Supplementary file 1

**A) Summary of sequencing experiments**

Number of reads; from each experiment, that are left after removal of rRNA and tRNA contaminants, that are unique matches and that map to CDS regions of the genome.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Experiment | Raw reads | Reads that pass clipping and trimming | After removal of rRNA and tRNA | Tophat mapped reads | Unique match reads | Reads that map to ORFs |
| Small polysomal  footprint | 99,973,197 | 93,838,021 | 6,250,297 (6.66%) | 5,498,415  (87.97%) | 3,748,512  (68.17%) | 2,870,975  (76.59%) |
| Large polysomal  footprint | 67,230,885 | 59,191,477 | 3,579,490 (6.05%) | 2,625,211  (73.34%) | 2,371,751  (90.34%) | 1,906,050  (80,36%) |
| Total cytoplasmic mRNA 1 | 3,737,677 | 3,296,829 | 2,249,793 (68.24%) | 2,006,335  (89.17%) | ND | 1,619,013  (80.7%) |
| Total cytoplasmic mRNA 2 | 8,698,975 | 4,374,205 | 2,911,316 (66.56%) | 2,559,302  (87.91%) | ND | 1,956,992  (76.47%) |
| Small  polysomal  footprint  extensive | 189,631,476 | 188,066,263 | 20,008,723 (10.64%) | 9,160,610  (45.78%) | 8,133,854  (88.79%) | 5,904,132  (72.59%) |
| -rRNA all polysomes  footprint | 14,092,100 | 12,759,694 | 4,144,111 (32.48%) | 3,190,896  (77.0%) | 2,972,003  (93.14%) | 2,355,257  (79.25%) |

**B) Summary of smORF embryo RNA-seq data**

Number of translated smORFs expressed throughout embryonic stages of *Drosophila melanogaster,* according to RNAseq data (FlyBase).

|  |  |
| --- | --- |
| Number of Embryo Stages | Number of smORFs (%) |
| 12 | 88 (41.7%) |
| 11 | 16 (7.6%) |
| 10 | 11 (5.2%) |
| 9 | 17 (8.1%) |
| 8 | 9 (4.3%) |
| 7 | 8 (3.8%) |
| 1-6 | 47 (22.3%) |
| 0 | 15 (7.1%) |
| **Total** | **211** |
| NA | 17 |