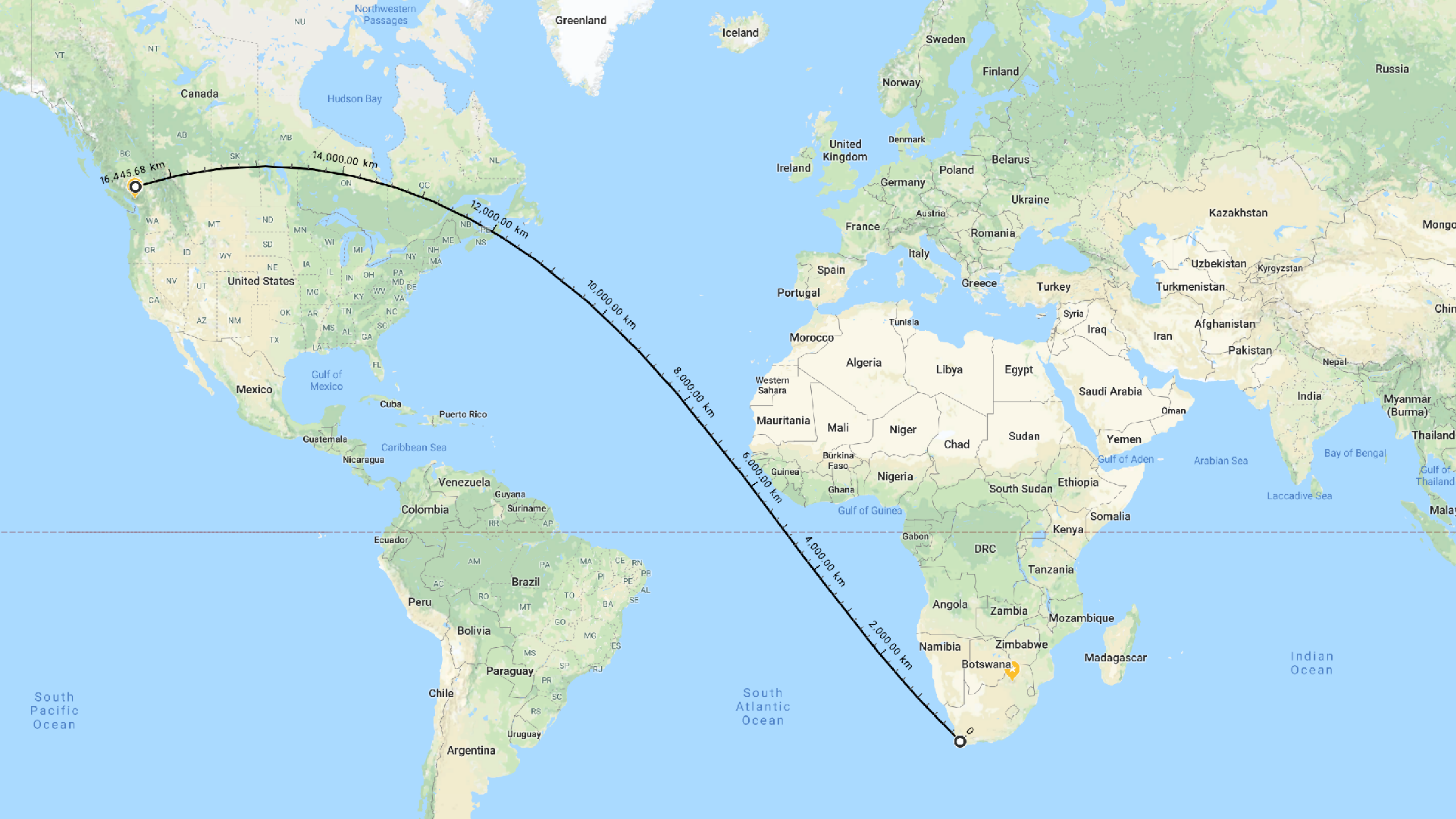


A thousand genomes in a Galaxy server far, far away...

Charles Hefer
Galaxy Africa
3-5 April 2018







Genetic Improvement of Poplar Trees as a Canadian Bioenergy Feedstock



Carl Douglas



Shawn Mansfield



Quentin Cronk



GenomeCanada

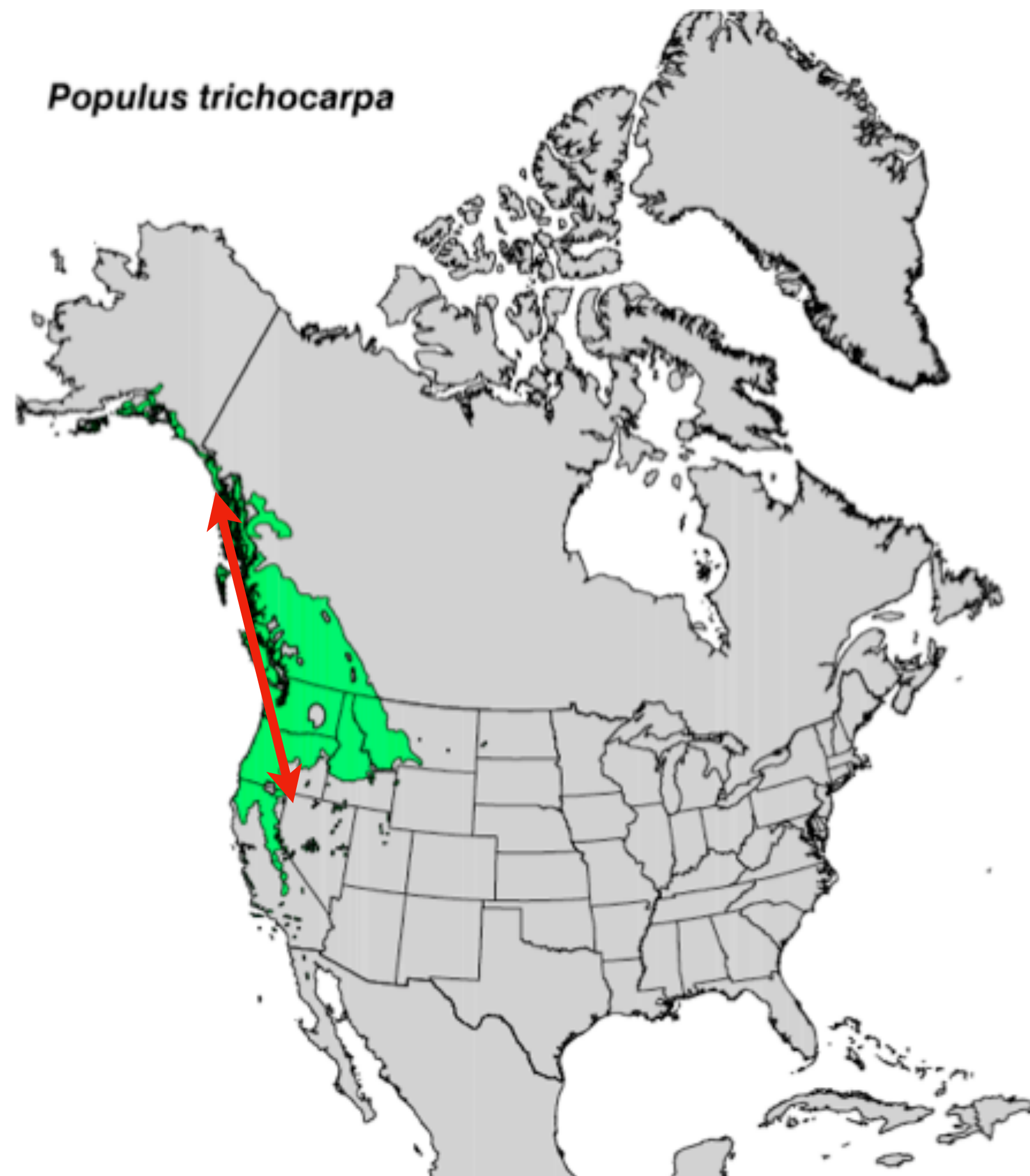
Poplar genome resources

- Poplar - First woody genome sequenced (back in 2006)
 - Followed by *Eucalyptus*, *Pinus*, *Picea*
 - Relatively small genome ~460Mbp
 - 19 Chromosomes
 - 41,335 loci and 73,031 proteins (www.phytozome.org)
- Genus consists of 29 species (6 sections)

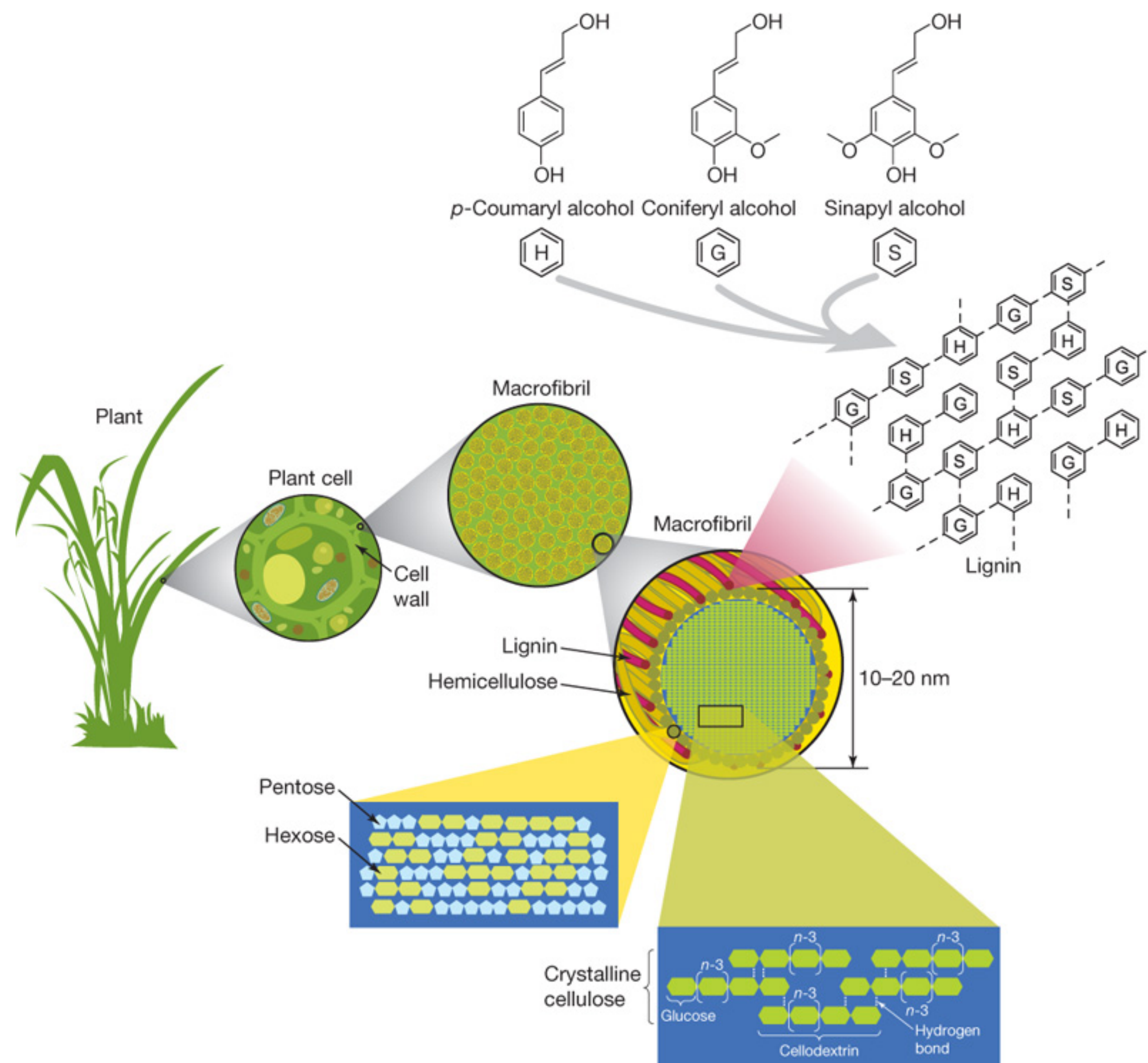


Tuscan *et al*, 2006

Poplar



Lignocellulose as bioenergy feedstock



Poplar as a cellulose crop

Cellulose crop	ton/ha/yr
Sorghum	40 - 55
Sugarcane	80 - 120
Switchgrass	14 - 18
Miscanthus	30 - 41
Corn Stover	3 - 5
Poplar	12 - 24



The PopCan project

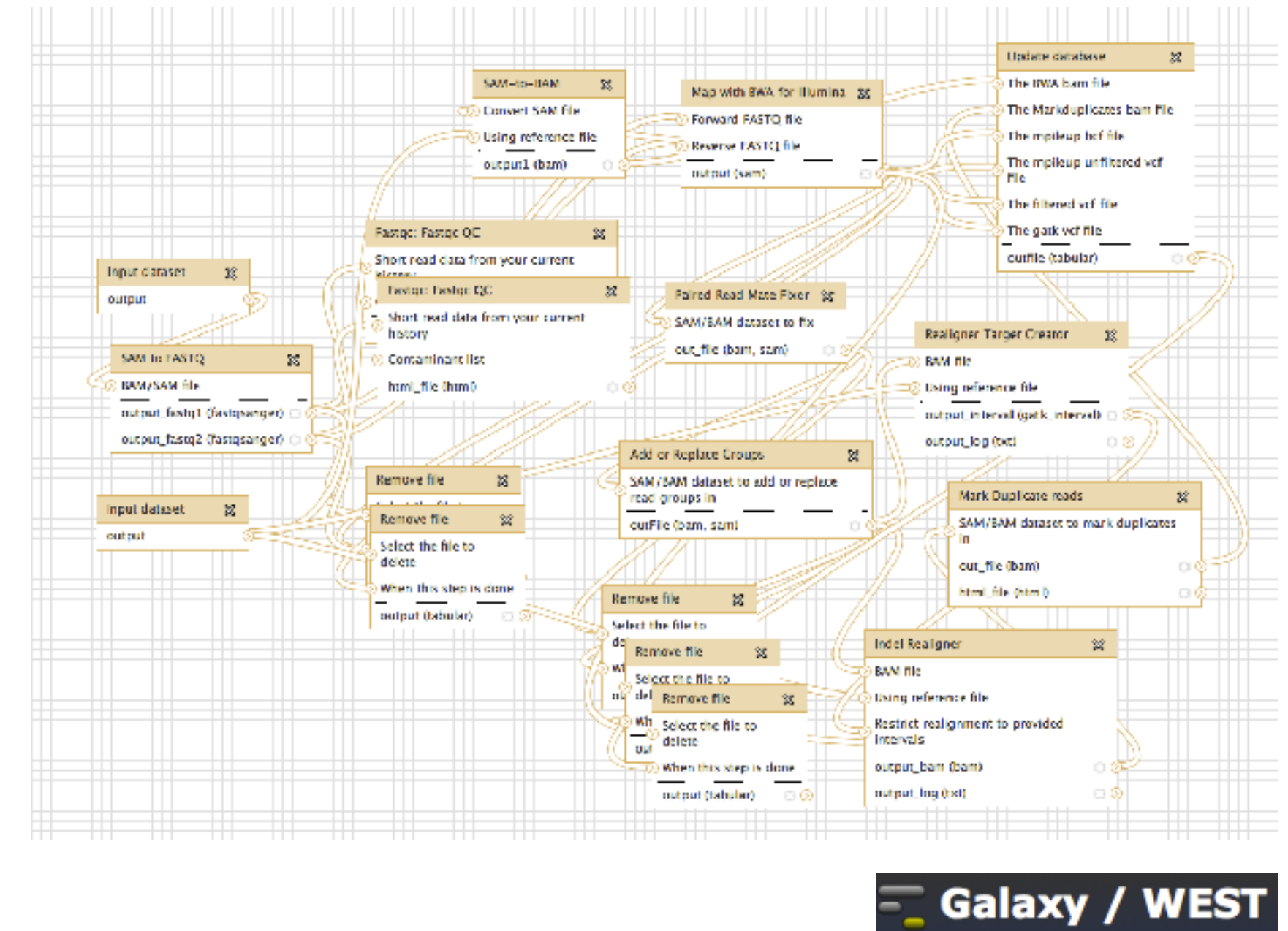
- Genetic improvement of Poplar sp. for biofuel production
- 2 Main aims
 - Search for adaptive traits within and between species
 - Search for trait associated with:
 - growth
 - wood properties (cellulose to ethanol)
- In essence: Characterize poplar in terms of variation, adaptive traits and functional elements associated with growth and wood properties



P. trichocarpa

Genome Resequencing

- 1,113 samples sequenced over 3 years
 - Between 15X and 30X coverage
 - Illumina HiSeq 2000
- Samples completed in batches
 - rsync from sequencing centre
- Single workflow needed to handle all the samples.
 - Genome resequencing, focus on SNP discovery



Hardware



R910

64 CPUs
512Gb RAM



R910

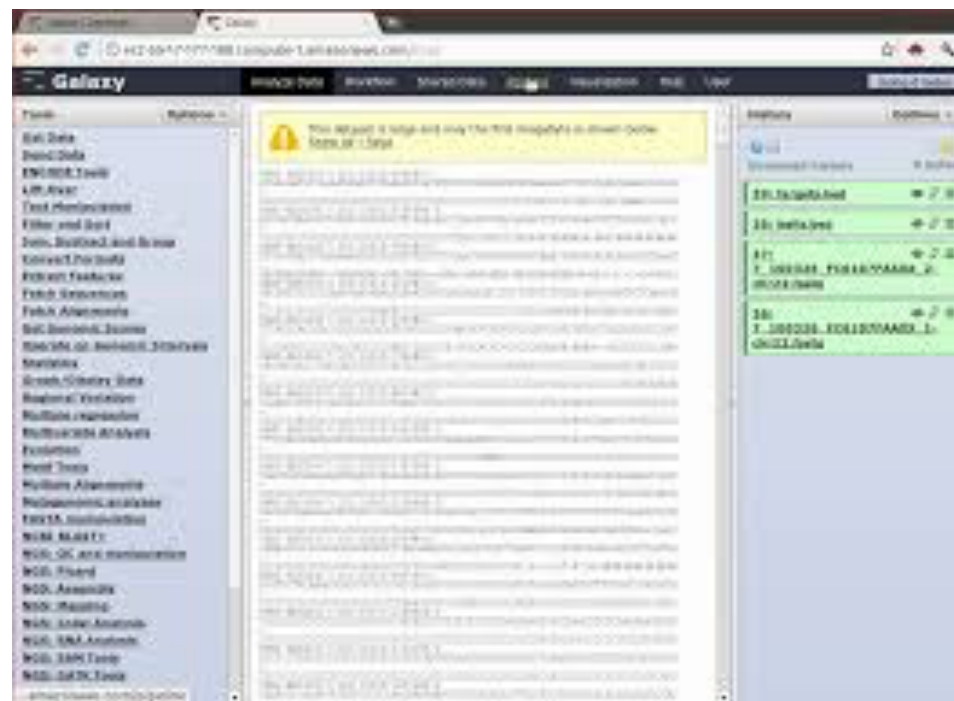


72Tb

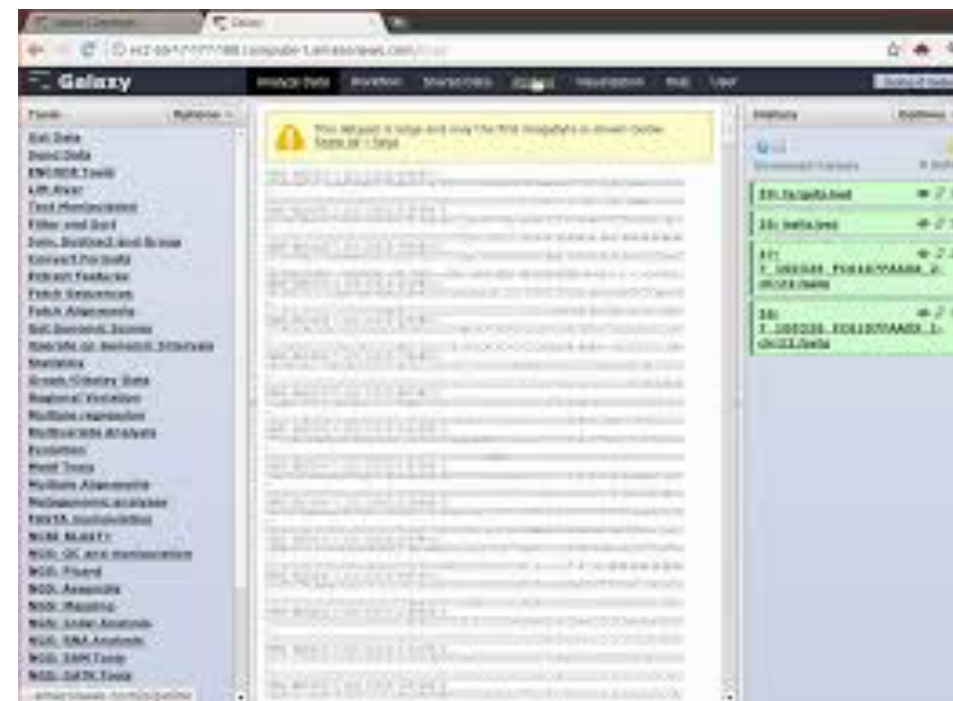
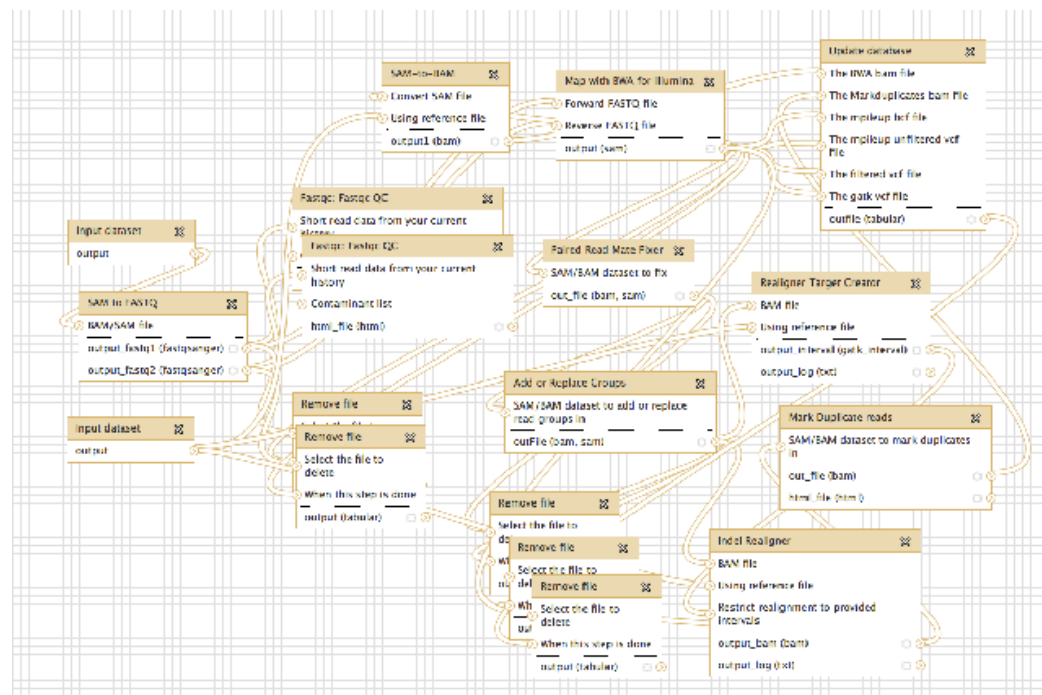
72Tb



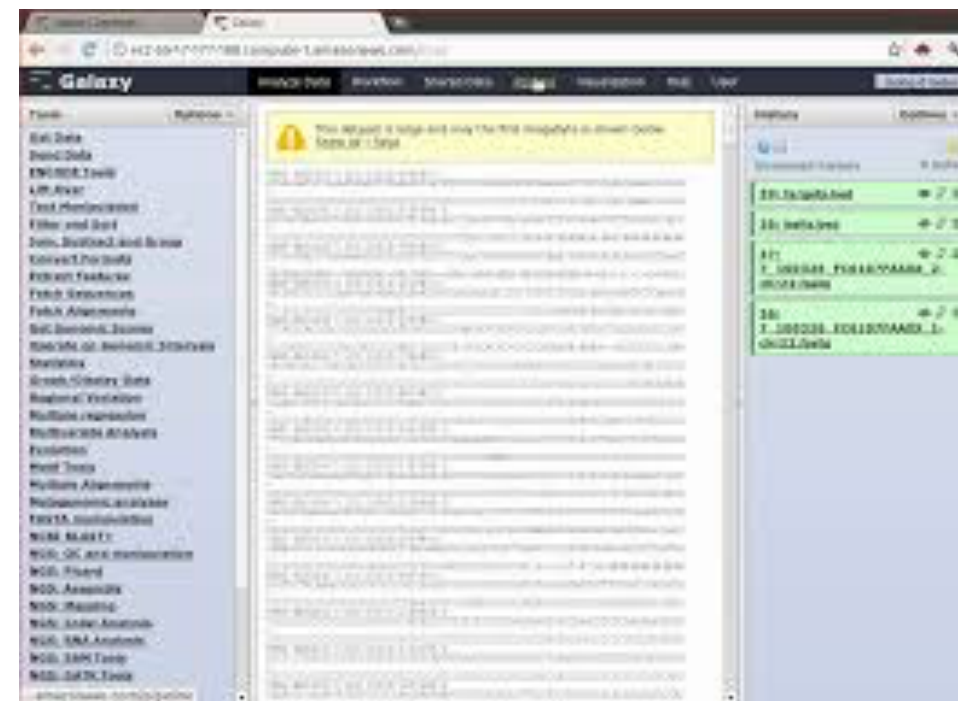
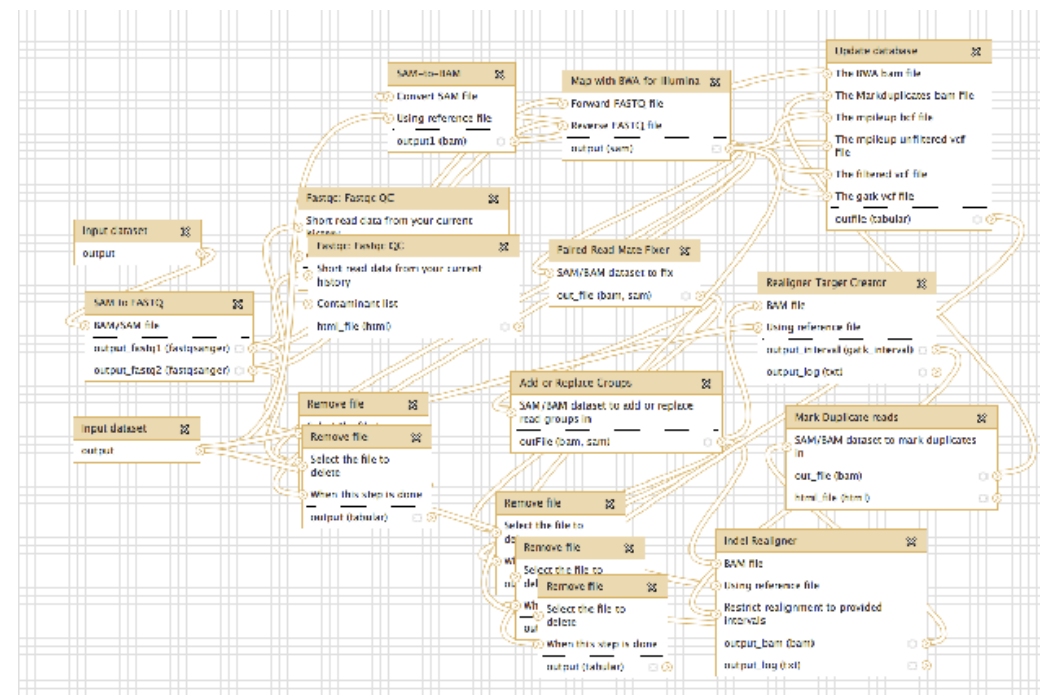
Software



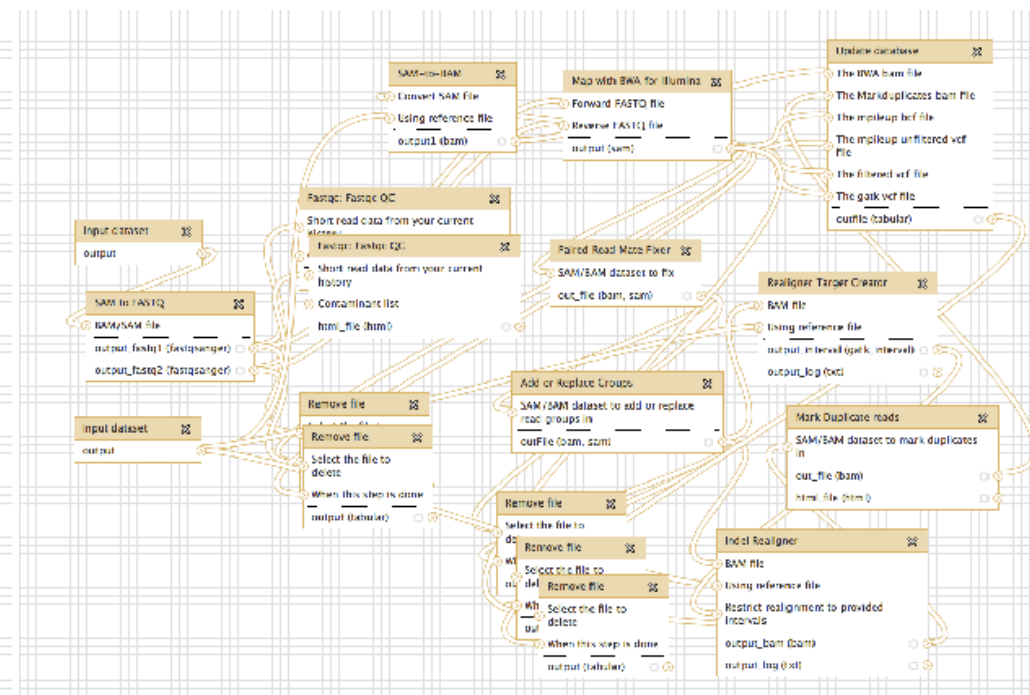
Whole Genome



Transcriptome

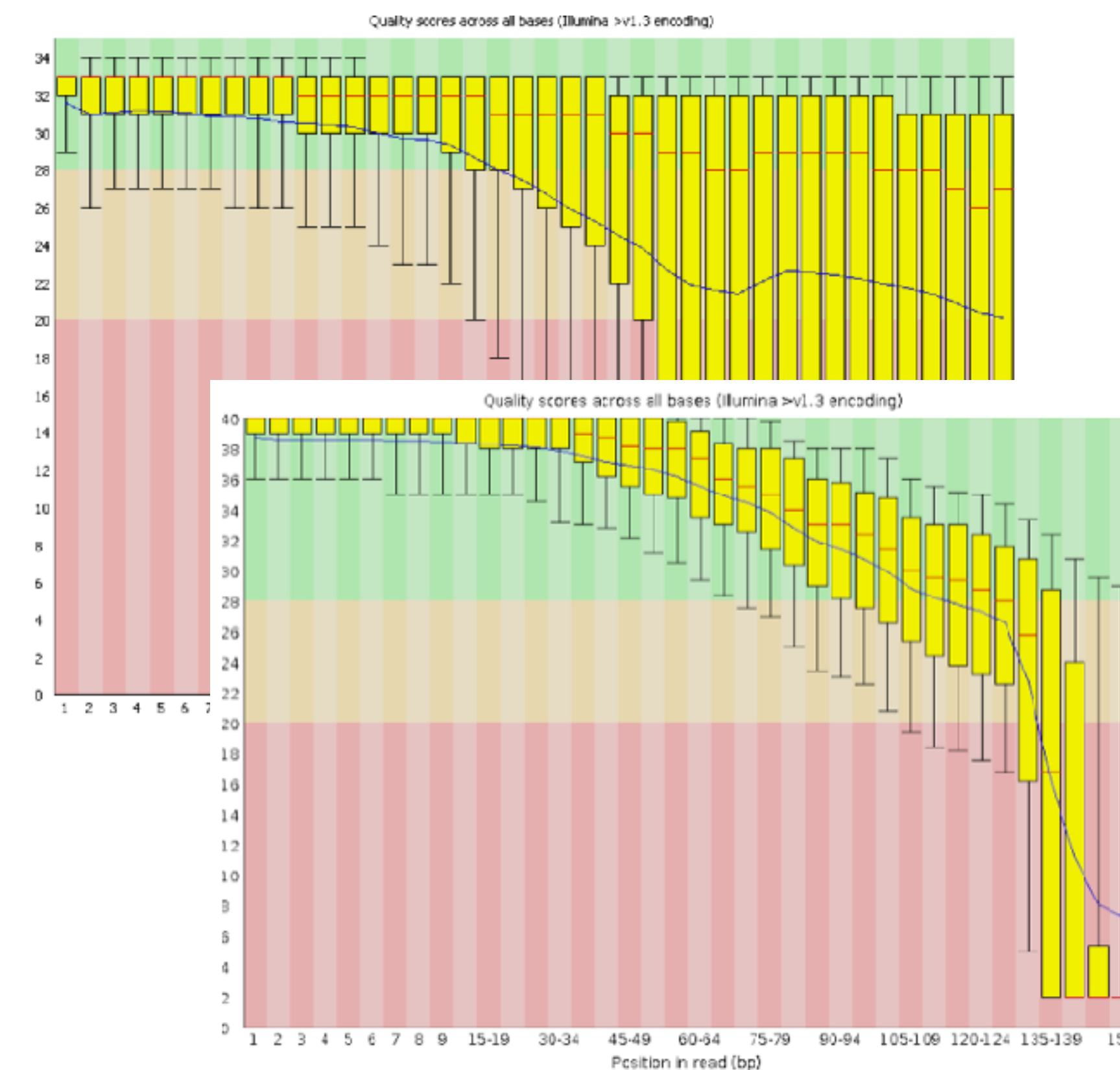


BiSulfite

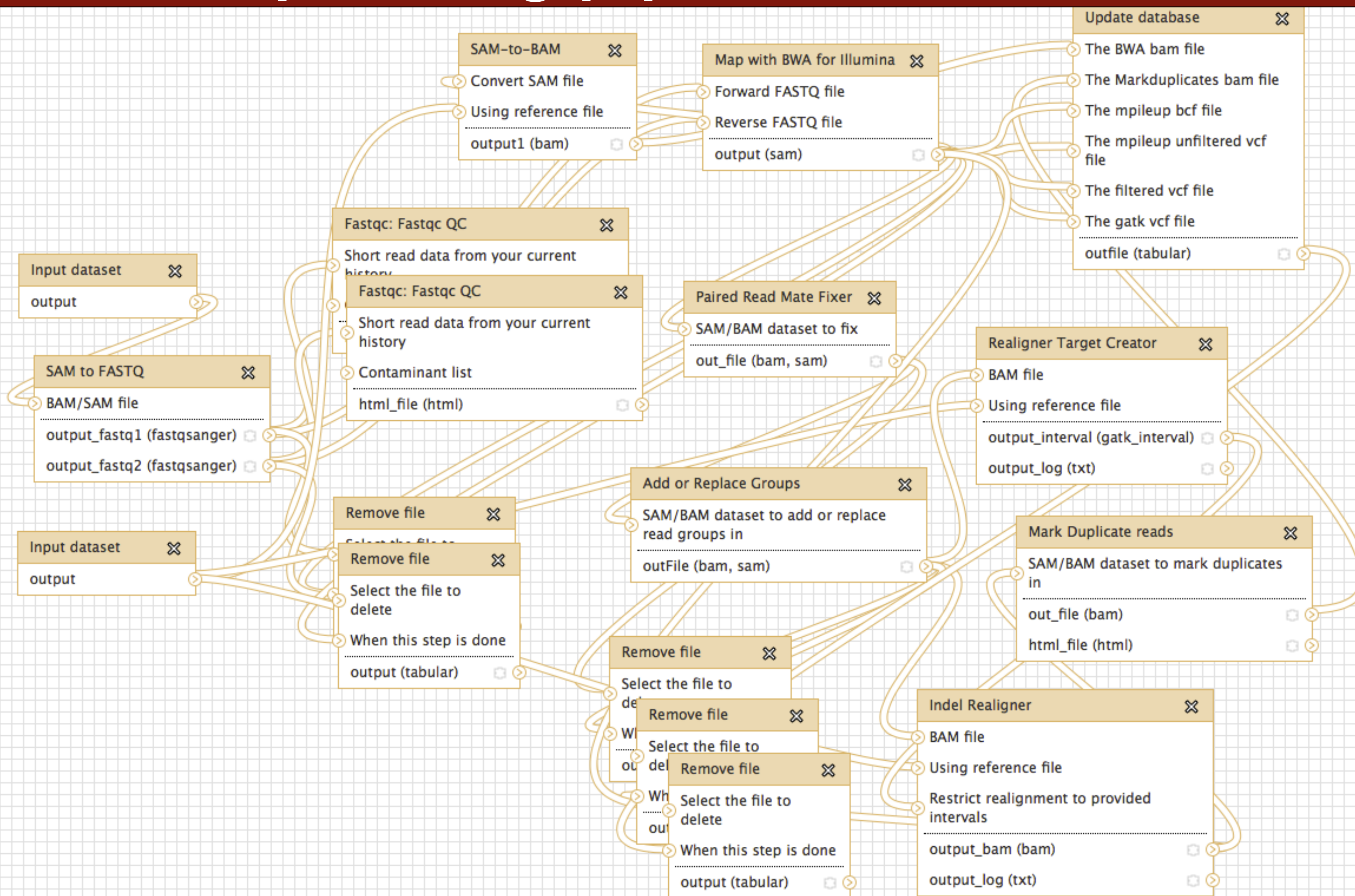


Research team setup

- Small bioinformatics team (1)
- Quick verification of input data
 - Quality control -> within 14 days
 - Data only available for 30 days on FTP server
- Research team setup
 - Very specific samples needed to be processed fast and distributed to postdocs
 - Verification of samples -> IBD from SNPs
 - Duplicate samples

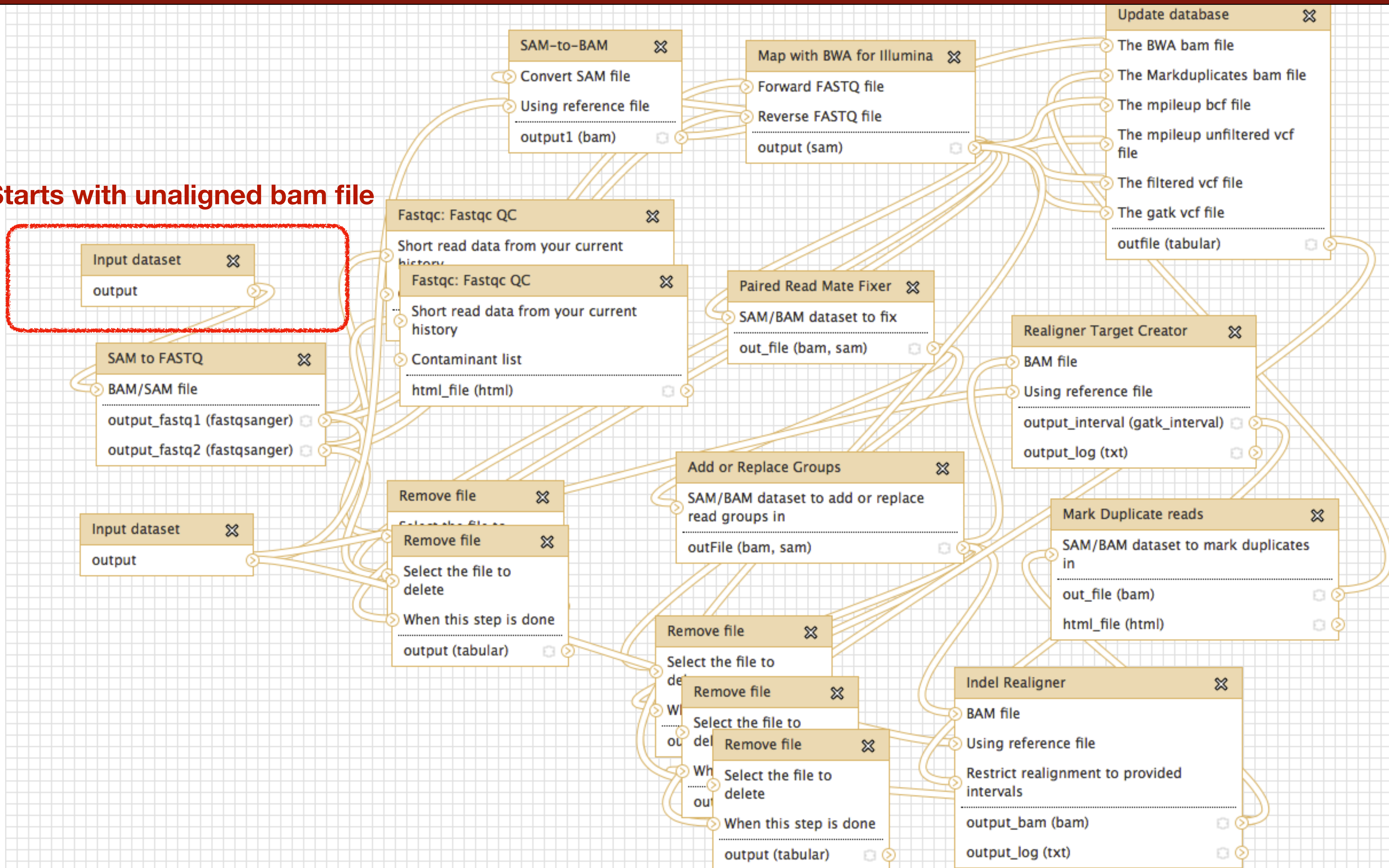


Genome Resequencing pipeline

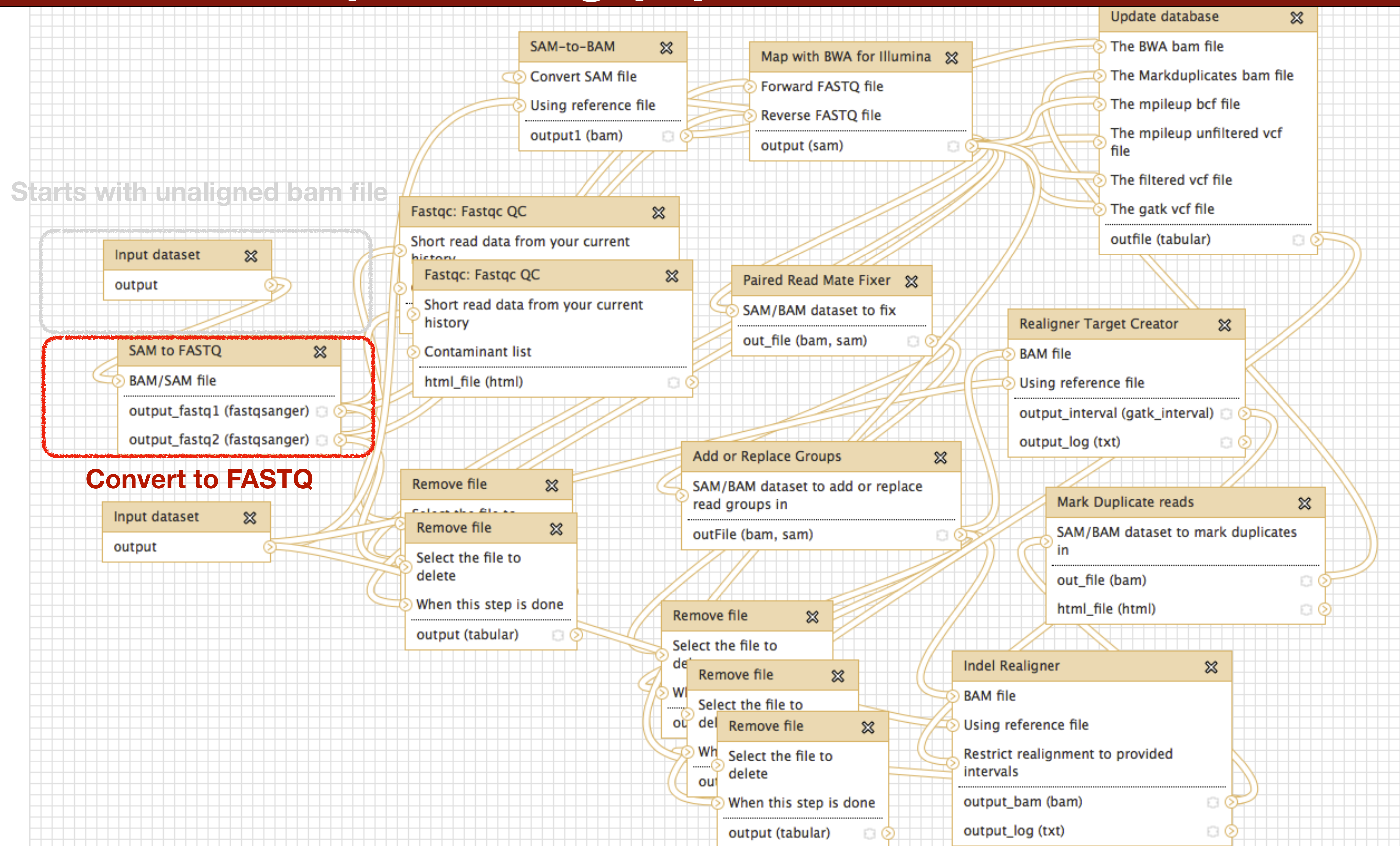


Genome Resequencing pipeline

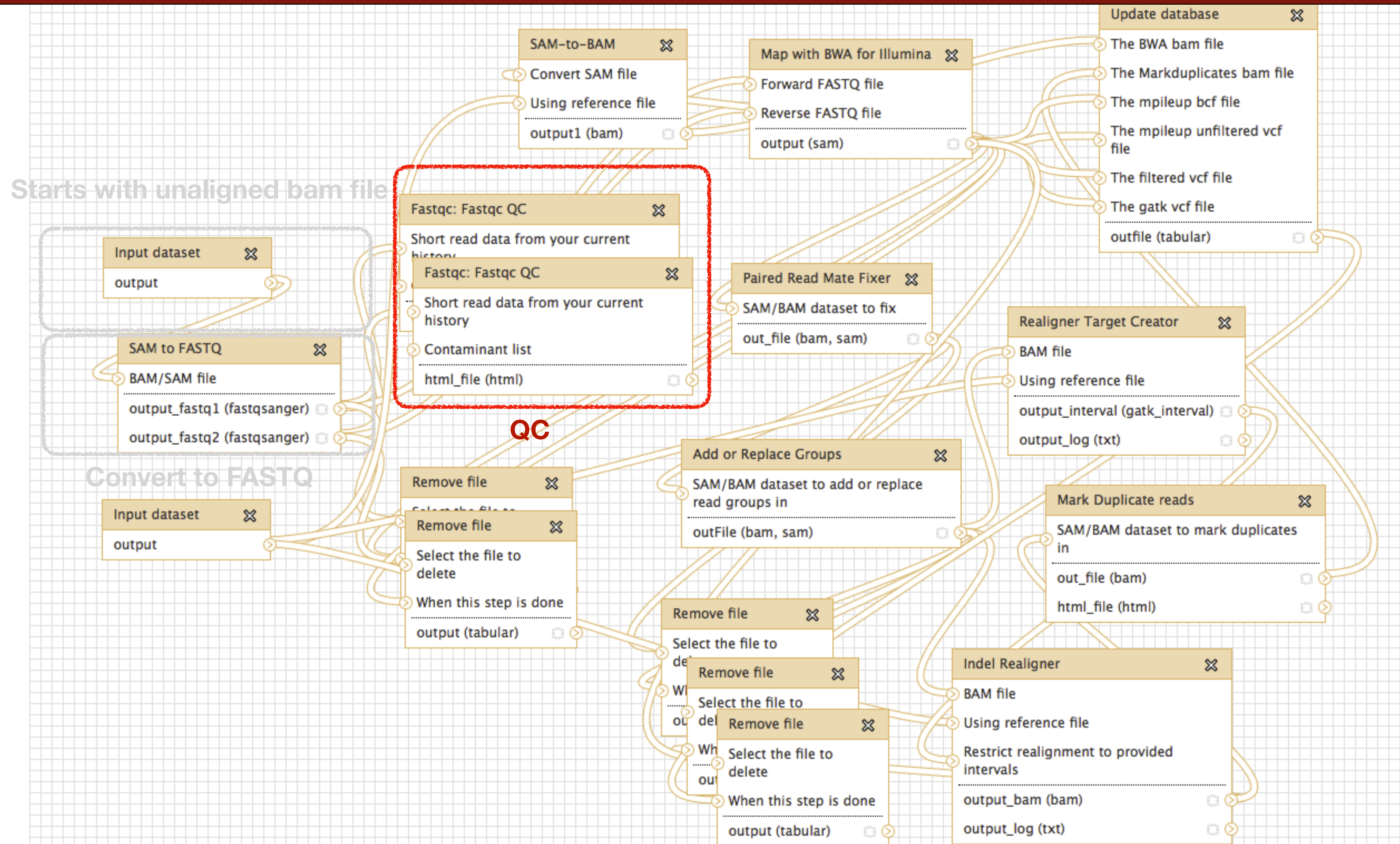
Starts with unaligned bam file



Genome Resequencing pipeline



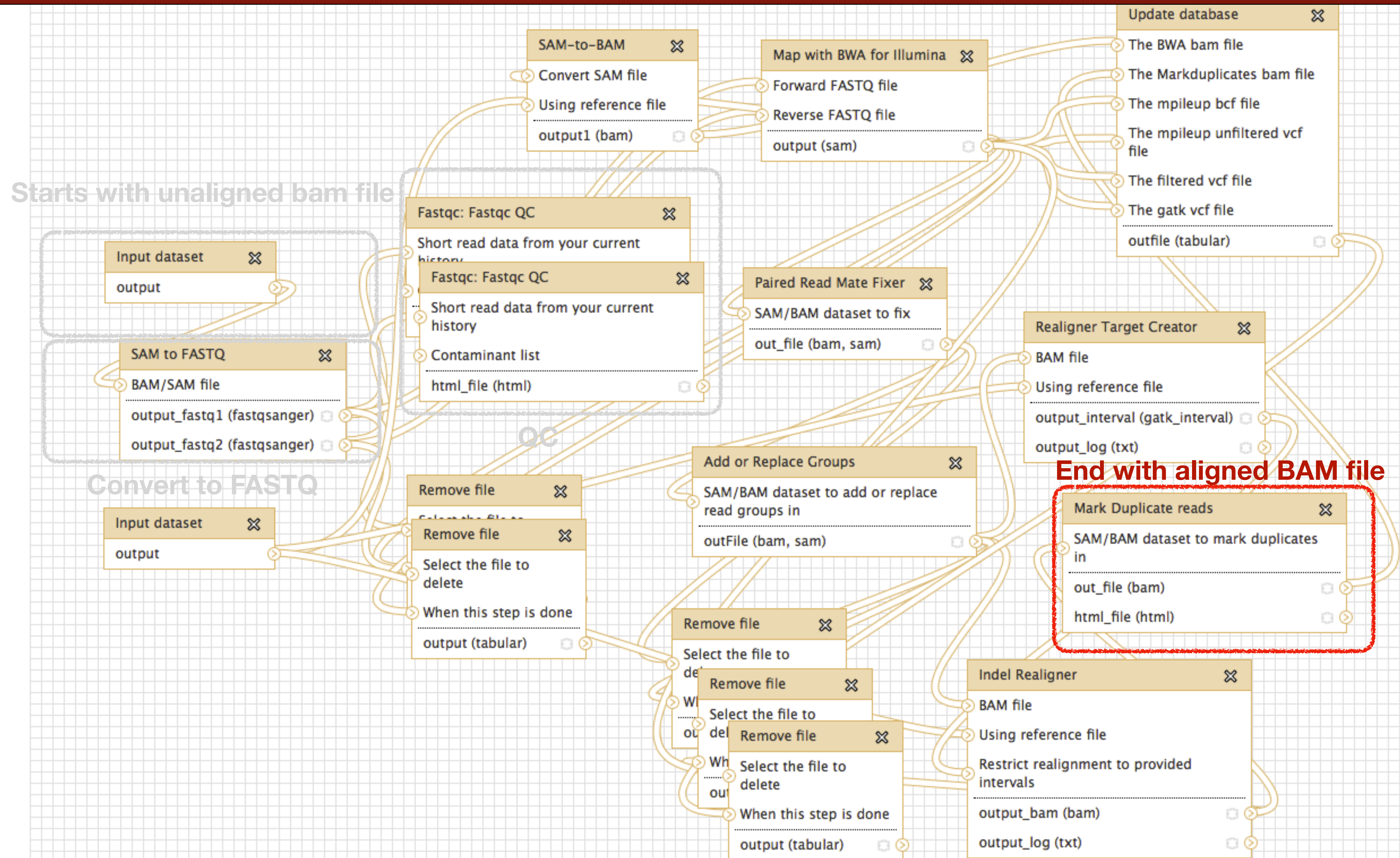
Genome Resequencing pipeline



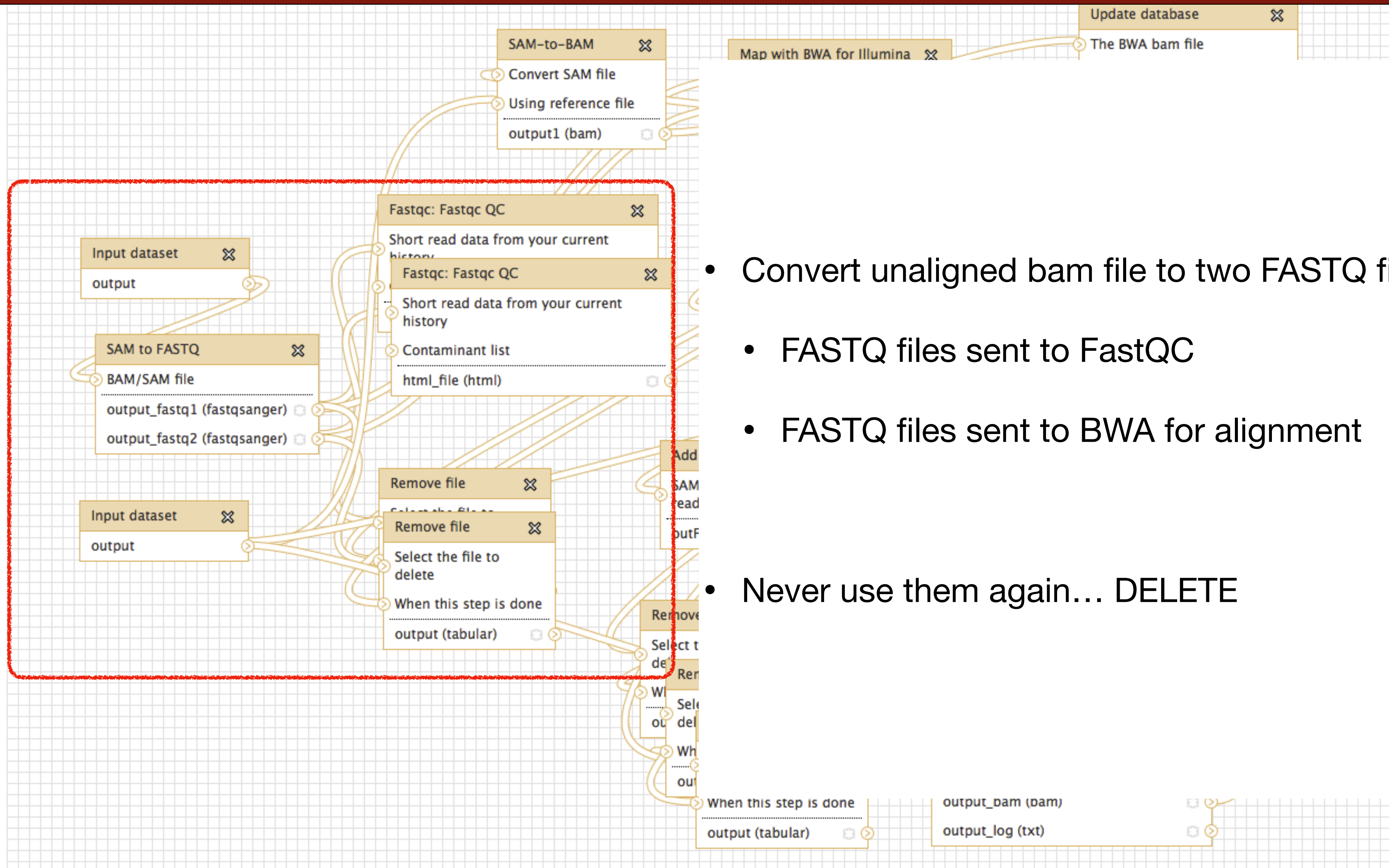
Genome Resequencing pipeline



Genome Resequencing pipeline

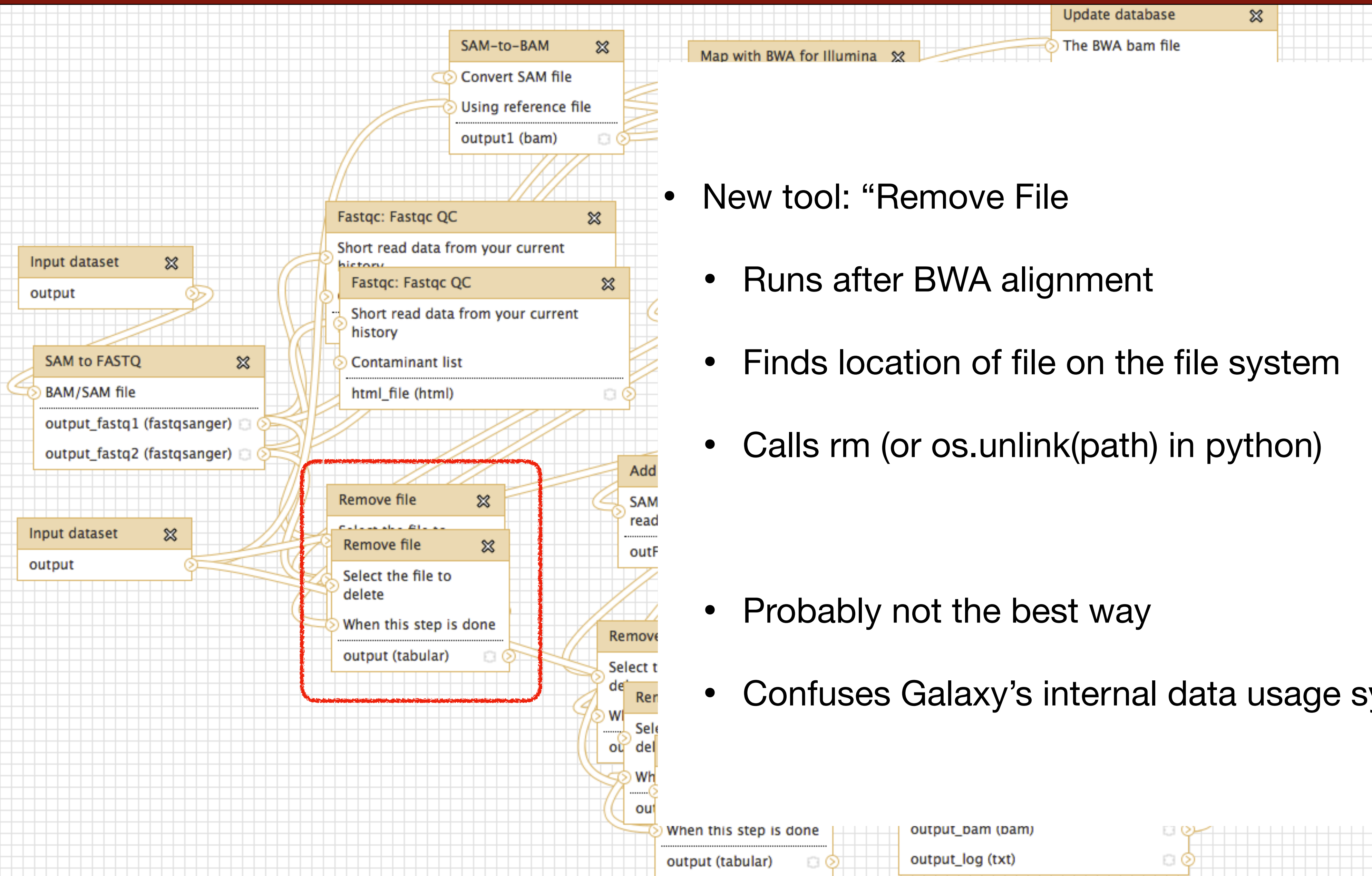


Genome Resequencing pipeline



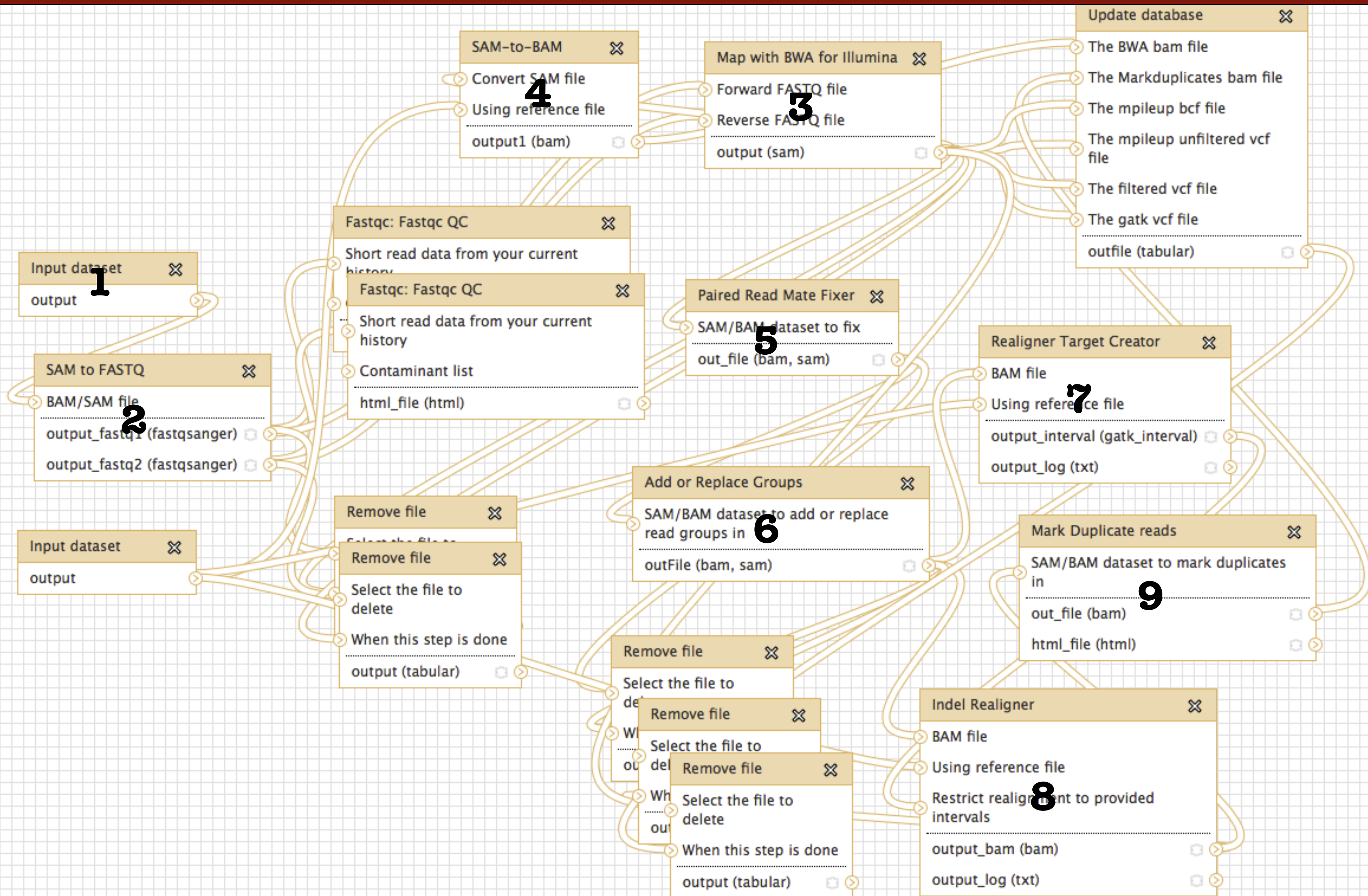
- Convert unaligned bam file to two FASTQ files
 - FASTQ files sent to FastQC
 - FASTQ files sent to BWA for alignment
- Never use them again... DELETE

Genome Resequencing pipeline

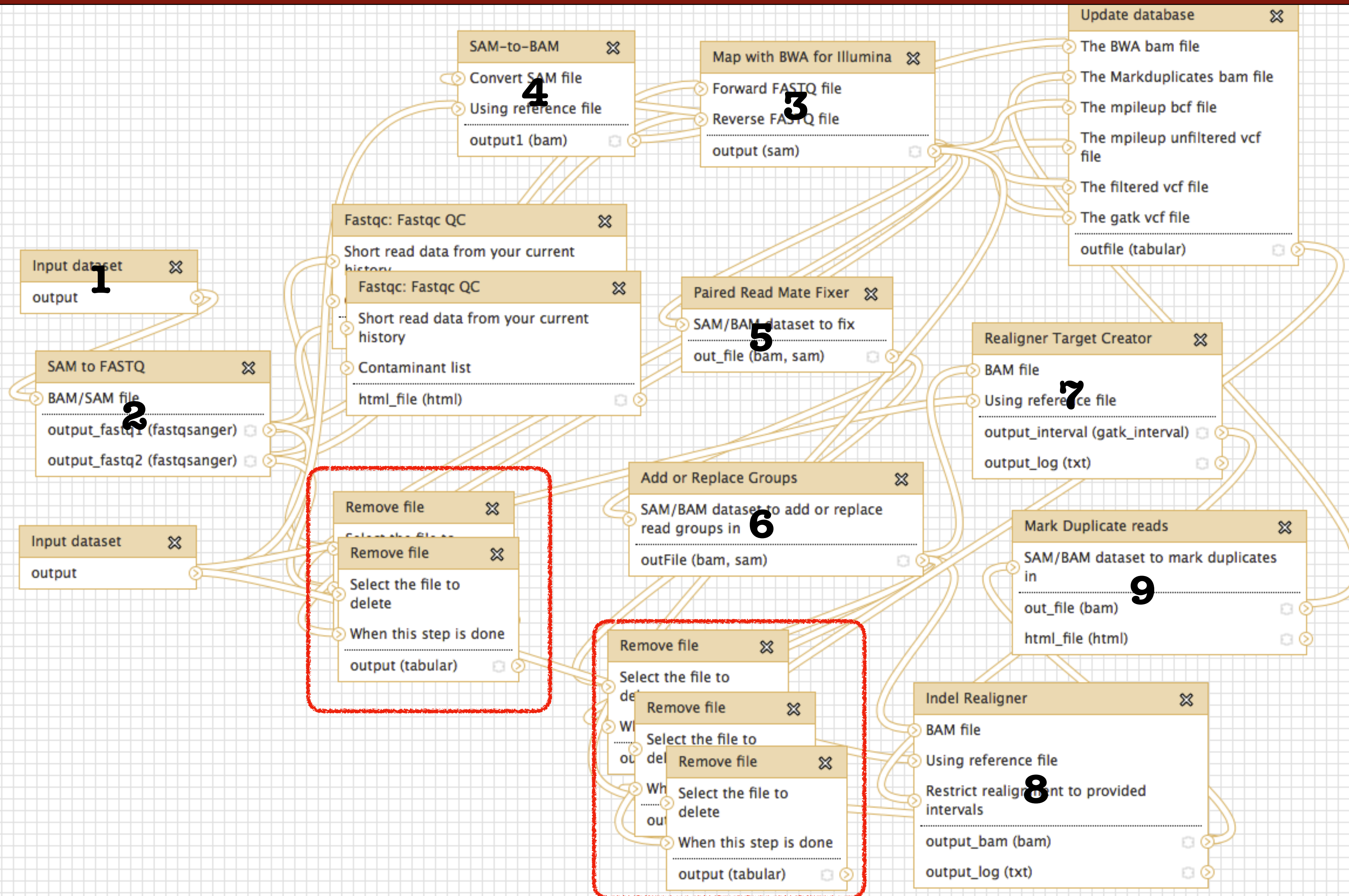


- New tool: “Remove File”
 - Runs after BWA alignment
 - Finds location of file on the file system
 - Calls `rm` (or `os.unlink(path)` in python)
- Probably not the best way
- Confuses Galaxy’s internal data usage system

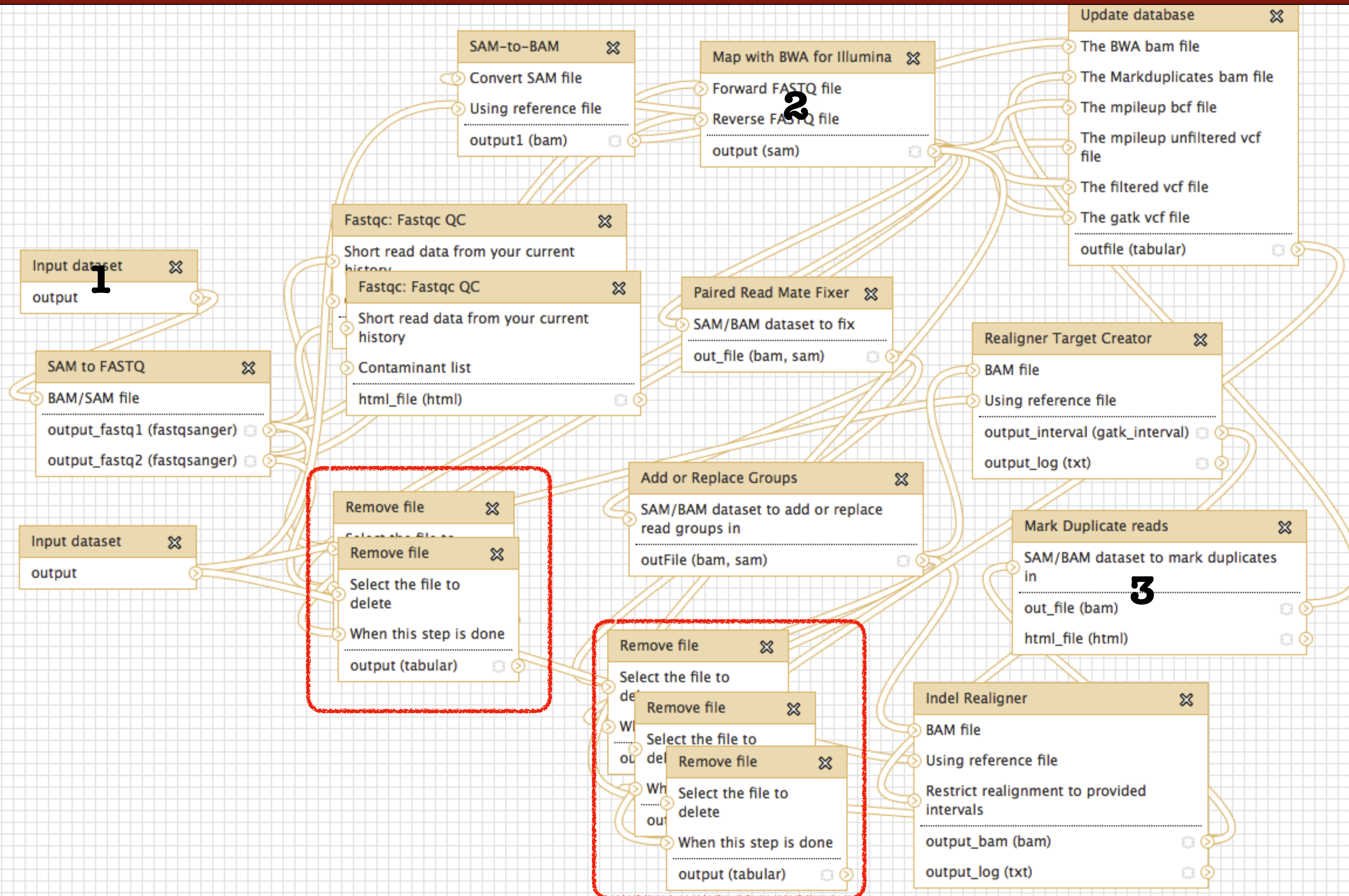
Genome Resequencing pipeline



Genome Resequencing pipeline

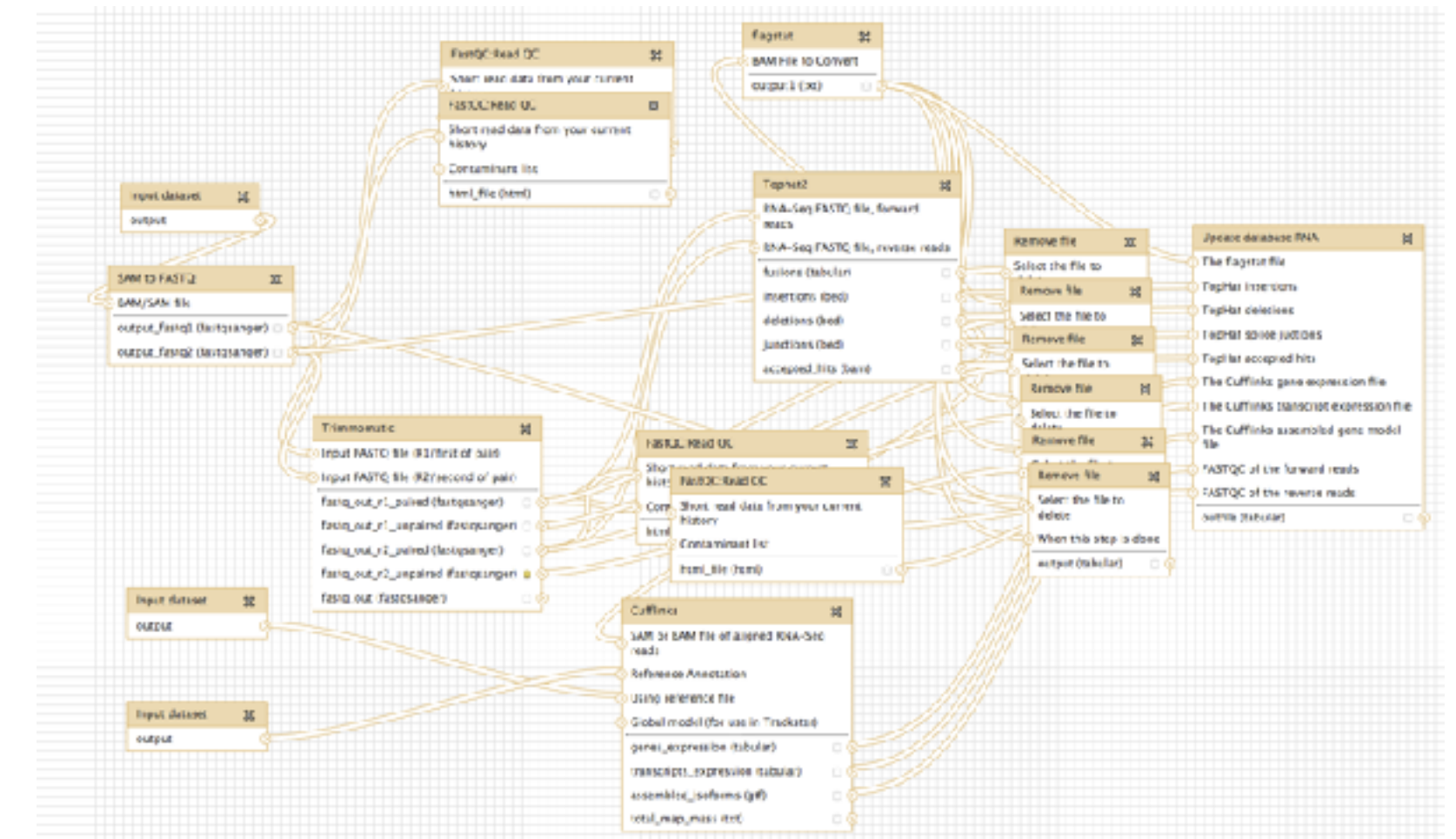


Genome Resequencing pipeline



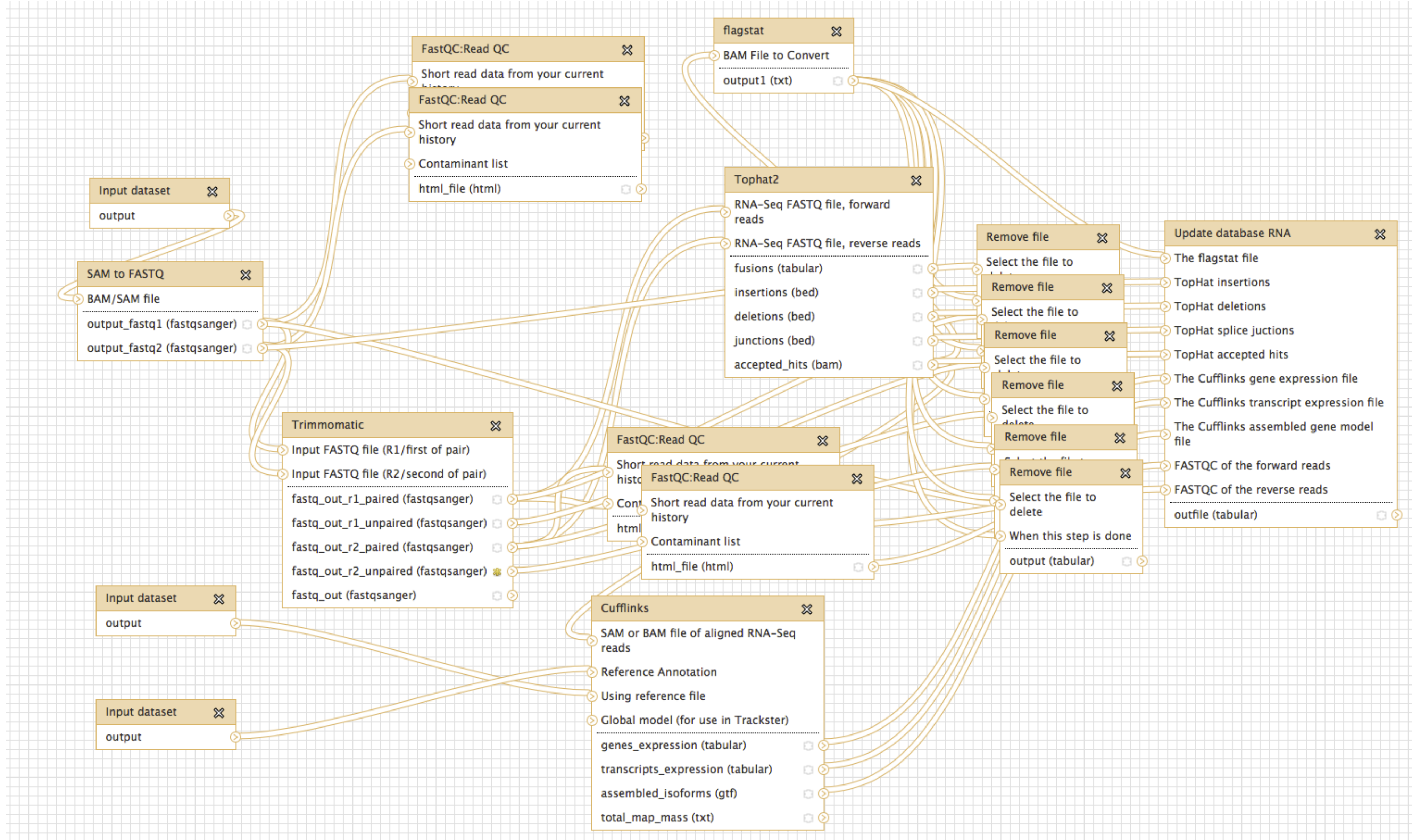
Transcriptome Sequencing

- 866 transcriptome samples
 - Between 40 and 80 million reads
 - Illumina HiSeq 2000
-
- Single workflow needed to handle all the samples.
 - Gene expression

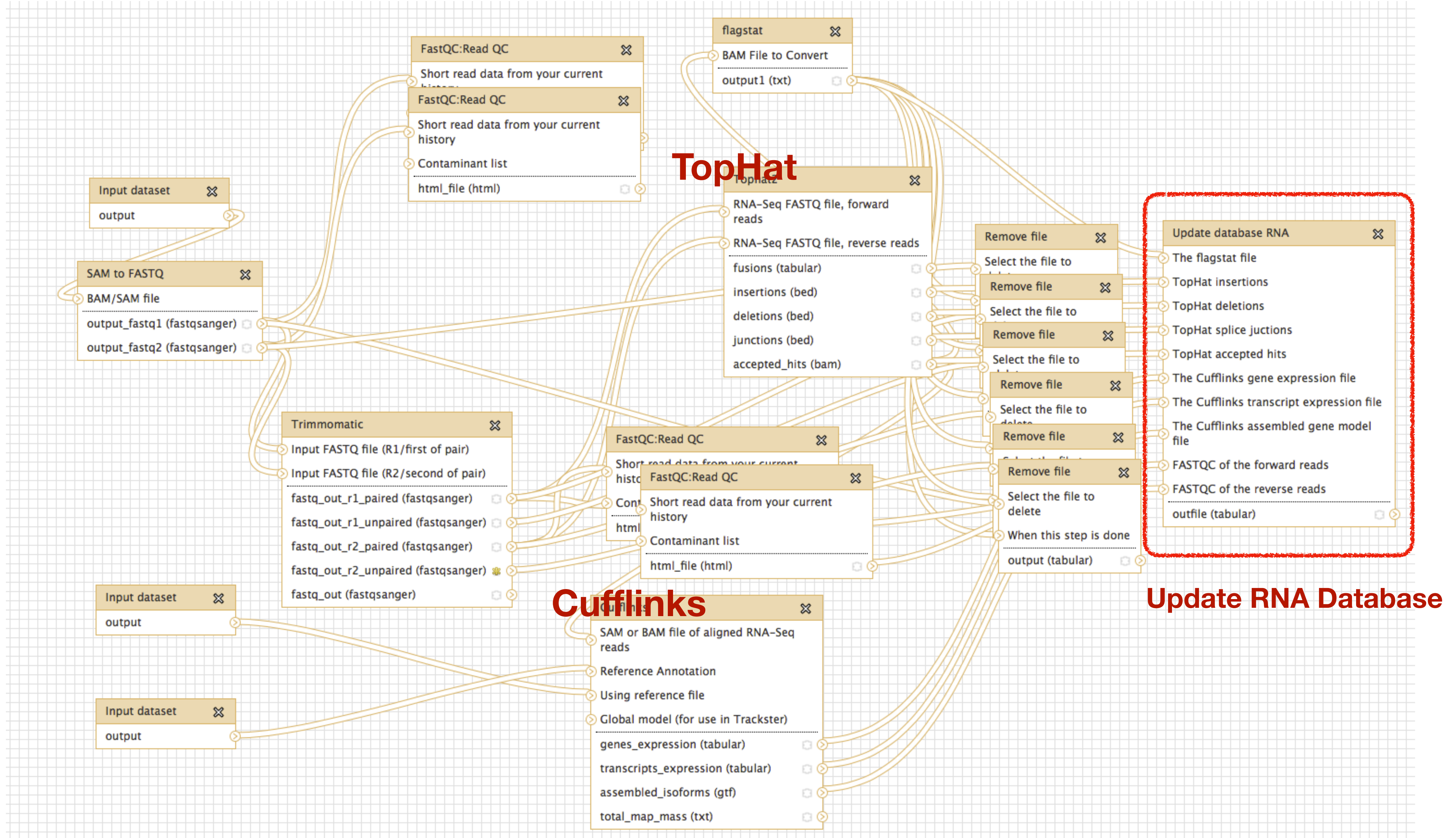


Galaxy / WEST

Transcriptome Sequencing



Transcriptome Sequencing



Transcriptome Sequencing

```
CREATE TABLE `PopCanRNA`.`RNA_file_uri` (  
    `hostname` text,  
    `sample_name` text NOT NULL,  
    `flagstat` text,  
    `cufflinks_gene_expression` text,  
    `cufflinks_transcript_expression` text,  
    `cufflinks_gene_models` text,  
    `fastqc_fwd` text,  
    `fastqc_rev` text,  
    `tophat_insertions` text,  
    `tophat_deletions` text,  
    `tophat_splice_junctions` text,  
    `tophat_bam` text,  
    `tophat_bam_md5sum` text,  
    PRIMARY KEY (`sample_name`(30)) USING BTREE  
    ) ENGINE=InnoDB DEFAULT CHARSET=utf8
```

Transcriptome Sequencing

- Independent MySQL database
- Allows for programmatic querying of results

Untitled @ localhost via socket

Back Next

SELECT * FROM PopCanRNA.RNA_file_uri R

Execute Stop

Query

hostname	sample_name	flagstat	cufflinks_gene_expression	cufflinks_transcript_expression	cufflinks_gene_m
west	11332_RNAOr85	/data/raid5A/LocalGalaxyRNADData/013/	/data/raid5A/LocalGalaxyRNADData/013/	/data/raid5A/LocalGalaxyRNADData/013/	/data/raid5A/LocalGalaxyRNADData/013/
east	ALAA20-4_Rep1_RNA1_1L	/data/raid5A/LocalGalaxyRNADData/009/	/data/raid5A/LocalGalaxyRNADData/009/	/data/raid5A/LocalGalaxyRNADData/009/	/data/raid5A/LocalGalaxyRNADData/009/
east	ALAA20-4_Rep2_RNA1_2L	/data/raid5A/LocalGalaxyRNADData/011/	/data/raid5A/LocalGalaxyRNADData/011/	/data/raid5A/LocalGalaxyRNADData/011/	/data/raid5A/LocalGalaxyRNADData/011/
west	ALAA20-4_TCol26_TRaw6_RNA182	/data/raid5A/LocalGalaxyRNADData/017/	/data/raid5A/LocalGalaxyRNADData/017/	/data/raid5A/LocalGalaxyRNADData/017/	/data/raid5A/LocalGalaxyRNADData/017/
east	ALAA20-4_TCol50_TRaw40_RNA369	/data/raid5A/LocalGalaxyRNADData/010/	/data/raid5A/LocalGalaxyRNADData/010/	/data/raid5A/LocalGalaxyRNADData/010/	/data/raid5A/LocalGalaxyRNADData/010/
west	AMER13-1addrep1L_RNA201	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/
west	AMER13-1addrep1L_RNA202	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/
east	AMER13-1addrep1L_RNA203	/data/raid5A/LocalGalaxyRNADData/010/	/data/raid5A/LocalGalaxyRNADData/010/	/data/raid5A/LocalGalaxyRNADData/010/	/data/raid5A/LocalGalaxyRNADData/010/
west	AMER13-1addrep2L_RNA205	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/
east	AMER13-1addrep2L_RNA206	/data/raid5A/LocalGalaxyRNADData/009/	/data/raid5A/LocalGalaxyRNADData/009/	/data/raid5A/LocalGalaxyRNADData/009/	/data/raid5A/LocalGalaxyRNADData/009/
west	AMER13-1addrep2L_RNA207	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/	/data/raid5A/LocalGalaxyRNADData/018/

Transcriptome Sequencing

- Dynamic Building of Gene Expression Results table

Untitled @ localhost via socket

BackNext

SELECT * FROM PopCanRNA.popcan_leaf_gene_expression p

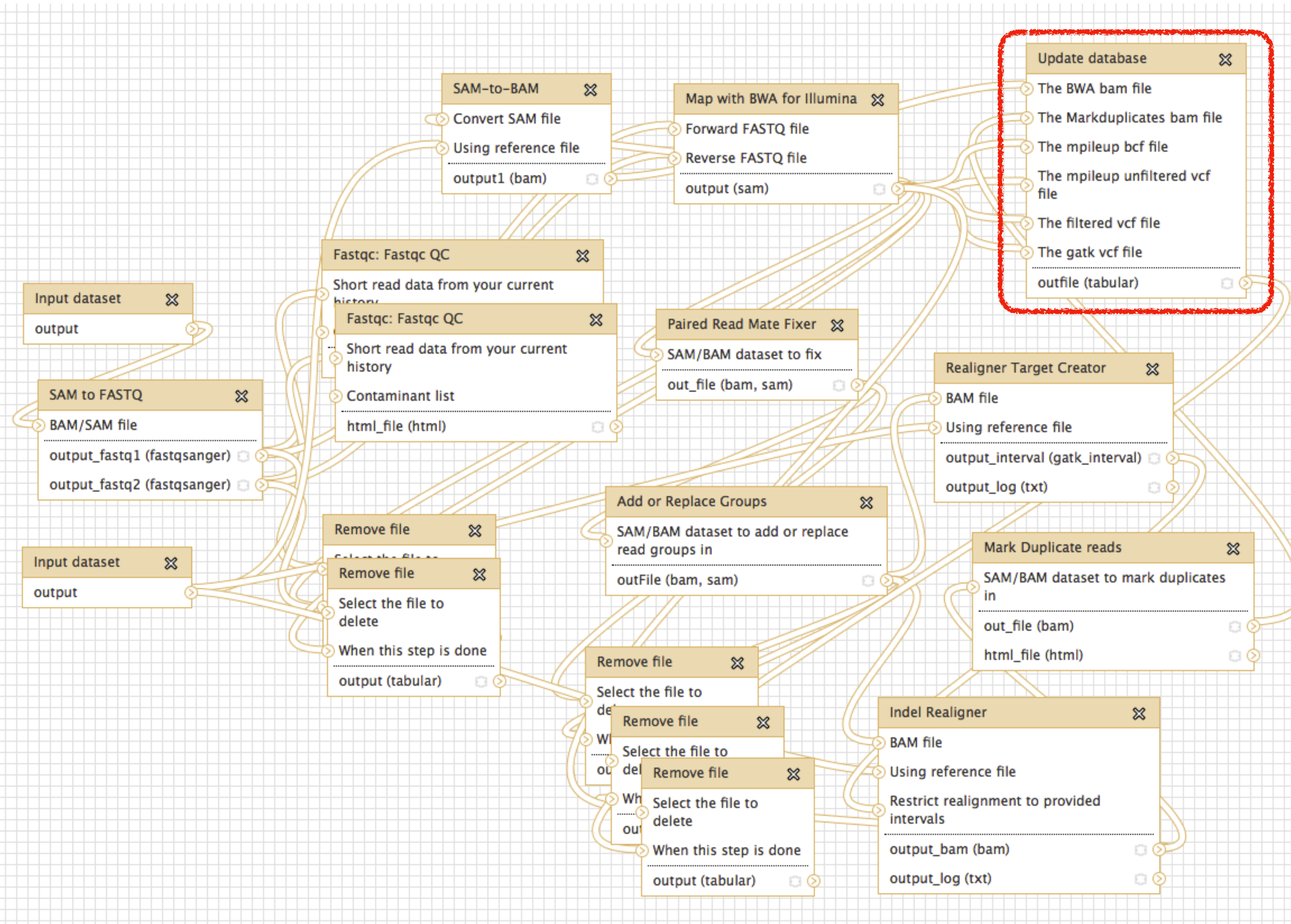
ExecuteStop

Query

gene_number	gene_id	tissue	ALAA20-3_AMER13-1addrep1L_RNA201	ALAA20-3_AMER13-1addrep1L_RNA202	ALAA20-3_AMER13-1addrep1L_RNA203	ALAA20-3_AMER13-1
4	Potri.001G000400	first uncurled leaf	41.0282	29.491	28.6132	32.8809
5	Potri.001G000500	first uncurled leaf	0	0	0	0
6	Potri.001G000600	first uncurled leaf	0.0251797	0	0	0
7	Potri.001G000700	first uncurled leaf	33.0592	22.9251	26.793	25.8353
8	Potri.001G000800	first uncurled leaf	0.325482	0.220098	0.155444	0.175966
9	Potri.001G000900	first uncurled leaf	86.5151	84.2777	88.5511	82.5777
10	Potri.001G001000	first uncurled leaf	6.20827	1.92059	4.45097	2.79066
11	Potri.001G001100	first uncurled leaf	1.30059	1.72912	1.65504	1.70034
12	Potri.001G001200	first uncurled leaf	0.130111	0.0590433	0.144995	0.0438737
13	Potri.001G001300	first uncurled leaf	40.5672	24.2564	30.4796	30.8496
14	Potri.001G001400	first uncurled leaf	2.67977	2.76517	3.07866	2.72547
15	Potri.001G001500	first uncurled leaf	9.02086	10.1361	9.35012	9.2488
16	Potri.001G001600	first uncurled leaf	81.5252	61.6698	82.7354	73.2591
17	Potri.001G001700	first uncurled leaf	7.5533	5.45722	6.34066	7.18672
18	Potri.001G001800	first uncurled leaf	3.93523	1.58916	2.33021	2.78344

Combining Data

Genome Pipeline



- Genome BAM files
- GATK pipeline
- Different species
- Selected individuals
- Fast, up to date sharing of the results

Results

The Plant Cell, Vol. 29: 2000–2015, August 2017, www.plantcell.org © 2017 ASPB.
http://orcid.org/0000-0001-7527-5461



Exploiting Natural Variation to Uncover an Alkene Biosynthetic Enzyme in Poplar^{OPEN}

Eliana Gonzales-Vigil,^a Charles A. Hefer,^b Michelle E. von Loessl,^a Jonathan La Mantia,^c and Shawn D. Mansfield^a

^a Department of Botany,
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Research

Comparative interrogation of the developing xylem transcriptomes of two wood-forming species: *Populus trichocarpa* and *Eucalyptus grandis*

Charles A. Hefer¹, Eshchar Mizrachi², Alexander A. Myburg², Carl J. Douglas¹ and Shawn D. Mansfield³

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Altering carbon allocation in hybrid poplar (*Populus alba* × *grandidentata*) impacts cell wall growth and development

Faride Unda¹, Hoon Kim^{2,3}, Charles Hefer⁴, John Ralph^{2,3}, Shawn D. Mansfield^{1,3}

MOLECULAR ECOLOGY

Molecular Ecology (2015) 24, 3243–3256

doi: 10.1111/mec.13126

FROM THE COVER

Recent Y chromosome divergence despite ancient origin of dioecy in poplars (*Populus*)

A. GERALDES,^{*1} C. A. HEFER,^{*1} A. CAPRON,^{*} N. KOLOSOVA,^{*} F. MARTINEZ-NUÑEZ,^{*} R. Y. SOOLANAYAKANAHALLY,[†] B. STANTON,[‡] R. D. GUY,[§] S. D. MANSFIELD,[¶] C. J. DOUGLAS^{*} and Q. C. B. CRONK^{*}

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Introgression from *Populus balsamifera* underlies adaptively significant variation and range boundaries in *P. trichocarpa*

Adriana Suarez-Gonzalez¹, Charles A. Hefer^{1,2}, Christian Lexer³, Carl J. Douglas^{1†} and Quentin C. B. Cronk¹

¹Department of Botany, University of British Columbia, Vancouver, BC Canada, V6T 1Z4; ²Biotechnology Platform, Agricultural Research Council, Private Bag X05, Onderstepoort 0110,

Botany and Biodiversity Research, University of

GENOMICS OF HYBRIDIZATION

Genomic and functional approaches reveal a case of adaptive introgression from *Populus balsamifera* (balsam poplar) in *P. trichocarpa* (black cottonwood)

ADRIANA SUAREZ-GONZALEZ,^{*} CHARLES A. HEFER,^{*1} CAMILLE CHRISTE,[†] OLIVER COREA,[‡] CHRISTIAN LEXER,^{†§} QUENTIN C. B. CRONK^{*} and CARL J. DOUGLAS^{*}

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Stefan Jansson
USFS Forest Products Lab - Dan Cullen
BC Ministry of Forests and Range - Alvin Yanchuk
Greenwood Resources - Brian Stanton
Kruger Inc. – Dan Carson
University of Pretoria – Zander Myburg

Applied Genomics Innovation Program

Large Scale Applied Research Program