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Eco-evolutionary dynamics of prior selfing rate promotes the coexistence without niche partitioning under reproductive interference

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Running head: coexistence promoted by the evolution of selfing

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Abstract

1. When the two or more plants species share the same pollinators, pollinator-mediated reproductive interference make coexistence difficult. Recent studies suggested prior autonomous selfing mitigate reproductive interference, could enabling coexistence without pollination niche partitioning (pre-emptive selfing hypothesis). However, there are no studies to test whether evolution of prior selfing promote the coexistence, considering eco-evolutionary dynamics of population size, selfing rate and inbreeding depression.

2. To examine conditions that the evolution of prior selfing promote coexistence under mutual reproductive interference especially in the point of view for pollinator availability and dynamics of inbreeding depression, we constructed individual-based model in which two plant species compete against each other in the form of mutual reproductive interference and can evolve prior autonomous selfing rate. We expected that purging of deleterious mutations could cause evolutionary rescue because inferior species could rescue population density through the evolution of prior selfing if the strength of inbreeding depression decreases with an increase of population's selfing rate.

3. Our simulation demonstrated that the evolution of prior selfing could promote the coexistence while reproductive interference caused competitive exclusion without evolution. We found that lower pollinator availability tended to prefer rapid evolutionary shift to higher prior selfing rate, it neutralizes the negative effect of reproductive interference, and population dynamics exhibit neutral random walk in both species. When the strength of inbreeding depression decreased with an increase in population's selfing rate, moderate pollinator availability resulted in long-term coexistence in which relative-abundance-dependent selection on the prior selfing rate

45   rescue population density of inferior species intermittently.

46   **4.** *Synthesis.* We showed that the evolution of prior selfing could increase population

47   growth rate of inferior species and consequently enable the long-term coexistence with

48   evolutionary rescue. This is the new mechanisms explaining co-evolutionary

49   coexistence of closely related plant species without niche partitioning and consistent

50   with recent studies reported that closely related mixed-mating species are sympatrically

51   growing even under the mutual reproductive interference.

52

53   **Key-words:** co-evolution, evolutionary rescue, inbreeding depression, individual based

54   model, mixed mating, pollinator-mediated competition, selfing syndrome

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57   **Introduction**

58   Clarifying the conditions under which competing species can coexist is a traditional and

59   most fundamental subject in ecology (May, 1974; Chesson, 2000). Numerous empirical

60   and theoretical works have shown that niche partitioning between competing species is

61   required for their coexistence: i.e. the intraspecific competition should be larger than

62   interspecific competition (Chesson, 2000; Silvertown, 2004). Closely related species,

63   which potentially share the same or very similar resources and reproductive habits, are

64   therefore expected to heavily compete against each other, likely being unable to coexist

65   (Gröning & Hochkirch, 2008; Burns & Straus, 2011; Whitton, Sears & Maddison;

66   2017).

67   In flowering plants, when two or more plant species depend on the same

68   pollinators for reproduction, interspecific pollinator-mediated pollen transfer can cause

69   strong reproductive interference that results in competitive exclusion of either species or

70   niche partitioning between species (Levin & Anderson, 1970; Takakura, Nishida,

71   Matsumoto & Nishida, 2008; Runquist & Stanton, 2013; Moreira-Hernández &

72   Muchhala, 2019). Reproductive interference via interspecific pollen transfer can occur

73   when both or either of increase in heterospecific pollen deposition on the stigma and

74   conspecific pollen loss on heterospecific flower (Mitchell, Flanagan, Brown, Waser &

75   Karron, 2009; Morales & Traveset, 2008). Specifically, Pollen deposition from closely

76   related heterospecies is known to strongly decrease female reproductive success owing

77   to pollen tube growth competition in the style, ovule discounting and/or hybridization

78   (Harder, Cruzan & Thomson, 1993; Nishida, Kanaoka, Hashimoto, Takakura & Nishida,

79   2014; Whitton et al., 2017). Thus, reproductive interference via heterospecific pollen

80   deposition may favour spatiotemporal segregation in flowering or floral trait

81   displacement, with a consequence that they use different, or different body parts of the

82   same, pollinator species (e.g. Runquist 2012; van der Niet & Johnson 2012; Huang &

83   Shi, 2013).

84   Selfing has gathered much recent attention as an alternative mechanism that

85   can mitigate reproductive interference by heterospecific pollen transfer from competing

86   relatives (Fishman & Wyatt, 1999; de Waal, Anderson & Ellis, 2015; Katsuhara &

87   Ushimaru, 2019). Recent studies have suggested that selfing constitutes a reproductive

88   barrier among sympatrically coexisting related species (Fishman & Wyatt, 1999; Martin

89   & Willis, 2007; Goodwillie & Ness 2013; Brys, van Cauwenberghe & Jaquemyn, 2016).

90   Selfers with small and inconspicuous flower (selfing syndrome) which therefore

91   receives fewer pollinator visits are likely to coexist with outcrossing relatives (Sicard &

92   Lenhard, 2011; Kalisz et al., 2012). Thus, reduced heterospecific pollen deposition

93   owing to fewer visits and/or reproductive assurance via self-pollination might mitigate

94   the negative effect of reproductive interference in selfers, although it is difficult to

95   clarify their relative importance in general (Fishman & Wyatt, 1999; Martin & Willis,

96   2007; de Waal et al., 2015; Brys et al., 2016).

97   Recent studies further hypothesize that "prior" rather than "delayed"

98   autonomous selfing can mitigate the negative effect of reproductive interference via

99   interspecific pollen transfer and promote species coexistence independent of the

100   presence of pollinator visitations (the pre-emptive selfing hypothesis; Randle, Spigler &

101   Kalisz, 2018; Katsuhara & Ushimaru, 2019). Theoretical and empirical studies have

102   suggested that prior selfing unlikely evolves with frequent pollinator visitations (Lloyd,

103   1992; Kalisz, Vogler & Hanley, 2004; Eckert et al., 2010). However, in the presence of

104   reproductive interference by an abundant competitor species, frequent pollinator

105   visitations largely reduce outcrossing success of an inferior species. In such a situation,

106   the evolution of prior selfing can mitigate the negative effect of reproductive

107   interference from the competitor and would rescue the inferior species from competitive

108   exclusion.

109   The pre-emptive selfing hypothesis should be tested in the context of eco-

110   evolutionary dynamics of population size, selfing rate and inbreeding depression.

111   Because the negative effect of reproductive interference that decreases outcrossing

112   success becomes greater with an increase in the relative abundance of competing

113   species (Levin & Anderson, 1970; Katsuhara & Ushimaru, 2019), population dynamics

114   of mutually competing species should be an important driving factor of the evolution of

115   prior autonomous selfing of a given species. The evolution of prior selfing could rescue

116   the population density of competitively inferior species via mitigation of reproductive

117   interference while it could also result in self-extinction due to the negative effect of

118   inbreeding depression on population growth rate depends on the strength of inbreeding

119   depression (Cheptou, 2019; Katsuhara & Ushimaru, 2019). Therefore, dynamics of

120   inbreeding depression is an important factor influencing the evolution of selfing because

121   the strength of inbreeding depression is often expressed as a decreasing function of

122   population's sefling rate due to "purging" of deleterious, recessive alleles (Schemske &

123   Lande, 1985; Chaelesworth, Chaelesworth & Morgan, 1990; Lloyd, 1992; Husband &

124   Schemske, 1996; Crnokrak & Barrett, 2002; Goodwillie, Kalisz & Eckert, 2005;

125   Charlesworth & Willis, 2009). Thus, dynamics of population size, the degree of selfing

126   rate and inbreeding depression of competing species are ideally considered to examine

127   the adaptive significance of prior selfing under reproductive interferences. To the best of

128   our knowledge, however, no studies have examined the eco-evolutionary dynamics of

129   these variables, and therefore little is known about the possibility of coexistence under

130   reproductive interference, followed by evolution of prior selfing.

131   In this study, to examine the pre-emptive selfing hypothesis, we constructed a

132   model in which two plant species sharing the same pollination niche and can evolve

133   prior autonomous selfing compete against each other in the form of mutual reproductive

134   interference (i.e. eco-evolutionary dynamic model). Using the model, we addressed

135   following questions. Can prior selfing evolve under mutual reproductive interference

136   and promote their coexistence as an evolutionary rescue agent? Is inbreeding depression

137   an important determinant for the joint dynamics of population size and selfing rate? By

138   answering to these questions, we discuss the conditions in which the evolution of prior

139   selfing promotes the long-term coexistence of closely related species sharing the same

140   pollination niche.

141

Model

143 *Community structure, pollination, seed production and germination processes*

144 We develop an individual-based model of competition between two annual flowering

145 plant species (species with discrete generation) within a site whose carrying capacity is

146 K: K individuals of both or either of species 1 (sp$_1$) and 2 (sp$_2$) lives in the site for each

147 generation (default K value is 2,000). Here, the relative abundance of sp$_i$ at the t-th

148 generation are denoted as $q_{i,t}$ where i is either 1 or 2 and $q_{1,t} + q_{2,t} = 1$ holds. Thus,

149 the number of individuals of sp$_i$ equals $K q_{i,t}$. In the model, we assume that ecological

150 niches of sp$_1$ and sp$_2$ completely overlap with each other, although the two species

151 produce no hybrids.

152   First, we describe the pollination and fertilization processes in the model. Each

153 individual of both species produces n ovules which are fertilized via prior autonomous

154 selfing and outcrossing mediated by pollinators and g pollen grains. The j-th individual

155 of sp$_i$ fertilizes their ovules via prior autonomous selfing at the rate of $r_{i,j,t}$ in the t-th

156 generation ($1 \leq j \leq Kq_{i,t}$ for $i = 1$ or 2). Thus, an integral number of ovules obtained

157 by rounding $nr_{i,j,t}$ are fertilized via prior selfing and the others were remained for

158 pollinator-mediated outcrossing. Here, we assumed that an integral number obtained by

159 rounding proportion P ($0 \leq P \leq 1$) of the $n(1 - r_{i,j,t})$ ovules are pollinated with

160 outcrossed conspecific and/or heterospecific pollen grains by pollinators. Using $P < 1$,

161 we can formulate pollinator limitation. Here, we assume that pollinators

162 indiscriminately visit flowers of both species and carried their pollen in proportion to

163 their relative flower abundances.

164   Pollen parent of each outcrossed ovule of individual j is randomly assigned to

165   conspecies with the probability of $\dfrac{\sum_{j=1}^{Kq_{i,t}} g(1-r_{i,j,t})}{\sum_{j=1}^{Kq_{i,t}} g(1-r_{i,j,t}) + c_h \sum_{j=1}^{(1-q_{i,t})K} g(1-r_{h,j,t})}$,

166   where c_h, $r_{i,j,t}$ and $r_{h,j,t}$ $(0 \leq c_h,\ q_{i,t},\ r_{i,j,t}$ and $r_{h,j,t} \leq 1)$ are the strength of

167   reproductive interference from heterospecies (sp$_h$) and the rates of prior selfing of the j-

168   th individuals of sp$_i$ and sp$_h$, respectively. The parameter c_h is interpreted as the

169   competitive ability of a heterospecific pollen grain relative to that of a conspecific one

170   to get fertilization with individual j's ovule. Besides, the probabilities of which pollen

171   grains of individual j fertilize conspecific and heterospecific ovules are described as

172    $\dfrac{g(1-r_{i,j,t})}{\sum_{j=1}^{Kq_{i,t}} g(1-r_{i,j,t}) + c_h \sum_{j=1}^{(1-q_{i,t})K} g(1-r_{h,j,t})}$ and $\dfrac{c_h g(1-r_{i,j,t})}{c_h \sum_{j=1}^{Kq_{i,t}} g(1-r_{i,j,t}) + \sum_{j=1}^{(1-q_{i,t})K} g(1-r_{h,j,t})}$,

173   respectively. In this formulation, we assume that outcross pollen proportionally

174   decreases with the prior selfing rate in respective individuals of both species. This

175   assumption mimics increased pollen discounting and/or anther-stigma interference with

176   increasing the prior selfing rate (Lloyd and Webb, 1986; Webb and Lloyd, 1986;

177   Karron, Jackson, Thumser & Schlicht 1997; Fishman, 2000; Barrett 2002). Because we

178   assumed random pollination, it should be noted the ovule of j-th individual is fertilized

179   by pollen of j-th individual (pollinator-mediated self-pollination; geitonogamy) with the

180   above probability and they are treated as self-fertilized pollen as well as the ovules

181   fertilized by prior autonomous selfing.

182   Second, we denote the seed production process. We assume that only ovules

183   fertilized by self- and outcrossed-conspecific pollen can develop seeds whereas those

184   fertilized by heterospecific pollen produce no seeds. A cost of selfing relative to

185   outcrossing is also assumed as follows. In sp$_i$, all outcrossed ovules develop sound seeds

186   whereas selfed ovules set seeds at the rate of $1 - I_{i,t}$, where $I_{i,t}$ $(0 \leq I_{i,t} \leq 1)$ is the

187   strength of inbreeding depression at generation t in sp$_i$. Here, $I_{i,t}$ is described as a

188   function of the population's selfing rate at the $(t-1)$th generation of sp$_i$, $S_{i,t-1}$:

189      $$I_{i,t} = \beta e^{-\alpha S_{i,t-1}}.$$

190      $S_{i,t-1}$ is calculated as total number of selfed sound seeds divided by total number of

191   sound seeds in sp$_i$ in the last generation. $I_{i,t}$ can be interpreted as an evolutionary

192   variable, which decreases with an increase in the population's selfing rate in the parental

193   generation owing to the accumulation–purging balance of deleterious mutations

194   (Schemske & Lande, 1985; Husband & Schemske, 1996; Crnokrak & Barrett, 2002).

195   The coefficient α expresses how inbreeding depression steeply decreased with

196   increasing the population's selfing rate whereas the intercept β ($0 \leq \beta \leq 1$) indicates the

197   level of inbreeding depression when complete outcrossing occurs in the population. We

198   simulated various α and β values to test various scenarios in the eco-evolutionary

199   dynamics of population size and selfing rate.

200   As the final process, K seedlings from all seeds produced by both species are

201   randomly selected and construct generation $t+1$. We assume no interspecific

202   differences in competitive ability at germination and establishment processes as well as

203   other ecological and genetic features: c_h, α, and β are also equal for both sp$_1$ and sp$_2$. In

204   addition, our model has no spatial structure.

205

206   *Inheritance and mutation of the rate of prior autonomous selfing*

207   To describe the evolutionary dynamics of prior autonomous selfing, our model assumes

208   that the prior selfing rate of individual j in the next generation $r_{i,j,t+1}$ is determined as

209   the parental average. Thus, the prior selfing rate is assumed to be a quantitative genetic

210   trait value which can be influenced by various quantitative traits such as the degrees of

211   herkogamy and/or dichogamy and the proportion of cleistogamous flowers (Culley &

212   Klooster, 2007; Kalisz et al., 2012). In addition, $r_{i,j,t+1}$ can be mutated to be slightly

213   lower or higher than the parental mean (a random value between $-\sigma$ and $+\sigma$ is added to

214   the parental mean) with a probability μ. μ and σ are the rate and effect size of mutation,

215   respectively. We used 0.05 and 0.1 for μ and σ as default values, respectively. If mutated

216    $r_{i,j,t+1}$ becomes larger than 1 or smaller than 0, we use the values 1 and 0, respectively.

217

218   *Simulation settings and categorization of eco-evolutionary consequence*

219   To explore conditions for the coexistence of the two species, we examined the effects of

220   pollinator availability (P) and inbreeding depression-selfing rate relationship (α and β)

221   on the consequences of evolution. We tested two following scenarios for inbreeding

222   depression-selfing rate relationship. $I_{i,t}$ is fixed ($\alpha = 0$; $\beta = 0.1, 0.3, 0.5, 0.7$ or 0.9) or

223   it varies in concert with the population's selfing rate ($\alpha = 0, 0.5, 1, 2, 4$ or 8; $\beta = 0.9$),

224   with the whole parameter range of $0 \leq P \leq 1$ (Table 1). In each simulation run, the

225   initial numbers of individuals for both species are equal as $K/2$. The initial autonomous

226   selfing rates for individuals were generated randomly with normal distribution whose

227   mean and standard deviation are r_{intial} (1/2) and sd_{intial} (1/6) for both species. Each run

228   continues for 2,000 generations or until either species goes extinct.

229   After 50 simulation runs for each parameter setting, we classified the eco-

230   evolutionary dynamics into four categories based on ecological and evolutionary status

231   of the species. When the simulation terminated by the extinction of either species and

232   the population mean of prior autonomous selfing rate in survivors was higher or lower

233   than 0.5, the result was categorized as (1) competitive exclusion by selfer or (2) that by

234   outcrosser, respectively. Meanwhile, the runs in which the two species still coexisted at

235   the 2,000th generation were also divided into two categories, (3) a coexistence with

236   evolutionary rescue by prior selfing and (4) a coexistence with neutral dynamics, based

237   on following procedures.

238   To detect the evolutionary rescue, we calculated the population growth rate and

239   selection gradient in each generation of sp_i. Population growth rate ($W_{i,t}$) for the t-th

240   generation is calculated as $Kq_{i,t+1}/Kq_{i,t}$ $(= q_{i,t+1}/q_{i,t})$. For clarifying the selection

241   gradient on the prior selfing rate, we identified seed and pollen parents of all seeds and

242   calculate a correlation coefficient between selfing rate $r_{i,j,t}$ and seeding and siring

243   success of each individual as the selection gradient ($G_{i,t}$) at the t-th generation. The

244   positive (or negative) $G_{i,t}$ means that the higher (or lower) rate was adaptive at the

245   generation in sp_i. Then, the evolutionary rescue by prior selfing is defined as a state

246   following two conditions are satisfied simultaneously: (1) a significant negative

247   correlation between $q_{i,t}$ and $G_{i,t}$ (i.e., a population decline facilitates the evolution of

248   selfing), (2) a significant positive correlation between the population mean of prior

249   selfing rate $\sum_{j=1}^{Kq_{i,t}} r_{i,j,t}/Kq_{i,t}$ and $W_{i,t}$ (i.e., the evolution of selfing increases

250   population growth). Significances of these two correlations were examined by a

251   permutation test. We permutated the variables of the last 500 generations in each run

252   10,000 times and made null distribution and the 95% prediction interval of each

253   correlation to test the significance of the value of the run. When both or either of the

254   correlation coefficients were not significant, the run was categorized into the

255   coexistence with neutral dynamics (Fig. 1).

256   We compared difference in long-term stability between coexistences with

257   neutral dynamics and evolutionary rescue. We selected a typical parameter set for each

258   coexistence type: $P = 0.4$, $\alpha = 0.5$, $\beta = 0.9$ for that with evolutionary rescue (ER set) and

259    $P = 0.1$, $\alpha = 4$, $\beta = 0.9$ for coexistence with neutral dynamics (ND set). For each

260   parameter set, we conducted 200 simulations for 10,000 generations and recorded the

261   generation until which two species coexisted.

262   We also checked how simulation results change depending on the strength of

263   reproductive interference and the initial population's mean selfing rate. We examined

264   simulations in which c_h (= 0.0, 0.25, 0.5, 0.75 or 1.0) and $r_{initial}$ (= 0.0, 0.25, 0.5, 0.75 or

265   1.0) varied with the above parameter settings (ER and ND sets) and run 50 simulations

266   for each parameter set. Moreover, to check the population dynamics of the two species

267   with the fixed population's prior selfing rates, we conducted simulation runs in which

268   sp_1 and sp_2 had the same or different fixed prior selfing rates ($0 \leq r_{initial} \leq 1$) with the

269   same two parameter settings (ER and ND sets). Finally, we run simulations with the

270   fixed abundance of two species to examine the effect of fixed population size on the

271   evolution of prior selfing rate in the two parameter settings (ER and ND sets).

272

273   **Results**

274   *Eco-evolutionary dynamics with fixed inbreeding depression*

275   We found that lower pollinator availability preferred the evolution of higher selfing rate

276   in both species, often promoting their coexistence with neutral dynamics (Fig. 2).

277   Conditions for the coexistence with neutral dynamics was more limited by higher

278   inbreeding depression (Fig. 2). The coexistences with neutral dynamics were always

279   realized when the two species evolved the prior selfing rate close to 1.0, which

280   neutralized their mutual reproductive interference (Fig. 1). During the coexistence,

281   population dynamics of both species exhibited a random walk. Therefore, the

282   coexistence with neutral dynamics is not stable in the long term and the extinction of

283   either species occurred when simulations continued for more generations (see the

284   section below, *Long-term stability of the coexistences*). Meanwhile, when either or both

285   of I and P are large, the eco-evolutionary dynamics tended to be terminated by

286   competitive exclusion (Fig. 2). Especially when both of I and P are large, competitive

287   exclusion by outcrosser always terminated the eco-evolutionary dynamics (Fig. 2). The

288   coexistence with evolutionary rescue rarely occurred when the inbreeding depression

289   was fixed and independent of the population's selfing rate (Fig. 2).

290

291   *Eco-evolutionary dynamics with variable inbreeding depression*

292   In the scenarios with variable inbreeding depression, conditions for both types of

293   coexistence were more relaxed compared to those assuming fixed inbreeding depression

294   (Figs. 2, 3). Interestingly, conditions with intermediate levels of pollinator availability

295   and the slope of inbreeding depression function α more frequently facilitated the

296   coexistence with evolutionary rescue or neutral dynamics than other conditions (Fig. 3).

297   When inbreeding depression sharply decreases as the population's selfing rate

298   increases ($\alpha \geq 4.0$), competitive exclusion by selfer occurred in wider conditions as in

299   those with lower fixed inbreeding depression ($\alpha = 0$ and $\beta = 0.1$ or 0.3; Figs. 2, 3).

300   Meanwhile, when inbreeding depression more gently decreased with increasing the

301   population's selfing rate ($\alpha = 0.5$), competitive exclusion by outcrosser tended to occur

302   in the presence of higher pollinator availability like in the cases both of I and P are large

303   in fixed inbreeding depression scenario.

304

305   *Long-term stability of the coexistences with neutral dynamics and evolutionary rescue*

306   The coexistence with evolutionary rescue continued until the 10,000-th generation if the

307   fluctuations of the relative abundances ($q_{i,t}$) and the prior selfing rates ($r_{i,t}$) have once

308   started, while the coexistence with neutral dynamics never coexisted before reaching the

309   10,000-th generation (Fig. 4).

310

311   *Dependence of simulation consequences on c_h and $r_{initial}$*

312   In the simulations with ER sets, we found that coexistence with evolutionary rescue

313   nearly always occurred with high initial population's selfing rate ($r_{initial} \geq 0.75$) and

314   presence of mutual reproductive interference ($c_h > 0.0$). When the initial population's

315   selfing rate was low ($r_{initial} \leq 0.25$), the both types of coexistence rarely or very

316   infrequently occurred in both the ER and ND sets (Fig. 5). Moreover, no competitive

317   exclusion by outcrosser was found when the initial population's selfing rate was high

318   ($r_{intial} \geq 0.75$). Meanwhile, the strength of reproductive interference (c_h) seems unlikely

319   to largely influence the coexistence with neutral dynamics with the ND parameter

320   setting. However, the coexistence of evolutionary rescue never occurred without mutual

321   reproductive interference ($c_h = 0.0$) with the ER set.

322

323   *Simulation consequences with fixed prior selfing rate and fixed population size*

324   We found that coexistence for 2,000 generations very rarely occurred when the prior

325   selfing rates were fixed in the two species for the both parameter settings except when

326   both species had the same and very high prior selfing rates (Fig. 6). Winners were

327   always the species having higher prior selfing rates with the ND parameter set whereas

328   winners were usually the species having the lower and higher prior selfing rates in the

329   below and above areas of the line of $r_2 = -r_1 + 0.6$, respectively, with the ER set (Fig.

330   6). In the simulations with the fixed abundance of two species, the evolutionary shift to

331   the higher prior selfing rate was favored only when the relative abundance of focal

332   species was lower than 1/2 with the ER parameter set (Fig. 7). Meanwhile, under the

333   ND set, very high prior selfing rate was always favored independent on their abundance

334   (Fig. 7).

335

336   **Discussion**

337   Our model revealed that the evolution of prior selfing can promote the coexistence in

338   the presence of mutual reproductive interference while the coexistence rarely occurred

339   without the evolution of prior selfing (Figs. 2, 3, 6). In the variable inbreeding

340   depression scenario (inbreeding depression decreases with an increase in the

341   population's selfing rate), both types of coexistence tended to be more occurred than in

342   the fixed inbreeding depression scenario when comparing same pollinator availability

343   (Figs. 2, 3). Especially when the strength of inbreeding depression gently decreased and

344   pollinator availability was intermediate level, the coexistence with evolutionary rescue

345   often occurred and stably continued for very long-term (Figs. 3, 4).

346   Firstly, we discuss the processes enabling the coexistence with evolutionary

347   rescue in our model (Fig. 1). At the early generations, the stochastic process makes

348   slight difference in population size and selfing rate between the two species and

349   reproductive interference by more abundant species with higher selfing rate enlarge the

350   difference. In such a situation, low outcross success due to increased heterospecific

351   pollen deposition facilitates the evolution of high prior selfing rate in the inferior

352   species. This evolutionary shift toward high selfing rate improves the population growth

353   rate via an increase in selfed seed production (i.e., evolutionary rescue by prior selfing

354   occurs) especially when inbreeding depression is weakened with increasing the

355   population's selfing rate in the inferior species. Meanwhile, once becoming the

356   dominant species, reproductive interference from the competitor is getting weaker so

357   that inbreeding depression favors lower prior selfing rate in the species. Lower prior

358   selfing rate, in turn, can reduce total seed production of the population when pollinator

359   availability is not high, leading to lower population growth rate compared to the

360   competitor. This relative-abundance-dependent selection on the prior selfing rate

361   promotes a negative relationship between and fluctuations of the prior selfing rate and

362   population size through generations. Due to this out-of-phase fluctuations occurring

363   both in two species, the long-term coexistence of the two species is realized under

364   mutual reproductive interference.

365   Here, suitable conditions for the coexistence with evolutionary rescue are

366   discussed by comparing to empirical knowledge. Our simulation demonstrated that the

367   coexistence with evolutionary rescue occurred with moderate pollinator limitation,

368   variable and moderate levels of inbreeding depression, the relatively higher initial prior

369   selfing rate and the presence of reproductive interference. High pollinator availability

370   always favors competitive exclusion by either outcorsser or selfers depending on the

371   level of inbreeding depression. In other words, the long-term coexistence under the

372   reproductive interference requires pollinator limited conditions which are prevailing in

373   wild flowering plants (Larson & Barrett, 2000). Gently variable inbreeding depression

374   still function as the cost of selfing even when the population's selfing rate of given

375   species is very high. While inbreeding depression due to deleterious recessive alleles are

376   thought to be rapidly purged with increasing population's selfing rate, weak late acting

377   inbreeding depression caused by weakly deleterious mutations and heterozygous

378 advantage due to overdominance cannot be purged even in predominantly selfing

379 species (Charlesworth et al., 1990; Husband & Schemske, 1996; Crnokrak & Barrett,

380 2002; Charlesworth & Wills, 2009). Additionally, although it may not be surprising, we

381 found that higher initial prior selfing rate widens the possibility of the coexistence with

382 evolutionary rescue (Fig. 5). The finding suggests that only a pair of predominantly

383 selfing or of mixed-mating species can coexist stably under reproductive interference,

384 being consistent with recent studies on the coexistence under mutual reproductive

385 interference (Tokuda et al., 2015; Katsuhara & Ushimaru, 2019; Nishida et al.

386 unpublished data). Without reproductive interference, this type of coexistence never

387 occurred even when other parameter settings are suitable for the coexistence (Fig. 5).

388 This result is very interesting and proposes that mutual reproductive interference can act

389 the cost of outcrossing and promoting more selfing (Katsuhara & Ushimaru, 2019),

390 likely making a fluctuation pattern in the prior selfing rate throughout the generation.

391   The coexistence with neutral dynamics was often found in conditions with

392 lower pollinator availability and weak fixed or moderately variable inbreeding

393 depression (Figs. 2, 3). In such conditions, the higher prior selfing rate evolves very

394 quickly to be almost completely 1.0 in both species (Fig. 1), which should be free from

395 the negative effect of reproductive interference from competitor. Both species exhibit

396 population dynamics of neutral random walk (Hubbell, 2001; Chave, 2004) and coexist,

397 so that stochastic events will stop this type of coexistence at some point in time (Fig. 4).

398 In our model, this type of coexistence was usually found in the parameter conditions

399 where competitive exclusion by selfer frequently occurred, suggesting that these

400 consequence categories do not differ qualitatively (Figs. 2, 3, 5). The rate of

401 evolutionary change in prior selfing rate differed between these categories and the

402   coexistence occurred when the high prior selfing rate evolved more rapidly in both

403   species (Fig. 1). Many predominately selfing weeds usually coexist in human-disturbed

404   habitats where pollinators are often limited (Baker, 1974), most likely being explained

405   by this type of coexistence. Empirical studies have shown the evolutionary shift to

406   higher prior selfing rate (often via reduction of herkogamy) can rapidly occurred under

407   pollinator limitation (Roels & Kelly, 2011; Brys & Jacquemyn, 2012; Gravasi &

408   Schiestl, 2017; Cheptou, 2019). To apply our results to selfing-species coexistence in

409   the field, the rate of evolutionary change of the prior selfing rate and pollinator

410   availability are better to be examined in future studies.

411   Under conditions with high pollinator availability and strong inbreeding

412   depression, mutual reproductive interference causes very rapid competitive exclusion by

413   outcrosser, being consistent with expectations in the previous works that considered no

414   limitation in outcross gamete transfer (Fig. 1, 2, 3; Levin & Anderson, 1970; Kishi &

415   Nakazawa, 2013). Besides, competitive exclusion by selfer is frequently occurred under

416   conditions with weak inbreeding depression and/or low pollinator availability. The

417   exclusion occurred more slowly comparing to the exclusion by outcrosser (Fig. 1). The

418   difference was likely due to that reproductive interference no more reduced seed

419   production in highly selfing species.

420   In both types of coexistence, co-evolutionary shifts to extremely high prior

421   selfing rate (over 0.9) was necessary in both competing species (Fig. 1). Many previous

422   empirical studies, however, reported coexistences of an extremely selfer and a related

423   outcrosser (Fishman & Wyatt, 1999; Brys et al., 2016; Randle et al., 2018). This

424   difference between the field observations and our results might be explained by in two

425   possible mechanisms which are not assumed in our model. First, some kinds of

426   ecological differences, such as competitive ability for germination and strength of

427   inbreeding depression, might exist between the study species, promoting the

428   coexistence of species with different mating systems. Second, selfers in these studies

429   always exhibited reduced floral attractiveness which minimized the negative effect of

430   reproductive interference via receiving few pollinator visits (Fishman & Wyatt, 1999;

431   Brys et al., 2016; Randle et al., 2018). Meanwhile, our model assumed only the rate of

432   prior selfing was evolvable as in the previous model (Cheptou, 2019). Such an evolution

433   of selfing floral syndrome could enable obligative selfers to coexist with outcrossing

434   relatives. Some recent studies reported the mutual reproductive interference between

435   two sympatrically growing mixed-mating species which have showy flowers with

436   frequent pollinator visitations and traits promoting prior autonomous self-pollination

437   (cleistogamous flowers in *Impatiens noli-tangare* and *I. textori*, Tokuda et al., 2015; bud

438   pollination in *Commleina communis* and *C. c.* f. *ciliata*, Katsuhara & Ushimaru, 2019).

439   The coexistences found in these study systems could be explained by prior-selfing

440   mediated evolutionary rescue, which are predicted by our model. To test this possibility,

441   monitoring of eco-evolutionary dynamics of these competing species in the fields will

442   be required. Although the complete test will require much time and effort, to examine

443   the relationships among population's selfing rate, inbreeding depression and relative

444   abundance in the field should improve our understanding of co-evolutionary

445   coexistence mechanisms without pollination niche partitioning as the first step.

446   In conclusion, our model successfully showed that the evolution of prior

447   selfing could increase population growth rate of inferior species and consequently

448   enable the long-term coexistence with evolutionary rescue. We successfully showed that

449   evolutionarily variable inbreeding depression based on accumulation–purging balance

450   of deleterious mutations expand the possibility of coexistence and promote the long-

451   term coexistence. The result suggests that genetic dynamics of inbreeding depression

452   within a given species may largely influence dynamics of community where pollinator-

453   mediated competition occurs. Finally, we propose new mechanisms explaining co-

454   evolutionary coexistence of closely related species under mutual without any kinds of

455   niche differentiation and spatial structures. The applicability and generality of the

456   proposed mechanisms should be investigated empirically in future.

457

Acknowledgements

465

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611

612   **Table 1.** List of parameters.

Parameter	Definition	Default value
$r_{i,j,t}$	The ratio of ovules fertilized via prior autonomous selfing in the j-th individual of sp$_i$ at the t-th generation.	0-1
$q_{i,t}$	Relative abundance of sp$_i$ at the t-th generation	0-1
P	Pollinator availability	0-1
c_h	Strength of reproductive interference	1
α	Slope of inbreeding depression function	0, 0.5, 1, 2, 4, 8
β	Intercept of inbreeding depression function	0.1, 0.3, 0.5, 0.7, 0.9
μ	mutation rate	0.05
σ	Effect size of mutation	0.1
K	Carrying capacity (number of individual plant)	2000
n	Number of ovules per individual plant	200
$r_{initial}$	Mean of initial prior autonomous selfing rate	0.5
$sd_{initial}$	Standard deviation of initial prior autonomous selfing rate	1/6

613
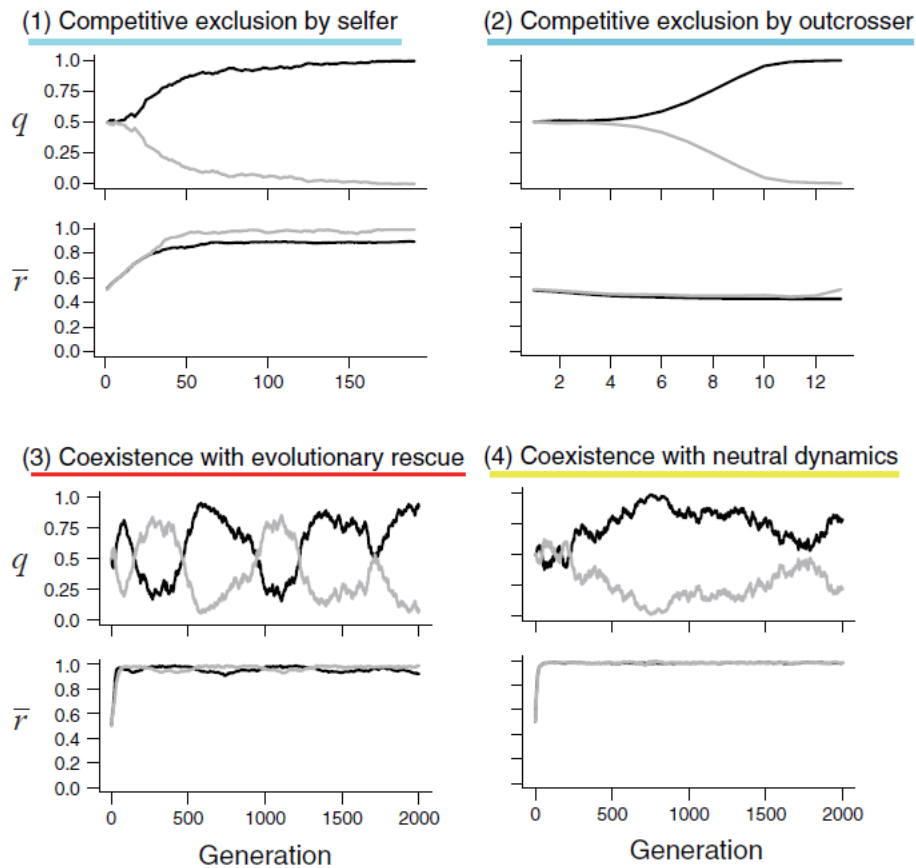
Fig. 1. Examples of four consequences of eco-evolutionary dynamics in our simulations: (1) competitive exclusion by selfer; (2) competitive exclusion by outcrosser; (3) coexistence with evolutionary rescue; (4) coexistence with neutral dynamics. Upper and lower graphs of each category show relative abundance and population mean of prior selfing rate dynamics of sp_1 (black line) and sp_2 (grey line), respectively.
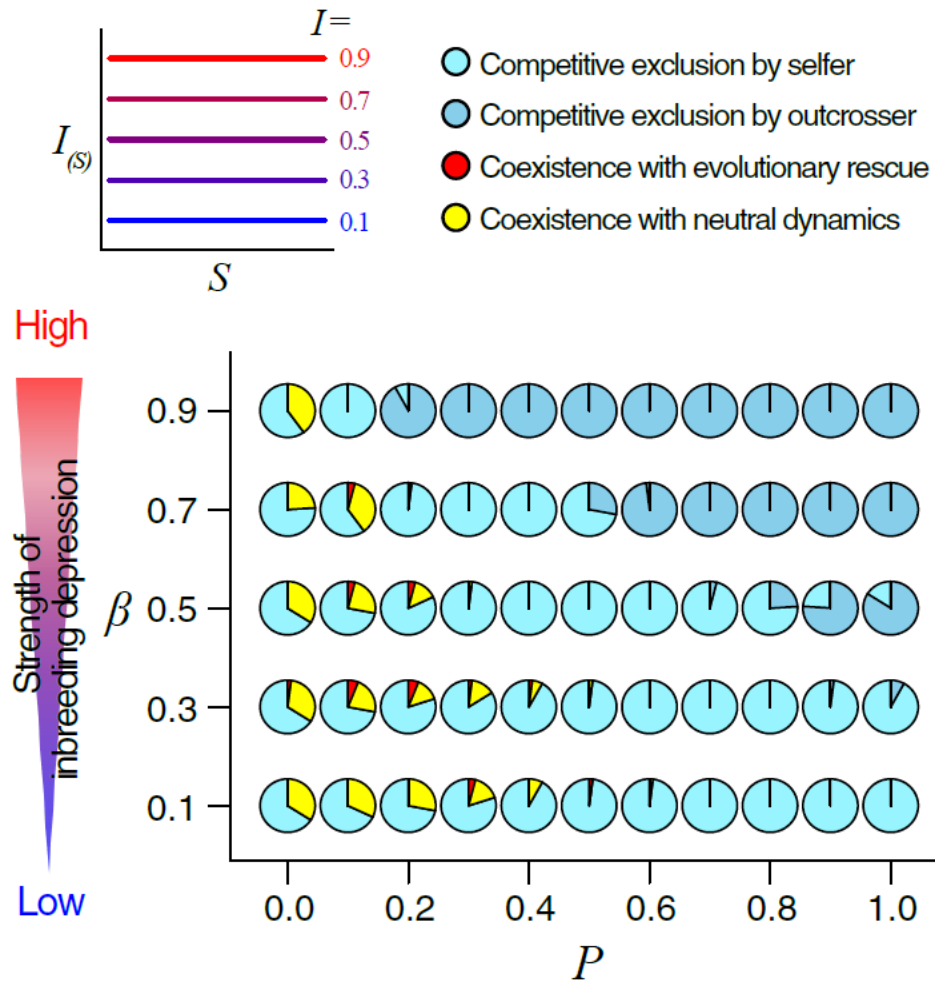
Fig. 2. Effects of pollinator availability and the strength of inbreeding depression on simulation consequence in fixed inbreeding depression scenarios. Each pie chat shows frequencies of four categories of eco-evolutionary consequences of 50 simulation runs (Fig. 1).

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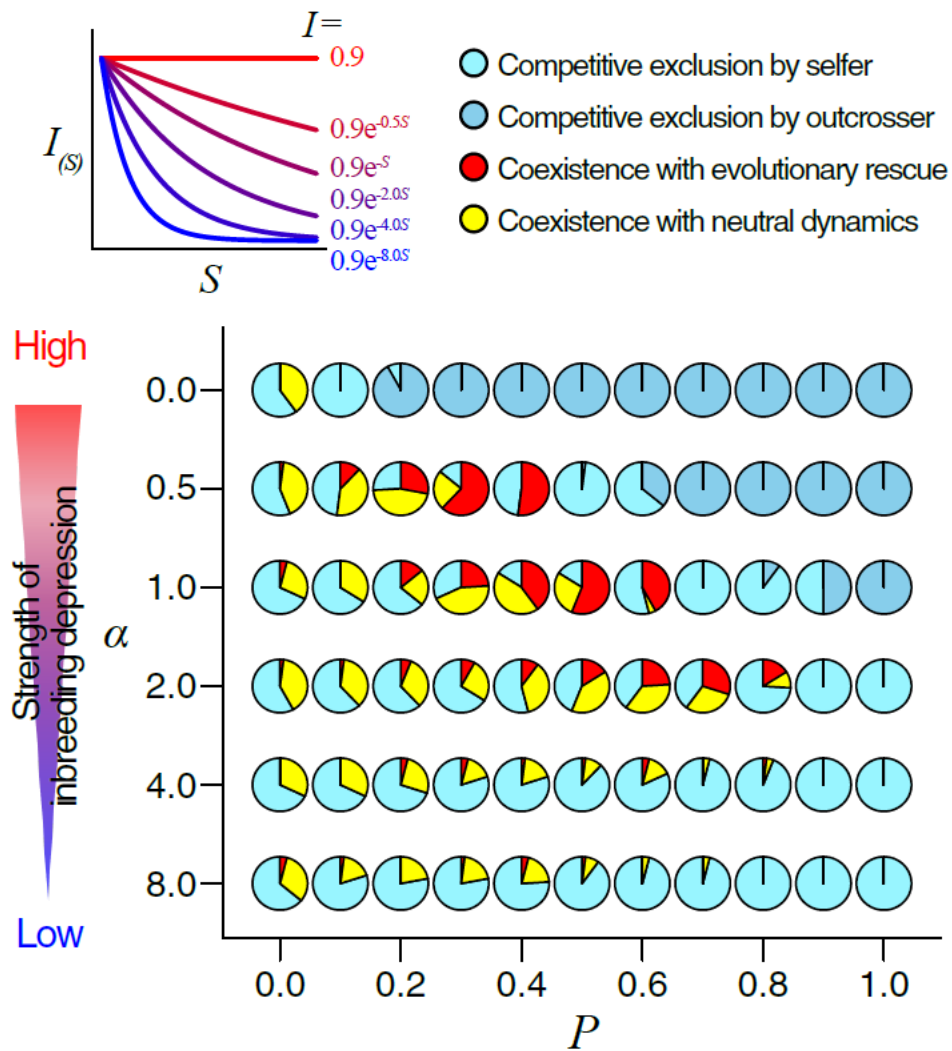
**Fig. 3**. Effects of pollinator availability and the strength of inbreeding depression on the simulation consequence in variable inbreeding depression scenarios. Each pie chat shows frequencies of four categories of eco-evolutionary consequences of 50 simulation runs (Fig. 1).

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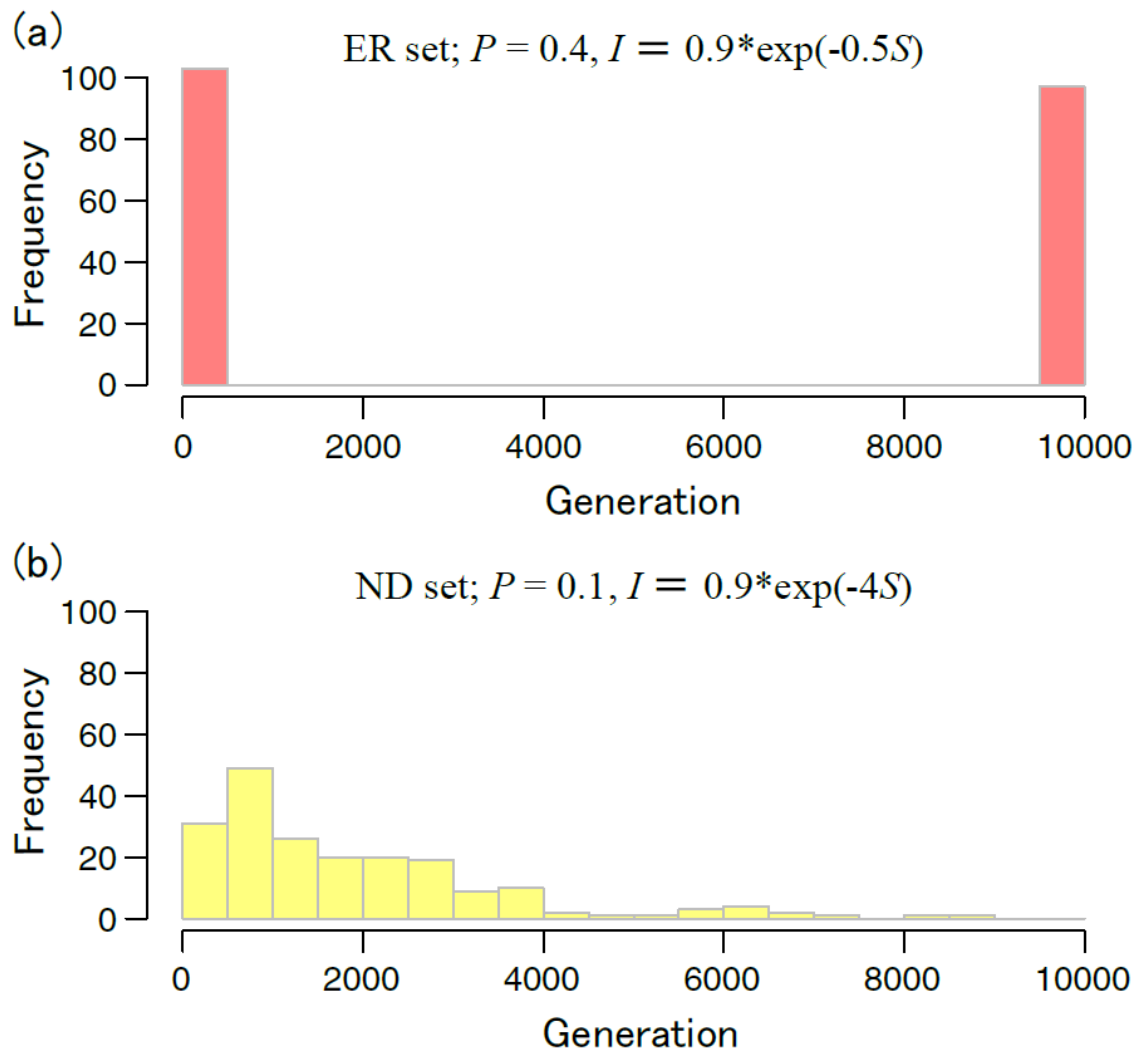
Fig. 4. Histograms of generation until when two species coexisted in two typical

parameter sets for coexistences with evolutionary rescue (a, ER set) and with neutral

dynamics (b, ND set), respectively. The graphs made from the outcomes of 200

simulation runs.
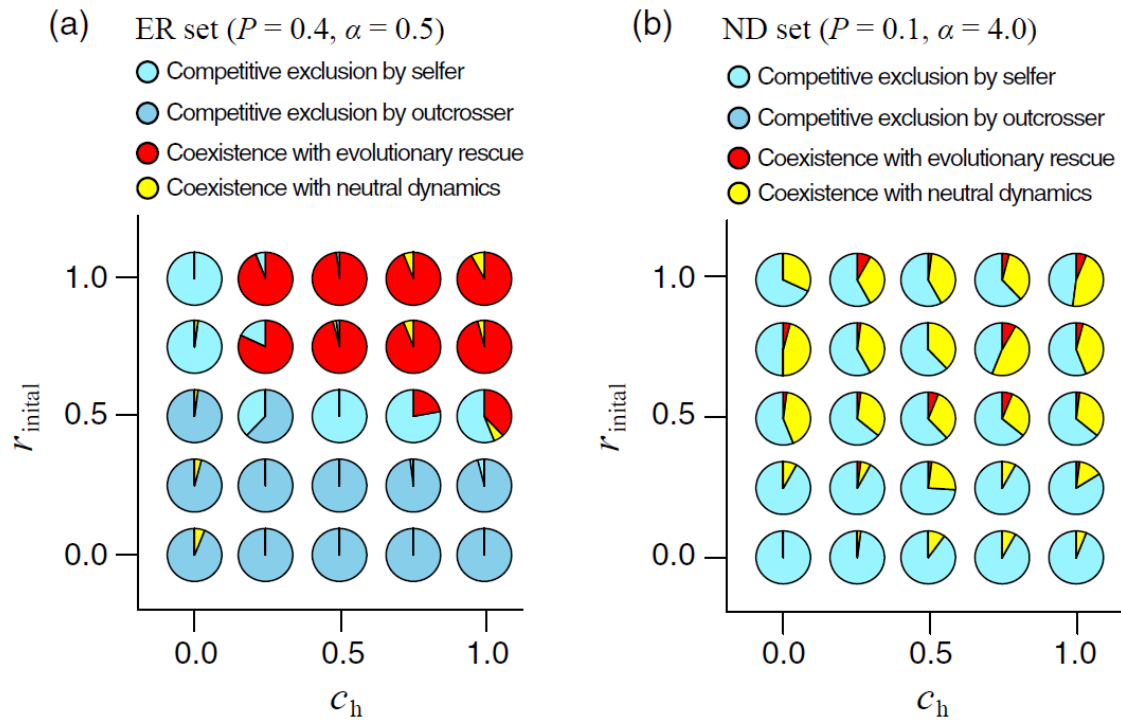
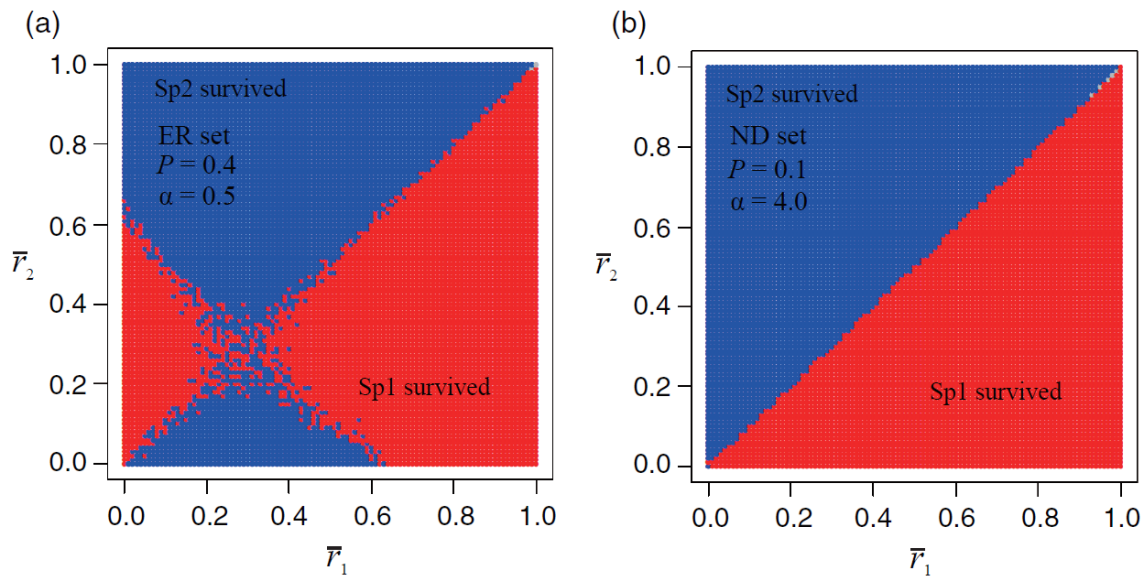**Fig. 5**. Effects of strength of reproductive interference and initial population's mean selfing rate in two typical parameter sets for coexistences with evolutionary rescue (a, ER set) and with neutral dynamics (b, ND set), respectively. Each pie chat shows frequencies of four categories of eco-evolutionary consequences of 50 simulation runs (Fig. 1).
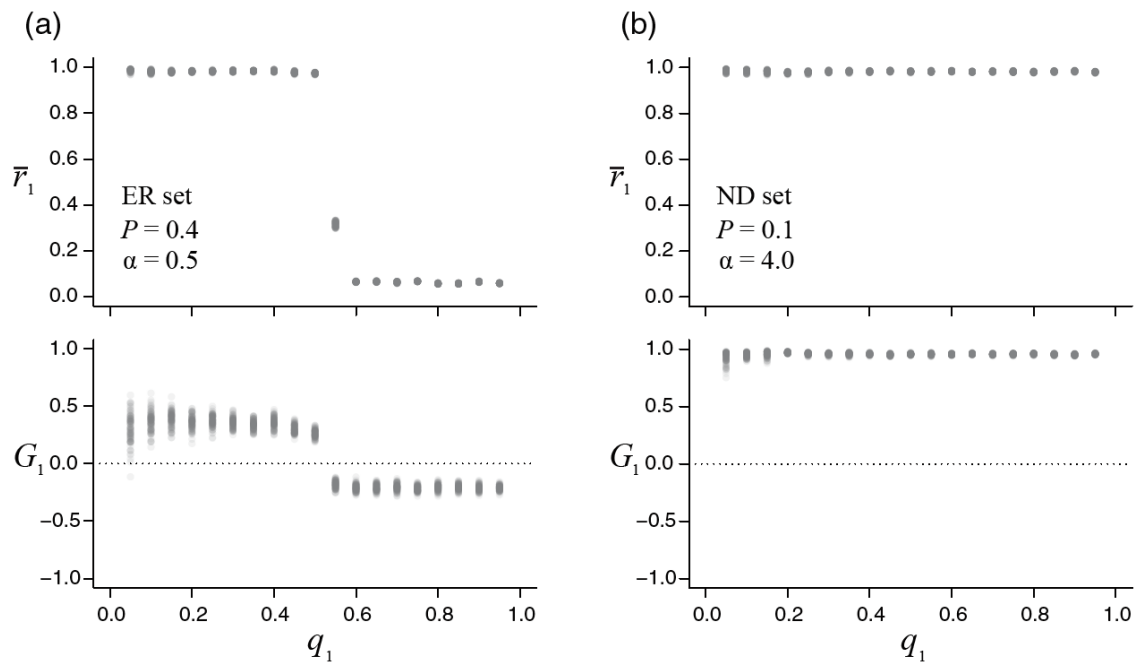
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Fig. 6. Ecological consequences with fixed population's prior selfing rate in two typical

parameter sets for coexistences with evolutionary rescue (a, ER set) and with neutral

dynamics (b, ND set), respectively. X and Y axes indicate population's mean prior

selfing rate of sp_1 and sp_2, respectively. Blue and red areas mean that survivor is sp_1 and

sp_2, respectively, and grey regions (shown upper right corner of each panel) indicated

that coexistence continued for 2,000th generations.

653

Fig. 7. Evolutionary consequences with fixed relative abundance in two typical parameter sets for coexistences with evolutionary rescue (a, ER set) and with neutral dynamics (b, ND set), respectively. Upper and lower graphs indicate the correlations between population's mean prior selfing rate and selection gradient, and relative abundance of the focal species in last 500 of 2,000 generations, respectively.