Review of: "The Assembly of the Y Chromosome Reveals Amplification of Genes Regulating Male Fertility in Bactrocera Dorsalis"

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Potential competing interests: No potential competing interests to declare.

The Article: "The Assembly of the Y Chromosome Reveals Amplification of Genes Regulating Male Fertility in Bactrocera Dorsalis" reports the first successful assembly of the Y chromosome in the oriental fruit fly, Bactrocera dorsalis, an invasive pest species. The authors used a combination of PacBio HiFi sequencing, Hi-C analysis, and resequencing data from male and female flies to achieve a chromosome-level genome assembly. This allowed them to identify and characterize the Y chromosome, which is typically difficult to assemble due to its highly repetitive nature.

The article's main findings are: - The Y chromosome of B. dorsalis is relatively small (~7.6 Mb) and contains 61 genes, including nine multi-copy genes. - The M factor (MoY), a key male-determining gene in Tephritidae, was found to have multiple copies in B. dorsalis. - The gene gyf-like on the Y chromosome (gyfY) was found to have undergone significant amplification, with 32 copies identified. RNA interference (RNAi) experiments showed that gyfY is essential for male fertility.

Methodological Assessment:
The methodology used in this study appears to be robust and well-suited to address the research question. The combination of PacBio HiFi sequencing, Hi-C analysis, and resequencing data allowed for a high-quality genome assembly, including the challenging Y chromosome. The use of ddPCR to verify the copy number of MoY and gyfY provides further validation of the genomic findings. The RNAi experiments were appropriately designed and conducted, with the use of dsGFP as a negative control and the assessment of sperm viability as a clear phenotypic readout.

This study makes several significant contributions to the field of genetics and cytogenetics:
It provides the first complete assembly of the Y chromosome in a Tephritidae species, opening up new avenues for research on sex determination and male fertility in this important group of insects.
The discovery of multiple copies of MoY and the amplification of gyfY highlights the unique evolutionary trajectory of the Y chromosome in B. dorsalis.
The functional analysis of gyfY provides valuable insights into the genetic regulation of male fertility in this species.

Suggestions for Improvement: The authors mention that the genome assembly is "highly completed" based on a BUSCO score of 98.3%. While this is a good score, it's worth noting that it still implies that 1.7% of the expected genes are missing or fragmented. It would be helpful to provide more context on the limitations of the assembly. The discussion of the repeat

content of the X chromosome could be expanded. The authors suggest that the high repeat content may be due to reduced selection efficacy, but other possibilities could be explored.

This is a well-conducted study that provides valuable insights into the structure and function of the Y chromosome in B. dorsalis. The findings have important implications for understanding the biology of this invasive pest and for developing novel control strategies. The suggestions for improvement are minor and do not detract from the overall significance of the work.