

File S3: The role of recombination

From now on, we assume that the population is large enough that we can approximate the number of Ab and aB mutants in the standing genetic variation by their expected number, Eq. (S1.14). For the number of double mutants prior to the environmental change, we use F_{AB} , Eq. (S1.17). In order to keep the equations simple, we usually assume $\sigma_{Ab} = \sigma_{aB} = \sigma$. Generalization to unequal selection coefficients for single mutants before the environmental change is straightforward.

S3.1 Single mutants are lethal in the new environment

The wildtype is lethal too. In the absence of any other types, a single rescue type individual establishes a permanent lineage with probability $p_{\text{est}}^{(AB)}$, Eq. (S1.20). In the first generation after the switch, with our choice of the life cycle (mutation and recombination before selection), the wildtype and the single mutants are, however, still present in the population (leading to the generation and deletion of AB mutants). A single rescue type individual present at the time of environmental change will hence not establish a permanent lineage with probability $\exp[-p_{\text{est}}^{(AB)}(1+s_{AB})(1-r)]$, and the probability that no new successful lineage is generated by recombination or mutation in this first generation is given by $\exp\left[-\left(r\frac{\bar{n}_{Ab}\bar{n}_{aB}}{N}(1+s_{AB}) + u(\bar{n}_{Ab} + \bar{n}_{aB})(1+s_{AB})(1-r)\right)p_{\text{est}}^{(AB)}\right]$. With this, the probability of evolutionary rescue is given by

$$P_{\text{rescue}} = 1 - F_{AB}(e^{-(1+s_{AB})(1-r)}p_{\text{est}}^{(AB)}) \times e^{-\left(r\frac{\bar{n}_{Ab}\bar{n}_{aB}}{N_0}(1+s_{AB}) + u(\bar{n}_{Ab} + \bar{n}_{aB})(1+s_{AB})(1-r)\right)p_{\text{est}}^{(AB)}}. \quad (\text{S3.1})$$

With $p_{\text{est}}^{(AB)} \approx 2s_{AB}$ and $\sigma_{Ab} = \sigma_{aB} = \sigma$, we can approximate

$$\begin{aligned} F(e^{-(1+s_{AB})(1-r)}p_{\text{est}}^{(AB)}) &\approx F(1 - 2s_{AB}(1-r)) \\ &= \left(1 + \frac{2s_{AB}(1+\sigma_{AB})(1-r)^2}{2(1-r)(1+\sigma_{AB}) - 2s_{AB}(1-r)^2(1+\sigma_{AB}) - 2}\right)^{\frac{2(1+\sigma_{AB})\left[r\frac{u^2N_0}{\sigma^2}(1+\sigma)^2 + \frac{u^2N_0}{-\sigma}(1+\sigma)(1-r)\right]}{(1+\sigma_{AB})(1-r)}} \\ &= \left(1 + \frac{s_{AB}(1-r)^2}{-s_{AB}(1-r)^2 + (1-r)\sigma_{AB} - 1}\right)^{-\frac{2u^2N_0}{\sigma^2}\left[2\sigma - \frac{r}{1-r}\right]} \\ &\approx \left(\frac{r - (2\sigma + E_1)(1-r)}{s_{AB}(1-r)^2 + r - (2\sigma + E_1)(1-r)}\right)^{-\frac{2u^2N_0}{\sigma^2}\left[2\sigma - \frac{r}{1-r}\right]} \\ &\approx \left(\frac{r - 2\sigma - E_1}{s_{AB}(1-r)^2 + r - 2\sigma - E_1}\right)^{-\frac{2u^2N_0}{\sigma^2}\left[2\sigma - \frac{r}{1-r}\right]}, \end{aligned} \quad (\text{S3.2})$$

where the first approximation is a series expansion of the exponential function up to first order in the exponent and the second approximation is based on dropping higher order terms in σ_{AB} and σ in the numerator, the denominator, and the exponent. The approximation in the last line consists in approximating $r - (1 - r)(2\sigma + E_1) \approx r - 2\sigma - E_1$ since the second term only matters when r is small, i.e. when $1 - r \approx 1$. If we furthermore ignore new mutations after the switch in the environment, we obtain:

$$P_{\text{rescue}} \approx 1 - \left(\frac{r - 2\sigma - E_1}{s_{AB}(1 - r)^2 + r - 2\sigma - E_1} \right)^{-\frac{2u^2 N_0}{\sigma^2} [2\sigma - \frac{r}{1-r}]} e^{-2s_{AB}r \frac{u^2 N_0}{\sigma^2}}. \quad (\text{S3.3})$$

If we do not take stochasticity in the number of double mutants in the standing genetic variation into account, we get

$$\begin{aligned} P_{\text{rescue}}^{\text{det}} &= 1 - e^{-(n_{AB})(1+s_{AB})(1-r)} p_{\text{est}}^{(AB)} \times e^{-\left(r \frac{\bar{n}_{Ab}\bar{n}_{aB}}{N} (1+s_{AB}) + u(\bar{n}_{Ab} + \bar{n}_{aB})(1+s_{AB})(1-r)\right) p_{\text{est}}^{(AB)}} \\ &\approx 1 - e^{-2 \frac{u^2 N_0}{\sigma^2} s_{AB} \left[1 - \frac{(1-r)(2-2\sigma-E_1)}{r-2\sigma-E_1}\right]} \\ &\approx 1 - e^{-2s_{AB} \frac{u^2 N_0}{\sigma^2} \frac{r-2\sigma}{r-2\sigma-E_1}}, \end{aligned} \quad (\text{S3.4})$$

where the first approximation makes use of the approximation for $\langle n_{AB} \rangle$ (Eq. S1.19) and furthermore uses $p_{\text{est}}^{(AB)} \approx 2s_{AB}$ and $1 + s_{AB} \approx 1$ and ignores new mutations from generation 0 to 1.

With this, we can compare the probability of evolutionary rescue (1) without epistasis and without drift (Eq. S3.4 with $E_1 = 0$), (2) without epistasis but with drift (Eq. S3.1 with $E_1 = 0$), (3) with epistasis but without drift (Eq. S3.4 with $E_1 \neq 0$), and (4) with epistasis and with drift (Eq. S3.1 with $E_1 \neq 0$). Fig. S3.1 shows all four cases. Note that the establishment of the rescue type after the environmental change is in any case subject to strong stochasticity.

Last, we want to estimate the influence of drift on the rescue probability

$$d = \frac{P_{\text{rescue}} - P_{\text{rescue}}^{\text{det}}}{P_{\text{rescue}}^{\text{det}}}. \quad (\text{S3.5})$$

For this, we approximate by a Taylor expansion up to leading order in s_{AB} (and similar approximations as in Eq. S3.4):

$$\begin{aligned} P_{\text{rescue}} - P_{\text{rescue}}^{\text{det}} &\approx \left(e^{-2s_{AB}\langle n_{AB} \rangle(1-r)} - \langle e^{-2s_{AB}n_{AB}(1-r)} \rangle \right) e^{-2s_{AB}r \frac{\bar{n}_{Ab}\bar{n}_{aB}}{N_0}} \\ &\approx -2s_{AB}^2(1-r)^2 \text{Var}[n_{AB}] + \mathcal{O}(s_{AB}^3). \end{aligned} \quad (\text{S3.6})$$

This leaves us with

$$\begin{aligned}
d &\approx -\frac{s_{AB}(1-r)^2 \text{Var}[n_{AB}]}{(1-r)\langle n_{AB} \rangle + r \frac{\bar{n}_{Ab}\bar{n}_{aB}}{N_0}} + \mathcal{O}(s_{AB}^2) \\
&= -\frac{-s_{AB}(1-r)^2 \frac{\text{Var}[n_{AB}]}{\langle n_{AB} \rangle}}{(1-r) + r \frac{\bar{n}_{Ab}\bar{n}_{aB}}{N_0 \langle n_{AB} \rangle}} + \mathcal{O}(s_{AB}^2) \\
&\approx \frac{\text{Var}[n_{AB}]}{\langle n_{AB} \rangle} \cdot \frac{-s_{AB}(1-r)^2}{1 + r \frac{E_1}{r-2\sigma}} + \mathcal{O}(s_{AB}^2).
\end{aligned} \tag{S3.7}$$

For the last line, we used Eq. (S1.19) and $\bar{n}_{Ab} = \bar{n}_{aB} \approx -\frac{uN_0}{\sigma}$. For the ratio of variance to mean, we obtain:

$$\begin{aligned}
\frac{\text{Var}[n_{AB}]}{\langle n_{AB} \rangle} &= \frac{F''_{AB}(1) + F'_{AB}(1) - F'_{AB}(1)^2}{F'_{AB}(1)} \\
&= \frac{1}{2} \left(1 + \frac{1}{r(1 + \sigma_{AB}) - \sigma_{AB}} \right),
\end{aligned} \tag{S3.8}$$

which is a decreasing function of r , i.e., the relative importance of drift decreases with r . Note that the variance itself depends on epistasis and is not decreasing over the entire parameter range (it can be increasing, decreasing, or be non-monotonic).

For $|\sigma|$ and $|\sigma_{AB}|$ small, we can further approximate

$$d \approx -\frac{\text{Var}[n_{AB}]}{\langle n_{AB} \rangle} s_{AB}(1-r)^2 \approx -\frac{1}{2}(1-r)^2(1+r) \frac{s_{AB}}{r - \sigma_{AB}}. \tag{S3.9}$$

Although the approximation deviates from the exact result for small r , we can read off the qualitative behavior: d is negative and monotonically increasing with r , i.e., the larger r , the less drift reduces P_{rescue} . We can distinguish two regimes: (1) If $|\sigma_{AB}| \gg s_{AB}$, drift does not play a significant role, irrespective of r . (2) If $|\sigma_{AB}| \ll s_{AB}$, drift has a significant influence unless $r \gg s_{AB}$.

The wildtype remains. If the wildtype population size decays slowly after the environmental change, the establishment probability of a single rescue mutant is well approximated by Eq. (S1.23). Analogous to before, we again have

$$P_{\text{rescue}} = 1 - F_{AB}(e^{-(1+s_{AB})(1-r)} p_{\text{est}}^{(AB)}) \times e^{-\left(r \frac{\bar{n}_{Ab}\bar{n}_{aB}}{N_0} (1+s_{AB}) + u(\bar{n}_{Ab} + \bar{n}_{aB})(1+s_{AB})(1-r)\right) p_{\text{est}}^{(AB)}}. \tag{S3.10}$$

Actually, $e^{-(1+s_{AB})(1-r)(1-q_{AB})} = q_{AB}$ (where q_{AB} is the exact extinction probability of a branching process with Poisson distributed offspring numbers with mean $(1+s_{AB})(1-r)$), and so we could simply use $F_{AB}(1 - p_{\text{est}}^{(AB)})$. Since we use an approximation for q_{AB} (which is our approximation $1 - p_{\text{est}}^{(AB)}$), we prefer the above form for consistency with the previous paragraph.

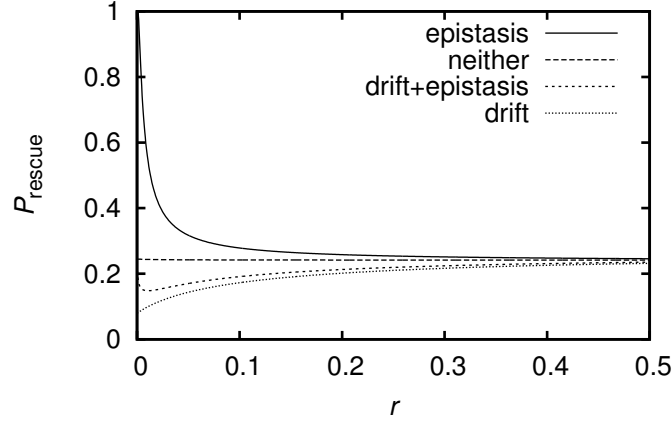


Fig. S3.1: Probability of evolutionary rescue as a function of recombination (cf. Fig. 1).

The curves are based on Eq. (S3.1) (drift) and Eq. (S3.4) (no drift). Parameter values are: $\sigma_{AB} = -0.0199$ (no epistasis) and $\sigma_{AB} = -0.0001$ (epistasis), $\sigma_{Ab} = \sigma_{aB} = -0.01$, $u = 10^{-5}$, $N_0 = 10^6$, $s_{AB} = 0.15$, $s_{Ab} = s_{aB} = s_{ab} = -1$.

As before, we can derive an approximation, ignoring stochasticity in the number of double mutants

$$\begin{aligned}
 P_{\text{rescue}}^{\text{det}} &= 1 - e^{-(n_{AB})(1+s_{AB})(1-r)p_{\text{est}}^{(AB)}} \times e^{-\left(r\frac{\bar{n}_{Ab}\bar{n}_{aB}}{N}(1+s_{AB})+u(\bar{n}_{Ab}+\bar{n}_{aB})(1+s_{AB})(1-r)\right)p_{\text{est}}^{(AB)}} \\
 &\approx \begin{cases} 1 - e^{-2\frac{u^2N_0}{\sigma^2}(s_{AB}-r)\left[1-\frac{(2-2\sigma-E_1)(1-r)}{r-2\sigma-E_1}\right]} \approx 1 - e^{-2(s_{AB}-r)\frac{u^2N_0}{\sigma^2}\frac{r-2\sigma}{r-2\sigma-E_1}} & \text{if } s_{AB} - r > 0, \\ 0 & \text{else,} \end{cases} \quad (\text{S3.11})
 \end{aligned}$$

where we approximate $p_{\text{est}}^{(AB)} \approx \max[2(s_{AB} - r), 0]$.

The wildtype is quite unfit. If the wildtype is not very fit, we need to resort to the more accurate approximation Eq. (S1.29) for the establishment probability of the double mutant. For the probability of rescue, we obtain as before:

$$P_{\text{rescue}} = 1 - F_{AB}\left(e^{-(1+s_{AB})(1-r)p_{\text{est}}^{(AB)}(1)}\right) \times e^{-\left(r\frac{\bar{n}_{Ab}\bar{n}_{aB}}{N_0}(1+s_{AB})+u(n_{Ab}+n_{aB})(1+s_{AB})(1-r)\right)p_{\text{est}}^{(AB)}(1)}. \quad (\text{S3.12})$$

Sensitivity of the approximation. How sensitive are the approximations to the assumption of lethality of the single mutants? Fig. S3.2 compares the approximations (assuming $s_{Ab} = s_{aB} = -1$) to simulations with $s_{Ab} = s_{aB} = -0.99$ (Panel A) and $s_{Ab} = s_{aB} = -0.9$ (Panel B). The fitter the wildtype the less sensitive is the approximation to deviations from strict lethality of the single mutants. For a lethal wildtype, even a slight increase in the fitness of mutants above lethality drastically increases P_{rescue} .

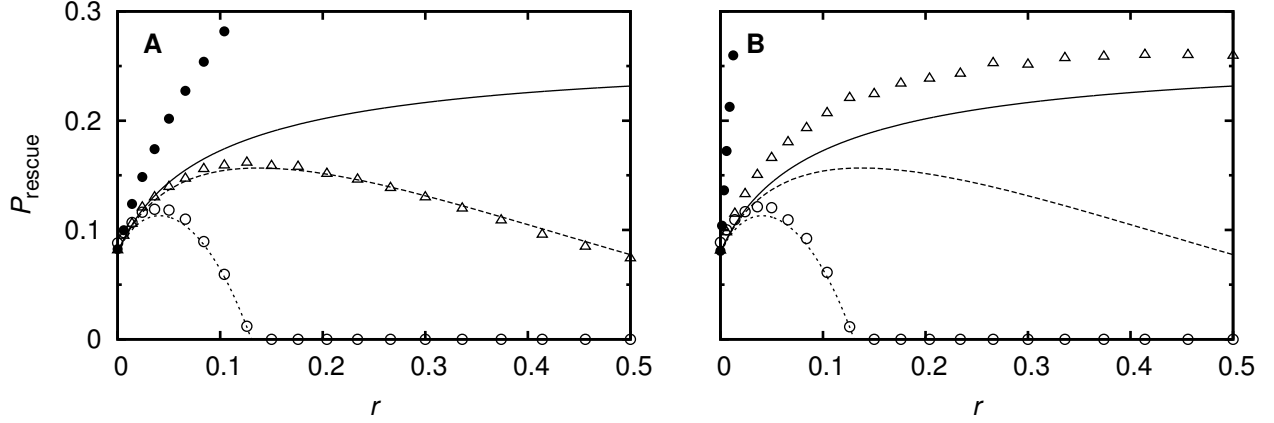


Fig. S3.2: Probability of evolutionary rescue as a function of recombination. The figure is identical to Fig. 1 except for that we set $s_{Ab} = s_{aB} = -0.99$ (Panel A) and $s_{Ab} = s_{aB} = -0.9$ (Panel B) in the simulations. The growth parameter of the wildtype is $s_{ab} = -1$ (solid lines, filled circle), $s_{ab} = -0.99$ (dashed line, triangles), $s_{ab} = -0.005$ (dotted line, empty circles). Circles and triangles denote simulation results. Each simulation point is the average of 10^5 replicates.

S3.2 One single mutant is viable, the other lethal

Let us now consider the situation $s_{Ab} > -1$ and $s_{aB} = -1$ after the environmental change.

The wildtype is lethal. The presence of one of the single mutant types after the environmental change opens up a new rescue pathway: new double mutants can be generated by mutation after generation 0. Analogous to before, the probability that the population is rescued via this pathway is given by

$$1 - e^{-(\bar{n}_{Ab} + uN_0)(1+s_{Ab})p_{\text{est}}^{(Ab)}} \quad (\text{S3.13})$$

with $p_{\text{est}}^{(Ab)}$ given by Eq. (S1.21). Combination with Eq. (S3.1) yields the total probability of evolutionary rescue:

$$P_{\text{rescue}} = 1 - F_{AB} \left(e^{-(1+s_{AB})(1-r)p_{\text{est}}^{(AB)}} \right) \times e^{-\left(r \frac{\bar{n}_{Ab}\bar{n}_{aB}}{N_0} (1+s_{AB}) + u(\bar{n}_{Ab} + \bar{n}_{aB})(1+s_{AB})(1-r) \right) p_{\text{est}}^{(AB)}} \times e^{-(\bar{n}_{Ab} + uN_0)(1+s_{Ab})p_{\text{est}}^{(Ab)}}. \quad (\text{S3.14})$$

We can estimate the respective significance of the contributions by a comparison of Eq. (S3.13) with Eq. (S3.4), assuming $\sigma_{Ab} = \sigma_{aB}$. Approximating $\bar{n}_{Ab} \approx \frac{uN_0}{-\sigma_{Ab}}$ and $1 + s_{Ab} \approx 1$ and ignoring the term that accounts for new mutations ($\sim uN_0$) in Eq. (S3.13) and setting $E_1 = 0$ in Eq. (S3.4), we arrive at the condition

$$p_{\text{est}}^{(Ab)} > 2 \frac{uS_{AB}}{-\sigma_{Ab}} \quad (\text{S3.15})$$

for the contribution of new rescue mutations after the environmental change being larger than the contribution by double mutants from the standing genetic variation. With the last approximation for $p_{\text{est}}^{(Ab)}$ in Eq. (S1.21), this condition simplifies to

$$\frac{2uS_{Ab}}{-s_{Ab}} > \frac{2uS_{Ab}}{-\sigma_{Ab}} \Leftrightarrow -\sigma_{Ab} > -s_{Ab}. \quad (\text{S3.16})$$

If $s_{Ab} > 0$, rescue is not contingent on the generation of the double mutant. Depending on the mutation rate and the fitness effects of mutations, generation of the double mutant might still help rescue or be negligible. In the latter case, results from single step rescue apply (Orr and Unckless, 2008; Bell and Collins, 2008; Uecker *et al.*, 2014). Formation of the double mutant after the environmental change cannot be ignored in Eq. (S3.13) if

$$\begin{aligned} 2s_{Ab} &\ll p_{\text{est}}^{(Ab)} \\ \Leftrightarrow 2s_{Ab} &\ll s_{Ab} - u + \sqrt{(s_{Ab} - u)^2 + 4s_{AB}u} \\ \Leftrightarrow s_{Ab} + u &\ll \sqrt{(s_{Ab} - u)^2 + 4s_{AB}u} \\ \stackrel{s_{Ab} \gg u}{\Leftrightarrow} s_{Ab} &\ll \sqrt{s_{Ab}^2 + 4s_{AB}u} = s_{Ab} \cdot \sqrt{1 + \frac{4s_{AB}u}{s_{Ab}^2}} \\ \Leftrightarrow 4s_{AB}u &\gg s_{Ab}^2. \end{aligned} \quad (\text{S3.17})$$

Altogether, generation of the double mutant cannot be ignored if

$$\begin{aligned} 2s_{Ab} \frac{uN_0}{-\sigma} &\ll p_{\text{est}}^{(Ab)} \frac{uN_0}{-\sigma} + 2s_{AB} \frac{u^2N_0}{\sigma^2} \frac{r-2\sigma}{r-2\sigma-E_1} \\ \Leftrightarrow 2s_{Ab} &\ll s_{Ab} - u + \sqrt{(s_{Ab} - u)^2 + 4s_{AB}u} + 2s_{AB} \frac{u^2N_0}{\sigma^2} \frac{r-2\sigma}{r-2\sigma-E_1} \\ \Leftrightarrow s_{Ab} + u &\ll \sqrt{(s_{Ab} - u)^2 + 4s_{AB}u} + 2s_{AB} \frac{u^2N_0}{\sigma^2} \frac{r-2\sigma}{r-2\sigma-E_1} \\ \stackrel{s_{Ab} \gg u}{\Leftrightarrow} s_{Ab} &\ll s_{Ab} \cdot \sqrt{1 + \frac{4s_{AB}u}{s_{Ab}^2}} + 2s_{AB} \frac{u^2N_0}{\sigma^2} \frac{r-2\sigma}{r-2\sigma-E_1} \\ \Leftrightarrow 4s_{AB}u &\gg s_{Ab}^2 \quad \text{or} \quad 2s_{AB} \frac{u^2N_0}{\sigma^2} \frac{r-2\sigma}{r-2\sigma-E_1} \gg s_{Ab}. \end{aligned} \quad (\text{S3.18})$$

The wildtype is at least as fit as the viable single mutant. Viability of the wildtype has two consequences: (1) The double mutant can be broken up by recombination. (2) The wildtype can generate new *Ab* mutants on its course to extinction. Modeling the wildtype deterministically, we obtain for the probability of rescue by de-novo generated double mutants

$$1 - e^{-\bar{n}_{Ab}(1+s_{Ab})p_{\text{est}}^{(Ab)}} \times e^{-\frac{uN_0}{-s_{Ab}}(1+s_{Ab})p_{\text{est}}^{(Ab)}}. \quad (\text{S3.19})$$

Combination with Eq. (S3.10) yields again the total probability of evolutionary rescue:

$$P_{\text{rescue}} = 1 - F_{AB} \left(e^{-(1+s_{AB})(1-r)} p_{\text{est}}^{(AB)} \right) \times e^{-\left(r \frac{\bar{n}_{Ab} \bar{n}_{aB}}{N_0} (1+s_{AB}) + u (\bar{n}_{Ab} + \bar{n}_{aB}) (1+s_{AB})(1-r) \right) p_{\text{est}}^{(AB)}} \times e^{-\left(\bar{n}_{Ab} + \frac{u N_0}{s_{Ab}} \right) (1+s_{Ab}) p_{\text{est}}^{(Ab)}}. \quad (\text{S3.20})$$

As before, we can compare the different pathways to rescue, (a) from double mutants from the standing genetic variation, (b) mutation of single mutants from the standing genetic variation after the change in the environment, (c) complete de-novo generation via the wildtype after the environmental switch. Pathway (c) is more important than pathway (b) if

$$-s_{ab} < -\sigma_{Ab}. \quad (\text{S3.21})$$

Pathway (b) is more important than pathway (a) if

$$-s_{Ab} < -\sigma_{Ab}. \quad (\text{S3.22})$$

If $s_{Ab} > 0$, analogous to the previous paragraph, formation of the double mutant after the environmental change cannot be ignored if

$$\begin{aligned} 2s_{Ab} &\ll p_{\text{est}}^{(Ab)} \\ \Leftrightarrow 2s_{Ab} &\ll s_{Ab} - u + \sqrt{(s_{Ab} - u)^2 + 4 \max[(s_{AB} - r), 0]u} \\ \Leftrightarrow s_{Ab} + u &\ll \sqrt{(s_{Ab} - u)^2 + 4 \max[(s_{AB} - r), 0]u} \\ \stackrel{s_{Ab} \gg u}{\Leftrightarrow} s_{Ab} &\ll \sqrt{s_{Ab}^2 + 4s_{AB}u} = s_{Ab} \cdot \sqrt{1 + \frac{4 \max[(s_{AB} - r), 0]u}{s_{Ab}^2}} \\ \Leftrightarrow 4 \max[(s_{AB} - r), 0]u &\gg s_{Ab}^2. \end{aligned} \quad (\text{S3.23})$$

Altogether, it cannot be ignored if

$$\begin{aligned} 2s_{Ab} \frac{u N_0}{-\sigma} &\ll p_{\text{est}}^{(Ab)} \frac{u N_0}{-\sigma} + \max[2(s_{AB} - r), 0] \frac{u^2 N_0}{\sigma^2} \frac{r - 2\sigma}{r - 2\sigma - E_1} \\ \Leftrightarrow 2s_{Ab} &\ll s_{Ab} - u + \sqrt{(s_{Ab} - u)^2 + 4 \max[(s_{AB} - r), 0]u} + \max[2(s_{AB} - r), 0] \frac{u^2 N_0}{\sigma^2} \frac{r - 2\sigma}{r - 2\sigma - E_1} \\ \Leftrightarrow s_{Ab} + u &\ll \sqrt{(s_{Ab} - u)^2 + 4 \max[(s_{AB} - r), 0]u} + \max[2(s_{AB} - r), 0] \frac{u^2 N_0}{\sigma^2} \frac{r - 2\sigma}{r - 2\sigma - E_1} \\ \stackrel{s_{Ab} \gg u}{\Leftrightarrow} s_{Ab} &\ll s_{Ab} \cdot \sqrt{1 + \frac{4 \max[(s_{AB} - r), 0]u}{s_{Ab}^2}} + \max[2(s_{AB} - r), 0] \frac{u^2 N_0}{\sigma^2} \frac{r - 2\sigma}{r - 2\sigma - E_1} \\ \Leftrightarrow 4 \max[(s_{AB} - r), 0]u &\gg s_{Ab}^2 \quad \text{or} \quad \max[2(s_{AB} - r), 0] \frac{u^2 N_0}{\sigma^2} \frac{r - 2\sigma}{r - 2\sigma - E_1} \gg s_{Ab}. \end{aligned} \quad (\text{S3.24})$$

S3.3 Both single mutants are viable

Finally, we consider the case $s_{Ab} = s_{aB} = s > -1$. With $\sigma_{Ab} = \sigma_{aB} = \sigma$, deterministically, the number of Ab mutants and aB mutants is hence equal at any point of time. In the following, we formulate equations in terms of type Ab .

The wildtype is lethal. Ignoring recombination, from generation 0 to generation 1, the number of Ab individuals changes to

$$n_{Ab}(1) = (\bar{n}_{Ab}(1 - 2u) + uN_0)(1 + s). \quad (\text{S3.25})$$

From then on, it evolves according to the recursive equation

$$\begin{aligned} n_{Ab}(t+1) &= (1+s)(1-2u) \left(n_{Ab}(t) - r \frac{n_{Ab}(t)n_{aB}(t)}{n_{Ab}(t) + n_{aB}(t)} \right) \\ &= (1+s)(1-2u) \left(n_{Ab}(t) - \frac{r}{2} n_{Ab}(t) \right), \end{aligned} \quad (\text{S3.26})$$

where the second line holds since $n_{Ab}(t) = n_{aB}(t)$. With this, we have

$$n_{Ab}(t+1) = n_{Ab}(1) \left((1+s)(1-2u) \left(1 - \frac{r}{2} \right) \right)^t. \quad (\text{S3.27})$$

From generation 1 on, the number of newly generated AB individuals follows a Poisson distribution with parameter

$$\left(u(n_{Ab}(t) + n_{aB}(t)) + \frac{r}{2} n_{Ab}(t) \right) (1 + s_{AB}). \quad (\text{S3.28})$$

Putting all together and using again $n_{Ab}(t) = n_{aB}(t)$, we obtain for rescue from generation 1 on:

$$1 - e^{-\sum_{t=0}^{\infty} (2u + \frac{r}{2}) n_{Ab}(t+1) (1+s_{AB}) p_{\text{est}}^{(AB)}}. \quad (\text{S3.29})$$

With

$$\sum_{t=0}^{\infty} n_{Ab}(t+1) = \sum_{t=0}^{\infty} n_{Ab}(1) \left((1+s)(1-2u) \left(1 - \frac{r}{2} \right) \right)^t = n_{Ab}(1) \frac{1}{1 - (1+s)(1-2u) \left(1 - \frac{r}{2} \right)}, \quad (\text{S3.30})$$

this yields

$$1 - e^{-\left(\frac{(1+s_{AB})(2u + \frac{r}{2}) n_{Ab}(1)}{1 - (1+s)(1-2u) \left(1 - \frac{r}{2} \right)} \right) p_{\text{est}}^{(AB)}} \approx 1 - e^{-2s_{AB} \frac{r}{2} \frac{uN_0(1+s)}{\frac{r}{2} + 2u - s}}. \quad (\text{S3.31})$$

Combining with Eq. (S3.1), we obtain for the total probability of evolutionary rescue

$$\begin{aligned} P_{\text{rescue}} &= 1 - F_{AB} \left(e^{-(1+s_{AB})(1-r)} p_{\text{est}}^{(AB)} \right) \times e^{-\left(r \frac{\bar{n}_{Ab} \bar{n}_{aB}}{N_0} (1+s_{AB}) + u(\bar{n}_{Ab} + \bar{n}_{aB})(1+s_{AB})(1-r) \right) p_{\text{est}}^{(AB)}} \\ &\quad \times e^{-\left(\frac{(1+s_{AB})(2u + \frac{r}{2}) n_{Ab}(1)}{1 - (1+s)(1-2u) \left(1 - \frac{r}{2} \right)} \right) p_{\text{est}}^{(AB)}}. \end{aligned} \quad (\text{S3.32})$$

The wildtype is as fit as the single mutants. As a second scenario, we consider the special case $s_{ab} = s_{Ab} = s_{aB} = s$. If we ignore mating between single mutants (note that unlike in the previous scenario, they are now relatively rare), we obtain for the deterministic dynamics

$$n_{ab}(t+1) = (1+s)(n_{ab}(t) - 2un_{ab}(t)), \quad (\text{S3.33a})$$

$$n_{Ab}(t+1) = (1+s)(n_{Ab}(t) + un_{ab}(t)), \quad (\text{S3.33b})$$

$$n_{aB}(t+1) = (1+s)(n_{aB}(t) + un_{ab}(t)) \quad (\text{S3.33c})$$

with the solutions

$$n_{ab}(t) = \bar{n}_{ab}((1+s)(1-2u))^t, \quad (\text{S3.34a})$$

$$n_{Ab}(t) = n_{aB}(t) = \frac{1}{2} (N_0(1+s)^t - \bar{n}_{ab}((1+s)(1-2u))^t), \quad (\text{S3.34b})$$

and $\bar{n}_{ab} = N_0 - \bar{n}_{Ab} - \bar{n}_{aB}$. Type AB is generated at rate

$$r \frac{n_{Ab}(t)n_{aB}(t)}{N(t)} (1+s_{AB}) + u(n_{Ab}(t) + n_{aB}(t))(1+s_{AB})(1-r) \quad (\text{S3.35})$$

and establishes with probability $p_{\text{est}}^{(AB)}$ as given by Eq. (S1.23). This yields for the probability of evolutionary rescue via this pathway

$$1 - e^{-\sum_{t=1}^{\infty} \left(r \frac{n_{Ab}(t)n_{aB}(t)}{N(t)} (1+s_{AB}) + u(n_{Ab}(t) + n_{aB}(t))(1+s_{AB})(1-r) \right) p_{\text{est}}^{(AB)}}. \quad (\text{S3.36})$$

Evaluating the sums yields

$$\begin{aligned} & \sum_{t=1}^{\infty} \frac{n_{Ab}(t)n_{aB}(t)}{N(t)} \\ &= -\frac{N_0}{4s} - \frac{N_0 - \bar{n}_{Ab} - \bar{n}_{aB}}{2(1 - (1+s)(1-2u))} + \frac{(N_0 - \bar{n}_{Ab} - \bar{n}_{aB})^2}{4N_0} \frac{1}{1 - (1+s)(1-2u)^2} - \frac{\bar{n}_{Ab}\bar{n}_{aB}}{N_0}, \end{aligned} \quad (\text{S3.37a})$$

$$\sum_{t=1}^{\infty} (n_{Ab}(t) + n_{aB}(t)) = -\frac{N_0}{s} - \frac{N_0 - \bar{n}_{Ab} - \bar{n}_{aB}}{1 - (1+s)(1-2u)} - \bar{n}_{Ab} - \bar{n}_{aB}. \quad (\text{S3.37b})$$

Putting it all together, we obtain:

$$\begin{aligned} P_{\text{rescue}} &= 1 - F(e^{-(1+s_{AB})(1-r)p_{\text{est}}^{(AB)}}) \times e^{-\left(r \frac{\bar{n}_{Ab}\bar{n}_{aB}}{N_0} (1+s_{AB}) + u(\bar{n}_{Ab} + \bar{n}_{aB})(1+s_{AB})(1-r) \right) p_{\text{est}}^{(AB)}} \\ &\quad \times e^{-\left(r(1+s_{AB}) \sum_{t=1}^{\infty} \frac{n_{Ab}(t)n_{aB}(t)}{N(t)} + u(1-r)(1+s_{AB}) \sum_{t=1}^{\infty} (n_{Ab}(t) + n_{aB}(t)) \right) p_{\text{est}}^{(AB)}}. \end{aligned} \quad (\text{S3.38})$$

The wildtype is fitter than the single mutants. If $s_{Ab} = s_{aB} = s$ and $s_{ab} > s$, we can proceed as in the previous section. The dynamics of the wildtype population are again given by

$$n_{ab}(t) = \bar{n}_{ab}(1 + s_{ab})^t(1 - 2u)^t. \quad (\text{S3.39})$$

The dynamics of the single mutants follow

$$n_{Ab}(t + 1) = n_{aB}(t + 1) = (1 + s)(n_{Ab}(t) + un_{ab}(t)), \quad (\text{S3.40})$$

yielding

$$n_{Ab}(t) = \frac{(uN_0(1 + s) + \bar{n}_{Ab}(s - s_{ab})(1 - 2u))(1 + s)^t - u\bar{n}_{ab}(1 + s)(1 + s_{ab})^t(1 - 2u)^t}{s - s_{ab} + 2u(1 + s_{ab})}. \quad (\text{S3.41})$$

With the abbreviations

$$\begin{aligned} C &:= s - s_{ab} + 2u(1 + s_{ab}), \\ \alpha &:= u\bar{n}_{ab}(1 + s), \\ \beta &:= uN_0(1 + s) + \bar{n}_{Ab}(s - s_{ab})(1 - 2u), \end{aligned} \quad (\text{S3.42})$$

and $N(t) \approx n_{ab}(t)$ we obtain

$$\sum_{t=1}^{\infty} (n_{Ab}(t) + n_{aB}(t)) = \frac{\beta \sum_{t=1}^{\infty} (1 + s)^t - \alpha \sum_{t=1}^{\infty} (1 + s_{ab})^t(1 - 2u)^t}{C} = \frac{\beta \frac{1+s}{-s} - \alpha \frac{(1+s_{ab})(1-2u)}{1-(1+s_{ab})(1-2u)}}{C}, \quad (\text{S3.43a})$$

$$\begin{aligned} \sum_{t=1}^{\infty} \frac{n_{Ab}(t)n_{aB}(t)}{N(t)} &= \frac{\alpha^2(1 + s_{ab})^{2t}(1 - 2u)^{2t} - 2\alpha\beta(1 + s_{ab})^t(1 - 2u)^t(1 + s)^t + \beta^2(1 + s)^{2t}}{C^2\bar{n}_{ab}(1 + s_{ab})^t(1 - 2u)^t} \\ &= \frac{1}{C^2\bar{n}_{ab}} \left(\alpha^2 \frac{(1 + s_{ab})(1 - 2u)}{1 - (1 + s_{ab})(1 - 2u)} - 2\alpha\beta \frac{1 + s}{-s} + \beta^2 \frac{(1 + s)^2}{(1 + s_{ab})(1 - 2u) - (1 + s)^2} \right). \end{aligned} \quad (\text{S3.43b})$$

Since the wildtype dominates at all times (unless rescue has occurred), we can again approximate $p_{\text{est}}^{(AB)} = 2 \max[(s_{AB} - r), 0]$.

Fig. S3.3 shows P_{rescue} for various values of s_{ab} with all other parameter values as in Fig. 3C.

S3.4 Both single mutants have fitness greater than one

We here formalize the special case $s_{ab} = -1$, $s_{Ab} = s_{aB} = s > 0$. For this, we consider pairs consisting out of one Ab and one aB mutant. Such a pair reproduces at rate $\frac{1}{2} + \hat{s}$ and dies at rate $\frac{1}{2} - \hat{s}$ with $\hat{s} = \ln(1 + s)$. At rate $\frac{r}{2}(1 + s_{AB})$, it turns into an individual of type AB (this

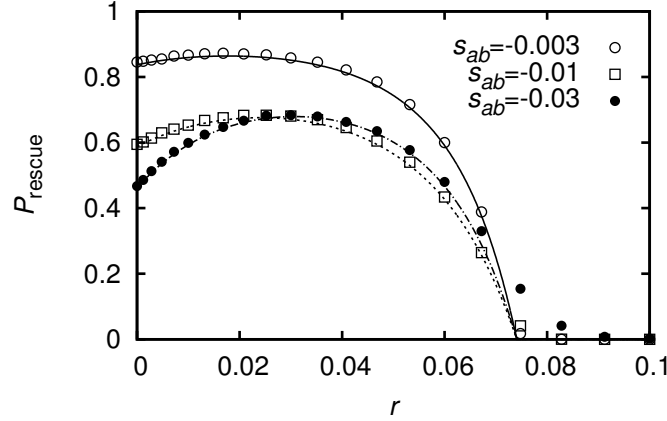


Fig. S3.3: Probability of evolutionary rescue as a function of recombination for various values of s_{ab} . All other parameter values are chosen as in Fig. 3C. Theoretical predictions are based on Eq. (S3.38) with Eq. (S3.43a). Symbols denote simulation results. Each simulation point is the average of $5 \cdot 10^4$ replicates. For the simulations with $s_{ab} = -0.003$, we considered a population as rescued when the number of double mutants reached $0.2N_0$ (changing the criterion to $0.3N_0$ did not alter the results).

ignores mutation). The growth rate of a pair is $2\hat{s}$, since in reality, we are not interested in pairs but establishment of any type (Ab , aB , AB) is fine, and each single mutant has growth rate s . However, it is pairs that convert into double mutants, and with this approximation, we assume that for every single mutant of type Ab , there is a single mutant of type aB to recombine with and vice versa. A single individual of type AB establishes a permanent lineage with probability $p_{\text{est}}^{(AB)} \approx 2s_{AB}$. Using Eq. (S1.8), we can calculate the survival probability of a process founded by exactly one pair:

$$p_{\text{est}}^{(Ab,aB)} = 1 - \frac{1 + \hat{s} + \frac{r}{2}(1 + s_{AB}) - \sqrt{(\hat{s} - \frac{r}{2}(1 + s_{AB}))^2 + (1 + 2\hat{s})r(1 + s_{AB})p_{\text{est}}^{(AB)}}}{1 + 2\hat{s}} \quad (\text{S3.44})$$

$$\approx 2s - \frac{r}{2} + \sqrt{\left(2s - \frac{r}{2}\right)^2 + 2s_{AB}r}.$$

The probability of evolutionary rescue from generation 1 on is given by

$$1 - e^{-\bar{n}_{Ab}(1+s_{Ab})p_{\text{est}}^{(Ab,aB)}}. \quad (\text{S3.45})$$

Neglecting the contribution of double mutants from the standing genetic variation to rescue, the possibility to generate the double mutant has a significant effect if either

$$p_{\text{est}}^{(Ab,aB)} \gg 4s \quad \text{or} \quad p_{\text{est}}^{(Ab,aB)} \ll 4s. \quad (\text{S3.46})$$

These conditions simplify in few steps to

$$s_{AB} \gg 2s \quad \text{or} \quad s_{AB} \ll 2s. \quad (\text{S3.47})$$

S3.5 Two-step rescue vs single-step rescue

We briefly discuss some instances where two-step rescue (as analyzed in this paper) is more likely to happen than single-step rescue (where there are only two types – the wildtype and the rescue type – and a single mutational step between them). For easier comparison, we denote the wildtype by ab and the rescue genotype by AB for single-step rescue as well. Mutation from wildtype to rescue mutants may happen with probability u_s . With Eq. (S1.4), the p.g.f. for the number of rescue mutations in the standing genetic variation is derived to be

$$F_{AB}^{\text{ssr}}(y) = \left(\frac{2\sigma_{AB}}{y + \sigma_{AB}y + \sigma_{AB} - 1} \right)^{2u_s N_0}. \quad (\text{S3.48})$$

The probability of evolutionary rescue for single-step rescue is given by

$$\begin{aligned} P_{\text{rescue}}^{\text{ssr}} &= 1 - F_{AB}^{\text{ssr}}(e^{-(1+s_{AB})p_{\text{est}}^{(AB)}}) e^{-\frac{u_s N_0}{-s_{ab}}(1+s_{AB})p_{\text{est}}^{(AB)}} \\ &= 1 - e^{-p_{\text{est}}^{(AB)}(1+s_{AB})\left[\frac{u_s N_0}{-\sigma_{AB}}(1+\sigma_{AB}) - \frac{u_s N_0}{-s_{ab}}\right]} \approx 1 - e^{-2s_{AB}\left[\frac{u_s N_0}{-\sigma_{AB}} - \frac{u_s N_0}{-s_{ab}}\right]}. \end{aligned} \quad (\text{S3.49})$$

where the first summand in the brackets accounts for the contribution of standing genetic variation and the second one for new mutations after the environmental change (cf. also Orr and Unckless (2008, 2014); Bell and Collins (2008); Uecker *et al.* (2014)).

In the following, we focus on scenarios where the wildtype is lethal in the new environment and approximate single-step rescue by

$$P_{\text{rescue}}^{\text{sgv}} \approx 1 - e^{-2s_{AB}\frac{u_s N_0}{-\sigma_{AB}}}. \quad (\text{S3.50})$$

Lethal single mutants. For two-step rescue, we use approximation Eq. (8):

$$P_{\text{rescue}} \approx 1 - e^{-2s_{AB}\frac{u^2 N_0}{\sigma^2}\frac{r-2\sigma}{r-2\sigma-E_1}} \stackrel{r \text{ large}/E_1=0}{\approx} 1 - e^{-2s_{AB}\frac{u^2 N_0}{\sigma^2}}. \quad (\text{S3.51})$$

Comparing with Eq. (S3.50) shows that two-step rescue is more likely if

$$\frac{u^2}{\sigma^2}\frac{r-2\sigma}{r-2\sigma-E_1} > \frac{u_s}{-\sigma_{AB}}. \quad (\text{S3.52})$$

For large recombination, this reduces to

$$\frac{u^2}{\sigma^2} > \frac{u_s}{-\sigma_{AB}}. \quad (\text{S3.53})$$

For $E_1 = 0$ (which implies $\sigma_{AB} \approx 2\sigma$):

$$\frac{u^2}{-\sigma} > \frac{u_s}{2}. \quad (\text{S3.54})$$

One viable single mutant. Following section S3.2, two-step rescue can be approximated by

$$1 - e^{-2s_{AB} \frac{u^2 N_0}{\sigma^2} \frac{r-2\sigma}{r-2\sigma-E_1}} \times e^{-\frac{u N_0}{-\sigma} \frac{2s_{AB} u}{-s_{Ab}}} . \quad (\text{S3.55})$$

Under these conditions, two-step rescue is more likely than single-step rescue if

$$\frac{u^2}{\sigma^2} \frac{r-2\sigma}{r-2\sigma-E_1} + \frac{u^2}{\sigma s_{Ab}} > \frac{u_s}{-\sigma_{AB}} . \quad (\text{S3.56})$$

Again, for strong recombination:

$$\frac{u^2}{\sigma^2} + \frac{u^2}{\sigma s_{Ab}} > \frac{u_s}{-\sigma_{AB}} . \quad (\text{S3.57})$$

And for $E_1 = 0$:

$$\frac{u^2}{-\sigma} + \frac{u^2}{-s_{Ab}} > \frac{u_s}{2} . \quad (\text{S3.58})$$

Viable single mutants. Last, we consider a scenario with both single mutants viable. With Eq. (10), the probability of evolutionary rescue is given by

$$1 - e^{-2s_{AB} \frac{u^2 N_0}{\sigma^2} \frac{r-2\sigma}{r-2\sigma-E_1}} \times e^{-2s_{AB} \frac{u N_0}{-\sigma} (1+s_{Ab}) \frac{r}{r-2s_{Ab}}} . \quad (\text{S3.59})$$

This yields for the condition that two-step rescue is more likely than single-step rescue

$$\frac{u^2}{\sigma^2} \frac{r-2\sigma}{r-2\sigma-E_1} + (1+s_{Ab}) \frac{u}{-\sigma} \frac{r}{r-2s_{Ab}} > \frac{u_s}{-\sigma_{AB}} , \quad (\text{S3.60})$$

which for strong recombination simplifies to

$$\frac{u}{-\sigma} \left(\frac{u}{-\sigma} + (1+s_{Ab}) \frac{r}{r-2s_{Ab}} \right) > \frac{u_s}{-\sigma_{AB}} . \quad (\text{S3.61})$$

For $E_1 = 0$:

$$\frac{u^2}{-\sigma} + (1+s_{Ab}) \frac{ur}{r-2s_{Ab}} > \frac{u_s}{2} . \quad (\text{S3.62})$$