Here, we focus on one such barrier to gene flow, hybrid sterility. Hybrid sterility can be caused by a variety of mechanisms that can generally be classified into two categories: incompatibilities between diverged chromosomes (such as large-scale chromosomal rearrangements and anti-recombination) and incompatibilities between individual genes from the diverging populations.

Here we show that repressing anti-recombination dissolves the reproductive barrier between two yeast species, S. cerevisiae and S. paradoxus, increasing their production of viable hybrid gametes by 70-fold (Figure 1A). We did this by repressing the meiotic expression of just two highly conserved genes, SGS1 and MSH2. Msh2 is a component of the mismatch repair system that removes base-pair mismatches in duplex DNA, both to repair misincorporations in newly synthesized DNA and to inhibit recombination between diverged sequences (anti-recombination). The former activity reduces mutations, and the latter can help maintain genome integrity by limiting ectopic recombination between non-homologous chromosomes and dispersed repeats. Sgs1 is a DNA helicase that is assumed to act downstream of mismatch recognition by Msh2 to unwind nascent recombination intermediates containing a high density of mismatches, but also plays a more general function in recombination to disassemble joint-molecule intermediates that could lead to crossovers. Thus, although completely deleting MSH2 enhances meiotic recombination between the diverged chromosomes of S. cerevisiae x S. paradoxus hybrids, increasing proper chromosome segregation and therefore hybrid spore viability, this benefit is countered by elevated mutagenesis and genome instability in mitotically dividing cells, reducing viability. We therefore replaced the native promoters of MSH2 and SGS1 with the CLB2 promoter, which represses gene expression during meiosis but not mitosis.

Meiotic repression of either gene alone significantly increased hybrid spore viability (Figure 1A, MSH2 p = 7.99 x 10^-6; SGS1 p = 2.2 x 10^-9). Overall spore viability rose from 0.46%
in the wild-type hybrid to 3.18% in the pCLB2-MSH2 strain and to 20.08% in the pCLB2-SGS1 strain. Spore viability was further improved to 32.65% when both genes were repressed (p < 2.2 × 10^-16). Although hybrid fertility was not increased to the level of the parents — the S. cerevisiae and S. paradoxus parent fertilities were 83.75% and 92.25%, respectively — it was well within the range of fertilities of non-hybrid crosses formed from diverged populations of one species or the other. For example, 32–87% for S. paradoxus or S. cerevisiae crosses with collinear genomes7, 14–86% for wild S. paradoxus crosses8,9. These results show that anti-recombination determines most of the hybrid sterility barrier between our S. cerevisiae and S. paradoxus strains.

This remarkable restoration of hybrid fertility allowed us to produce a large sample of perfectly euploid hybrid gametes. Any viable gametes produced by a hybrid are usually aneuploid10, and this remains the case even when MSH2 is knocked out11.

By dramatically improving hybrid fertility, we significantly increased the production of hybrid tetrads in which all four spores were viable from 0% in the wild-type hybrid to 5.3% in the double mutant hybrid (Figure 1B; 0 out of 269 versus 108 out of 2,037, respectively, p = 2.04 × 10^-5). Because all chromosomes are essential in yeast, we can infer that these full tetrads contain only euploid hybrid gametes. Generation of these hybrids enables the unambiguous analysis of recombination and trait mapping, both of which were previously confounded by aneuploidy in sampled hybrid spores10.

Finally, in order to map the genome-wide distribution of crossovers in our pCLB2-MSH2 pCLB2-SGS1 double mutant hybrid, we sequenced the genomes of the 336 hybrid spores from mutant hybrid, we sequenced the genomes of our recombinant hybrid spores revealed that the suppression of anti-recombination activity was evenly distributed across the genomes of both species, rather than being locally enriched at hotspot regions (Figure S1A).

This study shows that repressing the meiotic expression of just two genes, SGS1 and MSH2, overcomes the anti-recombination barrier between two yeast species, restoring the fertility of their hybrids to intra-specific levels, and allowing them to produce viable, euploid, recombinant gametes (Figures 1C and S1B). We demonstrate directly that anti-recombination is the major cause of post-zygotically reproductive isolation between these species. By enabling recombination between such diverged species, our method can be used to identify any intrinsic genetic incompatibilities, or speciation genes, to map the genetics underlying diverged phenotypes, or to produce recombinant hybrids with novel properties for commercial or research use.

SUPPLEMENTAL INFORMATION

Supplemental Information includes one figure, experimental procedures, author contributions, and supplemental references and can be found with this article online at https://doi.org/10.1016/j.cub.2020.12.038.

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DECLARATION OF INTERESTS

The authors declare no competing interests.

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