–248.

Wright S. 1943. Isolation by distance. *Genetics* **28:** 114–138.

Wu CI. 2001. The genic view of the process of speciation. *Journal of Evolutionary Biology* **14:** 851–865.

Xie X, Rull J, Michel A, Velez S, Forbes AA, Lobo N, Aluja M, Feder JL. 2007. Hawthorn-infesting populations of *Rhagoletis pomonella* in Mexico and speciation mode plurality. *Evolution* **61:** 1091–1105.