Genome Annotation with Companion (Part 1)

For this exercise we will start with an assembled genome that is unannotated. We will obtain the assembled FASTA files from EuPathDB sites.

Companion is housed at Sanger and can be accessed here:

https://companion.sanger.ac.uk

Companion, is an online pipeline that employs different software to annotate and compare an assembled sequence to a reference-annotated genome.

The figure below illustrates the Companion pipeline, the software used and the expected output.



Each group will download one of the genomes as indicated below.

Group 1 – Cryptosporidium baileyi TAMU-09Q1

http://tinyurl.com/hgtddbz

Group 2 – Cryptosporidium meleagridis UKMEL1

http://tinyurl.com/zskwvf5

Group 3 – Cryptosporidium hominis UKH1

http://tinyurl.com/zoxy48u

Group 4 – Plasmodium coatneyi Hackeri

http://tinyurl.com/je4hftj

Group 5 – Acanthamoeba palestinensis Reich (only the largest 3000 contigs)

http://tinyurl.com/j2rd9lq

All genomes in EuPathDB sites are available for download form the "Data File" download section, which you can access from the Downloads menu in the gray tool bar.

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Selecting the Data Files option takes you to the download directories where you can navigate to the genome and data type you are looking for.

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To download specific contigs/scaffolds/chromosomes you can use a genomic sequence search and place the desired sequences into your basket.

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- Once you have downloaded your sequence file, go to the Companion site: https://companion.sanger.ac.uk

- Click on the "Annotate your sequence" link.



-Follow the instructions as described on the Companion website:

1. Provide basic information about the job you are about to submit. This includes a job name, species prefix (usually the first letter of the genus and the first three letters of the species: *Acanthamoeba palestinensis* = Apal).

Submit a new annotation job

Step 1: Basic job properties					
First of all, please specify a free-text name for your new job. It should reflect the purpose of your job, and should probably include the organism you are annotating.					
Example: My new species annotation					
Job name					
Please also give a short species prefix that will be used to name entities (such as genes, pseudochromosomes, etc.) generated during the annotation run. It should not contain spaces or special characters.					
Example: LDON					
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2. In step 2, choose the assembly file that you downloaded.

3. In step 3, indicate if you will be using RNAseq evidence to guide the annotation – in this exercise we will **not** use any RNAseq data.

4. In step 4, select the reference sequence you would like to use to transfer the annotation and to compare your sequence to. Typically you would like to use a reference that is closely related, so a phylogenetic tree might be useful to look at. Here are examples for *Plasmodium* and *Cryptosporidium*. There is only one reference for *Acanthamoeba*.

http://tolweb.org/Plasmodium/68071

http://tolweb.org/Cryptosporidium/124803

Step 2: Target sequence

Please upload a **target sequence file** to be annotated from your local filesystem using the button below. The file (FASTA, EMBL or GenBank format) can be gzip- or bzip2-compressed. In this case it must have a .gz or .bz2 suffix.

Note: The maximal size of your uploaded file is 64 MB, and the maximum number of individual sequences in it is 3000.

Choose File no file selected

Here is an example sequence input file for a *Plasmodium falciparum* IT chromosome 5 sequence that can be used with the *Plasmodium falciparum* 3D7 example reference set (choose below in step 4) for a quick example run. To use it, please download it to your local machine and upload it using the button above.

Step 3: Transcript evidence

The Companion pipeline can optionally make use of assembled transcripts in the GTF format as created by Cufflinks.

○ Yes, use transcript evidence.

No, do not use transcript evidence.

Step 4: Reference organism

Please pick a (if possible closely related) reference organism for this annotation run. This organism will be used to specify the models for gene finding, functional annotation transfer and pseudochromosome contiguation.

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Please select a reference species

5. In step 5, there a few more parameters you may one to examine. For the purpose of our exercise we will keep these at the default values.

Step 5: Pseudochromosome contiguation
The contiguation step will try to orientate the sequences in your input file to align with the chromosomal sequences of the reference organism to build pseudochromosomes, which will then be used as the target sequences for gene annotation. This step is optional; if it is not desired then no modifications will be made to the input sequences.
Yes, contiguate pseudochromosomes.
○ No, do not modify my input sequences.
Select minimum required match length for contig placement: 500 bp
200 20000
Select minimum required match similarity for contig placement: 85 %

6. Enter your email address to get an update when your job starts running and when it is complete. Next, click on the "I'm not a robot" captcha (Completely Automated Public Turing test to tell Computers and Humans Apart). Finally, click on the "Submit Job" link.

Step 6: Advanced settings (click chevron to the right to show/hide)	*
Your contact information (optional)	
You can leave your email address if you want to be notified when your job starts and finishes. This is absolutely optional, if you choose not t share your email address, you can always manually check the status of your job using a private link provided by us after submission.	D
Email	

To protect the service from automated bots, please prove that you are a human by ticking the box below.



Submit job