

GEP Annotation Workflow

Select gene prediction

Click on gene prediction and the “**Predicted Protein**” link

Identify *D. melanogaster* ortholog (workflow)

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

Review *D. melanogaster* gene structure

Determine number of isoforms and unique coding exons (CDS):

- *Gene Record Finder* tables
- Genome Browsers (e.g., *JBrowse*)

Select isoform to annotate

- Obtain *D. melanogaster* CDS sequence from *Gene Record Finder*
- Use NCBI **blastx** → **Align two or more sequences** to compare contig against CDS
 - ❖ Record **start** and **end** positions, **coverage**, and **reading frame**

Map each coding exon (CDS) to contig

- Identify the best supported donor (**GT/GC**) and acceptor sites (**AG**)
- Minimize changes compared to *D. melanogaster*

Determine CDS coordinates (workflow)

Provide explanations for:

- Errors and warnings in checklist
- **Large gaps in dot plot**
- **Gaps near splice sites**

Verify isoform model

UCSC Genome Browser

Gene Record Finder

Gene Model Checker

FlyBase

NCBI BLAST (*bl2seq*)

Repeat for each isoform