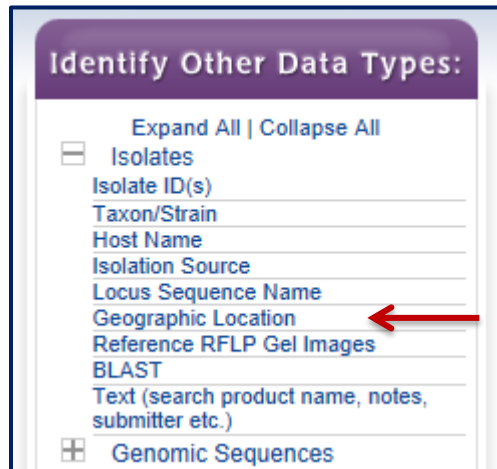


Exploring Isolate Data Exercise 8

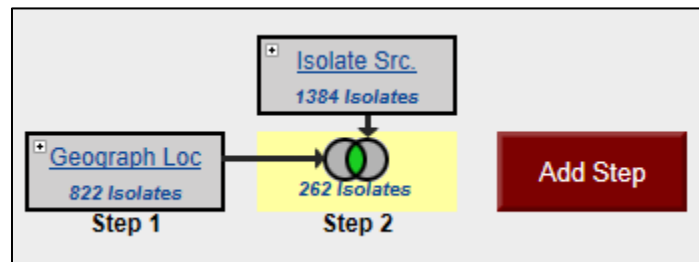
8.1 Exploring isolates in *Cryptosporidium* and using the alignment tool.

Note: For this exercise use <http://www.cryptodb.org>

- a. Identify all *Cryptosporidium* isolates from Europe.
Hint: search for isolates by geographic location in the “Identify Other Data Types” section.



- b. How many of the *Cryptosporidium* isolates collected in Europe were isolated from feces?
Hint: add another isolate search step.



- c. What is the general distribution of these isolates in Europe? (hint: you can do this quickly in two ways: sort the geographic location column by clicking on the sort arrows, then look at the represented countries; or use the “Isolate Geographical Location” tab to view a map and results summary table).

Sort by clicking on the arrows

Isolate ID	Geographic Location	Organism	Strain/Isolate Name	Host	Isolation Date
AB247224	Serbia	Cryptosporidium parvum	#6	Unknown	fecal sample
AB247225	Serbia	Cryptosporidium parvum	#24	Unknown	fecal sample
AB247226	Serbia	Cryptosporidium parvum	#42	Unknown	fecal sample
AB247227	Serbia				

Country	Number of Isolates	Isolate Type	Latitude	Longitude
Belgium	1	Sequencing Typed	50.503887	4.469936
Czech Republic	73	Sequencing Typed	49.817492	15.472962
Germany	90	Sequencing Typed	51.165691	10.451526
Ireland	4	Sequencing Typed	53.41291	-8.24389
Italy	11	Sequencing Typed	41.87194	12.56738
Lithuania	1	Sequencing Typed	55.169438	23.881275
Netherlands	41	Sequencing Typed	52.132633	5.291266

- d. Out of those in step ‘b’, how many are unclassified *Cryptosporidium* species? Hint: add another isolate search step.
- e. How many of step ‘b’ isolates originated from humans?
- f. How many of the isolates in step ‘b’ were typed using GP40/15 (GP60)? (hint: you can insert a step within a strategy. Click on the name of the step you want to insert a step before, then click on “Insert step before”).

Rename | View | Revise | Make Nested Strategy | **Insert Step Before** | Delete

STEP 3 : Host Name

Host : Mammals - Human
Isolate assay type : HTS, Sequencing Typed

Results: 1210 Isolates

+ Give this search a weight

- g. Compare some of these isolates using the multiple sequence alignment tool (ClustalW). Do you see any sequences with insertions or deletions?

My Step Result:


Geograph Loc - step 3 - 3 Isolates Add 3 Isolates to Basket | Download 3 Isolates

Isolates Select Columns

Advanced Paging

Isolate Id	Country	Organism	Strain	Host	Isolation Source
EF519704 <input checked="" type="checkbox"/>	Italy	Cryptosporidium sp. CrIT-20	CrIT-20	Testudo marginata	feces
EF547155 <input checked="" type="checkbox"/>	Italy	Cryptosporidium sp. CrIT-20	CrIT-20	Testudo marginata	feces
EU331243 <input checked="" type="checkbox"/>	Czech Republic	Cryptosporidium sp. pig genotype II	H199	Homo sapiens	stool sample

Please select at least two isolates to run ClustalW. Note: only isolates from a single results page will be aligned. Increase the page size in advanced paging to increase the number that can be aligned.



Advanced Paging

- h. Take a look at the 'guide tree' that was built using this alignment. Change the isolates that you selected for alignment - how does the tree change? Do isolates from the same country cluster together?

8.2 Typing an unclassified isolate.

Note: For this exercise use <http://www.cryptodb.org>

- a. Run a search to find all unclassified *Cryptosporidium* isolates and find one that was typed using 18S small subunit ribosomal RNA. (Hint: Identify Isolates based on Taxon/Strain and choose 'unclassified' under *Cryptosporidium*. Add a column for Gene Product and sort the column).

Select Columns

Update Columns

clear all | expand all | collapse all
reset to current | reset to default

- Organism
- Strain/Isolate Name
- Host
- Isolation Source
- Geographic Location
- EuPathDB Annotated Host
- EuPathDB Annotated Isolation Source
- EuPathDB Annotated Geographic Location
- Gene Product (Sequence Typed Only)
- Description
- Is Reference
- Isolate Type
- Weight

clear all | expand all | collapse all
reset to current | reset to default

Update Columns

Gene Product (Sequence Typed Only)
18S rRNA
COWP
COWP

- b. Go to the isolate record page and copy the DNA sequence.
- c. Go to search for isolates based on BLAST, select isolates and make sure only the reference isolates are selected in the target organism window.
- d. Paste the DNA sequence in the input window and select the Blastn program. Click on "Get Answer".
- e. Explore your results. Based on the similarity which reference isolate is this one closest to?

Target Data Type ? Transcripts
 Proteins
 Genome
 EST
 ORF
 Isolates

BLAST Program ? blastn
 blastp
 blastx
 tblastn
 tblastx

Target Organism ? select all | clear all | expand all | collapse all | reset to default
 Cryptosporidium Isolates minus Reference
 Cryptosporidium Reference Isolates

Input Sequence ?

```
AATTAGAGTCCTTAAAGCAGGC
ATTGCGCTTGAATACTCCAGCATGGATAATATTTAAG
ATTTTATCTTCTTATTGGTT
CTAAGATAAAAATAATGATTAATAGGCAGTTGGGGG
CATTGTATTAA
```

Note: only one input sequence allowed.
maximum allowed sequence length is 31K bases.

Expectation value ? 10

Maximum descriptions (V) ? 50

Maximum alignments (B) ? 50

Low complexity filter ? yes

+ Give this search a weight

+ Give this search a name

Get Answer

8.3 Exploring isolates in *Plasmodium*.

Note: For this exercise use <http://www.plasmodb.org>

- a. Identify all isolates from Mexico.
- b. How many of those are *P. falciparum*? How many *P. vivax*?
- c. What about all of North and South America?
Hint: revise the first step in your strategy to include all countries in both continents.
- d. For these results, add columns such as isolate product and length. Sort these columns and explore your results. For example, what product is mainly used in typing *P. falciparum* isolates? What about *P. vivax* isolates?