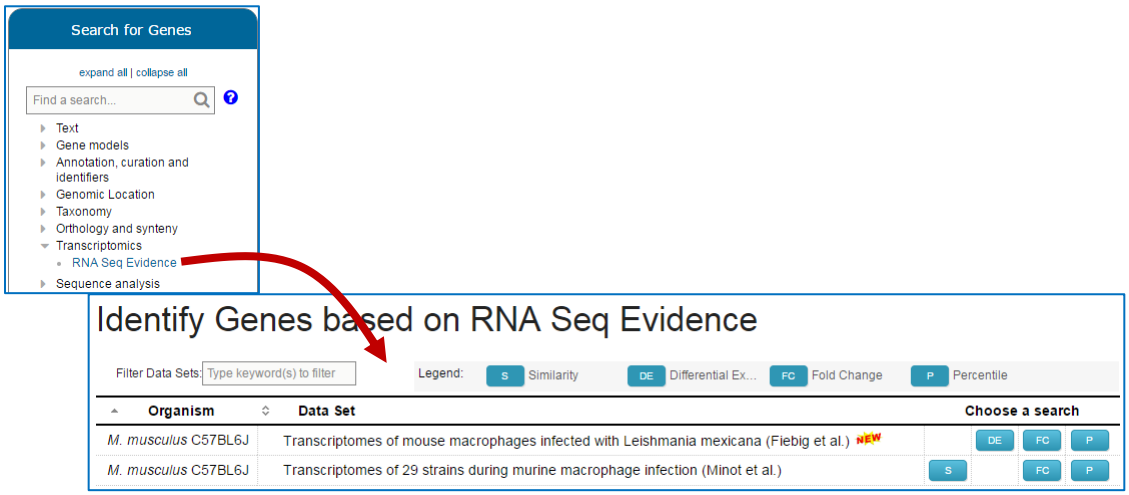
Host Response

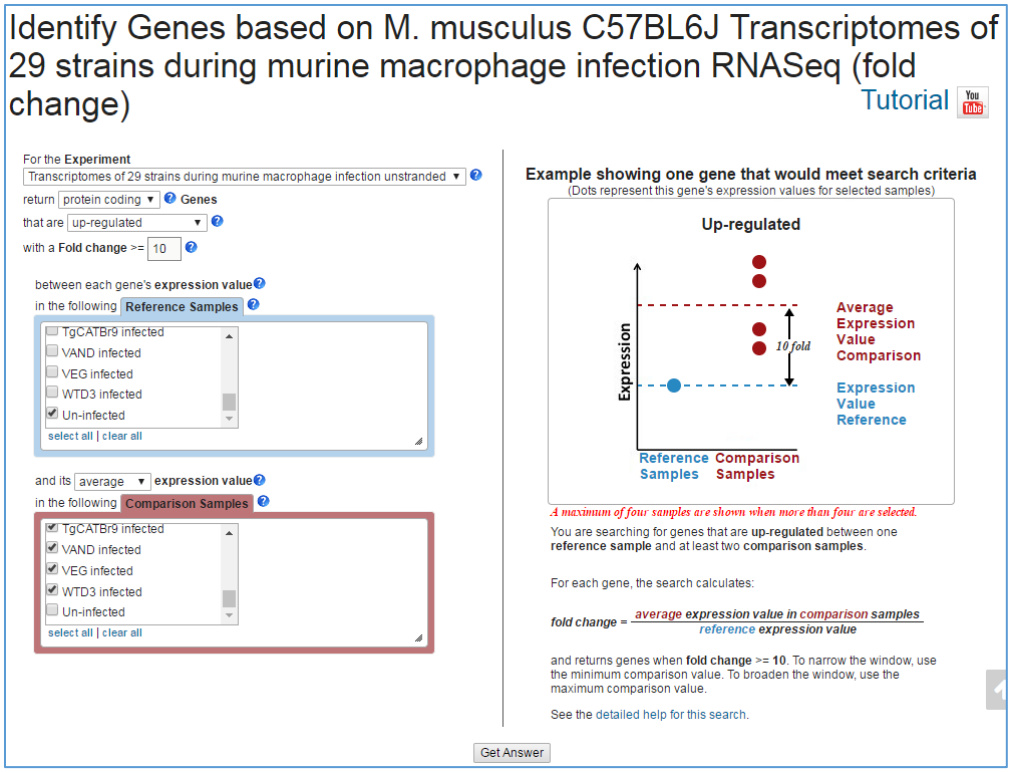
1. Find host genes that are upregulated in infected mouse cells compared to uninfected ones. For this exercise use http://hostdb.org

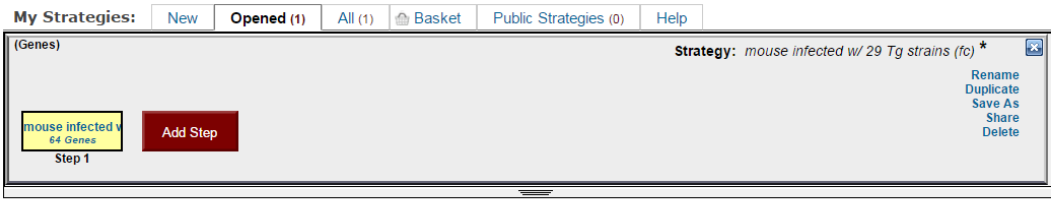
   a. Navigate to the "Transcriptomics" section then select "RNA Seq Evidence". Select the fold change query for the "Transcriptomes of 29 strains during murine macrophage infection (Minot et al.)" experiment.

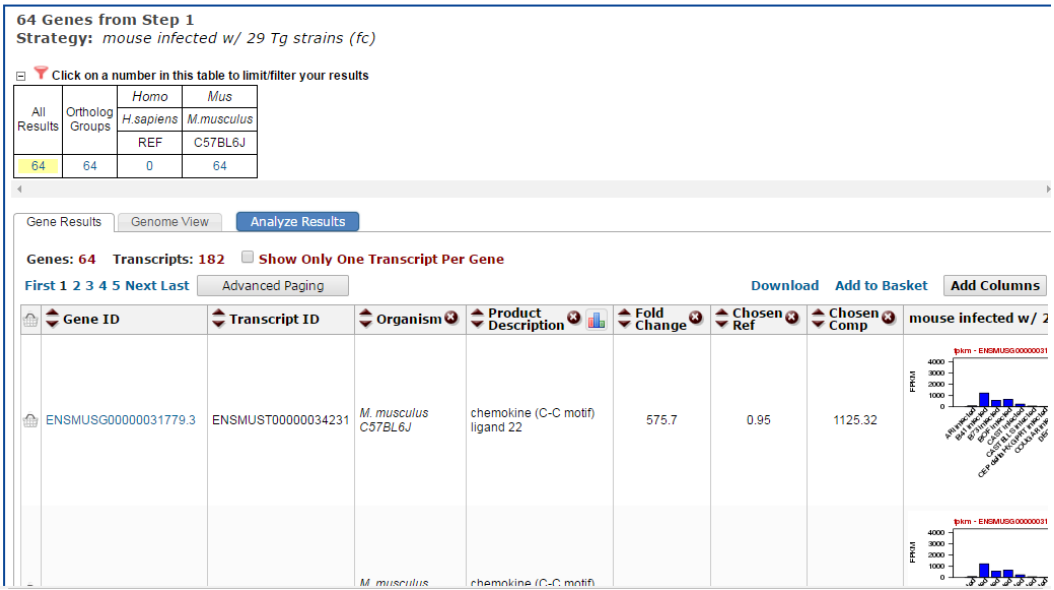


   b. Configure the search to compare all infected samples to the uninfected control. Make sure to select upregulated. In the example below a fold change of 10 was selected and the "average" operation was applied on the comparison samples.

      - What do these genes represent?


Identify Genes based on M. musculus C57BL6J Transcriptomes of 29 strains during murine macrophage infection RNASeq (fold change)
Tutorial
For the Experiment
Transcriptomes of 29 strains during murine macrophage infection unstranded
return protein coding Genes
that are up-regulated
with a Fold change >= 10
between each gene's expression value
in the following Reference Samples
TgCATBr9 infected
VAND infected
VEG infected
WTD3 infected
Un-infected
select all | clear all
and its average expression value
in the following Comparison Samples
TgCATBr9 infected
VAND infected
VEG infected
WTD3 infected
Un-infected
select all | clear all
Example showing one gene that would meet search criteria
(Dots represent this gene's expression values for selected samples)
Up-regulated
Expression
10 fold
Average Expression Value Comparison
Expression Value Reference
Reference Samples
Comparison Samples
A maximum of four samples are shown when more than four are selected.
You are searching for genes that are up-regulated between one reference sample and at least two comparison samples.
For each gene, the search calculates:
fold change = average expression value in comparison samples / reference expression value
and returns genes when fold change >= 10. To narrow the window, use the minimum comparison value. To broaden the window, use the maximum comparison value.
See the detailed help for this search.
Get Answer


My Strategies:
New
Opened (1)
All (1)
Basket
Public Strategies (0)
Help
(Genes)
Strategy: mouse infected w/ 29 Tg strains (fc) *
Rename
Duplicate
Save As
Share
Delete
mouse infected v
64 Genes
Step 1
Add Step


64 Genes from Step 1
Strategy: mouse infected w/ 29 Tg strains (fc)
Click on a number in this table to limit/filter your results
All Results
Ortholog Groups
Homo H.sapiens REF
Mus M.musculus C57BL6J
64
64
0
64
Gene Results
Genome View
Analyze Results
Genes: 64 Transcripts: 182
Show Only One Transcript Per Gene
First 1 2 3 4 5 Next Last
Advanced Paging
Download
Add to Basket
Add Columns
Gene ID
Transcript ID
Organism
Product Description
Fold Change
Chosen Ref
Chosen Comp
mouse infected w/ 2
ENSMUSG00000031779.3
ENSMUST00000034231
M. musculus C57BL6J
chemokine (C-C motif) ligand 22
575.7
0.95
1125.32

c. What are the functional characteristics of the genes in this result? What kinds of GO terms are enriched? Does the host immune response appear to be turned on?
Hint: click on the "Analyze Results" tab and perform a GO enrichment analysis for the biological process ontology.

Gene Results | Genome View | Gene Ontology Enrichment | Analyze Results

[Rename This Analysis | Copy These Parameter Values

Gene Ontology Enrichment

Find Gene Ontology terms that are enriched in your gene result. *Read More*

▸ **Parameters**

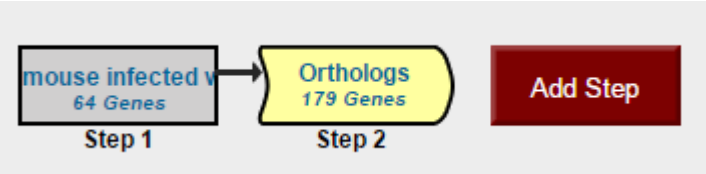
This analysis result may be lost if you change your gene result. To save this analysis result, please Download Analysis Results

Analysis Results:

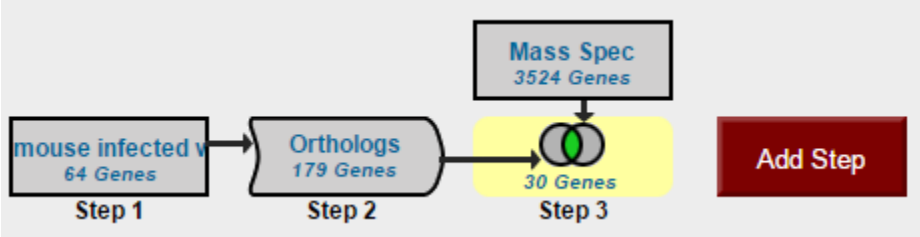
Got a total of 1,258 results Filter :

GO ID	GO Term	Genes in the bkgd with this term	Genes in your result with this term	Percent of bkgd Genes in your result	Fold enrichment	Odds ratio	P-value	Benjamini	Bonferroni
GO:0006955	immune response	619	20	3.2	15.49	22.91	1.06e-18	1.33e-15	1.33e-15
GO:0034097	response to cytokine	388	16	4.1	19.76	26.75	1.36e-16	5.97e-14	1.71e-13
GO:0002376	immune system process	1069	22	2.1	9.86	15.13	1.42e-16	5.97e-14	1.79e-13
GO:0071345	cellular response to cytokine stimulus	300	14	4.7	22.37	29.01	2.68e-15	8.42e-13	3.37e-12
GO:0019221	cytokine-mediated signaling pathway	187	11	5.9	28.19	34.42	3.06e-13	7.70e-11	3.85e-10
GO:0071310	cellular response to organic substance	968	18	1.9	8.91	12.39	8.09e-13	1.70e-10	1.02e-9
GO:0006952	defense response	714	16	2.2	10.74	14.36	1.25e-12	2.04e-10	1.58e-9
GO:0051239	regulation of multicellular organismal process	1497	21	1.4	6.72	9.89	1.30e-12	2.04e-10	1.63e-9
GO:0010033	response to organic substance	1526	21	1.4	6.6	9.69	1.86e-12	2.60e-10	2.34e-9
GO:0001817	regulation of cytokine production	309	12	3.9	18.61	23.11	2.53e-12	3.18e-10	3.18e-9
GO:0006954	inflammatory response	234	11	4.7	22.53	27.46	3.13e-12	3.58e-10	3.94e-9

d. Expand the result set to include orthologs/paralogs of these genes. *Hint:* add a "Transform by Orthology" step choosing Homo sapiens.

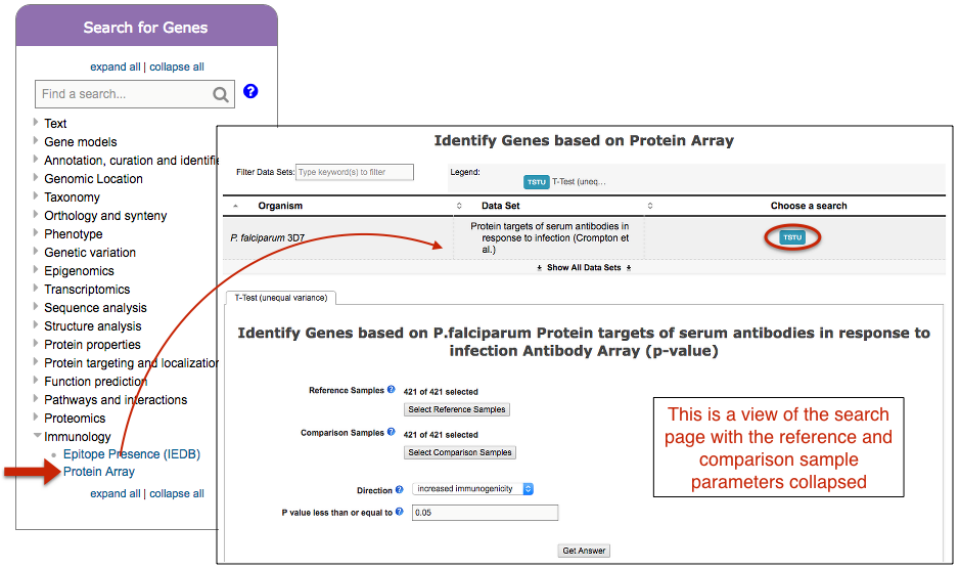


e. Does this set of human genes also show enriched GO terms? What, if any, are the enriched GO terms?
f. Do any of these human genes also have peptide evidence for their expression during infection? *Hint:* add a step and explore the "Mass Spec Evidence" data in the protein expression section. Run the search using the default parameters.

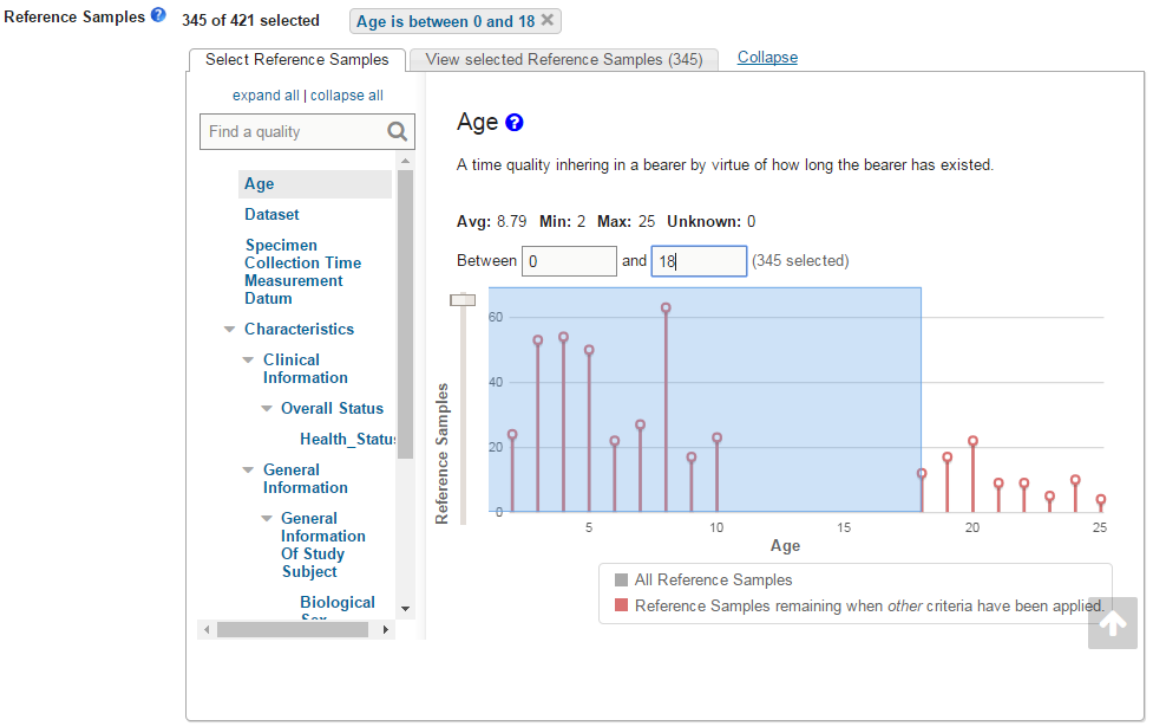

Mass Spec
3524 Genes
mouse infected v
64 Genes
Orthologs
179 Genes
30 Genes
Add Step
Step 1
Step 2
Step 3

2. Find *Plasmodium falciparum* antigens that are immunogenic.
For this exercise use http://plasmodb.org

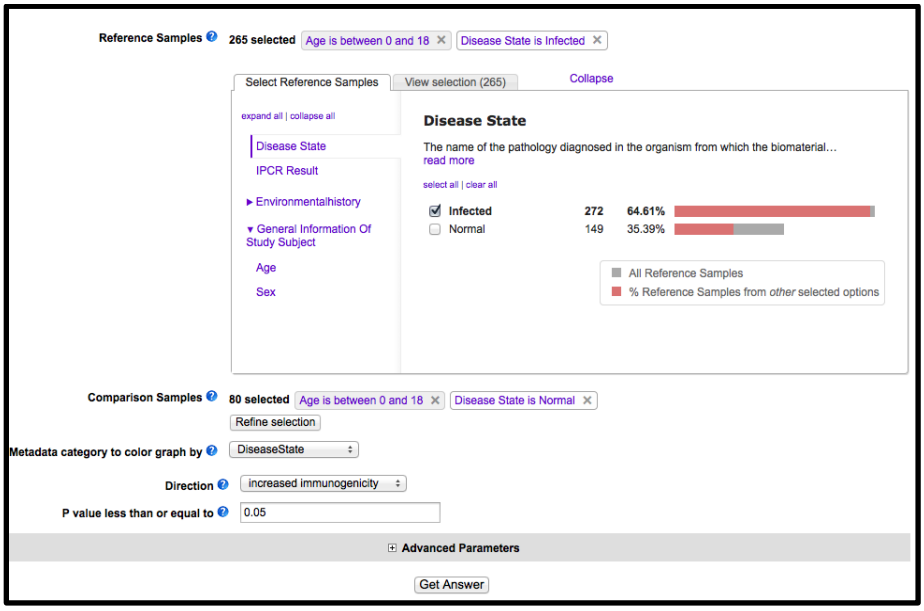
a. Identify antigens (genes) that exhibited an increased immunogenicity in children (ages 0-18) with no disease (normal) compared to children with disease (malaria). *Hint*: the "Protein Array" search is available in the "Immunology" menu item in the "Identify Genes By" section of the home page. Choose the experiment Protein targets of serum antibodies in response to infection (Crompton et al.).



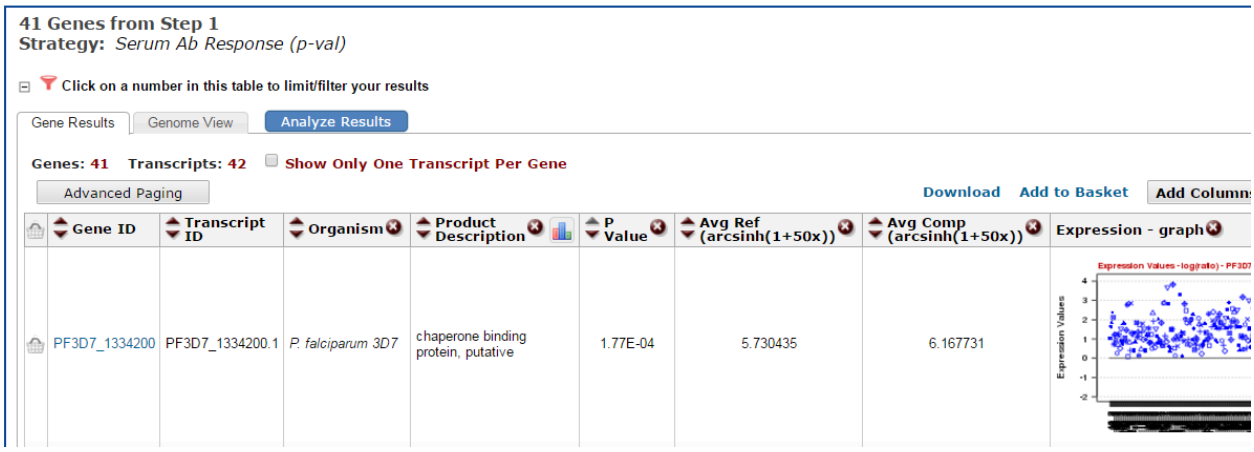
In this example, your **comparison samples will be normal children** and your **reference samples will be infected children**. Each set of samples (reference and comparison) has two parameters that need to be set, age and health status. Set the age parameter to 0-18 years for both the reference and the comparison samples.



Now set the **health status for the reference samples to malaria**. Move on the comparison samples and set the age to 0-18 and the disease state to normal. The default settings for other parameters are good – increased immunogenicity and p-value = 0.05.



You are ready to click Get Answer! What do your results look like? Could these represent potential protective antigens? (result image below)



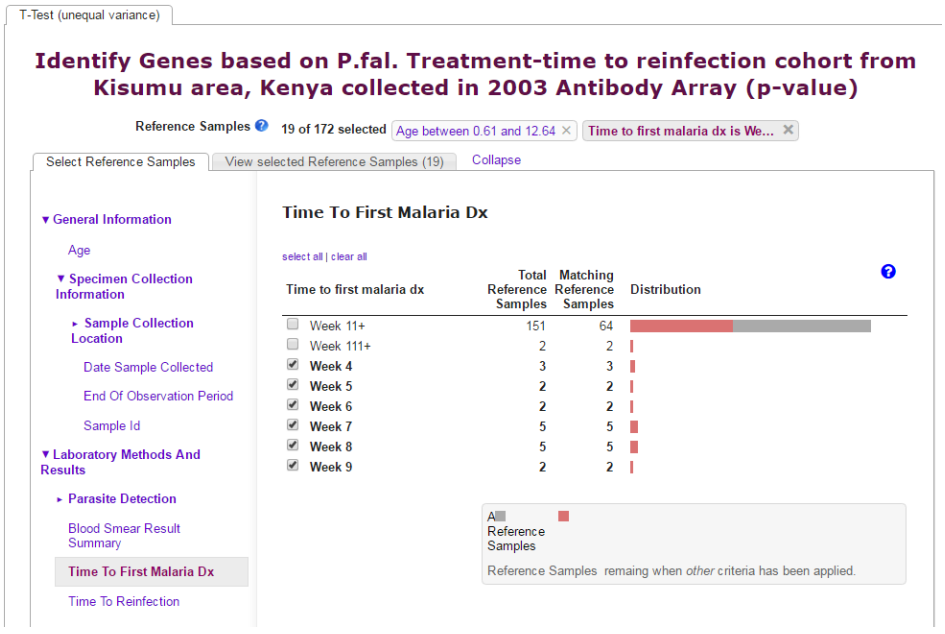
3. Find *falciparum* antigens that may be protective from reoccurrence of malaria (and potentially reinfection)

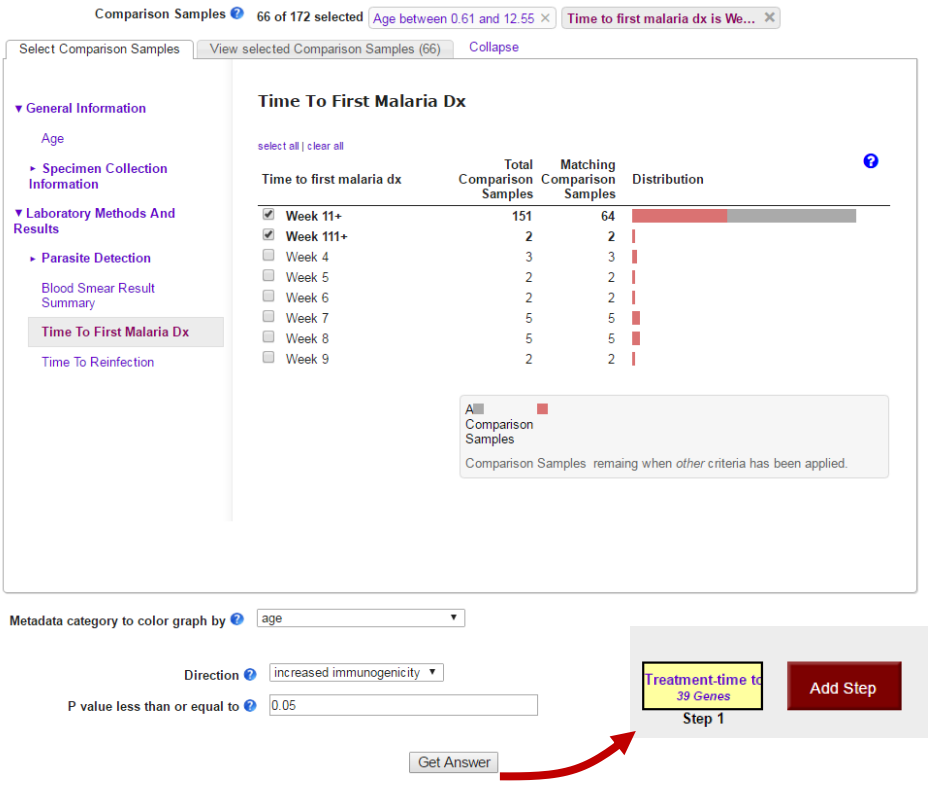
   For this exercise use http://plasmodb.org

   A recently published study from Kenya (view paper) where participants were followed for 12 weeks following an initial screening for malaria and treatment with anti-malarials is available in PlasmoDB. Each week patients were assessed for presence of parasites and clinical symptoms of malaria. Select the **"Treatment-time to reinfection cohort from Kisumu area, Kenya collected in 2003 (Dent et al.)"** experiment from the protein array searches and configure the parameters to see if you can reproduce the results of the paper. They concluded that increased antigenicity was present in children who did not show clinical symptoms of malaria, and suggest that these antigens are protective in children who did not get a recurrence of symptomatic malaria (compared to those children who did exhibit malaria symptoms). They also concluded that there did not appear to be a correlation between antigenicity and time to re-infection (could be asymptomatic). Test both these conclusions.

   Hint: compare children (age 0-12.5) who got clinical malaria during the study (time to first malaria Dx weeks 4-9) compared to those who didn't (week 11+). Try running with increased immunogenicity then revise and change to decreased immunogenicity. See image below for help configuring the search.
   Do these results make sense?

   Ask the same question (age 0-12.5) except compare time to re-infection weeks 3 and 4 with time to reinfection weeks 9,10,11,11+. Do you get significant results? Does this agree with the conclusions of the paper? Revise the search and remove the age limits, just keeping the times to re-infection.




Comparison Samples
66 of 172 selected
Age between 0.61 and 12.55
Time to first malaria dx is We...
Select Comparison Samples
View selected Comparison Samples (66)
Collapse
▾ General Information
Age
▸ Specimen Collection Information
▾ Laboratory Methods And Results
▸ Parasite Detection
Blood Smear Result Summary
Time To First Malaria Dx
Time To Reinfection
Time To First Malaria Dx
select all | clear all
Time to first malaria dx
Total Comparison Samples
Matching Comparison Samples
Distribution
Week 11+ 151 64
Week 111+ 2 2
Week 4 3 3
Week 5 2 2
Week 6 2 2
Week 7 5 5
Week 8 5 5
Week 9 2 2
Comparison Samples
Comparison Samples remaing when other criteria has been applied.

Metadata category to color graph by
age
Direction
increased immunogenicity
P value less than or equal to
0.05
Get Answer
Treatment-time to
39 Genes
Step 1
Add Step