Searching for sympatric speciation in the genomic era

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Abstract

Sympatric speciation illustrates how natural and sexual selection may create new species in isolation without geographic barriers. However, recent genomic reanalyses of classic examples of sympatric speciation have revealed complex histories of secondary gene flow. Thus, there is a need to revisit how to connect the diverse theoretical models of sympatric speciation and their predictions to empirical case studies in the face of widespread gene flow. We summarize theoretical differences between sympatric speciation and speciation-with-gene-flow models and propose genomic analyses for distinguishing which models apply to case studies based on the timing and function of adaptive introgression. Investigating whether secondary gene flow contributed to reproductive isolation is necessary to test whether predictions of theory are ultimately borne out in nature.

What is sympatric speciation?

As Mayr famously quipped, sympatric speciation is like the Lernean Hydra: "which grew two new heads whenever one of its heads was cut off" (p. 451; [1]). The latest incarnation of this phenomenon has occurred over the past decade: sympatric speciation now means two different things to different research groups.

In the classic definition, sympatric speciation represents the most extreme endpoint on the divergence with gene flow continuum: panmictic gene flow and no initial divergence at the start of speciation [2–4]. In the context of theoretical speciation models, this classic process of sympatric speciation is the most difficult because the starting conditions involve no pre-existing divergence, potentially tied with physical linkage, among loci involved in reproductive isolation. Instead, **linkage disequilibrium** (see Glossary) must build up through time within a population through the action of disruptive natural selection and strong assortative mating by ecotype, despite the countervailing eroding force of recombination [5–8].

Recently, the definition of sympatric speciation has been expanded to focus more on biogeographical context [9], in which the speciation process is defined as sympatric a) as long as diverging populations are within 'cruising range' of each other and b) regardless of whether secondary gene flow provided alleles contributing to reproductive isolating barriers in sympatry. Cruising range provides a practical empirical definition of gene flow between diverging sympatric populations, allowing for some geographic or microallopatric population structure (e.g. [10,11]). However, allowing for secondary gene flow of alleles contributing some or all of the reproductive isolation between sympatric populations expands the definition of sympatric speciation to include both hard and easy processes under one umbrella. Theoretical models show

69 that speciation is much easier from starting conditions that involve some level of initial
70 divergence and/or restricted gene flow, for example, if the alleles necessary for reproductive
71 isolation first become physically linked in allopatry [12].

72 These two definitions, the classic theoretical population genetic and the recent
73 biogeographic, reflect different perspectives on the value of studying sympatric speciation. The
74 biogeographic definition, with its broader range of starting conditions that are easier to verify in
75 nature, increases, perhaps vastly, the number of empirical speciation events that could be
76 categorized as examples of sympatric speciation. This definition values the frequency of
77 sympatric diverging populations in nature compared to allopatric speciation, as an estimate of the
78 overall importance of "sympatry" in contributing to biodiversity. The classic definition, with its
79 narrow set of starting conditions that remain challenging to verify in nature, finds value in
80 studying the difficult process of sympatric speciation itself. Namely it values the theoretical
81 possibility of creating new species solely through the power of divergent selection alone,
82 regardless of whether this process is common in nature. This opinion piece focuses on the types
83 of questions that genomic data now allow us to ask to improve the search for examples of the
84 latter process and understand the range of speciation mechanisms found in nature from among
85 those shown to be plausible in theory.

86 Under the biogeographical definition of sympatric speciation, there is little difference in
87 terms of the speciation mechanisms involved in scenarios that start with initial panmixia (e.g.
88 classic sympatric speciation) versus those that start with some geographic or microallopatric
89 population structure (e.g. [10,11]). This contrasts sharply with the rich theoretical literature
90 differentiating models of sympatric speciation from other models of speciation with gene flow.

Indeed, theory teaches us that the classic process of sympatric speciation (without the aid of secondary gene flow contributing to reproductive isolation) is uniquely and notoriously difficult [13], in part because quite specific conditions of resource availability (e.g., [6,14]), mating traits and preferences (e.g., [15,16]), and search costs (e.g., [17]) must be met for it to occur. Some claim that the effort to discern the exact geographic scenario and initial conditions of speciation would be better spent on finding loci involved in reproductive isolation (i.e. 'barrier loci' [18,19]). This is an important first step and we can glean something about the process of speciation from gene annotations of barrier loci and linkage architecture. However, understanding whether any one particular locus or potential mechanism was necessary for speciation often requires placing genomic discoveries in the context of speciation models that compare the importance of such processes, models whose outcomes are highly dependent on the initial conditions before sympatric divergence.

Different mechanistic processes underlying divergence in sympatry

Regardless of definition, it is necessary to distinguish among different sympatric divergence processes to understand which classes of speciation models and predictions apply to specific case studies. We here distinguish different scenarios (Fig. 1) that will result in two sister species in sympatry based on whether **secondary gene flow** aided in population divergence: 1) classic sympatric speciation without gene flow; 2) sympatric speciation in the presence of a) neutral secondary gene flow or b) after differential sorting of an ancestral **hybrid swarm**. In the latter case, it is important to distinguish whether the ancestral hybrid swarm population achieved panmixia before later divergence (i.e. sympatric divergence); otherwise, differential sorting of

113 haplotypes within the hybrid swarm is better described by secondary contact speciation with
114 gene flow models rather than sympatric speciation models. 3) Speciation may also be aided by
115 secondary gene flow that a) triggers initial sympatric divergence or b) increases divergence after
116 initial divergence in sympatry becomes stalled, an outcome of many sympatric speciation models
117 without sufficiently strong disruptive selection [20–23]. Finally, 4) secondary contact after a
118 period of allopatry between two populations can result in coexistence or reinforcement, if there is
119 not collapse into a single admixed population [12,24–26] or extinction of one or both
120 populations. We consider scenarios 1 and 2a to be examples of classic sympatric speciation
121 models, the 'hard' process, whereas scenarios 3 and 4 would be examples of speciation aided by
122 secondary gene flow, a much easier process in theory. Interestingly, hybrid swarm scenarios (2b)
123 exist in a gray area, since substantial initial gene flow from multiple sources may increase
124 ecological or preference variation within a population that is sufficient to trigger later sympatric
125 divergence, even without segregating inversions or genetic incompatibilities [27–29]. So far, we
126 know of no examples of scenario 1 within any case study of sympatric sister species examined
127 using genomic tools; even long diverged species show some evidence of **introgression** in their
128 past (e.g. [30]). In contrast, sympatric speciation with neutral gene flow (Scenario 2a, and
129 conditionally Scenario 2b) and speciation aided by gene flow (Scenarios 3 and 4) frequently
130 appear to operate concurrently even within a single sympatric adaptive radiation (e.g. [31–33]).
131 It is important to distinguish these scenarios because theoretical models predict that
132 sympatric divergence unaided by any form of secondary gene flow is substantially more difficult
133 than other speciation with gene flow scenarios (Box 1). Gene flow throughout the speciation
134 process allows recombination to break down linkage disequilibrium among alleles associated

135 with ecological divergence and assortative mating. There are actually three different types of
136 sympatric speciation models to consider: the most difficult process involves independently
137 segregating loci for ecotype, female preferences, and male traits within the population, whereas
138 sympatric divergence is much easier if any of these three types of traits are combined, such as
139 assortative mating based on phenotype matching instead of separate loci for preference and traits
140 [34,35] or "magic" traits (such as assortative mating based on microhabitat preference; [6,7,36]).
141 Sympatric speciation by sexual selection alone is also theoretically possible (albeit considered
142 highly unlikely) if there is substantial preference variation either initially within the population
143 or through secondary gene flow [16,27].

144 Any form of linkage disequilibrium among ecological and mate choice loci formed in
145 allopatry, whether due to physical linkage, selection, or drift, can tend to shift the initial starting
146 conditions of panmixia in favor of sympatric divergence [24]. However, linkage disequilibrium
147 without physical linkage subsides within a relatively small number of generations after
148 secondary sympatry and thus may not allow sufficient time for the evolution of assortative
149 mating within the population. In contrast, pre-existing physical linkage among ecological loci
150 has been shown to increase the probability of divergence, especially when it captures already
151 divergent alleles, as is more likely after a period of divergence in allopatry before secondary
152 contact [37,38]. Similarly, physical linkage can cause preference and trait alleles to mimic
153 phenotype matching, although even tight linkage can break down over long timescales (shown in
154 a model with population structure: [39]). Segregating inversions in the ancestral population are
155 now well-known empirical examples of physical linkage promoting divergence in sympatry

156 [37,40]. Sympatric divergence is also limited by many other restrictive conditions regarding the
157 costs of female choosiness and strengths of disruptive selection and assortative mating.
158 Despite extensive searches for examples of sympatric speciation in the wild, there are few
159 convincing case studies due to the difficulty of ruling out historical allopatric scenarios (see
160 below) and the new difficulty of ruling out a role of introgression in speciation. Furthermore, the
161 role of magic traits or matching vs. preference/trait mechanisms is not fully understood in any
162 existing case study. Thus, we still have very limited empirical tests of an extensive theoretical
163 literature and diverse competing models of the notoriously difficult process of sympatric
164 speciation [34,35,41,42].
165

166 **The classic problem of sympatric speciation**

167 There are four traditional criteria for demonstrating classic sympatric speciation (Scenario 1): 1)
168 sister species which are reproductively isolated, 2) form a **monophyletic** group, 3) largely
169 overlap in ranges, and 4) have biogeographic and evolutionary histories that make periods of
170 allopatric divergence highly unlikely [43]. Very few case studies have been able to meet these
171 rigorous criteria despite intensive searches [4,43]. This has led to the prominent status of crater
172 lake cichlid radiations as some of the best examples of sympatric speciation in the wild due to
173 the uniform shape of isolated volcanic lakes which convincingly rule out phases of allopatry due
174 to water level changes (Box 2; [44]).
175 The monophyly criterion assumes that monophyly arises only when a single ancestral
176 population underlies the present-day daughter species. This is typically met by inferring a single
177 phylogeny from one or more loci. This single point-estimate view of evolutionary history is

178 problematic because it obscures the presence of non-bifurcating relationships among organisms
179 (e.g. sister species which derived ancestry from multiple source populations due to extensive
180 gene flow or hybrid speciation) and the real variation in evolutionary histories among genes
181 across the genome itself (e.g. [45]). Few regions of the genome may initially contribute to
182 reproductive isolation resulting in a heterogeneous genomic landscape of differentiation among
183 incipient species [46], a pattern now extensively supported across case studies (e.g. [47–49]).
184 Therefore, monophyletic relationships are consistent with, but not exclusive to a scenario of
185 sympatric speciation. Examining heterogeneous evolutionary histories across regions relevant to
186 speciation is thus crucial for understanding the processes and conditions under which sympatric
187 divergence can occur.
188

189 **The 'new' problem of sympatric speciation**

190 While genomics has increased our ability to resolve evolutionary relationships among organisms,
191 it has also revealed more complex evolutionary histories of multiple colonizations and extensive
192 secondary gene flow in nearly all examples of sympatric speciation that have been examined
193 with genomic data so far ([50–59]; e.g. to our knowledge Lord Howe Island palms and
194 indigobirds have not yet been directly examined for secondary gene flow with an outgroup).
195 Indeed, only a handful of genes may directly contribute to the speciation process whereas the rest
196 of the genome is porous to gene flow while reproductive isolation is incomplete [46,60].
197 Examples of sympatric speciation without secondary gene flow (Scenario 1) are now even rarer
198 after applying modern genomic tools to search for introgression. Instead, it is still possible that
199 sympatric speciation occurs in the face of secondary gene flow in nearly all these examples

200 (Scenario 2; [56]). Importantly, most evidence of secondary gene flow impacting putative
201 examples of sympatric speciation comes from genome-wide tests of introgression from outgroup
202 lineages that do not look at how that secondary gene flow has impacted gene flow between
203 diverging populations in sympatry (e.g. [53,54]). It is also possible in cases studies of sympatric
204 speciation that involve radiations of species, that secondary gene flow impacted some, but not all
205 of the divergence events such that some species within a radiation may better represent classic
206 sympatric speciation scenarios than others. Therefore, introgression detected at the genome-wide
207 level from lineages outside the speciation event tells us that secondary gene flow has occurred,
208 but little about the divergence process among incipient sympatric species and how that gene flow
209 shaped the process of speciation.

210 The challenge of understanding the hard process of classic sympatric speciation in the
211 genomic era is establishing or rejecting a functional role for the ubiquitous secondary gene flow
212 present during the speciation process, in effect ruling out scenarios 3 and 4 in favor of scenario 2
213 (Fig. 1). Even if signatures of secondary gene flow are detected, speciation could still have
214 occurred solely via mechanisms of sympatric speciation, in the classical sense, if that secondary
215 gene flow did not play a causal role in divergence (Scenario 2a, conditionally Scenario 2b). In
216 contrast, secondary gene flow could play a causal role if it introduced novel genetic variation or
217 physically linked alleles (e.g. a segregating inversion) that promoted speciation before the start
218 of divergence, through mechanisms such as inflating variance through the creation of a hybrid
219 swarm **(**Scenario 2b**;** ([28,29,32,61]), adaptive introgression (Scenario 3; [62–64]), **transgressive**
220 **segregation** (Scenarios 2-3; [65,66]), or hybrid speciation ([67]). Here we propose and discuss
221 genomic analyses that may help to establish or reject a functional role of secondary gene flow in

222 the speciation process (Fig. 1). This is necessary to identify putative cases of the classic
223 sympatric speciation process when gene flow appears to be nearly universal in the wild,
224 particularly among sympatric diverging populations.
225
226 **Searching for sympatric speciation in the genomic era**
227 Although genome-wide analyses of introgression provide a starting point, ultimately
228 consideration of the time of arrival and functional role of each introgressed region within extant
229 sympatric sister species pairs will be necessary to distinguish between the classic 'hard' process
230 of sympatric speciation in which incidental gene flow does not contribute to reproductive
231 isolating barriers (Scenario 2a) or speciation aided by secondary gene flow (Scenario 3; e.g.
232 segregating inversions [68,69] or balancing selection on regions containing multiple barrier loci
233 [70,71]). We suggest four major types of genomic analyses to address questions about the role of
234 secondary gene flow and identify sympatric speciation with gene flow: analyses to1) estimate the
235 timing of introgression into sympatric sister species relative to their divergence time, 2) infer the
236 presence and timing of selective sweeps within sympatric sister species, 3) annotate candidate
237 adaptive introgression regions for functional elements or trait associations that may be relevant
238 to speciation, and 4) if closely related non-speciating outgroups are available, confirm the lack of
239 selective sweeps of these regions in outgroups. These analyses are by no means trivial, but
240 recently developed methods have made it possible to start addressing such challenging questions.
241 The geographic context and breeding locations of populations are also needed to sort
242 classic sympatric divergence from parapatric or microallopatric divergence scenarios. In
243 particular, sympatric taxa which segregate in different microhabitats, such as different depths or

244 host types, exhibit a type of automatic magic trait (i.e. 'mate where you eat') and belong to the
245 easiest class of sympatric speciation models if these microallopatric traits diverged in sympatry
246 [68,72]. Combining geographic and natural history context with population genetic statistics will
247 aid in distinguishing where case studies fall along the speciation with gene flow continuum and
248 whether the starting conditions of panmixia in sympatric speciation models will apply (Fig. 1).
249
250 *1) Is the observed secondary gene flow concurrent with divergence times?*
251 Estimating the duration of gene flow and the timing of introgression into the sister species from
252 an outgroup relative to the timing of divergence between sympatric sister species will help
253 distinguish between scenarios of sympatric speciation, speciation with gene flow, and secondary
254 contact. If populations diverged in sympatry independent of any concurrent secondary gene flow
255 (Scenario 2), we might expect to see weak concordance of the timing of gene flow with
256 divergence times among species, for example discrete gene flow events that date well before or
257 after divergence times among species (Fig 1A). In the case of both discrete gene flow events
258 surrounding divergence time estimates or continuous gene flow from the time of colonization to
259 the present, more information about function and selection on regions introgressed near the time
260 of speciation will be needed. Increasingly sophisticated approaches for detecting fine-scale
261 patterns of introgression and inferring the timing and duration of gene flow from genomic data
262 are becoming available (Box 3).
263
264 *2) Are any of the introgressed regions under selective sweeps and does the timing of these*
265 *sweeps align with species divergence time?*

266 We can use information about selective sweeps of introgressed variation to further characterize
267 the role of secondary gene flow in sympatric divergence. When an allele is selectively favored in
268 a population, positive selection may cause it to increase in frequency and form a localized
269 selective sweep of reduced genetic variation surrounding the adaptive variant [73]. Such regions
270 of high differentiation in recently diverged species are often targeted as candidates for speciation
271 genes, although other processes not directly associated with speciation can lead to similar
272 patterns of high heterogeneity in differentiation across a genome (reviewed in [19,74,75]). If
273 speciation was recent or ongoing, there may be strong signatures of a selective sweep for
274 particular haplotypes in at least one of the sister species for regions involved in the divergence
275 process (Fig. 1B). If secondary gene flow was neutral with respect to speciation, we may find no
276 signatures of selective sweeps in those introgressed regions.

277 Importantly, a sweep of the same introgressed region in both sympatric sister species may
278 be interpreted as adaptation to the same new environment, which may not contribute to
279 reproductive isolation between the pair (dependent on their respective genetic backgrounds; e.g.
280 [76,77]). However, this pattern is also consistent with the sweep of a region contributing to a
281 'one-allele' mechanism of mate choice [5,34,35], such as increased female choosiness in both
282 sympatric sister species (e.g. [78]), which *would* contribute to reproductive isolation. Thus,
283 selective sweeps of an introgressed region in both sympatric sister species do not rule out its role
284 in aiding the speciation process.

285 Alternatively, if selective sweeps are detected, the timing of selective sweeps can give
286 indirect evidence about their role in speciation. If the timing of introgression predates the timing
287 of the selective sweep, it is challenging to infer the importance of an introgressed region for

288 speciation because linkage disequilibrium among loci relevant to speciation may take time to
289 build up. However, the absence of selective sweeps or introgression until long after species
290 divergence would suggest that introgression was not relevant to speciation (Scenario 2a).
291
292 *3) Is there support for a causal role of secondary gene flow based on functional genetic analyses*
293 *of variants in the region?*
294 Another potential source of evidence for the functional importance of gene flow can come from
295 genome-wide association studies (GWAS) between variants in introgressed regions and traits
296 involved in ecological or sexual isolation between sister species. The conservation of sequences
297 within introgressed regions across taxa may also provide strong evidence of a functional role
298 (e.g. PhastCons [79]). However, many complex traits are driven by a large number of variants of
299 small effect and ruling out a functional role for gene flow from gene annotations is difficult (e.g.
300 see the omnigenic model; [80]). Finally, and most powerfully, genome editing and gene
301 expression reporter systems are increasingly tractable in non-model systems (e.g. [81,82]). This
302 is ultimately an asymmetric problem: finding evidence that an introgressed region may have
303 contributed to reproductive isolation is far easier than demonstrating that no introgressed regions
304 contributed to reproductive isolation in any way [56]. Finding evidence for sympatric speciation
305 in the wild is now the difficult problem of functional genetic analyses of introgressed regions.
306
307 *4) Are there similar patterns of selection or divergence in the introgessed regions in closely*
308 *related outgroup populations?*

309 Thorough investigation of these same regions in outgroups to the sympatric species gives added
310 power to distinguish whether secondary gene flow aided sympatric divergence. If non-
311 diversifying, closely related species exist in similar environments and haven't diversified in a
312 similar manner but share signatures of selective sweeps in the same regions, then the observed
313 introgression may have been neutral relative to speciation, e.g. due to adaptations to shared
314 changes in climate or pathogens or shared regions of reduced recombination or increased
315 background selection. Similarly, several studies comparing genomic landscapes of differentiation
316 across closely related taxa have found that high differentiation observed in the same genomic
317 regions across taxa reflects the action of linked selection across low-recombination regions rather
318 than selection against gene flow at barrier loci [83–86].
319

320 **Concluding Remarks**

321 Sympatric speciation remains among the most controversial evolutionary processes, beloved by
322 theorists and long sought after by empiricists. While evidence of divergence under the
323 biogeographic definition of sympatry is mounting using traditional genetic criteria of monophyly
324 [4], genomic data has now revealed the ubiquity of secondary gene flow and introgression in
325 these many of these examples. Future fine-scale investigations of introgression will likely
326 continue to paint a complex picture of the role of secondary gene flow in speciation. Ruling out a
327 role for secondary gene flow in speciation and discerning which putative cases studies meet the
328 classic definition of sympatric speciation in the wild will be a formidable task, yet a worthwhile
329 one in its revelation of the sheer power of divergent selection to create species in nature.
330 Nearly all existing case studies of sympatric speciation involve some form of automatic
331 magic trait, such as assortative mating by habitat [68,87,88], along a depth gradient [52], or

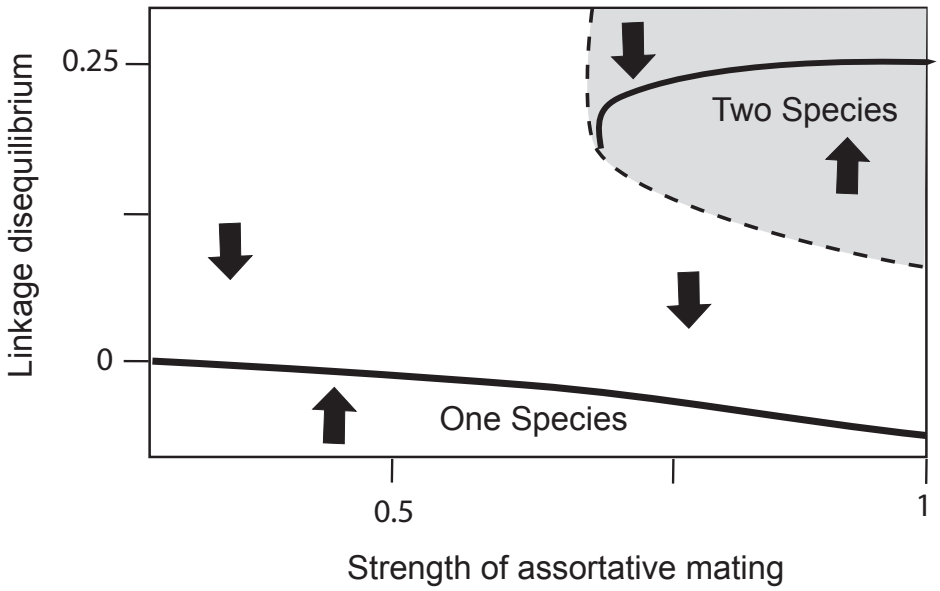
environment-induced phenology shifts [89]. We think an outstanding remaining question is whether the classic 'hard' process of sympatric speciation occurs in nature without the aid of some form of magic trait, as originally demonstrated to be possible in theory [6]. The highly polygenic and multi-dimensional nature of adaptation and mate choice suggests that an 'all-of-the-above' speciation scenario containing a mix of preference/trait, magic trait, and phenotype matching, each spread across a wide distribution of allelic effect sizes with varying times of arrival, will be the norm in nature. In contrast, although numerous and diverse, most speciation models continue to address these mechanisms in a piecemeal fashion with an assumption of large effect alleles. It remains unclear how different mechanisms, effect sizes, and times of arrival will interact and compete within a single model.

***Box 1*. Why do we care whether speciation is sympatric?**

Inferences from theoretical models predict that, under a scenario of speciation with gene flow (Scenario 3), introgression can make the process of speciation much easier in three ways. First, by introducing additional variation in ecological traits into the population, introgression could potentially facilitate a branching process due to competition for resources (although we are not aware of a model that assesses this precise situation, it can be inferred from the dynamics of [6]). Second, introgression of novel alleles for mating preferences may provide a boost in preference variation that could be an important trigger to aid the evolution of assortative mating

355 under a preference/trait mechanism, which requires preference variation to be large ([16,27]).
356 For example, we found evidence of secondary gene flow of olfactory alleles shortly before the
357 rapid divergence of a Cameroon cichlid radiation in Lake Ejagham, which may have boosted
358 preference variation [55]. Third, secondary sympatry may lead to increased linkage
359 disequilibrium between assortative mating and ecological loci or among ecological loci. It seems
360 logical that this might facilitate sympatric speciation as this metric is often described as progress
361 along the speciation continuum. However, initial linkage disequilibrium has been shown not to
362 matter much in at least some scenarios [5] because without physical linkage, linkage
363 disequilibrium breaks down quickly. However, physical linkage may enable these alleles to
364 remain in association for a sufficient time for assortative mating to evolve within the population
365 (e.g., [39]). Initial linkage disequilibrium may also increase the probability of allelic capture by
366 an inversion or for selection for new mutations within the inversion that may affect both ecology
367 and assortment [37]. Finally, higher linkage disequilibrium among ecological loci may in some
368 cases increase the probability of sympatric divergence, but this is in effect similar to varying
369 effect sizes of alleles at ecological loci (e.g. many small effect alleles within a region resemble a
370 large-effect locus [90]). These predictions could also apply to sympatric radiations. For example,
371 some classic sympatric speciation models (e.g. [6]) can yield many more that just two species if
372 left to run for more generations [14,22].
373 The fundamental difference between sympatric speciation and speciation with gene flow,
374 including secondary contact scenarios, lies in the fact that very often multiple equilibrium states
375 exist in speciation models, such that loss of divergence and maintenance of divergence in the
376 presence of gene flow are both possible outcomes, depending on the starting conditions of a

377 population (this is nicely illustrated for one measure of divergence by [24], Fig. I). In such
378 cases, speciation is much more easily reached from starting conditions that match those of two
379 populations that have diverged largely in allopatry due to the large amount of allelic variation or
380 pre-existing phenotypic bimodality and assortative mating. Even for scenarios of speciation with
381 gene flow that are much easier, such as geographic separation between two incipient species that
382 are undergoing gene flow, differentiation is much more difficult to reach or maintain from an
383 initially homogeneous population than from an initially differentiated one [91,92].
384
385 **Fig. I**



386
387
388 ***Fig I.*** *Two equilibrium cases exist for the linkage disequilibrium (LD), a proxy for*
389 *differentiation into two distinct "species" in this proof-of-concept model, that can be maintained*
390 *between two loci that are under disruptive selection and determine assortative mating. With*

little initial LD, the one-species equilibrium is likely to be reached even when the intensity of assortment is high. When LD in the traits is initially large, as can be the case if there is initially divergence in allopatry, the two-species equilibrium can be reached instead. Modified from [24].

Box 2. **Evidence for sympatric speciation from crater lake cichlid radiations**

There are relatively few volcanic chains of crater lakes containing fishes in the tropics, notably found in Cameroon, Nicaragua, Tanzania, Uganda, Madagascar, and Papua New Guinea [52,93,94]. Although sympatric radiations of endemic fishes are known from other isolated saline, alkali, postglacial, and ancient lakes, only four lineages of cichlids have radiated in the world's crater lakes (Fig. II). The most diverse radiation is Barombi Mbo, Cameroon with eleven endemic cichlid species, followed by Lake Bermin, Cameroon with nine [95]. Nicaraguan crater lakes reach up to five species [96], the East African craters never exceed two sympatric species [52,57], and Madagascar's crater lakes contain a single endemic cichlid [94]. It remains unknown why regional and lineage diversity varies so greatly because there appears to be no relationship between the occurrence of endemic cichlid radiations and crater lake size or age (up to approximately 5 km diameter and 2 million years old) [97].

The evidence for secondary gene flow is remarkably similar across all crater lake cichlid radiations examined with genomic data so far. Admixture proportions with outgroups are frequently detected within the range of 1-4%: 0.6% in Barombi Mbo *Sarotherodon* (percentage of polyphyletic trees in *Saguaro*: [56]), 1.1% in Massoko *Astatotilapia* (Patterson's *D*: [52]), 4.3% in Apoyo *Amphilophus* (demographic model:[54]), and 4.4% in Ejagham *Coptodon* (1,138

413 f_D outliers: [98]). No case studies have yet found evidence of substantial divergence in allopatry
414 followed by secondary contact (but see Lake Xiloá *Amphilophus* [54]). Instead, nearly all studies
415 have concluded sympatric divergence with periodic or continuous gene flow, potentially from an
416 initial hybrid swarm population (i.e. introgression from multiple outgroup populations).

417 Secondary gene flow may have triggered sympatric divergence in a radiation of three
418 *Coptodon* cichlids in Lake Ejagham: demographic analyses of whole genomes suggest that this
419 population did not diversify for 8,000 years despite frequent gene flow until an influx of
420 olfactory receptor alleles 1,000 years ago, coinciding with the first sympatric divergence in the
421 lake [98]. Similarly in Lake Victoria, segregating opsin alleles in riverine cichlid populations
422 were differentially sorted among Lake Victorian cichlids and may have triggered their
423 diversification [32].

424 Evidence for sympatric divergence in crater lake cichlids without the aid of secondary
425 gene flow remains elusive. Malinsky et al. [52] showed that 1.1% introgression occurred long
426 before the divergence of a shallow/deep-water sister species pair of cichlids in Lake Massoko,
427 Tanzania; however, this initial introgression may have aided later sympatric divergence (which
428 admittedly is very difficult to rule out). Very recent sympatric divergence in some crater lakes or
429 the proliferation of many species from a few colonization events may also suggest that
430 divergence occurred in sympatry without the aid of gene flow [99,100]; however, in the former
431 case it remains unclear if incipient divergence will become stalled as in other sympatric
432 radiations [31]. Very rare secondary gene flow into the Barombi Mbo cichlid radiation (0.6%
433 introgression) without a clear functional role provides weak evidence of sympatric divergence,
434 but more functional characterization and timing of introgression is needed [56]. The recent

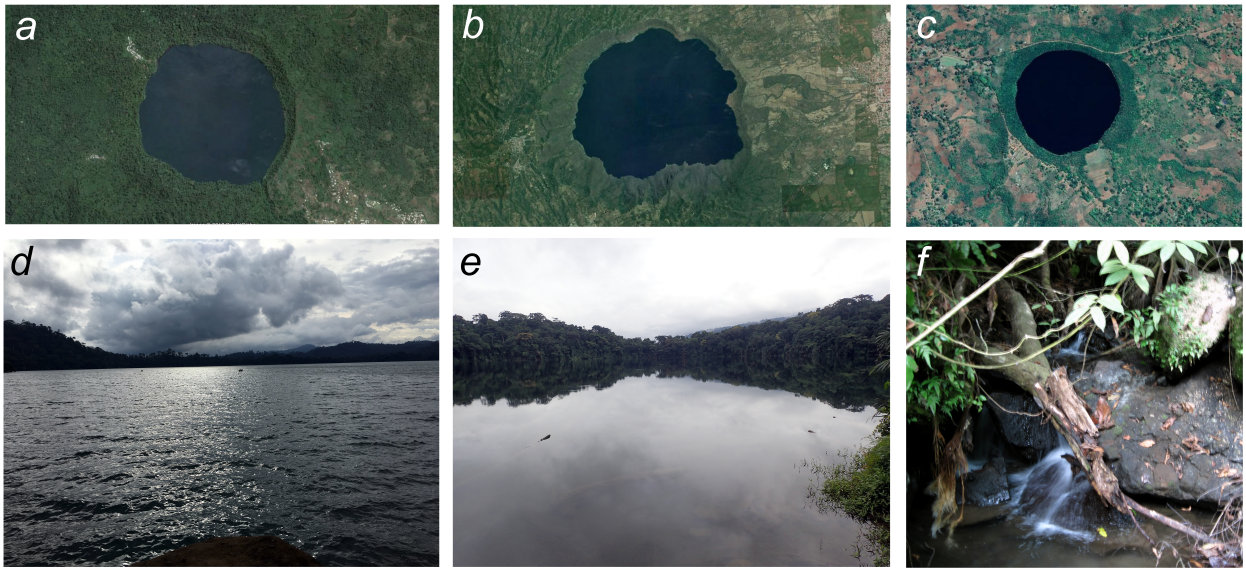
435 advent of transgenic reporters, CRISPR-Cas9, and *in situ* hybridization genetic tools within

436 Nicaraguan crater lake cichlids provides much promise for future investigations of the role of

437 introgression in sympatric divergence [81,101].

438

439 **Fig. II**



440

441 **Fig. II** Examples of volcanic crater lakes containing endemic cichlid radiations around the globe:

442 *a,d,f*) Barombi Mbo, Cameroon and its only outlet stream; *b*) Lake Apoyo, Nicaragua, *c*) Lake

443 Massoko, Tanzania, *e*) Lake Bermin, Cameroon. Satellite images (a-c) from Google Earth; (d-f)

444 by CHM.

445

446 ***Box 3.*** **Tools for detecting and timing adaptive introgression**

447 *1) Detecting and timing introgression*

448 Although there are a variety of tests to detect gene flow on a local scale or within sliding

449 genomic windows, currently three major types of demographic **coalescent** modeling approaches

450 can infer the timing of introgression based on different genomic information: 1) the distribution
451 of allele frequencies from genotype data (site frequency spectrum: e.g; [102,103]), 2) the
452 distribution of haplotype block lengths from phased genomes: e.g; [104,105], and 3) variation in
453 coalescent patterns among gene trees [106].
454
455 *2) Timing of selective sweeps*
456 Recent methods for estimating the age of a selective sweep exploit different aspects about the
457 pattern of variation surrounding the allele on its haplotypic background. These include heuristic
458 approaches that use point estimates of mean haplotype length or the number of derived mutations
459 within a chosen distance of the site [107,108], model-based approaches that use demographic
460 information and summary statistics of allele frequencies and linkage disequilibrium to model a
461 distribution of ages that fit the observed data [109–111], and full sequence approaches that
462 leverage the length of ancestral haplotypes surrounding the beneficial allele and the
463 accumulation of derived mutations [112,113].
464
465 3) *Functional analyses of introgressed variants*
466 Functional annotation of introgressed regions minimally involves searching an annotated
467 reference genome for genes with relevant functions known from model organisms. Intergenic
468 regions can be searched for evidence of strong sequence conservation across taxa [79] or
469 potential regulatory elements (reviewed in [114]). Additionally, genome wide association studies
470 (GWAS) can identify variants in introgressed regions correlated with reproductive isolating

barriers. Functional validation of gene and regulatory element variants through genome-editing experiments is also becoming increasingly tractable for non-model organisms (e.g. [81]).

Fig. 1

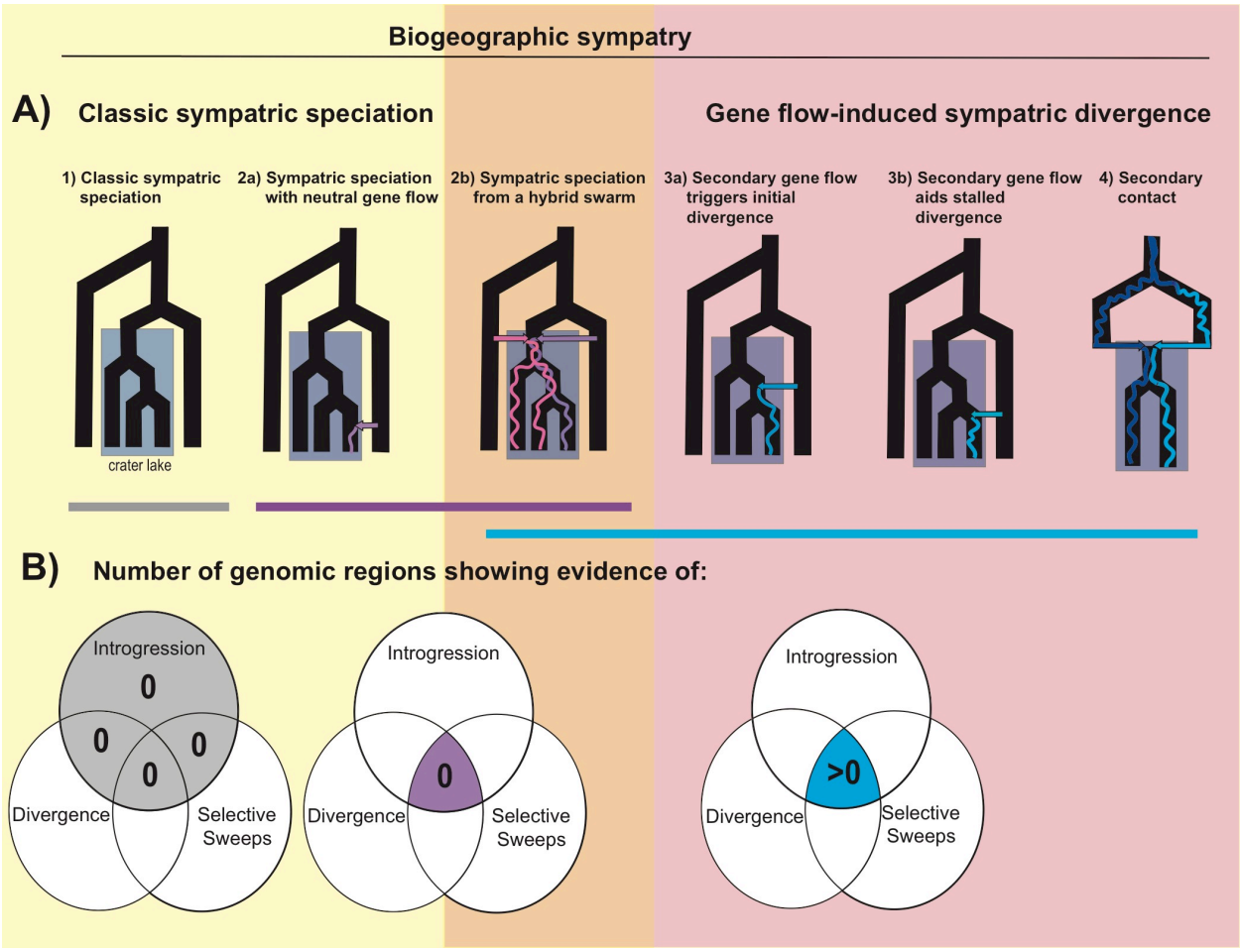


Figure 1: Genomic signatures of sympatric speciation and speciation with gene flow. Speciation scenarios are grouped into classic sympatric speciation scenarios (yellow box; harder process in theoretical models) and other divergence scenarios which can also occur under biogeographic sympatry (red box; easier processes, which we refer to broadly as sympatric divergence here). Speciation from a hybrid swarm (orange box) can fall under either class of scenarios and additional information is necessary to determine what category of speciation

482 models best describe this process. **A)** The timing of gene flow relative to divergence can be used
483 to distinguish between speciation scenarios. The colored arrows represent gene flow events and
484 the colored lines within the tree are simplified representations of a signature of introgression
485 from that gene flow event into the sympatric species. **B)** Venn diagrams illustrating the number
486 of genomic windows across the entire genome expected to have overlapping signatures of
487 introgression (e.g. f_d outliers), genetic divergence (e.g. F_{st} and D_{xy} outliers), and selective sweeps
488 (e.g. SweeD) for each speciation scenario (e.g. see [115]). The highlighted sections of the Venn
489 diagrams indicate the key signature that can be used to distinguish between the scenarios. The
490 scenarios that are expected to leave very similar signatures of overlap are grouped by the bars
491 colored with their respective Venn diagram.
492
493
494 **Glossary Box**
495
496 **Coalescence**: The event of two sampled lineages from different populations merging back in
497 time in a shared ancestral lineage.
498
499 **Hybrid swarm**: A genetically diverse population with unique allele combinations derived from
500 the hybridization of multiple distinct taxa and subsequent backcrossing with hybrids and crossing
501 between hybrids themselves.
502

503 **Introgression**: The movement and incorporation of genetic material from one distinct lineage
504 into another upon hybridization between the two and subsequent backcrossing with one of the
505 parent species.
506
507 **Linkage disequilibrium**: A non-random association of alleles at two or more loci.
508
509 **Monophyletic**: A group of lineages where the most recent ancestor of the group is not an
510 ancestor of any lineages outside the group.
511
512 **Secondary gene flow**: Any gene flow event from non-sympatric populations after the initial
513 colonization of the area that sympatric sister species diverged in. Introgression into the diverging
514 sister species following such events potentially brings in variation that has evolved in allopatry
515 that can aid the speciation process.
516
517 **Transgressive segregation**: The formation of extreme phenotypes in a segregating hybrid
518 population that are outside the range of phenotypes observed in parental species.
519
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525
526

References

1 Mayr, E. (1963) *Animal species and evolution*, Belknap.

2 Fitzpatrick, B.M. *et al.* (2009) Pattern, process and geographic modes of speciation. *J. Evol. Biol.* 22, 2342–2347

3 Fitzpatrick, B.M. *et al.* (2008) What, if anything, is sympatric speciation? *J. Evol. Biol.* 21, 1452–9

4 Bolnick, D.I. and Fitzpatrick, B.M. (2007) Sympatric speciation: models and empirical evidence. *Annu. Rev. Ecol. Evol. Syst.* 38, 459–487

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

6 Dieckmann, U. and Doebeli, M. (1999) On the origin of species by sympatric speciation. *Nature* 400, 354

7 Gavrilets, S. (2004) *Fitness landscape and origin of species*, Princeton University Press.

8 Doebeli, M. *et al.* (2005) What we have also learned: adaptive speciation is theoretically plausible. *Evolution (N. Y).* 59, 691–699

9 Mallet, J. *et al.* (2009) Space, sympatry and speciation. *J. Evol. Biol.* 22, 2332–41

10 Mallet, J. *et al.* (2009) Space, sympatry and speciation. *J. Evol. Biol.* 22, 2332–41

11 Foote, A.D. (2018) Sympatric Speciation in the Genomic Era. *Trends Ecol. Evol.* 33, 85–95

12 Otto, S.P. *et al.* (2008) Frequency-dependent selection and the evolution of assortative mating. *Genetics* 179, 2091–2112

13 Coyne, J.A. and Orr, A.H. (1989) Patterns of speciation in Drosophila. *Evolution (N. Y).* 43, 362–381

14 Polechová, J. *et al.* (2005) Speciation thrrough competition: a critical review. *Evolution (N. Y).* 59, 1194–1210

15 Norvaišas, P. and Kisdi, E. (2012) Revisiting Santa Rosalia to Unfold a Degeneracy of Classic Models of Speciation. *Am. Nat.* 180, 388–393

16 Weissing, F.J. *et al.* (2011) Adaptive speciation theory: a conceptual review. *Behav. Ecol. Sociobiol.* 65, 461–480

17 Kopp, M. and Hermisson, J. (2008) Competitive speciation and costs of choosiness. *J. Evol. Biol.* 21, 1005–1023

18 Nosil, P. and Schluter, D. (2011) The genes underlying the process of speciation. *Trends Ecol. Evol.* 26, 160–167

19 Ravinet, M. *et al.* (2017) Interpreting the genomic landscape of speciation: a road map for finding barriers to gene flow. *J. Evol. Biol.* 30, 1450–1477

20 Matessi, C. *et al.* (2001) Long-term Buildup of Reproductive Isolation Promoted by Disruptive Selection: How Far Does it Go? *Selection* 2, 41–64

21 Bolnick, D.I. and Doebeli, M. (2003) Sexual dimorphism: two sides of the same ecological coin. *Evolution (N. Y).* 57, 2433–2449

22 Bolnick, D.I. (2006) Multi-species outcomes in a common model of sympatric speciation. *J. Theor. Biol.* 241, 734–744

23 Bürger, R. and Schneider, K.A. (2006) Intraspecific Competitive Divergence and Convergence under Assortative Mating. *Am. Nat.* 167, 190–205

24 Kirkpatrick, M. and Ravigné, V. (2002) Speciation by Natural and Sexual Selection: Models and Experiments. . *Am. Nat.* 159, S22–S35

25 Turelli, M. *et al.* (2001) Theory and speciation. *Trends Ecol. Evol.* 16, 330–343

26 Garner, A.G. *et al.* (2018) Genomic signatures of reinforcement. *Genes (Basel).* 9,

27 van Doorn, G.S. and Weissing, F.J. (2001) Ecological versus Sexual Selection Models of Sympatric Speciation: A Synthesis. *Selection* 2, 17–40

28 Seehausen, O. (2004) Hybridization and adaptive radiation. *Trends Ecol. Evol.* 19, 198–207

29 Seehausen, O. (2013) Conditions when hybridization might predispose populations for adaptive

radiation. *J. Evol. Biol.* 26, 279–281
30 Turissini, D.A. and Matute, D.R. (2017) Fine scale mapping of genomic introgressions within the Drosophila yakuba clade. *PLOS Genet.* 13, e1006971
31 Martin, C.H. (2012) Weak Disruptive Selection and Incomplete Phenotypic Divergence in Two Classic Examples of Sympatric Speciation: Cameroon Crater Lake Cichlids. *Am. Nat.* 180, E90–E109
32 Meier, J.I. *et al.* (2017) Ancient hybridization fuels rapid cichlid fish adaptive radiations. *Nat. Commun.* 8, 14363
33 Wagner, C.E. and McCune, A.R. (2009) Contrasting patterns of spatial genetic structure in sympatric rock-dwelling cichlid fishes. *Evolution (N. Y).* 63, 1312–1326
34 Servedio, M.R. and Boughman, J.W. (2017) The Role of Sexual Selection in Local Adaptation and Speciation. *Annu. Rev. Ecol. Evol. Syst.* 48, annurev-ecolsys-110316-022905
35 Kopp, M. *et al.* (2017) Mechanisms of Assortative Mating in Speciation with Gene Flow: Connecting Theory and Empirical Research. *Am. Nat.* 191, 1–20
36 Servedio, M.R. *et al.* (2011) Magic traits in speciation: 'magic' but not rare? *Trends Ecol. Evol.* 26, 389–397
37 Kirkpatrick, M. and Barton, N.H. (2006) Chromosome inversions, local adaptation and speciation. *Genetics* 173, 419–434
38 Feder, J.L. and Nosil, P. (2009) Chromosomal inversions and species differences: when are genes affecting adaptive divergence and reproductive isolation expected to reside within inversions? *Evolution (N. Y).* 63, 3061–3075
39 Servedio, M. and Bürger, R. The Effects on Parapatric Divergence of Linkage between Preference and Trait Loci versus Pleiotropy. , *Genes* , 9. (2018)
40 Fuller, Z.L. *et al.* (2018) Ancestral polymorphisms explain the role of chromosomal inversions in speciation. *PLOS Genet.* 14, e1007526
41 Gavrilets, S. *et al.* (2007) Case studies and mathematical models of ecological speciation. 1. Cichlids in a crater lake. *Mol. Ecol.* 16, 2893–909
42 Gavrilets, S. (2014) Models of speciation: Where are we now? *J. Hered.* 105, 743–755
43 Coyne, J.A. and Orr, A. (2004) *Speciation*, Sutherland, MA.
44 Barluenga, M. *et al.* (2006) Sympatric speciation in Nicaraguan crater lake cichlid fish. *Nature* 439, 719–723
45 Hahn, M.W. and Nakhleh, L. (2016) Irrational exuberance for resolved species trees. *Evolution (N. Y).* 70, 7–17
46 Wu, C.-I.I. (2001) The genic view of the process of speciation. *J. Evol. Biol.* 14, 851–865
47 McGirr, J.A. and Martin, C.H. (2016) Novel candidate genes underlying extreme trophic specialization in Caribbean pupfishes. *Mol. Biol. Evol.* 34, 873–888
48 Fontaine, M.C. *et al.* (2015) Extensive introgression in a malaria vector species complex revealed by phylogenomics. *Science (80-.).* 347, 1258522–1258522
49 Campbell, C.R. *et al.* (2018) What is Speciation Genomics? The roles of ecology, gene flow, and genomic architecture in the formation of species. *Biol. J. Linn. Soc.* at <http://dx.doi.org/10.1093/biolinnean/bly063>
50 Geiger, M.F. *et al.* (2013) Crater Lake Apoyo revisited: population genetics of an emerging species flock. *PLoS One* 8, 1–17
51 Igea, J. *et al.* (2015) A comparative analysis of island floras challenges taxonomy-based biogeographical models of speciation. *Evolution (N. Y).* 69, 482–491
52 Malinsky, M. *et al.* (2015) Genomic islands of speciation separate cichlid ecomorphs in an East African crater lake. *Science (80-.).* 350, 1493–1498
53 Martin, C.H. *et al.* (2015) Complex histories of repeated gene flow in Cameroon crater lake cichlids cast doubt on one of the clearest examples of sympatric speciation. *Evolution (N. Y).* 69, 1406–1422
54 Kautt, A.F. *et al.* (2016) Multispecies Outcomes of Sympatric Speciation after Admixture with the

Source Population in Two Radiations of Nicaraguan Crater Lake Cichlids. *PLoS Genet.* 12, 1–33

55 Poelstra, J. *et al.* (2018) Speciation in sympatry with ongoing secondary gene flow and an olfactory trigger in a radiation of Cameroon cichlids. *bioRxiv* 229864,

56 Richards, E. *et al.* (2018) Don't throw out the sympatric speciation with the crater lake water: fine-scale investigation of introgression provides equivocal support for causal role of secondary gene flow in one of the clearest examples of sympatric speciation. *bioRxiv* at <http://biorxiv.org/content/early/2017/11/17/217984.abstract>

57 Machado-Schiaffino, G. *et al.* (2015) Parallel evolution in Ugandan crater lakes: Repeated evolution of limnetic body shapes in haplochromine cichlid fish. *BMC Evol. Biol.* 15,

58 Elmer, K.R. *et al.* (2014) Parallel evolution of Nicaraguan crater lake cichlid fishes via non-parallel routes. *Nat. Commun.* 5, 1–8

59 Barluenga, M. and Meyer, A. (2010) Phylogeography, colonization and population history of the Midas cichlid species complex (Amphilophus spp.) in the Nicaraguan crater lakes. *BMC Evol. Biol.* 10, 326

60 Wu, C.-I. and Ting, C.-T. (2004) Genes and speciation. *Nat. Rev. Genet.* 5, 114–122

61 Martin, C.H. (2016) The cryptic origins of evolutionary novelty: 1,000-fold-faster trophic diversification rates without increased ecological opportunity or hybrid swarm. *Evolution (N. Y).* DOI: 10.1111/evo.13046

62 Hedrick, P.W. (2013) Adaptive introgression in animals: Examples and comparison to new mutation and standing variation as sources of adaptive variation. *Mol. Ecol.* 22, 4606–4618

63 Huerta-Sanchez, E. *et al.* (2014) Altitude adaptation in Tibetans caused by introgression of Denisovan-like DNA. *Nature* 512, 194–197

64 Stankowski, S. and Streisfeld, M.A. (2015) Introgressive hybridization facilitates adaptive divergence in a recent radiation of monkeyflowers. *Proc. R. Soc. London B* 282, 20151666

65 Rieseberg, L.H. *et al.* (1999) Transgressive segregation, adaptation and speciation. *Heredity (Edinb).* 83, 363

66 Kagawa, K. and Takimoto, G. (2017) Hybridization can promote adaptive radiation by means of transgressive segregation. *Ecol. Lett.* 21, 264–274

67 Schumer, M. *et al.* (2014) How common is homoploid hybrid speciation? *Evolution (N. Y).* 68, 1553–1560

68 Feder, J.L. *et al.* (2003) Allopatric genetic origins for sympatric host-plant shifts and race formation in Rhagoletis. *Proc. Natl. Acad. Sci.* 100, 10314–10319

69 Fishman, L. *et al.* (2013) Chromosomal rearrangements and the genetics of reproductive barriers in mimulus (monkey flowers). *Evolution (N. Y).* 67, 2547–2560

70 Guerrero, R.F. and Hahn, M.W. (2017) Speciation as a sieve for ancestral polymorphism. *Mol. Ecol.* 26, 5362–5368

71 Nelson, T.C. and Cresko, W.A. (2018) Ancient genomic variation underlies repeated ecological adaptation in young stickleback populations. *Evol. Lett.* 2, 9–21

72 Malinsky, M. *et al.* (2015) Genomic islands of speciation separate cichlid ecomorphs in an East African crater lake. *Science (80-.).* 350, 1493–1498

73 Smith, J.M. and Haigh, J. (1974) The hitch-hiking effect of a favourable gene. *Genet. Res.* 23, 23–35

74 Nachman, M.W. and Payseur, B.A. (2012) Recombination rate variation and speciation: theoretical predictions and empirical results from rabbits and mice. *Philos. Trans. R. Soc. B Biol. Sci.* 367, 409–421

75 Cruickshank, T.E. and Hahn, M.W. (2014) Reanalysis suggests that genomic islands of speciation are due to reduced diversity, not reduced gene flow. *Mol. Ecol.* 23, 3133–3157

76 Stölting, K.N. *et al.* (2015) Genome-wide patterns of differentiation and spatially varying selection between postglacial recolonization lineages of *Populus alba* (Salicaceae), a widespread forest tree. *New Phytol.* 207, 723–734

77 Clarkson, C.S. *et al.* (2014) Adaptive introgression between Anopheles sibling species eliminates

a major genomic island but not reproductive isolation. *Nat. Commun.* 5, 4248
78 Ortíz-Barrientos, D. and Noor, M.A.F. (2005) Evidence for a One-Allele Assortative Mating Locus. *Science (80-.).* 310, 1467 LP-1467
79 Siepel, A. *et al.* (2005) Evolutionarily conserved elements in vertebrate, insect, worm, and yeast genomes. *Genome Res.* 15, 1034–1050
80 Boyle, E.A. *et al.* (2017) An Expanded View of Complex Traits: From Polygenic to Omnigenic. *Cell* 169, 1177–1186
81 Kratochwil, C.F. *et al.* (2017) Tol2 transposon-mediated transgenesis in the Midas cichlid (Amphilophus citrinellus) — towards understanding gene function and regulatory evolution in an ecological model system for rapid phenotypic diversification. *BMC Dev. Biol.* 17, 15
82 Cleves, P.A. *et al.* (2018) An intronic enhancer of Bmp6 underlies evolved tooth gain in sticklebacks. *PLOS Genet.* 14, e1007449
83 Vijay, N. *et al.* (2017) Genomewide patterns of variation in genetic diversity are shared among populations, species and higher-order taxa. *Mol. Ecol.* 26, 4284–4295
84 Van Doren, B.M. *et al.* (2017) Correlated patterns of genetic diversity and differentiation across an avian family. *Mol. Ecol.* 26, 3982–3997
85 Ma, T. *et al.* (2018) Ancient polymorphisms and divergence hitchhiking contribute to genomic islands of divergence within a poplar species complex. *Proc. Natl. Acad. Sci.* 115, E236 LP-E243
86 Delmore, K.E. *et al.* (2018) Comparative analysis examining patterns of genomic differentiation across multiple episodes of population divergence in birds. *Evol. Lett.* 2, 76–87
87 Bush, G.L. (1975) Modes of Animal Speciation. *Annu. Rev. Ecol. Syst.* 6, 339–364
88 Sorenson, M.D. *et al.* (2003) Speciation by host switch in brood parasitic indigobirds. *Nature* 424, 928–931
89 Savolainen, V. *et al.* (2006) Sympatric speciation in palms on an oceanic island. *Nature* 441, 210–213
90 Yeaman, S. *et al.* (2016) The evolution of genomic islands by increased establishment probability of linked alleles. *Mol. Ecol.* 25, 2542–2558
91 Cotto, O. and Servedio, M.R. (2017) The Roles of Sexual and Viability Selection in the Evolution of Incomplete Reproductive Isolation: From Allopatry to Sympatry. *Am. Nat.* 190, 680–693
92 Sachdeva, H. and Barton, N.H. (2017) Divergence and evolution of assortative mating in a polygenic trait model of speciation with gene flow. *Evolution (N. Y).* 71, 1478–1493
93 Seehausen, O. (2006) African cichlid fish: a model system in adaptive radiation research. *Proc. R. Soc. Biol. Sci.* 273, 1987–98
94 Sparks, J.S. (2004) Molecular phylogeny and biogeography of the Malagasy and South Asian cichlids (Teleostei: Perciformes: Cichlidae). *Mol. Phylogenet. Evol.* 30, 599–614
95 Schliewen, U.K. *et al.* (1994) Sympatric speciation suggested by monophyly of crater lake cichlids. *Nature* 368, 629–631
96 Kautt, A. *et al.* Multispecies outcomes of sympatric speciation after admixture with the source population in two radiations of Nicaraguan crater lake cichlids. *journals.plos.org*
97 Wagner, C.E. *et al.* (2014) Cichlid species-area relationships are shaped by adaptive radiations that scale with area. *Ecol. Lett.* 17, 583–592
98 Poelstra, J.W. *et al.* (2018) , Speciation in sympatry with ongoing secondary gene flow and a potential olfactory trigger in a radiation of Cameroon cichlids. , *Molecular Ecology*, 27, 1–19
99 Kautt, A.F. *et al.* (2016) Incipient sympatric speciation in Midas cichlid fish from the youngest and one of the smallest crater lakes in Nicaragua due to differential use of the benthic and limnetic habitats? *Ecol. Evol.* 6, 5342–5357
100 Elmer, K.R. *et al.* (2010) Rapid sympatric ecological differentiation of crater lake cichlid fishes within historic times. *BMC Biol.* 8,
101 Kratochwil, C.F. *et al.* (2015) Embryonic and larval development in the Midas cichlid fish species flock (Amphilophus spp.): A new evo-devo model for the investigation of adaptive novelties and species differences Evolutionary developmental biology. *BMC Dev. Biol.* 15, 1–15

730 102 Excoffier, L. *et al.* (2013) Robust Demographic Inference from Genomic and SNP Data. *PLOS*
731 *Genet.* 9, e1003905
732 103 Gutenkunst, R.N. *et al.* (2009) Inferring the Joint Demographic History of Multiple Populations
733 from Multidimensional SNP Frequency Data. *PLOS Genet.* 5, e1000695
734 104 Loh, P.-R. *et al.* (2013) Inferring Admixture Histories of Human Populations Using Linkage
735 Disequilibrium. *Genetics* 193, 1233 LP-1254
736 105 Vernot, B. and Akey, J.M. (2014) Resurrecting Surviving Neandertal Lineages from Modern
737 Human Genomes. *Science (80-.).* 1245938,
738 106 Gronau, I. *et al.* (2011) Bayesian inference of ancient human demography from individual genome
739 sequences. *Nat. Genet.* 43, 1031
740 107 Hudson, R.R. (2007) The Variance of Coalescent Time Estimates from DNA Sequences. *J. Mol.*
741 *Evol.* 64, 702–705
742 108 Coop, G. *et al.* (2008) The Timing of Selection at the Human FOXP2 Gene. *Mol. Biol. Evol.* 25,
743 1257–1259
744 109 Beleza, S. *et al.* (2013) The Timing of Pigmentation Lightening in Europeans. *Mol. Biol. Evol.* 30,
745 24–35
746 110 Nakagome, S. *et al.* (2016) Estimating the Ages of Selection Signals from Different Epochs in
747 Human History. *Mol. Biol. Evol.* 33, 657–669
748 111 Ormond, L. *et al.* (2016) Inferring the age of a fixed beneficial allele. *Mol. Ecol.* 25, 157–169
749 112 Chen, H. *et al.* (2015) A hidden Markov model for investigating recent positive selection through
750 haplotype structure. *Theor. Popul. Biol.* 99, 18–30
751 113 Smith, J. *et al.* (2018) Estimating Time to the Common Ancestor for a Beneficial Allele. *Mol.*
752 *Biol. Evol.* 35, 1003–1017
753 114 Chatterjee, S. and Ahituv, N. (2017) Gene Regulatory Elements, Major Drivers of Human Disease.
754 *Annu. Rev. Genomics Hum. Genet.* 18, 45–63
755 115 Richards, E.J. and Martin, C.H. (2017) Adaptive introgression from distant Caribbean islands
756 contributed to the diversification of a microendemic adaptive radiation of trophic specialist
757 pupfishes. *PLoS Genet.* 13, 1–35
758