

## Description of Additional Supplementary Files

File Name: Supplementary Data 1

Description: **EggNOG and Diamond Blast Annotation.**

File Name: Supplementary Data 2

Description: **Preprocessing parameter space exploration.**

UMAP visualisation of the 80,387 cells preprocessed with different parameters for minimum gene UMI counts (30-100), maximum number of genes (300-1000), maximum number of UMI counts (500-1200), number of top highly variable genes (4,000-22,000), number of neighbors (15-85), and number of principal components (15-145). For each parameter, UMAP feature plots of diagnostic markers PrileiEVm023936t1 (piwi+ cells), PrileiEVm008309t1 (epidermis), PrileiEVm011741t1 (muscle), PrileiEVm021316t1 (gut), PrileiEVm022250t1 (gut), PrileiEVm000325t1 (neurons), PrileiEVm013699t1 (eleocytes), and PrileiEVm020595t1 (unannotated).

File Name: Supplementary Data 3

Description: **Cluster Wilcoxon markers and Diamond Blast annotations.**

Marker genes per cluster calculated using the Wilcoxon method with maximum p-values of 0.05.

File Name: Supplementary Data 4

Description: **Cluster Logistic Regression markers and Diamond Blast annotations.**

Top 30 markers per cluster calculated using the Logistic Regression method.

File Name: Supplementary Data 5

Description: **Pristina leidy cell cluster markers.**

UMAP visualisation of 75,218-cell Pristina leidy single-cell transcriptomic cell atlas. In the top left panel of each page each cluster is coloured. In the remaining plots, UMAP feature plots of markers of the top 8 common markers among the Wilcoxon and the Logistic Regression methods. If there are less than 8 common markers the plots only display the UMAP visualisation without any feature gene expression.

File Name: Supplementary Data 6

Description: **HCR in situ probes.**

File Name: Supplementary Data 7

Description: **WGCNA modules.**

File Name: Supplementary Data 8

Description: **WGCNA modules GO terms.**

File Name: Supplementary Data 9

Description: **Transcription Factor annotations.**

File Name: Supplementary Data 10

Description: **piwi+ cells COGs.**

File Name: Supplementary Data 11

Description: **Top 200 TFs expressed in piwi+ cells.**

Expression (in log-ratio of counts in a given broad cell type vs piwi+ cells) of the top 200 variable (CV > 1) transcription factors in *Pristina leidyi*; as well as the top hit in the DIAMOND annotation. See Methods for further details.

File Name: Supplementary Data 12

Description: **Top 200 TFs expressed in piwi+ cells UMAP visualisation.**

File Name: Supplementary Data 13

Description: **Transcriptomic profile of piwi+ cells and Diamond Blast annotations.**

File Name: Supplementary Data 14

Description: **Annotation of *Pristina* epigenetic factor homologues.**