Overview of steps in AMR++ with merged reads

Confirm everything works

You'll need to load the typical AMR++ environment, plus add "FLASH" and "SeqKit". Or you can simply re-install the AMR++ conda environment using the recipe as shown in the installation tutorial.

You'll still need to run the "demo" as this downloads the github with the SNP confirmation software. If you forget to do this, make sure your sbatch script includes the module that allows internet connection (e.g. "module load WebProxy").

Download a new AMR++ repository and switch to "dev" branch

```
git clone https://github.com/Microbial-Ecology-Group/AMRplusplus

cd AMRplusplus/

git pull origin dev
```

Full pipeline

You can run the full AMR++ pipeline (without kraken) by specifying the path to your `--reads` and using the `--pipeline merged_AMR` flag. This could work well if you re-direct the results or the work directory `-w /path/to/shared_drive`.

Step 1 : Run the "eval_qc" pipeline as normal with `--reads`

Step 2: Run "trim_qc" pipeline as normal with `--reads`

Modify trimming parameters as needed in the `params.txt` file.

Step 3: Run new "merge_reads" pipeline as normal with `--reads`

Parameters that have to change:

- `--pipeline` ==> `--pipeline merge_reads`

This will output two files per sample, the "extendedFrags" (merged) and "notCombined" (unmerged).

Step 4: Run "merged_rm_host" with a new flag, `--merged_reads`

Parameters to change

Parameters that have to change:

- `--pipeline` ==> `--pipeline merged_rm_host`
- `--merged_reads` ==> `--merged_reads 'Merged_AMR++_analysis/Flash_reads/*.{extendedFrags,notCombined}.fastq.gz'`
    - Remember, it's very important to use the single quote (') and not the backtick or backquote (`) that's on the same key as the tilde.
- `host` ==> `--host "/path/to/your/host/chr21.fasta.gz"`
    - remember, you can change this in `params.config` file or add it to your nextflow command.
    - On grace, bovine: `/scratch/group/big_scratch/SHARED_resources/host_genome/GCF_002263795.3_ARS-UCD2.0_genomic.fna`

Example command:

```
nextflow run main_main++.nf --pipeline merged_rm_host --output
Merged_AMR++_analysis --merged_reads 'Merged_AMR++_analysis/Flash_reads/*.
{extendedFrags,notCombined}.fastq.gz' -profile local
```

Step 5: Run "merged_resistome" and point to non host reads with `--merged_reads`

Parameters that have to change:

- `--pipeline` ==> `--pipeline merged_resistome`
- `--merged_reads` ==> `--merged_reads 'Merged_AMR++_analysis/HostRemoval/NonHostFastq/*.{merged,unmerged}.non.host.fastq.gz'`

SNP confirmation and count deduplication is performed by default.

Example command:

```
nextflow run main_main++.nf --pipeline merged_resistome --output
Merged_AMR++_analysis --merged_reads 'Merged_AMR++_analysis/Flash_reads/*.
{extendedFrags,notCombined}.fastq.gz' -profile local
```