Reproductive compensation and embryo competition drive the evolution of polyembryony

November 17, 2020

Institutional affiliations

Yaniv Brandvain: Department of Plant and Microbial Biology, University of Minnesota, St. Paul, Minnesota 55108

Alexander Harkness: Department of Ecology, Evolution, and Behavior, University of Minnesota, St. Paul, Minnesota 55108

Tanja Pyhäjärvi: Department of Ecology and Genetics, University of Oulu, FI-90014 Oulu, Finland.

Corresponding author contact details

Corresponding author: Yaniv Brandvain. 1500 Gortner Ave. St. Paul, MN, USA 55108. ybrandva@umn.edu

Keywords

Self-Incompatibility

Polyembryony

Sibling Rivalry

Abstract

Simple polyembryony – where a single gametophyte produces multiple embryos with different sires but the same maternal haplotype – is common in conifers, ferns, horsetails and other vascular plants. Polyembryony could be favored as a mechanism of reproductive compensation, providing a backup for inviable embryos, or as a mechanism of embryo competition and eliminating plants with low fitness, perhaps acting as a mechanism of Self-Incompatibility (SI). However as the evolution of polyembryony from monoembryony has not been modeled these long standing verbal models have not been evaluated. We develop an infinite-site, forward population genetics model to test how these factors can favor the evolution of polyembryony, and how these underlying benefits of polyembryony shape the genetic load under a range of selfing rates, dominance, and selection coefficients. We find that the benefit of reproductive compensation strongly favors the evolution of polyembryony, while the benefits of embryo competition are much weaker. Importantly, when embryo competition favors the evolution of polyembryony it increases embryo competitiveness, but does not act as an SI mechanism, as it does not effectively trade low-fitness selfed offspring for high fitness outcrossed offspring. We find that the impact of polyembryony on the genetic load depends on its function – increasing the embryo load when acting as a mechanism of embryo compensation and decreasing the embryo load when acting as a mechanism of competition.

27 Nature is, above all, profligate. Don't believe them when they
28 tell you how economical and thrifty nature is.

29 – Annie Dillard 1974.

30 Not only do most parents produce more offspring than will survive, but
31 most organisms that provide parental care make more offspring than they
32 will likely be able to nurture to independence. Frequent siblicide in the great
33 egret, *Casmerodius albus*, provides a dramatic example of this – siblings kill
34 one another, presumably over the ability to monopolize small food items
35 (Mock 1984); Why then do egret mothers continue laying eggs that will
36 develop into offspring that will kill one another? Could such overproduction
37 allow parents to screen for offspring quality (Forbes and Mock 1998), or
38 does the “diverse portfolio” of offspring born over the breeding season allow
39 parents to hedge their bets (Forbes 2009)?

40 Simple polyembryony provides an even more extreme, but perhaps less
41 dramatic, example of this problem. With simple polyembryony, a single ma-
42 ternal gametophyte is fertilized by multiple sperm cells to produce multiple
43 embryos with genetically identical maternally derived genomes but distinct
44 paternal genomes (Buchholz 1922; Schnarf 1937, cited in Dogra 1967). Here
45 we present an infinite sites forward-in-time population genomic simulation
46 to test the *competition* (akin to egret mothers screening for offspring qual-
47 ity) and *compensation* (akin to egret mothers hedging their bets) theories
48 for the evolution of polyembryony, and to investigate how polyembryony
49 changes the genetic architecture of embryonic and postembryonic fitness.

50 Simple polyembryony is ubiquitous in gymnosperms (Willson and Bur-
51 ley 1983), and is found in many seedless vascular plants including ferns and
52 horsetails (Buchholz 1922). The number of archegonia per seed typically

varies from two to four in the genus *Pinus*, but can reach up to 200 (as reported in *Widdringtonia juniperoides* Saxton 1934). In gymnosperms, from this base of numerous archegonia, typically only a single embryo survives in mature seed (Chamberlain 1966).

Evolutionary theorists have investigated the evolutionary consequences of polyembryony – specifically how polyembryony (or less mechanistically explicit forms of reproductive compensation) could shape the genetic load (Latta 1995; Sakai 2019; Porcher and Lande 2005; Kärkkäinen et al. 1996) and the exposure of inbreeding depression (Kärkkäinen and Savolainen 1993; Hedrick et al. 1999). However, theories for the evolutionary origin of simple polyembryony are less well developed. Here, we develop theory for the evolution of simple polyembryony. We do not consider cleavage polyembryony, in which a fertilized zygote can split into numerous genetically identical embryos (Agapito-Tenfen et al. 2012), or nucellar polyembryony, in which maternal tissue asexually develops into embryos (Lakshmanan and Ambegaokar 1984), sometimes competing with sexually derived embryos, as they are likely favored by distinct mechanisms (Ganeshaiah et al. 1991).

We consider the two major advantages of simple polyembryony described by Kärkkäinen and Savolainen (1993): reproductive compensation – improved seed set, and embryo competition – the potentially improved post-embryonic fitness of surviving embryos compared to the projected fitness of unsuccessful embryos (Sorensen 1982; Porcher and Lande 2005). Reproductive compensation is an increase in seed set that occurs when embryo mortality is counteracted by an expanded supply of embryos. Polyembryony provides reproductive compensation if a lone embryo is less likely to develop into a successful seed than is a collection of sibling embryos. So, for example, if a proportion p of embryos are inviable, a second embryo increases the

80 probability that a seed contains a surviving embryo from $1 - p$ to $1 - p^2$
81 (Lindgren 1975).

82 Alternatively, if embryonic and post-embryonic fitness are positively cor-
83 related, embryo competition (dubbed Developmental Selection by Buchholz
84 (1922)), could favor the evolution of polyembryony. Such a correlation can
85 arise either through pleiotropy across the life cycle, or if embryonic fitness
86 determined by one set of loci predicted post-embryonic fitness produced by
87 another set of loci. This latter option seems particularly likely if inbred
88 offspring are unfit across the life cycle, and as such, simple polyembryony
89 is often interpreted as an inbreeding avoidance mechanism (e.g. Dogra
90 1967; Sorensen 1982) analogous to the self-incompatibility systems (here-
91 after SI) found in angiosperms. Koski (1971) and others contend that this
92 gives way to evolution of the so-called “Embryo Lethal System” – an ap-
93 parently coordinated self destruction mechanism revealed upon inbreeding
94 (Koski 1971; Sarvas 1962, e.g. page 162 onwards) in pines – as a mech-
95 anism evolved to prevent selfing. Under this model, polyembryony does
96 not prevent self-fertilization *per se*, but dampens self-fertilization’s deleter-
97 ious effects by allowing competition and something of a maternal choice
98 among the selfed and outcrossed progeny before major maternal resource
99 allocation (Willson and Burley 1983; Sorensen 1982). This potential form
100 of postzygotic mate choice could circumvent the constraint imposed by the
101 unenclosed gymnosperm seed, which precludes prezygotic mate choice (e.g.
102 SI systems Dogra 1967; Sorensen 1982; Willson and Burley 1983).

103 Critically, the embryo competition model assumes that possibility of ef-
104 fective competition between embryos in a seed, a topic of much debate.
105 Based on extensive experimental work on *P. sylvestris*, Sarvas (1962) stated
106 that embryo competition and “struggle for life” is quite apparent under mi-

107 crosscopic observation. However, others argue that selfed embryo death pri-
 108 marily occurs after the dominant embryo is determined (Williams 2008), and
 109 embryo survival is determined by chance physical factors Williams (2007);
 110 Mikkola (1969), undercutting the embryo competition model. Empirical
 111 studies evaluating these ideas are quite rare, and the evidence from these
 112 studies is mixed. For example, O'Connell and Ritland (2005) conducted
 113 controlled pollinations with varying levels of self-pollen with *Thuja plicata*,
 114 and found that the effect of embryo competition became apparent with a
 115 probability of selfing (0.75), that exceeds reasonable estimates of the fre-
 116 quency of self-pollination in most conifers. However, subtle effects at lower
 117 selfing rates are plausible.

118 In addition to various selective forces favoring the evolution of polyem-
 119 bryony, polyembryony itself can have striking evolutionary consequences.
 120 Previous models (Porcher and Lande 2005; Klekowski 1982; Sorensen 1982)
 121 examined the effect of reproductive compensation on the number of deleteri-
 122 ous mutations, mean population fitness, the extent of inbreeding depression,
 123 and the realized selfing rate. These models generally show that because re-
 124 moving selfed embryos early in development, polyembryony will prevent the
 125 effective purging of deleterious recessive mutations (Klekowski 1982; Haig
 126 1992), will increases the number of deleterious mutations at equilibrium, in-
 127 crease the extent of inbreeding depression, and decrease the realized selfing
 128 rate while increasing population mean fitness. As such, polyembryony is
 129 often suggested as an explanation for the joint observation of high inbreed-
 130 ing depression (gymnosperms have an estimated 5-10 lethal equivalents per
 131 haploid genome Lynch and Walsh 1998; Williams and Savolainen 1996) and
 132 low realized selfing rates in gymnosperms (Kärkkäinen and Savolainen 1993;
 133 Hedrick et al. 1999). Likewise, polyembryony could explain the absence of

134 a relationship between inbreeding depression and the primary selfing rate
135 in gymnosperms (Husband and Schemske 1996). However other models of
136 polyembryony make drastically different predictions — for example, Latta
137 (1995) modelled the embryo competition component of polyembryony and
138 found that e.g., under a mild mutations model, polyembryony decreased
139 the number of deleterious mutations per individual. In this work we un-
140 cover that much of these differences are attributable to implicit modelling
141 decisions that consider polyembryony as a mechanism of embryo choice or
142 reproductive compensation.

143 The previous work described above provide some insight into the evo-
144 lutionary consequences of polyembryony, but contains numerous modelling
145 assumptions that limit their applicability to major questions in the evolu-
146 tion of polyembryony and its consequences. For example, comparing cases
147 with and without reproductive compensation, Porcher and Lande (2005)
148 showed that reproductive compensation can favor the evolution of selfing
149 and can allow for the maintenance of mixed mating systems, while Sakai
150 (2019) showed that selective embryo abortion could allow for the mainte-
151 nance of high levels of inbreeding depression in selfing species. But if the
152 mating system of an initially monoembryonic population affects whether
153 polyembryony evolves in the first place, this initial condition may affect
154 subsequent mating system evolution after the transition to polyembryony.
155 A second limitation with current theory of the evolutionary consequences
156 of polyembryony is that each model has focused on a single dominance and
157 selection coefficient. As such, while current theory predicts evolution of
158 the number and frequency of deleterious mutations, the magnitude of ge-
159 netic load, it cannot predict evolution of the distributions of dominance or
160 selection coefficients, the architecture of genetic load. This limitation has

161 prevented theory from addressing Koski’s 1971 hypothesis that the “Embryo
162 Lethal System” evolved as an altruistic mechanism by which inbred embryos
163 sacrifice themselves to prevent their mothers from selfing, as opposed to the
164 parsimonious alternative that selfing simply exposes the elevated number of
165 deleterious mutations that can accumulate under polyembryony.

166 Here we present a series of infinite-sites forward population genetic sim-
167 ulations of polyembryony. This model allows us to evaluate the relative
168 importance of competition and compensation to the evolution of polyem-
169 bryony, the evolutionary consequences of alternative models of embryo se-
170 lection, and if polyembryony can favor the evolution of an increased recessive
171 load as a mechanism to prevent inbreeding.

172 **Methods**

173 *Overview*

174 We present a series of models to disentangle the contribution of the potential
175 evolutionary benefits of compensation and competition to the evolution of
176 polyembryony. To better understand how and when these factors favor
177 the evolution of polyembryony, we vary the distribution of dominance and
178 fitness effects and the probability of selfing. Importantly, by changing the
179 underlying probability of selfing, we can address the possibility that when
180 polyembryony evolves as a mechanism of embryo competition, that it may in
181 fact be favored as a mechanism of preventing the generation of self-fertilized
182 offspring. That is, we may hypothesize that polyembryony can evolve to
183 minimize the realized selfing rate. When polyembryony does evolve, we ask
184 how its evolution shapes the genetic load and its architecture.

185 The life cycle begins with $N = 1000$ diploid seeds, each of which has

one or two embryos, depending on whether mothers are mono- or polyembryonic. Following embryo selection, surviving seed parents for the next generation are chosen with replacement with a probability reflecting their post-embryonic fitness. Each time a seed parent is chosen, it generates one of the N seeds in the next generation, thus maintaining a constant population size. Each embryo in the seed is fertilized independently. If directly selfed, which occurs with probability equal to the selfing rate, the seed parent of an embryo is also its pollen parent, otherwise the pollen parent is selected at random and in proportion to adult fitness. Next, gametes are formed by free recombination and each gamete acquires mutations. Finally, fusion between gametes generates the seeds and embryos for the next generation (??).

Parameters and model details

Genome structure and mutation rate: Every generation, each haploid genome expects a Poisson distributed number (mean U) of *de novo* deleterious mutations to arise, each at any one of an infinite number of unlinked sites (i.e. an infinite sites model). We investigate cases with $U = 0.5$ mutations per haploid chromosome per generation.

The timing of mutational effects: We focus on the case in which half of *de novo* deleterious mutations impact embryonic fitness and the other half impact post-embryonic fitness. Our code also allows mutations to influence both pre- and post-embryonic fitness pleiotropically. However our initial early investigation showed that this pleiotropic effect trivially favored the evolution of polyembryony, so we do not investigate this pleiotropic model here.

Figure 1: **Overview of the life cycle model:** The life cycle starts with N seeds, each with one or two embryos, followed by embryo and seed selection. Because of seed inviability, the number of plants will be less than n . Seed parents are chosen with replacement in proportion to their post-embryonic fitness, and directly self with probability p_{self} . Both embryos of polyembryonic mothers are fertilized independently, and pollen parents of non-selfed seed are sampled with from the population with replacement in proportion to each genotypes post-embryonic fitness. Seed parents carrying the dominant polyembryony allele, produce two archegonia per seed, while those without this allele produce only one.

211 **The distribution of fitness and dominance effects of new mutations:**

212 For all parameter values, fitness effects (s) across loci are multiplicative
 213 and independent (Bramlett and Bridgwater 1986, as cited in Williams and
 214 Savolainen 1996), such that the fitness of the i^{th} individual, w_i , equals the
 215 product of one minus the deleterious effect of their genotype at the k^{th}
 216 locus, taken across all loci (i.e. $w_i = \prod_k (1 - s_{ik})$). To investigate the impact

217 of mutational architecture on the evolution of polyembryony, we compare
 218 models with a different value of fitness (s), and dominance (h) effects of new
 219 mutations. For s we present cases with $s = 0.1$, $s = 0.5$, $s = 1$, and $s =$
 220 $\text{Uniform}(20/N, 1)$. Dominance, h , can take any value between 0 and 1, but
 221 we present cases with full recessivity ($h = 0$) and full additivity ($h = 0.5$), as
 222 well as a case where the dominance of each mutation takes a random value
 223 between zero and one from the uniform distribution ($h = \text{Uniform}(0, 1)$).
 224 Thus, mutation effects span the range from quite deleterious to lethal, but
 225 will not reach fixation by random genetic drift. Practically, this means that
 226 we save considerable computational resources, and that we do not consider
 227 weakly deleterious mutations whose fixation is not effectively prevented by
 228 selection. In all simulations, we assumed that the distribution of fitness and
 229 dominance effects did not differ for mutations impacting the embryo and
 230 adult.

231 **Selfing:** With a probability equal to the p_{self} (which we systematically
 232 varied from zero to one in increments of 0.2) the seed parent was also cho-
 233 sen to be the pollen parent. Otherwise, mating was random, with pollen
 234 parents chosen and with replacement in proportion to adult fitness, using
 235 the `sample()` function in R (R Core Team 2020). We note that this random
 236 mating does not preclude selfing. Therefore, even with $p_{\text{self}} = 0$, one of
 237 every N_e embryos (approximately 0.001 when $N = 1000$, depending on seed
 238 survival rates and the variance in post-embryo fitness) is expected to have
 239 identical pollen and seed parents.

240

Evolution

241 **Burn in:** For all parameter combinations, we forward simulated ten repli-
 242 cates process for 2000 generations, ensuring that populations achieved mutation-
 243 selection-drift balance by visually examining the variability in the number
 244 of deleterious mutations over time and among replicates (Figure S1). For
 245 most parameter values, equilibrium was reached within this time frame (Fig-
 246 ure S1). However, for recessive mutations in predominantly outcrossing
 247 populations (with selfing rates of 0, 0.2, or 0.4) this was not enough time
 248 to reach equilibrium. For these slowly equilibrating cases, we increased
 249 the burn-in period until 3000 generations, at which point equilibrium was
 250 largely achieved. Finally, with complete selfing and a non-recessive load
 251 with $s = 0.1$, the number of deleterious mutations seems somewhat unsta-
 252 ble (Figure S1).

253 **Invasion of polyembryony:** For each burn in replicate, we ran many
 254 introductions of a dominant acting polyembryony allele, introduced at a
 255 frequency of $1/2N$, and kept track of the fate of this allele (loss or fixa-
 256 tion) for each introduction. Due to computational considerations, we varied
 257 the number of introductions from 500 to 1000 for each model of polyem-
 258 bryony (below) for each burn-in replicate. That is, when polyembryony
 259 was strongly favored, a given simulation took longer to complete (because
 260 fixation from $1/2N$ takes more time than loss from $1/2N$). By contrast,
 261 when polyembryony is not strongly favored, individual simulations are faster
 262 (because loss occurs more quickly than fixation) and more precision was
 263 needed to distinguish fixation rates from neutrality. The R (R Core Team
 264 2020) code for these forward simulations is available on github [https:](https://github.com/robertmcclellan/evolution)

Figure 2: **Monoembryony and polyembryony:** (A) Under monoembryony, the seed viability is defined by the viability of the single embryo. (B) In simple polyembryony with two embryos, the seed has three possible combinations of viable and inviable embryos. In cases with 1-2 viable embryos, the outcome is defined by the four embryo selection models (see text for details)

265 [//github.com/ybrandvain/polyembryony](https://github.com/ybrandvain/polyembryony).

266 *Models of polyembryony*

267 We aim to dissect the contribution of reproductive compensation and embryo
268 competition to the evolution of polyembryony.

269 The reproductive compensation benefit of polyembryony is that having

two potential embryos in a seed increases the probability that a seed will contain a viable embryo. Thus, the benefit of reproductive compensation occurs if the survival of at least one embryo ensures a viable seed. We can effectively remove this benefit of polyembryony by allowing seed viability to be determined by survival of an arbitrarily chosen embryo among that seed's two embryos.

The embryo competition benefit of polyembryony is that, if there is any relationship between early embryo viability and adult fitness, mothers can increase their expected inclusive fitness by allowing embryos to compete and the more vigorous embryo to become the dominant embryo. We allow for the benefit of embryo competition by selecting the embryo that makes it to the seed among surviving embryos with probabilities in proportion to their embryonic fitness. We can effectively remove this benefit of polyembryony by randomly choosing a single surviving embryo in a seed to become the dominant embryo and continue development. Factorially combining these options results in four models for the evolution of polyembryony (Figure 2).

All advantages: In this model, each embryo in the seed of a polyembryonic mother survives independently with a probability determined by its embryonic fitness. If only one embryo survives, this embryo develops into the seed, and if both embryos survive, the embryo that develops in the seed is chosen in proportion to the relative embryonic fitness of each embryo (i.e. if both embryos survive, the probability an embryo develops into a seed equals its fitness divided by the sum of the fitness of both surviving embryos. Thus, this model includes both potential benefits of polyembryony.

294 **Compensation only:** In this model, each ovule in the seed of a polyem-
 295 bryonic genotype survives independently with a probability determined by
 296 its embryonic fitness. If only one embryo survives, this embryo develops
 297 into the seed, and if both embryos survive, the embryo that develops in the
 298 seed is chosen at random. As such, polyembryony provides the benefit of
 299 increasing the probability that a seed survives, but does not provide the
 300 added benefit of embryo competition. This model resembles the case of re-
 301 productive compensation (Porcher and Lande 2005), as inviable genotypes
 302 can be replaced. This type of selection is much like hard selection – embryo
 303 viability is not dependent on the fitness of the other embryo, however em-
 304 bryos with inviable siblings are more likely to become seed than are embryos
 305 with viable siblings.

306 **Competition only:** In this model, a seed in a polyembryonic genotype
 307 survives with a probability equal to the embryonic fitness of an arbitrarily
 308 chosen embryo (embryo 1 in our simulation). Thus, if this embryo dies but
 309 the other lives, the seed still dies. As such, monoembryonic and polyembry-
 310 onic genotypes will have the same probability of developing a viable seed
 311 (i.e. in both cases seed survival is determined by the fitness of a single
 312 random embryo). However, if both embryos survive, the embryo that devel-
 313 ops in seed is chosen in proportion to the relative embryonic fitness of each
 314 embryo.

315 **No benefit:** In this model, seed survival is determined based on the fitness
 316 of a random embryo in that seed, as in the embryo competition model, while
 317 embryo selection follows the bet-hedging model, in which each embryo in a
 318 seed has an equal probability of surviving. As such, there should be no

319 advantage of polyembryony. This model acts as a control to ensure that our
320 simulation scheme meets neutral expectations and that our control for each
321 potential benefit of polyembryony is properly implemented.

322 Results

323 .

324 *Burn-in simulations*

325 We discuss the results from our burn-in simulations, as they set the scene
326 for the evolution of polyembryony. Throughout the discussion of burn-in
327 results, we focus on mutations impacting embryo fitness, as results for post-
328 embryonic fitness follow similar qualitative and quantitative patterns (Figure
329 S1). Genomes saved at the end of the burn-in are available for download
330 here.

331 **Comparison to published analytical results:** Before discussing spe-
332 cific results, we evaluate whether our simulations behave sensibly by compar-
333 ing model output to known analytical results – namely, the expected number
334 of recessive lethal mutations per diploid genome in a panmictic population.
335 Based on classic results of Li and Nei (1972), Gao et al. (2015) show that
336 the expected number of recessive lethals per (diploid) individual in a finite,
337 panmictic population equals $U\sqrt{2\pi N_e}$, where U is the mutation rate per
338 haploid genome, and N_e is the effective population size. For the case of re-
339 cessive embryo lethals in outcrossers, we find a mean of 18.6 mutations per
340 diploid genome, a value remarkably consistent with the predicted value of
341 18.4 (Compare dashed white line to simulation results in Figure 3A). That

342 is, if $U = 0.25$, as we are only concerned with mutations impacting em-
 343 bryo fitness (half of total mutations), and $2N_e \approx 1745$, the mean number of
 344 surviving embryos across replicates in the final generation. Additional ex-
 345 ploratory simulations (not shown) found a consistent agreement with theory
 346 across a range of mutation rates.

347 **Novel Burn-in Results:** Recessive lethal mutations are effectively purged
 348 with predominant selfing (selfing rate > 0.5), while a large number of dele-
 349 terious mutations accumulate with predominant outcrossing (Figure 3A).
 350 Intriguingly, with an intermediate selfing rate of 0.4, the population appears
 351 to reach an equilibrium, relatively modest number of recessive mutations,
 352 until this rapidly and dramatically increases, presumably reflecting a tran-
 353 sition from effective purging to interference among deleterious mutations
 354 (Lande et al. 1994; Porcher and Lande 2016). Across all parameter com-
 355 binations, the number of deleterious mutations at equilibrium decreased as
 356 mutations became more deleterious and more additive (Figure S1A). Ad-
 357 ditionally, across all dominance and selection coefficients, the number of
 358 deleterious alleles in a population decreased with the selfing rate. However,
 359 the results for obligate selfers were somewhat unstable with weak selection
 360 and non-recessive dominance coefficients.

361 When mutations are recessive ($h = 0$), mean fitness is lowest with in-
 362 termediate selfing rates, and is generally highest with high levels of selfing
 363 or outcrossing (Figure 3B). This pattern is most pronounced when reces-
 364 sive mutations are lethal ($s = 1$), exceptionally deleterious ($s = 0.5$), or
 365 where selection coefficients were drawn from a uniform distribution, and
 366 more subtle with a selection coefficient of $s = 0.1$ (Figure 3B). By contrast,
 367 when mutational effects are additive ($h = 0.5$) or are drawn from a uniform

Figure 3: Results from burn-in: (A) The mean number of recessive lethal alleles per haploid genome over time. Each line is one of ten replicates for each selfing rate, designated by color. The dashed white line shows the theoretical expectation for a randomly mating population in our simulation, while the larger colored line shows the mean across replicates. Results with different dominance and selection coefficients are presented in Figure S1. Figures (B-D) show features of the population 'burn-in' populations after the load equilibrates. Points are slightly jittered to show the data - with one value for each replicate simulation for a given combination of selfing rates on the x, and fitness effects of new mutations in color, lines connect means. In C, the one to one line is shown by the dashed black line. All mutations are fully recessive. Results with different dominance coefficients are presented in Figure S2.

368 distribution, mean fitness increases with the selfing rate, with significantly
369 positive slopes ranging between 0.045, and 0.082 (Figure S2A, Table S1),
370 presumably because selfing increases the variance in fitness, allowing for
371 more effective selection. In these non-recessive cases, mean embryo fitness is
372 roughly similar, regardless of the fitness effects of individual mutations (Fig-

ure S2, modelling mean fitness = $f(\text{selfing}, s)$, the p-value for the effect of s is 0.059 and 0.25, for cases with a uniform and additive load, respectively, Table S2). Reassuringly, this grand mean fitness under obligate outcrossing for non-recessive alleles of 0.78 is in line with Haldane's 1937 classic result that mean fitness equals e^{-U} (where U is the mutation rate per haploid genome, which equals 0.5 divided by two, as half of mutations will impact embryo fitness). Somewhat surprisingly, mean post-embryonic fitness does depend on the selection coefficient (Fig. S2D), suggesting that selection at one life stage impacts outcomes at another as suggested by (Sakai 2019).

With intermediate selfing rates and recessive gene action, we observe a much higher primary than realized selfing rate, suggesting that inbreeding depression underlies much of the embryo death in these cases (Fig. 3C). By contrast, we observe a nearly perfect relationship between primary and realized selfing rates under non-recessivity (Fig. 3B). We observe a strong positive correlation between embryo and post-embryo fitness for recessive gene action and intermediate selfing rates, but no relationship otherwise (Fig. 3D, and Fig. S2D). Together these results support the intuition that if competition acts to remove selfed embryos, this benefit of polyembryony will be most relevant when mutations are recessive.

Invasion of polyembryony

We compare the fixation probability of a new mutant that confers polyembryony, across all models described above. We find that, when the polyembryony allele fixes, it tends to fix more quickly when polyembryony provides reproductive compensation than when it does not (Fig. 4A, Fig. S4, Table S3). Similarly, polyembryony is most likely to fix when it provides repro-

ductive compensation – in some cases, single mutations have up to a fifteen percent chance of reaching fixation, a 300-fold increase in the probability, relative to neutral expectations (Fig. 4B & 4C). The benefits of competition alone also favored the evolution of polyembryony, but had a more modest effect – in some cases, single mutations have up to a one percent chance of reaching fixation, a 20-fold increase in the probability, relative to neutral expectations. Reassuringly, fixation proportions from the no benefits model matched neutral expectations, with approximately $1/2N = 0.0005$ introductions resulting in fixation (See Table S3, and compare the solid lines to the dashed line in Figure 4B).

Other biological parameters such as the selfing rate, and the dominance and selective coefficients of deleterious mutations also impact on the evolution of polyembryony, often depending on their interaction. Below we discuss the effects of selfing rate and additive vs. recessive modes of gene action, noting that results from the uniform mode of gene action are qualitatively similar to the additive model (Fig. S3, Table S3).

The benefit of reproductive compensation strongly favored the evolution of polyembryony for all biological parameters investigated (Fig. 4). Figure 4C displays the fixation proportions for the compensation models (row two in Fig. 4B) on a linear scale to reveal the effect of selfing rate and selective effects of new mutations.

Under recessivity, the probability of fixation is maximized (approximately 15%) at intermediate selfing rates, suggesting that polyembryony can evolve to make up for offspring lost to early acting-inbreeding depression. Again assuming recessivity, obligate outcrossing more strongly favors the evolution of polyembryony than does obligate selfing (compare an ap-

Figure 4: The fixation of an allele conferring polyembryony: A) Example trajectories of the fixation of the polyembryony allele with all benefits (black), the benefit of compensation (pink), the benefit of competition (orange), and no benefit (blue). Note that generation on x increases on the log10 scale, but specific values are noted with their linear value. **B)** The proportion of introductions resulting in fixation of the polyembryony allele as a function of the selfing rate (x), the fitness effect of new mutations (color), the mode of gene action (columns), and the benefit of polyembryony (rows). The dashed pink line displays the expectation under neutrality. Note that fixation proportion on y increases on the $\log_{10}$ scale, but specific values are noted with their linear value. **C)** The proportion of introductions resulting in fixation of the polyembryony allele as a function of the selfing rate (x), the fitness effect of new mutations (color), the mode of gene action (columns), with all the benefits of polyembryony. The values are identical to those in the first row of **B**, but are presented on a linear scale to highlight the effect of selfing rate on fixation probability.

proximately 10% fixation probability under obligate outcrossing to a 2.5% fixation probability under obligate selfing, Fig. 4C), presumably reflecting the higher within-seed variance in fitness under obligate outcrossing leading to higher impact of polyembryony. The fitness effect of recessive deleterious mutations have only a modest effect on fixation proportion, varying slightly across selfing rate.

However, the compensation model also strongly favors the evolution of polyembryony with an additive load, suggesting that overcoming inbreeding depression is not the only driver of the evolution of polyembryony. (Second row, second column, Figure 4B). In cases with additive gene action, the fixation probability of a polyembryony allele decreases with the selfing rate, again reflecting the lack of within-seed variance in fitness. Additionally, under additivity (or if mutations take their dominance coefficients from a uniform distribution, Table S3, Figure S3) a load composed of highly deleterious mutations is less likely to foster the evolution of polyembryony than a load composed of a larger number of mild mutations (compare $s = 1$ (blue) to $s = 0.5$ (orange) or $s = \text{uniform}$ (teal) to $s = 0.1$ (grey), Fig. 4). This surprising result might reflect the fact that while mean fitness does not depend on fitness effects of new mutations, the survival of maternal sib-embryos becomes more dependent on one another as mutational effects get larger (Figure S5). As such, with large effect mutations, a backup embryo is less useful as if one dies the other is likely to die as well.

The benefit of embryo competition also favors the evolution of polyembryony. However, fixation probabilities are approximately five- to ten-fold lower for this model than for the reproductive compensation model. With a recessive load and intermediate selfing rates (0.20 or 0.40), the benefit

of embryo choice results in the fixation of the polyembryony allele in approximately one percent of introductions, a twenty-fold increase relative to the neutral expectation of 0.05%. Somewhat surprisingly, the embryo competition model favors polyembryony for a non-recessive load (Third row, second column of Fig. 4B), even though embryo fitness was uncorrelated with post-embryo fitness in these models (Fig. S2). This likely reflects the benefit of producing grand-children with higher embryonic fitness who will out-compete their siblings (analogous to models of “runaway sexual selection” Kirkpatrick 1982). Under both additivity and a uniform distribution of mutational effects, the probability of fixation of an allele conferring competitive polyembryony is greatest in predominantly outcrossing populations (selfing rates of 0.40 or less), decreasing as the selfing rate increases. Here, the probability of fixation is greatest when the load is composed of alleles of large effect, a result that runs counter to that found in the compensation model with an additive load.

All benefits results in fixation probabilities qualitatively similar to the reproductive compensation model (Fig. S3, Table S3) – reflecting the importance of the benefits of reproductive compensation, to the evolution of polyembryony.

No benefits results in fixation probabilities consistent with neutral expectations (Fig. S3, Table S3).

Evolutionary consequences of polyembryony

We compare how different models of the evolution of polyembryony shape key features of a population, including the proportion of surviving seeds,

the realized selfing rate and the architecture of genetic load. Although a strict version of the competition only model is unlikely to occur in nature, its inclusion allows us to distinguish the individual effects of competition and compensation when both would be operating in nature (i.e. the all benefits model). Because results were qualitatively similar across all selection coefficients (save the decrease in fitness with recessive mutations, $s = 0.1$ and high selfing rates, which did not always converge Fig. S1), and because results from the additive model and the uniformly distributed dominance coefficient model did not differ qualitatively, we focus on results from the cases in which the selection coefficients of new mutations are selected at random from a uniform distribution, exploring cases in which mutations are recessive or additive (Figure 5).

Curiously, the benefit of embryo competition alone did not impact the realized selfing rate (Fig. 5), even with recessive mutations and intermediate selfing rates. This result is a consequence of features of both our model and biological reality. Specifically, there are limited opportunities for competition between selfed and outcrossed embryos in a seed (Williams 2007), as this only occurs with a probability equal to two times the variance in the selfing rate (the probability that exactly one of two embryos is from a self-fertilization event) times the probability that both are destined to survive. By contrast, with a recessive load a benefit of compensation decreases the realized selfing rate, and increases the number of mutations impacting embryo fitness in partially selfing populations (Fig. 5).

The benefit of compensation (in both the compensation and all benefits model) resulted in a strong increase in seed survival. Under a recessive load, this effect was maximized with intermediate selfing rates, while it decreased steadily with selfing rate under a (partially) additive load. By selecting for

Figure 5: **The evolutionary impact of polyembryony.** How the evolution of polyembryony impacts the per individual number of mutations impacting embryo fitness, expected seed survival, mean embryo fitness, the realized selfing rate, and mean post-embryo fitness (rows), when mutations are additive or recessive (columns), across selfing rates (x-axis), for each model of polyembryony (color).

501 higher fitness embryos, the benefit of competition alone subtly increased
 502 seed survival for all models of dominance investigated so long as the selfing
 503 rate was not too large (Fig. 5). Consequently, the expected embryo fitness of
 504 the surviving seeds subtly increases with the benefit of embryo competition,
 505 but decreases with compensation. These benefits appear to act additively,
 506 such that the expected embryo fitness decreases in the all benefits model
 507 but does so less severely than in the compensation model.

508 Regardless of the mode of gene action, the competition model does not
 509 increase the expected post-embryo fitness of surviving seeds (Fig. 5). While

510 post-embryo fitness modestly increases with the evolution of polyembryony
 511 under a model of competition with intermediate selfing rates and a recessive
 512 load, this increase pales in comparison to the benefits of higher seed viability
 513 which accompanies reproductive compensation. Together, these lines of evi-
 514 dence suggest that polyembryony does not evolve as a mechanism to prevent
 515 self-fertilization, and is not analogous to the system of self-incompatibility
 516 observed in angiosperms.

517 Additional evidence against the hypothesis that the embryonic lethal
 518 system evolves as an SI-like mechanism comes from the allele frequency
 519 spectrum of deleterious mutations (Fig. 5). If the prevention of the forma-
 520 tion of inviable selfed seeds by eliminating selfed embryos favored a system
 521 to destroy selfed embryos, we would expect an increase in very rare reces-
 522 sive lethal alleles so as to ensure the death of selfed embryos. Contrary to
 523 this expectation, we see no such shift in the frequency spectrum. Figure 6
 524 shows that the allele frequency spectrum is comparable in the no benefit and
 525 competition model, arguing against the idea that competition favored self-
 526 sacrifice in the form of an excess of rare recessive lethals. By contrast, there
 527 is a slight increase in the count of deleterious recessive mutations across all
 528 frequency classes in the compensation and all benefits models, reflecting the
 529 relaxation of embryo selection in these cases.

530 Discussion

531 We present four models to test the plausibility of the compensation and
 532 competition theories for the evolution of polyembryony. We find that the
 533 evolutionary benefit of compensation – that is, the opportunity for a backup
 534 embryo to replace an inviable one – strongly favors the evolution of polyem-

Figure 6: **The allele frequency spectrum** for embryo acting allele following the evolution of polyembryony under the recessive lethal model for a selfing rate of 0.00, 0.40, and 0.80 (left to right). Lines display averages of ten simulation replicates, and colors note the model of polyembryony. Note that the x axis with a selfing rate of 0.80 (right panel) is truncated relative to the other selfing rates, reflecting the effective purging of early acting recessive mutations with high selfing rates.

535 bryony. Relative to neutral expectations, the benefit of compensation results
 536 in between a twenty-fold increase in fixation probability above the neutral
 537 expectation with high selfing rates, and a two hundred-fold increase with in-
 538 termediate to low selfing rates and a recessive load, all across a broad range
 539 of selection and dominance coefficients. By contrast, the benefit of embryo
 540 competition more weakly favored the evolution of polyembryony, resulting
 541 in between a zero-fold increase with high selfing rates, and a twenty-fold
 542 increase, with intermediate to low selfing rates and a recessive load, relative
 543 to neutral expectations.

544 Our work highlights the previously underappreciated result that the con-
 545 sequences of polyembryony depend on its function. When polyembryony
 546 functions as a compensation mechanism, mean embryo fitness is reduced,
 547 but the probability of seed survival increases, consistent with previous work
 548 (Latta 1995; Porcher and Lande 2005). By contrast, competition between
 549 embryos alone increases embryo fitness, but has a negligible effect on seed

550 survival, in line with models of competition alone (Latta 1995). With a re-
 551 cessive load, the benefit of compensation acts to decrease the effective selfing
 552 rate, while competition did not. With both the benefits of competition and
 553 compensation the evolutionary consequences of polyembryony is somewhere
 554 in between but is often closer to those expected from compensation.

555 **The limited role of embryo competition in the evolution of polyem-**
 556 **bryony:** It has long been assumed that a major benefit of polyembryony
 557 is that it provides an opportunity for embryos to compete (Sarvas 1962;
 558 Koski 1971), and to create high fitness offspring offspring. Not only was
 559 the benefit of competition a comparatively weak force in the evolution of
 560 polyembryony, but it did not reliably increase seed fitness. In fact, under
 561 most selfing rates and dominance coefficients, competition more strongly fa-
 562 vored the evolution of polyembryony with an additive load (in which there
 563 is no relationship between embryo and post embryo fitness) than a recessive
 564 load (in which there was such a relationship). This is surprising because
 565 there is a limited scope for selection on polyembryony when it cannot affect
 566 post-embryo fitness. In this case, selection on polyembryony only occurs
 567 within polyembryonic seeds themselves, and, since the embryos' maternal
 568 genomes are identical, only among the paternal genomes. Like runaway sex-
 569 ual selection (Fisher 1915), the automatic transmission advantage of selfing
 570 (Fisher 1941), or meiotic drive (Rhoades 1942), embryo competition is an
 571 example of a selective advantage that does not make a population necessarily
 572 more adapted to its environment.

573 **Why doesn't embryo competition effectively remove selfed off-**
 574 **spring and increase post-embryonic fitness (and could it ever)?**

575 We considered only two embryos and only allowed for competition if both
 576 embryos were destined to live in our model of embryo competition. As such,
 577 embryo competition can only weed out selfed offspring when one seed is
 578 selfed and the other is not and both seeds are destined to live. Because
 579 predominant selfing (selfing rate > 0.5) purges the recessive load, and pre-
 580 dominant outcrossing (selfing rate < 0.5) generates large inbreeding depres-
 581 sion, in most cases in which selfed and outcrossed embryos could compete,
 582 their fitness is either nearly equal or the selfed embryo is destined to die.
 583 As such, embryo competition does not offer a more refined view into post-
 584 embryo fitness than is automatically accounted for by “hard selection” on
 585 seed viability imposed in our model. Our observation that embryo compe-
 586 tition leads to more competitive embryos rather than higher fitness plants
 587 is consistent with the claim of McCoy and Haig (2020) that Goodhart’s law
 588 – ‘When a measure becomes a target it ceases to be a good measure’ – can
 589 undermine effective embryo selection. Despite our focus on the evolution
 590 of polyembryony, these results apply broadly and suggest that verbal mod-
 591 els predicting that selective embryo abortion could limit the mating costs
 592 of selfing in plants with mixed mating systems (e.g. Huang et al. 2020),
 593 require more rigorous scrutiny.

594 Nonetheless, it is possible that pure “soft selection” (Wallace 1968, 1975)
 595 on embryos could reliably increase post-embryo fitness. However we had
 596 trouble implementing this model computationally (e.g. we could not define
 597 the allele frequency spectrum of alleles determining success in soft selection
 598 before embryo competition evolved), nor could we map this onto a plausible
 599 biological mechanism. Nonetheless, this challenge could reflect a shortcom-
 600 ing in our imagination, rather than a biological impossibility. Additionally,
 601 we note that even if the benefits of compensation initially favored the evo-

602 lution of polyembryony, it is possible that the evolution of polyembryony
603 was followed by novel recessive mutations experiencing soft selection and
604 that therefore the benefits of competition could maintain but not drive the
605 evolution of polyembryony.

606 **The embryo lethal system:** Since Buchholz (1922), it has been argued
607 that the embryo-lethal system, an apparently coordinated process of embryo
608 death, could achieve a similar function to angiosperm self-incompatibility in
609 the self-compatible gymnosperms. That is, intentional, coordinated death in
610 the embryo stage would give way to highly outbred surviving adults (Sarvas
611 1962; Koski 1971). This would be an altruistic act in which an embryo
612 sacrifices its predictably low fitness for a half sibling. We did not observe
613 the evolution of an embryo lethal system in response to the evolution of
614 polyembryony, as would be expected if polyembryony favored altruistic self-
615 destruction of more inbred embryos (e.g. we did not see a change in the allele
616 frequency spectrum towards an excess of low-frequency recessive mutations).
617 However, by relaxing selection on embryo viability, embryo compensation
618 could indirectly result in an increase in the number of highly deleterious
619 recessive mutations impacting embryo fitness. As such, while we cannot
620 exclude possibilities which we did not model (e.g. pure soft selection, above,
621 or pleiotropy across life stages (below)), it appears that the embryo lethal
622 system could reflect an elevated load tolerance rather than an exquisite
623 adaptation (Gould and Lewontin 1979), as Williams (2007) argued forcefully
624 based on developmental and genetic evidence.

625 **Which has driven the evolution of polyembryony – Compensation**
626 **or competition?** We find that the benefit of embryo compensation favors

the evolution of polyembryony more strongly than does embryo competition. However, we caution that whether compensation or competition have actually favored the evolution of polyembryony depends on their biological plausibility and whether they reflect effective solutions to the problems they address. That is, we must consider biological processes outside of our model as we interpret our model results. For example, embryo competition could perhaps be most effectively achieved by placing more embryos in a seed, while compensation could be more effectively achieved by producing more seeds per plant.

Our models provide competing testable predictions to distinguish between predictions of the compensation and competition model at within seed level, for simple polyembryony, assuming no pleiotropic effects. For example, we show that the evolution of polyembryony and its consequences depend on the selfing-rate and dominance coefficient. Specifically, with a recessive genetic load, embryo competition most strongly favors the evolution of polyembryony at intermediate selfing rates (Fig. 4B,C). The estimates of selfing rates for modern conifers can reach 0.30 - 0.60 (Sarvas 1962; Sorensen 1982), a range that favors polyembryony. We note, of course, that estimates of the primary selfing rate from extant conifers rate may differ substantially from the primary selfing rates of the population in which polyembryony arose.

Additionally, the two models make subtly different predictions about the difference between the realized and primary selfing rate. Relative to a monoembryonic ancestral population, polyembryony favored by embryo competition alone does not result in decrease in the difference between realized and primary selfing rates. By contrast, with a recessive load and intermediate selfing rates, polyembryony favored by compensation strongly amplified the

654 difference between the realized and primary selfing rates. In nature, differ-
 655 ences between primary and realized selfing rates are often observed in species
 656 with simple polyembryony (Lindgren 1975; Sorensen 1982; Kärkkäinen and
 657 Savolainen 1993; Lande et al. 1994), further emphasizing the probable role
 658 of compensation in the evolution and maintenance of polyembryony.

659 **Alternative Models for the Evolution of Polyembryony:** We as-
 660 sumed no pleiotropy across life stages – that is, mutations either impacted
 661 embryo or post embryo fitness. However, this is clearly untrue. For ex-
 662 ample, severe loss of function mutations in key genes would likely decrease
 663 both embryo and seed fitness. Preliminary investigations of this scenario
 664 (not shown) showed that this model so strongly favored the evolution of
 665 polyembryony that it was theoretically trivial, and we therefore did not
 666 pursue this possibility in greater detail. Theoretical triviality does not im-
 667 ply biological irrelevance, and as such we cannot exclude this biologically
 668 plausible model.

669 On the other hand, an allele could antagonistically increase embryo fit-
 670 ness while decreasing post embryonic fitness. When such a mutation occurs
 671 with embryo competition, it could generate an ontogenic conflict. Empirical
 672 studies, e.g. mapping and measuring of inbreeding depression at different life
 673 stages (Koelewijn 1998), comparing gene expression across embryo develop-
 674 ment and later life stages (Raheison et al. 2012), and signatures of negative
 675 and positive selection in such genes would be valuable to further evaluate
 676 the potential importance of pleiotropy in the evolution of polyembryony.

677 **Competition, compensation and conflict in a pine nutshell:** Gym-
 678 nosperm seed with a maternal haploid megagametophyte, multiple geneti-

679 cally distinct embryos, genetically identical (cleavage) embryos, and strong
680 inbreeding depression is a stage of evolutionary drama that deserves more
681 attention, and we hope that the provided model will be used to broaden the
682 investigations on the evolutionary dynamics outside the angiosperm sphere.
683 For example, in contrast to the opportunity for altruism to favor the embryo-
684 lethal system, polyembryony also provides avenues for parental and embry-
685 onic conflict.

686 In simple polyembryony, embryos are potentially derived from different
687 sires. A paternal genome carrying a mutation that sabotaged rival embryos
688 carrying different paternal genomes could possess a net advantage even if
689 doing so would reduce the probability that a viable seed is formed at all.
690 Sabotage and anti-sabotage alleles would only be beneficial when expressed
691 in a particular parental genome, so genomic imprinting that prevented ex-
692 pression in the wrong parental genome would also be advantageous.

693 Conifers and other gymnosperms provide unique opportunities to test
694 key questions of plant mating system evolution and evolutionary conflict
695 from a novel angle, especially now that their genomic resources are no longer
696 seriously hindered by their large genome sizes. From the practical/empirical
697 perspective, large seed size and gametophytic tissue allow easy identification
698 of maternal haplotypes and alleles. Thus expression patterns and genetic di-
699 versity for example in potentially imprinted genes should be easy to quantify
700 and identify in many conifer species.

Acknowledgments

TP received funding from Academy of Finland (grant nrs. 287431 and 319313) and acknowledges CSC, IT Center For Science for computational resources. AH was funded through the University of Minnesota Doctoral Dissertation Fellowship. The authors thank Outi Savolainen for insightful comments on the manuscript.

Literature Cited

- Agapito-Tenfen, S. Z., N. Steiner, M. P. Guerra, and R. O. Nodari. 2012. Patterns of polyembryony and frequency of surviving multiple embryos of the brazilian pine *Araucaria angustifolia*. *Australian Journal of Botany* 59:749–755.
- Bramlett, D. L., and F. E. Bridgwater. 1986. Segregation of recessive embryonic lethal alleles in a F1 population of virginia pine. *In* IUFRO Conference on Breeding Theory, Williamburg Va.
- Buchholz, J. T. 1922. Developmental selection in vascular plants. *Botanical Gazette* 73:249–286.
- Chamberlain, C. J. 1966. *Gymnosperms: Structure and Evolution*; With 397 Fig. Dover Publ.
- Dillard, A. 1974. *Pilgrim at Tinker Creek*. Literature (Harper & Row). HarperPerennial.
- Dogra, P. D. 1967. Seed sterility and disturbances in embryogeny in conifers with particular reference to seed testing and tree breeding in pinaceae. *Studia forestalia Suecica* pages 1–97.
- Fisher, R. A. 1915. The evolution of sexual preference. *Eugenics Review*

725 7:184–192.

726 ———. 1941. Average excess and average effect of a gene substitution.
727 *Annals of Eugenics* 11:53–63.

728 Forbes, L. S., and D. W. Mock. 1998. Parental optimism and progeny choice:
729 When is screening for offspring quality affordable. *Journal of Theoretical*
730 *Biology* 192:3–14.

731 Forbes, S. 2009. Portfolio theory and how parent birds manage investment
732 risk. *Oikos* 118:1561–1569.

733 Ganeshaiah, K. N., R. Uma Shaanker, and N. V. Joshi. 1991. Evolution
734 of polyembryony: Consequences to the fitness of mother and offspring.
735 *Journal of Genetics* 70:103–127.

736 Gao, Z., D. Waggoner, M. Stephens, C. Ober, and M. Przeworski. 2015. An
737 estimate of the average number of recessive lethal mutations carried by
738 humans. *Genetics* 199:1243–1254.

739 Gould, S. J., and R. C. Lewontin. 1979. The spandrels of san marco and
740 the panglossian paradigm: a critique of the adaptationist programme.
741 *Proceedings of the Royal Society B: Biological Sciences* 205:581–598.

742 Haig, D. 1992. Brood reduction in gymnosperms. Pages 63–84 *in* M. Elgar
743 and B. Crespi, eds. *Cannibalism: ecology and evolution among diverse*
744 *taxa*. Oxford University Press, Oxford.

745 Haldane, J. B. S. 1937. The effect of variation on fitness. *American Natu-*
746 *ralist* 71:337–349.

747 Hedrick, P. W., O. Savolainen, and K. Karkkainen. 1999. Factors influencing
748 the extent of inbreeding depression: an example from scots pine. *Heredity*
749 82 Pt 4:441–450.

750 Huang, Q., W. Wang, S. C. H. Barrett, and M. Ren. 2020. Plasticity in se-

751 lective embryo abortion may limit the mating costs of geitonogamy in self-
752 compatible plants: a hypothesis. *American Journal of Botany* 107:390–
753 393.

754 Husband, B. C., and D. W. Schemske. 1996. Evolution of the magnitude
755 and timing of inbreeding depression in plants. *Evolution* 50:54–70.

756 Kärkkäinen, K., V. Koski, and O. Savolainen. 1996. Geographical variation
757 in the inbreeding depression of scots pine. *Evolution* 50:111–119.

758 Kärkkäinen, K., and O. Savolainen. 1993. The degree of early inbreeding
759 depression determines the selfing rate at the seed stage: model and results
760 from *Pinus sylvestris* (scots pine). *Heredity* 71:160–166.

761 Kirkpatrick, M. 1982. Sexual selection and the evolution of female choice.
762 *Evolution* 36:1–12.

763 Klekowski, E. J. 1982. Genetic load and soft selection in ferns. *Heredity*
764 49:191–197.

765 Koelewijn, H. P. 1998. Effects of different levels of inbreeding on progeny
766 fitness in *Plantago coronopus*. *Evolution* 52:692–702.

767 Koski, V. 1971. Embryonic lethals of *Picea abies* and *Pinus sylvestris*.
768 Metsäntutkimuslaitos.

769 Lakshmanan, K. K., and K. B. Ambegaokar. 1984. Polyembryony. Pages
770 445–474 in B. M. Johri, ed. *Embryology of Angiosperms*. Springer Berlin
771 Heidelberg, Berlin, Heidelberg.

772 Lande, R., D. W. Schemske, and S. T. Schultz. 1994. High inbreeding de-
773 pression, selective interference among loci, and the threshold selfing rate
774 for purging recessive lethal mutations. *Evolution* 48:965–978.

775 Latta, R. G. 1995. The effects of embryo competition with mixed mating
776 on the genetic load in plants. *Heredity* 75:637–643.

777 Li, W. H., and M. Nei. 1972. Total number of individuals affected by a single
778 deleterious mutation in a finite population. *American Journal of Human*
779 *Genetics* 24:667–679.

780 Lindgren, D. 1975. The relationship between self-fertilization, empty seeds
781 and seeds originating from selfing as a consequence of polyembryony. *Stu-*
782 *dia Forestalia Suecica* 126:1–24.

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

785 McCoy, D. E., and D. Haig. 2020. Embryo selection and mate choice: Can
786 ‘honest signals’ be trusted? *Trends in Ecology and Evolution* 35:308–318.

787 Mikkola, L. 1969. Observations on interspecific sterility in picea. *Annales*
788 *Botanici Fennici* 6:285–339.

789 Mock, D. W. 1984. Siblicidal aggression and resource monopolization in
790 birds. *Science* 225:731–733.

791 O’Connell, L. M., and K. Ritland. 2005. Post-pollination mechanisms pro-
792 moting outcrossing in a self-fertile conifer, *Thuja plicata* (cupressaceae).
793 *Canadian Journal of Botany* 83:335–342.

794 Porcher, E., and R. Lande. 2005. Reproductive compensation in the evolu-
795 tion of plant mating systems. *New Phytologist* 166:673–684.

796 ———. 2016. Inbreeding depression under mixed outcrossing, self-
797 fertilization and sib-mating. *BMC Evolutionary Biology* 16:105.

798 R Core Team. 2020. *R: A Language and Environment for Statistical Com-*
799 *puting*. R Foundation for Statistical Computing, Vienna, Austria.

800 Raherison, E., P. Rigault, S. Caron, P.-L. Poulin, B. Boyle, J.-P. Verta,
801 I. Giguère, C. Bomal, J. Bohlmann, and J. MacKay. 2012. Transcriptome
802 profiling in conifers and the PiceaGenExpress database show patterns

803 of diversification within gene families and interspecific conservation in
804 vascular gene expression. BMC Genomics 13:434.

805 Rhoades, M. M. 1942. Preferential segregation in maize. Genetics 27:395–
806 407.

807 Sakai, S. 2019. Maintenance of high inbreeding depression in selfing popu-
808 lations: Effects of coupling of early- and Late-Acting mutations. bioRxiv
809 page 748699.

810 Sarvas, R. 1962. Investigations on the flowering and seed crop of *Pinus*
811 *silvestris*. Metsätieteellisen tutkimuslaitoksen julkaisuja .

812 Saxton, W. T. 1934. Notes on conifers: IX. the ovule and embryogeny of
813 widdringtonia. Annals of Botany 48:429–431.

814 Schnarf, K. 1937. Anatomie der Gymnospermen-Samen, vol. 10. Gebr.
815 Borntraeger.

816 Sorensen, F. C. 1982. The roles of polyembryony and embryo viability in
817 the genetic system of conifers.

818 Wallace, B. 1968. Polymorphism, population size and genetic load,
819 in “population biology and Evolution” (RC lewontin, ed.) .

820 ———. 1975. Hard and soft selection revisited. Evolution 29:465–473.

821 Williams, C. G. 2007. Re-thinking the embryo lethal system within the
822 pinaceae. Canadian Journal of Botany 85:667–677.

823 ———. 2008. Selfed embryo death in *Pinus taeda*: a phenotypic profile.
824 New Phytologist 178:210–222.

825 Williams, C. G., and O. Savolainen. 1996. Inbreeding depression in conifers:
826 Implications for breeding strategy. Forest Science 42:102–117.

827 Willson, M. F., and N. Burley. 1983. Mate Choice in Plants: Tactics, Mech-
828 anisms, and Consequences. Princeton University Press.

Table S1: Slope of the relationship between selfing rate and mean embryo fitness after burn-ins for non-recessive variants. All t values are associated with 59 degrees of freedom.

h	s	estimate	lower 95% CI	upper 95% CI	t	p-value
0.5	0.1	0.054	0.031	0.076	4.719	0.000015
0.5	0.5	0.085	0.076	0.094	19.114	<10-11
0.5	1	0.051	0.039	0.062	8.600	<10-11
0.5	uniform	0.082	0.072	0.091	16.413	<10-11
uniform	0.1	0.045	0.015	0.074	2.980	0.004203
uniform	0.5	0.078	0.068	0.088	15.313	<10-11
uniform	1	0.065	0.055	0.076	12.021	<10-11
uniform	uniform	0.083	0.074	0.091	19.445	<10-11

Table S2: Effect of selection coefficient on mean fitness following burn in for non-recessive variants.

h	estimate	$F_{3,225}$	p-value
0.5	0.054	1.39	0.059
uniform	0.083	2.52	0.247

Table S3: The proportion of introductions of the polyembryony allele resulting in fixation.

p_{self}	s	h	All benefits	Compensation	Competition	No benefit
0	uniform	uniform	0.1084 (n=5000)	0.1025 (n=10000)	0.0041 (n=10000)	0.0012 (n=10000)
0	uniform	recessive	0.089 (n=5000)	0.08937 (n=9500)	0.00027 (n=7500)	0.00027 (n=7500)
0	uniform	additive	0.1058 (n=5000)	0.1038 (n=10000)	0.0056 (n=10000)	7e-04 (n=10000)
0	0.1	uniform	0.129 (n=5000)	0.131 (n=10000)	0.0017 (n=10000)	2e-04 (n=10000)
0	0.1	recessive	0.09517 (n=6000)	0.10129 (n=7000)	0.00013 (n=7500)	0 (n=6500)
0	0.1	additive	0.1306 (n=5000)	0.128 (n=10000)	0.001 (n=10000)	1e-04 (n=10000)
0	0.5	uniform	0.1128 (n=5000)	0.0997 (n=10000)	0.0051 (n=10000)	4e-04 (n=10000)
0	0.5	recessive	0.095 (n=5000)	0.0885 (n=10000)	4e-04 (n=7500)	0.00031 (n=6500)
0	0.5	additive	0.1028 (n=5000)	0.1037 (n=10000)	0.0049 (n=10000)	4e-04 (n=10000)
0	1	uniform	0.0844 (n=5000)	0.0875 (n=10000)	0.0049 (n=10000)	0.001 (n=10000)
0	1	recessive	0.0792 (n=5000)	0.0817 (n=10000)	0.00042 (n=9500)	0.00012 (n=8500)
0	1	additive	0.0874 (n=5000)	0.077 (n=10000)	0.0092 (n=10000)	1e-04 (n=10000)
0.2	uniform	uniform	0.0946 (n=5000)	0.0885 (n=10000)	0.0033 (n=10000)	2e-04 (n=10000)
0.2	uniform	recessive	0.1398 (n=5000)	0.1539 (n=10000)	0.00462 (n=6500)	0 (n=9000)
0.2	uniform	additive	0.0952 (n=5000)	0.0901 (n=10000)	0.0054 (n=10000)	2e-04 (n=10000)

p_{self}	s	h	All benefits	Compensation	Competition	No benefit
0.2	0.1	uniform	0.1178 (n=5000)	0.1211 (n=10000)	0.0015 (n=10000)	3e-04 (n=10000)
0.2	0.1	recessive	0.14436 (n=5500)	0.138 (n=6000)	0.01154 (n=6500)	0 (n=7000)
0.2	0.1	additive	0.1164 (n=5000)	0.1258 (n=10000)	0.0028 (n=10000)	2e-04 (n=10000)
0.2	0.5	uniform	0.0912 (n=5000)	0.089 (n=10000)	0.0048 (n=10000)	5e-04 (n=10000)
0.2	0.5	recessive	0.154 (n=5000)	0.1427 (n=10000)	0.00322 (n=9000)	0 (n=8500)
0.2	0.5	additive	0.0884 (n=5000)	0.0826 (n=10000)	0.0041 (n=10000)	5e-04 (n=10000)
0.2	1	uniform	0.0808 (n=5000)	0.0672 (n=10000)	0.0055 (n=10000)	4e-04 (n=10000)
0.2	1	recessive	0.1428 (n=5000)	0.1378 (n=10000)	0.00056 (n=9000)	0.00084 (n=9500)
0.2	1	additive	0.0746 (n=5000)	0.0669 (n=10000)	0.0062 (n=10000)	6e-04 (n=10000)
0.4	uniform	uniform	0.0762 (n=5000)	0.0736 (n=10000)	0.004 (n=10000)	2e-04 (n=10000)
0.4	uniform	recessive	0.1598 (n=5000)	0.1534 (n=10000)	0.0086 (n=10000)	0.00021 (n=9500)
0.4	uniform	additive	0.0824 (n=5000)	0.0738 (n=10000)	0.0048 (n=10000)	5e-04 (n=10000)
0.4	0.1	uniform	0.105 (n=5000)	0.109 (n=10000)	0.0017 (n=10000)	1e-04 (n=10000)
0.4	0.1	recessive	0.131 (n=5000)	0.1275 (n=10000)	0.00778 (n=9000)	0 (n=8500)
0.4	0.1	additive	0.1086 (n=5000)	0.103 (n=10000)	0.0028 (n=10000)	5e-04 (n=10000)
0.4	0.5	uniform	0.071 (n=5000)	0.0681 (n=10000)	0.0053 (n=10000)	3e-04 (n=10000)

p_{self}	s	h	All benefits	Compensation	Competition	No benefit
0.4	0.5	recessive	0.1672 (n=5000)	0.1617 (n=10000)	0.0107 (n=10000)	9e-04 (n=10000)
0.4	0.5	additive	0.0714 (n=5000)	0.0718 (n=10000)	0.0083 (n=10000)	5e-04 (n=10000)
0.4	1	uniform	0.0542 (n=5000)	0.0547 (n=10000)	0.0066 (n=10000)	0 (n=10000)
0.4	1	recessive	0.1468 (n=5000)	0.149 (n=10000)	5e-04 (n=10000)	4e-04 (n=10000)
0.4	1	additive	0.0626 (n=5000)	0.0532 (n=10000)	0.0074 (n=10000)	8e-04 (n=10000)
0.6	uniform	uniform	0.0576 (n=5000)	0.0578 (n=10000)	0.0043 (n=10000)	7e-04 (n=10000)
0.6	uniform	recessive	0.0702 (n=5000)	0.0672 (n=10000)	0.0013 (n=10000)	7e-04 (n=10000)
0.6	uniform	additive	0.0618 (n=5000)	0.058 (n=10000)	0.0057 (n=10000)	5e-04 (n=10000)
0.6	0.1	uniform	0.0886 (n=5000)	0.0917 (n=10000)	0.0021 (n=10000)	3e-04 (n=10000)
0.6	0.1	recessive	0.0938 (n=5000)	0.1021 (n=10000)	2e-04 (n=10000)	5e-04 (n=10000)
0.6	0.1	additive	0.0906 (n=5000)	0.091 (n=10000)	0.0036 (n=10000)	2e-04 (n=10000)
0.6	0.5	uniform	0.0546 (n=5000)	0.0512 (n=10000)	0.006 (n=10000)	7e-04 (n=10000)
0.6	0.5	recessive	0.0634 (n=5000)	0.0607 (n=10000)	0.0031 (n=10000)	6e-04 (n=10000)
0.6	0.5	additive	0.0594 (n=5000)	0.0557 (n=10000)	0.006 (n=10000)	2e-04 (n=10000)
0.6	1	uniform	0.0488 (n=5000)	0.0399 (n=10000)	0.0035 (n=10000)	8e-04 (n=10000)
0.6	1	recessive	0.048 (n=5000)	0.0417 (n=10000)	4e-04 (n=10000)	6e-04 (n=10000)

p_{self}	s	h	All benefits	Compensation	Competition	No benefit
0.6	1	additive	0.0516 (n=5000)	0.0445 (n=10000)	0.0053 (n=10000)	4e-04 (n=10000)
0.8	uniform	uniform	0.0374 (n=5000)	0.0433 (n=10000)	0.0035 (n=10000)	4e-04 (n=10000)
0.8	uniform	recessive	0.042 (n=5000)	0.0455 (n=10000)	0.0023 (n=10000)	2e-04 (n=10000)
0.8	uniform	additive	0.042 (n=5000)	0.0416 (n=10000)	0.0026 (n=10000)	5e-04 (n=10000)
0.8	0.1	uniform	0.0658 (n=5000)	0.0665 (n=10000)	0.0022 (n=10000)	5e-04 (n=10000)
0.8	0.1	recessive	0.0678 (n=5000)	0.0747 (n=10000)	7e-04 (n=10000)	4e-04 (n=10000)
0.8	0.1	additive	0.066 (n=5000)	0.066 (n=10000)	0.0018 (n=10000)	4e-04 (n=10000)
0.8	0.5	uniform	0.0386 (n=5000)	0.0354 (n=10000)	0.0042 (n=10000)	0.0012 (n=10000)
0.8	0.5	recessive	0.0366 (n=5000)	0.0323 (n=10000)	0.0023 (n=10000)	3e-04 (n=10000)
0.8	0.5	additive	0.0404 (n=5000)	0.0375 (n=10000)	0.0034 (n=10000)	1e-04 (n=10000)
0.8	1	uniform	0.0308 (n=5000)	0.0301 (n=10000)	0.0032 (n=10000)	3e-04 (n=10000)
0.8	1	recessive	0.033 (n=5000)	0.0285 (n=10000)	4e-04 (n=10000)	5e-04 (n=10000)
0.8	1	additive	0.0388 (n=5000)	0.0322 (n=10000)	0.0041 (n=10000)	2e-04 (n=10000)
1	uniform	uniform	0.0132 (n=5000)	0.0138 (n=10000)	0.0011 (n=10000)	6e-04 (n=10000)
1	uniform	recessive	0.0102 (n=5000)	0.0072 (n=10000)	8e-04 (n=10000)	2e-04 (n=10000)
1	uniform	additive	0.013 (n=5000)	0.0127 (n=10000)	0.0013 (n=10000)	4e-04 (n=10000)

p_{self}	s	h	All benefits	Compensation	Competition	No benefit
1	0.1	uniform	0.0242 (n=5000)	0.0223 (n=10000)	4e-04 (n=10000)	2e-04 (n=10000)
1	0.1	recessive	0.0214 (n=5000)	0.0235 (n=10000)	9e-04 (n=10000)	8e-04 (n=10000)
1	0.1	additive	0.0196 (n=5000)	0.0151 (n=10000)	5e-04 (n=10000)	3e-04 (n=10000)
1	0.5	uniform	0.0148 (n=5000)	0.0154 (n=10000)	0.0021 (n=10000)	6e-04 (n=10000)
1	0.5	recessive	0.012 (n=5000)	0.0084 (n=10000)	0.0028 (n=10000)	5e-04 (n=10000)
1	0.5	additive	0.0168 (n=5000)	0.0161 (n=10000)	0.0023 (n=10000)	2e-04 (n=10000)
1	1	uniform	0.023 (n=5000)	0.0228 (n=10000)	0.0023 (n=10000)	6e-04 (n=10000)
1	1	recessive	0.0268 (n=5000)	0.0267 (n=10000)	8e-04 (n=10000)	6e-04 (n=10000)
1	1	additive	0.0246 (n=5000)	0.025 (n=10000)	0.0027 (n=10000)	5e-04 (n=10000)

Figure S1: **Overview of the life cycle model:** The number of deleterious mutations impacting embryo fitness over time in burn in simulations, across selective (s) and dominance (h) coefficients, and selfing rates (on the x-axis).

Figure S2: **Summaries of our simulated populations at equilibrium:** (A) The mean embryo fitness, (B) Realized selfing rate, (C) Correlation between embryo and postembryonic fitness, (D) Post-embryo fitness, across selective (s , colors and shapes) and dominance (h , facets) coefficients, and selfing rates (on the x-axis). The number of deleterious mutations impacting embryo fitness over time in burn in simulations, across selective (s) and dominance (h) coefficients, and selfing rates (on the x-axis).

Figure S3: **Proportion of introductions resulting in fixation** as a function of selfing rate (x-axis), the benefit of polyembryony (rows), selection against new mutations (columns), and the dominance of new mutations (color). The purple line denotes neutral expectations. Note that fixation probabilities for additive mutations and those taking their value from a uniform distribution are very similar.

Figure S4: **Mean time to fixation** of the polyembryony allele across selective (s, colors and shapes) and dominance (h, faceted columns) coefficients, and selfing rates (on the x-axis), for each model (faceted rows).

Figure S5: **Correlation in fitness** of the hypothetical two embryos in a seed, before polyembryony evolves as a function of the fitness effect of new mutations (on the x-axis), the selfing rate (color), and the dominance effect of new mutations (columns).