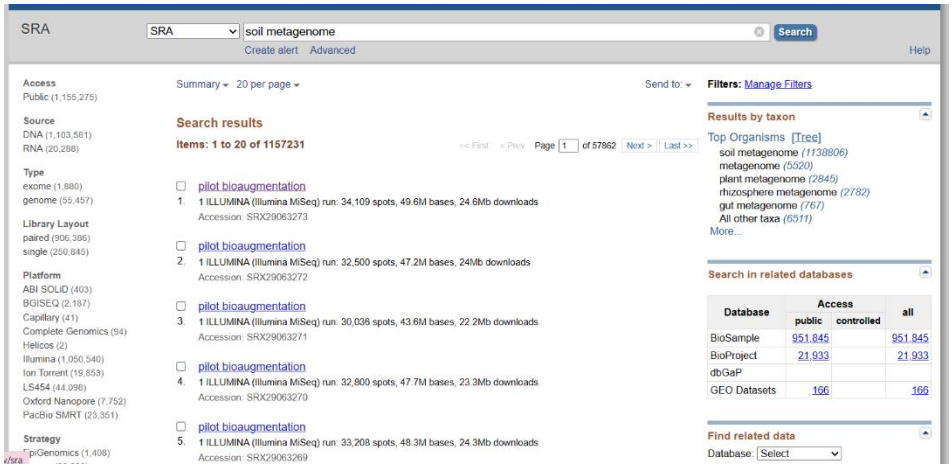
METAGENOMICS PROJECT:

For this project, I analysed a soil microbiome to identify antimicrobial resistance (AMR) genes. Soil is one of the most biologically diverse environments on Earth, home to vast microbial communities that play critical roles in nutrient cycling, plant growth, and ecosystem stability. Importantly, it also serves as a **natural reservoir of antimicrobial resistance (AMR) genes**. Many antibiotics used in medicine and agriculture are originally derived from soil bacteria, which have naturally evolved resistance mechanisms over millions of years. This helps -detect novel AMR Genes**,** understand resistance**,** monitor environmental spread and evaluate impact of human activity

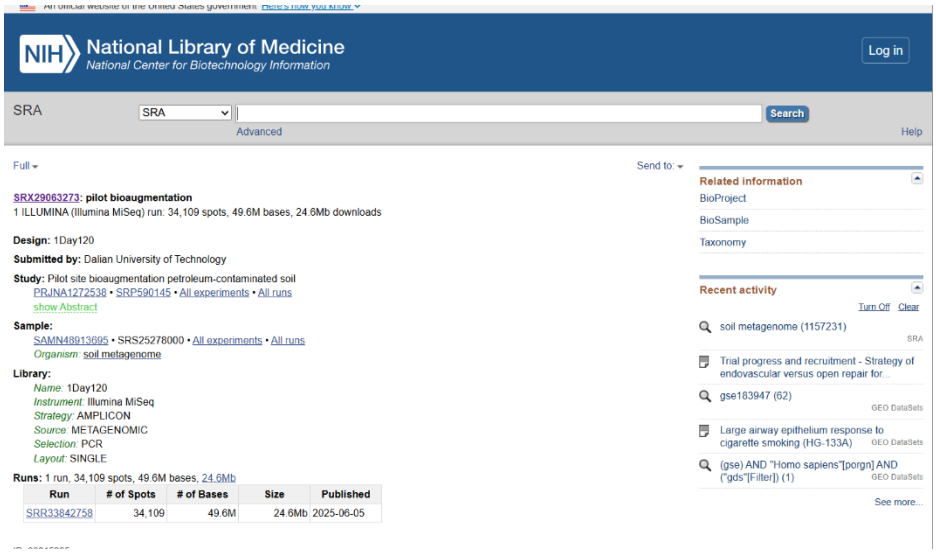
Aim:

Perform antibiotic resistance gene profiling on metagenomic sample **SRR287818** using the CARD-RGI tool and NGS TOOLS

Step1: SRA ID was obtained by searching soil metagenome in SRA database of NCBI



Step 2: After obtaining SRA ID details about it such as it single or paired end was checked



Step 3: Using prefetch and fastq dump split files commands in WSL the Sequence was downloaded

>fastq-dump SRR33842758

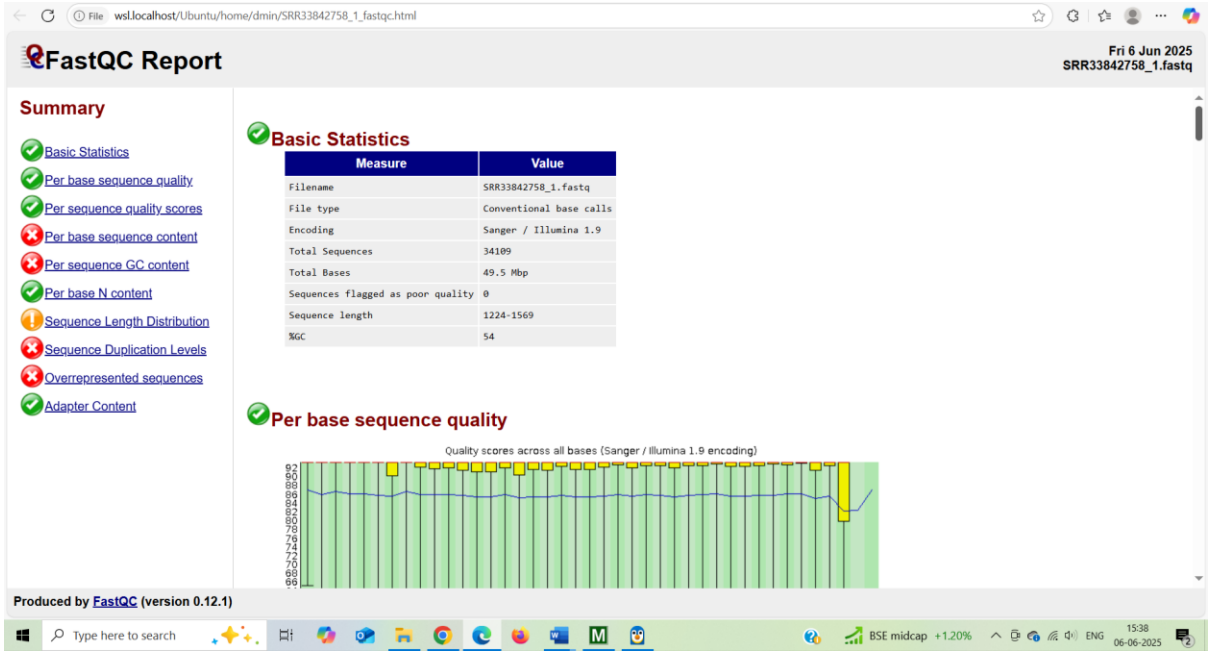
output

>Read 34109 spots for SRR33842758

Written 34109 spots for SRR33842758

Step 4: FastQC report of SRR33842758 was obtained

>fastqc SRR33842758.fastq



Step 5: To improve the quality of sequence, trimming using trim galore was performed

> trim_galore --quality 30 --length 20 –paired SRR33842758_1_fastq.gz

Step 6: Used Megahit tool to develop contigs

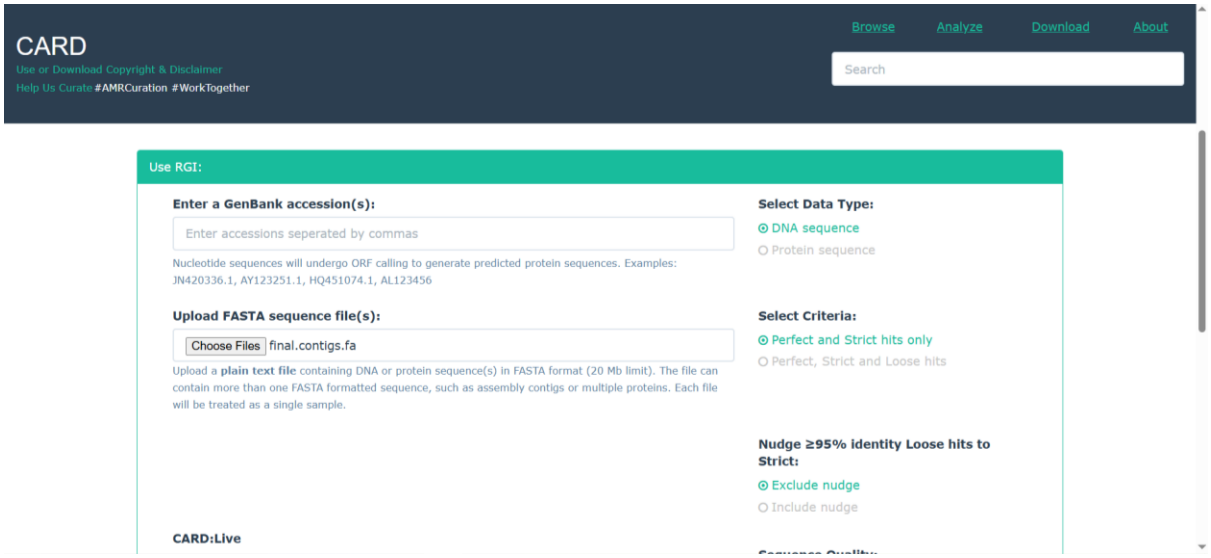
> sudo apt install megahit

> megahit -r SRR33842758_1_trimmed.fq -o megahit_output

> ls

> cd megahit_output

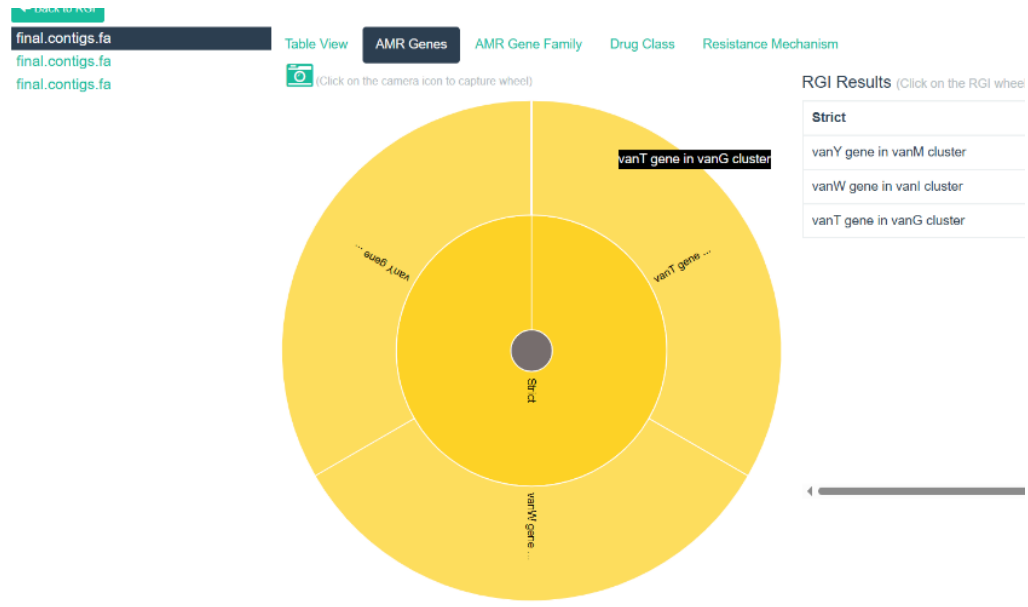
Step 7: Upload final.contigs.fa to CARD Database



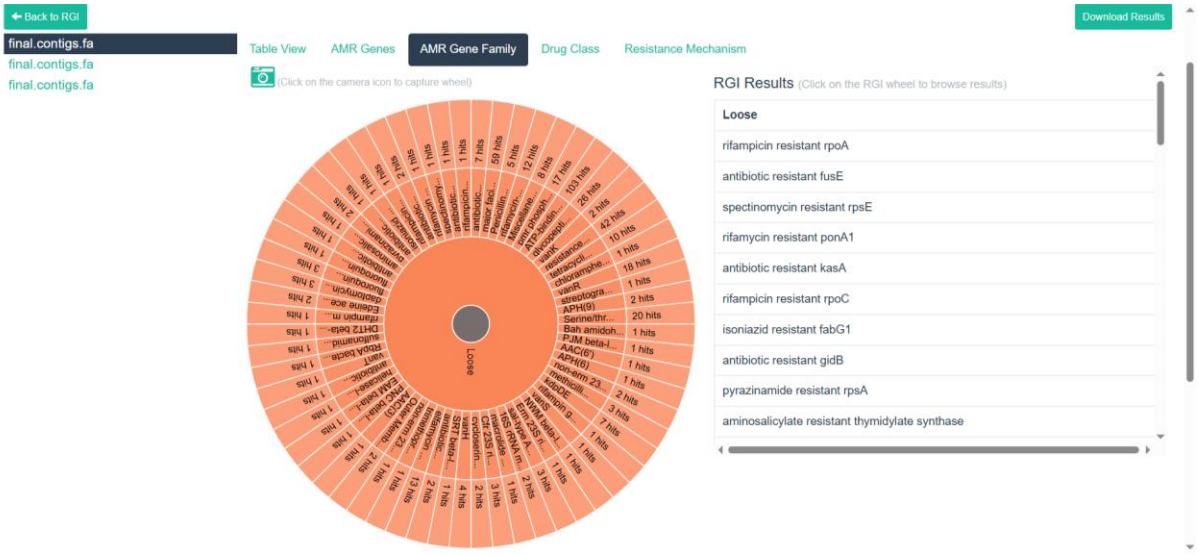
Step 8: Analyse drug resistant genes in Strict and lose hits

Hit	Criteria	Bitscore	Cut-Off	Percent Identity
vanT gene in vanG cluster	Strict	188	175	32.26
vanW gene in vanI cluster	Strict	98.2	50	34.5
vanY gene in vanM cluster	Strict	87.4	50	29.23

Strict Hit



Lose Hits



Step 9: Analyse different antibiotic resistance mechanisms involved-

reduced permeability to antibiotic
antibiotic inactivation
antibiotic target protection
antibiotic target replacement
antibiotic efflux
antibiotic target alteration

Conclusion:

For this project, I analyzed a soil microbiome to identify antimicrobial resistance (AMR) genes. I began by retrieving the relevant dataset from the NCBI SRA using its SRA ID and downloaded the raw sequencing data with prefetch and fastq-dump. After performing quality control with FastQC, I trimmed the reads using Trimmomatic to remove adapters and low-quality sequences. The cleaned reads were then assembled into contigs using MEGAHIT. Finally, I uploaded the assembled contigs to the CARD (Comprehensive Antibiotic Resistance Database) to identify AMR genes and determine the resistance mechanisms present in the sample.