ASSOCIATION OF GUT MICROBIOME WITH COGNITIVE IMPAIRMENT IN PUERTO RICANS

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ABSTRACT

Purpose: Alzheimer's disease (AD) is the most common type of dementia, a debilitating disorder that could result in complete loss of mental functions and death. In Puerto Rico, AD is the fourth leading cause of death, while in the United States is the sixth. Evidence suggests that the gut microbiota plays a role in the pathophysiology of AD through neuroinflammation and amyloid deposition at brain. *Our objective: study the fecal microbiota composition and diversity of Puerto Ricans with AD compared to cognitive-intact controls and associate the microbiota with cognitive impairment.*

Methods: We recruited 53 participants, 28 with AD and 25 controls, who underwent clinical and cognitive assessments (MoCA/CDR). Genomic DNA extractions performed on collected fecal samples. NextGen Illumina MiSeq was used to sequence 16S rRNA genes (V4 region) and analyzed with standard pipelines for microbiome species.

Results: Preliminary analyses showed no statistically significant differences between AD and controls in bacterial diversity and richness. However, AD participants showed an abundance of *Euryachaeota,* while controls had higher levels of *Bacteroidetes*. We found significant differences in alpha diversity with cognitive decline and a reduction of *Roseburia* -a known butyrate producer with protective and anti-inflammatory properties- in participants with severe cognitive impairment.

Conclusion: First study in Puerto Rico comparing a neuro-degenerative disease common in the aging population with the gut microbiota. The study of the Gut-Brain-Axis may open the possibility for preventive microbiota-based therapies and strategies for a healthy microbiome resulting in better outcomes for our patients with and without AD.

METHODOLOGY

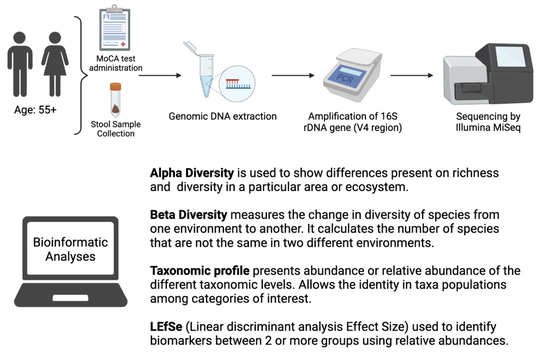

Figure 1. Overview of the collection of 53 samples, laboratory protocols, and analyses. We analyzed a total of 1,713,977 reads with a rarefaction level of 3,700 per sample

RESULTS

	Alzheimer Patient n = 28	Control n = 25
Sex		
Female	19	19
Male	9	6
Age Range		
55 - 65	1	7
66 - 76	10	15
Greater than 77	17	3
MoCA		
Normal (26 – 30)	0	3
Mild (18 – 25)	8	17
Moderate (10 – 17)	7	5
Severe (≤ 9)	13	0

Table 1. Selected clinical variables of study participants

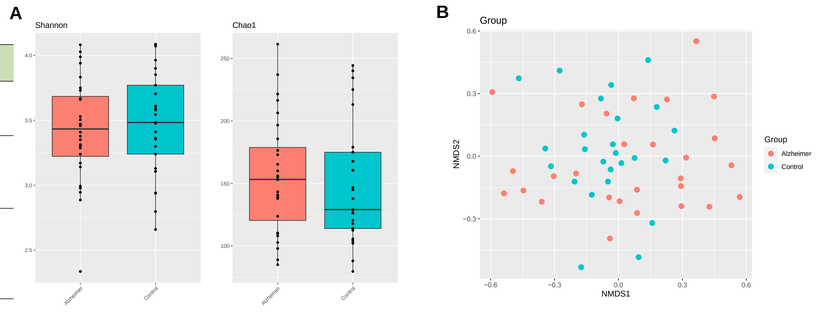

Figure 2. Microbial alpha diversity (Shannon and Chao1) (Pairwise NS) (A) and beta diversity analyses (NMDS) between groups (B) .

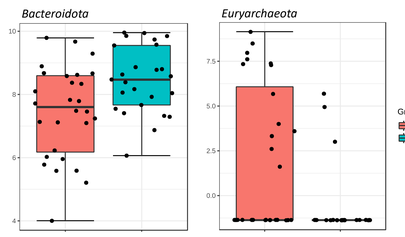

Figure 3. Selected significant phyla-level taxa distinguishing between groups.

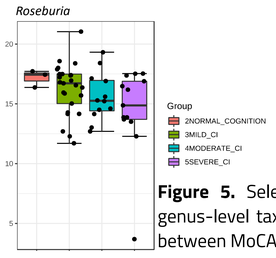

Figure 5. Selected significant genus-level taxa distinguishing between MoCA classes.

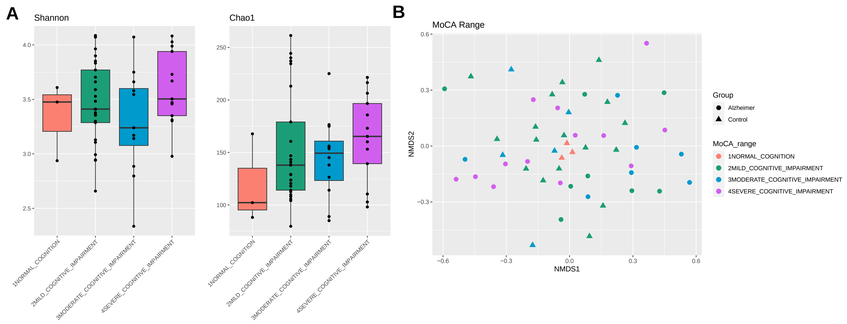

Figure 4. Microbial alpha diversity (Shannon and Chao1) (Pairwise NS) (A) and beta diversity analyses (NMDS) (B) between MoCA classes.

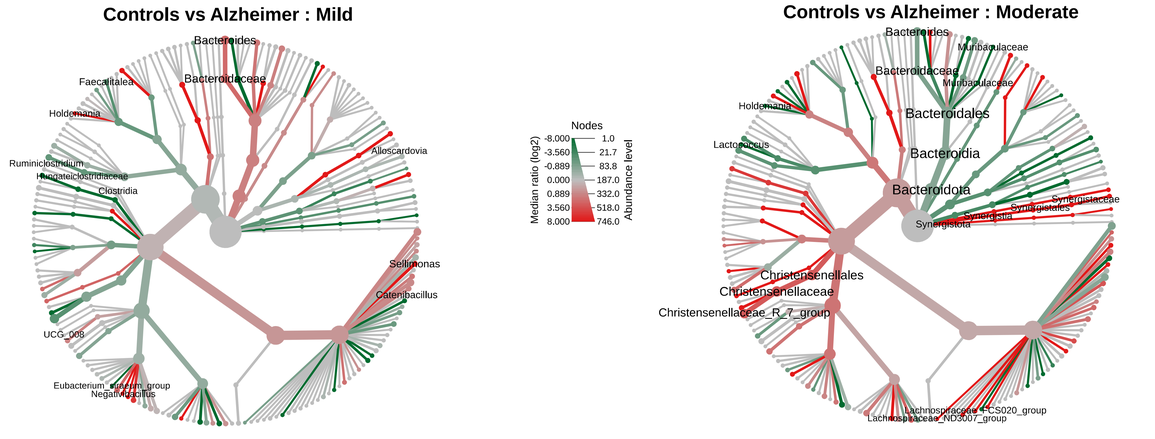

Figure 6. Heat tree analysis depicting alterations in microbiome composition between controls and AD with different levels of cognitive impairment (Mild < Moderate). A red branch indicates an increase in AD while green ones show an increase in controls or decrease in AD (p-value < 0.05).

DISCUSION

1. Are there any differences in the gut microbiome of AD patients?

- Given that this preliminary data comes from a small cohort of only 53 participants, Alpha diversity analysis found no significant differences nor richness between AD patients and controls (p value > 0.05) (Figure 2).
- Linear discriminant analysis Effect Size (LEfSe) indicates there is a significant dominance of *Euryarchaeota* in Alzheimer's disease patients while *Bacteroidota* dominated in the control group (Figure 3).
- *Euryarchaeota* taxa is known to produce intramembrane aspartyl proteases capable of degrading signal peptides which could potentially aggregate and induce cytotoxicity.
- *Bacteroidota* are anti-inflammatory taxa and butyrate producers.

2. Are there any differences in the gut microbiome due to cognitive capacity of participants?

- Alpha diversity analysis showed significant difference between microbial diversity according to the Montreal Cognitive Assessment (MoCA) test classes (p value < 0.05) (Figure 4A).
- Diversity analyses (Fisher and Chao1) revealed there was a decrease in gut microbial diversity, as cognitive condition declined (severe cognitive impairment) (Figure 4A).
- Beta diversity analysis revealed no significant differences in community composition due to cognitive condition (p > 0.05) (Figure 4B).
- A relative decline was observed in anti-inflammatory *Roseburia* (NS) (Figure 5).
- Heat tree analyses confirms *Bacteroides* in higher abundance in controls (Figure 6).

CONCLUSION

- This is a preliminary analyses on a subset of participants. Although there are no significant changes in the gut microbiome structure of Alzheimer's patients compared to controls, there were differences in composition at the phyla and genus levels.
- We found slight differences in composition including an abundance of *Euryachaeota* in AD while controls had higher levels of *Bacteroidota*.
- This to our knowledge, is the first study in Puerto Rico, combining a chronic disease in an aging population with high-resolution gut microbiome analyses. This may open the possibility for preventive microbiota-based therapies however a bigger sample size is required.

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