

Supplementary Figures and Tables

for

The House Fly Y Chromosome is Young and Minimally Differentiated from its Ancient X Chromosome Partner

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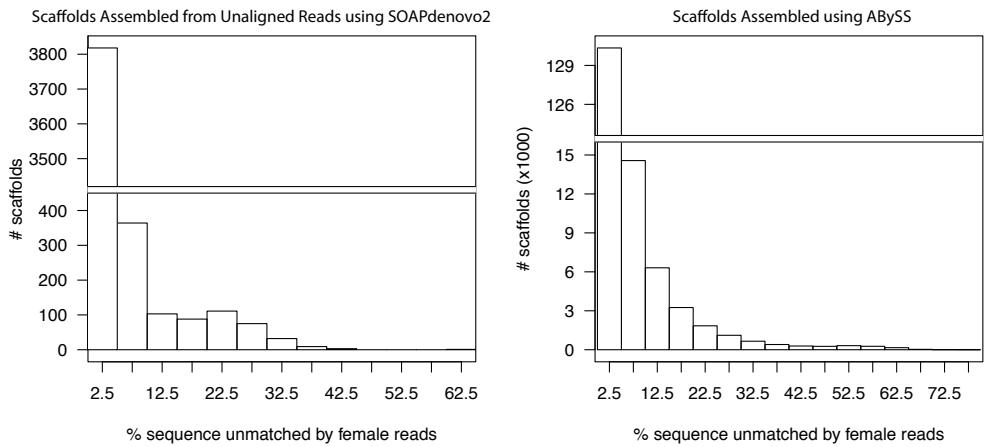
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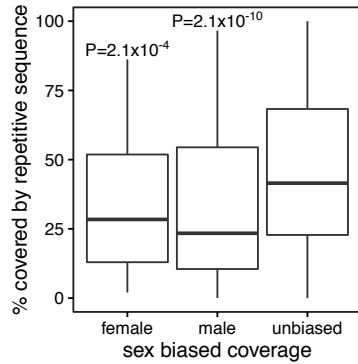
Running title: House fly sex chromosomes

Keywords: sex chromosomes; neo-Y chromosome; *Musca domestica*

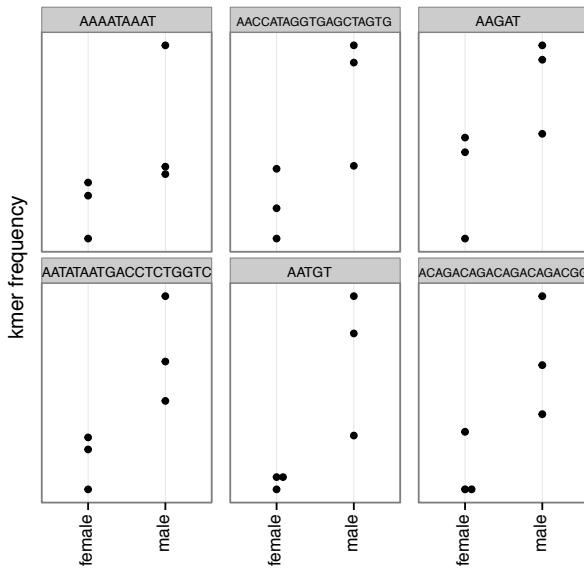
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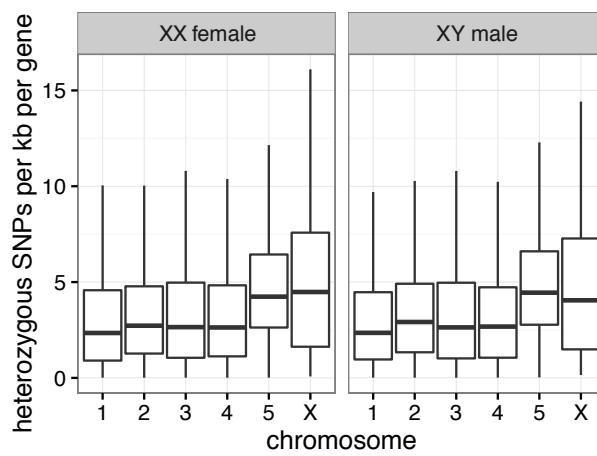
Supplemental Fig S1: Histograms of female read mapping coverage to scaffolds from male genomes assembled using SOAPdenovo2 with reads that did not align to the female reference genome (left) or using ABySS (right).



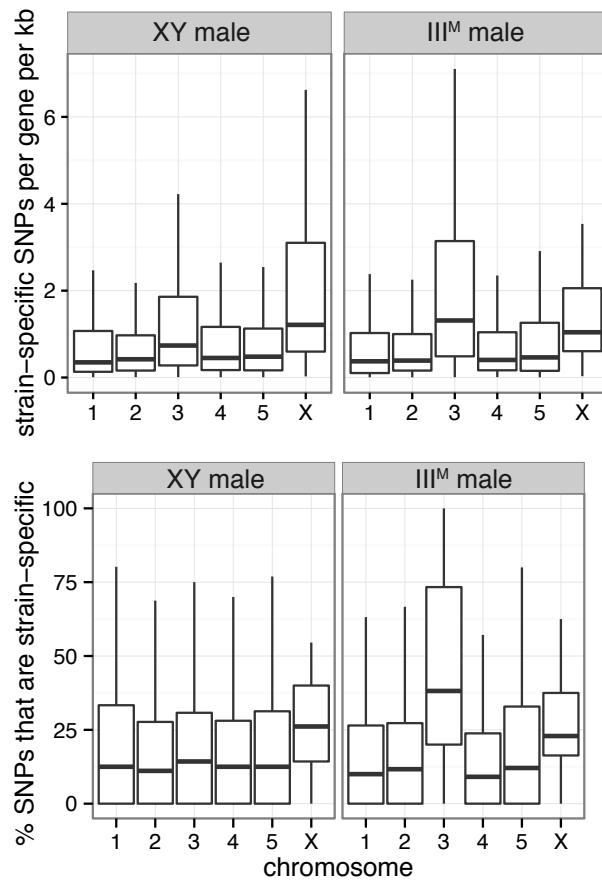
Supplemental Fig S2: Boxplots show the distribution of the percent of 1 kb windows that contain predicted repetitive sequence. Three different types of 1 kb windows are plotted: those with female-biased read mapping coverage ($\log_2 \frac{M}{F} < -1$), those with male-biased coverage ($\log_2 \frac{M}{F} > 1$), and those with insignificant differences in coverage (unbiased). P values comparing the female- and male-biased windows with the unbiased windows from a Mann-Whitney test are shown.



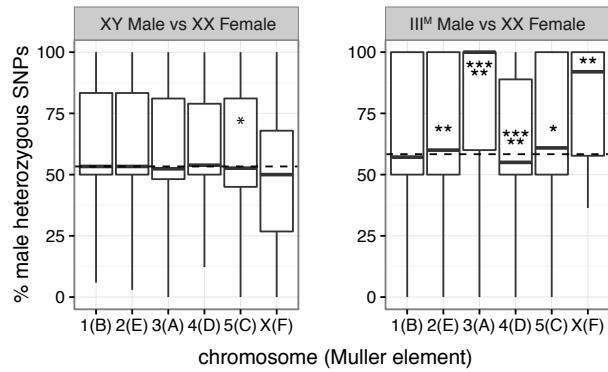
Supplemental Fig S3: The frequency of the six *k*-mers over-represented in males is plotted for each of the 3 female and 3 male libraries.



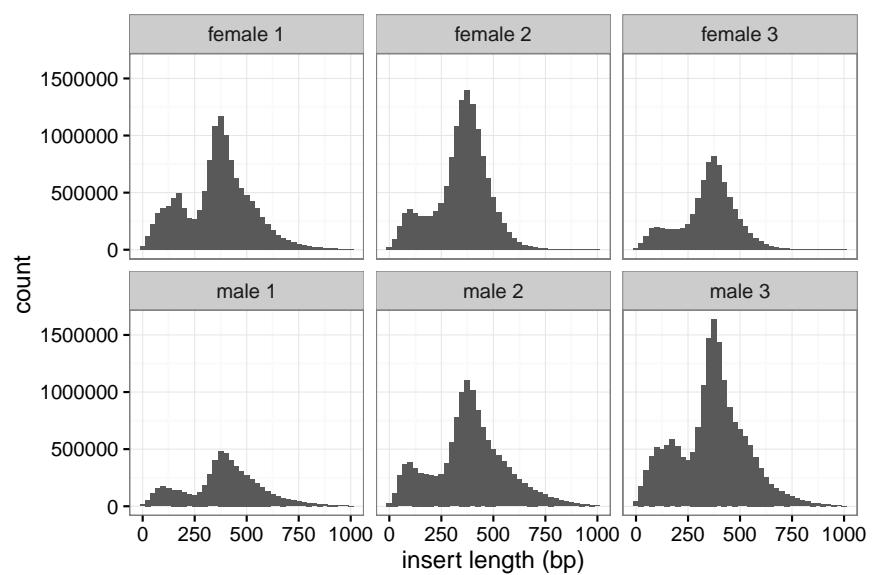
Supplemental Fig S4: Boxplots show the distributions of heterozygous SNPs per kb per gene for each chromosome from the re-sequencing of aabyss females and males.



Supplemental Fig S5: Boxplots show the distributions of strain-specific SNPs per gene per kb (top) and the percent of SNPs that are strain-specific per gene (bottom) for each chromosome from RNA-seq data collected in XY and III^M males.



Supplemental Fig S6: Box plots show the distributions of the percent of heterozygous SNPs within genes on each chromosome in males relative to females from strains with XY males (left) or III^M males (right). The median across all autosomes is indicated by a dashed line. Asterisks indicate significant differences between a chromosome and all other autosomes in a Mann-Whitney test (* $P < 0.05$, ** $P < 0.005$, **** $P < 0.000005$).



Supplemental Fig S7: Distributions of insert sizes for the 6 male and female aabys libraries.

| strain | library | mapped | mapped_single | mapped_distant | unmapped |
|--------|------------------|----------|---------------|----------------|----------|
| aabys | female1-26111291 | 13049678 | 887676 | 6292401 | 501177 |
| aabys | female2-26123107 | 13712814 | 809792 | 7125338 | 427100 |
| aabys | female3-26124109 | 7972753 | 550477 | 4716107 | 250508 |
| aabys | male1-26127107 | 6081087 | 656412 | 4333290 | 221249 |
| aabys | male2-26119223 | 13559671 | 1177151 | 8930649 | 514483 |
| aabys | male3-26113283 | 18323724 | 1275507 | 9500131 | 563536 |
| A3 | 1_A3_female_S10 | 21641219 | 2157816 | 5795725 | 1116445 |
| A3 | 2_A3_female_S7 | 22594865 | 2070878 | 6009122 | 1099204 |
| A3 | 3_A3_female_S6 | 28633153 | 2966799 | 8121340 | 1412310 |
| A3 | 4_A3_male_S9 | 22766977 | 2407270 | 6228309 | 1206439 |
| A3 | 5_A3_male_S5 | 21817355 | 2136473 | 5675312 | 1140114 |
| A3 | 6_A3_male_S4 | 19437196 | 1902804 | 4873879 | 1077971 |
| LPR | 7_LPR_female_S3 | 16238580 | 2325002 | 5586155 | 1062667 |
| LPR | 8_LPR_female_S12 | 16779395 | 2388949 | 5921294 | 1174171 |
| LPR | 9_LPR_female_S2 | 19773472 | 3284957 | 7356944 | 1317193 |
| LPR | 10_LPR_male_S8 | 15172543 | 2379298 | 5457376 | 1070268 |
| LPR | 11_LPR_male_S11 | 14438841 | 2420096 | 5374716 | 1041780 |
| LPR | 12_LPR_male_S1 | 14150680 | 2252137 | 5148394 | 1023886 |

Supplemental Table S1: Read mapping statistics for aabys, A3, and LPR male and female sequencing libraries. The number of paired reads that map to the same scaffold (mapped), where only one of two reads in a pair mapped (mapped_single), where two reads map to different scaffolds (mapped_distant), and where both reads failed to map (unmapped) are shown.

| Assembler | Reads | N50 | Scaffolds/Contigs | Total Length |
|-------------|---------------|------|-------------------|--------------|
| SOAPdenovo2 | All | 2491 | 231066 | 522258915 |
| SOAPdenovo2 | MaleUnaligned | 1403 | 4604 | 6769258 |
| ABYSS | All | 1936 | 159804 | 300356116 |

Supplemental Table S2: Assembly statistics for male genomes are shown for the three approaches: SOAPdenovo2 with all reads, SOAPdenovo2 with only reads that did not align to the reference genome, and ABYSS. The statistics are scaffold/contig N50, the number of scaffolds/contigs in the assembly, and the total length of the assembled genome. Only scaffolds/contigs that are at least 1 kb long were included in the statistics.