

# Configuration Manual

MSc Research Project  
Data Analytics

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**National College of Ireland**  
**MSc Project Submission Sheet**  
**School of Computing**



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**Programme:** Master of Science in Data Analytics                      **Year:** SEP 2022-23.  
 .....  
**Module:** Research Project  
 .....  
**Lecturer:** Qurrat Ul Ain  
 .....  
**Submission Due Date:** 14/08/2023  
 .....  
**Project Title:** Application of Supervised Learning Classifiers on Gut Microbial data to Predict Parkinson Disease  
 .....  
**Word Count:** ..... **Page Count:** .....

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# Configuration Manual

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Student ID: 21208808

## 1 Introduction

The step-by-step procedure for setting up the environment and carrying out this research study is thoroughly described in this configuration handbook. This document contains details on the tools, software programming language, library packages, and system hardware settings.

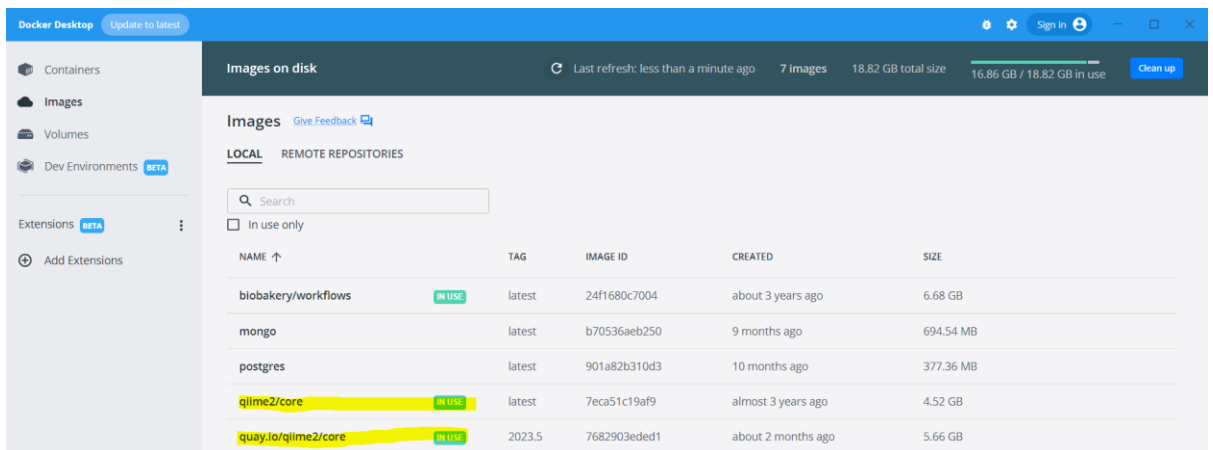
The document also covers the steps like input, output artifacts and command line instructions required from installation step up to end outcomes of the research project.

## 2 Environment set up Instructions.

### 2.1 Software Specifications

The tools required for this project are as follows.

- 1) Based the Operating system environment QIIME can be installed on any platform as mentioned in install documentation (QIIME 2 user documentation, n.d.). This research project installed QIIME2 tool on Docker<sup>1</sup>.



- 2) Latest Version QIIME 2 releases: 2023.5, (QIIME 2 user documentation, n.d.) is used for this project using Docker. This project installed QIIME on Docker.

<sup>1</sup> <https://www.docker.com/products/docker-desktop/>

```
Command Prompt - docker run -t -i -v C:/Users/madhu/./data quay.io/qiime2/core:2023.5
Microsoft Windows [Version 10.0.19045.3208]
(c) Microsoft Corporation. All rights reserved.

C:\Users\madhu>docker run -t -i -v C:/Users/madhu/./data quay.io/qiime2/core:2023.5
```

3) FastQC tool <sup>2</sup> for checking the quality of the sequences downloaded.

Software	Version
Docker	v0.11.2-desktop.1
QIIME 2	2023.5
FastQC	Version 0.12.0

## 2.2 Hardware Specifications

Below are the hardware specifications with which this research project is carried out.

Hardware	Specifications
Operating System	Windows 10 Home Version 22H2
Processor	Intel(R) Core (TM) i7-8550U CPU @ 1.80GHz 2.00 GHz
RAM	16.0 GB
Hard Disk	450 GB

## 2.3 Source data

The data for this is collected from open-source database European Nucleotide Archive (ENA) which is a repository storing annotated DNA and RNA sequences. The 17 selected samples from the project PRJEB27564 <sup>3</sup> are used for this study, out of which 8 faecal samples are from healthy individuals and 9 faecal samples from Parkinson's patient.

The raw sequence fastq files which are pair end reads with separate files for forward and reverse reads are first downloaded on to the local disk which is mounted on Docker Desktop.

<sup>2</sup> <https://www.bioinformatics.babraham.ac.uk/projects/fastqc/>

<sup>3</sup> <https://www.ebi.ac.uk/ena/browser/view/PRJEB27564>

```

Command Prompt - docker run -t -i -v C:/Users/madhur/data/quay.io/qiime2/core:2023.5
Application config directory
/opt/conda/envs/qiime2-2023.5/var/q2cli

Getting help
To get help with QIIME 2, visit https://qiime2.org
(qiime2-2023.5) root@92247823ecf6:/data# wget -nc ftp.sra.ebi.ac.uk/vol1/run/ERR273/ERR2730240/P-53-0_R1_001.fastq.gz
will not apply HSTS. The HSTS database must be a regular and non-world-writable file.
ERROR: could not open HSTS store at '/home/qiime2/.wget-hsts'. HSTS will be disabled.
--2023-08-12 10:33:55-- http://ftp.sra.ebi.ac.uk/vol1/run/ERR273/ERR2730240/P-53-0_R1_001.fastq.gz
Resolving ftp.sra.ebi.ac.uk (ftp.sra.ebi.ac.uk)... 193.62.193.138
Connecting to ftp.sra.ebi.ac.uk (ftp.sra.ebi.ac.uk)[193.62.193.138]:80... connected.
HTTP request sent, awaiting response... 200 OK
length: 392968 (384K) [application/x-gzip]
Saving to: 'P-53-0_R1_001.fastq.gz'

P-53-0_R1_001.fastq.gz      100%[=====] 383.76K  2.33MB/s   in 0.2s

2023-08-12 10:34:01 (2.33 MB/s) - 'P-53-0_R1_001.fastq.gz' saved [392968/392968]

(qiime2-2023.5) root@92247823ecf6:/data# wget -nc ftp.sra.ebi.ac.uk/vol1/run/ERR273/ERR2730240/P-53-0_R2_001.fastq.gz
will not apply HSTS. The HSTS database must be a regular and non-world-writable file.
ERROR: could not open HSTS store at '/home/qiime2/.wget-hsts'. HSTS will be disabled.
--2023-08-12 10:34:05-- http://ftp.sra.ebi.ac.uk/vol1/run/ERR273/ERR2730240/P-53-0_R2_001.fastq.gz
Resolving ftp.sra.ebi.ac.uk (ftp.sra.ebi.ac.uk)... 193.62.193.138, 193.62.193.138
Connecting to ftp.sra.ebi.ac.uk (ftp.sra.ebi.ac.uk)[193.62.193.138]:80... connected.
HTTP request sent, awaiting response... 200 OK
length: 468536 (458K) [application/x-gzip]
Saving to: 'P-53-0_R2_001.fastq.gz'

P-53-0_R2_001.fastq.gz      100%[=====] 457.55K  861KB/s   in 0.5s

2023-08-12 10:34:06 (861 KB/s) - 'P-53-0_R2_001.fastq.gz' saved [468536/468536]

(qiime2-2023.5) root@92247823ecf6:/data#

```

Below attachment gives the list of all the sample files used for this study downloaded from ENA repository.



Raw\_seq\_files\_list\_EN  
A download.txt

### 3 Implementation

Implementation for this study is mainly carried on QIIME 2 tool for the analysis of the 16SrRNA sequence to derive the features, ASV's and taxonomy abundance. With QIIME 2 installed on docker the implementation instructions are through command line interface.

All the commands from the beginning of installation to model evaluation is provided in the below attachment file.



q2cli\_parkison\_sampl  
e\_prediction.bash

#### 3.1 Importing

The quality control of the downloaded raw files is first checked with external tool FastQC software.

Then the downloaded sample files on local disk are imported into the QIIME 2 tool for analysis. This tool as a framework to process these files (QIIME 2 development team, n.d.). Depending on the input files format and layout importing to QIIME 2 tool the steps differ. For this study the sample files are 16S rRNA demultiplexed pair end sequences fastq files. All possible file formats and importing instructions are provided in QIIME documentation (QIIME 2 user documentation, n.d.) (QIIME2 development team, n.d.).Manifest file format import is used,this file has sample id's and its absolute path for import.

sample-id	forward-absolute-filepath	reverse-absolute-filepath
C118	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-118-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-118-O_R2_001.fastq.gz
P60	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-60-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-60-O_R2_001.fastq.gz
C18	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-18-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-18-O_R2_001.fastq.gz
C30	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-30-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-30-O_R2_001.fastq.gz
C48	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-48-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-48-O_R2_001.fastq.gz
C51	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-51-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-51-O_R2_001.fastq.gz
C80	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-80-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-80-O_R2_001.fastq.gz
C82	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-82-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-82-O_R2_001.fastq.gz
C137	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-137-N_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/C-137-N_R2_001.fastq.gz
P16	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-16-N_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-16-N_R2_001.fastq.gz
P31	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-31-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-31-O_R2_001.fastq.gz
P53	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-53-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-53-O_R2_001.fastq.gz
P77	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-77-N_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-77-N_R2_001.fastq.gz
P99	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-99-N_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-99-N_R2_001.fastq.gz
P103	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-103-N_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-103-N_R2_001.fastq.gz
P116	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-116-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-116-O_R2_001.fastq.gz
P120	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-120-O_R1_001.fastq.gz	/data/ENA_PRJEB27564_SAMPLES/ENA_PRJEB27564_SAMPLES/P-120-O_R2_001.fastq.gz

```

C:\ Command Prompt - docker run -t -i -v C:/Users/madhu/;/data quay.io/qiime2/core:2023.5

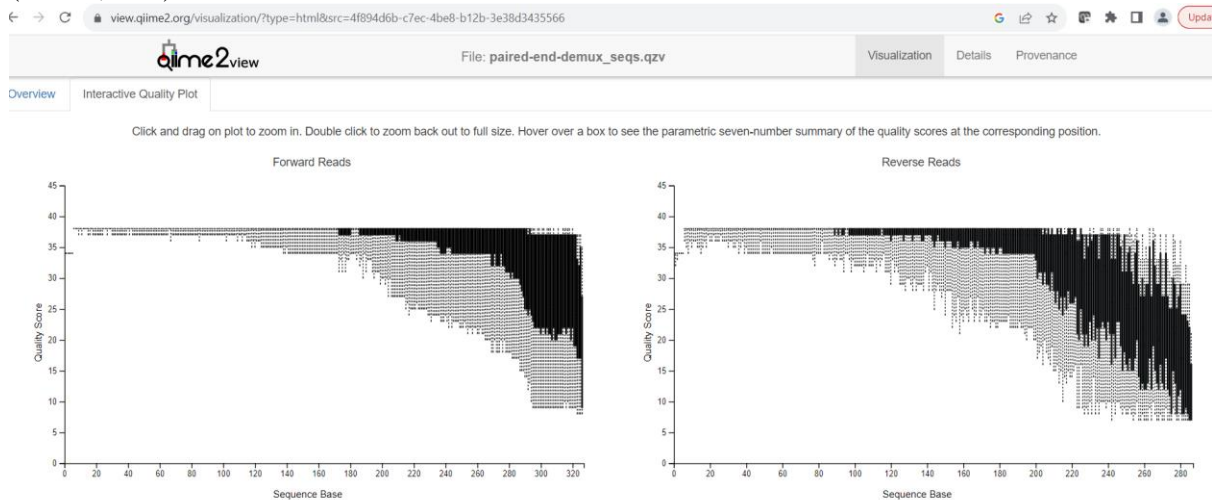
```

```

(qiime2-2023.5) root@92247823ecf6:/data#
(qiime2-2023.5) root@92247823ecf6:/data# qiime tools import \
> --type 'SampleData[PairedEndSequencesWithQuality]' \
> --input-format PairedEndFastqManifestPhred33V2 \
> --input-path ./pair_end_manifest.tsv \
> --output-path ./paired-end-demux.qza
Imported ./pair_end_manifest.tsv as PairedEndFastqManifestPhred33V2 to ./paired-end-demux.qza
(qiime2-2023.5) root@92247823ecf6:/data# qiime demux summarize \
> --i-data ./paired-end-demux.qza \
> --o-visualization ./paired-end-demux_seqs.qzv
Saved Visualization to: ./paired-end-demux_seqs.qzv
(qiime2-2023.5) root@92247823ecf6:/data#

```

QIIME2 offers a interface to the view .qza and .qzv artifacts files on a browser directly. (Anon., n.d.).



### 3.2 Data Analysis

In this step sequence quality control is carried out. Denoising the sequences using deblur methods and feature table and representative sequences are generated. This is the most important and critical step for further analysis as the artifacts generated at this step are used for various analysis like taxonomy classification, finding alpha diversity, beta diversity and sample classification.

### A) Joining Pair end Reads:

As the fastq files are pair end reads, sequences need to be joined because deblur method works on single end layout files. Using merge pairs in vsearch method forward and reverse read files are joined. Below statements accomplish this task in QIIME 2.

```
cmd Select Command Prompt - docker run -t -i -v C:/Users/madhu/;/data quay.io/qiime2/core:2023.5
(qiime2-2023.5) root@92247823ecf6:/data# Y
bash: Y: command not found
(qiime2-2023.5) root@92247823ecf6:/data# qiime vsearch merge-pairs \
> --i-demultiplexed-seqs paired-end-demux.qza \
> --o-merged-sequences paired-joined-reads.qza \
> --p-threads 8
Saved SampleData[JoinedSequencesWithQuality] to: paired-joined-reads.qza
(qiime2-2023.5) root@92247823ecf6:/data# qiime quality-filter q-score \
> --i-demux paired-joined-reads.qza \
> --o-filtered-sequences paired-joined-filtered.qza \
> --o-filter-stats paired-joined-filter-stats.qza
Saved SampleData[JoinedSequencesWithQuality] to: paired-joined-filtered.qza
Saved QualityFilterStats to: paired-joined-filter-stats.qza
(qiime2-2023.5) root@92247823ecf6:/data# qiime demux summarize \
> --i-data paired-joined-filtered.qza \
> --o-visualization paired-joined-filtered.qzv
Saved Visualization to: paired-joined-filtered.qzv
```

### B) Denoising using Deblur:

Once the pair end read are joined the out artifacts are single end reads and these reads are input artifacts to the deblur denoise-16S method available in QIIME 2 as a method. The sequences are trimmed to length 300 where quality score of the sequences started to decrease.



Select Command Prompt - docker run -t -i -v C:/Users/madhu:/data quay.io/qiime2/core:2023.5

```
(qiime2-2023.5) root@92247823ecf6:/data# qiime deblur denoise-16S \
> --i-demultiplexed-seqs paired-joined-filtered.qza \
> --p-trim-length 300 \
> --p-sample-stats \
> --p-jobs-to-start 10 \
> --o-stats paired-deblur-stats.qza \
> --o-representative-sequences paired-rep-seqs-deblur.qza \
> --o-table paired-table-deblur.qza
Saved FeatureTable[Frequency] to: paired-table-deblur.qza
Saved FeatureData[Sequence] to: paired-rep-seqs-deblur.qza
Saved DeblurStats to: paired-deblur-stats.qza
(qiime2-2023.5) root@92247823ecf6:/data# qiime deblur visualize-stats \
> --i-deblur-stats paired-deblur-stats.qza \
> --o-visualization paired-deblur-stats.qzv
Saved Visualization to: paired-deblur-stats.qzv
(qiime2-2023.5) root@92247823ecf6:/data# qiime feature-table tabulate-seqs \
> --i-data paired-rep-seqs-deblur.qza \
> --o-visualization paired-rep-seqs-deblur.qzv
Saved Visualization to: paired-rep-seqs-deblur.qzv
(qiime2-2023.5) root@92247823ecf6:/data# qiime feature-table summarize \
> --i-table paired-table-deblur.qza \
> --m-sample-metadata-file pair_end_meta.tsv \
> --o-visualization paired-table-deblur.qzv
Saved Visualization to: paired-table-deblur.qzv
```

## Feature table view

view.qiime2.org/visualization/?type=html&src=899a3177-54d0-4221-bb99-3c5b8ef32e2f

qiime2view File: paired-table-deblur.qzv Visualization Details Provenance

Overview Interactive Sample Detail Feature Detail

	Frequency	# of Samples Observed In
2578d4cc429aedab5958b83b36f08643	1,327	16
7397e3519223287db339e7c9561502e8	1,179	13
c80f6892bba04e804f733a349a9940e5	1,168	14

## Denoise statistics view

view.qiime2.org/visualization/?type=html&src=3a2519cf-1594-47dc-98bc-2b37c3ca1df4

qiime2view File: paired-deblur-stats.qzv Visualization Details Provenance

### Per-sample Deblur stats

Click on a Column header to sort the table.  
Mouse over a Column header to get a description.

sample-id	reads-raw	fraction-artifact-with-minsize	fraction-artifact	fraction-missed-reference	unique-reads-derep	reads-derep	unique-reads-deblur	reads-deblur	unique-reads-hit-artifact	reads-hit-artifact	unique-reads-chimeric	reads-chimeric	unique-reads-hit-reference	reads-hit-reference	unique-reads-missed-reference	reads-missed-reference	
0	P53	2415	0.583023	0.0	0.0	345	1007	306	680	0	0	37	46	146	378	0	0
1	P31	2935	0.428279	0.0	0.0	397	1678	306	1150	0	0	22	35	215	991	0	0
2	C118	3374	0.408121	0.0	0.0	453	1997	339	1294	0	0	26	44	216	1050	0	0
3	P103	25346	0.366133	0.0	0.0	2224	16066	1363	7563	0	0	302	582	500	5554	0	0
4	C18	27322	0.351914	0.0	0.0	2283	17707	1337	8195	0	0	303	711	499	6209	0	0
5	P16	10041	0.332238	0.0	0.0	1236	6705	800	3782	0	0	85	134	422	3011	0	0

## Representative sequence's view

The screenshot shows the qiime2view interface for a file named 'paired-rep-seqs-deblur.qzv'. It features two main sections: 'Sequence Length Statistics' and 'Seven-Number Summary of Sequence Lengths'.

**Sequence Length Statistics:**

Sequence Count	Min Length	Max Length	Mean Length	Range	Standard Deviation
2165	300	300	300.0	0	0.0

**Seven-Number Summary of Sequence Lengths:**

Percentile:	2%	9%	25%	50%	75%	91%	98%
Length* (nts):	300	300	300	300	300	300	300

\*Values rounded down to nearest whole number.

**Sequence Table:**

To BLAST a sequence against the NCBI nt database, click the sequence and then click the View report button on the resulting page.

Download your sequences as a raw FASTA file

Click on a Column header to sort the table.

Feature ID	Sequence Length	Sequence
c80f6892bba04e804f733a349a9940e5	300	TAGTGTCTACCGGAGGCAGCAGTGGGGAATATTGCACAATGGGCGAAAGCTGATGCAGCGACGCCGCGTGAGCGAAGAAGTATTTCGGTATGTAAAGCTCTATCAGCAGGGAAGATAATGACGGTACTGACTAAGAAGCACCGGCT

### C) Taxonomy Classification using Feature Classifier:

Once the feature table is extracted by the deblur method, further analysis on taxonomy classification using pretrained classifier with silva and green genes reference database is performed.

```

C:\> Select Command Prompt - docker run -t -i -v C:/Users/madhu/;/data quay.io/qiime2/core:2023.5
(qiime2-2023.5) root@92247823ecf6:/data# qiime feature-classifier classify-sklearn
taxonomy.qza
Saved FeatureData[Taxonomy] to: ./taxonomy.qza
(qiime2-2023.5) root@92247823ecf6:/data# qiime metadata tabulate \
> --m-input-file paired-deblur-taxonomy_gg.qza \
> --o-visualization paired-deblur-taxonomy-summary.qzv
Saved Visualization to: paired-deblur-taxonomy-summary.qzv
(qiime2-2023.5) root@92247823ecf6:/data# qiime metadata tabulate \
> --m-input-file paired-deblur-taxonomy_silva.qza \
> --o-visualization paired-deblur-taxonomy-summary_silva.qzv
Saved Visualization to: paired-deblur-taxonomy-summary_silva.qzv
  
```

Taxonomy summary report is generated to see the levels and abundance of the microbial species per feature. With level of confident the species is predicted with reference database.

The screenshot shows the qiime2view interface for a file named 'paired-deblur-taxonomy-summary\_silva.qzv'. It displays a table with taxonomy information for several features.

Download metadata TSV file

This file won't necessarily reflect dynamic sorting or filtering options based on the interactive table below.

Search:

Feature ID	Taxon	Confidence
0012dea73e04186111f35484c67ae63f	d__Bacteria; p__Firmicutes; c__Clostridia; o__Clostridia_UCG-014; f__Clostridia_UCG-014; g__Clostridia_UCG-014	0.9999984555068843
0016d5082628fd1329d4de018f629ecc	d__Bacteria; p__Bacteroidota; c__Bacteroidia; o__Bacteroidales; f__Prevotellaceae; g__Prevotella	0.9998674573837184
002b22d996b390fb4044f66b41710c3	d__Bacteria; p__Firmicutes; c__Clostridia; o__Lachnospirales; f__Lachnospiraceae; g__Butyrivibrio; s__Butyrivibrio_crossotus	0.8347686539798219
003192b088bd7bc09ab579d00cf4574	d__Bacteria; p__Bacteroidota; c__Bacteroidia; o__Bacteroidales; f__Bacteroidaceae; g__Bacteroides; s__Bacteroides_fragilis	0.8751521885467743

```

❏ Select Command Prompt - docker run -t -i -v C:/Users/madhu:/data quay.io/qiime2/core:2023.5
(qiime2-2023.5) root@92247823ecf6:/data# qiime taxa collapse --i-table paired-table-deblur.qza
silva.qza
Saved FeatureTable[Frequency] to: paired-collapsed-deblur-table_silva.qza
(qiime2-2023.5) root@92247823ecf6:/data# qiime taxa collapse \
> --i-table paired-table-deblur.qza \
> --i-taxonomy paired-deblur-taxonomy_gg.qza \
> --p-level 6 \
> --o-collapsed-table paired-collapsed-deblur-table_gg.qza
Saved FeatureTable[Frequency] to: paired-collapsed-deblur-table_gg.qza
(qiime2-2023.5) root@92247823ecf6:/data# qiime feature-table relative-frequency \
> --i-table paired-collapsed-deblur-table_gg.qza \
> --o-relative-frequency-table paired-relative-abundance-deblur_gg.qza
Saved FeatureTable[RelativeFrequency] to: paired-relative-abundance-deblur_gg.qza
(qiime2-2023.5) root@92247823ecf6:/data# qiime feature-table summarize \
> --i-table paired-relative-abundance-deblur_gg.qza \
> --o-visualization paired-relative-abundance-deblur-summary_gg.qzv
Saved Visualization to: paired-relative-abundance-deblur-summary_gg.qzv
(qiime2-2023.5) root@92247823ecf6:/data#
(qiime2-2023.5) root@92247823ecf6:/data# qiime feature-table filter-samples \
> --i-table ./paired-table-deblur.qza \
> --p-min-frequency 2000 \
> --o-filtered-table ./table_2k.qza
Saved FeatureTable[Frequency] to: ./table_2k.qza
(qiime2-2023.5) root@92247823ecf6:/data# qiime taxa barplot \
> --i-table ./table_2k.qza \
> --i-taxonomy ./paired-deblur-taxonomy_silva.qza \
> --m-metadata-file ./pair_end_meta.tsv \
> --o-visualization ./taxa_barplot_silva.qzv
Saved Visualization to: ./taxa_barplot_silva.qzv
(qiime2-2023.5) root@92247823ecf6:/data# qiime taxa barplot \
> --i-table ./table_2k.qza \
> --i-taxonomy ./paired-deblur-taxonomy_gg.qza \
> --m-metadata-file ./pair_end_meta.tsv \
> --o-visualization ./taxa_barplot_gg.qzv
Saved Visualization to: ./taxa_barplot_gg.qzv
(qiime2-2023.5) root@92247823ecf6:/data#

```

### 3.3 Model

Supervised machine learning classifier algorithms built based on sci-kit learn python packages provided in QIIME 2 to predict the samples susceptible to Parkinson's disease. —p-estimator argument to the plugin gives a list of classifiers to choose (scikit-learn developers, n.d.). Meta data file is provided as one of the input artifacts to the sample classifier which give the samples meta data as shown. Based the meta data column DiseaseStatus which serves as labelled column for the model training and predicts the test samples.

SampleID	Group	Description	SubjectID	TimePoint	Hostbodyproduct	Geographiclocation	Age	Sex	DiseaseStatus
C118	Control	human-gut	C-118-0	baseline	Stool	Finland	N/A	N/A	HC
P60	Case	human-gut	P-60-0	baseline	Stool	Finland	N/A	N/A	PD
P116	Case	human-gut	P-116-0	baseline	Stool	Finland	N/A	N/A	PD
P120	Case	human-gut	P-120-0	baseline	Stool	Finland	N/A	N/A	PD
C80	Control	human-gut	C-80-0	baseline	Stool	Finland	N/A	N/A	HC
C51	Control	human-gut	C-51-0	baseline	Stool	Finland	N/A	N/A	HC
C82	Control	human-gut	C-82-0	baseline	Stool	Finland	N/A	N/A	HC
P77	Case	human-gut	P-77-0	baseline	Stool	Finland	N/A	N/A	PD
C18	Control	human-gut	C-18-0	baseline	Stool	Finland	N/A	N/A	HC
C48	Control	human-gut	C-48-0	baseline	Stool	Finland	N/A	N/A	HC
P103	Case	human-gut	P-103-0	baseline	Stool	Finland	N/A	N/A	PD
C137	Control	human-gut	C-137-0	baseline	Stool	Finland	N/A	N/A	HC
C30	Control	human-gut	C-30-0	baseline	Stool	Finland	N/A	N/A	HC
P99	Case	human-gut	P-99-0	baseline	Stool	Finland	N/A	N/A	PD
P16	Case	human-gut	P-16-0	baseline	Stool	Finland	N/A	N/A	PD
P31	Case	human-gut	P-31-0	baseline	Stool	Finland	N/A	N/A	PD
P53	Case	human-gut	P-53-0	baseline	Stool	Finland	N/A	N/A	PD

## Sample classifier with default inputs

```

C:\ Command Prompt - docker run -t -i -v C:/Users/madhu/./data quay.io/qiime2/core:2023.5
(qiime2-2023.5) root@92247823ecf6:/data# qiime sample-classifier classify-samples \
> --i-table ./paired-table-deblur.qza \
> --m-metadata-file ./pair_end_meta.tsv \
> --m-metadata-column DiseaseStatus \
> --output-dir ./pairend-classifier-results-E3
Saved SampleEstimator[Classifier] to: ./pairend-classifier-results-E3/sample_estimator.qza
Saved FeatureData[Importance] to: ./pairend-classifier-results-E3/feature_importance.qza
Saved SampleData[ClassifierPredictions] to: ./pairend-classifier-results-E3/predictions.qza
Saved Visualization to: ./pairend-classifier-results-E3/model_summary.qzv
Saved Visualization to: ./pairend-classifier-results-E3/accuracy_results.qzv
Saved SampleData[Probabilities] to: ./pairend-classifier-results-E3/probabilities.qza
Saved Visualization to: ./pairend-classifier-results-E3/heatmap.qzv
Saved SampleData[TrueTargets] to: ./pairend-classifier-results-E3/training_targets.qza
Saved SampleData[TrueTargets] to: ./pairend-classifier-results-E3/test_targets.qza

```

## Sample classifier with estimator Random Forest Classifier with n estimators decision trees to 50.

```

C:\ Command Prompt - docker run -t -i -v C:/Users/madhu/./data quay.io/qiime2/core:2023.5
(qiime2-2023.5) root@92247823ecf6:/data# qiime sample-classifier classify-samples \
> --i-table paired-table-deblur.qza \
> --m-metadata-file pair_end_meta.tsv \
> --m-metadata-column DiseaseStatus \
> --p-optimize-feature-selection \
> --p-parameter-tuning \
> --p-estimator RandomForestClassifier \
> --p-n-estimators 50 \
> --p-test-size 0.3 \
> --output-dir paired-end-classifier-E4
Saved SampleEstimator[Classifier] to: paired-end-classifier-E4/sample_estimator.qza
Saved FeatureData[Importance] to: paired-end-classifier-E4/feature_importance.qza
Saved SampleData[ClassifierPredictions] to: paired-end-classifier-E4/predictions.qza
Saved Visualization to: paired-end-classifier-E4/model_summary.qzv
Saved Visualization to: paired-end-classifier-E4/accuracy_results.qzv
Saved SampleData[Probabilities] to: paired-end-classifier-E4/probabilities.qza
Saved Visualization to: paired-end-classifier-E4/heatmap.qzv
Saved SampleData[TrueTargets] to: paired-end-classifier-E4/training_targets.qza
Saved SampleData[TrueTargets] to: paired-end-classifier-E4/test_targets.qza

```

## Sample classifier with estimator LinearSVC

Command Prompt - docker run -t -i -v C:/Users/madhu:/data quay.io/qiime2/core:2023.5

```
(qiime2-2023.5) root@92247823ecf6:/data# qiime sample-classifier classify-samples \  
> --i-table paired-table-deblur.qza \  
> --m-metadata-file pair_end_meta.tsv \  
> --m-metadata-column DiseaseStatus \  
> --p-optimize-feature-selection \  
> --p-parameter-tuning \  
> --p-test-size 0.3 \  
> --p-estimator LinearSVC \  
> --p-random-state 123 \  
> --output-dir paired-end-classifier-E5  
Saved SampleEstimator[Classifier] to: paired-end-classifier-E5/sample_estimator.qza  
Saved FeatureData[Importance] to: paired-end-classifier-E5/feature_importance.qza  
Saved SampleData[ClassifierPredictions] to: paired-end-classifier-E5/predictions.qza  
Saved Visualization to: paired-end-classifier-E5/model_summary.qzv  
Saved Visualization to: paired-end-classifier-E5/accuracy_results.qzv  
Saved SampleData[Probabilities] to: paired-end-classifier-E5/probabilities.qza  
Saved Visualization to: paired-end-classifier-E5/heatmap.qzv  
Saved SampleData[TrueTargets] to: paired-end-classifier-E5/training_targets.qza  
Saved SampleData[TrueTargets] to: paired-end-classifier-E5/test_targets.qza
```

Your third section. Change the header and label to something appropriate.

→ view.qiime2.org/visualization/?type=html&src=288ab37b-3656-4b22-bf45-02b390447dd8

qiime2view File: model\_summary.qzv Visualization

### Model parameters

	Parameter setting
est_bootstrap	True
est_ccp_alpha	0.0
est_class_weight	None
est_criterion	entropy
est_max_depth	16
est_max_features	0.1
est_max_leaf_nodes	None
est_max_samples	None

## 4 References

Anon., n.d. *QIIME2 View*. [Online]

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QIIME 2 user documentation, n.d. *Installing QIIME 2*. [Online]

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QIIME2 development team, n.d. *Importing data*. [Online]  
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Available at: [https://scikit-learn.org/stable/supervised\\_learning](https://scikit-learn.org/stable/supervised_learning)