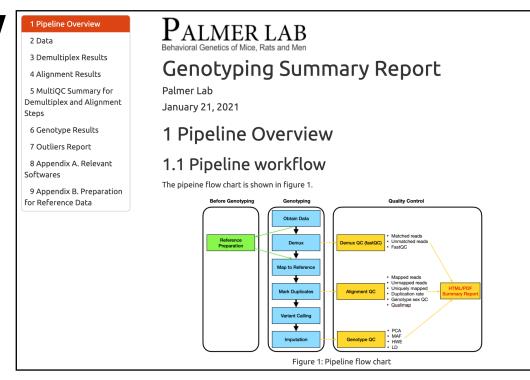
HS Rats Genotyping Pipeline

Pipeline Summary Report Design

Pipeline Overview



2.1 Sequence Data

Sample strain: Heterogenous stock
Number of samples: 376
Flow cell run ID(s): 201218_A00953_0203_BHN5HYDSXY
Riptide library preparation: Riptide31, Riptide32, Riptide39, Riptide40
Metadata: /projects/pspalmer/hs_rats/201218_A00953_0203_BHN5HYDSXY/data/kn04_fastq_sample_metadata_n952.csv

Run ID
Library ID
Sample Project

201218_A00953_0203_BHN5HYDSXY Riptide31 u01_olivier_george_cocaine
1
201218_A00953_0203_BHN5HYDSXY Riptide31 u01_olivier_george_oxycodone
83
201218_A00953_0203_BHN5HYDSXY Riptide31 u01_olivier_george_scrub
4
201218_A00953_0203_BHN5HYDSXY Riptide31 u01_suzanne_mitchell
3007788
201218_A00953_0203_BHN5HYDSXY Riptide31 u01_suzanne_mitchell
3007788
201218_A00953_0203_BHN5HYDSXY Riptide31 u01_suzanne_mitchell
3007788
201218_A00953_0203_BHN5HYDSXY Riptide32 u01_olivier_george_cocaine
91
201218_A00953_0203_BHN5HYDSXY Riptide32 u01_olivier_george_scrub
5
201218_A00953_0203_BHN5HYDSXY Riptide32 u01_olivier_george_scrub
5
201218_A00953_0203_BHN5HYDSXY Riptide32 u01_olivier_george_scrub
5
201218_A00953_0203_BHN5HYDSXY Riptide39 u01_olivier_george_scrub
6
2007788

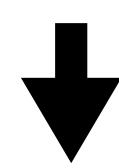
201218_A00953_0203_BHN5HYDSXY Riptide40 u01_peter_kalivas_italy

STITCH variant calling reference panel: /projects/ps-palmer/hs_rats/reference_genome/rn6_refPnls
BEAGLE imputation genetic map: /projects/ps-palmer/hs_rats/reference_genome/map_files

2.2 Reference Data

| Page |

Submission Script



Flowchart

Summary Report

Pipeline Overview

2.1 Sequence Data

2.2 Reference Data

3 Demultiplex Results 4 Alignment Results

MultiQC Summary for

6 Genotype Results

8 Appendix A. Relevant

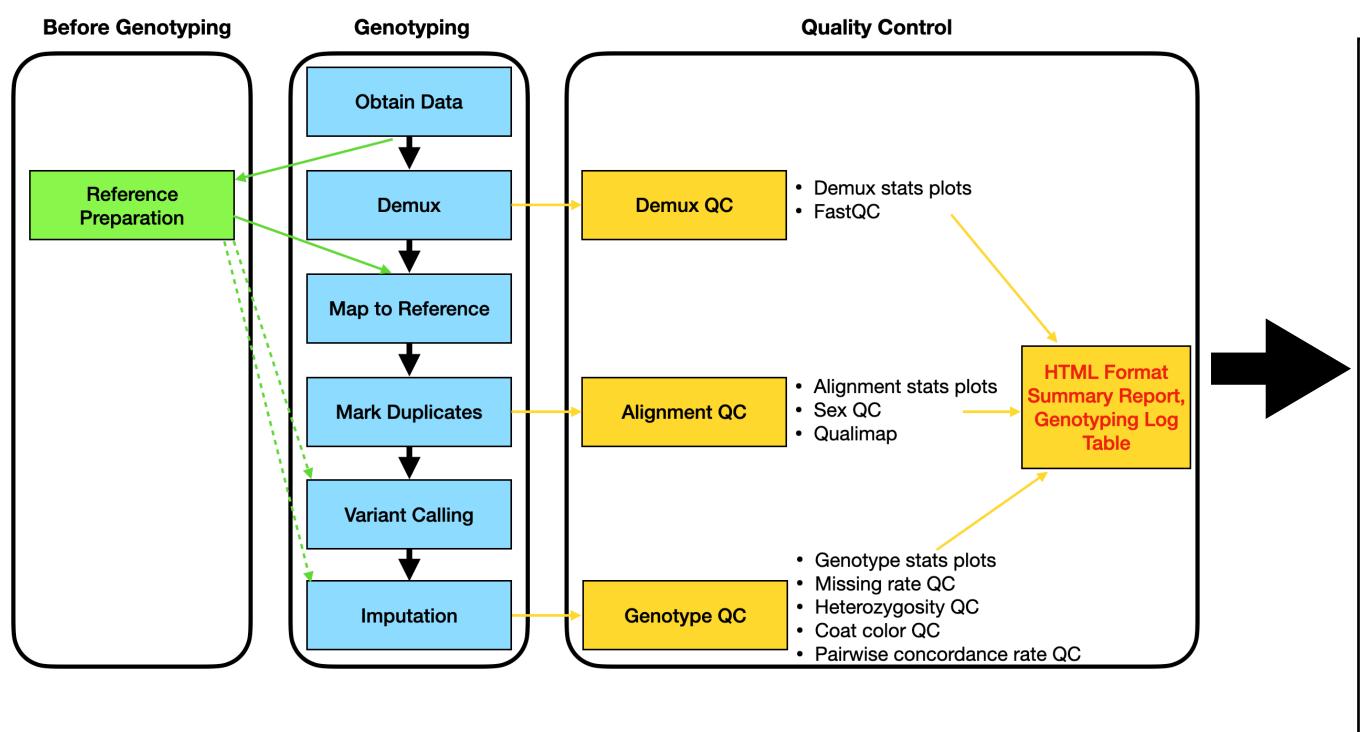
9 Appendix B. Preparation

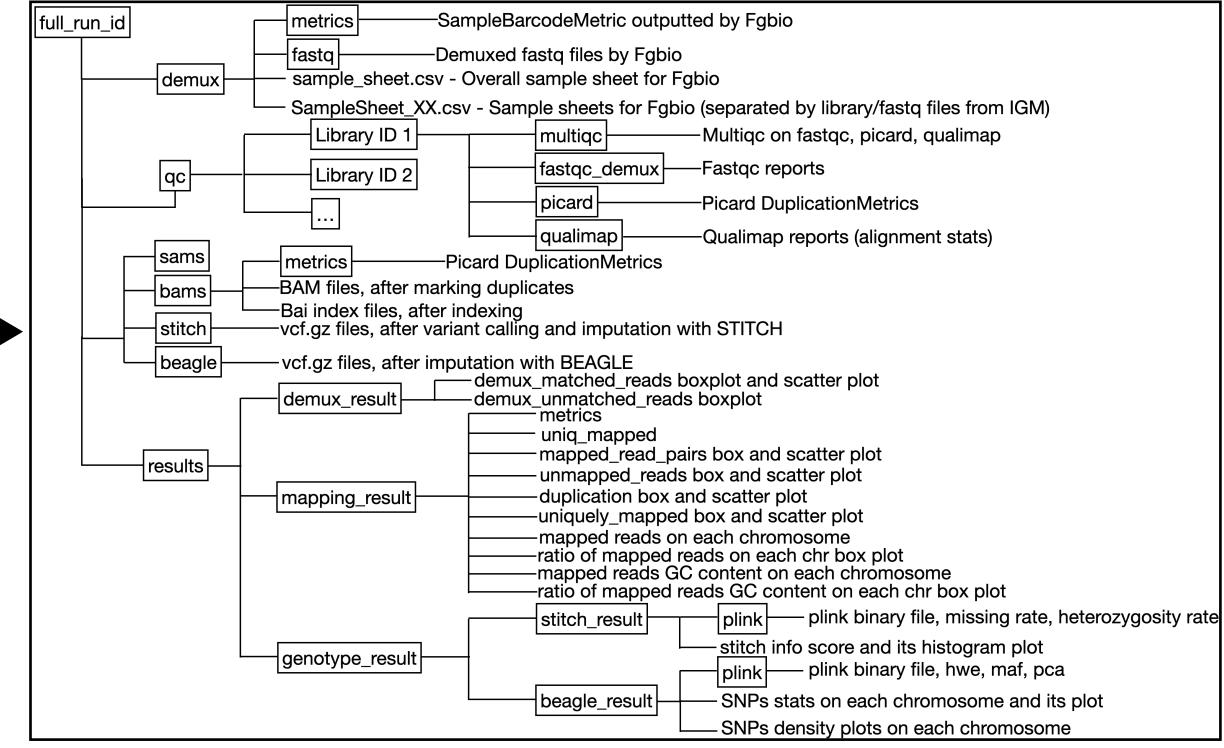
7 Outliers Report

multiplex and Alignmer



Genotyping Log Table





Directory Structure

Pipeline Arguments

- Line 1: home directory
- Line 2: Flow cell directory
- Line 3: Flow cell metadata Line 4: Sequencing data directory
- Line 5: Reference genome
- Line 6: Reference panels for STITCH
- Line 7: Genetic map for BEAGLE
- Line 8: Directory where you keep the code for the pipeline
- Line 9: The general name of this run

previous_flow_cells_metadata

Paths to previous flow cells' metadata.

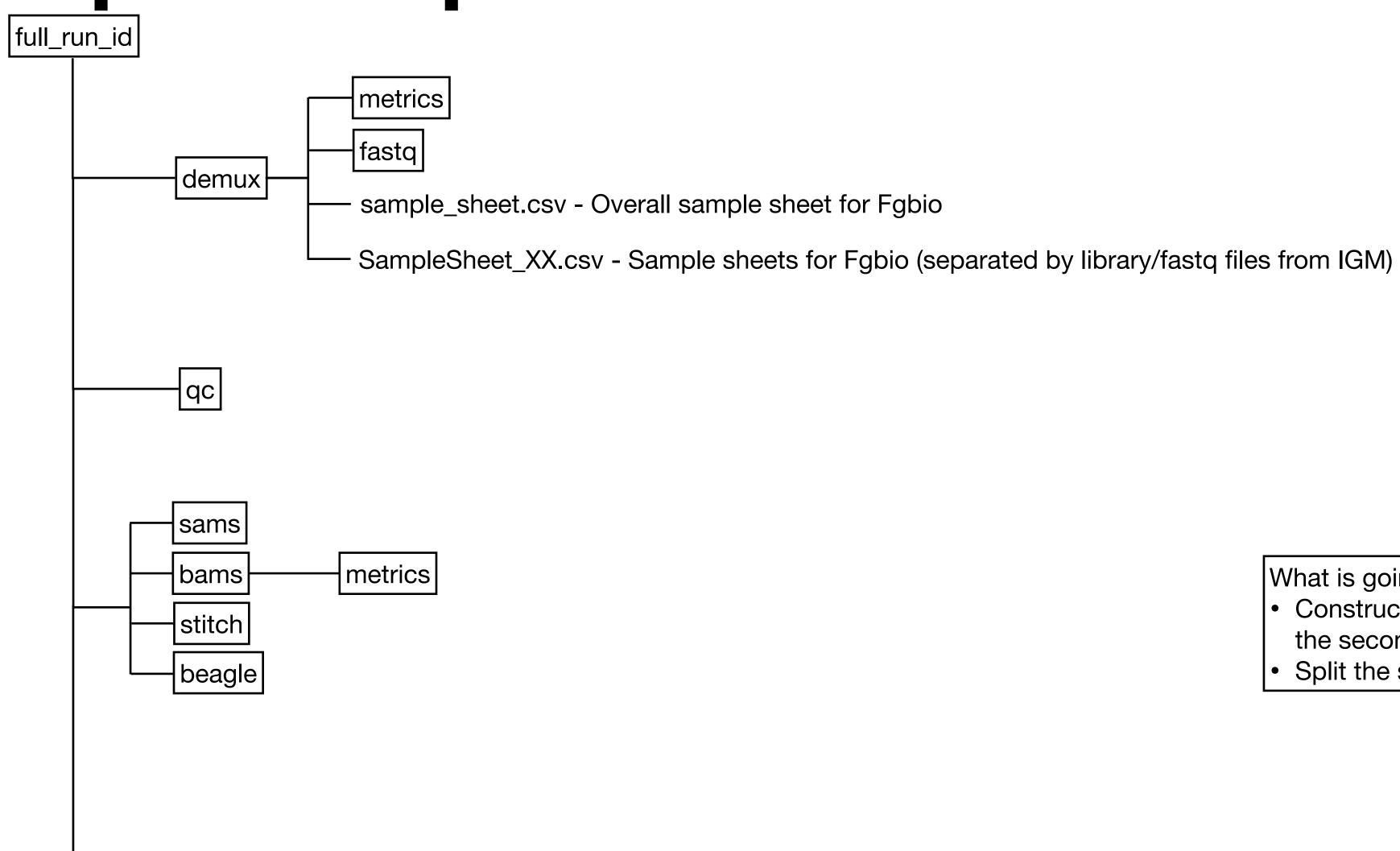
previous_flow_cells_bams

Paths to previous flow cells' BAM files.

pedigree_data

Paths to all flow cells' pedigree data.

Step 1 - Preparation



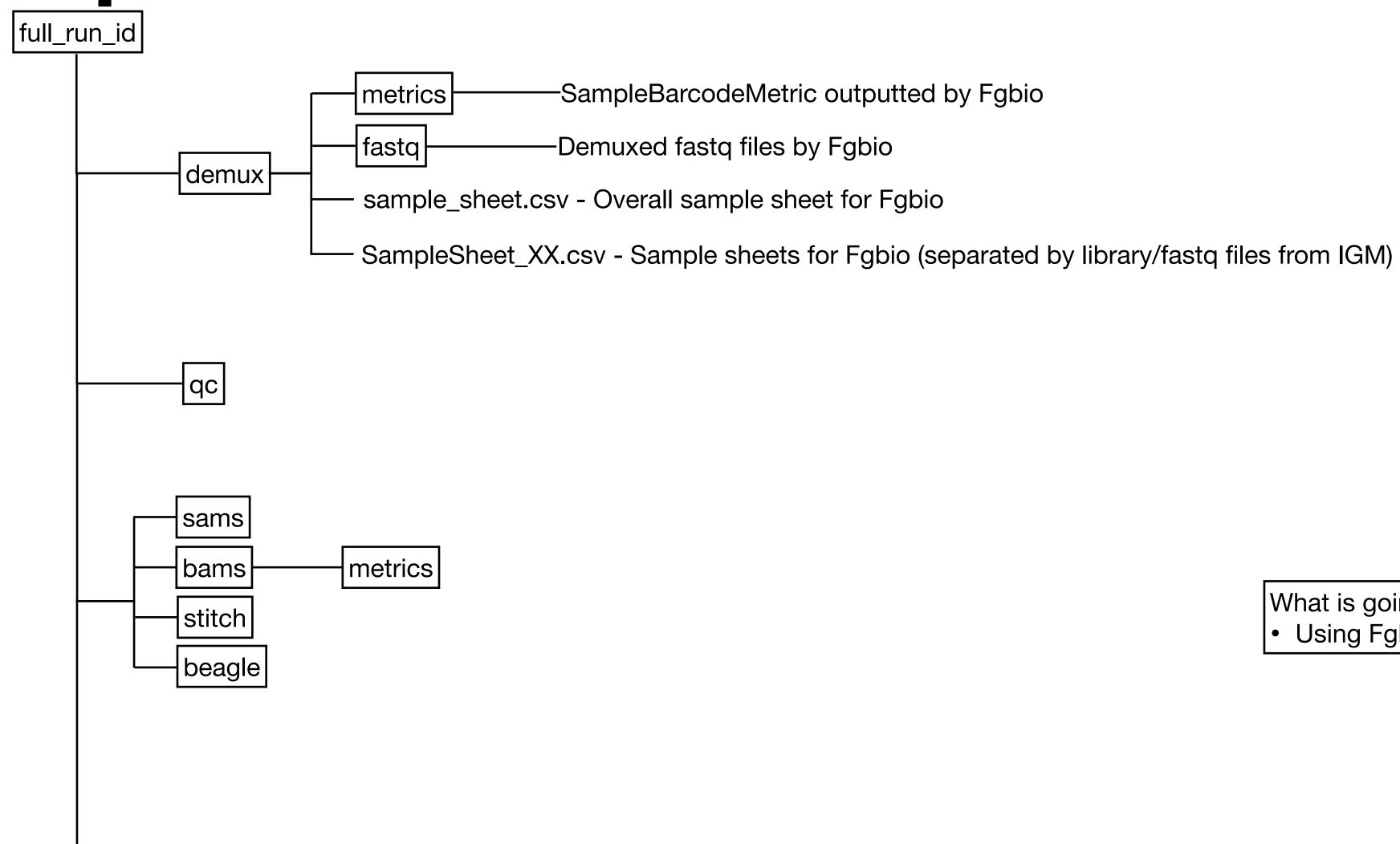
results

What is going on:

- Construct the basic structure of the directory from the second line of the Pipeline Argument file
- Split the sample sheet for each library prep for Fgbio

Step 2 - Demux

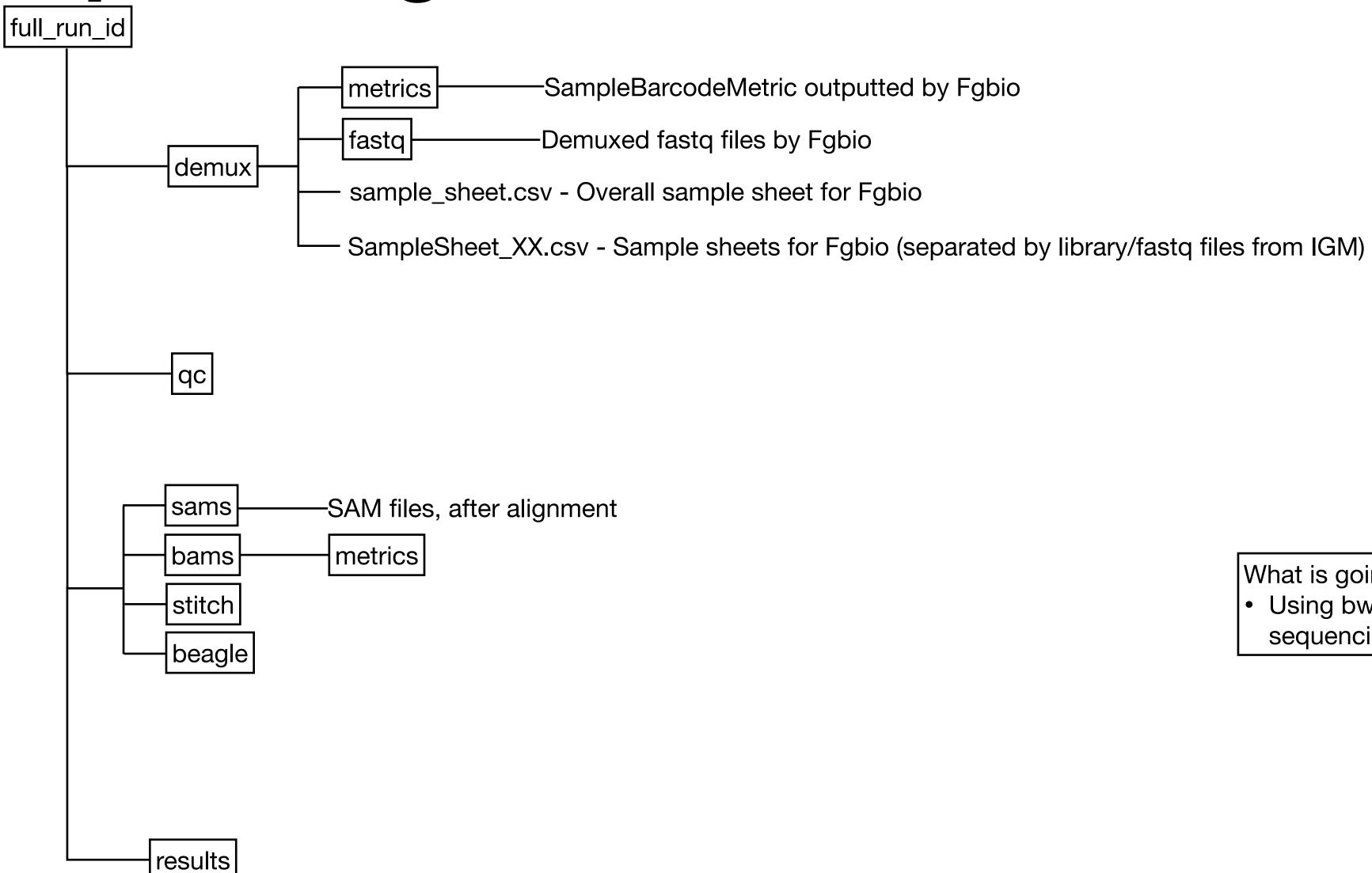
results



What is going on:

Using Fgbio to demultiplex the fastq files

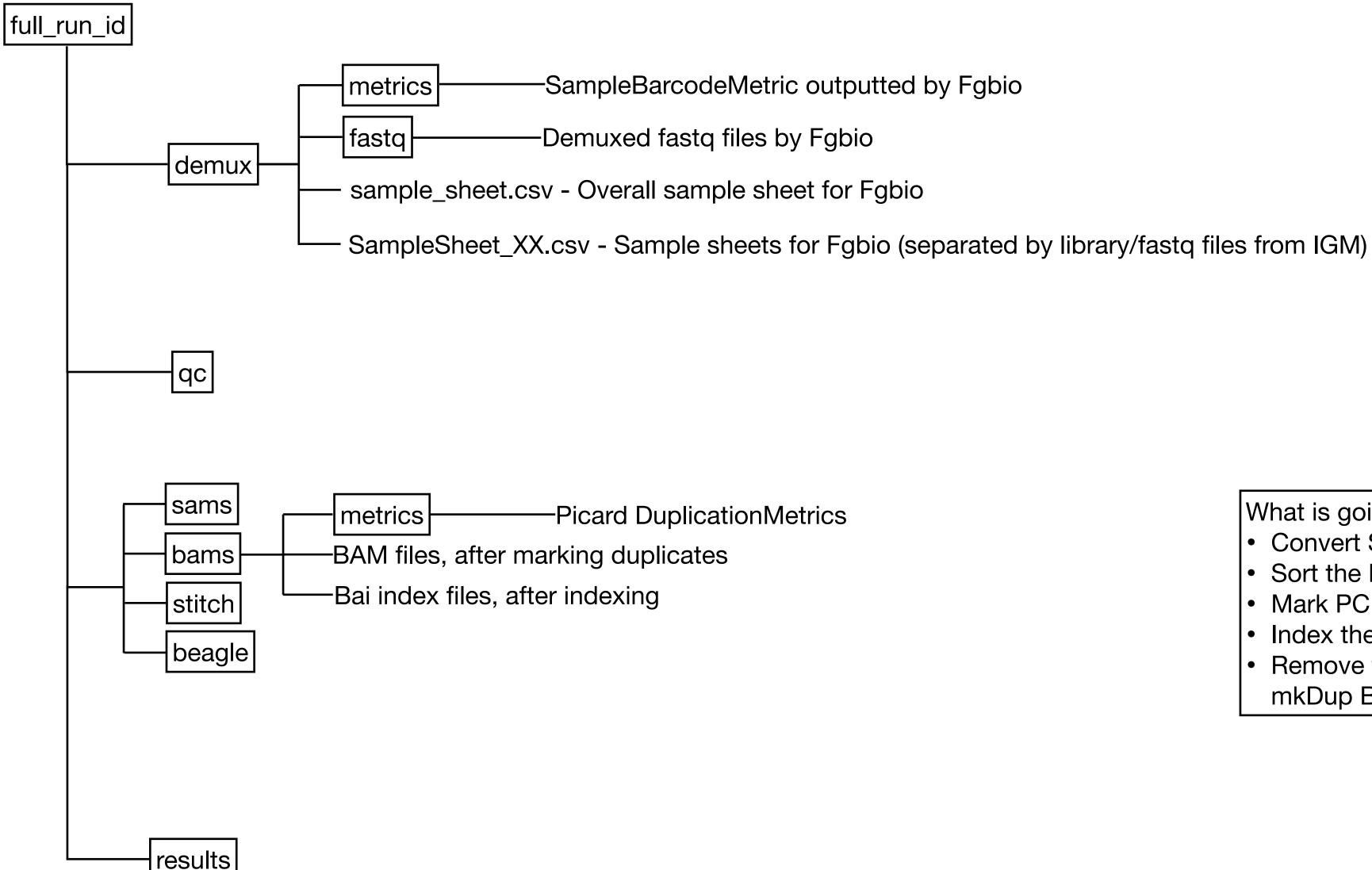
Step 3 - Alignment



What is going on:

• Using bwa mem to map the demultiplexed sequencing reads to reference genome

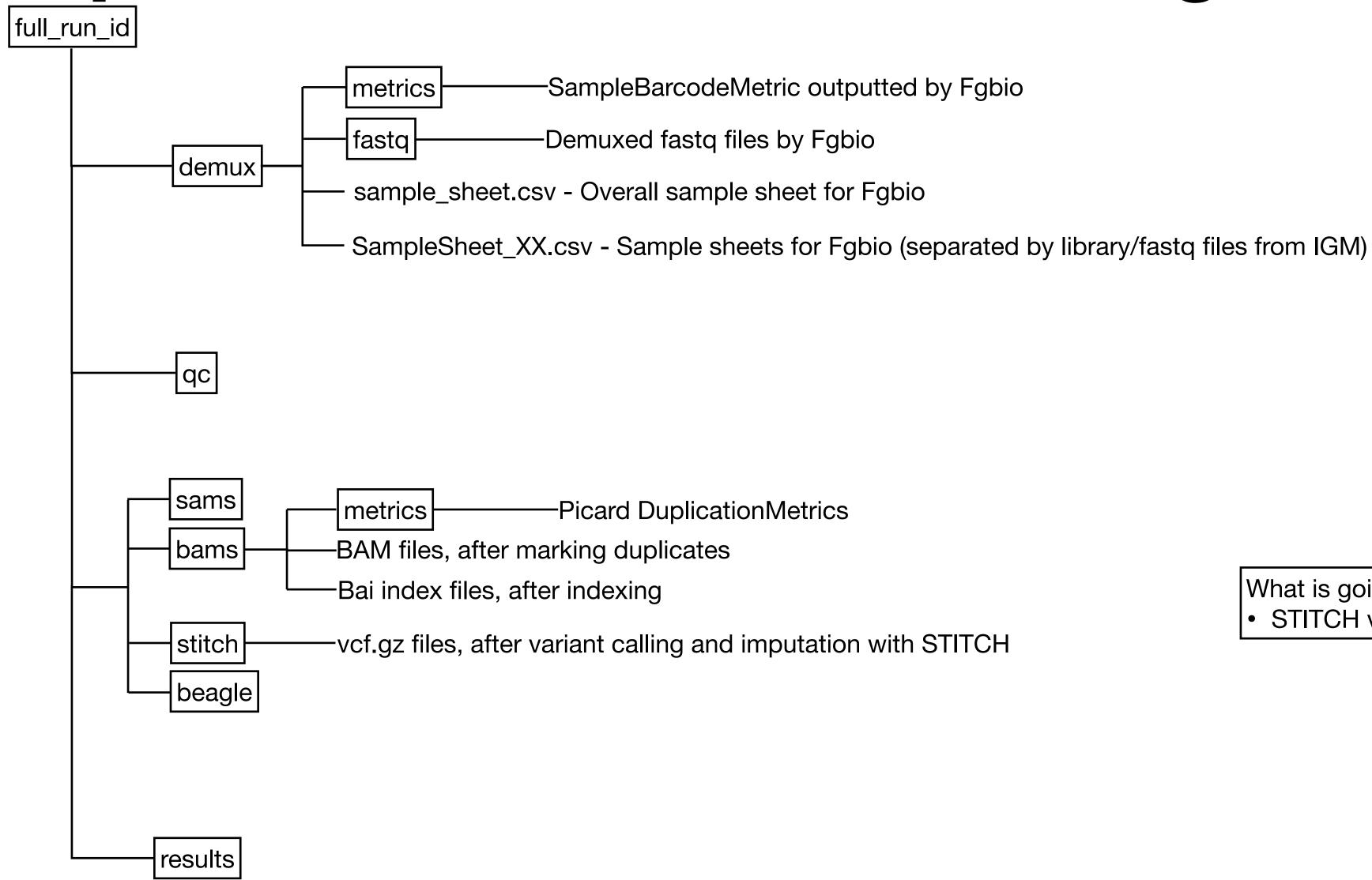
Step 4 - Mark Duplicates



What is going on:

- Convert SAM files to BAM files
- Sort the BAM files
- Mark PCR duplicates
- Index the marked duplicates BAM files
- Remove the SAM files, unsorted BAM files, and nonmkDup BAM files

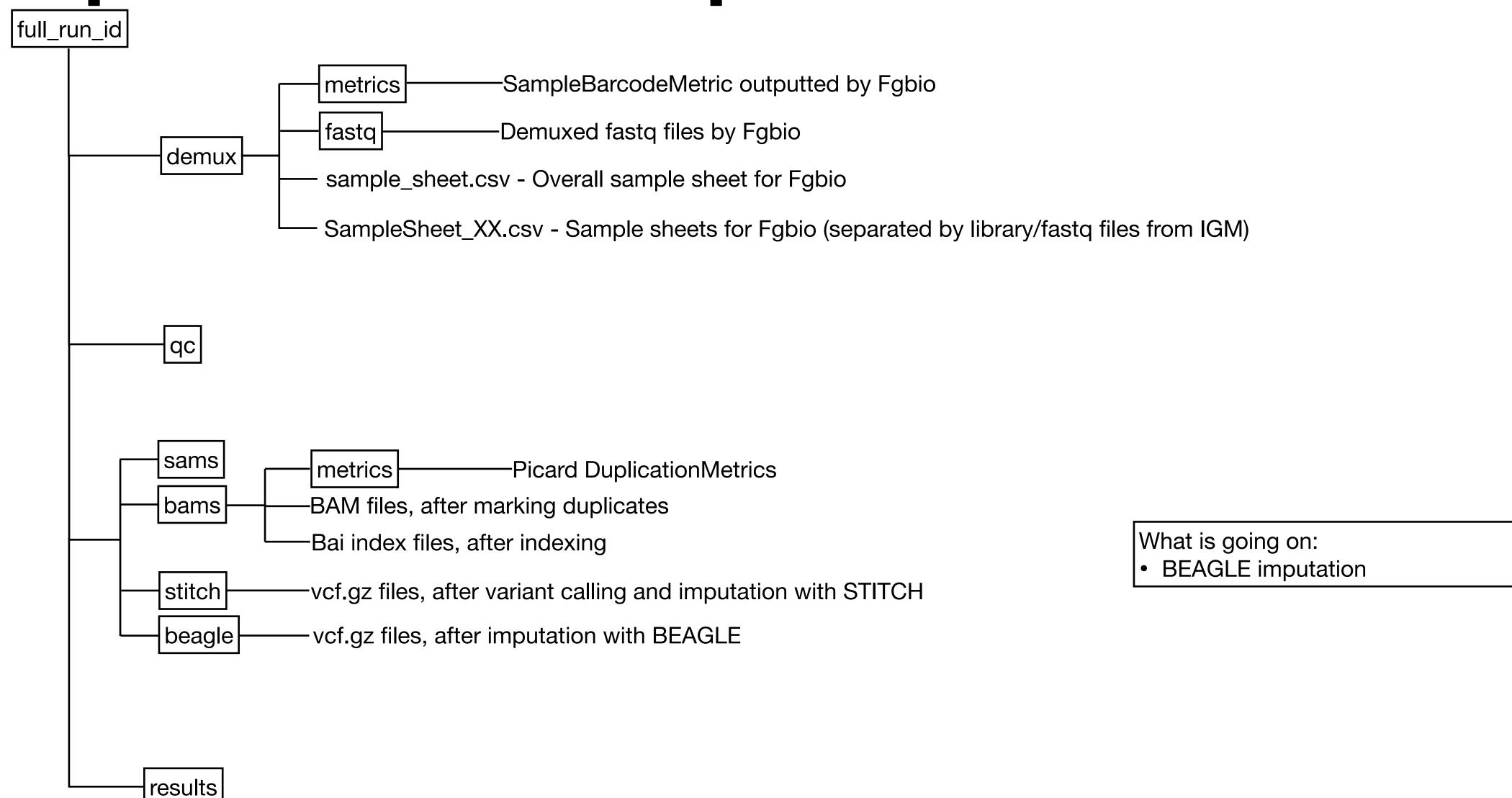
Step 5 - STITCH Variant Calling



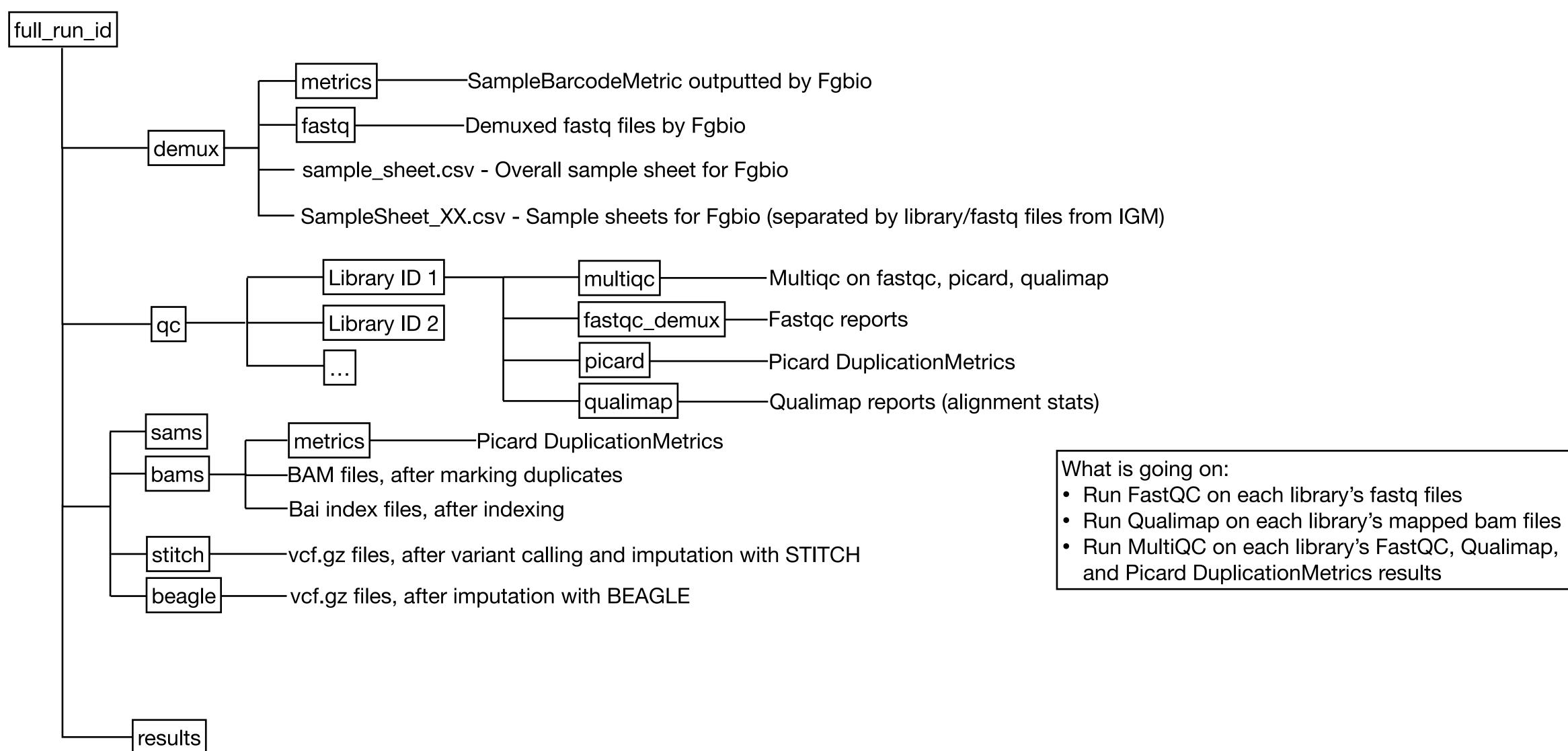
What is going on:

STITCH variant calling

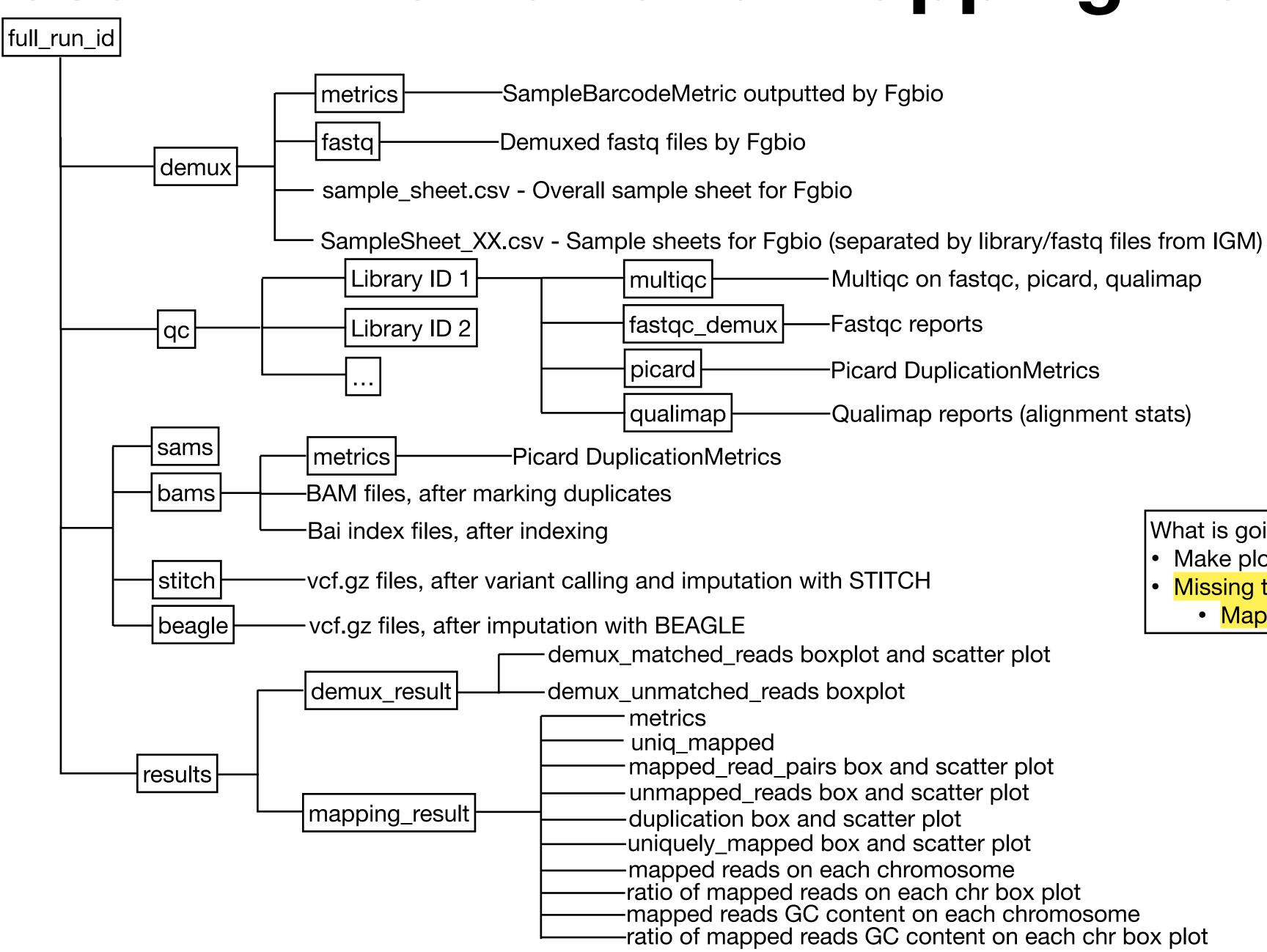
Step 6 - BEAGLE Imputation



Result 1 - MultiQC



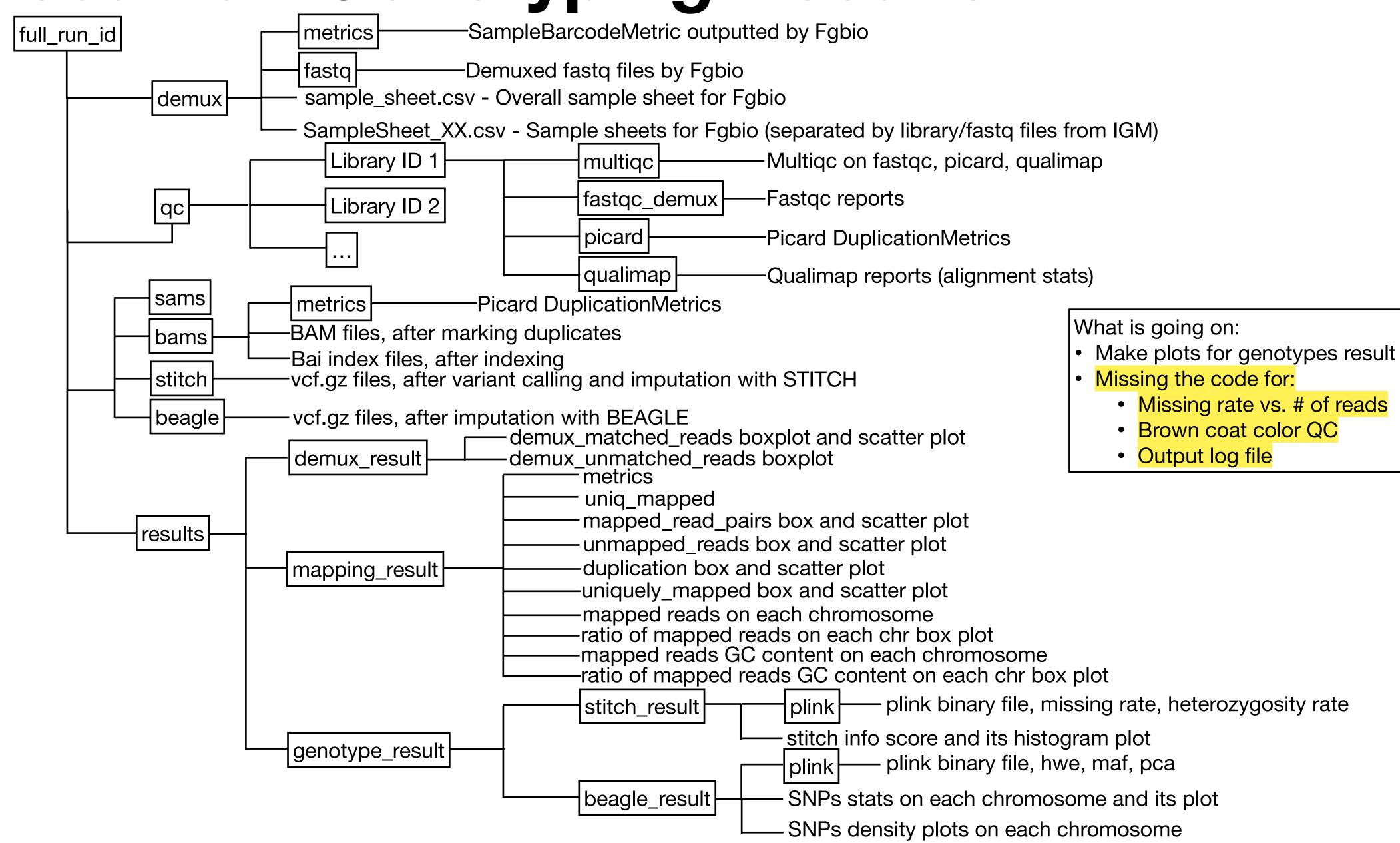
Result 2 - Demux and Mapping Results



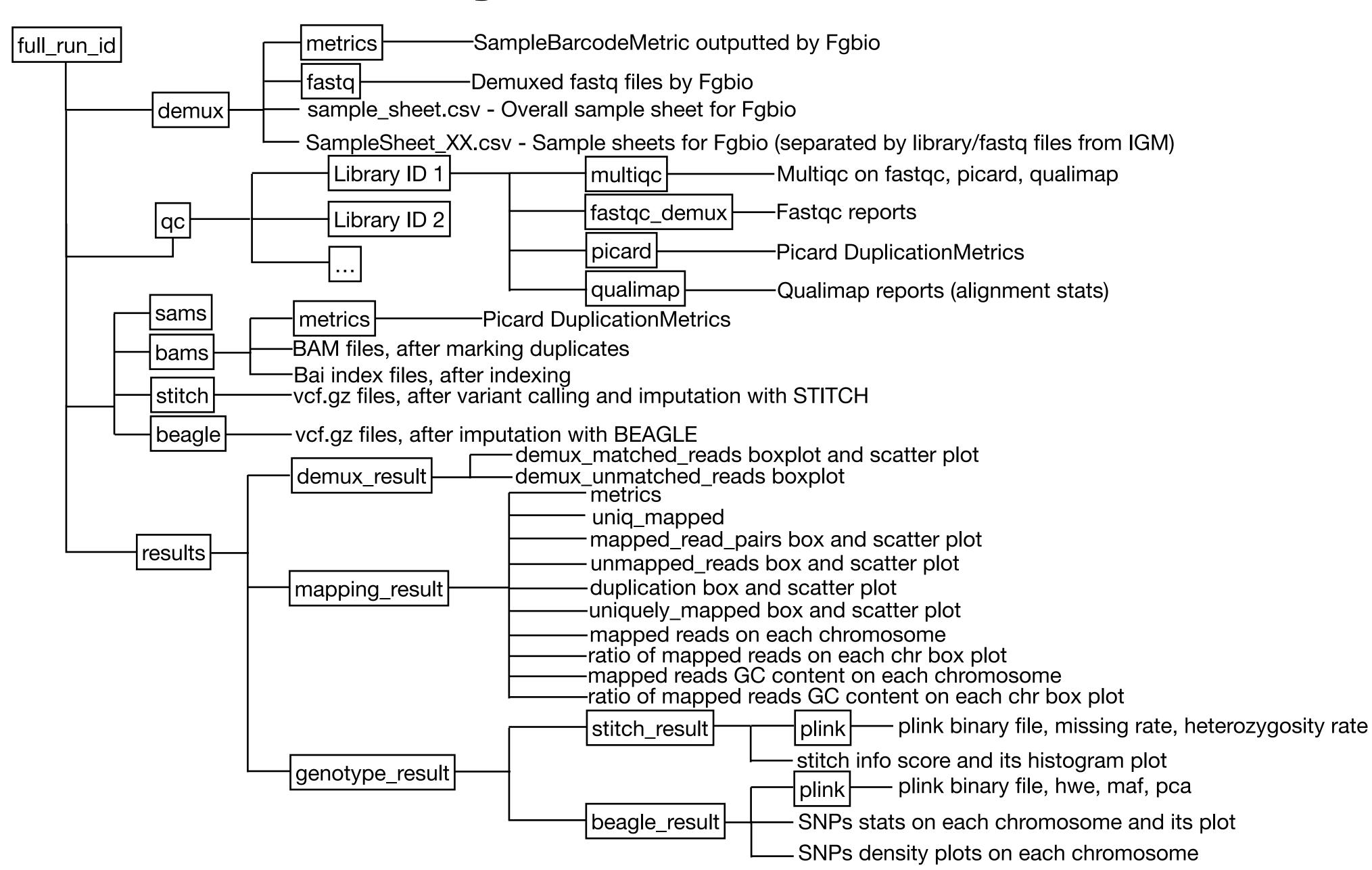
What is going on:

- Make plots for demux result and mapping result
- Missing the code for:
 - Mapping quality histogram

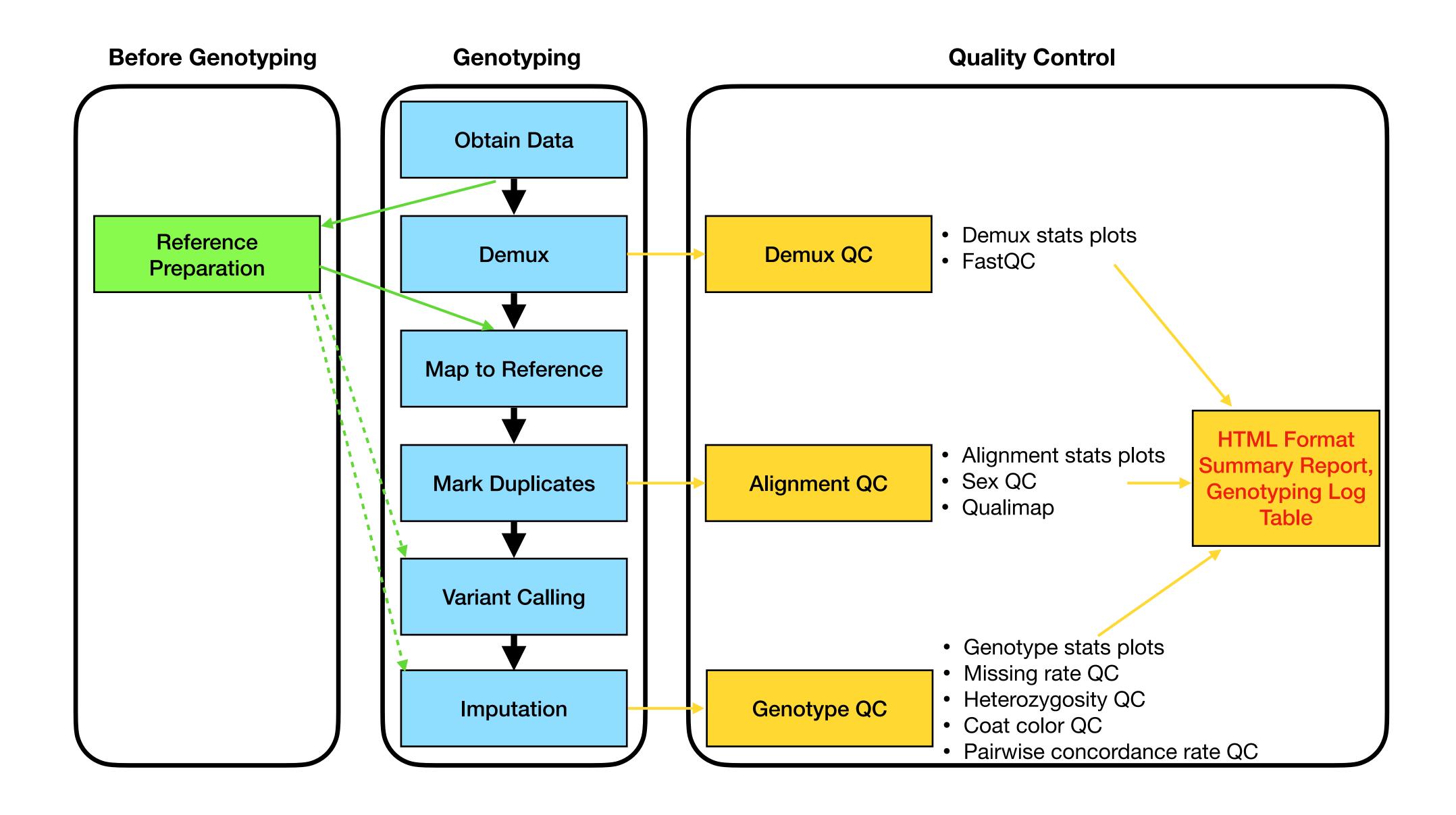
Result 3 - Genotyping Results



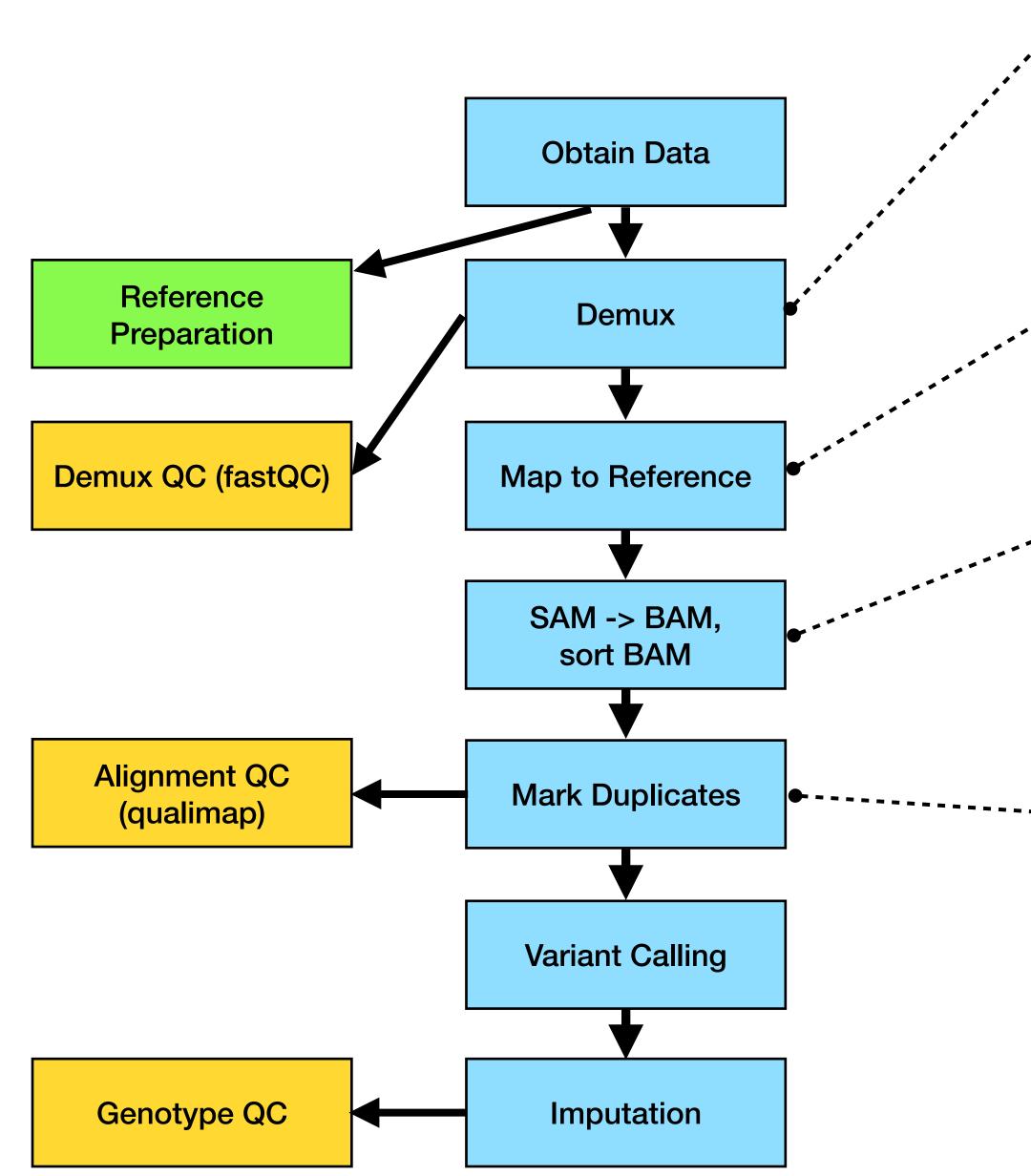
TSCC Directory Structure



Pipeline Overview

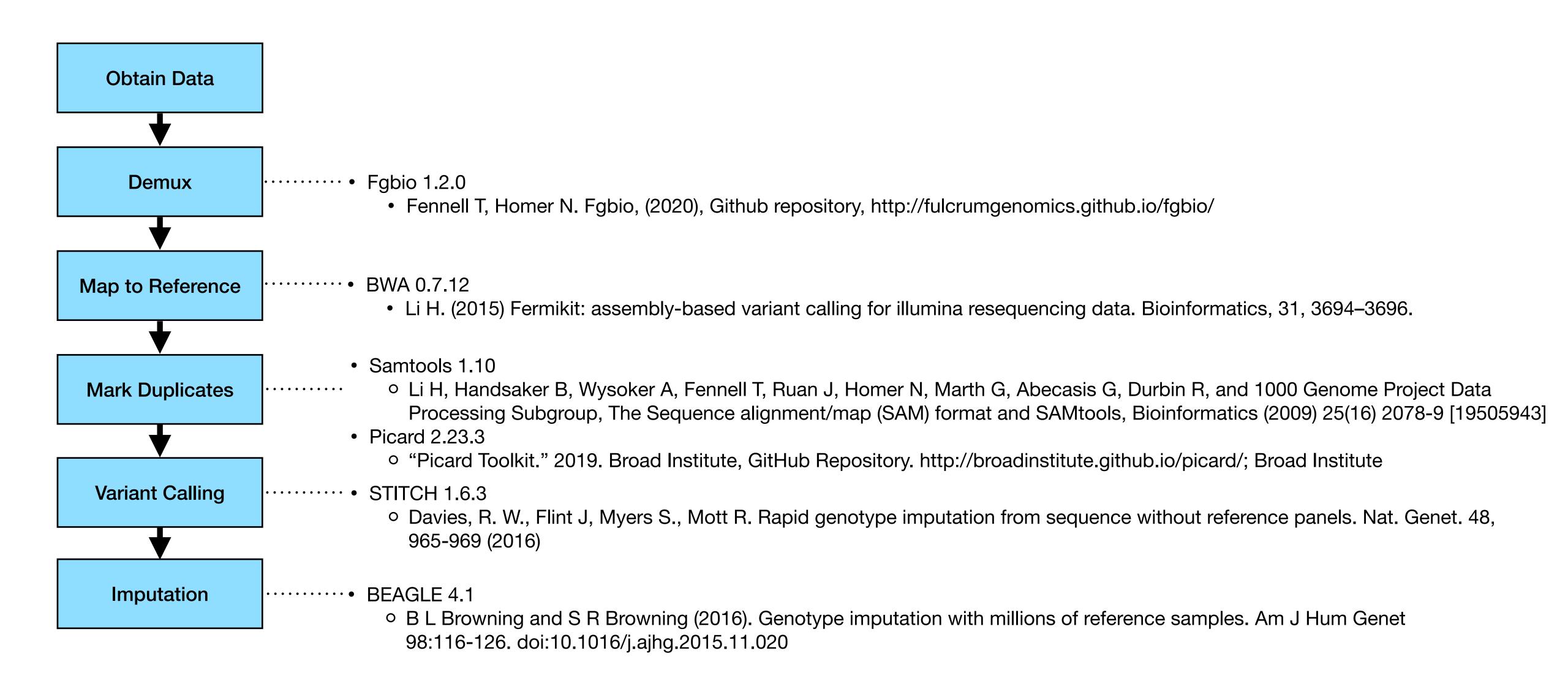


Pipeline Flowchart



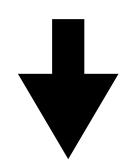
```
java -Xmx40G -XX:+AggressiveOpts -XX:+AggressiveHeap \
   -jar /projects/ps-palmer/software/local/src/fgbio-1.2.0/fgbio-1.2.0.jar DemuxFastqs \
   --inputs ${pre_demux_fastq_R1} ${pre_demux_fastq_R2} \
   --metadata ${sample_sheet} \
   --read-structures 8B12M+T 8M+T \
   --output-type=Fastq \
   --threads $ncpu \
   --output ${out_path}/demux/fastq \
   --metrics ${out_path}/demux/metrics/${fastq_temp}demux_barcode_metrics.txt
/projects/ps-palmer/software/local/src/bwa-0.7.12/bwa mem -aM -t 2\
-R "@RG\tID:${instrument_name}.${run_id}.${flowcell_id}.${flowcell_lane}\tLB:${library_id}
\tPL:ILLUMINA\tSM:${sample_id}\tPU:${flowcell_id}.${flowcell_lane}.${sample_barcode}" \
${reference_data} ${demux_data}/${f}_R1.fastq.gz \
${demux_data}/${f}_R2.fastq.gz > ${out_path}/sams/${f}.sam &
/projects/ps-palmer/software/local/src/samtools-1.10/samtools view -h -b \
 -t ${reference_data} -o ${out_path}/bams/${f}.bam ${mapped_data}/${f}.sam
/projects/ps-palmer/software/local/src/samtools-1.10/samtools sort -m 30G \
 -o ${out_path}/bams/${f}_sorted.bam ${out_path}/bams/${f}.bam
java -Xmx20G -XX:+AggressiveOpts -XX:+AggressiveHeap\
 -jar /projects/ps-palmer/software/local/src/picard-2.23.3/picard.jar MarkDuplicates \
 --INPUT ${out_path}/bams/${f}_sorted.bam \
 --REMOVE_DUPLICATES false \
 --ASSUME_SORTED true \
 --METRICS_FILE ${out_path}/bams/metrics/${f}_sorted_mkDup_metrics.txt \
 --OUTPUT ${out_path}/bams/${f}_sorted_mkDup.bam &
/projects/ps-palmer/software/local/src/samtools-1.10/samtools index \
 ${out_path}/bams/${f}_sorted_mkDup.bam ${out_path}/bams/${f}_sorted_mkDup.bai
```

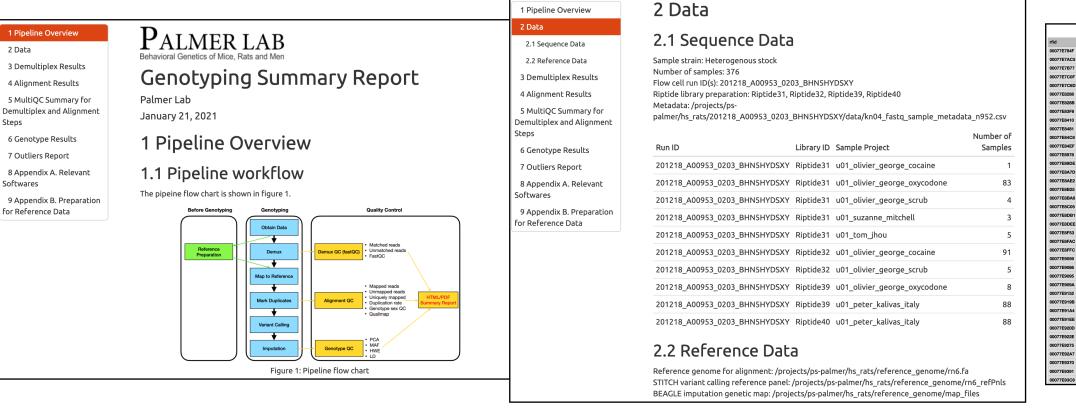
Relevant Software

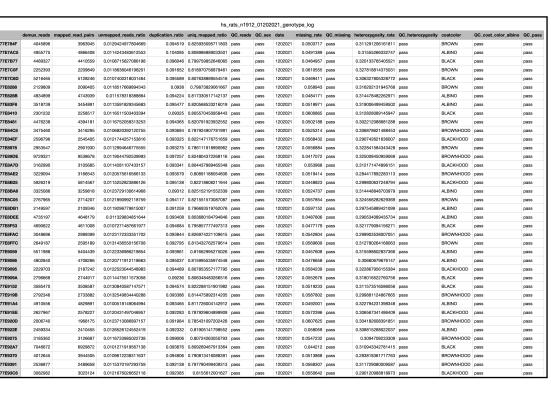


Pipeline on TSCC

Submission Script



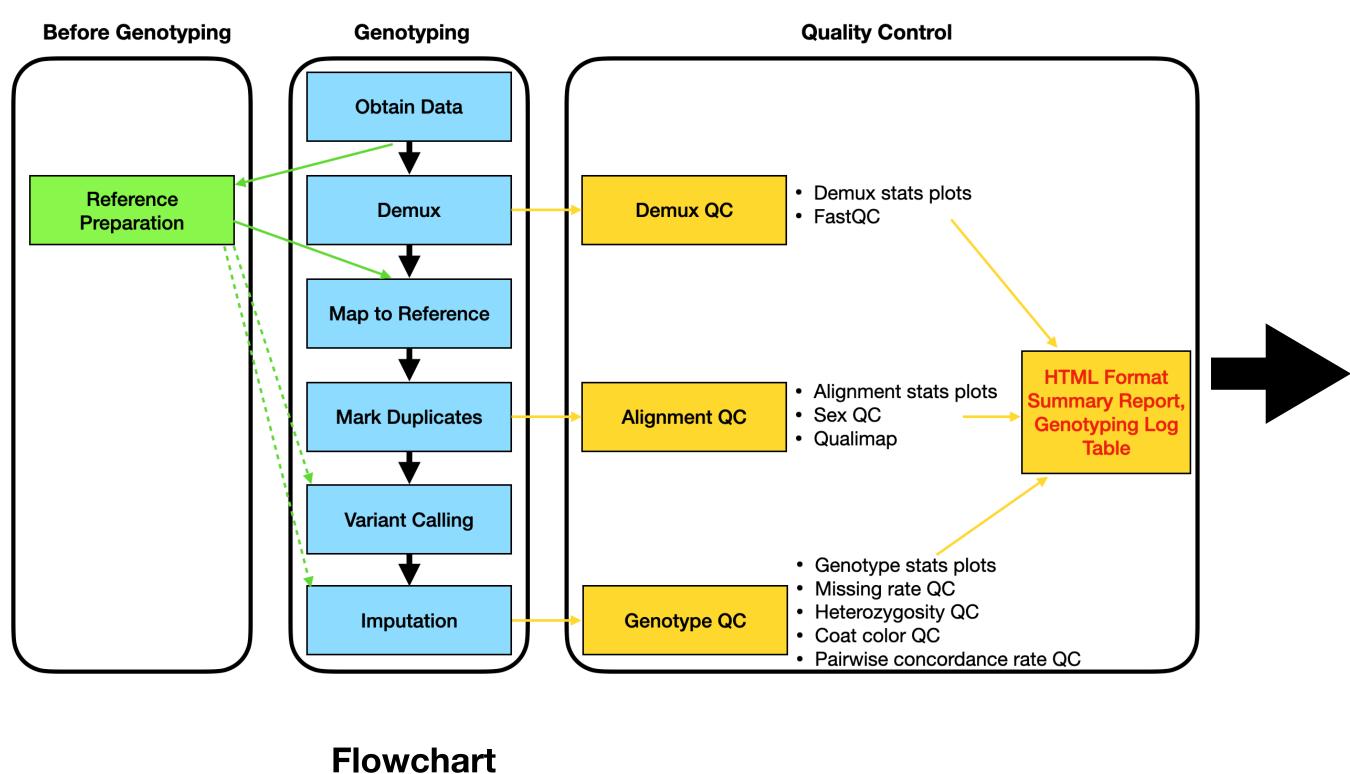


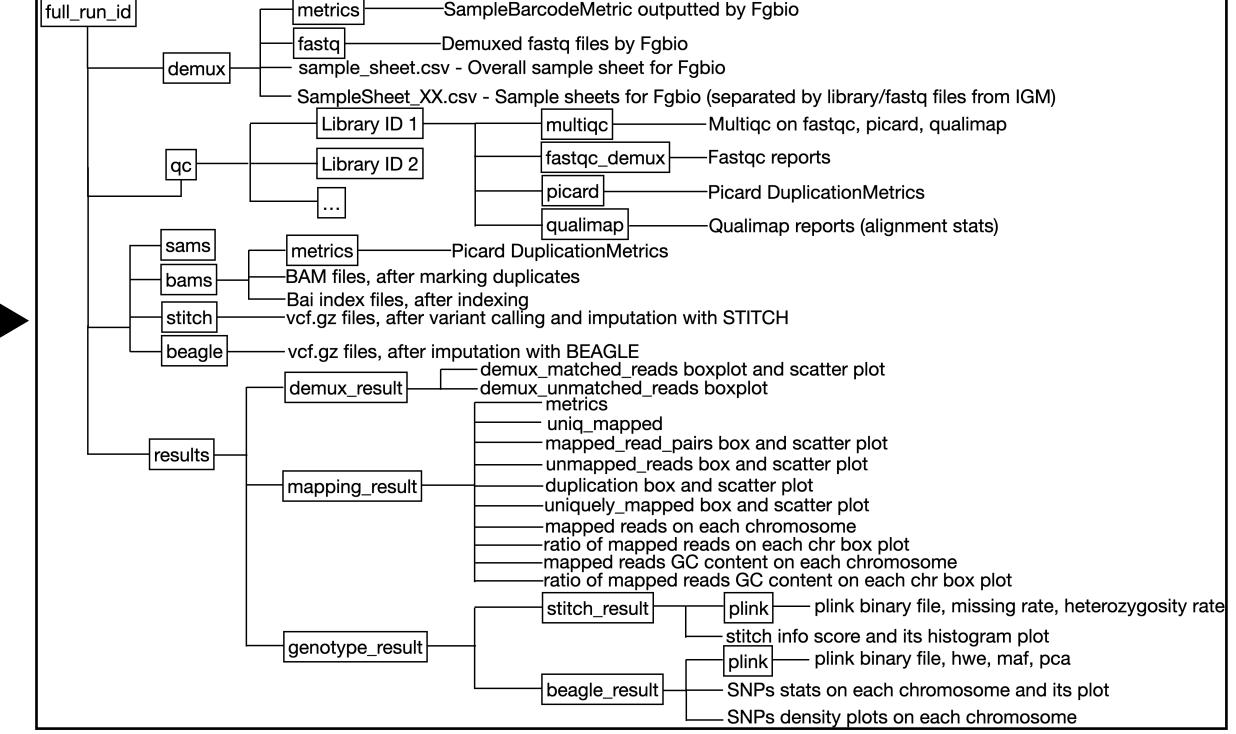


Summary Report



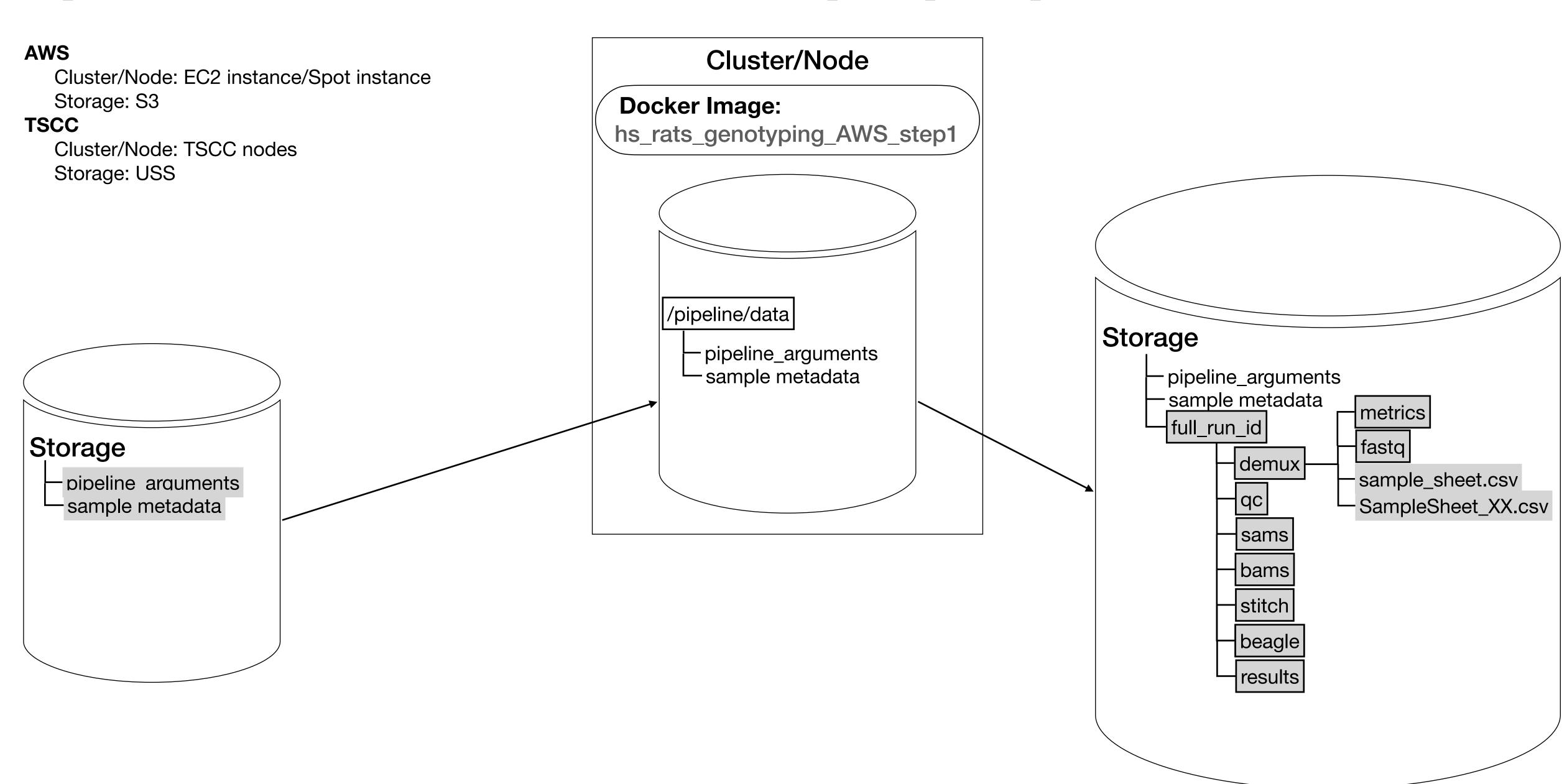
Genotyping Log Table



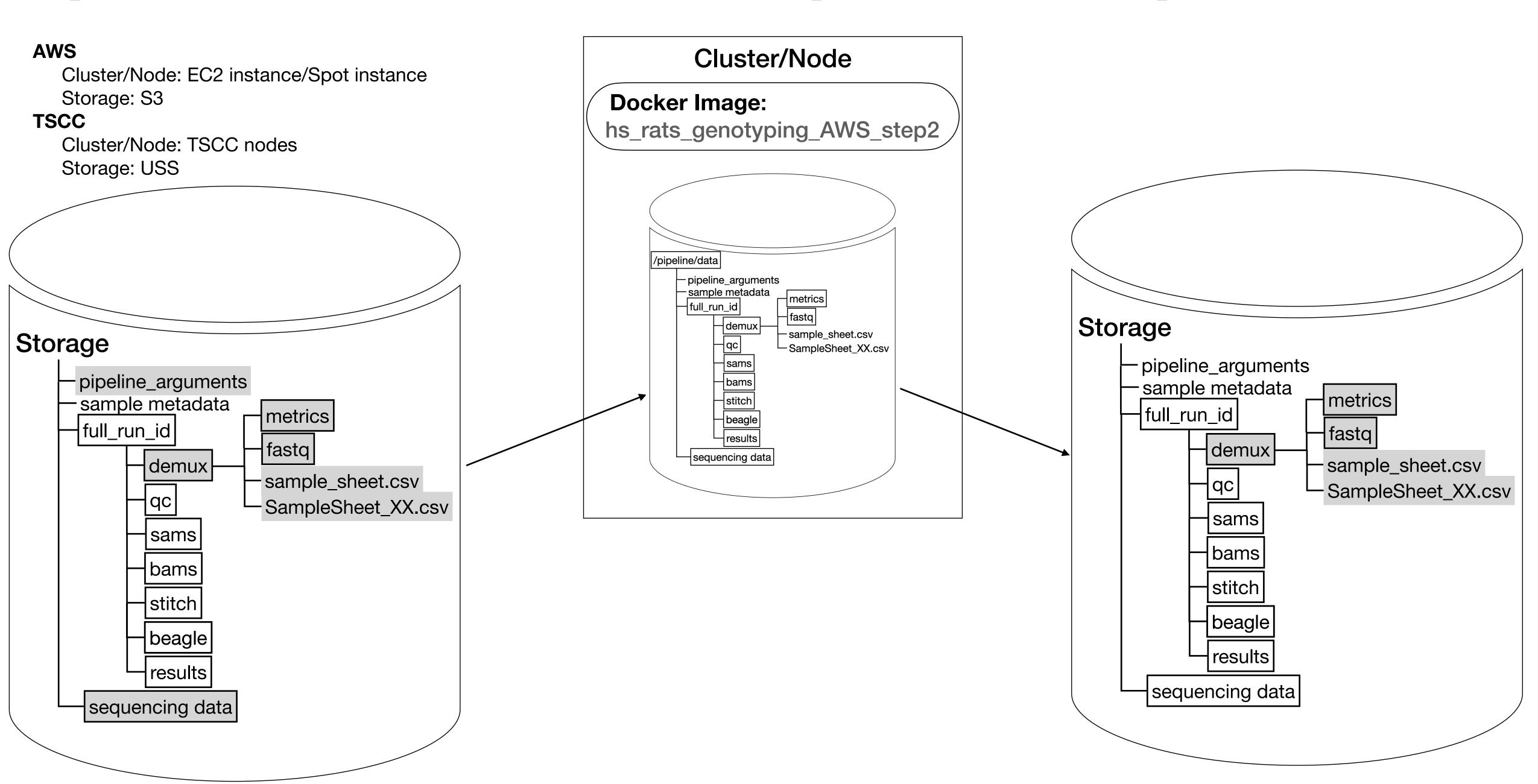


Directory Structure

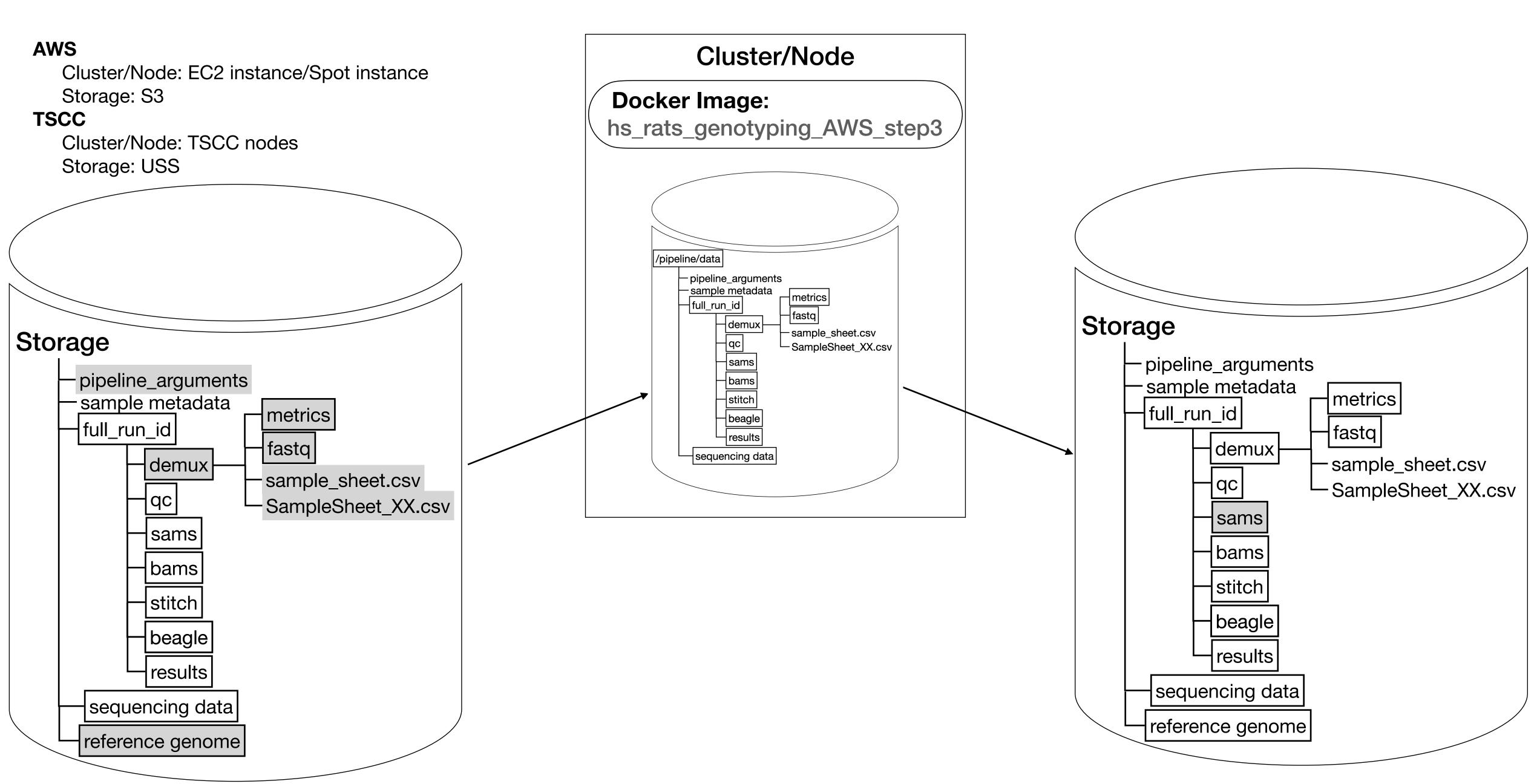
Pipeline with Docker - step1 preparation



Pipeline with Docker - step2 demultiplex



Pipeline with Docker - step3 alignment



Pipeline with Docker - step4 mark duplicates

