**Supplementary SX: Cytological observations of B chromosomes in experimental lines**

B chromosome screening was carried out through cytological preparations of laid embryos and the testes of second-instar males. B chromosomes could be easily identified from the embryonic stage when present in a line (Fig 1A) although. The number of paternal chromosomes could also be counted at the beginning of meiosis in male testes (Fig 1B; (i) and (iii)) and provided an alternative screening stage when chromosomes in the embryonic stage were not properly separated. Finally, preparation of cells at the end of male meiosis in B carrying lines allowed to visualize B segregation to the paternal or maternal genome (Fig 1B (iv) and (v)).

A picture containing graphical user interface

Description automatically generated  
***Figure 1.*** Cytological preparations of *Pseudococcus viburni* lines without and carrying B chromosomes. (A) Embryos with (i) only A chromosomes 2n=10, (i) A chromosomes 2n=10 + 1B chromosome, (iii) A chromosomes 2n=10 + 2B chromosomes. (B) Testes of second-instar males with cells at the beginning of meiosis, where the paternal chromosomes are showing either n=5 (i) or n=5 +2B (ii); and cells at the end of meiosis where the future gametes do not carry B in 0B lines (ii) or where the B chromosome either segregates with the paternal genome (iv) or the maternal genome (v). ADD which samples were used for sequencing. PV13 PV04 PV21 PV23