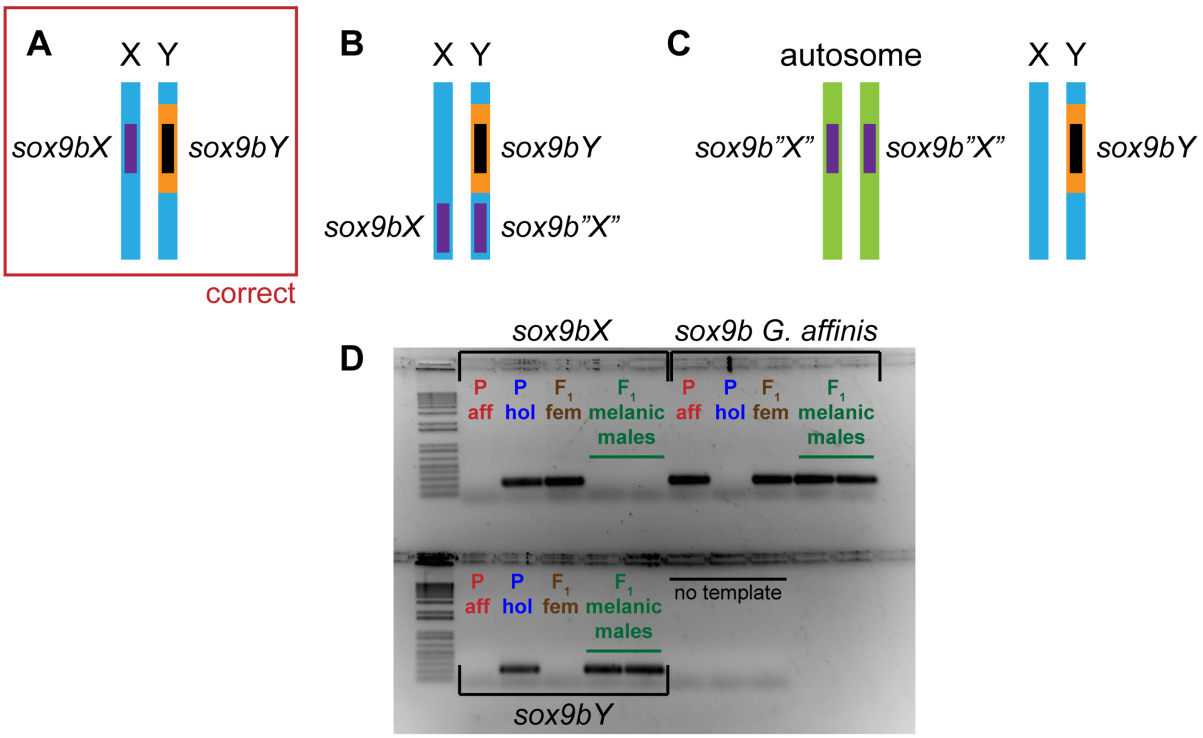

Figure S6 *Sox9bY* was generated by allelic diversification.

(A) Hypothesis 1: *sox9bY* and *sox9bX* underwent allelic diversification due to repressed recombination between the X and Y. (B) Hypothesis 2: *sox9bY* represents a duplication of *sox9bX*, which is located on the same linkage group. (C) Hypothesis 3: *sox9bY* represents a duplication *of sox9bX*, which is located on a different linkage group. (D) Gel electrophoresis of a PCR using primers distinguishing between *G. holbrooki sox9bX*, *G. holbrooki sox9bY*, and *G. affinis sox9b*. DNA from a *G. affinis* female, a melanic *G. holbrooki* male, and F_1 fish of a cross between the two fish was used. If (B) or (C) would be correct, F_1 males would have a copy of *G. affinis sox9b*, *sox9bY*, and *sox9bX*, which is not the case. Hypothesis (A) is therefore correct. The position and orientation of *sox9bX* and *sox9bY* on the *G. holbrooki* sex chromosome pair are not known. Orange, differentiated region on the Y. P aff, parental *G. affinis* female; P hol, parental melanic *G. holbrooki* male; F_1 fem, F_1 wildtype female.