OrthoMCL 7

Map Proteins to OrthoMCL with Diamond blastp: A Tutorial[1]

Learning Objectives

- Understand the purpose of the OrthoMCL protein mapping tool

- Learn how to prepare and upload sets of proteins for mapping

- Explore the output and understand the DIAMOND job result page

Introduction

- OrthoMCL is a genome-scale algorithm that uses protein sequence similarity and phylogenetic relationships to create groups of orthologous protein sequences both within and across species. OrthoMCL includes all VEuPathDB species plus additional Core species that broadly represent the diversity across the Tree of Life.
- **Purpose**: The protein mapping tool allows users to **map a set of proteins of interest**, usually a complete proteome from an organism, to existing OrthoMCL groups.
- This tool uses **DIAMOND blastp**, an alternative to NCBI BLAST which is 10,000 times faster while being only 0.1- 1% less sensitive.

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[1] Updated on April 18, 2025

- Access the tool from the **Tools** menu in the header > **Map proteins to OrthoMCL with DIAMOND blastp** (red arrow below)
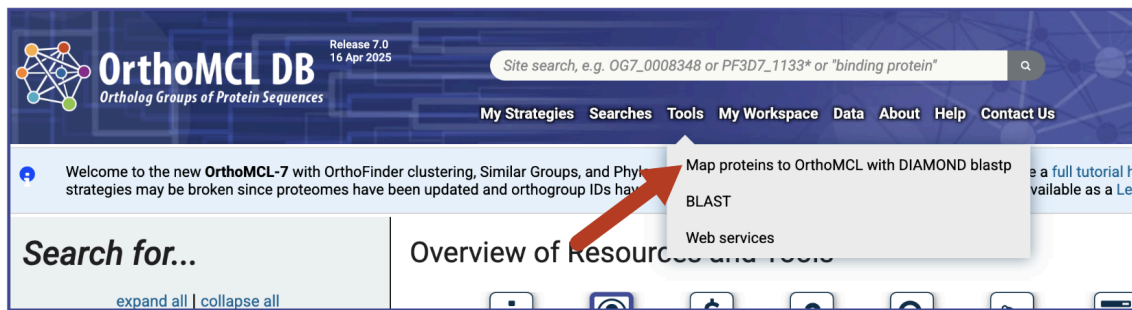
Layout of the DIAMOND blastp protein mapping page

There are **three tabs** (circled in red)

- **All**: A list of all of your previous jobs. These are saved in your account and persist across visits to the website
- **New job**: Interface that allows you to upload a FASTA formatted file of protein sequences
- **Help**: Tips for using the tool
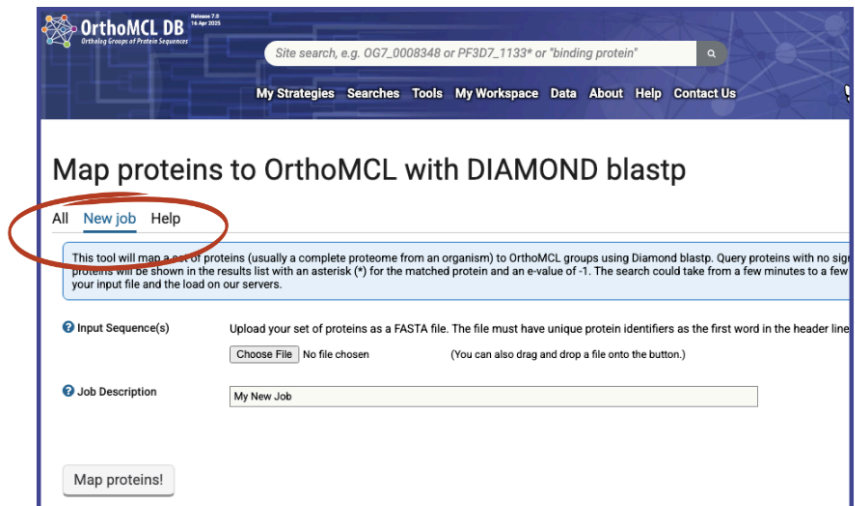
Preparing your data: Your set of proteins must be formatted as a plain text FASTA file.

- The single-line description/header for each protein sequence in the FASTA file must begin with a unique protein identifier.
- Header line must start with a greater than (>) symbol and end with a carriage return.

The figure on the right shows a properly formatted FASTA file.

Uploading data: Do the following steps in the "New job" tab (refer to figure below)

- *Input sequence(s)*: Choose a FASTA-formatted data file with protein sequences from your computer
- *Job description***:** Add brief text describing your set of proteins
- **Start the job**: Click on the *Map proteins!* button to start the job. You will see a message with a job ID assignment.

Understanding the output: The output page has two components

- **The results table** (see below). This is a preview of the matching results for the first 100 sequences in your query file.
- **A blue download button** at the top right (see red arrow below). The complete result can be downloaded as a tab delimited file (tsv) with one best match for each query protein with the following columns:
    - **Query sequence id**: the identifier for the sequence in your input file
    - **Subject sequence id:** the identifier for the best matching OrthoMCL sequence
    - **OrthoMCL group id**: the orthogroup containing the best matching OrthoMCL sequence
    - **Description**: description of the best matching OrthoMCL sequence
    - **Alignment length**: length of the aligned region between Query and Subject sequences
    - **Percent identical matches**: percent identity between Query and Subject sequences
    - **Expect value**: BLAST significance score for the alignment between Query and Subject sequences. The expect value (E-value) cutoff is 0.05, allowing you to filter the output file more stringently if required.

Note: Unmatched query proteins (no significant match) are included in the results file without an OrthoMCL protein or group listed. For example, see the red rectangle below.

DIAMOND Mapping Job - result

<< All my DIAMOND Mapping Jobs

Job Id: ca0b3bc15bc76c8b779cab30c37f2ddd
Description: My New Job
Program: diamond-blastp

Showing all 75 sequences in your query file.

Download as a tsv file

Query sequence id	Subject sequence id	OrthoMCL group id	Description	Alignment length	Percent identical matches	Expect value
g1.t1	tpse|B8LDJ4	OG7_0006976	gene=B8LDJ4 product=Radical SAM core domain-containing protein (Fragment)	305	51.8	1.65e-94
g2.t1	tpse|B8BVT1	OG7_0001781	gene=B8BVT1 product=Methyltransferase type 11 domain-containing protein	268	57.5	1.85e-98
g4.t1	vbra|Vbra_363	OG7_0001280	gene=Vbra_363 product=unknown	120	26.7	1.49e-04
g5.t1	aalf|AALF000687	OG7_0001280	gene=AALF000687 product=unknown	324	26.5	1.39e-20
g6.t1	tsti|TSTA_009530	OG7_0001280	gene=TSTA_009530 product=RNA-directed DNA polymerase [Source:UniProtKB/TrEMBL;Acc:B8MFW4]	103	35.9	5.32e-03
g7.t1	tpse|B8BU27	OG7_0009784	gene=B8BU27 product=Orc1-like AAA ATPase domain-containing protein	615	27.0	1.40e-69
g8.t1	tpse|B8LDV1	OG7_0009784	gene=B8LDV1 product=Orc1-like AAA ATPase domain-containing protein (Fragment)	481	26.0	2.65e-44
g9.t1	tpse|B8C6X7	OG7_0020726	gene=B8C6X7 product=DUF1995 domain-containing protein	341	60.7	9.37e-146
g9.t2	tpse|B8C6X7	OG7_0020726	gene=B8C6X7 product=DUF1995 domain-containing protein	341	60.7	1.26e-145
g10.t2	tpse|B8BWV7	OG7_0003575	gene=B8BWV7 product=Major facilitator superfamily (MFS) profile domain-containing protein (Fragment)	399	48.4	2.50e-114
g10.t1	tpse|B8BWV7	OG7_0003575	gene=B8BWV7 product=Major facilitator superfamily (MFS) profile domain-containing protein (Fragment)	399	48.4	1.68e-114
g11.t1	tsti|TSTA_111600	OG7_0001280	gene=TSTA_111600 product=RNA-directed DNA polymerase [Source:UniProtKB/TrEMBL;Acc:B8M929]	175	29.1	8.19e-08
g12.t1	*	-1	-1	-1	N/A	N/A
g13.t1	aalf|AALF006108	OG7_0001280	gene=AALF006108 product=unknown	684	23.4	4.99e-33

Questions? Comments?

Contact us- help@veupathdb.org