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Find Proteins of Unknown Function (PUFs) using Plantannot - Protocol F

Forked from Find Proteins of Unknown Function (PUFs) using Plantannot - Protocol E

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External link: https://www.machado.cnptia.embrapa.br/plantannot

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Protocol status: Working
It is working

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Abstract

The Plant annotation software provides several filters and a text search box that allows searching for molecules by its desired annotation features. These filters are needed to obtain PUFs and to try to relate them to abiotic stresses using RNA-seq expression data and co-expression networks.

The Filters menu is separated in 8 fields, of those we are going to use only five: “Organism”, “Feature type”, “Orthology”, “Orthologs_coexpression” and “Analyses”. The “Feature Type” filter has three molecule types, from those the polypeptide box is the only that is going to be always checked and the others

blank. By using the other 4 remaining filters, 6 protocols were created as examples of different ways to selecting PUFs. Protocol A: using lack of both homology and protein domain signatures. Protocol B: using lack of homology, presence of domain signatures - trying to select Domains of Unknown Function

(DUF) from PFAM, and the text search “Unknown function”.Protocol C: using homology, lack of protein domain signatures and the text search “Unknown function”. Protocol D-F: same protocols of A-C but using ortholog groups to find homolog proteins with co-expression data related to abiotic stress.

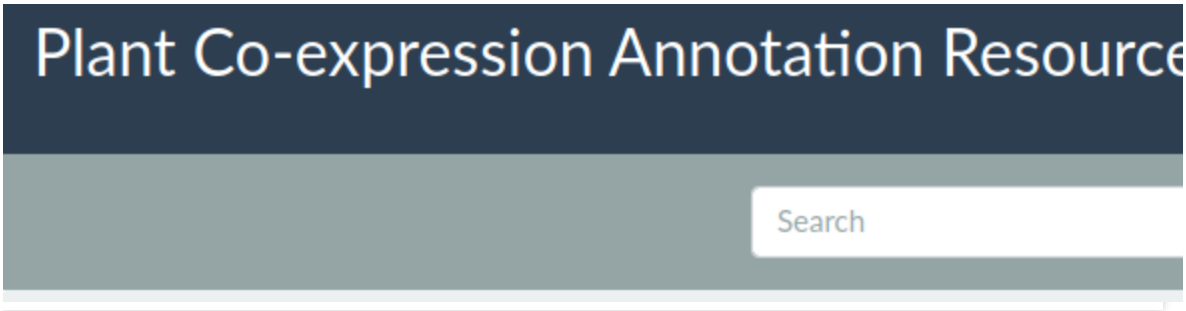
Protocol F is intended to find PUFs from organisms that proteins are already public in the NCBI´s "nr" database and have no protein domain found by Interproscan. Proteins will be selected using the text search "Unknown function". Also, ortholog groups and co-expression networks will be

used to relate proteins to abiotic stresses.

Entering application

1 Enter the Plantannot Result's page, with empty filters and text box search:
https://www.machado.cnptia.embrapa.br/plantannot/find/?q=

Or you can enter the **https://www.machado.cnptia.embrapa.br/plantannot** initial page and click on the magnifying glass with the text box empty as well.



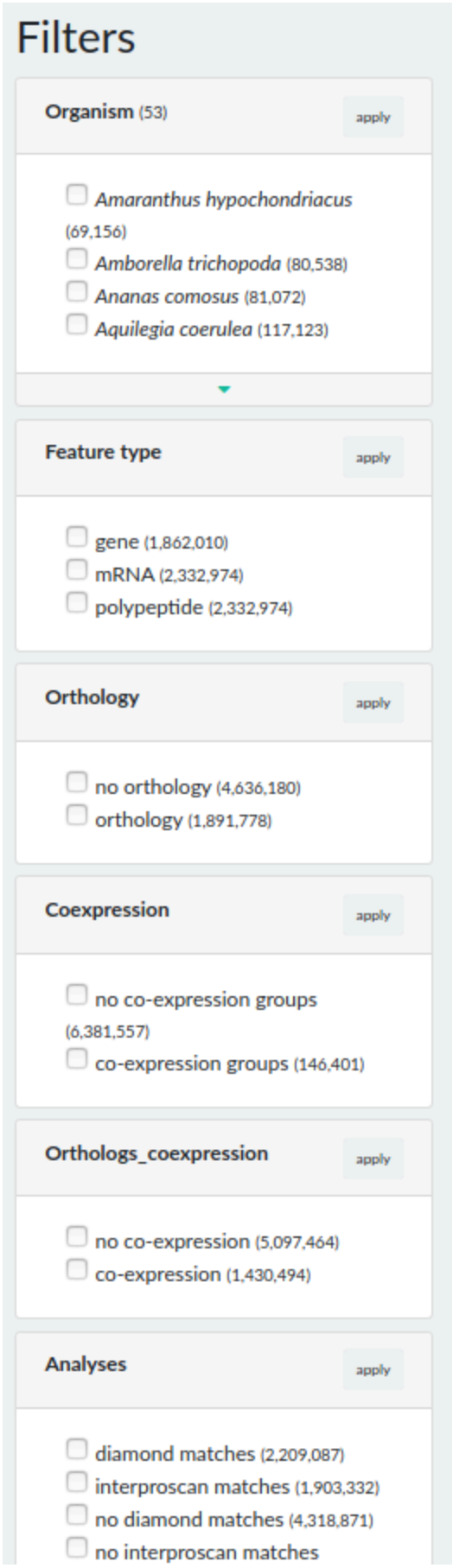
https://www.machado.cnptia.embrapa.br/plantannot

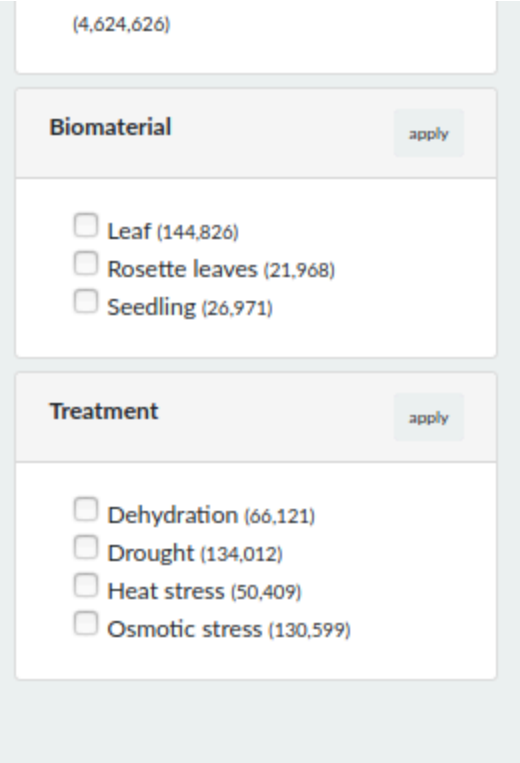
Filtering

2

Find PUFs from organisms that proteins are already public in the NCBI´s "nr" database and have no protein domain found by Interproscan. Proteins will be selected using the text search "Unknown function". Also, ortholog groups and co-expression networks will be used to relate proteins to abiotic stresses.

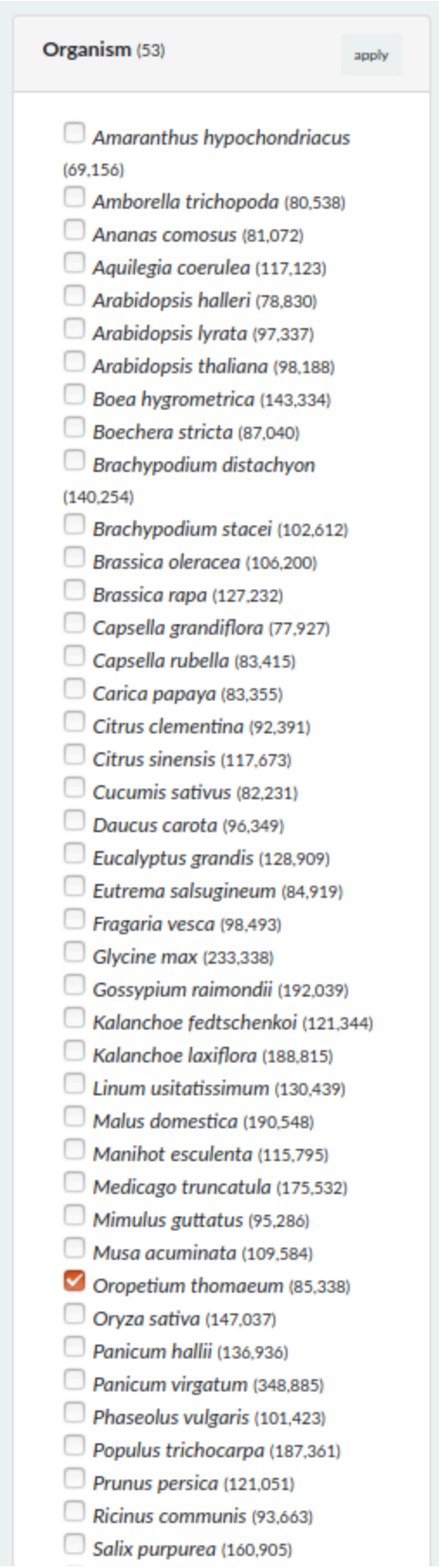
Visualize the "Filters" card on the left of the page from step1:

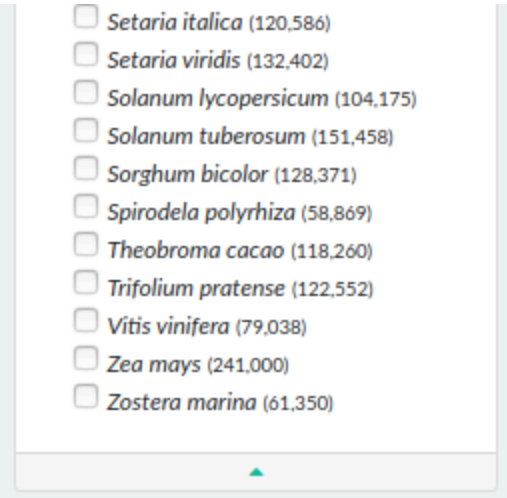

Filters
Organism (53)
apply
Amaranthus hypochondriacus (69,156)
Amborella trichopoda (80,538)
Ananas comosus (81,072)
Aquilegia coerulea (117,123)
Feature type
apply
gene (1,862,010)
mRNA (2,332,974)
polypeptide (2,332,974)
Orthology
apply
no orthology (4,636,180)
orthology (1,891,778)
Coexpression
apply
no co-expression groups (6,381,557)
co-expression groups (146,401)
Orthologs_coexpression
apply
no co-expression (5,097,464)
co-expression (1,430,494)
Analyses
apply
diamond matches (2,209,087)
interproscan matches (1,903,332)
no diamond matches (4,318,871)
no interproscan matches



https://www.machado.cnptia.embrapa.br/plantannot/find/?q=

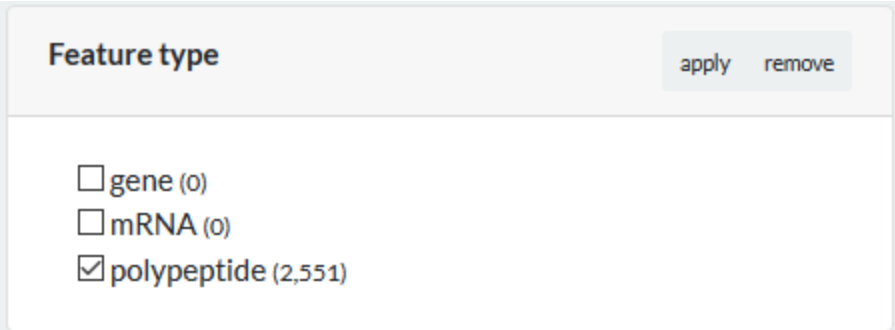
2.1 In the "Organisms" filter, select any organisms (expand the organism's list using the green arrow) or select all by leaving all boxes empty. We will use *Oropetium tomaeum* as example. Click "apply" to execute the filter:


Organism (53)
apply
Amaranthus hypochondriacus (69,156)
Amborella trichopoda (80,538)
Ananas comosus (81,072)
Aquilegia coerulea (117,123)
Arabidopsis halleri (78,830)
Arabidopsis lyrata (97,337)
Arabidopsis thaliana (98,188)
Boea hygrometrica (143,334)
Boechera stricta (87,040)
Brachypodium distachyon (140,254)
Brachypodium stacei (102,612)
Brassica oleracea (106,200)
Brassica rapa (127,232)
Capsella grandiflora (77,927)
Capsella rubella (83,415)
Carica papaya (83,355)
Citrus clementina (92,391)
Citrus sinensis (117,673)
Cucumis sativus (82,231)
Daucus carota (96,349)
Eucalyptus grandis (128,909)
Eutrema salsugineum (84,919)
Fragaria vesca (98,493)
Glycine max (233,338)
Gossypium raimondii (192,039)
Kalanchoe fedtschenkoi (121,344)
Kalanchoe laxiflora (188,815)
Linum usitatissimum (130,439)
Malus domestica (190,548)
Manihot esculenta (115,795)
Medicago truncatula (175,532)
Mimulus guttatus (95,286)
Musa acuminata (109,584)
Oropetium thomaeum (85,338)
Oryza sativa (147,037)
Panicum hallii (136,936)
Panicum virgatum (348,885)
Phaseolus vulgaris (101,423)
Populus trichocarpa (187,361)
Prunus persica (121,051)
Ricinus communis (93,663)
Salix purpurea (160,905)



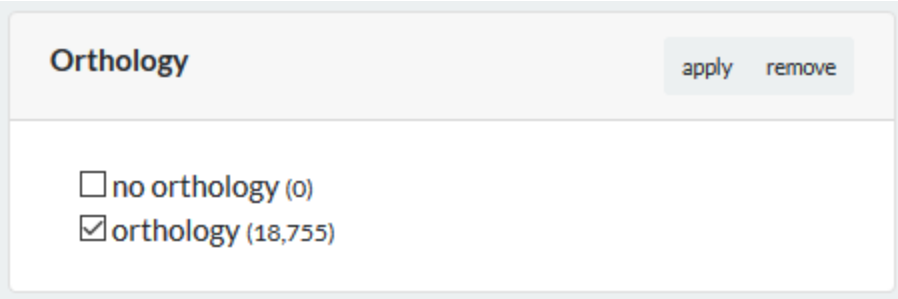
https://www.machado.cnptia.embrapa.br/plantannot/find/?q=&selected_facets=organism%3AOropetium+thomaeum

2.2 In the "Feature type" filter, select "polypeptide", and click "apply" to execute the filter:



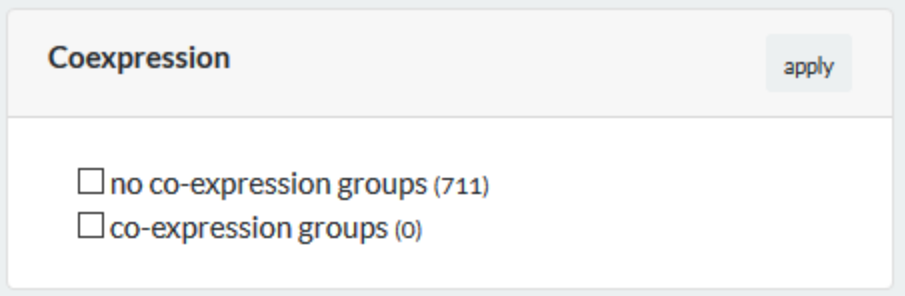
https://www.machado.cnptia.embrapa.br/plantannot/find/?q=&selected_facets=organism%3AOropetium+thomaeum&selected_facets=so_term%3Apolypeptide

2.3 In the Orthology filter, select "orthology", and click "apply" to execute the filter:



https://www.machado.cnptia.embrapa.br/plantannot/find/?q=&selected_facets=organism%3AOropetium+thomaeum&selected_facets=so_term%3Apolypeptide&selected_facets=orthology%3A1

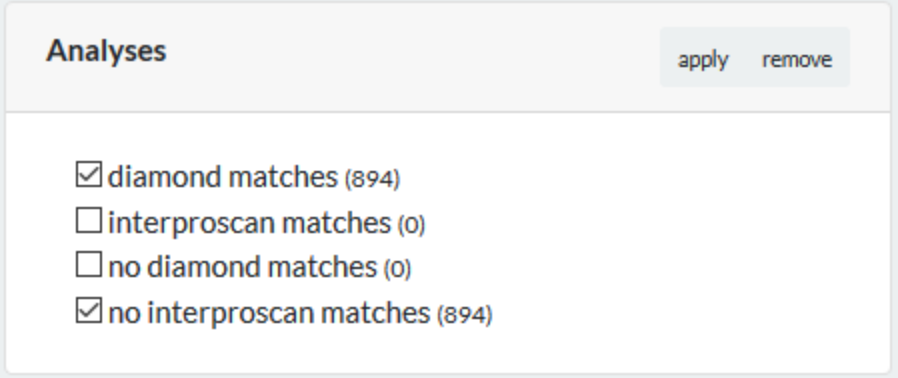
2.4 Leave the "Coexpression" filter empty:



2.5 In the "Orthologs_coexpression" filter, select "co-expression", and click "apply" to execute the filter:

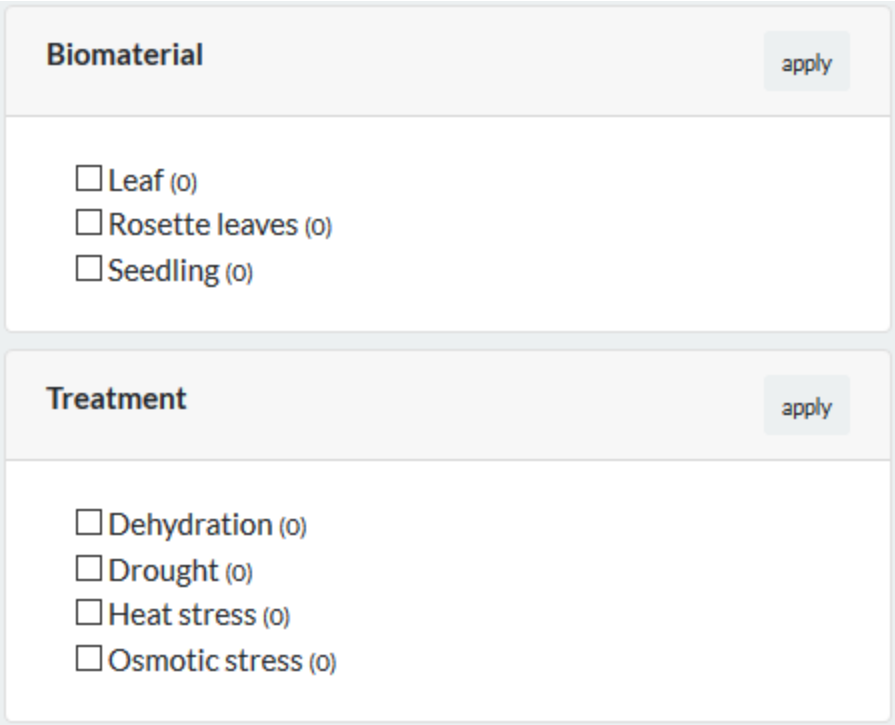
https://www.machado.cnptia.embrapa.br/plantannot/find/?q=&selected_facets=organism%3AOropetium+thomaeum&selected_facets=so_term%3Apolypeptide&selected_facets=orthology%3A1&selected_facets=orthologs_coexpression%3Atrue

2.6 In addition to the applied filters, text search for "Unknown function" in the search box. After entering the text click on the magnifying glass icon to apply the search.

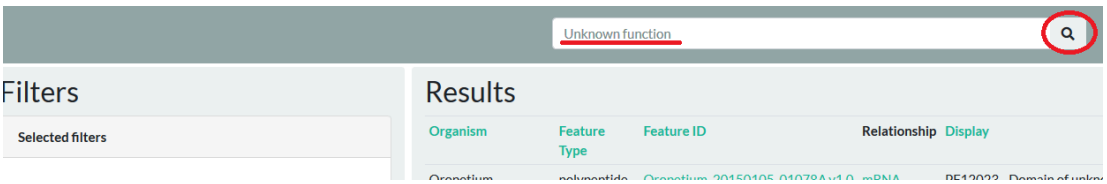


https://www.machado.cnptia.embrapa.br/plantannot/find/?q=&selected_facets=organism%3AOropetium+thomaeum&selected_facets=so_term%3Apolypeptide&selected_facets=orthology%3A1&selected_facets=orthologs_coexpression%3Atrue&selected_facets=analyses%3Adiamond+matches&selected_facets=analyses%3Ano+interproscan+matches

2.7 Leave the "Biomaterial" and "Treatment" filters empty:



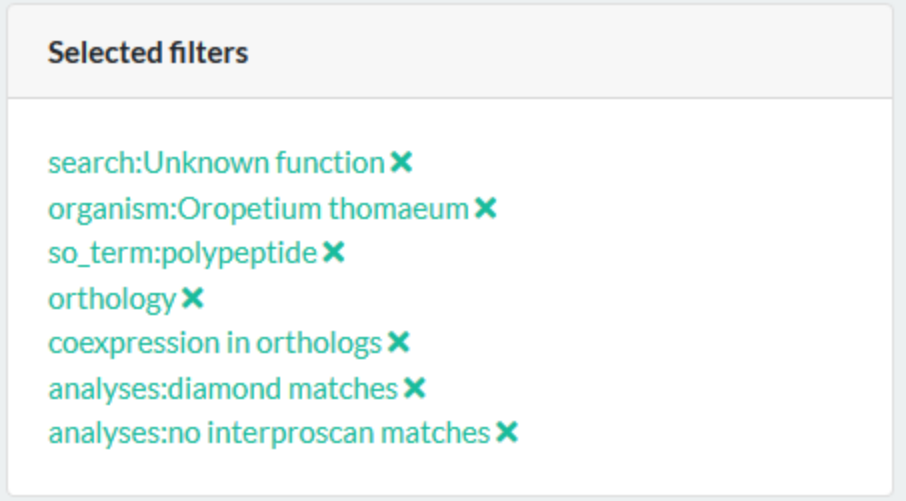
2.8 In addition to the applied filters, text search for "Unknown function" using the search box. After entering the text click on the magnifying glass icon.



https://www.machado.cnptia.embrapa.br/plantannot/find/?q=Unknown+function&selected_facets=organism%3AOropetium+thomaeum&selected_facets=so_term%3Apolypeptide&selected_facets=orthology%3A1&selected_facets=orthologs_coexpression%3Atrue&selected_facets=analyses%3Adiamond+matches&selected_facets=analyses%3Ano+interproscan+matches

Filters

3 After execution of all filters we will have the following list of filters:



https://www.machado.cnptia.embrapa.br/plantannot/find/?q=Unknown+function&selected_facets=organism%3AOropetium+thomaeum&selected_facets=so_term%3Apolypeptide&selected_facets=orthology%3A1&selected_facets=orthologs_coexpression%3Atrue&selected_facets=analyses%3Adiamond+matches&selected_facets=analyses%3Ano+interproscan+matches

Viewing results

4 Visualize the "Results" card on the center-right of the screen. There will be the resulting list of *Oropetium's* PUFs. 4 PUFs were filtered:

Results

Organism	Feature Type	Feature ID	Relationship	Display	Orthologous Group	Coexpression Group
Oropetium thomaeum	polypeptide	Oropetium_20150105_04788A.v1.0	mRNA	PF14365 - Domain of unknown function (DUF4409) (DUF4409) (1 of 10)	plantannot2042	
Oropetium thomaeum	polypeptide	Oropetium_20150105_05984A.v1.0	mRNA	PF03478 - Protein of unknown function (DUF295) (DUF295) (1 of 52)	plantannot22362	
Oropetium thomaeum	polypeptide	Oropetium_20150105_05988A.v1.0	mRNA	PF03478 - Protein of unknown function (DUF295) (DUF295) (1 of 52)	plantannot22362	
Oropetium thomaeum	polypeptide	Oropetium_20150105_06185A.v1.0	mRNA	PF04578//PF13968 - Protein of unknown function, DUF594 (DUF594) // Domain of unknown function (DUF4220) (DUF4220) (1 of 12)	plantannot17852	