




Effect of hybridization with genome exclusion on extinction risk

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Abstract: Human-induced habitat changes may lead to the breakdown of reproductive barriers between distantly related species. This phenomenon may result in fertile first-generation hybrids (F_1) that exclude the genome of one parental species during gametogenesis, thus disabling introgression. The species extinction risk associated with hybridization with genome exclusion is largely underappreciated because the phenomenon produces only F_1 hybrid phenotype, leading to the misconception that hybrids are sterile and potentially of minor conservation concern. We used a simulation model that integrates the main genetic, demographic, and ecological processes to examine the dynamics of hybridization with genome exclusion. We showed that this mode of hybridization may lead to extremely rapid extinction when the process of genome exclusion is unbalanced between the interbreeding species and when the hybridization rate is not negligible. The coexistence of parental species was possible in some cases of asymmetrical genome exclusion, but show this equilibrium was highly vulnerable to environmental variation. Expanding the exclusive habitat of the species at risk allowed its persistence. Our results highlight the extent of possible extinction risk due to hybridization with genome exclusion and suggest habitat management as a promising conservation strategy. In anticipation of serious threats to biodiversity due to hybridization with genome exclusion, we recommend a detailed assessment of the reproductive status of hybrids in conservation programs. We suggest such assessments include the inspection of genetic content in hybrid gametes.

Keywords: biodiversity, demographic flow, extinction, hemiclones, hybridogenesis, invasive species

Efectos de la Hibridación con Exclusión Genómica sobre el Riesgo de Extinción

Resumen: Los cambios en el hábitat inducidos por el humano pueden derivar en la disrupción de las barreras reproductivas entre especies relacionadas lejanamente. Este fenómeno puede resultar en híbridos fértiles de primera generación (F_1) que excluyen el genoma de una de las especies parentales durante la gametogénesis, inhabilitando así la introgresión. El riesgo de extinción de la especie asociado con la hibridación con exclusión genómica es poco apreciado debido a que el fenómeno produce solamente el fenotipo híbrido F_1 , lo que conduce al malentendido de que los híbridos son estériles y potencialmente de menor interés para la conservación. Usamos un modelo de simulación que integra a los principales procesos genéticos, demográficos y ecológicos para examinar las dinámicas de hibridación con exclusión genómica. Mostramos que este modo de hibridación puede llevar a una extinción extremadamente rápida cuando el proceso de exclusión genómica

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no está balanceado entre las especies que se cruzan y cuando la tasa de hibridación no es insignificante. La coexistencia de especies parentales fue posible en algunos casos de exclusión genómica asimétrica, pero mostraron que este equilibrio era altamente vulnerable a la variación ambiental. La expansión del hábitat exclusivo de las especies en riesgo permitió su persistencia. Nuestros resultados resaltan el alcance del riesgo de una posible extinción por causa de la hibridación con exclusión genómica y sugieren al manejo del hábitat como una estrategia de conservación prometedora. En anticipación a amenazas serias para la biodiversidad debidas a la hibridación con exclusión genómica, recomendamos una evaluación detallada del estado reproductivo de los híbridos dentro de programas de conservación. Sugerimos que dichas evaluaciones incluyan la inspección del contenido genético en los gametos híbridos.

Palabras Clave: biodiversidad, especie invasora, extinción, flujo demográfico, hemiclones, hibridogénesis

摘要: 人类引起的生境变化可能打破远源物种之间的生殖隔离。这可能产生在配子形成时排斥亲本中一方基因组的可育第一代杂种 (F_1), 因而基因渐渗不会发生。与存在基因组排斥的杂交相关的物种灭绝风险远远被低估了, 这是因为这个现象只会产生 F_1 代杂种表型, 使人误以为杂种不可育, 可能不需要重点关注。我们用整合了遗传学、种群统计学和生态学过程的模拟模型来研究存在基因组排斥时的杂交动态。我们发现当基因组排斥在两个杂交的物种间不对等, 或是杂交率不可忽略时, 这样的杂交可能导致非常快速的灭绝。在基因组排斥不对等的某些情况下, 亲本物种有可能共存, 但这样的平衡很容易因环境变化而被打破。另外, 扩大有灭绝风险的物种独有的生境可以维持它们的续存。我们的结果突出了因存在基因组排斥的杂交导致的灭绝风险的程度, 并表明生境管理可以作为一种可行的保护策略。考虑到有基因组排斥的杂交可能对生物多样性造成严重威胁, 我们建议对保护项目中杂种的繁殖状况进行详细评估, 并且要检查杂种配子中的遗传物质。【翻译: 胡恰思; 审校: 魏辅文】

关键词: 杂种发生, 半克隆, 入侵物种, 种群流动, 灭绝, 生物多样性

Introduction

Interspecific hybridization is enhanced by human-induced disturbances such as habitat modification, species translocation, and climate change (Todesco et al. 2016). The interbreeding between distantly related species may lead to the production of fertile first-generation hybrids (F_1), which produce gametes containing the nonrecombined genetic material of one parent; that is, the genome of the other parent is excluded during gametogenesis (e.g. Tinti & Scali 1995; Som & Reyer 2006; Mee & Taylor 2012). Consequently, the offspring of F_1 hybrids is either a pure parental individual or a regenerated F_1 hybrid, depending on the mate (Yakovlev et al. 2000). Hybridization with genome exclusion may result in the extinction of 1 parental species through progressive replacement of that individuals genetic material with that of the other parent, a process referred to as demographic flow (Quilodrán et al. 2015).

In a hybrid system with genome exclusion, no recombination between homologous chromosomes occurs during meiosis and no introgressions of nuclear genes take place. The F_1 hybrids produce nonrecombinant haploid gametes: either a single type of gametes due to the systematic exclusion of one of the parental genomes (non-alternative genome exclusion) or 2 types of gametes that each contain 1 of the 2 parental genomes (alternative genome exclusion) (Fig. 1). Thus, F_1 hybrids clonally transmit the haploid genome of one or both of the parents. This process may result in hybridogenetic, gynogenetic, parthenogenetic, or polyploid forms (Lampert 2009).

Examples are frequently observed in freshwater fishes of the families Atherinidae, Cobitidae, Cyprinidae, and Poeciliidae (Schmidt et al. 2011). For example, the hybrids between 2 North American cyprinids, *Phoxinus eos* and *P. neogaeus*, produce haploid gametes of 1 parent, thereby potentially regenerating pure, nonhybrid nuclear genomes (Goddard & Schultz 1993). The regenerated *P. eos* was originally believed to be rare in nature (Binet & Angers 2005); however, recent studies have revealed the high abundance of such individuals, which have replaced all of the original populations of *P. eos* that lived in sympatry with hybrids (Mee & Taylor 2012). As the original crosses occurred between female *P. neogaeus* and male *P. eos*, all the hybrids and regenerated parents shared the mtDNA haplotypes of *P. neogaeus*. Therefore, it is unknown whether a similar replacement has occurred in *P. neogaeus* (Angers & Schlosser 2007; Mee & Taylor 2012).

Another example involves the F_1 hybrids between 2 European freshwater cyprinids, roach (*Rutilus rutilus*) and bream (*Abramis brama*). These hybrids are fertile (Wood & Jordan 1987); however, their progeny were long believed to be unfertile due to the absence of post- F_1 hybrid phenotypes observed in nature (Pitts et al. 1997; Hayden et al. 2010). However, detailed examination of the gametes produced by F_1 hybrids confirmed that they each contain the nonrecombined haploid genome of one or the other parental species and could potentially regenerate roach and bream (Yakovlev et al. 2000).

In other hybrid systems, F_1 hybrids may regenerate only one parental species, as in the hybridization between *Poeciliopsis monacha* and *P. lucida*. The F_1 hybrids that

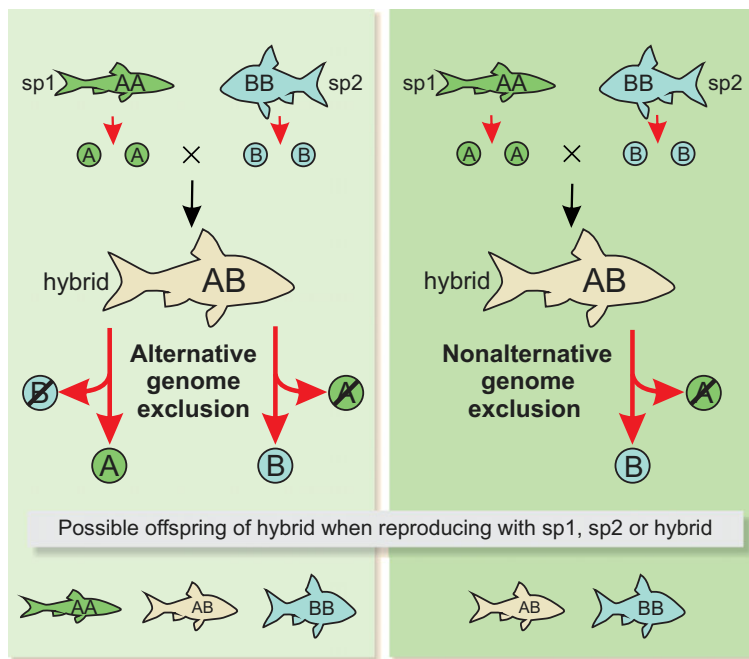


Figure 1. Interspecific hybridization with genome exclusion. Two types of genome exclusion are possible: alternative and nonalternative. This refers to the production of parental gametes by hybrids. Depending on the type of genome exclusion, either one or both parental species are regenerated by the hybrids in addition to the hybrid itself. The circles represent gametes.

produce gametes of *P. monacha* restore the *P. monacha* genome when crossing with *P. monacha*, whereas new hybrids are produced when they cross with *P. lucida* (Cimino 1972). Hybridization with genome exclusion has also been documented in stick insects of the genus *Bacillus* (Mantovani & Scali 1992), in the fish genus *Squalius* (Crespo-Lopez et al. 2006), and in *Ambystoma* salamanders (Charney 2012).

Hybridization with genome exclusion occurs in water frogs of the genus *Pelophylax*. In Western Europe, hybrids between *P. lessonae* and *P. ridibundus*, currently known as *P. esculentus*, discard the *P. lessonae* genome during gametogenesis and produce gametes that contain the haploid genome of *P. ridibundus*. The F_1 hybrids are thus regenerated when the hybrids mate with *P. lessonae*. Because the crosses between two hybrids are unviable and *P. ridibundus* retracted out of Western Europe during the last ice age, the system that comprises *P. lessonae* and the hybrid *P. esculentus* (LE system) has remained in equilibrium for 10,000 years. Recent translocations of *P. ridibundus* from Eastern Europe, where this species occurs naturally, to France and Switzerland during the 20th century are currently destabilizing the LE system. Hybridization between the introduced species and the native ones reinforces the invasive process and may lead to the potential replacement of both native frogs (Holsbeek & Jooris 2010). In a recent study, we used an original model to assess the conditions under which interspecific hybridization can lead to population extinction; the results provide a means to evaluate the efficiency of conservation actions (Quilodrán et al. 2015).

In our earlier model and in previous attempts by others (e.g. Hellriegel & Reyer 2000; Som & Reyer 2006), hy-

bridization with genome exclusion has been considered as a deterministic process and environmental variations have not been incorporated into the system. We made 2 improvements to our general model. First, we integrated environmental stochasticity. Second, we considered for the first time two types of genome exclusions: an alternative type in which one hybrid individual can produce gametes containing the pure haploid genome of one or the other parental species (in varying proportions) and a non-alternative type in which hybrids produce gametes containing the haploid genome of only one parental species, excluding systematically the other parental genome. We also devised guidelines to help stakeholders identify this threat, which requires rapid conservation action.

Methods

Our aim was to assess the threat interspecific hybridization with genome exclusion may represent for one or both parental species. We improved a previously published model of interspecific hybridization in which hybrids are unfertile or fertile but do not undergo chromosomal recombination during gametogenesis (Quilodrán et al. 2014, 2015) (Supporting Information).

There are three possible forms in our theoretical case study: two parental species and hybrids: 0, 1, and $\frac{1}{2}$. The parameter values are based on a previously published case study of Western Europe water frogs that undergo hybridization with genome exclusion (Quilodrán et al. 2015). We took from that study the parameter values corresponding to *Pelophylax ridibundus* but did not overlap generations (annual survival of adults set to zero)

Table 1. List of functions, parameters, and default values in the model of hybridization with genome exclusion.

Symbol	Definition
N_i	number of adult individuals of genotypic class i initial size: $N_0 = N_1 = 50$; $N_{1/2} = 0$
n_i	weighted number of matings that leads to offspring of class i
R_i	population growth rate
$\delta_{(t)}$	product of all of the stages before sexual maturity
AC	temporal autocorrelation
G	proportion of exclusion between parental genomes
Θ	time delay from hatching to age at maturity in $t + 1$ $\theta_0 = \theta_1 = \theta_{1/2} = 1$ (2 years)
C	clutch size $c_0 = c_1 = c_{1/2} = 1250$
S^0	survival of tadpoles $S_0^0 = S_1^0 = S_{1/2}^0 = 0.012$
S^1, S^2	survival of first- and second-year juveniles $S_0^1 = S_1^1 = S_{1/2}^1 = 0.5$; $S_0^2 = S_1^2 = S_{1/2}^2 = 0.4$
A	interspecific competition coefficient $\alpha_{01} = \alpha_{10} = 0$; $\alpha_{0^{1/2}} = \alpha_{1^{1/2}} = \alpha_{1/2^{1/2}} = \alpha_{1/2^{1/2}} = 0.5$
V	habitat size $V_0 = V_1 = V_{1/2} = 5000$
γ	interbreeding success rate $\gamma = \gamma_{01} = \gamma_{10} = 0 - 1$

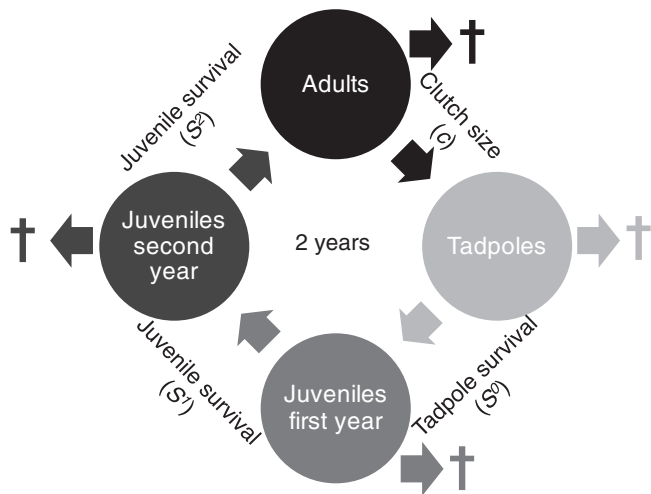


Figure 2. Life cycle of the theoretical species described in the model of hybridization with genome exclusion. Individuals take 2 years to reach sexual maturity. Equal sex ratio is assumed and parameter values are the same for parental species and hybrids. Parameter values are from a case of water frogs in Western Europe that undergo hybridization with genome exclusion (Quilodrán et al. 2015).

(Table 1). *Pelophylax ridibundus* is an invasive species that can hybridize with other *Pelophylax* water frogs (i.e., *P. perezi* and *P. lessonae* [Holsbeek & Jooris 2010]) and generate F_1 hybrids that undergo genome exclusion during their gametogenesis. Specimens of the *Pelophylax* complex take 2 years to reach sexual maturity, after going through tadpole and juvenile stages (Fig. 2). We assumed

the same demographic parameter values and equal sex ratios for all the interacting *Pelophylax* forms.

Depending on the species pairs that undergo hybridization with genome exclusion, the hybrids can generate gametes that either contain exclusively the genome of one of the two parental species or that contain the genome of one or the other parents in varying proportions (Yakovlev et al. 2000; Mee & Taylor 2012). To include this phenomenon in our model we introduced a parameter g to assess the effect of genome exclusion on different proportions. With this parameter, it is possible to model scenarios from the systematic exclusion of a given genome, either 00 or 11 ($g = 0$ or 1, respectively) to the exclusion of one or the other parental genome in a balanced way ($g = 0.5$). The parameter g is thus introduced in Eq. 3 of Quilodrán et al. (2014). It is defined for each interacting form in the following equations:

$$n_{0(t)} = N_{0(t)}M_{00} + (1 - g) \left(N_{0(t)}M_{0^{1/2}} + N_{1/2(t)}M_{1/20} + N_{1/2(t)}M_{1/2^{1/2}} \left(\frac{1}{2} + \left| g - \frac{1}{2} \right| \right) \right), \quad (1)$$

$$n_{1(t)} = N_{1(t)}M_{11} + g \left(N_{1(t)}M_{1^{1/2}} + N_{1/2(t)}M_{1/21} + N_{1/2(t)}M_{1/2^{1/2}} \left(\frac{1}{2} + \left| g - \frac{1}{2} \right| \right) \right), \text{ and} \quad (2)$$

$$n_{1/2(t)} = N_{0(t)}M_{01} + N_{1(t)}M_{10} + N_{1/2(t)}M_{1/2^{1/2}} \times \left(\frac{1}{2} - \left| g - \frac{1}{2} \right| \right) + g \left(N_{0(t)}M_{0^{1/2}} + N_{1/2(t)}M_{1/20} \right) + (1 - g) \left(N_{0(t)}M_{1^{1/2}} + N_{1/2(t)}M_{1/21} \right), \quad (3)$$

where M_{ij} is the probability of individuals i mating with individuals j , which is estimated as $M_{ij(t)} = \gamma_{ij}N_{j(t)}/\varphi_{i(t)}$, where $\varphi_{i(t)}$ is a normalization factor such that $\sum_i M_{ij} = 1$ and γ is the interbreeding success rate. Reproduction between the two parental species is controlled by γ . There is no reproduction between the interacting species when $\gamma = 0$, whereas reproduction is panmictic when this value is 1; intermediate values represent nonrandom mating preferences.

Equations 1, 2, and 3 are based on the assumption of Mendelian inheritance and equal fitness for all three forms. Therefore, if the value of g were equal to 0.5, it would be reduced to a case of single biallelic genes. These equations account for the presence of hybrids of both sexes. However, it is also possible to simulate cases in which hybrids display a sex bias. Supporting Information has an example of implementation of our equations in simulations that consider only female hybrids. Hybridization with genome exclusion generally produces only female hybrids (Burt & Trivers 2006), but a mix of both

sexes (Lehtonen et al. 2013) or only male hybrids have been documented (Schmidt et al. 2011).

The number of individuals that reach sexual maturity after intra- and interspecific density-dependent effects was computed with an extension of the Ricker model. This function is useful for species with a larval stage (Wilbur 1976) and in which distant hybridization has been observed frequently (i.e., fishes and amphibians).

$$N_{k(t+1)} = \text{round} \left[N_{k(t)} S_k + R_k n_{k(t-\theta)} e^{\left(-\frac{(n_{k(t-\theta)} + \sum_{k \neq l} \alpha_{kl} n_{l(t-\theta)})}{V_k} \right)} \right], \quad (4)$$

We rounded off the resulting value of Eq. 4 to account for the ‘‘lattice effect,’’ in which unexpected outputs may appear due to the noncontinuous but discrete nature of the individuals in a population (Henson et al. 2001). The first term of Eq. 4 denotes the number of adults that survive from one reproductive season to the next, in which S_k is the adult survival of individuals of class k . The second term denotes the number of offspring that survive until sexual maturity, where θ indicates the time to reach maturity (i.e., $\theta + 1$ in time $t + 1$). The parameter R_k is the number of progeny that survive until sexual maturity or the population growth rate; α_{kl} is the amount of competition between members of different classes relative to the amount of competition achieved by individuals of the same class k ; and V_k is habitat size as introduced by Henson et al. (2001). We assumed no competition between parental taxa ($\alpha_{01} = \alpha_{10} = 0$) and an intermediate level of competition between parental taxa and hybrids ($\alpha_{01/2} = \alpha_{1/20} = \alpha_{1/21} = \alpha_{1/21} = 0.5$). We made this assumption because hybridization with genome exclusion usually occurs between distantly related species and generally involves negligible competition between them and hybrids using intermediate habitats (e.g. Holenweg Peter et al. 2002; Hayden et al. 2011). Nevertheless, Supporting Information contains simulations incorporating competition between parental species.

We were also interested in introducing environmental stochasticity into the model. We incorporated it by adding a temporal dynamic to the growth-rate parameter estimated in eq. 8 of Quilodr  n et al. (2015), which accounts for temporal autocorrelation through the yearly survival of juvenile stages as follows:

$$R_{K(t)} = c_k \delta_{(t)}, \quad (5)$$

where c_k is the clutch size and $\delta_{(t)}$ is the product of all the survival probabilities across the developmental stages (S_k^l) before reaching sexual maturity ($\delta_{(0)} = \prod_{l=1}^b S_k^l$). The temporal dynamic is included through a beta distribution with mean $\delta_{(0)}$ and variance $\sigma^2 = (\delta_{(0)} - \delta_{(t)} \text{AC})^2$:

$$\delta_{(t+1)} = \delta_{(t)} \text{AC} + (1 - \text{AC}) \beta(\delta_{(0)}, \sigma^2), \quad (6)$$

where AC characterizes temporal autocorrelation and can take values ranging from 0 (completely stochastic process) to 1 (completely deterministic process).

We tested different values of hybridization frequency, given by the interbreeding success rate parameter (γ), to assess its combined effect with different proportions of genome exclusion (g), levels of temporal autocorrelation (AC), and habitat sizes (V) on the frequency of each genome. We simulated 200 generations of independent evolution for each parental species after the onset of hybridization. We present the results after a total of 400 generations. At this point all simulations had reached a stationary state.

By modeling interspecific hybridization with genome exclusion between virtual parental species, we projected the potential impact of this type of hybridization on species persistence. We considered diploid organisms with hybrid offspring that undergo different proportions of genome exclusion. We simulated two parental species (0 and 1) with genomes (00 and 11). Hybrids either systematically regenerate a single parental species when the proportion of genome exclusion is completely skewed to 0 or 1 or alternate between them with intermediate values of genome exclusion. Values of genome exclusion less than or greater than 0.5 favor the genomes 00 and 11, respectively.

Results

Because of the direct replacement of parental species by hybrid offspring, the extinction of one species was reached in a few generations when genome exclusion was completely skewed toward one parental species (Fig. 3a). In this case, when hybridization increased to the critical point at which the abundance of hybrids exceeded that of the nonregenerated species, both the hybrids and the nonregenerated species went extinct. At this point, the community was composed exclusively of the species favored by the genome exclusion. In the case where hybrids produced gametes of both parental species in equal proportions, the hybrids reached maximum abundance. When the values of genome exclusion varied from 0.2 to 0.8 and the interbreeding rate between both parental taxa was < 0.4 , all three forms persisted in the community. At smaller and larger values of genome exclusion and at higher interbreeding rates, the extinction of the species that was not favored by the genome exclusion occurred rapidly (27 generations). This extinction occurred even more rapidly when the genome exclusion was completely skewed toward one species and the interbreeding was panmictic (6 generations).

Environmental stochasticity was incorporated by varying the temporal autocorrelation of the yearly survival of the offspring (parameter AC). We considered a case in which the 3 forms persisted in the community even when

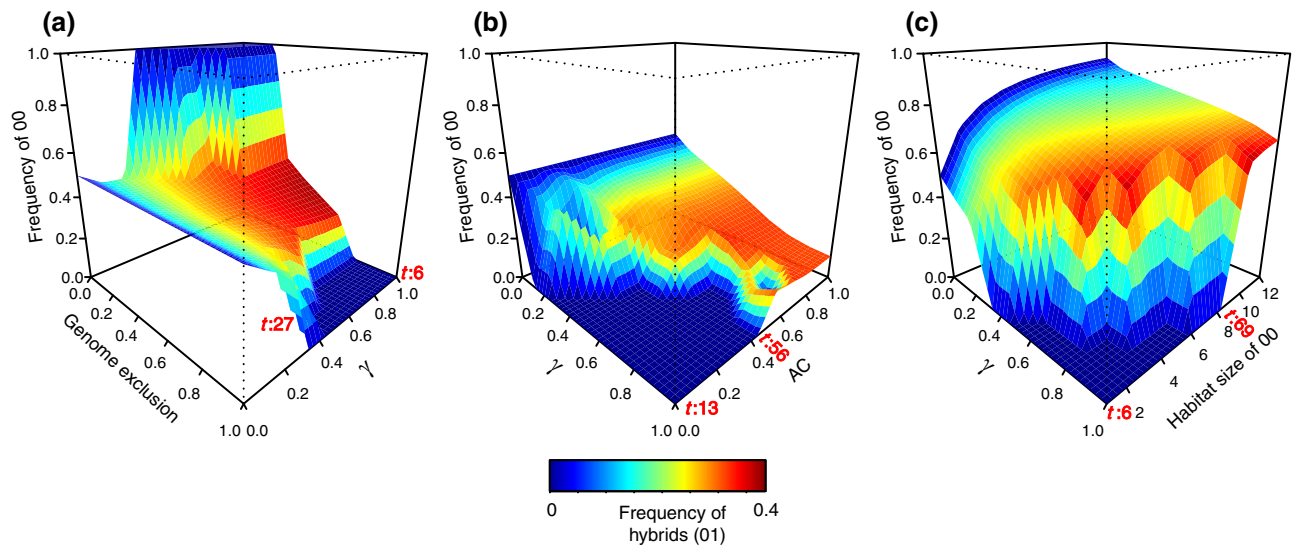


Figure 3. Simulation of the demographic impact of genome exclusion. We considered 2 parental species N_0 and N_1 with genomes 00 and 11, respectively. The hybrid $N_{1/2}$ has the genome 01 and generates fertile offspring that undergo genome exclusion. The frequency of hybridization is estimated through the interbreeding success rate (γ). The frequency of the genome 00 is estimated as $N_0/(N_0 + N_{1/2} + N_1)$ and that of the hybrid genome 01 as $N_{1/2}/(N_0 + N_{1/2} + N_1)$. (a) Proportion of genome exclusion in hybrid offspring (values from 0 to 1, systems ranging from completely skewed toward genome 00 [N_0] to completely skewed toward genome 11 [N_1]). (b) Incorporation of stochasticity in the model through autocorrelation in time (AC) (AC range 0–1; 0, completely stochastic model; 1, totally deterministic model). System is skewed toward one parental species but has all of the 3 forms (i.e., 2 parental species and the hybrids can persist in a deterministic and panmictic system; 0.75 of genome exclusion favoring genome 11). (c) Minimum habitat size required for species of genome 00 to avoid complete extinction when the exclusion is completely skewed toward the species with genome 11 and when reproduction is panmictic between species (t , number of generations in which interspecific hybridization with genome exclusion leads to the extinction of one of the parental species). No hybridization between the parental species was simulated during the first 200 generations. Data correspond to a situation projected after 400 generations.

reproduction was panmictic and genome exclusion was skewed toward 1 of the parental species (0.75 of genome exclusion, favors species 1 [Fig. 3a]). Even when all 3 forms were in equilibrium at different abundances in the deterministic model, the species that was not favored by the genome exclusion went extinct when environmental stochasticity was incorporated in the model under the same conditions (Fig. 3b). Thus, even when the apparent coexistence equilibrium of the 3 forms can be observed in nature, the long-term output will be strongly dependent on environmental variation.

We considered the worst-case scenario of genome exclusion (Fig. 3a), that is when it is completely skewed in favor of one species, to assess the minimum habitat size of the other species required to avoid extinction (in our simulations, favoring species 1, Fig. 3c). Increasing the exclusive habitat for the species that was disadvantaged due to genome exclusion (species 0) lengthened the time before extinction and eventually allowed the species to persist even when hybridization was at a maximum ($\gamma = 1$). In this case, because the reproduction between conspecific or heterospecific partners

was random, the reproductive cost for the species disadvantaged by the genome exclusion was at a maximum; thus, extinction was reached extremely rapidly (6 generations) when the habitat size was equal between species. In this critical case, the habitat size of a threatened species that is nine times larger than that of the favored species may save the threatened species from extinction (Fig. 3c).

When only female hybrids were produced, the number of generations to reach extinction was slightly higher (see Supporting Information). However, the general pattern of rapid extinction, much faster when the genome exclusion was skewed toward a single species, was still observed when only female hybrids were considered in the simulations. When interspecific competition between parental species was incorporated into the model and genome exclusion was marginally skewed in favor of one parental species, interspecific competition led to more rapid extinction of both hybrids and the disadvantaged parental species. A competitive advantage of the species disadvantaged by genome exclusion counterbalanced the negative effect of the demographic flow,

but rapid extinction still occurred with a high level of hybridization (Supporting Information).

Discussion

Hybridization with Genome Exclusion

Our results highlight the extreme consequences of hybridization with genome exclusion on species persistence. The direct replacement of a parental species by hybrid offspring lead to the collapse of one species within a few generations when the exclusion was skewed toward the other species. We thus demonstrated that such hybridization may precipitate extinction and showed this is strongly facilitated when environmental stability is challenged. The hybridizing species were also more affected by stochastic external stress such as extreme environmental changes. Therefore, the increasing frequency of extreme weather events worldwide (Cai et al. 2014) is an additional threat to the long-term coexistence of some apparently stable hybridogenetic and gynogenetic systems (Som & Reyer 2006; Mee & Otto 2010). It is thus important to consider environmental predictability when projecting the fate of interbreeding species in the context of extreme meteorological events such as those produced by global climate change.

We considered conservation actions that could improve species persistence by improving the habitat of the disadvantaged species. Because hybridization with genome exclusion is generally observed between distant species (Yakovlev et al. 2000), these species have differentiated habitat requirements (Holenweg Peter et al. 2002; Toscano et al. 2010). We highlight that taking advantage of these differences and managing the habitat in favor of the handicapped species is a good way to avoid species loss. Habitat management is therefore emphasized as a potential way to protect species affected by hybridization and genome exclusion.

Selfish Genetic Elements

Genome exclusion of the nonalternative form and the alternative form with unbalanced production of parental gametes may be seen as a particular case of meiotic drive, in which allelic variants influencing the production of gametes ensure their transmission in higher proportion than expected under Mendel's law of equal segregation (Lindholm et al. 2016). Meiotic drive may affect a single locus or full chromosomes. The process occurring in hybridization with genome exclusion would correspond to a meiotic drive affecting all the chromosomes of one parental species. There is a range of literature on meiotic drive that may be applied to various cases of hybridization with genome exclusion (e.g. Sandler & Novitski 1957; Wood & Newton 1991; Finseth et al. 2015). Most of this

literature highlights the rapid fixation rate or the strong selection against the so-called selfish genetic elements if they reduce organism fertility (e.g. Nachman & Searle 1995; Jaenike 2001; Lindholm et al. 2016). This may also happen in hybridization with genome exclusion (Burt & Trivers 2006). In other cases, hybrid frequency can be maintained at intermediate levels by the evolution of behavioral mechanisms such as density-dependent mate choice (e.g. Moore 1976; Som et al. 2000; Mee & Otto 2010).

In previous models that simulate meiotic drive, efforts have been made to show the potential rapid fixation or the extinction risk associated to selfish genetic element (Holman et al. 2015; Hall & Dawe 2017). Gardner and Ross (2014) developed a mathematical model to explain a pattern of genome elimination due to selfish genetic elements and found that inbreeding promotes paternal genome elimination in the heterogametic sex, potentially leading to population extinction.

Real incidences of meiotic drive are difficult to evaluate in nature because the fixation of the selfish genetic elements may be reached in few generations (Jaenike 2001; Manser et al. 2011). Laboratory experiments with *Drosophila* show allelic fixation in less than 10 generations and a potential for population extinction when the segregation of a single sex chromosome is stimulated (Lyttle 1991).

However, hybridization with genome exclusion has the potential to produce outcomes differing from those of meiotic drive. This is the case, for instance, when alternative genome exclusion results in a balanced production of clonal gametes of both parental species. Previously developed models of meiotic drive cannot be applied to cases of interspecific hybridization with genome exclusion because such models consider intraspecific reproduction, panmictic among all interacting forms (e.g. Gardner & Ross 2014; Holman et al. 2015; Hall & Dawe 2017). Moreover, hybridization with genome exclusion most often occurs between distantly related species with species-specific ecological features that may contribute to the dynamics of the system (Holenweg Peter et al. 2002; Toscano et al. 2010; Hayden et al. 2011). This phenomenon is therefore driven by outbreeding rather than by an inbreeding effect. Our model was designed to fit most cases of hybridization with genome exclusion, including important demographic and ecological parameters of the interbreeding species and their hybrid form. For instance, it allows the exploration of various levels of hybridization rate and of how particular ecological or behavioral features may counterbalance the demographic flow against one of the interacting species.

Evolutionary Origin

The evolutionary origin of hybridization with genome exclusion is poorly known (Yamada et al. 2015). One

possible explanation posits that some selfish genetic elements present in all chromosomes were fixed through meiotic drive in the genome of one parental species. When this species hybridizes with another species devoid of such selfish genetic elements, the phenomenon of genome exclusion is observed in the meiosis of the hybrids (Burt & Trivers 2006). However, other explanations are also possible, thus further research is needed to disentangle the most likely origin of genome exclusion.

Persistence of Hybrids

We found that in a system of hybridization with genome exclusion, the three forms have the potential to persist and to be ecologically successful until reaching some equilibrium situation. In cases of hybridization with genome exclusion, the long-term persistence of the hybrid form may depend on several factors. Hybridogenetic systems in which the hybrids undergo nonalternative genome exclusion, for instance, produce gametes with the genome of parental species A, have hybrids that reproduce only with parental species B, and the genomic contribution of species A is clonally transmitted (e.g., Hellriegel & Reyer 2000). The absence of recombination in this part of the hybrid genome may reduce the adaptive potential of the hybrids in front of changing environmental conditions and thus affect their long-term survival (Burt & Trivers 2006). However, in hybridization with genome exclusion in which hybrids can still reproduce with both parental species, the chromosomes that are clonally transmitted by the hybrids can still undergo recombination if they are inherited by a nonhybrid. In this case, the overall reduction in recombination may depend on many factors related to the frequency of hybridization, but the impact on the long-term persistence of the hybrid form is likely less severe.

Incidence of Genome Exclusion

The true incidence of hybridization with genome exclusion in nature is unknown, but approximately 80 taxa of reptiles, amphibians, and fishes clonally transmit a single parental genome (Neaves & Baumann 2011). We focused our analyses on documented cases in animals, but this phenomenon may be widespread in plants. Indeed, hybridogenesis has been observed in plants that undergo genome exclusion after a hybridization event (Kashin 2012; Dubovets & Sycheva 2017). The appearance of selfish genetic elements, resulting in meiotic drive, has also been registered in plants (Nasuda et al. 2005); several cases of interspecific hybridizations have resulted in the elimination of parental chromosomes (Gernand et al. 2005; Comai 2014). We speculate that the incidence of this type of hybridization will increase in the near future because many historically allopatric species are undergoing secondary contact due to human-caused species

translocations or to the effects of climate change, such as habitat modification, changes in migration patterns, and changes in the timing of reproduction.

Guidelines to Identifying the Type of Hybridization

When hybridization with genome exclusion is suspected, we suggest stakeholders inspect the type of gametes produced by hybrids in a conservation program. A set of guidelines is presented in Fig. 4. When two species can hybridize, producing observable F_1 hybrids (with a phenotype that is approximately intermediate between the two parental species), it becomes crucial to understand the reproductive properties of the F_1 hybrids. If F_2 hybrids are observed (with phenotypes that are often intermediate between one parental species and the F_1 hybrid), this indicates the F_1 hybrids are not sterile. In this case, hybridization with genetic introgression is the most probably occurring. If no F_2 hybrids are observable, either the F_1 hybrids are sterile or they are fertile, but they undergo genome exclusion during their gametogenesis. We recommend determining whether the F_1 hybrid is fertile by conducting controlled breeding experiments. If this is not possible, monitoring the demography of the parental species can provide valuable information. We found that when hybrids are sterile, in most cases they do not affect substantially the demography of the parental species. To the contrary, when hybrids are fertile and undergo genome exclusion during gametogenesis, they can dramatically affect the demography of one or both parental species. The monitoring of the sex of hybrids may also bring to light information relevant to the potential production of hybrids undergoing genome exclusion of parental species. Although the production of male and female hybrids or only male hybrids have been documented, most cases of hybridization with genome exclusion favor the production of only female hybrids (Burt & Trivers 2006). Therefore, if demographic decline and sex-biased production of hybrids are observed, it could be indicative of a situation in which hybrids are fertile and undergoing genome exclusion.

To further characterize the type of hybridization with genome exclusion, it is necessary to determine whether the genome exclusion is of the alternative or the nonalternative type by analyzing the genome content of the gametes. Supporting Information and Fig. 4 show a genetic test that discriminates the type of genome exclusion produced by the gametes of hybrids. It is recommended to start with the simplest test, which consists of determining whether genome exclusion is of the nonalternative type, in which case a single allele per gene would be present across all the gametes of an individual. If this test results in the presence of 2 alleles for some loci in the pool of gametes, then the genome exclusion would be of the alternative type. In this case, a single-cell genotyping method should be implemented to determine the fraction

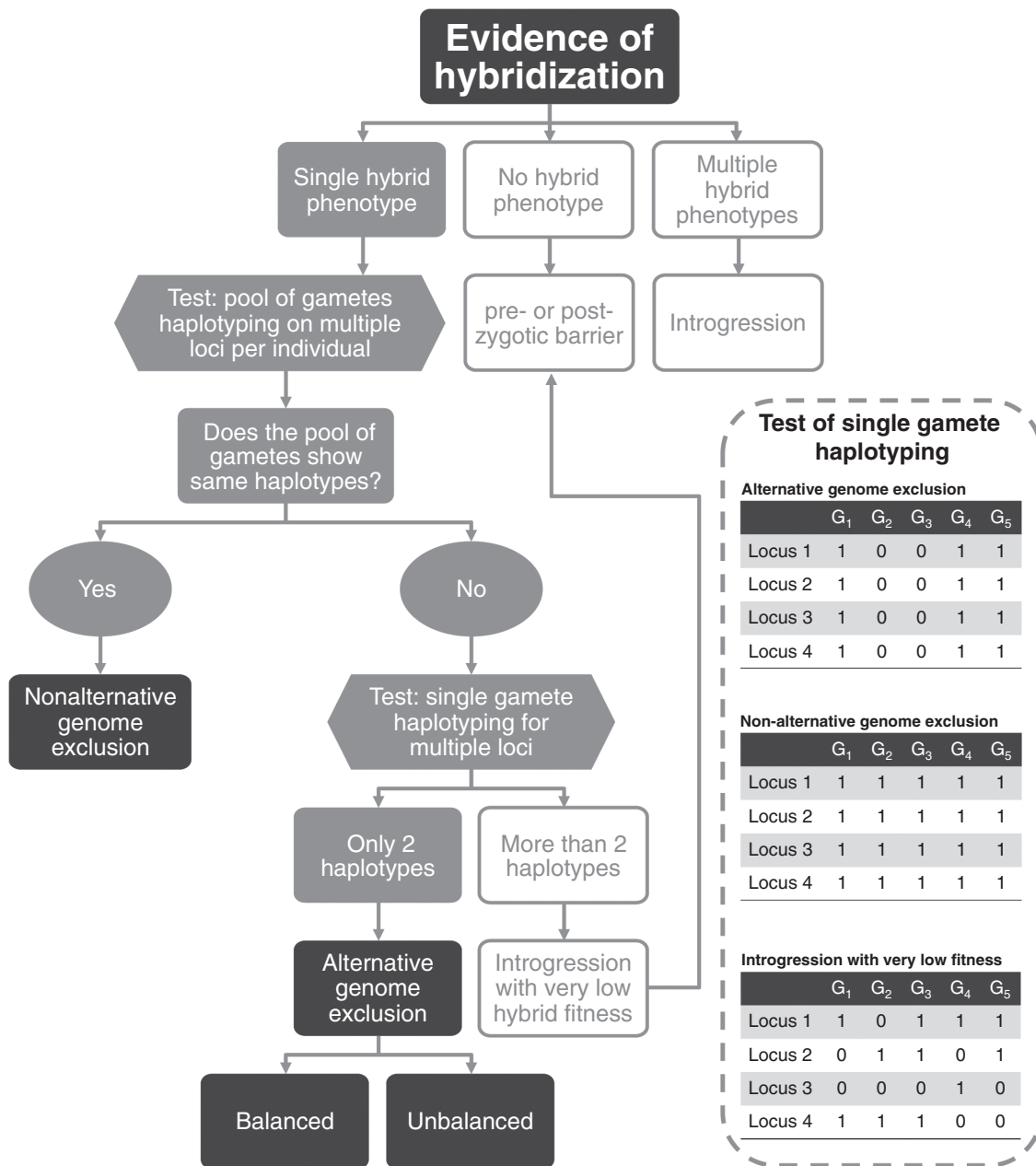


Figure 4. Steps to identify hybridization with genome exclusion. The dashed outline encircles examples of output of a single gamete haplotyping test in which 1 and 0 denote alleles from different parental species on each analyzed gamete (G_i).

of gametes corresponding to parental species 0 and to the parental species 1 (see Supporting Information for details about this method).

Conservation Programs

We emphasize that extinction risk associated with hybridization with genome exclusion is under appreciated because it generates only hybrids displaying the F_1 phenotype, as in hybridization with hybrid sterility that may

be considered of minor conservation concern (Allendorf et al. 2001). We therefore urge policy makers, managers, and stakeholders to include in conservation programs inspection of the genetic content of the gametes of hybrids where distant species hybridization is occurring and only the F_1 phenotype is observed. Because of the species extinction mediated by demographic flow against the disadvantaged species, conservation could benefit from greater awareness and understanding of this issue.

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Supporting Information

A detailed explanation of our general model (Appendix S1), simulations incorporating only female hybrids (Appendix S2) and parental species competition (Appendix S3), and a simple test to recognize hybridization with genome exclusion (Appendix S4) are available online.

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