



Supplementary Figure S2: Read length distribution for all small RNA libraries sequenced in this study. We extended our previous broad and deep analysis of *D. melanogaster* by sampling 11 additional *Drosophila* species as listed to the right, by analyzing mixed embryos, adult heads, male bodies and female bodies; a testis library was also generated for *D. simulans*. A subset of libraries were sequenced in replicates, especially ones where the expected dominant miRNA-sized peak (21-22 nt) peak was not initially observed. A piRNA peak is seen in most of the body libraries. Due to the technical difficulty in culturing *D. grimshawi*, it was only feasible to generate two libraries for male and female bodies.