

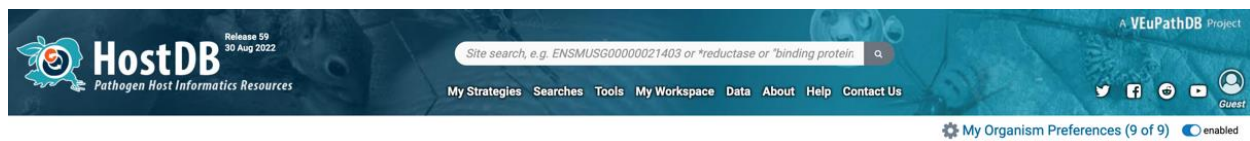
Host Response

Learning objectives:

- Exploring host responses by running a search strategy in HostDB
- Add steps in a search strategy
- Revising steps in a search strategy

1. Identify host genes that are differentially regulated in response to infection by different pathogens. For this exercise use <http://hostdb.org>

- The goal of this exercise is to find genes that are differentially regulated in the host during infection by multiple pathogens:
 - *Babesia rossi*
 - *Plasmodium vinckei*
 - *Toxoplasma gondii*
- Start by running search for genes that are differentially regulated (fold-change) in the experiment “*Babesia infection in dog (Smith et al. 2021)*”



Identify Genes based on RNA-Seq Evidence

Legend: S Similarity DE Differential Expression FC Fold Change P Percentile SA SenseAntisense		
Filter Data Sets: <input type="text"/> 38 rows		
Organism	Data Set	Choose a Search
<i>Bos taurus</i> breed Hereford	Transcriptome of <i>Bos taurus</i> during infection with virulent and avirulent <i>N. caninum</i> strains (Horcajo et al.)	DE FC P SA
<i>Bos taurus</i> breed Hereford	Host cell transcriptome in bovine cells infected with <i>Cryptosporidium parvum</i> (Widmer et al.)	DE FC P SA
<i>Canis lupus familiaris</i> isolate SID07034	<i>Babesia</i> infection in dog (Smith et al. 2021) NEW	DE FC P SA
<i>Cavia porcellus</i> 2N	mRNA-based anti-tick (<i>I. scapularis</i>) vaccine (Sajid et al 2021)	FC P SA
<i>Gallus gallus</i> isolate bGalGal1	Chicken caecal tissue during <i>E. tenella</i> infection (Sandholt et al 2021)	DE FC P SA

- Configure the search to return all genes that are up or down regulated by at least 2-fold when comparing all low and high infection samples to Day 0 (low and high).

Identify Genes based on C. lupus familiaris isolate SID07034 Babesia infection in dog RNA-Seq (fold change)

☐ Babesia infection in dog - Antisense

return Genes

that are

with a **Fold change** \geq 2

between each gene's expression value
(or a Floor of)

in the following **Reference Samples**

- ☒ Day 0- high dose
- ☐ Day 1- high dose
- ☐ Day 3- high dose
- ☐ Day 4- high dose
- ☐ Day 6- high dose
- ☐ Day 8- high dose
- ☒ Day 0- low dose
- ☐ Day 1- low dose
- ☐ Day 3- low dose

[select all](#) [clear all](#)

and its expression value
(or the Floor selected above)

in the following **Comparison Samples**

- ☒ Day 4- high dose
- ☒ Day 6- high dose
- ☒ Day 8- high dose
- ☐ Day 0- low dose
- ☒ Day 1- low dose
- ☒ Day 3- low dose
- ☒ Day 4- low dose
- ☒ Day 6- low dose
- ☒ Day 8- low dose

[select all](#) [clear all](#)

Up or down regulated

A maximum of four samples are shown when more than four are selected.

For each gene, the search calculates:

$$\text{fold change}_{\text{up}} = \frac{\text{average expression value in comparison}}{\text{average expression value in reference}}$$

$$\text{fold change}_{\text{down}} = \frac{\text{average expression value in reference}}{\text{average expression value in comparison}}$$

and returns genes when $\text{fold change}_{\text{up}} \geq 2$ or $\text{fold change}_{\text{down}} \geq 2$.

You are searching for genes that are up or down regulated between at least two reference samples and at least two comparison samples.

- How many genes did the search return? What is the host organism?
- Add a step and navigate to the RNA-Seq datasets
- Find the experiment “Mouse transcriptome during P. vinckei CY infection time course (Ramaprasad et al)” and select the fold-change search.
- Configure the search to return all genes that are differentially regulated (up or down) by at least 2-fold when comparing the 18hr and 24hr infection to the 6 and 12 hr infection samples.

[Add a step to your search strategy](#)

Differential Expression **Fold Change** Percentile SenseAntisense

Configure Search [Learn More](#) [View Data Sets Used](#)

For the Experiment

- ☒ Mouse transcriptome during P. vinckei CY infection time course - Sense
- ☐ Mouse transcriptome during P. vinckei CY infection time course - Antisense

return Genes

that are

with a **Fold change** \geq 2

between each gene's expression value
(or a Floor of)

in the following **Reference Samples**

- ☒ 6 hr
- ☒ 12 hr
- ☐ 18 hr
- ☐ 24 hr

[select all](#) [clear all](#)

and its expression value
(or the Floor selected above)

in the following **Comparison Samples**

- ☐ 6 hr
- ☐ 12 hr
- ☒ 18 hr
- ☒ 24 hr

[select all](#) [clear all](#)

Example showing one gene that would meet search criteria
(Dots represent this gene's expression values for selected samples)

Up or down regulated

For each gene, the search calculates:

$$\text{fold change}_{\text{up}} = \frac{\text{average expression value in comparison}}{\text{average expression value in reference}}$$

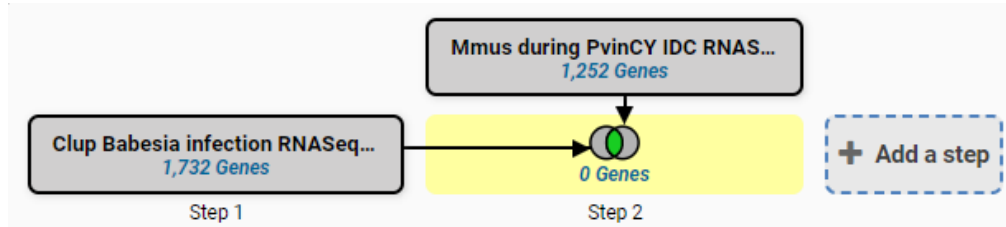
$$\text{fold change}_{\text{down}} = \frac{\text{average expression value in reference}}{\text{average expression value in comparison}}$$

and returns genes when $\text{fold change}_{\text{up}} \geq 2$ or $\text{fold change}_{\text{down}} \geq 2$.

You are searching for genes that are up or down regulated between at least two reference samples and at least two comparison samples.

[Run Step](#)

- How many results did you get?



- Is this result of 0 genes surprising? What can you do to change this? Hint: think about the host organism for each experiment, are they the same or different? What can you do to compare across species?

My Search Strategies

Opened (1) All (3) Public (2) Help

Unnamed Search Strategy *

Step 1: Clup Babesia infection RNASeq... (1,732 Genes)

Step 2: Mmus during PvinCY IDC RNAS... (1,252 Genes)

0 Genes (0 ortholog groups)

Organism Filter: select all | clear all | expand all | collapse all

Details for step: Mmus during PvinCY IDC RNAS... (1,252 Genes)

Experiment: Mouse transcriptome during P. vinckei infection time course - Sense

Direction: 5' to 3'

Reference Samples: 6 hr, 12 hr

Operation Applied to Reference Samples: average

Comparison Samples: 18 hr, 24 hr

Operation Applied to Comparison Samples: average

fold difference >= 2

Floor = 10 reads

Protein Coding Only: protein coding

Add a step to your search strategy ?

Your Genes from Step 1 will be converted into Orthologs

Organism

1 selected, out of 9

select only these | add these | clear these

mus

Mammalia

Rodentia

Muridae

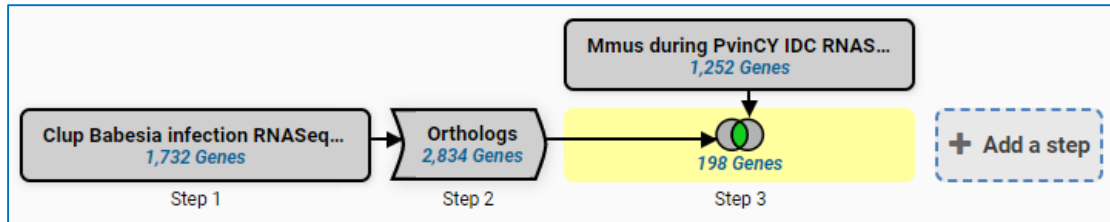
☒ Mus musculus C57BL6J [Reference]

select only these | add these | clear these

Syntenic Orthologs Only?

no

Run Step



- Use the same logic above and add a step to identify all genes that are differentially regulated in the experiment “Mouse transcriptome during early and late chronic infection with *T. gondii* (Garfoot et al.)”
- Configure the search to compare the 21- and 28-day infected female mice to the 21- and 28-day uninfected female mice.

Add a step to your search strategy ?

Differential Expression
Fold Change
Percentile

For the Experiment

☒ Mouse transcriptome during early and late chronic infection with *T. gondii* unstranded

return protein coding ? Genes

that are up or down regulated ?

with a **Fold change** ≥ 2 ?

between each gene's average ? expression value
(or a Floor of 10 reads ?)

in the following **Reference Samples** ?

☐ 21 days Tg Female
☒ 21 days Uninfected Female
☐ 28 days Tg Female
☒ 28 days Uninfected Female
☐ 3 mo Tg Female
☐ 3 mo Uninfected Female

[select all](#) | [clear all](#)

and its average ? expression value
(or the Floor selected above)

in the following **Comparison Samples** ?

☒ 21 days Tg Female
☐ 21 days Uninfected Female
☒ 28 days Tg Female
☐ 28 days Uninfected Female
☐ 3 mo Tg Female
☐ 3 mo Uninfected Female

[select all](#) | [clear all](#)

Example showing one gene that would meet search criteria
(Dots represent this gene's expression values for selected samples)

Up or down regulated

Up or down regulated

For each gene, the search calculates:

$$\text{fold change}_{\text{up}} = \frac{\text{average expression value in comparison}}{\text{average expression value in reference}}$$

$$\text{fold change}_{\text{down}} = \frac{\text{average expression value in reference}}{\text{average expression value in comparison}}$$

and returns genes when $\text{fold change}_{\text{up}} \geq 2$ or $\text{fold change}_{\text{down}} \geq 2$.

You are searching for genes that are **up or down regulated** between at least two reference samples and at least two comparison samples.

- How many genes did you get? Are there any gene with interesting functions in the list of results? Hint: Try a GO enrichment

