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 “Transcript Expression” section then select “RNA Seq Evidence”. Select the fold change query for the “Transcriptome during infection with 25 strains of T. gondii (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.
c. What are the functional characteristics of the genes in this result? What kinds of GO terms are enriched?  
*Hint*: click on the “Analyze Results” tab and select the GO enrichment analysis.
d. Expand the result set to include orthologs/paralogs of these genes. *Hint:* add a “Transform by Orthology” step choosing Homo sapiens.

```
mouse infected v
200 Genes
Orhologs
727 Genes
```

```
Add Step
```

```
Step 1
Step 2
```

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e. 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.

```
mouse infected v
200 Genes
Orhologs
727 Genes
Mass Spec
10887 Genes
```

```
139 Genes
```

```
Add Step
```

```
Step 1
Step 2
Step 3
```
2. Find *Plasmodium falciparum* antigens that are immunogenic.

For this exercise use [http://plasmodb.org](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 (infected). *Hint*: the “Protein Array” search is available in the “Host Response” 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.).

![Protein Array search](image)

In this example, your **comparison samples will be normal children** and your **reference samples with be infected children**. So each set of samples (reference and comparison) has two parameters that need to be set, age and disease state. The age parameter should be set to 0-18 years for both the reference and the comparison samples. To set the age parameter for the reference samples, choose the ‘General Information of Study Subject’ and then ‘Age’ from the left menu to reveal choices for the parameter on the right. Set the range to 0-18 (see image below).
Now set the disease state for the reference samples to infected. 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](http://plasmodb.org)

A recently published study from Kenya ([view paper](http://plasmodb.org)) 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.