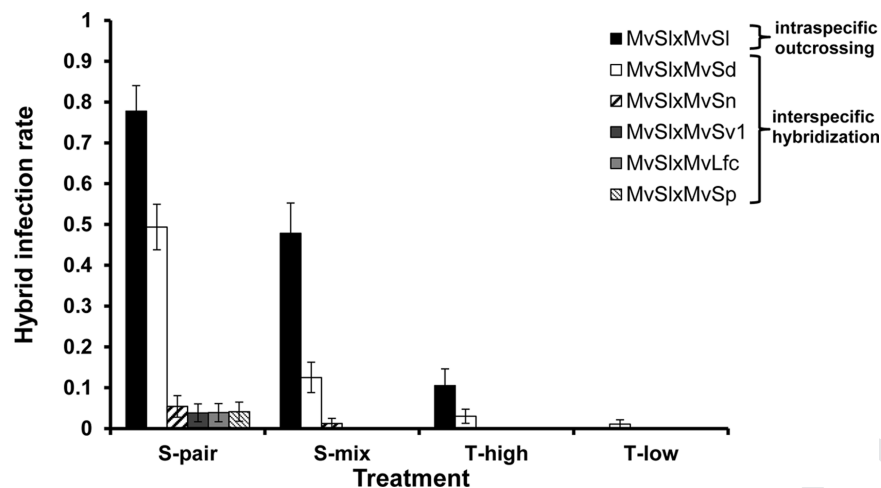


## SELFING AND SIBLING COMPETITION PREVENT GENE FLOW



**Figure 3.** Hybrid infection rate according to treatment, cross, and genetic distance. The hybrid infection rate is shown for each cross across all four treatments; see Figure 1 and the text for treatment definitions. Hybrid infection rate indicates the proportion of all inoculated plants that became infected with hybrid pathogens. The genetic distance between MvSI and the hybridizing pathogen species increases from left to right within each treatment. Note that MvSI  $\times$  MvSI crosses represent outcrossing rather than hybridization and are used for comparison to hybrid crosses. Error bars show the standard error of the proportion.

mating influences the probability of hybridization between *Microbotryum* species (Table 4, Fig. 3). In comparing only treatments with forced hybridization (S-pair) to those with the possibility of sporidial selfing (S-mix), the potential for selfing reduced the rate of hybrid infection by 39% for intraspecific crosses (MvSI  $\times$  MvSI) and by 75% for interspecific hybrid crosses with the closest species (MvSI  $\times$  MvSd). The possibility of selfing reduced the rate of hybrid infection by 77% for the next most distant cross (MvSI  $\times$  MvSn) and by 100% for the three most distant interspecific crosses (Fig. 3). When comparing treatments in which selfing was possible between sporidia (S-mix) to those in which intrapromycelial selfing was also possible (T-high), the potential of intrapromycelial mating reduced the rate of hybrid infection by 78% for intraspecific crosses and by 76% for the closest interspecific crosses (MvSI  $\times$  MvSd) (Fig. 3). This comparison was not informative for the four more distant interspecific crosses (MvSI  $\times$  MvSn, MvSI  $\times$  MvSp, MvSI  $\times$  MvSv1, MvSI  $\times$  MvLfc) because hybrid infection rates were 5% or less under forced hybridization (S-pair) and were already dramatically reduced to 0–1% in the S-mix treatment (Fig. 3).

The concentration of teliospores used in inoculation affected hybrid infection rates (Fig. 3, Table 4). As predicted, fewer hybrids resulted at low teliospore densities (T-low) relative to high teliospore densities (T-high) for intraspecific crosses (MvSI  $\times$  MvSI) and the closest interspecific crosses (MvSI  $\times$  MvSd). This comparison was not possible for more distant interspecific crosses due to the absence of hybrid infections in both treatments.

#### BARRIERS TO HYBRIDIZATION: ADDITION OF THE SIBLING COMPETITION ARENA

The intrinsic fitness reduction of hybrid progeny relative to selfed progeny, without accounting for competition, was estimated based upon the deviation of infection rates in the forced mating treatment (S-pair) from the mean infection rate of the three selfed MvSI  $\times$  MvSI crosses, which was calculated to be 0.617. Using this estimate of deviation, a significant negative correlation between intrinsic hybrid fitness and genetic distance between MvSI and the hybridizing species was found ( $r = 0.85$ ,  $P = 0.031$ ) (S-pair in Fig. 3). The hybrid infection rate in the S-mix treatment for MvSI  $\times$  MvSd was significantly lower than expected based upon the outcrossing rate measured in intraspecific S-mix crosses and the intrinsic reduction in MvSI  $\times$  MvSd hybrid fitness as measured by the S-pair hybrid infection rate ( $\chi^2 = 13.333$ ,  $df = 1$ ,  $P = 0.0003$ ) (Fig. 4). Thus, competition generated by the presence of selfed progeny further impeded hybrids from successfully infecting beyond their intrinsic fitness reduction and the selfing rate. This difference remained marginally significant when the intraspecific outcrossing rate was reduced from 0.478 to 0.30 to test for robustness to observed variation in outcrossing rates ( $\chi^2 = 3.411$ ,  $df = 1$ ,  $P = 0.0647$ ) (Giraud et al. 2005). The most relevant outcrossing rate here, however, remains the prior value of 0.478, as it was obtained directly from the experimental conditions applied across all treatments. Moreover, the hybrid infection rate in the T-high treatment for MvSI  $\times$  MvSd was marginally lower than expected based upon the expected S-mix hybrid infection rate under this intraspecific outcrossing rate and the 76% reduction in MvSI  $\times$  MvSd hybrid infection