Annotating a Genome in PATRIC

The following step-by-step workflow is intended to help you learn how to navigate the new PATRIC workspace environment in order to annotate and browse your genome on the PATRIC website.

Step1. Go to the PATRIC website and log on

www.patricbrc.orglogin: workshopdemoPassword: patric123

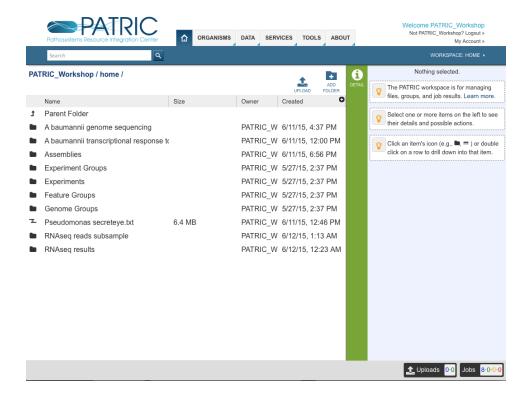


Step 2. Let's familiarize ourselves with the PATRIC workspace

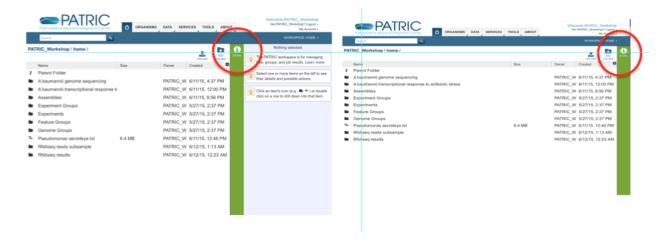
• Click on Workspace: Home in the top right corner to go to the PATRIC workspace



• The workspace is a series of folders that you can make for your projects



- The right hand panel shows links for information on how to use the workspace
- This can be displayed or hidden by clicking on the "DETAIL" button



 To create a folder or upload to a folder click on the buttons in the top right corner of the main panel



You can navigate into a folder by clicking on the folder icon



- The current working directory will be shown at the top
- The "Parent Folder" Icon is your "Back" button



Step 3. Download two Acinetobacter baumanii genomes to your computer

- Navigate to the "Assemblies" folder
- Click on the first genome, A. baumannii 1000160
- Click on the SRR1033639.auto assembly
- Download it to your computer
- Note that you may need to turn off the pop-up blocker on your browser





PATRIC_Workshop / home / Assemblies / A baumannii 1000160 / SRR1033693.auto

Genome Assembly Job Result

Start time	6/11/15, 7:05 PM	
End time	6/11/15, 8:31 PM	
Run time	1h26m3607s	
Parameters	baumannii 1000160","paired_end_libs":[{"re genome sequencing/Acinetobacter bauman 1000160/SRR1033693_2.fastq.gz","read1": genome sequencing/Acinetobacter bauman	/PATRIC_Workshop@patricbrc.org/home/A baumannii

Result Files



Make sure that the pop-up blocker is turned off on your browser

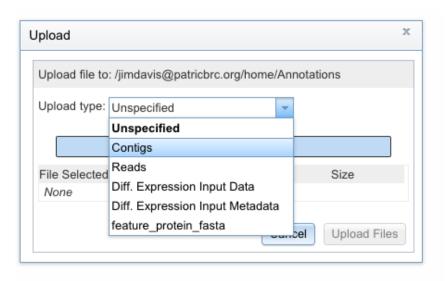
- Repeat this for *A. baumannii* 1592797.
- Select the SRR1019284.auto assembly

Step 4. Let's annotate our A. baumanii genomes in our own workspace

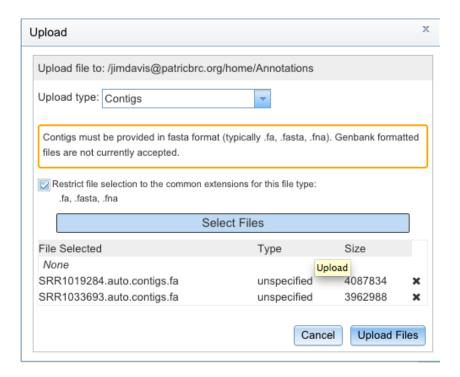
- Locate both files on your computer
- Add the file extension .fa to both files
- Log out of the "Patric_Workshop" workspace
- Log into your personal workspace
 - o If you do not have an account, please create one
- In your workspace create a new folder called "Annotations"
- Navigate to the Annotations folder
- Click the upload icon



Select "Contigs" as the file type



• Click "Select Files", find the two contig files and upload them

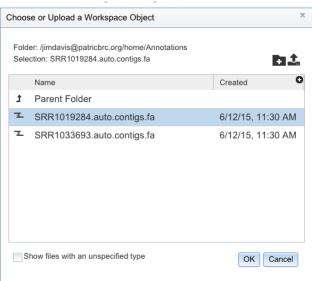


• Under the "Services" tab at the top of the page, select "Genome Annotation"



- First select the Annotations folder
- Then select one of the contig files

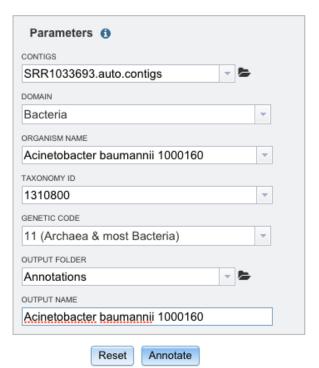




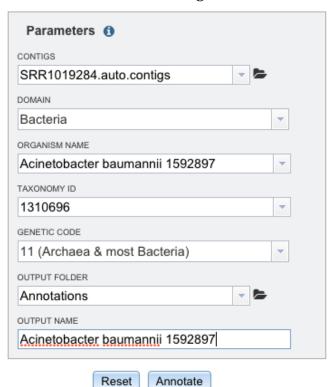
- Select SRR1033693.auto
- Fill out the form as shown below

Genome Annotation

Annotates genomes using RASTtk.



- Click "Annotate"
- Do the same for the other genome



- You can see the job status in the bottom right
- Clicking here will show the completed and queued jobs

Status	Submit	Арр	Output Name	Start	Completed
queued	6/12/15, 11:36 AM	GenomeAnnotation	Acinetobacter baumannii 1592897		
queued	6/12/15, 11:33 AM	GenomeAnnotation	Acinetobacter baumannii 1000160		
• failed	6/10/15, 3:41 PM	GenomeAnnotation	83333.1.anno	6/10/15, 3:41 PM	
completed	6/3/15, 4:37 PM	GenomeAnnotation	E_Coli_TEST	6/3/15, 4:37 PM	6/3/15, 4:42 PM

What is happening during our annotation job?

We are using a version of RAST that has been customized for PATRIC

- Calling rRNAs (16S, 23S, 5s)
- Calling tRNAs with tRNAscanSE
- Searching for repeat regions
- Finding special proteins
 - Selenoproteins
 - Pyrrolysylproteins
- If it is a Streptococcus genome, it will find special repeats
- Calling CRISPRs
 - clustered regularly interspaced short palindromic repeats
- Calling protein-encoding genes
 - Prodigal
 - Glimmer
- Assigning function
 - First attempt: annotates against CoreSEED
 - Second attempt: annotates against FIGFams
 - Third attempt: BLAST against close relatives
- Overlapping genes are resolved
- Annotates matches to ARDB, CARD, virulence factors, human homologs
- Assigns proteins to FigFams
- Finds closest neighbors

Step 5. Let's look at our annotation output

• If you click on the annotation job you will see job details:

Genome Annotation Job Result

Genome	Organism (Acinetobacter baumannii 1000160), Domain (Bacteria), Annotation ID (1310800.5), Feature count (3864)
Start time	6/12/15, 1:43 PM
End time	6/12/15, 2:31 PM
Run time	47m32s
Parameters	{"scientific_name":"Acinetobacter baumannii 1000160","domain":"Bacteria","contigs":"/PATRIC_Workshop@patricbrc.org/home/Assemblies/A baumannii 1000160/.SRR1033693.auto/SRR1033693.auto.contigs","output_file":"Acinetobacter baumannii 1000160","output_path":"/PATRIC_Workshop@patricbrc.org/home/Annotations","taxonomy_id":"1310800","code":"11"}

• And a set of files that you can download:

	Filename	Туре	File size
L	Acinetobacter baumannii 1000160.gff	gff	593.3 k
L	Acinetobacter baumannii 1000160.txt	string	5.3 MB
	Acinetobacter baumannii 1000160.embl	embl	8.4 MB
_	Acinetobacter baumannii 1000160.tar.gz	string	2.2 MB
	Acinetobacter baumannii 1000160.feature_protein.fasta	feature_protein_fasta	1.6 MB
_	Acinetobacter baumannii 1000160.merged.gb	genbank_file	8.1 MB
_	Acinetobacter baumannii 1000160.gb	genbank_file	8.0 MB
	Acinetobacter baumannii 1000160.xls	string	5.6 MB
_	Acinetobacter baumannii 1000160.features.txt	feature_table	573.1 k
_	Acinetobacter baumannii 1000160.genome	genome	10.6 ME
_	Acinetobacter baumannii 1000160.contigs.fasta	contigs	4.0 MB
	Acinetobacter baumannii 1000160.feature_dna.fasta	feature_dna_fasta	3.9 MB
_	sp_gene.json	json	104.1 k
_	load_files	folder	0 B
	genome.json	json	1.1 kB
	genome_feature.json	json	8.7 MB
_	genome_sequence.json	json	3.9 MB
_	pathway.json	json	2.5 MB

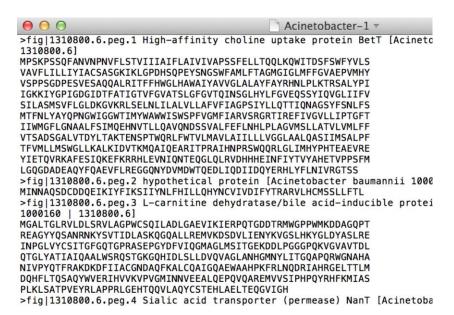
- If you click on the icon button, you will download the files to your folders.
- Typically, you will want the genbank or fasta formatted file.
- The .json and .genome files are java script files that are used by our system

٠	Acinetobacter baumannii 1000160.xls	string	5.6 MB
_	Acinetobacter baumannii 1000160.feature_dna.fasta	feature_dna_fasta	3.9 MB
٠	Acinetobacter baumannii 1000160.feature_protein.fasta	feature_protein_fasta	1.6 MB
_	Acinetobacter baumannii 1000160.gff	gff	593.3 kB
¥	load_files	folder	0 B
٠	genome.json	json	1.1 kB

• The download box may look different if you are using a PC

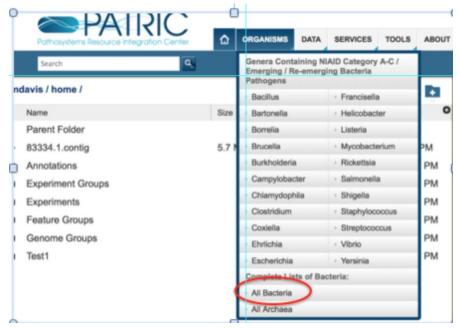


Using your favorite text editor you can see the proteins in fasta format (in this case).



Step 6. How do we find our private genome on the PATRIC website?

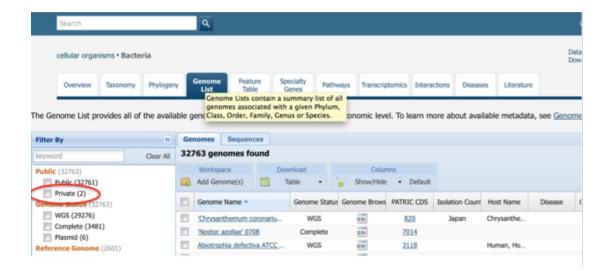
- When you annotate a private genome in PATRIC genome, it gets indexed in our database, but only you can see it
- The ability to share genomes is coming soon
- To see your private genomes, First click on the "Organisms" tab at the top of the page
- Then click on "All Bacteria"



- This will bring up the overview page for all bacteria in the database
- Click the "Genome List" tab



- This will bring up the list of all genomes
- You can filter the genome list by clicking on the checkboxes on the left hand panel
- Click the checkbox that says "Private"



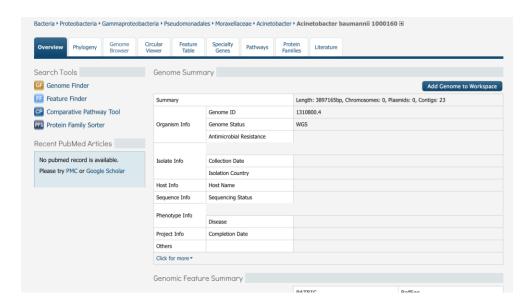
This will filter the list to the two Acinetobacter genomes that we annotated



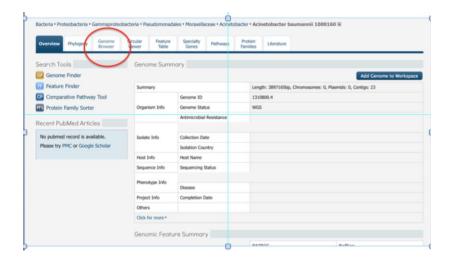
- Typically, when you browse the public genomes, you can select columns
- Being able to add these columns for private genomes is coming soon

Step 7. Let's look at the genomes

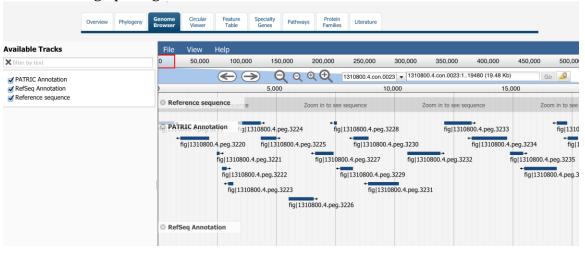
- First click on one of the genomes
- This will load the genome landing page



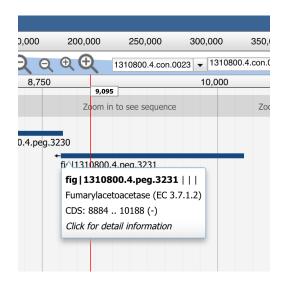
At the top of the page click on the "Genome Browser" tab



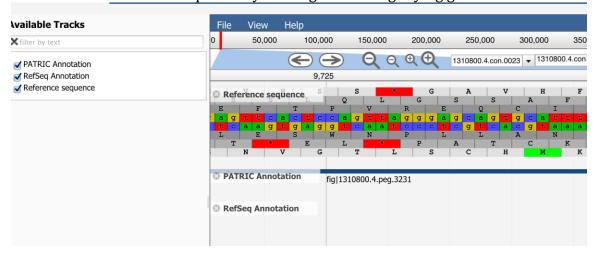
This will bring up the genome browser



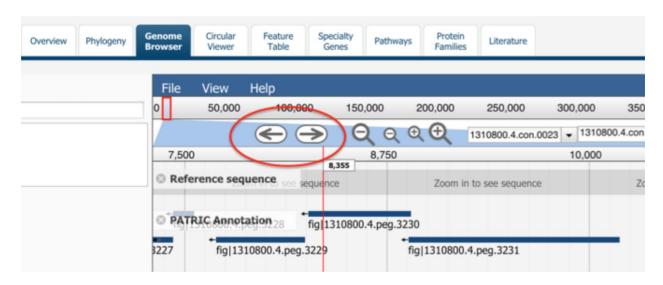
- Doing a mouse over will show the annotation of the gene.
- Arrows display direction of transcription.



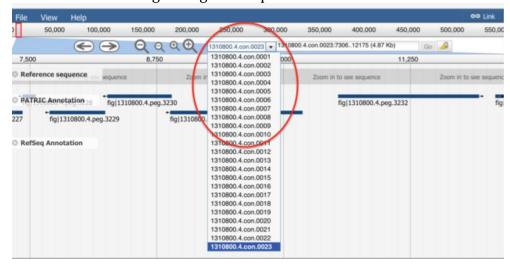
You can zoom to the sequence by clicking on the magnifying glass icon



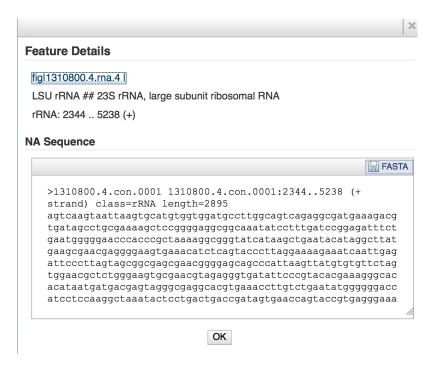
• You can walk the contig by clicking on the arrow buttons



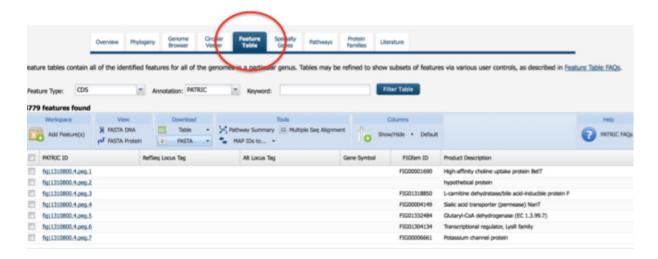
You can switch contigs using the dropdown menu



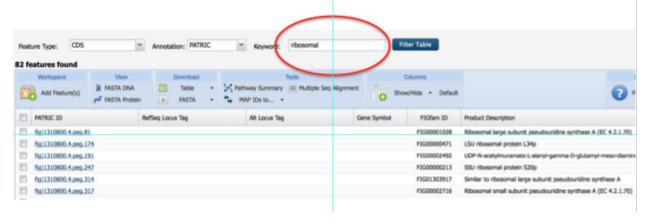
If you click on a gene, it will display the sequence in a pop up



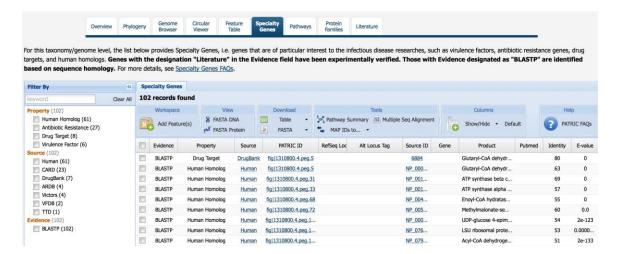
You can click on the "feature table" tab to view a table of all features in the genome



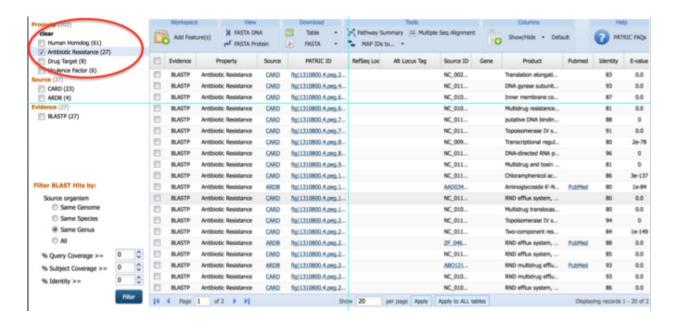
You can perform a keyword search to filter the table.



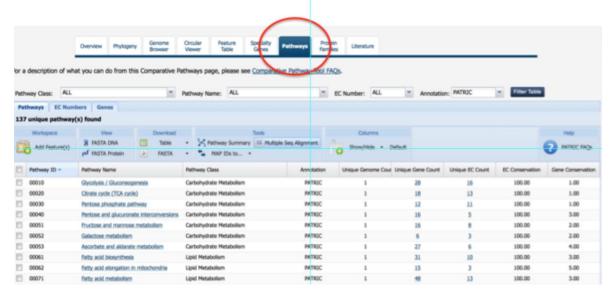
- Let's look at those weird specialty genes that we annotated
- Click on the "Specialty Genes" tab



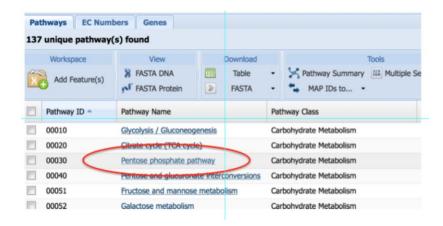
- Membership in this list is based on BLAST similarity to a reference database
- You can filter the list by database, blast score, and closest matching organism



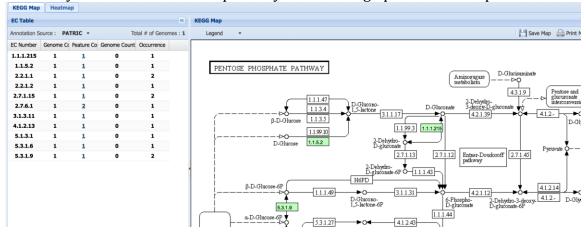
• If you click on the "Pathways" tab you can see all the genes with annotations that map to KEGG



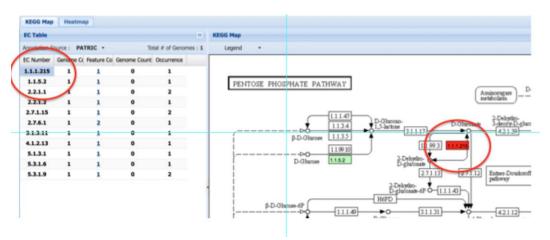
• If you click on one of the pathways it will bring up the KEGG map



If you click on one of the pathways it will bring up the KEGG map



• If you click an EC number in the side bar it will highlight the enzyme in the map



- Finally you can render a diagram of the genome.
- Click on the "Circular Viewer" tab.



- You can control the layout by clicking the checkboxes. You can download a publication-quality image by clicking the download button.

