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Review

Local adaptation and reproductive isolation: when does speciation start?

Roger K. Butlin^{1,2,*}, Rui Faria^{3,4}

¹Ecology and Evolutionary Biology, School of Biosciences, The University of Sheffield, Sheffield S10 2TN, United Kingdom

²Department of Marine Sciences, University of Gothenburg, Tjärnö Marine Laboratory, SE 45296 Strömstad, Sweden

³CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO, Laboratório Associado, Universidade do Porto, Vairão, Portugal

⁴BIOPOLIS Program in Genomics, Biodiversity and Land Planning, CIBIO, Campus de Vairão, 4485-661 Vairão, Portugal

*Corresponding author. Ecology and Evolutionary Biology, School of Biosciences, The University of Sheffield, Sheffield S10 2TN, United Kingdom.
E-mail: r.k.butlin@sheffield.ac.uk

ABSTRACT

The speciation process often takes a long time. The speciation continuum framework has been useful to reconstruct the evolutionary processes that result in the formation of new species but defining when this continuum starts is far from trivial. Although a panmictic population is often considered the initial condition of speciation, this is unrealistic for almost all species. Local or divergent adaptation are viewed by many researchers as processes that shape intraspecific diversity and thus are not part of speciation. We propose that speciation starts when reproductive isolation becomes greater than zero, arguing in favour of the alternative view that local adaptation necessarily involves some reproductive isolation, independently of whether it results in the completion of speciation. Given that local adaptation is widespread, the consequence is that most species are constantly in the process of speciating. The process of speciation is best represented as the formation of separate subnetworks, defined by reproductive isolation, within extended and fluid spatial networks of populations.

Keywords: barriers; coupling; gene flow; population structure; reinforcement; divergent selection; speciation continuum

INTRODUCTION

The ‘speciation continuum’ is a widely used and helpful framework for considering the evolutionary processes that lead to the origin of new species by the splitting of lineages. One version envisages a single axis of reproductive isolation (Stankowski and Ravinet 2021). However, since multiple processes are involved and their impacts on evolving populations can be documented in many different ways, the continuum may best be represented as a multidimensional hypercube, rather than on a single axis. Bolnick *et al.* (2023) suggest a hypercube with axes representing any measure of progress towards speciation, including genetic and phenotypic divergence as well as reproductive isolation. Johannesson *et al.* (2024) prefer to retain the focus on reproductive isolation but argue that a hypercube is needed because no single measure fully reflects both the organismal traits contributing to isolation and the properties of the resulting barriers to gene flow. Whether considering one or many axes, there has been much debate about the end-point of the continuum: when is speciation complete (Coyne and Orr 2004, Mallet

2005, Kulmuni *et al.* 2020)? This clearly relates to the perennial problem of defining species: here we adopt the conventional view under the Biological Species Concept that the end-point is complete reproductive isolation. However, there has been much less discussion concerning the other end of the continuum: when does speciation start? Stankowski and Ravinet (2021: fig. 2) and Johannesson *et al.* (2024: fig. 1) show the continuum as running from ‘one population’ to ‘two species’. Seehausen *et al.* (2014: box 2) show the start as ‘Panmictic populations’ and the end as ‘Two irreversibly isolated species’. The y-axes in these figures vary but they indicate, in different ways, that the speciation process starts from a state with neither spatial structure nor reproductive isolation.

This starting point raises various issues that we will discuss here. First, there is a semantic issue about what constitutes ‘speciation’. The continuum approach encourages the application of this term to the whole extended process of lineage splitting, and this is the way in which we will use the term. However, this use has its disadvantages, particularly because it places many pairs of

populations as being ‘in the process of speciation’ even though they will never complete the process (e.g. [Nosil *et al.* 2009](#), [Huang 2020](#)). The term has been applied by some authors in a more restricted sense, essentially describing the completion of the process. For example, [Schemske \(2010\)](#) asks (p.S11), ‘which barriers were in place at the time of speciation, that is, when gene flow between taxa essentially ceases?’. Since completion can be hard to define and to determine empirically, this use can also be problematic. Unfortunately, some authors have used both senses of the term within the same publication. For example, [Hendry \(2009\)](#) advocates a continuum view of ‘ecological speciation’ and considers any non-zero level of reproductive isolation as part of speciation (process sense) but also sometimes uses ‘speciation’ to mean completion of the process [as in ‘progress toward (or away from) ecological speciation’; p.1385]. As with many terms in the ‘language of speciation’, it helps to be clear about the usage of terms in order to communicate effectively ([Harrison 2012](#), [Stankowski *et al.* 2024](#)). Using ‘process of speciation’ to refer to the continuum and ‘completion of speciation’ when referring to the final step in the process, rather than just ‘speciation’, would help to maintain an important distinction.

We also wish to emphasize that ‘reproductive isolation’ (however defined, see [Westram *et al.* 2022a, b](#) and associated commentaries) is also a continuous measure. To say that two populations are reproductively isolated implies some reduction in gene flow but does not specify the extent of reduction. If the intended meaning is that gene exchange has ceased, then ‘complete reproduction isolation’, or similar, should be used. We see ‘divergence’ between populations as a more neutral term, implying genetic or phenotypic differences but not necessarily reproductive isolation.

If speciation is viewed as an extended process, where does it start? Real species do not exist as single panmictic populations: this starting point is only reasonable as a theoretical construct. In reality, species have spatially extended distributions, almost always on scales greater than their dispersal distance and, therefore, they exhibit some level of population structure. Typically, this structure is also influenced by historical events such as changes in range size or population size, and subdivision by physical or environmental barriers to dispersal or by behaviour (e.g. homing). Therefore, panmixia is not the starting point for speciation. However, it is still possible to consider the absence of reproductive isolation as the initial condition. This works when reproductive isolation is defined as a reduction in gene flow relative to that expected in the absence of genetic differences between populations that constrain gene exchange (the definition adopted by [Stankowski and Ravinet 2021](#), and advocated by [Butlin 2022](#), [Westram *et al.* 2022a, 2022b](#), but not universally accepted). It is then possible to have population structure (divergence) without reproductive isolation. However, genetic differences among populations within species that influence gene flow may be the norm, rather than the exception. The main issue that we wish to discuss here is the conceptual relationship between non-neutral population structure and the start of the process of speciation.

LOCAL ADAPTATION

Local adaptation is the pattern where ‘resident genotypes in each deme ... have on average a higher relative fitness in their

local habitat than genotypes originating from other habitats’ ([Kawecki and Ebert 2004](#): p.1225). Given variation in habitat features on a scale that is large enough relative to the dispersal distance of an organism ([Slatkin 1973](#)), this pattern is the predicted result of divergent selection and it is expected to arise rapidly following population establishment. [Hereford \(2009\)](#) found local adaptation to be common (>70% of studies) and strong (>40% fitness advantage to native populations over non-native populations). The studies surveyed were likely to be biased towards species with a strong a priori expectation of local adaptation, which might explain these high values. However, [Dittmar and Schemske \(2023\)](#) considered the Hereford survey to show less local adaptation than expected and certainly some cases will be missed due to limited power to detect small fitness effects.

Hereford’s results came from reciprocal transplant experiments but the patterns known as ‘isolation by environment’ ([Wang & Summers, 2009](#)) ([Wang and Summers, 2009](#)) or ‘isolation by adaptation’ ([Nosil *et al.* 2008](#)) point in the same direction. These patterns reveal greater genetic differentiation between populations in different environments or with different phenotypes, respectively, than expected from isolation-by-distance alone. This is interpreted as a result of lower effective gene exchange caused by divergent selection and local adaptation. The patterns appear to be widespread ([Nosil 2012](#), [Bagley *et al.* 2023](#)). Together, these observations suggest that some fitness advantage to resident populations is likely to be very common. This is consistent with ubiquitous variation in environmental variables, populations that are not too small (so that selection is effective relative to drift), and spatial extents that are large enough relative to dispersal for the impact of gene flow to be weak.

In our view, local adaptation necessarily implies reproductive isolation (see below for justification). However, this does not seem to have been universally accepted in either the speciation or the local adaptation literature. Classic models of sympatric speciation did not treat disruptive selection as part of reproductive isolation ([Maynard Smith 1966](#), [Felsenstein 1981](#), as noted by [Nosil *et al.* 2005](#)). [Schemske’s \(2010\)](#) discussion of the history of views on the role of adaptation in speciation suggests that the controversy goes back to Darwin: Darwin’s own view of speciation was centred on what we would now call divergent selection and local adaptation, but he was criticised for his ‘neglect of reproductive isolation’ and for leaving the speciation problem unsolved ([Schemske 2010](#), citing [Mayr 1982](#) and [Coyne and Orr 2004](#)). The implication is that there is no necessary link between local adaptation and reproductive isolation. This implication is also present in Schluter’s classic paper on ecological speciation ([Schluter 2001](#)), or at least there is some ambiguity. For example, in his opening definition:

‘ECOLOGICAL SPECIATION ... occurs when DIVERGENT SELECTION on traits between populations or subpopulations in contrasting environments leads directly or indirectly to the evolution of REPRODUCTIVE ISOLATION.’ [upper case in original; p.372]

or later:

‘Demonstrating a role for divergent selection in speciation, however, is only the first step to detecting an ecological

speciation event. The next step is to understand the process by which divergent selection has led to the evolution of reproductive isolation (e.g. by-product alone or with reinforcement, mechanisms of hybrid fitness, etc.).' [p.375]

It is unclear whether the author intended to advocate a separation between local adaptation and reproductive isolation, but both extracts can be read as suggesting that there can be a response to divergent selection, i.e. that there can be local adaptation, without the evolution of reproductive isolation. This lack of clarity has persisted. For example, [Hendry \(2009\)](#) lists ways in which adaptive divergence restricts gene flow (which he equates with reproductive isolation) and yet argues that evidence for ecological speciation requires that 'one must **also** demonstrate that this adaptive divergence has contributed to the evolution of reproductive isolation' [our emphasis; p.1385]. The current literature contains comments that suggest the same separation. To take just one recent example, ([Freedman et al. 2023](#)) say [p.2296], '... implies that reproductive isolation has evolved in response to local adaptation ...'. We have used this sort of equivocal wording in our own work too! For example, 'Nowadays, it is widely recognized that local adaptation to divergent environmental conditions (e.g. different habitats) can result in the formation of distinct morphs or ecotypes and in some instances lead to the emergence of reproductive barriers' ([Carvalho et al. 2016](#): p.503).

Others clearly recognize that it is not possible to have one without the other (for example, [Feder et al. 2012](#) treat 'direct selection' as the first stage of speciation). As soon as there is local adaptation, selection operates against migrants (or migrant alleles) arriving in a population from any population in a different habitat ([Fig. 1](#)). This has clearly been recognized in the case of the component of reproductive isolation dubbed 'immigrant inviability' by [Nosil et al. \(2005\)](#). This may include components of fitness other than viability (e.g. fertilization success; [Svensson et al. 2017](#)) and there need only be a fitness reduction, not necessarily complete inviability or infertility. It is possible that selection does not act against migrants themselves but instead against the maladapted alleles that they introduce (for example when fitness differences are concentrated in juveniles but it is adults that migrate between habitats). In this case, there is no 'immigrant inviability' but, nevertheless, selection against either migrant individuals or alleles clearly reduces gene flow relative to that expected in the absence of genetic differences between populations (m) and, therefore, fits the broad [Westram et al. \(2022a\)](#) definition of reproductive isolation. The barrier to gene flow that it creates is likely to be concentrated around loci that contribute to adaptive divergence. If these are few and scattered in the genome, and their effects are not extremely large, then the reduction in gene flow at unlinked neutral loci (reflecting 'effective migration', m_e) will be small ([Barton and Bengtsson 1986](#), [Feder and Nosil 2010](#)) but there must still be some reproductive isolation (RI), even according to the 'genetic' definition of [Westram et al. \(2022a\)](#); where $RI = 1 - [m_e/m]$). Indeed, the effect on unlinked neutral loci might remain small with stronger local adaptation and differentiation at more loci unless a threshold level of selection is reached, in relation to recombination among selected loci, permitting 'genomic coupling' ([Barton and Bengtsson 1986](#), [Nosil et al. 2021](#), and see [Dopman et al. 2023](#)

for a wider discussion of 'coupling'). This might be considered a shortcoming of placing too much emphasis on unlinked neutral loci in the definition of reproductive isolation ([Butlin 2022](#), [Mallet and Mullen 2022](#), [Moyle 2022](#)). For example, when loci contributing to local adaptation are contained within chromosomal inversions, a substantial proportion of the genome might experience a barrier to gene flow with little impact on gene flow in collinear regions ([Rieseberg 2001](#), and for recent examples see ([Le Moan et al. 2024](#), [Johannesson et al. 2024](#)).

We see the fitness costs to immigrant individuals or alleles as a component of reproductive isolation that is an inescapable consequence of local adaptation. In addition, local adaptation will often be associated with other components of reproductive isolation. It may allow the expansion of one population into habitats that cannot be occupied by the other population, generating a component of 'ecogeographic isolation' ([Sobel et al. 2010](#); and see below). While first-generation (F1) hybrids between locally adapted populations might in some cases experience heterosis, second-generation (F2 and backcross) and later-generation hybrids commonly have reduced fitness ('hybrid breakdown'). The fitness cost can be considered to have two components, one due to maladaptation (hybrid phenotypes do not fit well in either parental environment) and another due to segregation, which increases phenotypic variance and so generates a load that is experienced in all environments ('transgressive incompatibility' of [Chevin et al. 2014](#), 'intrinsic' isolation of [De Sanctis et al. 2023](#), see also [Schneemann et al. 2024](#)). These segregation effects include mismatched combinations of traits, as observed in sticklebacks ([Arnegard et al. 2014](#)). The expected magnitudes of heterosis and hybrid breakdown depend on divergence history ([Chevin et al. 2014](#), [Thompson et al. 2019](#), [De Sanctis et al. 2023](#), [Kulmuni et al. 2023](#)) but there are few scenarios where they do not result in an increase in reproductive isolation relative to that due to selection against immigrants alone.

When local adaptation occurs in the presence of gene flow, it can result in selection favouring additional forms of reproductive isolation. The classic case is reinforcement, where the production of unfit hybrid offspring selects for an increase in assortative mating or other forms of prezygotic isolation ([Servedio and Noor 2003](#)), but other cases are possible: collectively called 'adaptive coupling' by [Butlin and Smadja \(2018\)](#). Reduced migration, an example of a one-allele effect ([Felsenstein 1981](#), [Butlin et al. 2021](#)), and habitat choice (e.g. [Berner and Thibert-Plante; 2015](#)) provide examples. These processes potentially increase reproductive isolation and so move pairs of populations along the speciation continuum but this is not an inevitable progression ([Stankowski and Ravinet 2021](#)). Local adaptation may be stable ([Nosil et al. 2009](#), [Servedio and Hermisson 2020](#), [Barraclough 2024](#)) or there may be a breakdown of isolation following secondary contact or a change in the environment ([Anderson and Weir 2022](#)). The outcome may also depend on the genomic architecture of reproductive isolation, for instance whether barrier loci are located within chromosomal inversions ([Noor et al. 2001](#), [Rafajlović et al. 2021](#)).

The foregoing discussion of local adaptation largely ignored the spatial relationships among populations. In principle, adaptive divergence can evolve between two spatially congruent populations as a result of disruptive selection, but it is much more likely where populations are spatially separated ([Fig. 1](#)),

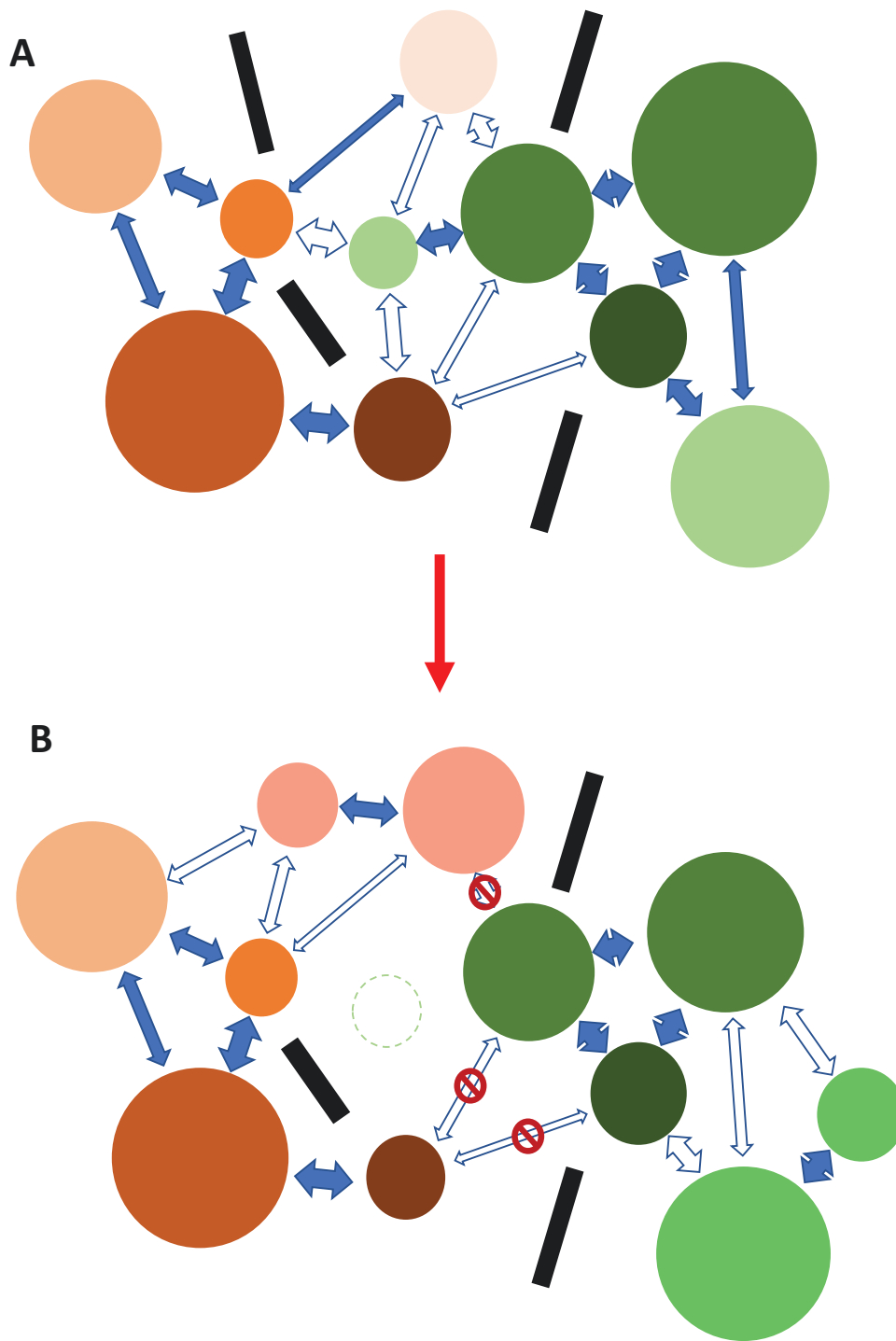


Figure 1. Representation of the speciation process in a spatial network of populations. A, local populations of a species are represented by circles and migration between them by arrows. The width of each arrow represents the migration rate, which is inversely proportional to distance. Black bars represent physical barriers to migration. Orange and green populations are adapted to different habitats. This reduces gene flow between them, relative to migration, and this is represented by the hollow arrows. Within habitats, population structure is represented by the variation in colour intensity. B, by the time reproductive isolation is complete between orange and green populations (if this happens), reproductive isolation has started to evolve among populations within each species (different hues). This occurs against a background of ongoing changes to the network of populations, with changes in population size (area of the circles), extinction (empty circle), gain of populations, and loss of physical barriers.

causing some prior reduction in gene flow (Maynard Smith 1966, Felsenstein 1981, but see also Dieckmann and Doebeli 1999, for example). Spatially separated populations can also accumulate incompatibilities by mutation-order processes

(Schluter 2009). This can occur with adaptation to similar environments but may also augment reproductive isolation due to divergent adaptation. A population might spread into a new region where it is initially not well-adapted to any local patch

and subsequently the ancestral and derived populations might diverge. Alternatively, a well-adapted population might spread into a neighbouring patch where it is initially maladapted, followed by adaptive divergence in the new habitat. In either case, local adaptation is something that follows from the occupation of a variable environment. However, it is also possible that adaptive divergence between populations favours their spread into different regions. When this happens, adaptive divergence generates an additional component of reproductive isolation that has been called ‘ecogeographic isolation’ (Sobel *et al.* 2010, Sobel and Chen 2014). This is the isolation that results from geographical separation of populations as a result of their genetic differences, rather than as a result of historical factors (which Sobel *et al.* 2010 call ‘effective geographic isolation’ and do not consider to be a component of reproductive isolation; but see Butlin 2022 for a slightly different view). Westram *et al.* (2022a) clearly consider ecogeographic isolation to be a component of reproductive isolation, although they dispute the way it should be quantified. In the context of the present discussion, the point is that ecogeographic isolation is another component of reproductive isolation that must commonly be associated with local adaptation.

CONCLUSION

Where does the speciation continuum start? One option would be to consider population structure due to local or divergent adaptation as a form of intraspecific variation and so to argue that the process of speciation does not begin until something else starts to happen. This is problematic. If one accepts, as we have argued here, that local adaptation is necessarily associated with components of reproductive isolation, then this viewpoint requires that reproductive isolation can be non-zero without speciation having begun. It would be necessary to define the start of the speciation process as the point at which other components of reproductive isolation are added to those that are direct consequences of adaptive divergence. This distinction is difficult, if not impossible, because both incompatibilities and components of prezygotic isolation can be by-products of adaptive divergence or can evolve in response to selection pressures generated by adaptive divergence (by-product and adaptive coupling in Butlin and Smadja 2018).

The alternative is to accept that the speciation process starts when reproductive isolation is greater than zero (Fig. 1). Local adaptation is then a part of the speciation process regardless of whether it will ultimately lead to the completion of speciation. This seems to us to be less problematic conceptually. The major implication, given that local adaptation is widespread, is that many, perhaps most, species contain populations that are in the process of speciating. Indeed, many species will have started speciating before separation from their sister species is complete (Fig. 1). Most of these speciation processes will not go to completion. This view is actually fully compatible with estimates of the rate and duration of speciation. In their classic survey, Coyne and Orr (2004) found a range of speciation intervals (the time between successive branching events that give rise to persistent species) from 0.08 to 5.5 Myr. They also found the duration of speciation (time to evolve complete reproductive isolation) to

range from 0.1 to 1 Myr in *Drosophila*, but much longer in some other taxa (e.g. ~10 Myr in birds). Many more, and much more sophisticated estimates are now available (e.g. Rabosky 2016) but the key point is unchanged: the ranges of these two estimates overlap broadly, even within clades, and this implies that incomplete reproductive isolation among populations must be a common feature of within-species variation. Others have emphasized similar conclusions. In particular, Harvey *et al.* (2019) argued that the formation of ‘isolated populations’ within species is common, with many of these populations either going extinct or re-fusing with other populations rather than continuing to diverge and completing the speciation process.

Why is this important? Many speciation researchers are interested in understanding why local adaptation leads towards complete reproductive isolation in some cases and not in (many?) others. However, speciation studies are often biased towards taxa already presenting a substantial degree of reproductive isolation (Faria *et al.* 2014). With this narrow focus, it is difficult to have a comprehensive understanding of why the process of speciation is not completed in some cases, and to identify key evolutionary events within species that ultimately result in new species. Since maintaining intraspecific diversity is key for the formation of new species, this has direct consequences in conservation biology. Focusing conservation efforts on networks of more or less reproductively isolated populations within species will be key for maintaining the evolutionary processes that lead to the evolution of new species.

In summary, it is not helpful to think of the speciation continuum as starting with a single population or with panmixia. Local adaptation is a common feature of population structure within species that is necessarily associated with reductions in gene flow between populations and so represents a component of reproductive isolation rather than something separate from reproductive isolation. Putting these two ideas together results in a more realistic view of speciation as an extended process in which patterns of reproductive isolation among populations in spatial networks change continuously (Fig. 1), occasionally generating completely isolated sub-networks that we call species.

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CONFLICT OF INTEREST

The authors have no conflicts of interest to declare.

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DATA AVAILABILITY

NA.

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