1 **Inbreeding reduces long-term growth of Alpine ibex populations.**

2

3 **Authors:** Claudio Bozzuto[1, ‡, +], Iris Biebach[1,+], Stefanie Muff[1, 2], Anthony R. Ives[3,*],

4 Lukas F. Keller[1,4,*].

5

6 **Affiliations:**

7 [1] Department of Evolutionary Biology and Environmental Studies, University of

8 Zurich, Winterthurerstrasse 190, 8057 Zurich, Switzerland.

9 [2] Department of Biostatistics, Epidemiology, Biostatistics and Prevention Institute,

10 University of Zurich, Hirschengraben 84, 8001 Zurich, Switzerland.

11 [3] Department of Zoology, University of Wisconsin-Madison, Madison, WI 53706,

12 USA.

13 [4] Zoological Museum, University of Zurich, Karl-Schmid-Strasse 4, 8006 Zurich,

14 Switzerland.

15

16 ‡ Current address: Wildlife Analysis GmbH, Oetlisbergstrasse 38, 8053 Zurich,

17 Switzerland.

18

19 + These authors contributed equally to this work.

20 * Correspondence to: lukas.keller@ieu.uzh.ch and arives@wisc.edu.

21

22

Introductory paragraph

Since Darwin's time we have known that inbreeding has negative effects on individuals, and conservation efforts to preserve rare species routinely employ strategies to reduce inbreeding. Despite this, there are few clear examples in nature of inbreeding decreasing the growth rates of populations and the extent of population-level effects of inbreeding in the wild remains controversial. Here we take advantage of a long-term data set spanning nearly 100 years and 26 reintroduced Alpine ibex (*Capra ibex ibex*) populations to show that inbreeding substantially reduced per capita population growth rates, increasingly so in harsher environmental conditions. Populations with high average inbreeding ($F \approx 0.2$) exhibited population growth rates reduced by 71% compared to populations with no inbreeding. Our results show that inbreeding can have long-term demographic consequences even in the presence of large environmental variation and when deleterious alleles may have been purged during bottlenecks. Thus, efforts to guard against inbreeding effects in populations of endangered species have not been misplaced.

39 Main text

40 Inbreeding depression, the harmful effects of inbreeding on the fitness of individuals,

41 is widespread among plants and animals, with recent genomic studies revealing an

42 even greater impact on individual fitness than previously thought[1]. However, reduced

43 fitness of individuals due to inbreeding does not necessarily lead to reduced

44 population growth rates[2,3,4] in the same way that natural selection need not impact

45 population growth[5]. Instead, theory predicts that the degree to which inbreeding

46 depression affects population growth will depend on several aspects of the ecology

47 and life-history of a species[3,6]. For example, in species experiencing density-

48 dependent population growth, even substantial inbreeding depression at the individual

49 level need not translate into reduced population growth because fitness reductions due

50 to inbreeding may be compensated by fitness gains due to relaxed competition. Under

51 such circumstances, inbred individuals may produce enough offspring to maintain

52 population growth (soft selection[2]).

53   Collecting unequivocal evidence for population-level effects of inbreeding is

54 difficult because it requires many replicated populations that differ in levels of

55 inbreeding to be monitored over many generations. Hence, the extent of population-

56 level effects of inbreeding in the wild remains controversial[7,8,9] and we currently lack

57 an understanding of the magnitude of the consequences of inbreeding depression for

58 long-term population growth in natural populations[10]. Here, we take advantage of a

59 long-term data set of 26 reintroduced Alpine ibex populations (Supplementary Figures

60 1 and 2) spanning 23-96 years to show that inbreeding can reduce long-term

61 population growth rates in the wild.

62   Alpine ibex were extirpated from the Alps by the end of the 19th century, with

63 only a single population surviving in the Gran Paradiso region in northern Italy[11].

64 Starting in 1906, Alpine ibex were taken from Gran Paradiso, bred in Swiss zoos, and

65 released back into their former habitat. These reintroductions are well documented[12],

66 with counts of the released individuals, subsequent time series of annual abundance

67 counts, and counts of the numbers of harvested animals (Supplementary Table 1).

68 Genetic data suggest little natural migration between populations after reintroductions

69 ceased[13], making the populations distinct replicates for the purpose of this study.

70   Ibex populations in our study experienced up to four reintroduction-associated

71 bottlenecks[13]. The first bottleneck occurred when the Swiss breeding program was

72 initiated with ~88 individuals from Gran Paradiso[11]. First reintroductions into the wild

73 with ibex from the Swiss breeding program caused a second set of bottlenecks

74 (founder population sizes: 18-78). The third set of bottlenecks took place when

75 individuals from the first wild founded populations were used to found additional wild

76 populations (founder population sizes: 9-137). Subsequent reintroductions sourced

77 some founder individuals from populations that already had experienced three

78 bottlenecks, thus causing a fourth bottleneck[13]. Genetically, the bottlenecks were

79 twice as pronounced as expected from the number of released founders because, on

80 average, only about half of the founders contributed genes to the following

81 generations[14].

82   These serial bottlenecks resulted in considerable genetic drift and inbreeding[15].

83 In this study, we use the term inbreeding to refer to the average identity by descent

84 across individuals that accumulates under random mating in a population of finite size

85 in concert with genetic drift[16,17 p. 167-171]. We quantified this inbreeding using 37

86 microsatellite loci and population-specific F_{ST} estimates, which measure the

87 probability of identity by descent of pairs of alleles at a locus within populations

88 relative to pairs of alleles from different populations[18,19]. Population-specific F_{ST}

89estimates are calculated for each population separately. Averaged across all

90populations they yield the familiar global F_{ST} estimate[19]. There is no evidence for

91inbreeding due to non-random mating within Alpine ibex populations ($F_{IS} \approx 0$), hence

92population-specific F_{ST} estimates quantify total inbreeding since the last common

93ancestral population[18,20] at the beginning of the reintroduction programm about 12.5

94generations ago[13]. Population-specific F_{ST} does not suffer from the same lack of power

95as individual inbreeding coefficients estimated from limited molecular data[10,15]

96because limited dispersal and population structure create identity disequilibrium and

97thus correlation in heterozygosity across loci[21].

98   To estimate long-term population growth rates, we fitted a nonlinear state-

99space population model to each of the 26 populations containing terms for the

100continuous rate of increase (r_0), density dependence in population growth, the number

101of reintroduced as well as harvested individuals, environmental and demographic

102stochasticity, and sampling variability. Fig. 1 shows two example populations and the

103fit of the state-space model to the data. To quantify the impact of inbreeding on

104population growth, we regressed r_0 estimates against inbreeding and other covariates.

105Using a conventional regression approach would substantially underestimate the

106inbreeding effects, since inbreeding levels are only known with uncertainty, thus

107violating the important assumption of regressions that covariates are known exactly[22].

108To obtain unbiased estimates of the effects of inbreeding on population growth, the

109uncertainty in population-specific F_{ST} estimates needs to be incorporated in the

110statistical analysis. We accounted for these uncertainties using Bayesian

111heteroscedastic measurement-error models[23]. In addition, we explicitly accounted for

112uncertainties in estimates of r_0, and because larger values of r_0 showed systematically

113larger variances (Supplementary Figure 3A), we log-transformed r_0 in all our

114statistical models. In addition to inbreeding, the models included as covariates the

115year when the time series of a population began and climatic variables known to

116affect ibex populations[12], but averaged across the entire length of the time series to

117capture environmentally induced spatial variation in population growth: mean daily

118summer and winter temperatures, mean daily summer and winter precipitation, and

119winter snow cover. Due to the Bayesian nature of the analysis, model selection was

120guided by DIC minimization.

121

122*Results*

123   The best-fitting model (Table 1), which captured 79% of the variation in log-

124transformed continuous rates of increase among the 26 populations (Supplementary

125Tables 5 and 6), revealed evidence for a negative effect of inbreeding on population

126growth rates in conjunction with climatic variables (Fig. 2). According to the model, a

127population-specific F_{ST} of 0.21 (the maximum inbreeding observed in this study)

128reduced the expected r_0 by 71% with respect to a hypothetical population with zero

129inbreeding, while a population-specific F_{ST} of 0.03 (the observed minimum) lead only

130to a 17% reduction. As expected from measurement error theory, regressions that did

131not account for the uncertainty in estimates of inbreeding yielded substantially

132downwardly biased estimates of the impacts of inbreeding (Supplementary Table 5).

133   Environmental factors represented by the averaged climate variables had

134strong effects on population growth rates, and the magnitude of inbreeding effects

135depended on these environmental conditions (Table 1): inbreeding effects on

136population growth rates were absent in areas with low summer precipitation, but were

137increasingly pronounced in areas with wetter summers (Fig. 3). This may represent a

138direct effect of summer precipitation through adverse effects on neonatal mortality or

139body growth[12]. However, given that the climate variables represent averages across

140the entire time series period, the effect may have different, indirect causes, which we

141cannot identify since our study was not designed to isolate the causes of

142environmental variation in r_0. Whatever the causality, our findings support theoretical

143predictions that ecological processes can modify the population-level effects of

144inbreeding[3], and they mirror many studies at the individual level that have shown

145inbreeding depression to vary with environmental conditions[24].

146   We have shown that inbreeding reduces long-term population growth rates.

147But population growth rates in turn can affect levels of inbreeding: low population

148growth rates will keep populations small, which increases genetic drift and hence the

149amount of inbreeding[15,25]. Could the observed effect of inbreeding on population

150growth be confounded by an effect of population growth on inbreeding? Theory does

151not predict a straightforward effect of population growth rate on inbreeding. Instead,

152the expected inbreeding in a randomly mating population is determined by the

153harmonic mean population size[26], which in turn is a nonlinear function of population

154growth rate and the founding population size (equation A3 in[27]). In our data, r_0 and

155harmonic mean population sizes were not correlated (r=-0.07, 95% CI: -0.44 to 0.33,

156p=0.74, N=26; rearranging equation A3 in[27] to obtain a linear relationship), and

157adding harmonic mean population sizes to our regression models for r_0 did not affect

158the overall conclusions (Supplementary Tables 1, 6 and 7). Thus, we found no

159evidence that variation in r_0 among the populations in our data set generated

160differences in inbreeding. Instead, variation in founder group size, admixture of the

161founder groups, and carrying capacity appear to be the major sources of variation in

162inbreeding that arose among ibex populations since the beginning of the

163reintroduction program[14,15].

164

165 *Discussion*

166   Our results support the emerging view that genetic processes can substantially

167 affect long-term population growth, even in populations that may have purged

168 deleterious recessive alleles during successive bottlenecks[28,29]. Our study design did

169 not allow the detection of purging, hence we can only speculate about purging in

170 Alpine ibex. Some purging may have occurred, as in some invasive species[28], because

171 conditions promoting decreases in population growth rates through inbreeding (i.e.,

172 hard selection) also lead to purging[6]. However, the efficiency of purging depends on

173 population size and the relatively small bottleneck sizes of the Alpine ibex

174 populations (mean number of founding chromosomes: 42)[14] imply that purging would

175 mostly remove strongly deleterious mutations[29,30]. The more weakly deleterious

176 alleles may have drifted to fixation during the bottlenecks, creating drift load[31] but

177 those that were not fixed may be purged now that population sizes increase[29]. The

178 combination of drift load and limited purging likely explains the substantial

179 inbreeding effects in reintroduced Alpine ibex populations. Thus, genetic rescue with

180 increased population growth may result if ibex were translocated among

181 populations[1,32].

182   The population time series we analysed included periods of rapid growth and

183 substantial declines, and show the impact of density dependence (Fig. 1). Yet the

184 inbreeding effects were strong enough to overcome density-dependent compensation

185 and reduce the growth of the reintroduced populations. One factor that may contribute

186 to the strong demographic consequences of inbreeding in Alpine ibex is the relatively

187 weak density dependence in many populations (Supplementary Table 4). Weak

188 density dependence suggests that Alpine ibex do not compete strongly for limited

189resources. This leads to hard selection and, hence, reduced population growth because

190deaths due to inbreeding do not substantially relax the already low competition[2,3].

191Thus, the relatively weak density dependence may have contributed to the observed

192demographic consequences of inbreeding in Alpine ibex.

193   Our results indicate that inbreeding can substantially lower long-term

194population growth even when deleterious alleles may have been purged during

195bottlenecks[28] and when populations are reintroduced into habitat to which they are

196adapted[24]. In line with theoretical predictions, we found that ecological conditions

197modify the extent to which inbreeding affects population growth, but they are unlikely

198to completely mask them[3]. Thus, when ecological conditions produce hard selection,

199for example when density dependence is relatively weak as in the case of most

200populations of conservation concern, inbreeding depression at the individual level can

201lead to large reductions in population growth. Ultimately, these effects can lead to

202increased extinction rates at the population and species level[33,34,35].

203   As we have shown, detecting population-level effects of inbreeding requires

204an exceptional dataset, with many populations that differ in inbreeding and enough

205environmental data to factor out other causes of reduced population growth rates.

206Even with the exceptional Alpine ibex data, detecting population-level inbreeding

207effects was a statistical challenge. Thus, it is not surprising that, despite the many

208examples of individual-level inbreeding effects in nature, population-level effects on

209the dynamics of unmanaged wild living populations have rarely been

210unambiguously documented.

211

212

213

Author contributions

I.B. and L.F.K. conceived and designed the study and collected the samples. I.B. performed the genetic analyses and compiled the demographic data. C.B. and A.R.I. analysed the time series data, and S.M. performed the measurement error modelling. All authors wrote the manuscript.

Competing interests

The authors declare no competing financial interests.

234**Tables**

235**Table 1:** Parameter estimates (posterior means and 95% credible intervals) for the ef-

236fects of inbreeding ($\hat{\beta}_F$), the year when the time series began ($\hat{\beta}_{year}$), mean precipita-

237tion in winter ($\hat{\beta}_{pw}$), mean precipitation in summer ($\hat{\beta}_{ps}$), and two interaction terms (

238$\hat{\beta}_{year \cdot ps}$ ∧ $\hat{\beta}_{F \cdot ps}$) on the log-transformed intrinsic per capita population growth rate r_0.

239Inbreeding reduced population growth in interaction with summer precipitation

240(Model 1) and when the interaction was omitted (Model 2). For modeling details see

241Methods.

242

Model	DIC	$\hat{\beta}_F$	$\hat{\beta}_{year}$	$\hat{\beta}_{pw}$	$\hat{\beta}_{ps}$	$\hat{\beta}_{year \cdot ps}$	$\hat{\beta}_{F \cdot ps}$
1	−6.61	−5.85 [−13.02, 0.84]	0.0223 [0.0111, 0.0343]	0.610 [0.457, 0.770]	−0.510 [−0.713, −0.325]	−0.0218 [− 0.0345, −0.0098]	−4.22 [−8.57, −0.71]
2	0.261	−8.56 [−15.61, −1.54]	0.0240 [0.0116, 0.0367]	0.622 [0.453, 0.792]	−0.486 [−0.673, −0.300]	−0.0213 [− 0.0341, −0.0087]	

243

244

245

246**Figure legends**

247

248**Fig. 1**.

249Time series of annual abundance counts (dots) together with the nonlinear state-space

250model fits (lines) for the relatively inbred population 2 (red, with point estimates

251r_0=0.14 and F_{ST}=0.18) and the relatively outbred population 23 (blue, with point

252estimates r_0=0.34 and F_{ST} =0.06). Numbers of released (> 0) and harvested animals (<

2530) are shown on separate x- and y-axes. Note the clear differences in population

254growth in the first 15-20 years, reflected in the corresponding differences in estimates

255of the per capita growth rates, r_0. Time series characteristics are summarized in

256Supplementary Table 1, and estimated model parameters are presented in

257Supplementary Tables 2 and 4.

258

259**Fig. 2.**

260(**A**) Main effect of inbreeding, measured by population-specific F_{ST}, on the intrinsic

261rate of population increase, r_0, among 26 populations of Alpine ibex. Values of r_0 were

262adjusted for all covariates in the model except for the main effects of F_{ST}. The area of

263each point is inversely proportional to the error variances in the estimates of r_0 from

264the time series, which are used in the error model to down-weight observations with

265large uncertainty. Panels (**B**) and (**C**) on the right give the absolute frequency

266distribution of estimates of r_0 and F_{ST} for the 26 Alpine ibex populations.

267

268**Fig. 3.**

269Visualization of the estimated interaction effects of inbreeding and summer

270precipitation on population growth rates (table S5), illustrated for minimal (left),

271mean (middle) and maximal values (right) of summer precipitation. Increased summer

272precipitation reduced population growth rates, implying that inbreeding effects on

273population growth rates were more pronounced in harsher environments. The

274shadowed areas represent the 95% credible ranges. Note the logarithmic scale for the

275y-axis when comparing Fig. 3 to Fig. 2.

276

METHODS

Study populations and population-related data. Alpine ibex populations in Switzerland have been monitored closely since their reintroductions, with yearly abundance counts conducted usually in spring[36]. During spring, ibex are found in fairly restricted areas below the snow line and just above timberline[37], and are therefore easier to count than most other ungulates. Hence, Saether *et al.*[38] found the error in population censuses to be small (median coefficient of variation across 28 ibex populations: 5.1%). In addition to abundance count data, we had time series for (i) the number of released animals, (ii) the number of harvested animals for all but a single non-hunted population (number 22), and (iii) the number of animals that were removed for translocations. Initial releases were performed at the end of winter. Harvest of ibex populations started in 1977 when many populations had grown to high densities. In some populations ibex were removed for translocation to other populations. In our study, these ibex were considered to be harvested and were added to the count of harvested animals in that year. This explains why the first year for harvesting can be prior to 1977 (Supplementary Table 1, column "Hunt"). We analysed data of 26 Alpine ibex populations, with time series ranging from 23 to 96 years (Supplementary Table 1). For this study we added genetic data from three populations (number 24, 25 and 26) to 23 populations that had been previously analysed genetically[13].

Inference of inbreeding level. An average of 36.9 (range 17–102) individuals from each of the 26 populations were genotyped at 37 neutral microsatellite loci as detailed in Biebach *et al.*[13]. Most inbreeding in the reintroduced Alpine ibex populations

302accumulated in concert with genetic drift during founder events and during the time

303when population sizes were low following reintroduction[15]. We quantified the

304inbreeding that arose since the reintroduction program started over 100 years ago

305using marker-based population-specific F_{ST} [18,19]. Population-specific F_{ST} of population

306i is defined as $F_{ST\_i} = (\theta_i - \theta_A) / (1 - \theta_A)$, where θ_i is the probability of a gene being

307identical by descent (IBD) within population i and θ_A is the probability of a gene

308being IBD between all populations[18]. In line with theory, simulations have shown that

309F_{ST} explains a substantial part of the effects of deleterious mutations on population

310fitness[39], making F_{ST} a suitable measure of average population inbreeding for this

311study.

312We used a Bayesian framework in a modified version of 2MOD[40] to estimate

313population-specific F_{ST} for 42 ibex populations[15]. For this study we used the results of

31426 populations for which we also had abundance data. All parameters were given

315uninformative flat prior distributions. We used the non-equilibrium drift model in

3162MOD that estimates inbreeding relative to the last common ancestral population. The

317model assumes that the reciprocal of the mutation rate is much longer than the

318divergence time[40], which is a reasonable assumption for the reintroduced ibex

319populations since they were founded no more than 12.5 generations ago. In our case,

320the ancestral reference population is the Gran Paradiso population in Italy, the single

321remaining population of Alpine ibex before animals were transferred to found the zoo

322populations that were the source for the first reintroductions. Thus, the estimated

323inbreeding coefficient measures the accumulated inbreeding from the start of the

324reintroduction program in 1906. The ancestral reference population used here differs

325from that used in previous studies of inbreeding in Alpine ibex[15], hence results are not

326directly comparable.

327   The observed degree of inbreeding will depend on the composition of the

328 founder population and the harmonic mean population size after founding (inbreeding

329 N_e). The length of the time series is not expected to influence the inbreeding

330 coefficient (r=–0.27, 95% CI: -0.60 to 0.13, N=26) because harmonic mean

331 population size is mainly determined by the small population sizes early in the

332 population development[15]. As expected given that inbreeding measures IBD and

333 homozygosity measures identity-in-state, mean observed homozygosity and

334 population-specific F_{ST} were only moderately correlated (r=0.55, 95% CI: 0.21 to

335 0.77, N=26) across the 26 populations. F_{ST} estimates are not only affected by

336 statistical sampling variance, but also by genetic sampling variance caused by genetic

337 drift[20], hence their credible intervals are substantial even when based on three dozens

338 of loci (Supplementary Table 2). We estimated mean, variance, and 95% credible

339 intervals of population-specific F_{ST} for each population from 450,000 iterations with

340 the R-package STATS[41].

341   Next generation sequencing methods offer alternative measures of individual

342 inbreeding coefficients[10] like runs of homozygosity (ROH). ROH (mean total length

343 >5 Mb) were estimated from 41 907 SNPs obtained with RAD-sequencing for 76 ibex

344 of 10 populations[xx], with the difference that the minimum SNP-density was set to 1

345 SNP per 100kb[xx]. Averaged ROH across each of the 10 populations yielded very

346 similar estimates of population-level inbreeding as the population-specific F_{ST}

347 estimates (r=0.81, 95% CI: 0.38 to 0.95, N=10, Supplementary Table 8). Thus, next

348 generation sequencing data confirm the population-level estimates of inbreeding

349 obtained with microsatellites in this study.

350

351 **Population dynamics: Model description.** In constructing the population dynamics

352model, we followed the dynamically important steps of a population through a year.

353We started the cycle with population size in spring, coinciding with the time when

354counts were made. We then added reproduction in summer. In autumn, hunting of

355adults takes place before the December-January rut[42]. Thus, only the proportion not

356killed was retained in the model. Winter is the season when most natural mortality

357occurs; thus, we added density-dependent mortality. Finally, we added the

358reintroduced animals.

359   Combining these components of ibex population dynamics leads to the

360discrete-time dynamical equation (1):

361

$$N_t = N_{t-1} e^{r_0} \left(1 - \frac{H_{t-1}}{N_{t-1}}\right) f\left(N_{t-1} - H_{t-1}\right) \left(1 + \frac{R_{t-1}}{e^{r_0}\left(N_{t-1} - H_{t-1}\right) f\left(N_{t-1} - H_{t-1}\right)}\right) e^{\varepsilon_t} e^{\phi_t} \tag{1}$$

362

363Here, N_t is the "true" (unobserved) population abundance before reproduction in

364spring in year t; r_0 is the density-independent (intrinsic) continuous rates of increase;

365H_t is the number of adult animals harvested in year t; $f\left(N_{t-1} - H_{t-1}\right)$ is a function

366giving density-dependent survival that depends on the number of individuals in the

367population, excluding kids born that year and harvested individuals; R_t is the number

368of individuals added to the population following overwintering survival; ε_t is a

369random variable giving the effect of environmental variation on population growth,

370and ϕ_t is a random variable for demographic stochasticity. We assumed that the

371function f is a Gompertz equation, so that $f\left(N_{t-1} - H_{t-1}\right) = \exp\left(b \log\left(N_{t-1} - H_{t-1}\right)\right)$,

372where $b \leq 0$ and smaller parameter values imply greater density dependence. Taking

373$x_t = \log\left(N_t\right)$ and rearranging gives the model

374

$$x_t = r_0 + \log\left(\left(e^{x_{t-1}} - H_{t-1}\right)^{|1+b|} + e^{-r_0} R_{t-1}\right) + \varepsilon_t + \phi_t \tag{2}$$

375

376   The year-to-year fluctuations in "true" population abundances, referred to as

377 process variation in state-space models, are assumed to have two components:

378 demographic stochasticity (ϕ_t) that decreases with increasing population size, and

379 environmental variability (ε_t). Environmental variation is assumed to have variance

380 independent of the mean (on a log scale), and thus we take ε_t as independent draws

381 from a Gaussian random variable with mean 0 and variance σ^2_{env}. In contrast,

382 demographic stochasticity depends on the mean log population size. Assuming that

383 demographic stochasticity follows a Poisson process, the resulting variation can be

384 approximated by treating ϕ_t as a Gaussian random variable with mean 0 and variance

385 $\log\left(\sigma^2_{dem} e^{-x_{t-1}} + 1\right)$.

386   Since the "true" population abundance N_t cannot be observed directly, our

387 model takes a state-space form to account for the observation process. We assume that

388 population counts follow a binomial process, and therefore observation error can be

389 approximated as a Gaussian random variable η_t with mean 0 and variance

390 $\log\left(\sigma^2_{obs} e^{-x_{t-1}} + 1\right)$. A full statement of the state-space model is

391

$$x_t = r_0 + \log\left(\left(e^{x_{t-1}} - H_{t-1}\right)^{|1+b|} + e^{-r_0} R_{t-1}\right) + \varepsilon_t + \phi_t \tag{3a}$$
$$y_t = x_t + \eta_t \tag{3b}$$
$$\varepsilon_t N\left(0, \sigma^2_{env}\right) \tag{3c}$$
$$\phi_t N\left(0, \log\left(\sigma^2_{dem} e^{-x_{t-1}} + 1\right)\right) \tag{3d}$$
$$\eta_t N\left(0, \log\left(\sigma^2_{obs} e^{-x_{t-1}} + 1\right)\right) \tag{3e}$$

392

393 where y_t is the log-transformed observed number of individuals in the population in

394 year t. Equations 3a and 3b are referred to as the process and observation equations,

395respectively, of the state-space model.

396

397**Population dynamics: Model fitting to data.** The model is nonlinear and in state-

398space form (Eqs. 3a-e), and therefore we used an extended Kalman filter (EKF) to

399calculate likelihoods and obtain the maximum likelihood parameter estimates[43], using

400a procedure similar to Schooler *et al.*[44]; see also Supplementary Information. Time

401series for several Alpine ibex populations have been analysed previously by Saether

402*et al.*[38] using a state-space model, although in a Bayesian context. In contrast to our

403study, Saether *et al.*[38] did not include the release periods in their analyses. We

404included release periods because some of them span a considerable number of years

405of early population growth (Supplementary Table 1) when density dependence was

406still low, therefore containing valuable information for estimating r_0. To prepare the

407time series for the EKF analysis, for each population we (i) discarded years prior to

408the first available census count, (ii) substituted subsequent counts of zero animals

409with 0.01 times the lowest non-zero census count, and (iii) substituted missing values

410with zero in the covariate time series of harvested and released animals.

411   We estimated simultaneously the five parameters r_0 (continuous rate of

412increase), b (density dependence), σ^2_{env} (environmental stochasticity), σ^2_{dem}

413(demographic stochasticity), and σ^2_{obs} (observation error). Further, because the initial

414population sizes were small and therefore prone to observation error, we treated the

415first point in each time series, x_0, as an additional parameter to be estimated[45].

416   We used simulated annealing to find optimal starting parameter values for the

417maximization routine, and then refined the results using the Nelder-Mead Simplex

418method[46]. Although it is theoretically possible to distinguish environmental,

419demographic, and observation variation solely from time series data, in practice this is

420often impossible due to small sample sizes and the similarity of effects of different

421sources of variability on the observed time series. Therefore, the estimated values of

422σ^2_{env}, σ^2_{dem}, and σ^2_{obs} are sometimes zero (Supplementary Table 4), even though in

423reality they will not be. These zero estimates, however, will have very little effect on

424the estimates of r_0, the main target of the analyses.

425   To quantify the uncertainty in the r_0 point estimates, we calculated 95%

426confidence intervals using profile likelihoods[47]. Because the 95% confidence intervals

427around r_0 were an important ingredient of the final regression analysis, we checked

428whether this uncertainty was significantly correlated to time series length or to the

429number of missing values (Supplementary Table 1). We found no significant

430correlation between $1/\hat{\sigma}^2_{\log(r_0)}$ and time series length (r=0.01, 95% CI: -0.38 to 0.39,

431N=26) or the number of missing data (r=–0.06, 95% CI: -0.44 to 0.33, N=26).

432

433**Regression analysis: Model description.** We log-transformed r_0 for the regression to

434account for three aspects of the estimates of r_0: (i) the uncertainty in estimates of r_0

435increased with the point estimate (Supplementary Figure 3a), (ii) most of the

436confidence intervals around single r_0 estimates were right-skewed, and (iii) the

437distribution of r_0 point estimates was right-skewed (see also Supplementary Figure

4383b). Log-transforming r_0 resolved all of these issues.

439   The regression models included as covariates the population-specific F_{ST}, five

440climate variables (see below), and the year when the time series of a population began

441(Supplementary Table 1, column "*Period*"). We included the year when the time

442series began to account for possible changes in the suitability of habitats as

443reintroductions progressed. The climate variables were included because studies have

444shown effects of weather conditions on Alpine ibex population growth[12]. We obtained

445relevant data from the Swiss Federal Office of Meteorology and Climatology

446MeteoSwiss. All relevant weather stations for this study are located in a population's

447habitat or in the immediate vicinity. From ecological knowledge of ibex, we split the

448year into summer (May – October) and winter (November – April). For each

449population we calculated one mean spanning the respective time series period for the

450following weather measures (Supplementary Table 3): Daily mean air temperature in

451summer (ts) and winter (tw) (both in degree Celsius), daily total precipitation in

452summer (ps) and winter (pw) (both in millimeter), and daily total snow cover in

453winter (sw) (in centimeter). Not all weather stations were recording data when the

454populations in this study were reintroduced. Thus, for some populations the climatic

455variables are calculated over a shorter time period than we had time series data

456(Supplementary Table 1). These climate variables, averaged across the entire length

457of the time series, are a measure of the climate zone a population inhabits and are

458used to account for environmentally induced spatial variation in population growth

459rates among populations. The effects captured by these averaged climate variables

460include indirect effects of variables that may covary with climate zone, such as spatial

461variation in food quality or quantity. The climate variables cannot, therefore, be

462interpreted in the same way that they have been in previous within-population

463studies[12,38].

464   All covariates were centred by subtracting their respective mean value. Due to

465the log-transformation of r_0, variances were transformed by the delta rule for variance

466transformations: $\hat{\sigma}^2_{\log(r_0)} = \hat{\sigma}^2_{r_0}/r_0^2$ (Supplementary Figure 3b). In order to account for the

467population-dependent (heteroscedastic) error in $\log(r_0)$, a random effects term with a

468population-specific variance, denoted as δ_y, was added to the linear regression model.

469   Importantly, not only the response $\log(r_0)$, but also the covariate of interest,

470F_{ST}, has been estimated with uncertainty, that is, with measurement error. It is,

471however, a fundamental assumption of regression models that covariates have been

472precisely measured, and a violation of this assumption may lead to biased estimates of

473the regression coefficients [22,48]. Here, population-specific estimates of the uncertainty

474in the F_{ST} point estimates were available (see Inference of inbreeding level), thus we

475could properly account for it by explicitly formulating an error model for this

476covariate. Note that measurement errors in covariates correlated with inbreeding

477could also bias the estimates of the inbreeding effects[48], but because inbreeding was

478not substantially correlated with other covariates in the regression model (all r $\leq$

4790.27), we only modelled measurement error in inbreeding. We formulated a Bayesian

480hierarchical measurement error model following the description in Muff $et\ al.$[23],

481where the first level is the Gaussian regression model relating population growth to

482the true covariates (Eq. 4a), the second level is the classical Gaussian error model for

483the observed F_{ST} that accounts for unequal variances (heteroscedasticity; Eq. 4b), and

484the third level is an independent Gaussian exposure model for the true but unobserved

485predictor F_{ST} (Eq. 4c):

486

$$\mathbf{log}(\boldsymbol{r_0}) = \beta_0\mathbf{1} + \beta_F\,\boldsymbol{F_{true}} + \boldsymbol{z'}\,\boldsymbol{\beta_z} + \boldsymbol{\delta}_y + \boldsymbol{\varepsilon}_y\,, \qquad \boldsymbol{\varepsilon}_y \sim N(\mathbf{0},\,\sigma^2_y\mathbf{I})\,, \qquad\qquad (4a)$$

$$\boldsymbol{\delta}_y \sim N(\mathbf{0},\,\mathbf{D}_y)$$
$$\boldsymbol{F_{ST}} \;\;= \boldsymbol{F_{true}} + \boldsymbol{u}\,, \qquad\qquad \boldsymbol{u} \sim N(\mathbf{0},\,\mathbf{D}_u) \qquad\qquad (4b)$$
$$\boldsymbol{F_{true}} \;\;= \boldsymbol{\mu_0} + \boldsymbol{\varepsilon}_{Ftrue}\,, \qquad\qquad \boldsymbol{\varepsilon}_{Ftrue} \sim N(\mathbf{0},\,\sigma^2_{Ftrue}\mathbf{I}) \qquad\qquad (4c)$$

487

488Bold-face notation indicates vectors or matrices. The vector $\boldsymbol{F_{true}}$ denotes the correct

489but unobserved inbreeding values, $\boldsymbol{z'}$ is the transposed of the matrix with rows

490containing the additional covariates, and β_0, β_F and $\boldsymbol{\beta}_z$ represent the intercept, slope

491coefficient of $\boldsymbol{F_{true}}$, and the vector of slope coefficients of $\boldsymbol{z}$, respectively. The random

492term δ_y accounts for the error in the observed $\log(r_0)$ values. $\boldsymbol{F_{ST}}$ is the vector of the

493estimated levels of inbreeding, and $\boldsymbol{u}$ is the error vector from the measurement error

494model. Classical covariate measurement error models require assigning a distribution

495to the predictor variable that is measured with error[22], and we selected a Gaussian

496distribution (Eq. 4c) with mean $\boldsymbol{\mu_0}=\boldsymbol{0}$ to match the observed (centred) distribution of

497F_{ST} scores (Fig. 2).

498   The variances σ^2_y and σ^2_{Ftrue} are the residual variance of the regression and the

499variance of $\boldsymbol{F_{true}}$, respectively. The entries in the diagonal matrices $\mathbf{D}_y$ and $\mathbf{D}_u$ account

500for population-specific (heteroscedastic) uncertainties in the regression and error

501model, respectively: the entries in the former are equal to the estimated error

502variances $\hat{\sigma}^2_{\log(r_0)}$, while the entries in the latter are equal to the estimated error

503variances $\hat{\sigma}^2_{F_{ST}}$ for the individual populations, and thus these properly account for

504uncertainty in the $\log(r_0)$ and F_{ST} estimates in each population, respectively. Matrix $\mathbf{I}$

505is the identity matrix of appropriate dimension.

506   It is straightforward to incorporate prior knowledge into such a Bayesian

507hierarchical model, and in particular prior uncertainty given by the variance estimates.

508To estimate the posterior marginal distributions, we used a fast and accurate

509alternative to MCMC sampling, namely integrated nested Laplace approximations

510(INLA)[49]. INLA is suitable for inference on latent Gaussian models, which are a

511subset of hierarchical models and compatible with our model[23].

512   We closely followed the procedure as described in Muff *et al.*[23] to assign

513priors according to expert/prior knowledge. We used independent $N(0,10^4)$ priors for

514all β-coefficients, and inverse Gamma distributions for the variances: $\sigma^2_y \sim IG(1,0.02)$

515and $\sigma^2_{Ftrue} \sim IG(1.9,0.001)$. The σ^2_y prior differed from Muff *et al.*[23] because the log-

516transformed version of the response variable was used. Finally, the $\mathbf{D}_y$ and $\mathbf{D}_u$ were

517assumed known and are thus fixed.

518

Regression analysis: Model selection and parameter estimates. Model selection was guided by minimization of the deviance information criterion DIC[50], where the main effect of interest, β_F, was always retained in the model. To illustrate the bias that would result if the uncertainty in the point estimates of population-specific F_{ST} were ignored, we also fitted the model with lowest DIC (termed "model 1") with a standard least squares approach using weighted regression with mean standardized weights proportional to $1/\hat{\sigma}^2_{\log(r_0)}$, but ignoring covariate error in inbreeding values (Supplementary Table 5, model 1(ML)). Further, we also retained a model that was identical to model 1, but that did not contain the interaction term $F_{ST} \cdot$ ps (Supplementary Table 5, model 2). Here, too, we additionally fitted the model using weighted regression [model 2 (ML)]. All analyses were performed using R version 3.3.2[41]. The hierarchical model (Eqs. 4a-c) was fitted with INLA, using the R-interface R-INLA (version built on June 20, 2017), which can be downloaded from www.r-inla.org.

533

534

Data and code availability

Data that support the findings of this study have been deposited in Dryad Digital Repository with the identifier (doi:xxx)[51].

538

539

References

541

542

543 1. Hedrick, P. W. & Garcia-Dorado, A. Understanding inbreeding depression, purging, and genetic

544   rescue. *Trends Ecol. Evol.* **31,** 940–952 (2016).

545 2. Wallace, B. Hard and soft selection revisited. *Evolution* **29,** 465–473 (1975).

546 3. Agrawal, A. F. & Whitlock, M. C. Mutation load: The fitness of individuals in populations where

547   deleterious alleles are abundant. *Annu. Rev. Ecol. Evol. Syst. Vol 43* **43,** 115–135 (2012).

548 4. Johnson, H. E., Mills, L. S., Wehausen, J. D., Stephenson, T. R. & Luikart, G. Translating effects of

549   inbreeding depression on component vital rates to overall population growth in endangered bighorn

550   sheep. *Conserv. Biol.* **25,** 1240–1249 (2011).

551 5. Reed, T. E., Grotan, V., Jenouvrier, S., Saether, B.-E. & Visser, M. E. Population growth in a wild

552   bird is buffered against phenological mismatch. *Science* **340,** 488–491 (2013).

553 6. Agrawal, A. F. & Whitlock, M. C. Environmental duress and epistasis: how does stress affect the

554   strength of selection on new mutations? *Trends Ecol. Evol.* **25,** 450–458 (2010).

555 7. Creel, S. Recovery of the Florida panther - genetic rescue, demographic rescue, or both? Response

556   to Pimm et al. (2006). *Anim. Conserv.* **9,** 125–126 (2006).

557 8. Wootton, J. T. & Pfister, C. A. Processes affecting extinction risk in the laboratory and in nature.

558   *Proc. Natl. Acad. Sci. U. S. A.* **112,** E5903–E5903 (2015).

559 9. Hufbauer, R. A. *et al.* Reply to Wootton and Pfister: The search for general context should include

560   synthesis with laboratory model systems. *Proc. Natl. Acad. Sci. U. S. A.* **112,** E5904–E5904 (2015).

561 10. Kardos, M., Taylor, H. R., Ellegren, H., Luikart, G. & Allendorf, F. W. Genomics advances the

562   study of inbreeding depression in the wild. *Evol. Appl.* **9,** 1205–1218 (2016).

563 11. Stuwe, M. & Nievergelt, B. Recovery of alpine ibex from near extinction: the result of effective

564   protection, captive breeding, and reintroductions. *Appl. Anim. Behav. Sci.* **29,** 379–387 (1991).

565 12. Grotan, V., Saether, B. E., Filli, F. & Engen, S. Effects of climate on population fluctuations of

566   ibex. *Glob. Change Biol.* **14,** 218–228 (2008).

567 13. Biebach, I. & Keller, L. F. A strong genetic footprint of the re-introduction history of Alpine ibex

568   (*Capra ibex ibex*). *Mol. Ecol.* **18,** 5046–5058 (2009).

569 14. Biebach, I. & Keller, L. F. Genetic variation depends more on admixture than number of founders

570   in reintroduced Alpine ibex populations. *Biol. Conserv.* **147,** 197–203 (2012).

571 15. Biebach, I. & Keller, L. F. Inbreeding in reintroduced populations: the effects of early

572   reintroduction history and contemporary processes. *Conserv. Genet.* **11,** 527–538 (2010).

573 16. Crow, J. F. Breeding structure of populations. II. Effective population number. in *Statistics and*

574 *Mathematics in Biology* (eds. Kempthome, O., Bancroft, T. A., Gowen, J. W. & Lush, J. L.) (Iowa

575 State University Press, 1954).

576 17. Jacquard, A. (ed) *The genetic structure of populations*. (Springer Verlag, 1974).

577 18. Vitalis, R., Dawson, K. & Boursot, P. Interpretation of variation across marker loci as evidence of

578 selection. *Genetics* **158,** 1811–1823 (2001).

579 19. Weir, B. S. & Goudet, J. A Unified Characterization of Population Structure and Relatedness.

580 *Genetics* **206,** 2085–2103 (2017).

581 20. Holsinger, K. E. & Weir, B. S. Genetics in geographically structured populations: defining,

582 estimating and interpreting F-ST. *Nat. Rev. Genet.* **10,** 639–650 (2009).

583 21. Vitalis, R. & Couvet, D. Estimation of effective population size and migration rate from one- and

584 two-locus identity measures. *Genetics* **157,** 911–925 (2001).

585 22. Fuller, W. A. *Measurement error models*. (John Wiley & Sons, 1987).

586 23. Muff, S., Riebler, A., Held, L., Rue, H. & Saner, P. Bayesian analysis of measurement error models

587 using integrated nested Laplace approximations. *J. R. Stat. Soc. Ser. C-Appl. Stat.* **64,** 231–252

588 (2015).

589 24. Yun, L. & Agrawal, A. F. Variation in the strength of inbreeding depression across environments:

590 Effects of stress and density dependence. *Evolution* **68,** 3599–3606 (2014).

591 25. Gilpin, M. E. & Soulé, M. E. Minimum viable populations: processes of extinction. in *Conservation*

592 *biology, the science of scarcity and diversity* (ed. Soulé, M. E.) 19–34 (Sinauer Assiocates, 1986).

593 26. Wright, S. Size of population and breeding structure in relation to evolution. *Science* **87,** 430–431

594 (1938).

595 27. Beaumont, M. A. Estimation of population growth or decline in genetically monitored populations.

596 *Genetics* **164,** 1139–1160 (2003).

597 28. Facon, B. *et al.* Inbreeding depression is purged in the invasive insect *harmonia axyridis*. *Curr.*

598 *Biol.* **21,** 424–427 (2011).

599 29. Garcia-Dorado, A. Understanding and Predicting the Fitness Decline of Shrunk Populations:

600 Inbreeding, Purging, Mutation, and Standard Selection. *Genetics* **190,** 1461–1476 (2012).

601 30. Glemin, S. How are deleterious mutations purged? Drift versus nonrandom mating. *Evolution* **57,**

602 2678–2687 (2003).

603 31. Kirkpatrick, M. & Jarne, P. The effects of a bottleneck on inbreeding depression and the genetic
604   load. *Am. Nat.* **155,** 154–167 (2000).

605 32. Whiteley, A. R., Fitzpatrick, S. W., Funk, W. C. & Tallmon, D. A. Genetic rescue to the rescue.
606   *Trends Ecol. Evol.* **30,** 42–49 (2015).

607 33. Saccheri, I. *et al.* Inbreeding and extinction in a butterfly metapopulation. *Nature* **392,** 491–494
608   (1998).

609 34. O'Grady, J. J. *et al.* Realistic levels of inbreeding depression strongly affect extinction risk in wild
610   populations. *Biol. Conserv.* **133,** 42–51 (2006).

611 35. Goldberg, E. E. *et al.* Species selection maintains self-incompatibility. *Science* **330,** 493–495
612   (2010).

613 36. Nievergelt, B. *Der Alpensteinbock (Capra Ibex L.) in seinem Lebensraum*. (Verlag Paul Parey,
614   Berlin, Germany, 1966).

615 37. Abderhalden, W. D. *Raumnutzung und sexuelle Segregation*. (Albert-Ludwigs-Universität Freiburg
616   i. Brsg., 2004).

617 38. Saether, B. E., Lillegard, M., Grotan, V., Filli, F. & Engen, S. Predicting fluctuations of
618   reintroduced ibex populations: the importance of density dependence, environmental stochasticity
619   and uncertain population estimates. *J. Anim. Ecol.* **76,** 326–336 (2007).

620 39. Jaquiery, J., Guillaume, F. & Perrin, N. Predicting the deleterious effects of mutation load in
621   fragmented populations. *Conserv. Biol.* **23,** 207–218 (2009).

622 40. Ciofi, C., Beaumont, M. A., Swingland, I. R. & Bruford, M. W. Genetic divergence and units for
623   conservation in the Komodo dragon *Varanus komodoensis. Proc. R. Soc. Lond. Ser. B-Biol. Sci.*
624   **266,** 2269–2274 (1999).

625 41. R Development Core Team R: *A Language and Environment for Statistical Computing* (R
626   Foundation for Statistical Computing, Vienna, 2016).

627 42. Aeschbacher, A. *Das Brunftverhalten des Alpensteinwildes*. (Rentsch, 1978).

628 43. Harvey, A. C. *Forecasting, structural time series models and the Kalman filter*. (Cambridge
629   University Press, 1989).

630 44. Schooler, S. S., Salau, B., Julien, M. H. & Ives, A. R. Alternative stable states explain unpredictable
631   biological control of Salvinia molesta in Kakadu. *Nature* **470,** 86–89 (2011).

632 45. Dennis, B., Ponciano, J. M., Lele, S. R., Taper, M. L. & Staples, D. F. Estimating density

633 dependence, process noise, and observation error. *Ecol. Monogr.* **76,** 323–341 (2006).

634 46. MathWorks. *MATLAB*. (The MathWorks Inc., 2012).

635 47. Kendall, M. & Stuart, A. *The advanced theory of statistics*. **2,** (Griffin, 1979).

636 48. Carroll, R., Ruppert, D., Stefanski, L. & Crainiceanu, C. *Measurement error in nonlinear models: a*

637 *modern perspective*. (Chapman & Hall, 2006).

638 49. Rue, H., Martino, S. & Chopin, N. Approximate Bayesian inference for latent Gaussian models by

639 using integrated nested Laplace approximations. *J. R. Stat. Soc. Ser. B-Stat. Methodol.* **71,** 319–392

640 (2009).

641 50. Spiegelhalter, D. J., Best, N. G., Carlin, B. R. & van der Linde, A. Bayesian measures of model

642 complexity and fit. *J. R. Stat. Soc. Ser. B-Stat. Methodol.* **64,** 583–616 (2002).

643 51. Bozzuto, C., Biebach, I., Muff, S., Ives, A.R. & Keller, L.F., Inbreeding reduces long-term growth

644 of Alpine ibex populations. *Dryad Digital Repository.* doi:xxx

645

646