

Microbiome Informatics Workshop 2019

16S rRNA Sequence Analysis: Intro and Advanced Courses

Center for Microbiome Research

Courses in sequencing and analysis of microbiome data held over two days.

Workshop Summary:

This workshop is designed to be a combination of the theory behind processing sequence data and practical applications of current software in studying the microbiome. The first day of the workshop will cover a primer on sequencing technology, information theory, ecological theory, and statistics as applied to microbial communities. The practical portions will focus on 16S sequence analysis through QIIME2. The goal is to provide a comprehensive survey of sequencing applications and hands-on experience with data analysis.

The second day is intended as an advanced analysis course where participants analyze their own data.

Goals:

By the end of the workshop, participants should:

1. Understand the basics of 16S biology and bioinformatics uses
2. Be able to analyze their own 16S data using QIIME2

Dates:

16S basics/intro course: Tuesday, January 29, 2019 (MFRC 3075)

Advanced 16S: Wednesday, January 30, 2019 (TBRC C2195)

Seminar Coordinators:

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Schedule:

Prior to the workshop *all participants* need to have QIIME2 installed on their computers, instructions will be provided in the week prior to the workshop.

Day 1 (16S basics): LOCATION MFRC 3075

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| 9-9:30am | Introductions & Sequencing and information theory <ul style="list-style-type: none">• DNA Extraction, Library Prep and Sample Considerations• Types of Sequencing Platforms• 16S rDNA gene structure and 16S Sequencing• Analysis Platforms• Taxonomic Assignment Methods• State of the Field for Sequencing Microbiomes |
| 9:30-10:30am | QIIME2 background, Basic command line, Importing data, Quality control, & “OTU” picking (practical) <ul style="list-style-type: none">• QIIME2 platform purpose and file/program structure• Using command line to interface with the platform• Importing data to a QIIME2-format• Quality Control of Sequences (PHRED scores and Trimming)• Choosing Operational Taxonomic Units (OTUs) |
| 11-11:15am | Break |
| 11:15-12pm | Statistics, Ecology, what does QIIME tell you <ul style="list-style-type: none">• Alpha diversity<ul style="list-style-type: none">○ What is alpha diversity○ Uses for alpha diversity○ Metrics for alpha diversity○ Interpreting alpha diversity metrics• Beta Diversity<ul style="list-style-type: none">○ What is beta diversity○ Uses for beta diversity○ Metrics for beta diversity○ Interpreting beta diversity metrics |
| 12-1pm | Lunch: on your own |
| 1-2:15pm | Practical: Ecological profiling of samples |
| 2:15-2:30pm | Break |
| 2:30-4pm | How to ask a question/Practical/remaining questions |

Day 2 (Advanced)*: LOCATION TBRC C2195

DATA FORMAT

Day 2 participants are expected to bring their own data. Your data should be in FASTQ formats as CASAVA-ONE-EIGHT, and the sequencing length and type (Paired End or Single) should be known. If your data is not in CASAVA-ONE-EIGHT format, please know the type (FASTA/FASTQ) and formatting of your data.

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|------------|--|
| 9-10am | Introductions & Summary of analysis steps <ul style="list-style-type: none">• Recap of day 1• Where we are now in the analysis pipeline |
| 10-10:15am | Break |
| 10:15-12pm | Work on your own data |
| 12-1pm | Lunch |
| 1-2pm | Background advanced topics <ul style="list-style-type: none">• Balances<ul style="list-style-type: none">○ Relative Abundance vs Actual Read Counts○ Log Trees○ Balances and the resolution they provide○ Statistical Models from Balances○ Step by step walk through• PICRUST2 (predicted functional assignments)<ul style="list-style-type: none">○ Limitations and Uses of PICRUST○ Interpreting PICRUST○ Step-by-step walkthrough |
| 2-4pm | Work on voted option/Continue own data |

*Day 2 schedule is a loose schedule, subject to change based on rate that people work/have problems.

Additional Topics We can Discuss on Day 2

- q2-longitudinal (longitudinal & paired sample comparisons)
- MaAsLin: Multivariate Association with Linear Models