

May 20, 2020

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

Forked from [Find Proteins of Unknown Function \(PUFs\) using Plantannot - Protocol B](#)

DOI

[dx.doi.org/10.17504/protocols.io.bgdjjs4n](https://dx.doi.org/10.17504/protocols.io.bgdjjs4n)

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

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**Protocol status:** Working

It is working

**Created:** May 13, 2020

**Last Modified:** May 20, 2020

**Protocol Integer ID:** 37003

## 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 stress using RNA-seq expression data and co-expression networks.

The Filter s 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

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(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 proto cols of A-C but using ortholog groups to find homolog proteins with co-expression data related to abiotic stress.

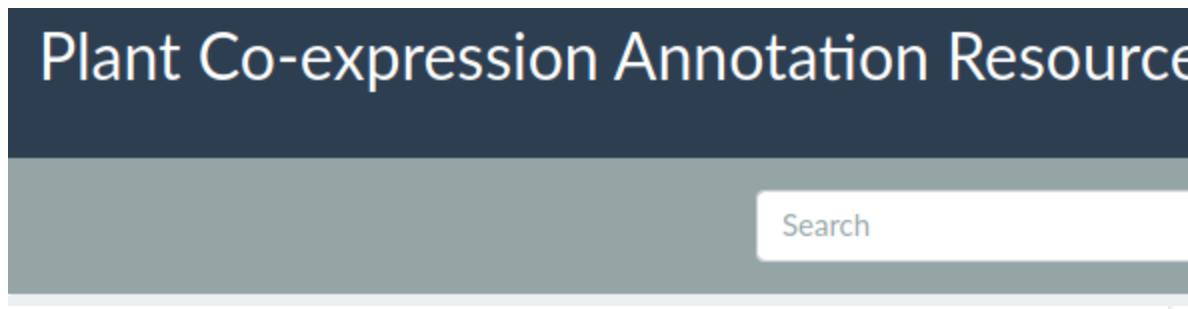
Protocol E is intended to find PUFs from organisms whose proteins are not yet in the NCBI "nr" database and have DUF domains found by InterProScan. Proteins will be selected using the text search "Unknown function". Also, ortholog groups and co-expression networks will be used to relate proteins.

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## 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 whose proteins are not yet in the NCBI "nr" database and have DUF domains from PFAM 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

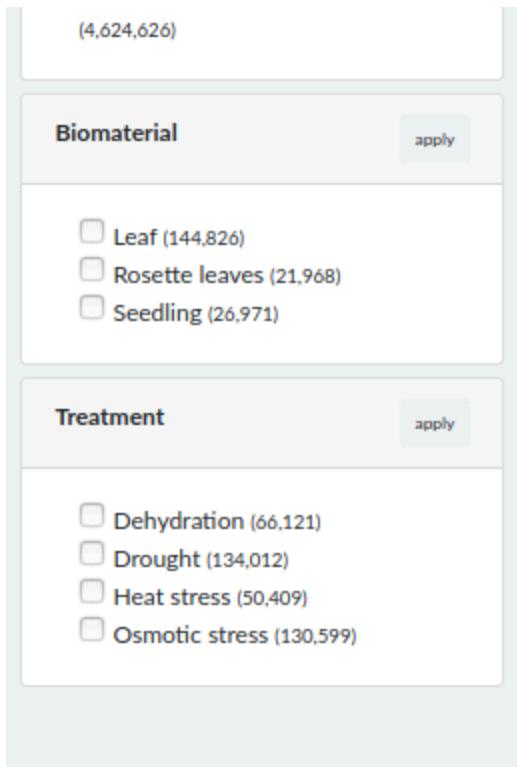
apply

- 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



The screenshot shows a search interface for plant annotations. At the top, a blue header bar displays '(4,624,626)' in white. Below it is a sidebar with two main sections: 'Biomaterial' and 'Treatment'. Each section has an 'apply' button to its right. Under 'Biomaterial', there are three filter options: 'Leaf (144,826)', 'Rosette leaves (21,968)', and 'Seedling (26,971)'. Under 'Treatment', there are four filter options: 'Dehydration (66,121)', 'Drought (134,012)', 'Heat stress (50,409)', and 'Osmotic stress (130,599)'. Each filter option consists of a small square checkbox followed by the category name and its count.

<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)

- Setaria italica* (120,586)
- Setaria viridis* (132,402)
- Solanum lycopersicum* (104,175)
- Solanum tuberosum* (151,458)
- Sorghum bicolor* (128,371)
- Spirodela polyrhiza* (58,869)
- Theobroma cacao* (118,260)
- Trifolium pratense* (122,552)
- Vitis vinifera* (79,038)
- Zea mays* (241,000)
- Zostera marina* (61,350)

[https://www.machado.cnptia.embrapa.br/plantannot/find/?q=&selected\\_facets=organism%3AOropetium+thomaeum](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:

Feature type	apply    remove
<ul style="list-style-type: none"><li><input type="checkbox"/> gene (0)</li><li><input type="checkbox"/> mRNA (0)</li><li><input checked="" type="checkbox"/> polypeptide (2,551)</li></ul>	

[https://www.machado.cnptia.embrapa.br/plantannot/find/?q=&selected\\_facets=organism%3AOropetium+thomaeum&selected\\_facets=so\\_term%3Apolypeptide](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:

**Orthology**

no orthology (0)  
 orthology (18,755)

[apply](#) [remove](#)

[https://www.machado.cnptia.embrapa.br/plantannot/find/?q=&selected\\_facets=organism%3AOropetium+thomaeum&selected\\_facets=so\\_term%3Apolypeptide&selected\\_facets=orthology%3A1](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:

**Coexpression**

no co-expression groups (711)  
 co-expression groups (0)

[apply](#)

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

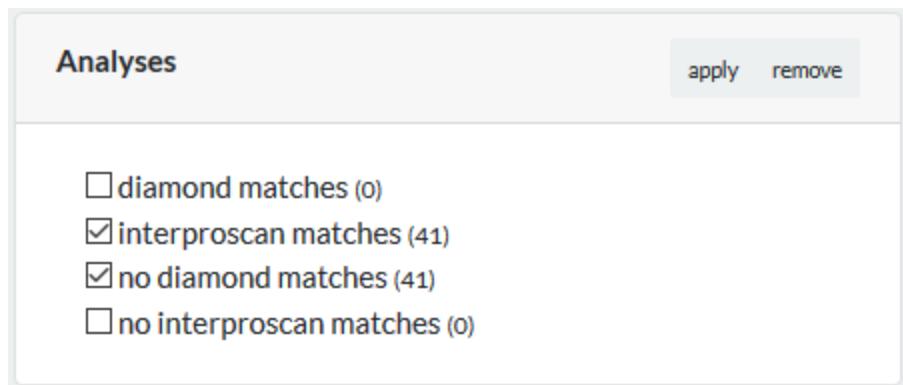
**Orthologs\_coexpression**

no co-expression (0)  
 co-expression (15,641)

[apply](#) [remove](#)

[https://www.machado.cnptia.embrapa.br/plantannot/find/?  
q=&selected\\_facets=organism%3AOropetium+thomaeum&selected\\_facets=so\\_term%3A  
polypeptide&selected\\_facets=orthology%3A1&selected\\_facets=orthologs\\_coexpression  
%3Atrue](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 the "Analyses" filter, select both "no diamond matches" and "interproscan matches", 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%3A  
polypeptide&selected\\_facets=orthology%3A1&selected\\_facets=orthologs\\_coexpression  
%3Atrue&selected\\_facets=analyses%3Ainterproscan+matches&selected\\_facets=analyses%3A  
no+diamond+matches](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%3Ainterproscan+matches&selected_facets=analyses%3Ano+diamond+matches)

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

### Biomaterial

[apply](#)

- Leaf (0)
- Rosette leaves (0)
- Seedling (0)

### Treatment

[apply](#)

- Dehydration (0)
- Drought (0)
- Heat stress (0)
- Osmotic stress (0)

- 2.8 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.



Filters	Results										
Selected filters	Results <table border="1" style="width: 100%; border-collapse: collapse; font-size: small;"> <thead> <tr> <th style="width: 10%;">Organism</th> <th style="width: 10%;">Feature Type</th> <th style="width: 10%;">Feature ID</th> <th style="width: 10%;">Relationship</th> <th style="width: 10%;">Display</th> </tr> </thead> <tbody> <tr> <td>Organism</td> <td>polypeptide</td> <td>Organism_20150105_01070A110</td> <td>mRNA</td> <td>DE12022_Domain of unknown</td> </tr> </tbody> </table>	Organism	Feature Type	Feature ID	Relationship	Display	Organism	polypeptide	Organism_20150105_01070A110	mRNA	DE12022_Domain of unknown
Organism	Feature Type	Feature ID	Relationship	Display							
Organism	polypeptide	Organism_20150105_01070A110	mRNA	DE12022_Domain of unknown							

[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%3Ainterproscan+matches&selected\\_facets=analyses%3Ano+diamond+matches](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%3Ainterproscan+matches&selected_facets=analyses%3Ano+diamond+matches)

## Filters

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

### Selected filters

search:Unknown function 

organism:Oropetium thomaeum 

so\_term:polypeptide 

orthology 

coexpression in orthologs 

analyses:interproscan matches 

analyses:no diamond matches 

[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%3Ainterproscan+matches&selected\\_facets=analyses%3Ano+diamond+matches](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%3Ainterproscan+matches&selected_facets=analyses%3Ano+diamond+matches)

## Viewing results

- 4 Visualize the "Results" card on the center-right of the screen, we will have the resulting list of *Oropetium*'s PUFs, 1 PUF were filtered:

Results						
Organism	Feature Type	Feature ID	Relationship	Display	Orthologous Group	Coexpression Grou
Oropetium thomaeum	polypeptide	Oropetium_20150105_24873Av1.0	mRNA	PF05097 - Protein of unknown function (DUF688) (DUF688) (1 of 13)	plantannot66633	

