



The Impact of Fecal Sample Preservation Conditions on the Identification of Gut Microbial Markers of Peripheral Artery Disease

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Introduction

- Proper handling and preservation of fecal samples is critical to accurate profiling of the microbial community using multi-omics approaches
- Home sample collection can be challenging for participants and affect compliance and feasibility
- Sample collection and preservation methods can affect microbial community composition and diversity
- The effect of different sample preservation methods on gut microbial profiling in patients with peripheral artery disease (PAD), which is caused by atherosclerosis and is known to affect the gut microbiome, is poorly understood

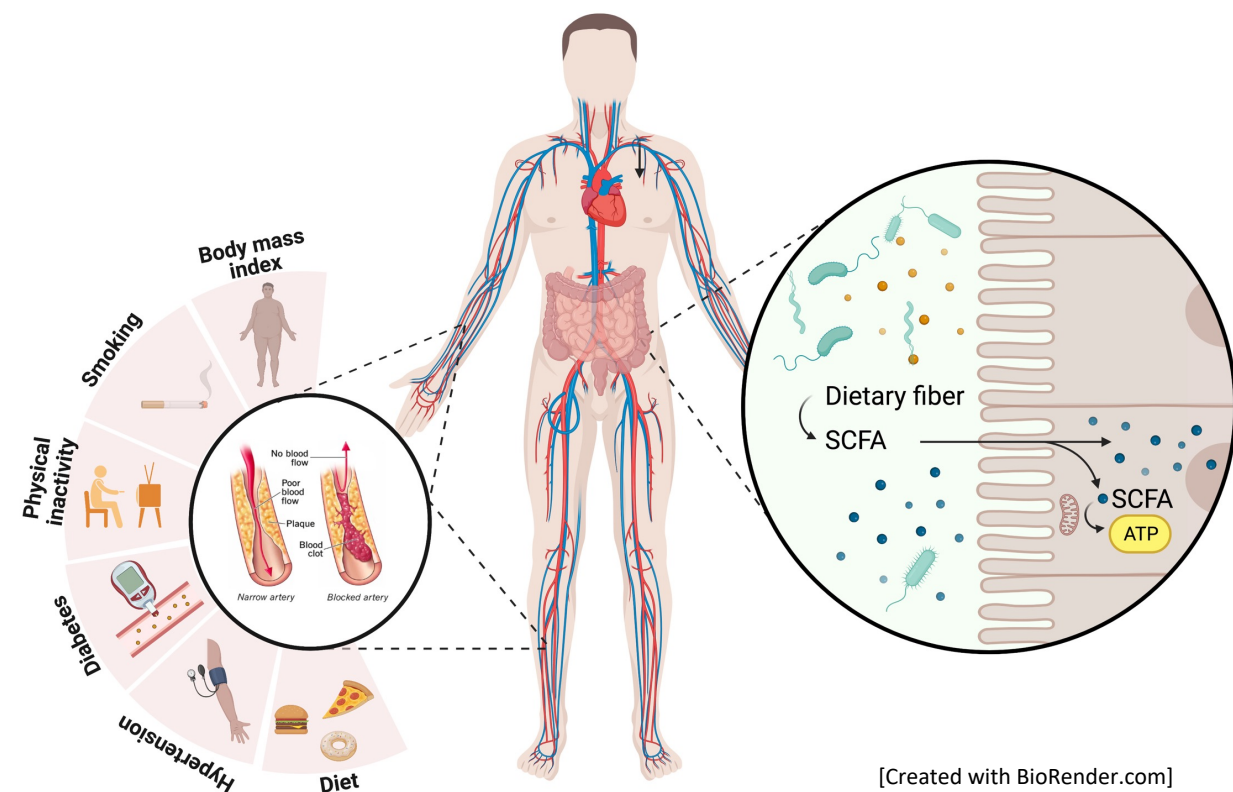


Figure 1. Peripheral artery disease (PAD) caused by atherosclerotic occlusions leads to pain and disability.

Objectives

- To identify gut microbial features associated with PAD
- To evaluate the effects of 3 preservation conditions on PAD and non-PAD fecal microbial community profiles: immediately frozen (FR), Cary-Blair media (CB) and OMNIgene-Gut (GT)

Methods

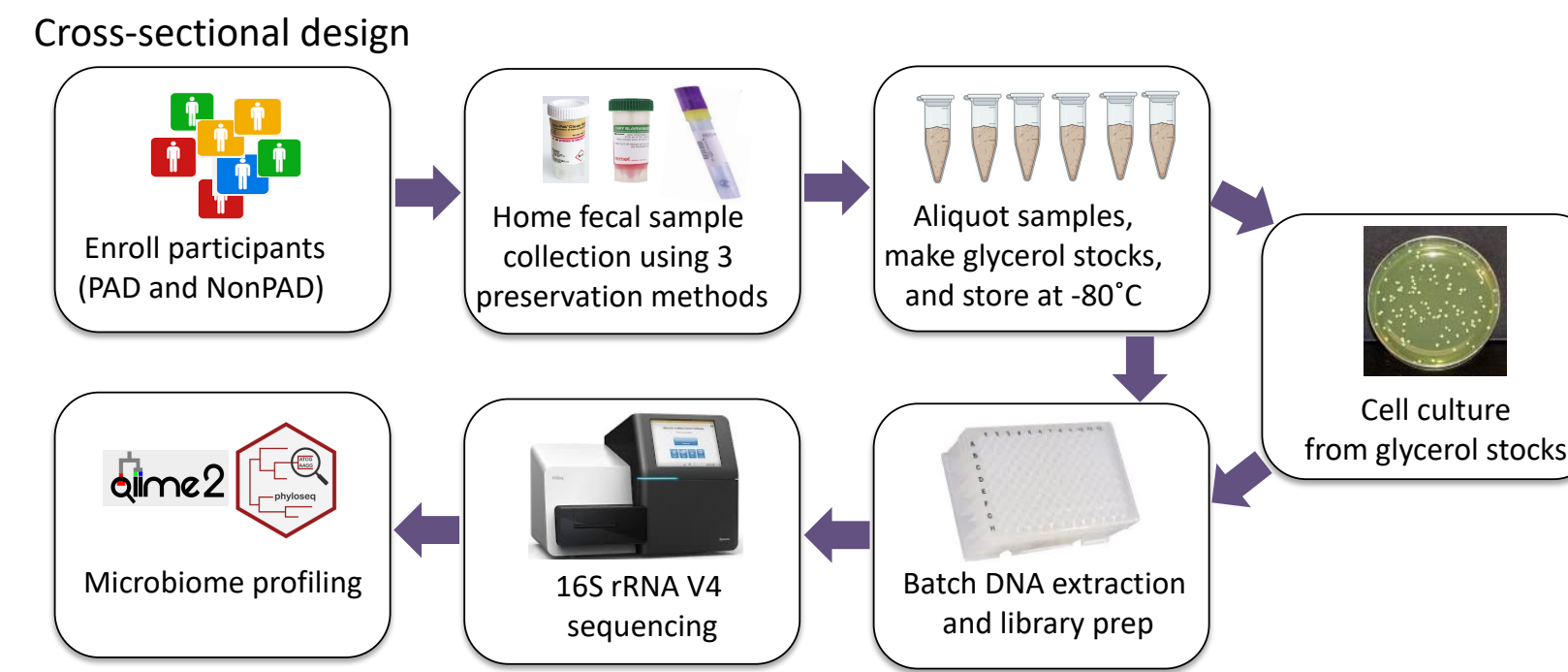


Figure 2. Schematic of overall workflow.

Results

1. Preservation conditions alter microbial composition.

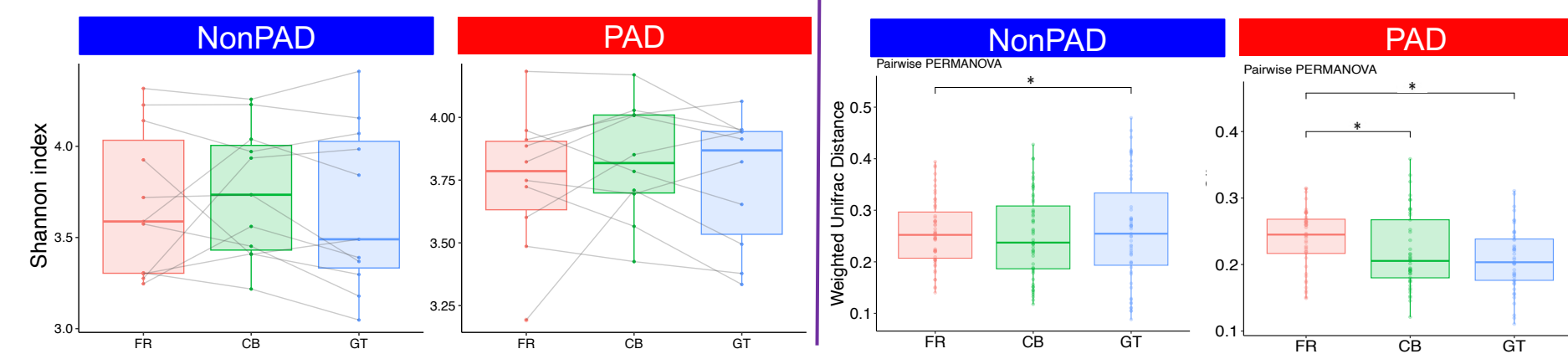


Figure 3. Overall effect of preservation conditions on alpha (Shannon index; left) and beta diversity (Weighted UniFrac matrix; right). Each dot represents an individual. [n=19 (NonPAD), 14 (PAD)]

2. Relative abundances of phyla vary among preservation methods.

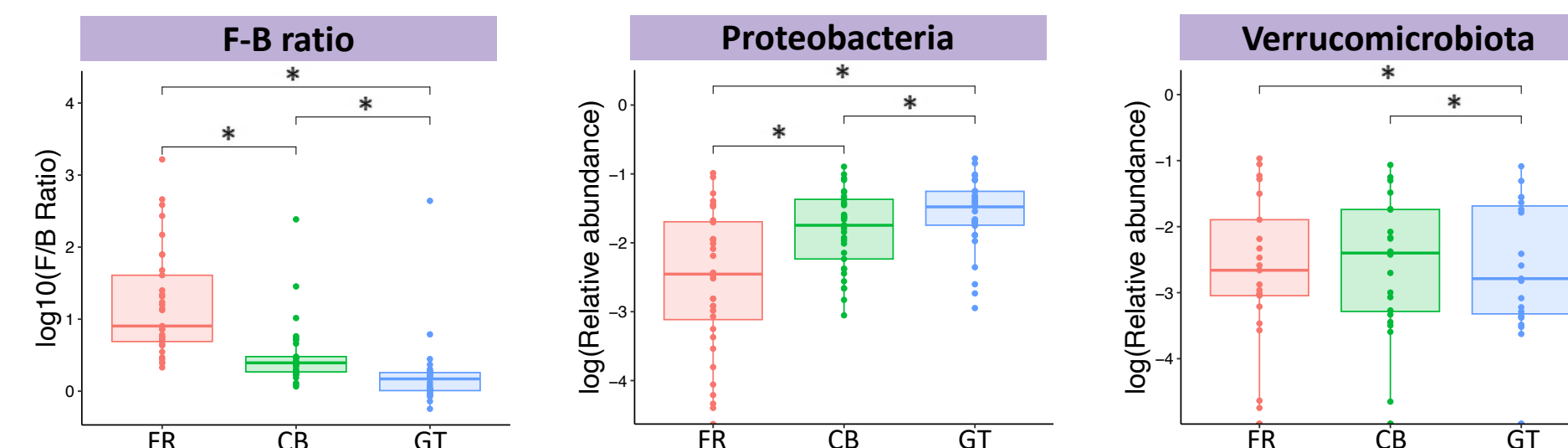


Figure 4. Relative abundance of taxa at phylum level. P values represent significant differences using Wilcoxon-sum rank test. F-B, Firmicutes-Bacteroidetes

3. Effects of storage conditions on differentially abundant taxa.

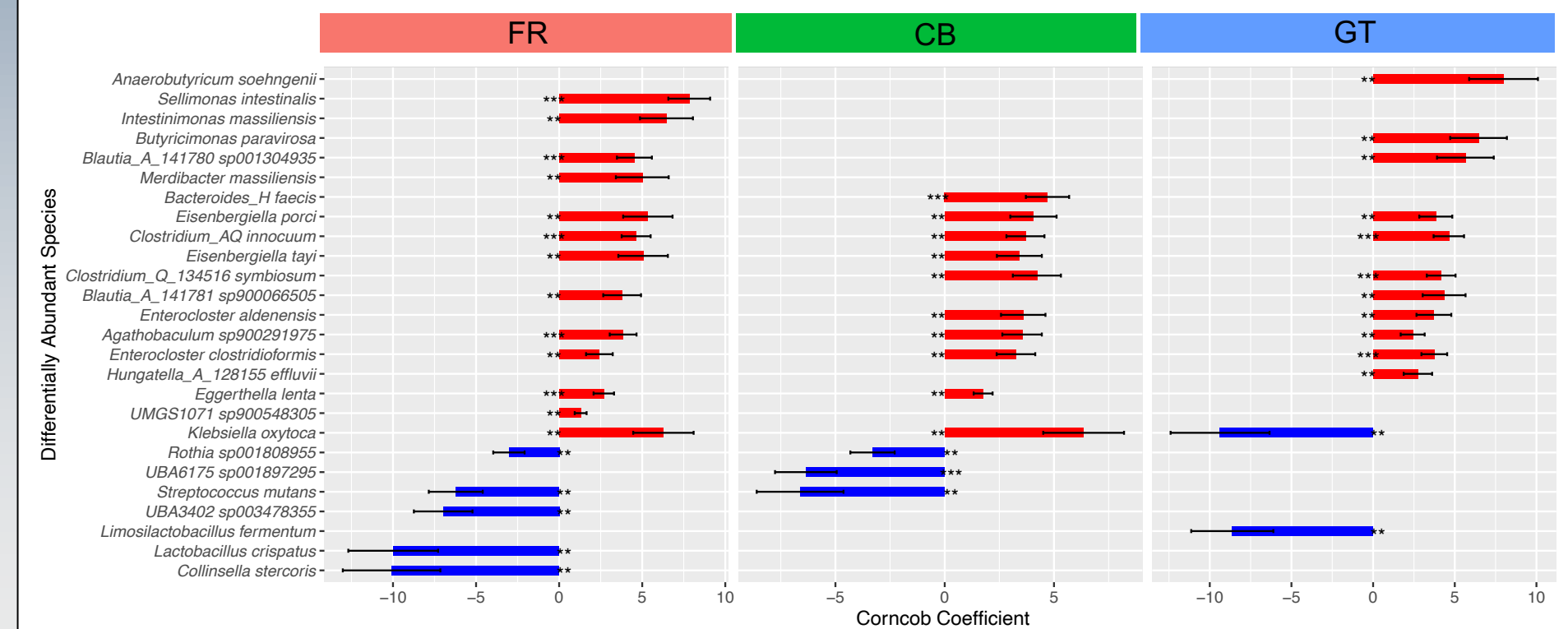


Figure 5. Differentially abundant taxa between PAD (red) and Non-PAD (blue) calculated using three preservation methods after adjustment for age.

4. Effects of preservation conditions on differentially enriched functions.

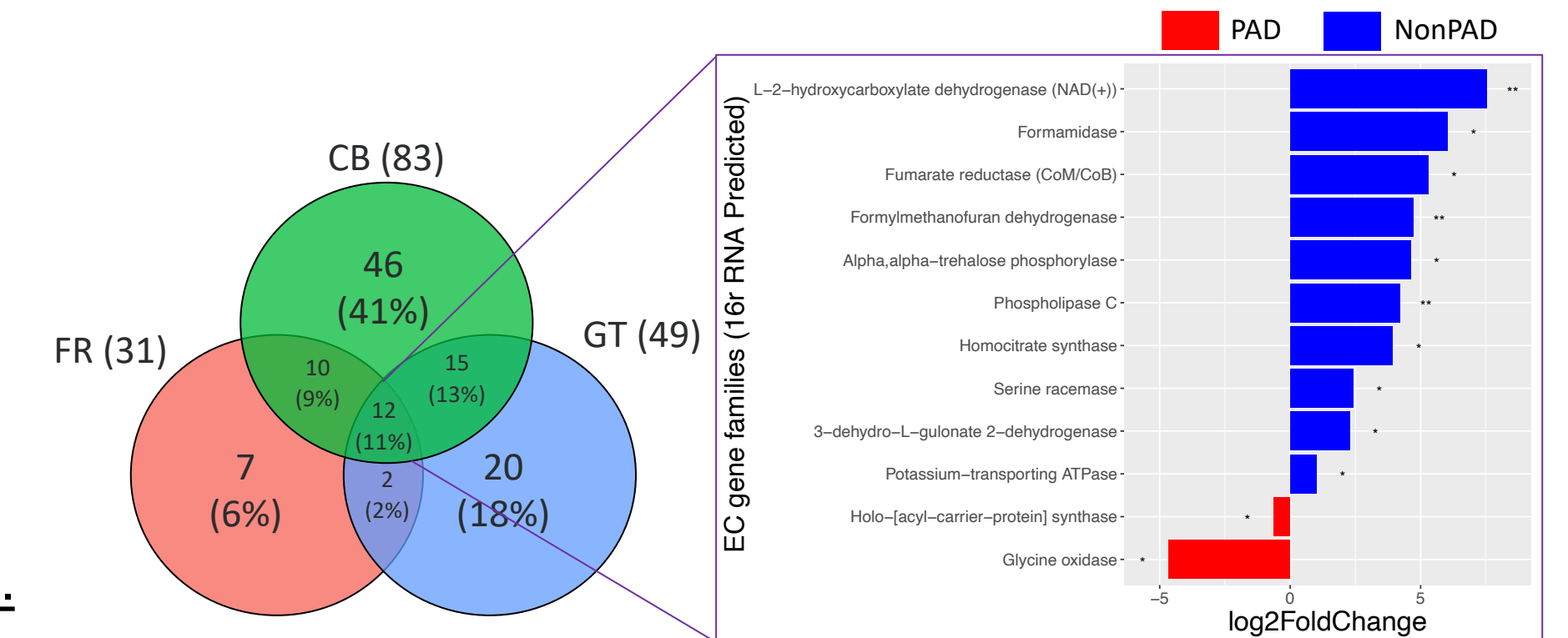


Figure 6. Differentially enriched predicted functions between PAD and non-PAD controls that are shared among three preservation methods.

Conclusions

Our study demonstrates the feasibility of home fecal sample collection for gut microbial profiling of patients with PAD. Storage conditions alter the overall microbiome composition and impact the outcome of comparative studies between PAD and non-PAD communities.