

# Identify *D. melanogaster* ortholog

FlyBase *blastp* search of predicted protein against the *D. melanogaster* “**Annotated Proteins**” database

**Consult instructor**

Potential novel gene

Probable misprediction

Yes

No

Has at least one significant match (E-value < 1e-5)?

No

NCBI *blastp* search of predicted protein against the **nr protein** database

Has at least one significant match (E-value < 1e-5)?

Matches multiple *D. melanogaster* genes?

No

Assign the only *D. melanogaster* match as the putative ortholog

Multiple matches with E-values similar to the best match?

No

Assign gene with the **lowest E-value** as the putative ortholog

Multiple matches with sequence identity similar to the best match?

No

Assign gene with the **highest sequence identity** as the putative ortholog

Multiple matches with alignment coverage similar to the best match?

No

Assign gene with the **highest alignment coverage** as the putative ortholog

Multiple matches located on the same Muller element as the best match?

No

Assign *D. melanogaster* gene located on the **same Muller element** as the contig in the target species as the putative ortholog

Multiple genes within the orthologous region in *D. melanogaster* (as defined by the placements of the adjacent genes)

No

Assign putative ortholog based on **synteny** with *D. melanogaster*

Yes

**Consult instructor**