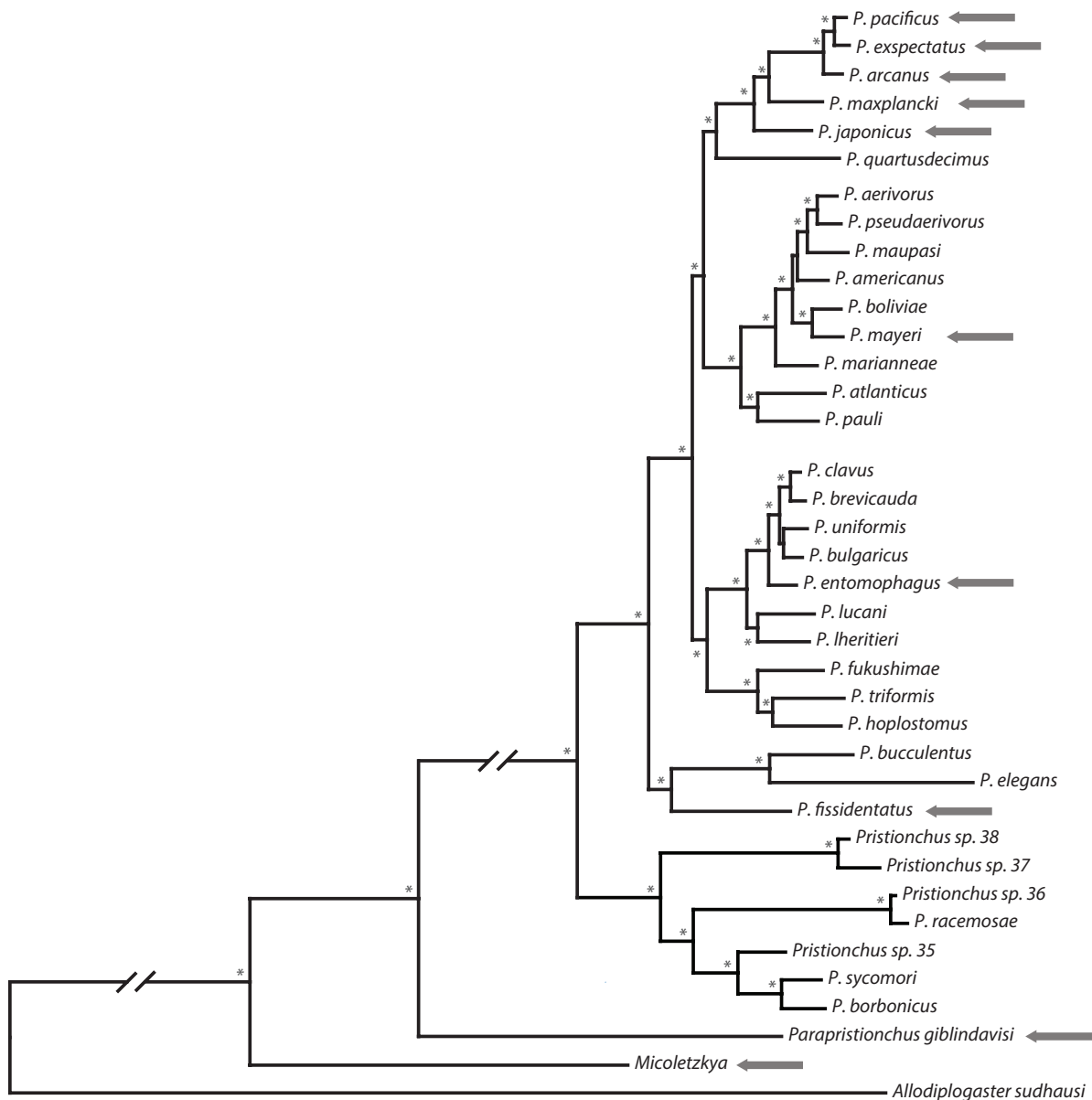
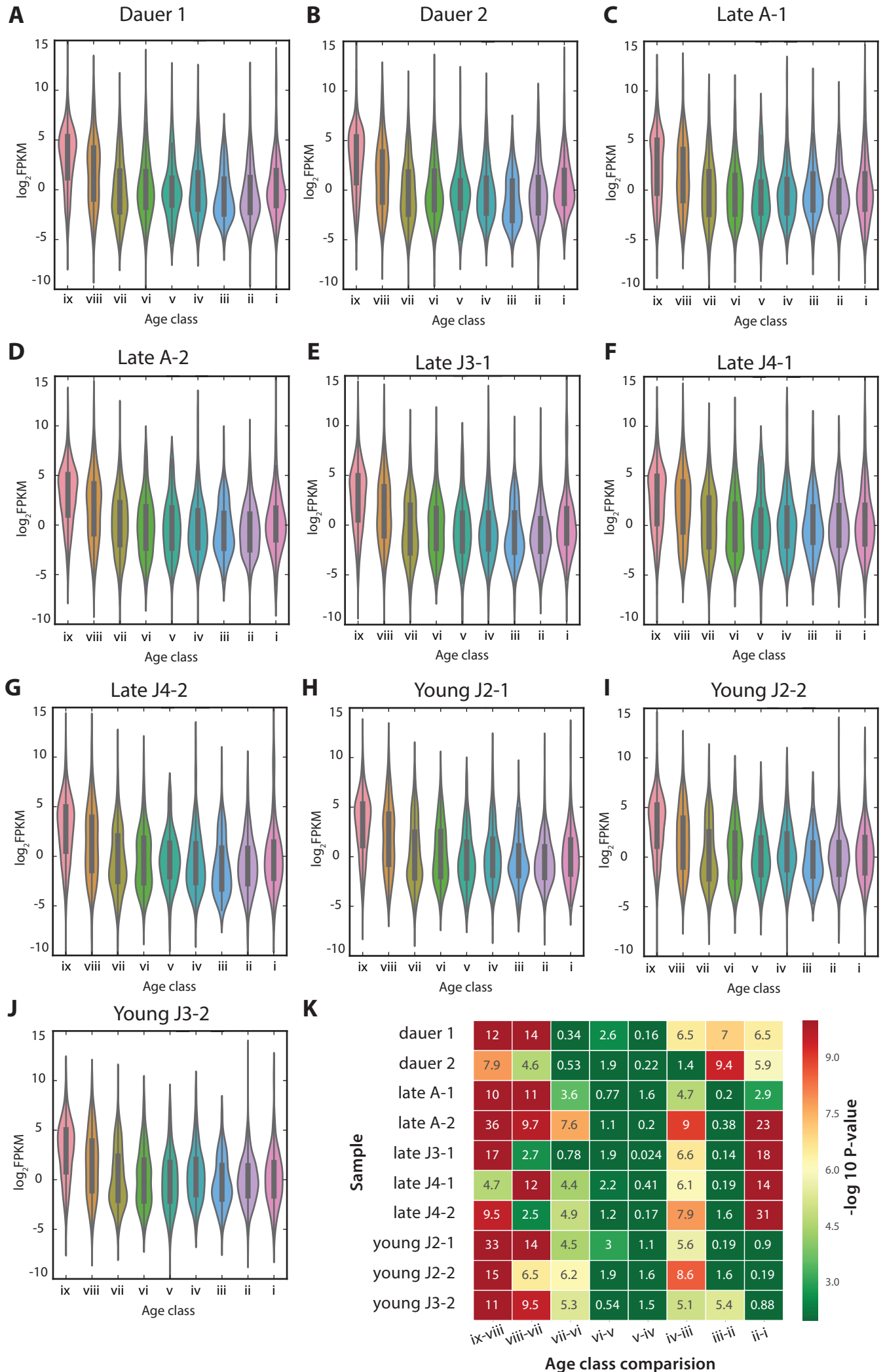
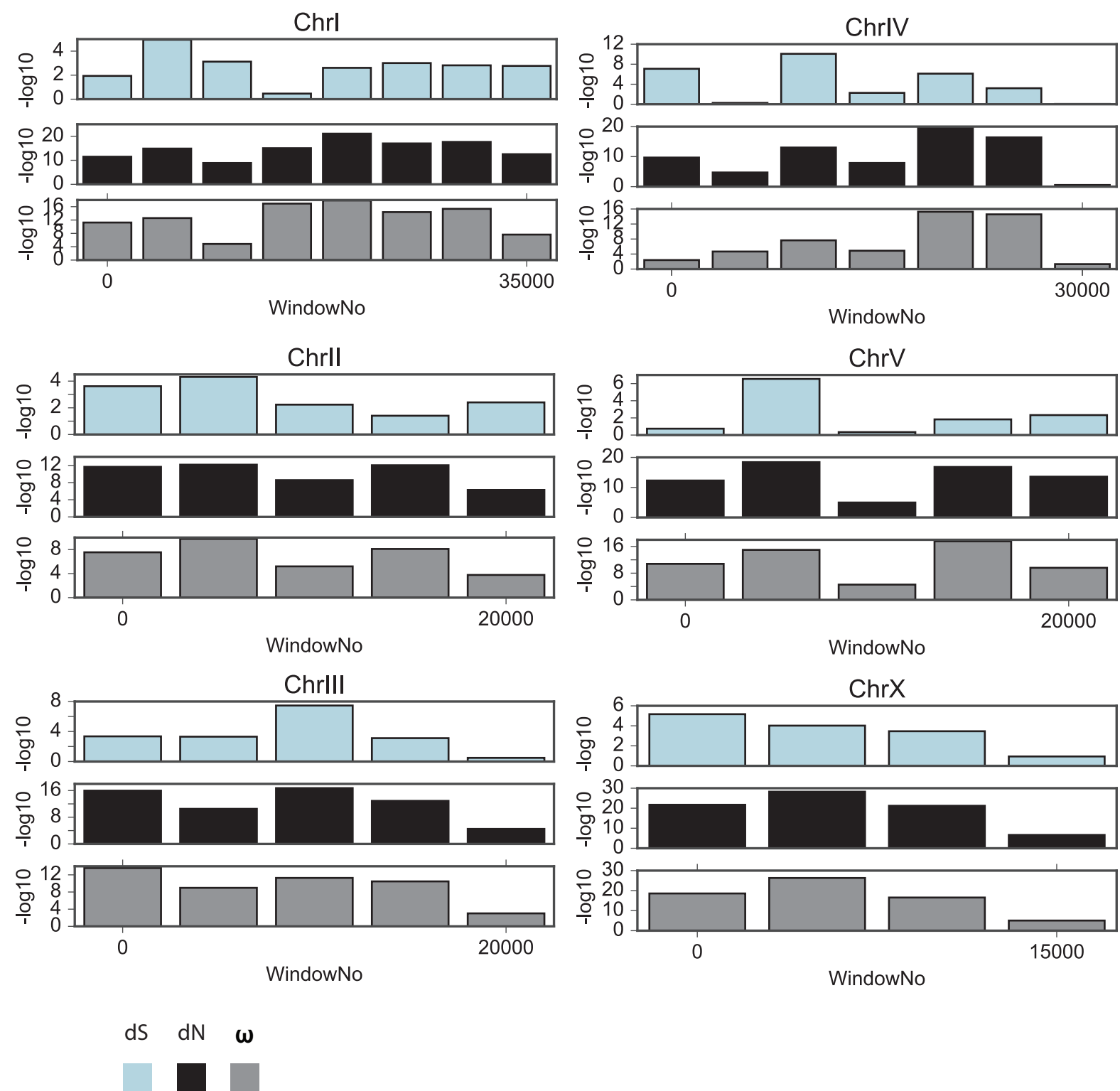




**Supplemental Figure S1.** Phylogeny of *Pristionchus* species inferred from 18S and 28S rRNA genes and 27 ribosomal protein genes by Susoy et al. (2016). Asterisk represent node support of 100% posterior probability. Arrows represent species analysed in this study.



**Supplemental Figure S2.** (A-J) Gene expression levels are correlated with gene age . Transcriptome expression values for *P. pacificus* genes from different Age classes in all 10 RNA seq samples. (K) Heat map of FDR corrected P-values of pairwise Wilcoxon ranksum test for expression values between subsequent age classes in all samples.



**Supplemental Figure S3.** On y-axis we have the P values for Wilcoxon ransum test for  $d_S$ ,  $d_N$  and  $\omega$  distributions between *P. pacificus* and *P. exspectatus* orthologs of Age class ix and Age class i-viii mapped on the chromosomes of *P. pacificus*, in a non-overlapping window of 5 Mb. Here, we see that both  $d_N$  and  $\omega$  show significant difference between young and old genes in multiple windows and  $d_S$  is least different.

| Species                 | RNA sequencing experiments | Assembled Transcriptome Size (Mb) | Gene Count | Exon Length (Mb) | Intron Length (Mb) |
|-------------------------|----------------------------|-----------------------------------|------------|------------------|--------------------|
| <i>P. pacificus</i>     | 6                          | 44                                | 21,311     | 30               | 84                 |
| <i>P. expectatus</i>    | 6                          | 64                                | 31,172     | 39               | 90                 |
| <i>P. arcanus</i>       | 6                          | 71                                | 35,909     | 45               | 112                |
| <i>P. maxplancki</i>    | 1                          | 51                                | 31,765     | 43               | 109                |
| <i>P. japonicus</i>     | 1                          | 49                                | 31,996     | 40               | 93                 |
| <i>P. mayeri</i>        | 1                          | 62                                | 36,554     | 37               | 114                |
| <i>P. entomophagus</i>  | 1                          | 49                                | 37,279     | 38               | 107                |
| <i>P. fissidentatus</i> | 1                          | 45                                | 25,634     | 30               | 110                |
| <i>P. giblindavisi</i>  | 1                          | 36                                | 35,770     | 30               | 91                 |
| <i>M. japonica</i>      | 1                          | 60                                | 24,971     | 26               | 99                 |

**Supplemental Table S1.** The table gives an overview about the genome annotations in all 10 species. The first two columns indicate the number of RNA sequencing experiments and the size of the resulting transcriptome assembly (including isoforms). The last three columns show the number of final gene annotations along with their total exon and intron length.