

# Type 1 Diabetes: Mice

## Study on the cage effect

Dea Rynanda Putri, Prof. Ziv Skhedy

January 19, 2017

# Introduction

- ▶ **RNA sequencing** of gut bacteria allows to study the microbiome evolution with and without interventions over time.
- ▶ **Important** to understand the factors that might potentially affect the observed OTUs in the gut.
- ▶ **Constraint:** OTU **not observed** at a certain time point in a subject (mouse) might actually have **positive abundance**.
- ▶ Although zero abundance not necessarily means OTU was not present, it is important to also understand the factors (other than interventions under study) that might explain the "appearance" of an OTU between two consecutive time points.

# Objective

- ▶ To answer whether "transmission" of bacteria between mice sharing the same cage is one of the factors that may influence the appearance of an OTU in a mouse.

# Data

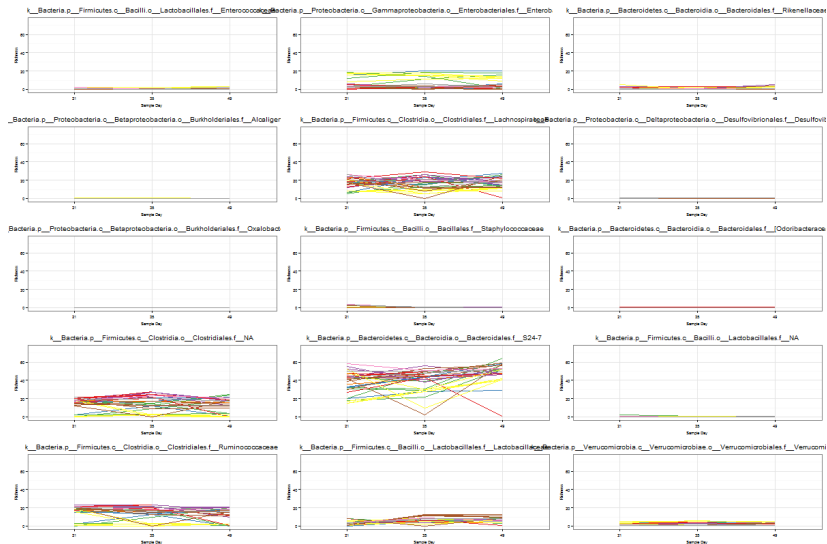
- ▶ OTU abundance measured in mice at 3 bi-monthly time points (Day 21, 35, and 49)
- ▶ Two groups: **control** (without antibiotic treatment) and **antibiotics** group. Only **control** group is used in this analysis

# OTU Richness

Time Point	Richness	Min.	Q1	Median	Mean	Q3	Max
Day 21	Absolute	48.0	75.0	100.0	98.9	120.0	167.0
	Chao1	53.6	91.2	123.3	120.8	140.1	228.2
Day 35	Absolute	4.0	86.8	109.5	105.2	124.0	157.0
	Chao1	4.0	109.6	127.7	126.8	147.5	206.5
Day 49	Absolute	5.0	94.8	121.0	114.0	132.0	169.0
	Chao1	8.0	121.0	140.4	136.2	156.7	190.0
<b>Overall</b>	<b>Absolute</b>	<b>4.0</b>	<b>85.0</b>	<b>110.0</b>	<b>106.0</b>	<b>127.0</b>	<b>169.0</b>
	<b>Chao1</b>	<b>4.0</b>	<b>104.7</b>	<b>130.1</b>	<b>127.9</b>	<b>150.1</b>	<b>228.2</b>

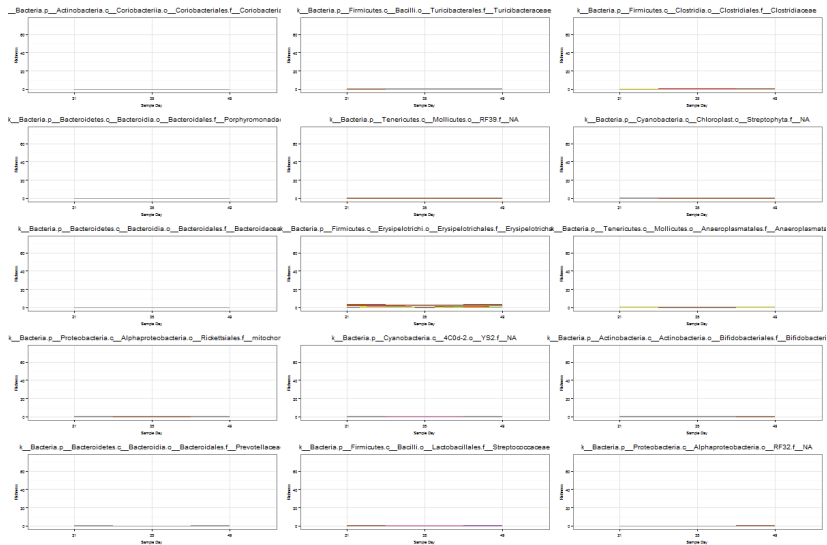
# Richness on Family Level

Control group only



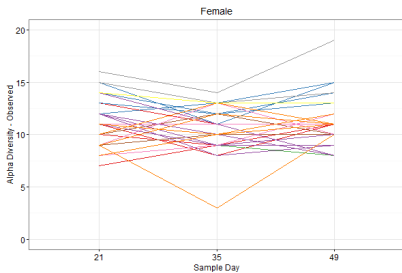
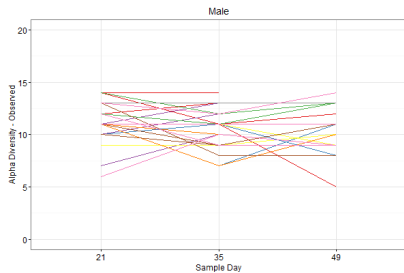
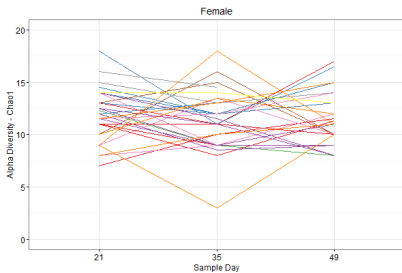
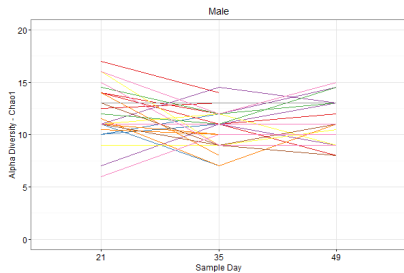
# Richness on Family Level (cont'd)

Control group only



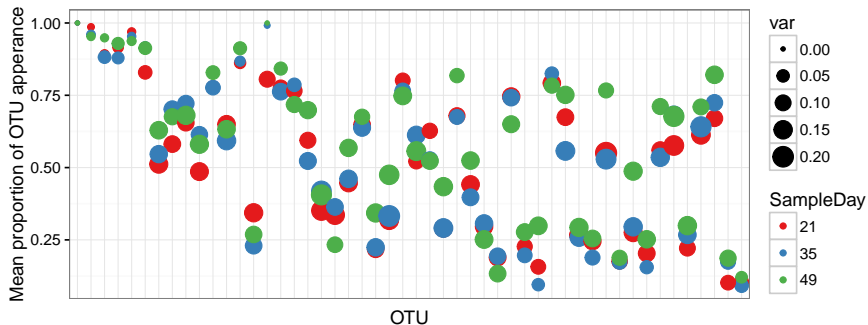
# Alpha diversity on family level

Control group only





# Proportion of OTU presence in a cage by OTU



# Fisher's exact test

Group	Day	Number of mice	Number of OTUs	Dependency between mice in same cage and mice with OTU in common, by OTU			
				Mean number of common OTUs for mice in same cage	Mean number of common OTUs for mice in different cages	Number of OTUs with significant p-value Fisher exact test	
						Not adjusted for multiplicity	Adjusted for multiplicity (Hochberg)
Control	Day 21	88	348	66	42	191 (55%)	70 (20%)
	Day 35	84	348	78	50	187 (54%)	77 (22%)
	Day 49	87	348	87	56	214 (61%)	72 (21%)

# Linear mixed effect models

## Model comparison:

$$M_0 : g(\pi) = \beta_0 + \beta_t \text{Time}$$

$$M_1 : g(\pi) = \beta_0 + \beta_t \text{Time} + a_k$$

$$M_2 : g(\pi) = \beta_0 + \beta_t \text{Time} + a_k + b_i$$

where :

- ▶  $g(\pi)$  is 0/1 value for OTU appearance (1: OTU appears)
- ▶  $\beta_t$  is the mean effect of time
- ▶  $a_k$  is a random effect of **cage**
- ▶  $b_i$  is a random effect of **mouse**

No OTU is having a significant random effect on neither Cage nor Mouse.

# Linear mixed effect models

## Model comparison:

$$M_0 : g(\pi) = \beta_0$$

$$M_1 : g(\pi) = \beta_0 + a_k$$

where :

- ▶  $g(\pi)$  is 0/1 value for OTU appearance (1: OTU appears)
- ▶  $\beta_t$  is the mean effect of time
- ▶  $a_k$  is a random effect of **cage**

Same case as before, no OTU is having a significant random effect on neither Cage.

Thank you