



Bioinformatics Workshop

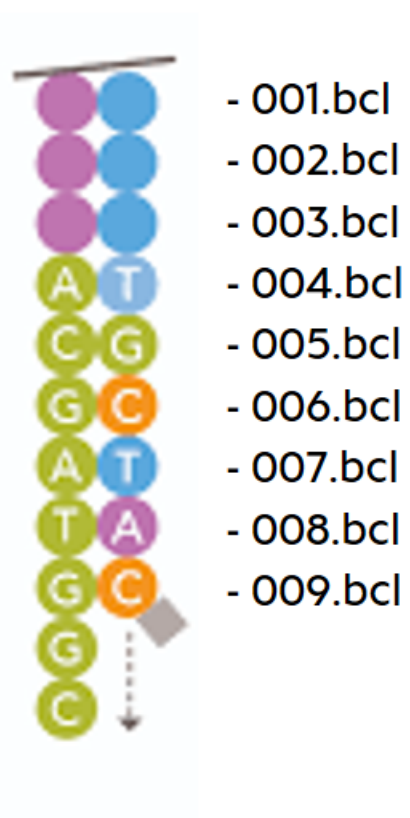
bcl2fastq

2.8.2023

Therese Muzeniek

NGS output (.bcl) to pipeline input (.fastq)

.bcl format



bcl2fastq

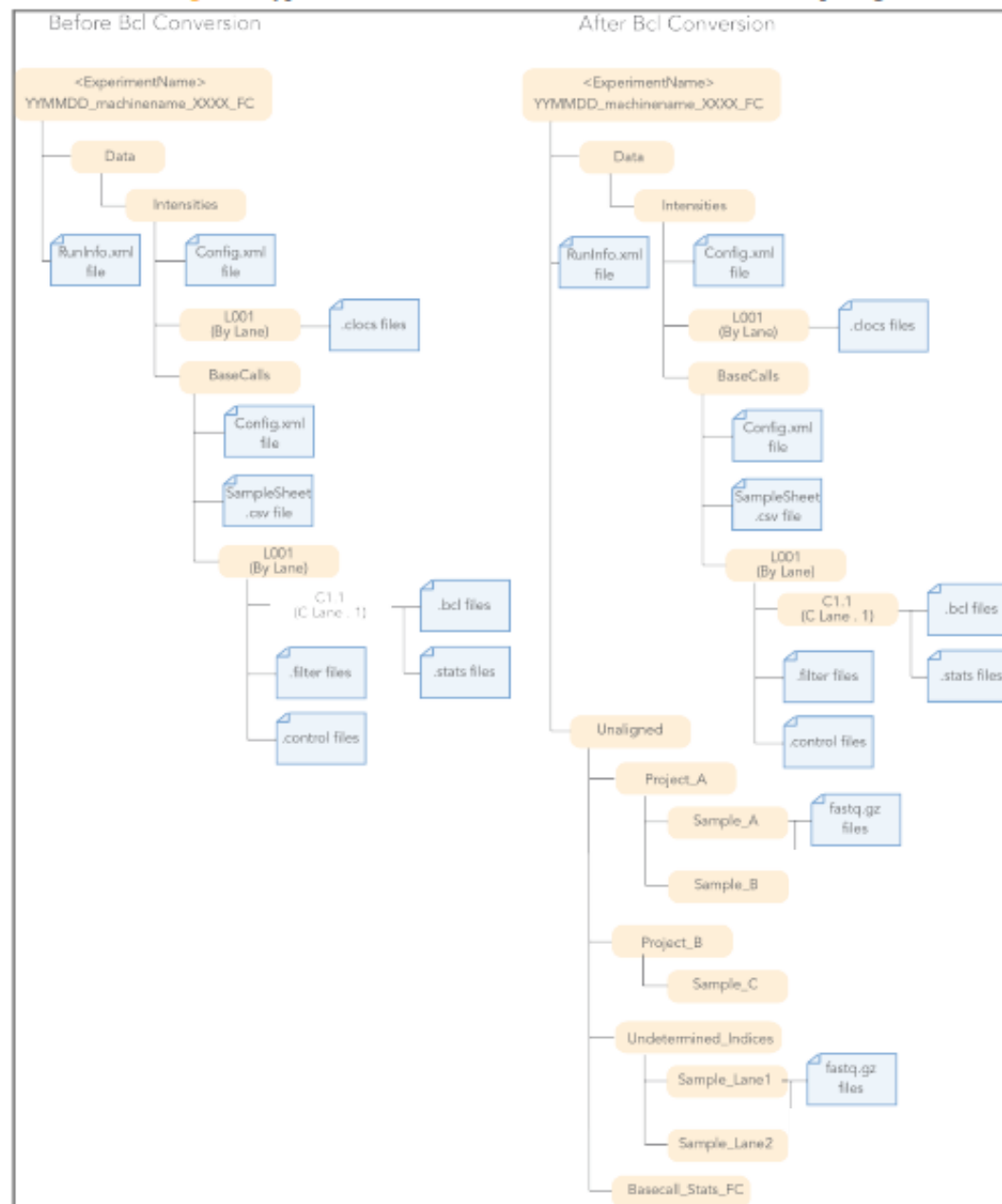
.FASTQ format



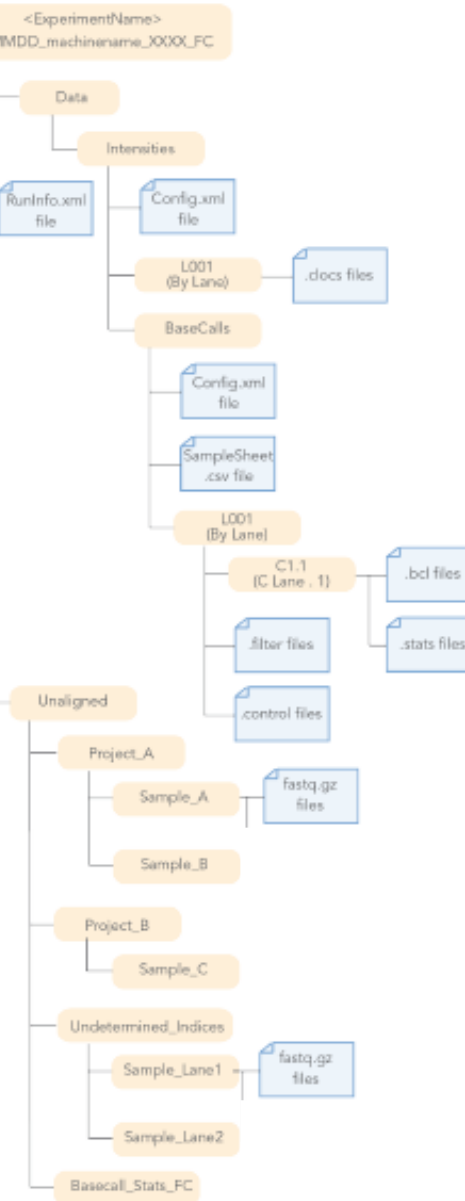
bcl2fastq

Illumina NGS run
Folder structure

Figure 1 Typical Run Folder Structure after Bcl Conversion and Demultiplexing



After Bcl Conversion



bcl2fastq



➤ .bcl files: How many files do we expect from a 300 cycle kit, paired end reads?

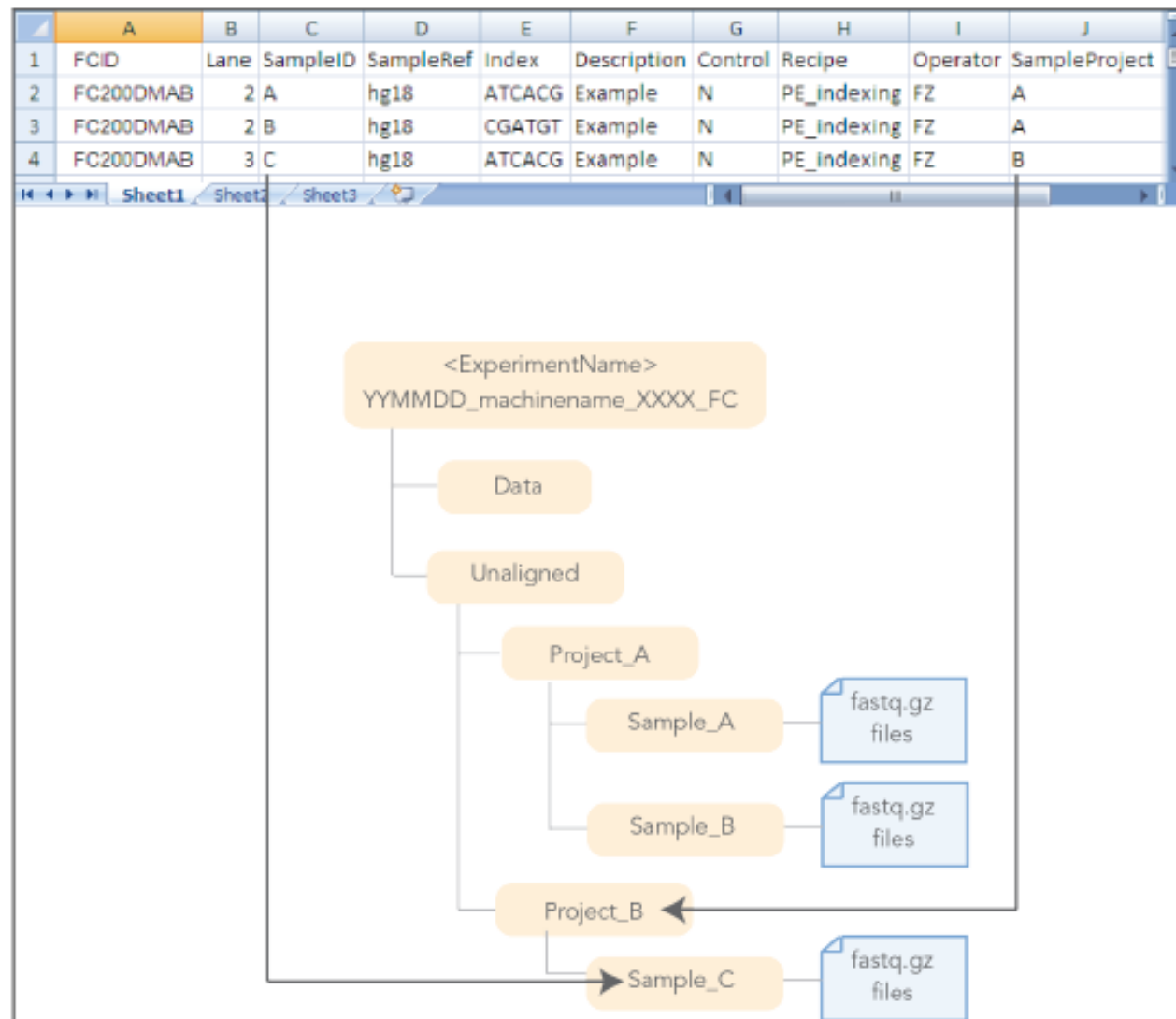
(318 files: 2 x 151 + 2 x 8 for the index sequencing)

➤ the bcl2fastq program needs a samples sheet (.csv) with sample information for demultiplexing

bcl2fastq

Sample sheet

Figure 2 Relation between Sample Sheet and Directory Structure



bcl2fastq

Input:

- Sample sheet in .csv format
 - Information to identify samples by their adapters
 - Sorting of reads by adapter to different folders
 - Incorrectly formatted data will cause an error
- .bcl files in the run folder

Output:

- FASTQ files, one folder per sample
- For PE: R1 and R2 in separate folders
 - Important for the next rule (fastp)