

Functional annotation in Apollo

There are two options to add function annotation in VEuPathDB:

- 1) Adding a user comment on the gene record page
- 2) Using the community annotation tool Apollo

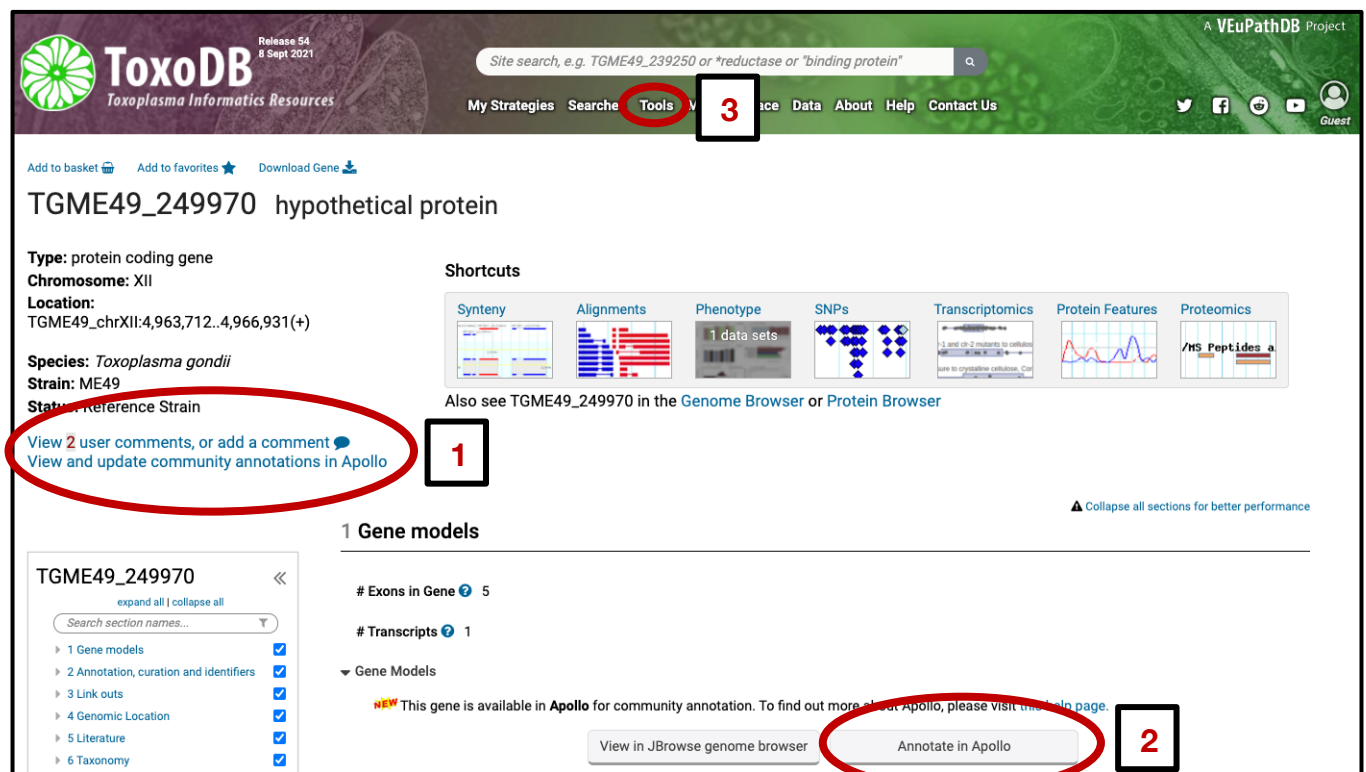
Functional annotation can involve:

- Adding or changing the description of a gene or product
- Assigning or changing a gene name/symbol
- Adding Gene Ontology (GO) terms
- Adding a publication
- Adding an EC number

In this manual we are showing you step by step how to add a gene name, product description and GO term to a gene in Apollo.

1) Accessing Apollo

To access Apollo go to the gene record page of your gene of interest and click on the link **View and update community annotations in Apollo** (1). You can also access Apollo from the gene models section by clicking on the button **Annotate in Apollo** (2). Alternatively, go to the **Tools** menu and choose Apollo from the drop-down list (3).

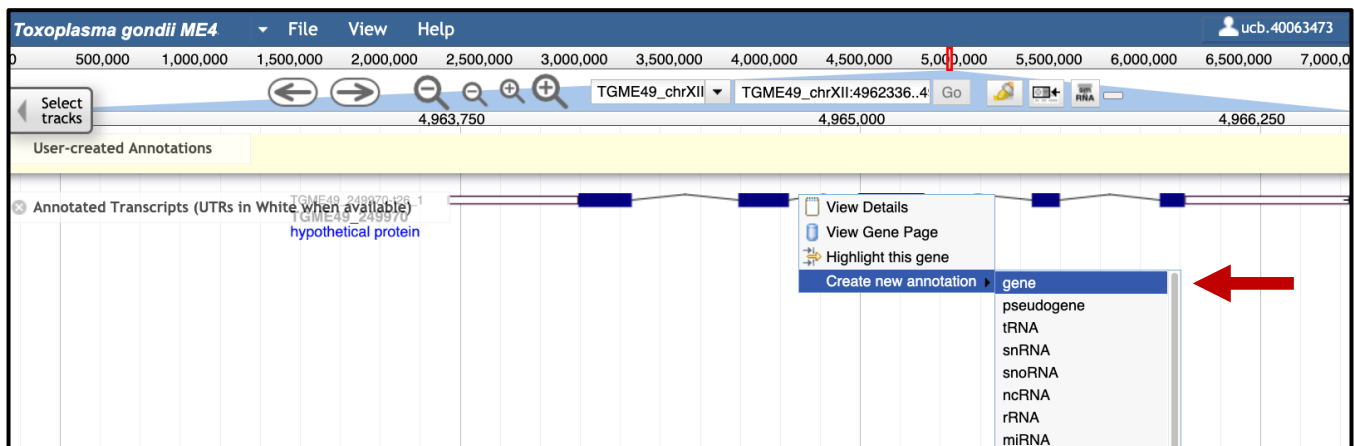


The screenshot displays the ToxoDB website interface. At the top, the ToxoDB logo and navigation menu are visible. The main content area shows the gene record for TGME49_249970, a hypothetical protein. A red circle highlights the link "View and update community annotations in Apollo" (1). Another red circle highlights the "Annotate in Apollo" button in the "Gene models" section (2). A third red circle highlights the "Tools" menu in the header (3).

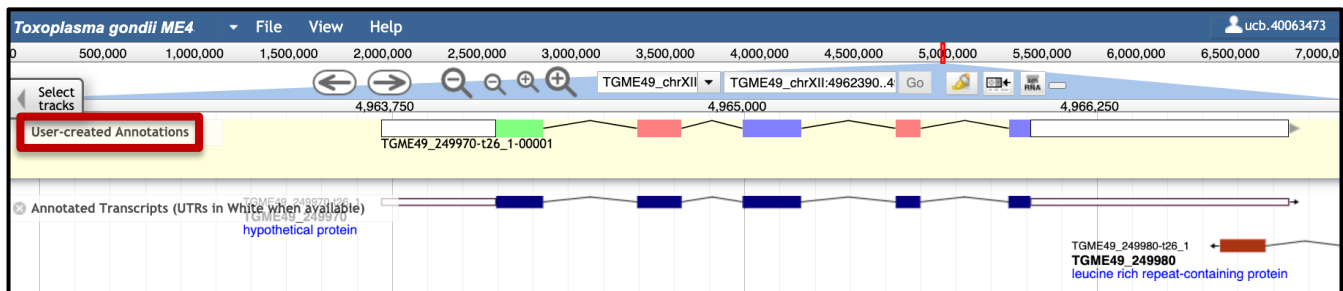
To use Apollo you need to be logged into VEuPathDB. If you have not done so yet log now into Apollo with your VEuPathDB user ID and password.

2) Dragging and dropping the annotation

Right click on the gene in the Annotated Transcripts track, select from the drop-down menu **Create new annotation > gene**.

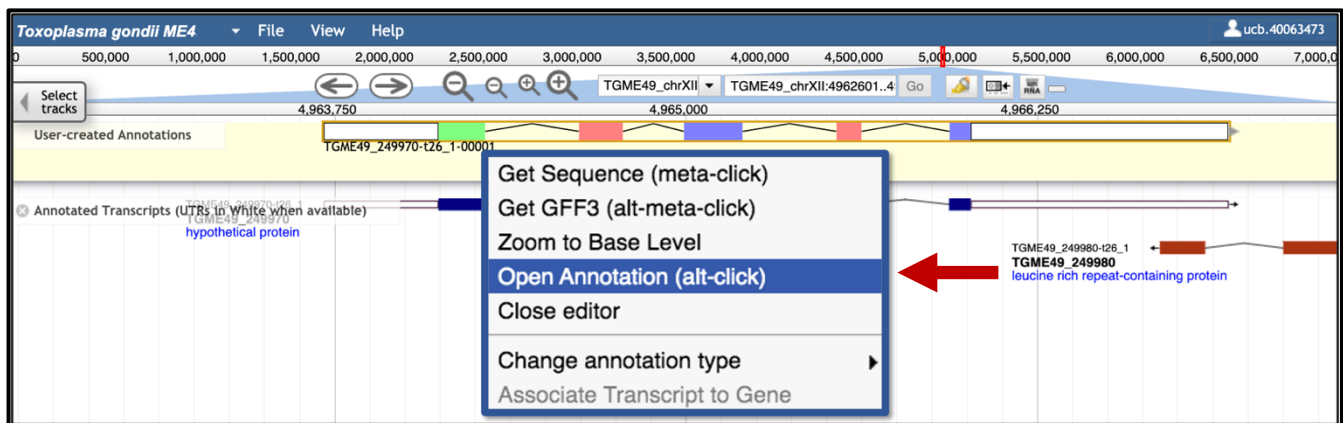


You can now see your gene of interest in the User-created Annotations track.



3) Opening of the Annotation editor window

Select the gene in the User-created Annotations track and with a right-click open the drop-down menu and choose **Open Annotation**. Alternatively, you can use the short-cut **alt-click**.



The annotation editor window is now shown on the right-hand side.

Annotation editor →

Annotations Tracks Ref Sequence Organism Users Groups Admin

Show All Show Visible Only

0cecc08e-205b-405f-8199- ID All Types GO GP Prov

Reference Sequence All Users All Statuses

Rows 25 1-1 of 1

Name	Seq	Type	Length	Updated
TGME49_249970-l26_1	TGME49_chrXII	gene	3,220	May 27, 2021
TGME49_249970-l26_1-00001		mRNA	3,220	May 27, 2021

gene: TGME49_249970-l26_1 Link to annotation Close(x)

Details GO Gene Product Provenance DbXref Comment Attributes

Go ID Sync name with transcript Obsolete Delete

Type gene Status No status select

Name TGME49_249970-l26_1

Symbol

Aliases (! separated)

Description

Location 4963712 - 4966931 strand(+) Partial: ☐ 3' ☐ 5'

Ref Sequence TGME49_chrXII

Owner ucb.40063473

Created May 27, 2021 03:13 PM

Updated May 27, 2021 03:13 PM

You can either select the gene or the mRNA.

Please note functional annotation should be added to the gene. For genes with alternative transcripts add the information to the mRNA.

4) Adding Functional annotation

In this example we are showing you how to improve the functional annotation of TGME49_249970. This gene is currently annotated as hypothetical protein. It has been experimentally characterised in the following publication:

<https://pubmed.ncbi.nlm.nih.gov/26962945/>

Gene name/symbol: APH

Gene product: Acylated pleckstrin-homology domain-containing protein

GO term: microneme

PMID: 26962945

Adding a gene name/symbol

Once the annotations panel (1) is open click on the details tab (2) and add the gene name in the field Symbol (3). In our example the new gene name/symbol is APH.

🔍

Toxoplasma gondii ME49 [Mar 22, ▾]

ucb.40063473

🔄

Annotations

1

Ref Sequence

Organism

Users

Groups

Admin

Show All

Show Visible Only

0cecc08e-205b-405f-8199-

☒ ID

All Types ▾

🔼

GO

GP

Prov

Reference Sequence

All Users ▾

All Statuses ▾

Rows 25 ▾

⏪

⏩

1-1 of 1

⏪

⏩

Name	Seq	Type	Length	Updated
TGME49_249970-t26_1	TGME49_chrXII	gene	3,220	May 27, 2021 ▾
TGME49_249970-t26_1-00001		mRNA	3,220	May 27, 2021

gene: TGME49_249970-t26_1

[Link to annotation](#)

[Close\(x\)](#)

Details

2

Gene Product

Provenance

DbXref

Comment

Attributes

🔍 Go

👤 ID

🔄 Sync name with transcript

☐ Obsolete

Delete

Type

gene

Status

No status select ▾

Name

TGME49_249970-t26_1

Symbol

APH

Aliases ('|' separated)

Description

Location

4963712 - 4966931 strand(+)

Partial: ☐ 3' ☐ 5'

Ref Sequence

TGME49_chrXII

Owner

ucb.40063473

Created

May 27, 2021 03:13 PM

Updated

May 27, 2021 03:13 PM

Adding a product description

To add a product description with metadata choose the tab Gene Product (1) and click on **New** at the bottom of the editor window (2).

Gene Product editor window for TGME49_249970-t26_1. The 'Gene Product' tab is selected and highlighted with a red box and a '1' in a black box. At the bottom left, the 'New' button is highlighted with a red box and a '2' in a black box. The window shows gene and mRNA details, a table with columns Name, Evidence, Based On, and Reference, and buttons for Details, GO, Gene Product, Evidence, DbXref, Comment, and Attributes.

Fill in the fields product, evidence and PMID. Click on Save. More information about evidence codes can be found here: <http://geneontology.org/docs/guide-go-evidence-codes>

Add new Gene Product to TGME49_249970-t26_1

GO Annotation Guidance

Product: Acylated pleckstrin-homology dor

Evidence: ECO:0000314

PMID: 26962945

Save

Please note, if possible always add a product description with metadata to the Gene Product tab (1). If you don't have any metadata, for example a publication you can add the product description in the field Description (2) in the Details tab. Don't add the product to both, the Gene Product tab and the Details tab.

gene: TGME49_249970-t26_1

Link to annotation Close(x)

Details GO **Gene Product** 1 DbXref Comment Attributes

Go ID Sync name with transcript Obsolete Annotations Delete

Type gene Status Finished

Name TGME49_249970-t26_1

Symbol APH

Aliases (| separated)

Description 2

Location 4963719 - 4966931 strand(+) Partial: ☐ 3' ☐ 5'

Adding GO terms

Choose the tab GO in the editor window, fill in the required fields and click on Save.

Add new GO Annotation to TGME49_249970-t26_1

GO Annotation Guidance

Aspect CC cellular component

Go Term GO:0020009 *microneme* (GO:0020009)

Relationship between Gene Product and GO Term part of

Evidence ECO:0000314 *IDA (ECO:0000314): direct assay evidence used in manual assertion Evidence Code Info*

With Prefix : ID + Add

Reference PMID : 26962945

Note + Add

Save Cancel

Adding a PubMed ID

Choose the Tab DbXref in the annotation editor window, add the PMID as shown in the screenshot. Click on the + sign.

The screenshot shows the 'DbXref' tab selected in the annotation editor. The window title is 'gene: TGME49_249970-t26_1'. The 'DbXref' tab is highlighted with a red box. Below the tab, there are two input fields: 'PMID' and 'Accession'. The 'PMID' field contains the value '26962945'. A red circle with a plus sign is positioned next to the 'PMID' field, indicating where to click to add the entry.

A small window will come up showing the title of the Pubmed Article. Click OK.

The screenshot shows a small window with the title 'Add article Phosphatidic Acid-Mediated Signaling Regulates Microneme Secretion in Toxoplasma.' The window has a close button (X) in the top right corner. At the bottom right, there are two buttons: 'Cancel' and 'OK'. The 'OK' button is highlighted with a red circle, indicating where to click.

You can also add additional database identifiers (DbXref), i.e. EC numbers.

The screenshot shows the 'DbXref' tab with multiple entries. The 'EC' field contains the value '2.7.1.1'. The 'PMID' field is empty. A red circle with a plus sign is positioned next to the 'EC' field, indicating where to click to add the entry.

5) Finalising the functional annotation

Go to the **Attributes** tab in the gene section, choose from the “Select canned tag” drop-down menu **annotation**.

The screenshot shows the 'Attributes' tab selected in the annotation editor. The window title is 'gene: TGME49_249970-t26_1'. The 'Attributes' tab is highlighted with a red box. Below the tab, there are two input fields: 'Tag' and 'Value'. The 'Tag' field has a dropdown menu open, showing options: 'Select canned tag', 'structural', 'user_comment', and 'annotation'. The 'annotation' option is highlighted with a blue background and a red arrow points to it. The 'Value' field is empty. A red circle with a plus sign is positioned next to the 'Value' field, indicating where to click to add the entry.

From the “Select canned value” drop-down menu choose **added_product**. Repeat this and choose **added_symbol**, **added_go** and **added_pmid**. Finally click on the + sign.

Details GO Gene Product Provenance DbXref Comment **Attributes**

Prefix Accession

annotat Value

annotation

- ✓ Select canned value
- added_comment
- split
- merge
- retain_previous
- added_product**
- added_go
- added_symbol
- added_alias
- added_pmid
- removed_product
- added_ec_number
- delete
- isoform
- new
- modify
- added_dbxref

gene: TGME49_249970-t26_1 [Link to annotation](#) [Close\(x\)](#)

Details GO Gene Product Provenance DbXref Comment **Attributes**

Prefix Accession

Tag Value

Select can Select can

annotation	added_symbol
annotation	added_go
annotation	added_product
annotation	added_pmid

In the last step, go back to the Details tab and add to the gene and mRNA the status **Finished**.

gene: TGME49_249970-t26_1 [Link to annotation](#) [Close\(x\)](#)

Details GO Gene Product Provenance DbXref Comment Attributes

Go ID Sync name with transcript ☐ Obsolete [Delete](#)

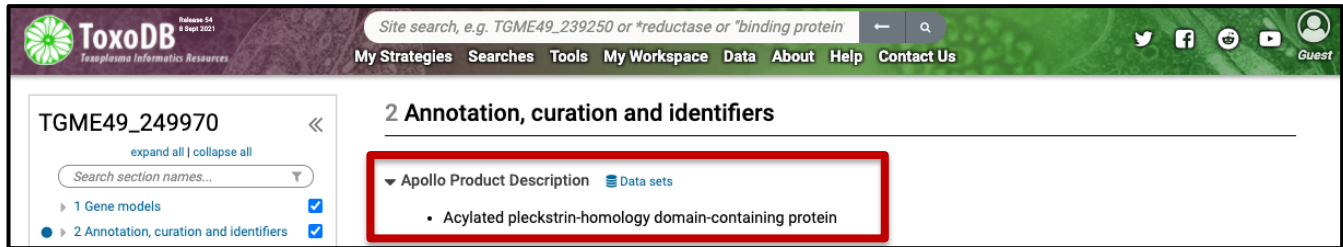
Type gene Status

Name TGME49_249970-t26_1

Symbol APH

✓ No status selected
Not Finished
Finished
Requires Curator

The new Apollo product description can be viewed the following day on the gene record page in the Annotation, curation and identifiers section.

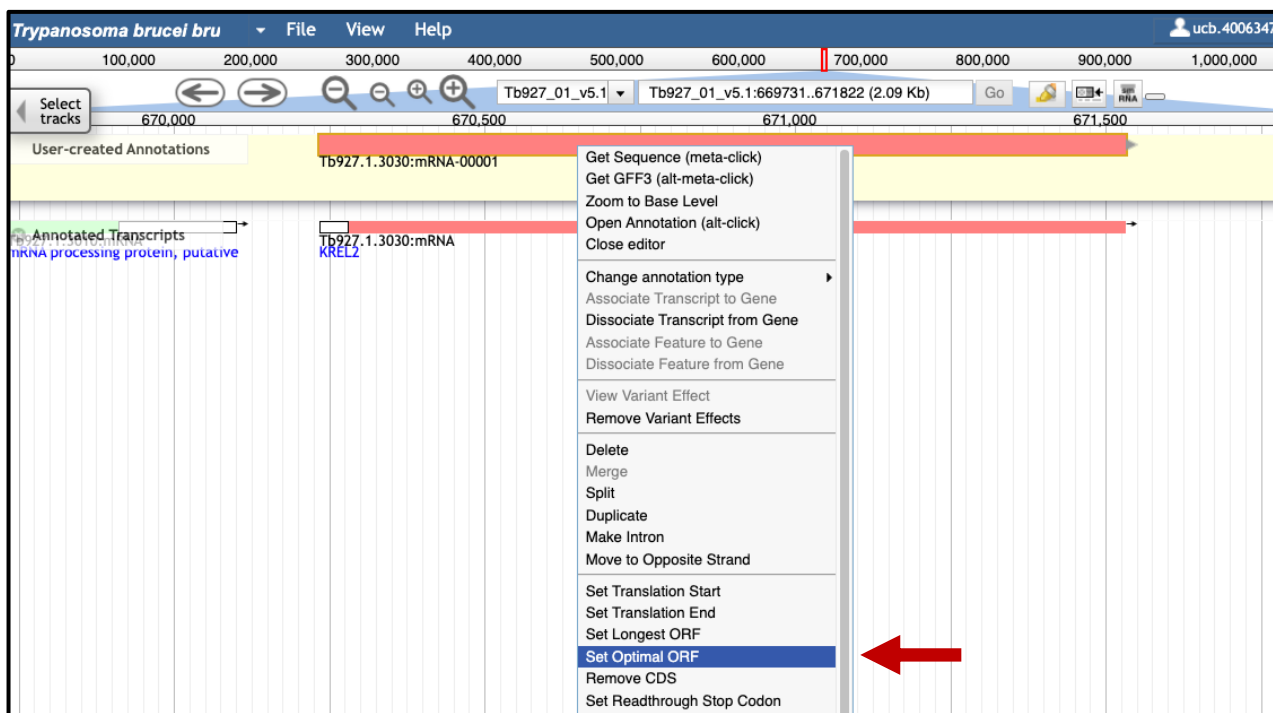


The screenshot shows the ToxoDB website interface. The top navigation bar includes the ToxoDB logo, a search bar, and links for My Strategies, Searches, Tools, My Workspace, Data, About, Help, and Contact Us. The main content area is titled '2 Annotation, curation and identifiers'. A red box highlights the 'Apollo Product Description' section, which contains the text 'Acylated pleckstrin-homology domain-containing protein'.

Done! For additional questions, please get in touch with the VEuPathDB help desk.

Additional note

On rare occasions when dragging and dropping a gene model in the User-created Annotations track, the gene model automatically changes and ends up without a correct start. To correct this right-click on the gene model in the User-created Annotations track and select from the menu **Set Optimal ORF**. This will correct the start.



The screenshot shows the Apollo genome browser interface. A right-click context menu is open over a gene model in the 'User-created Annotations' track. The menu includes options such as 'Get Sequence (meta-click)', 'Get GFF3 (alt-meta-click)', 'Zoom to Base Level', 'Open Annotation (alt-click)', 'Close editor', 'Change annotation type', 'Associate Transcript to Gene', 'Dissociate Transcript from Gene', 'Associate Feature to Gene', 'Dissociate Feature from Gene', 'View Variant Effect', 'Remove Variant Effects', 'Delete', 'Merge', 'Split', 'Duplicate', 'Make Intron', 'Move to Opposite Strand', 'Set Translation Start', 'Set Translation End', 'Set Longest ORF', 'Set Optimal ORF', 'Remove CDS', and 'Set Readthrough Stop Codon'. A red arrow points to the 'Set Optimal ORF' option.