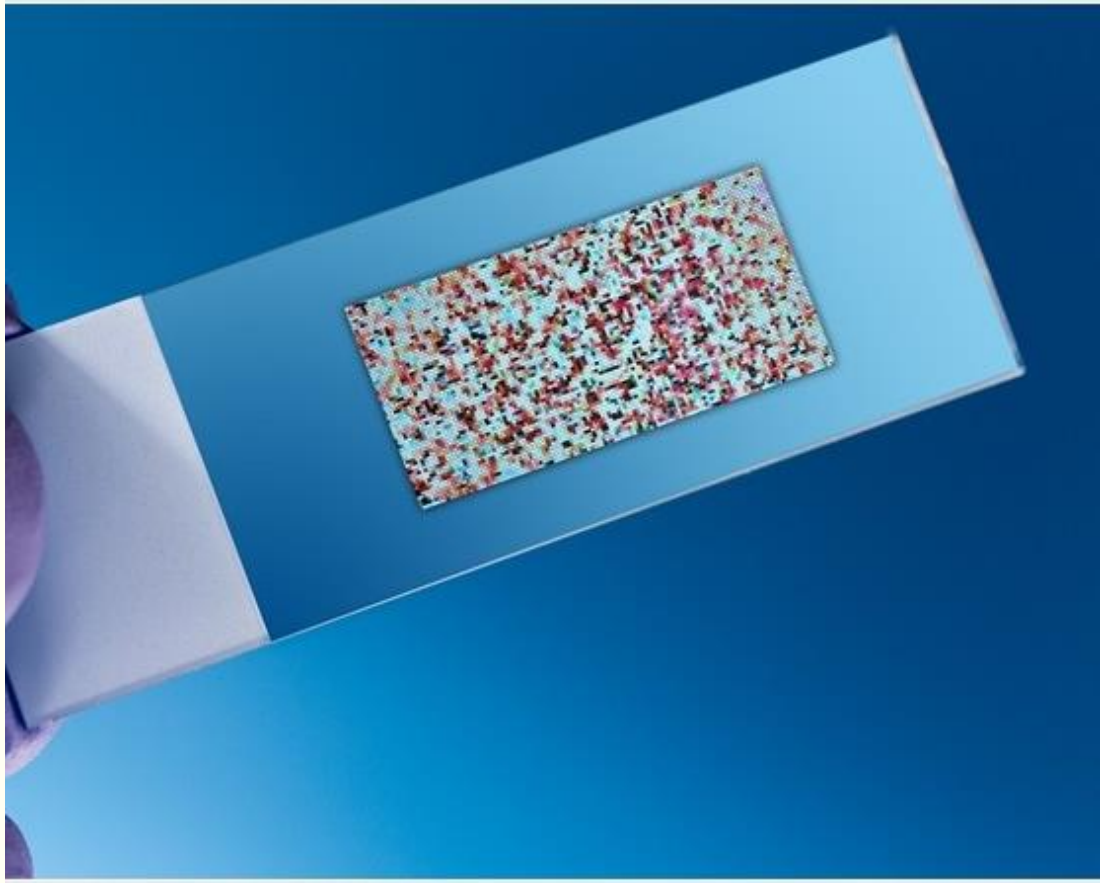
CHANGES IN GUT MICROBIOTA ABUNDANCE WITH DIETARY INTERVENTION

By Daniel Broderick



Content

- What is the microbiome and why is it important?
- The study
- The data
- Hypotheses
- Analysis
- Discussion

The microbiome

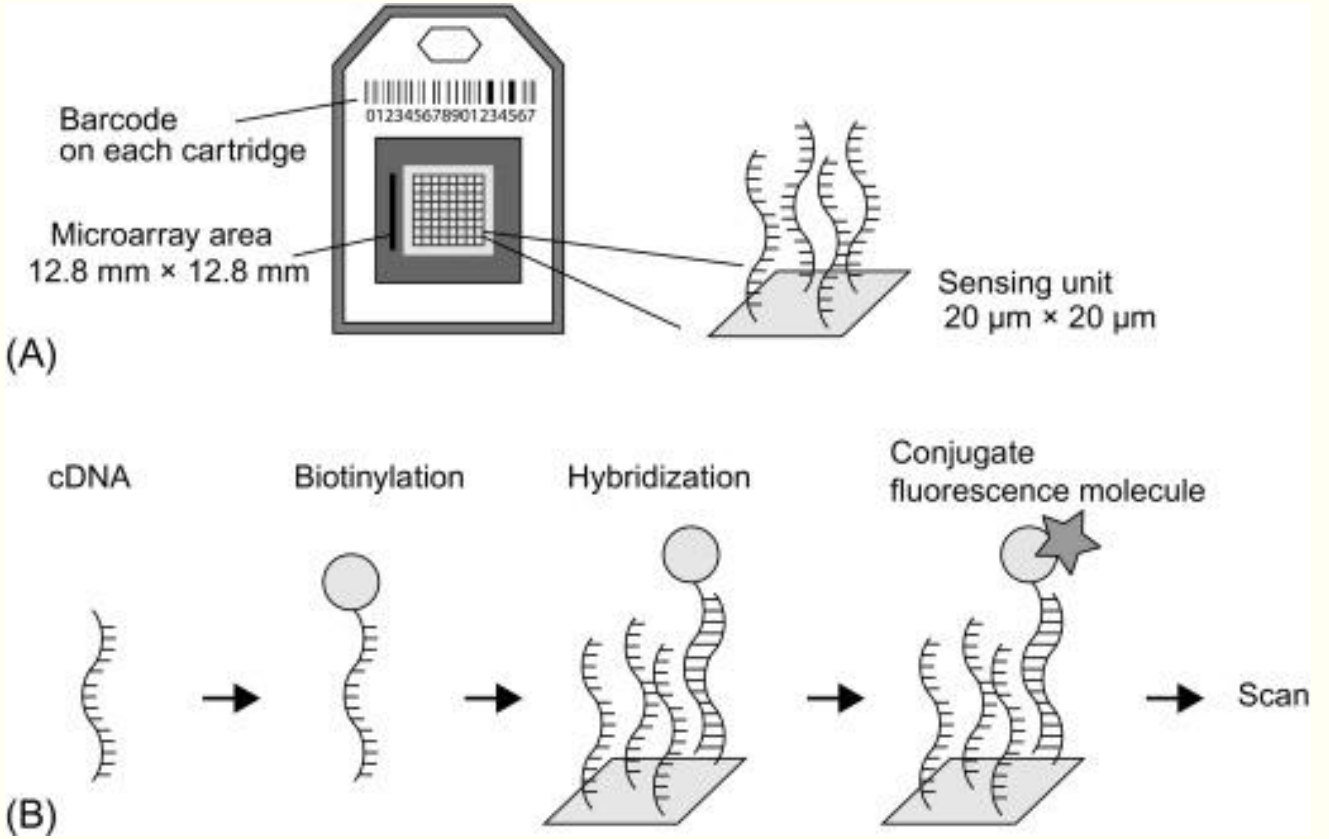
- Human microbiome project started in 2008
- Our guts have ~20,000 orthologous groups of bacteria, less than 5,000 of which are known, even fewer have their functions characterized in the literature [1]
- Responsible for digesting fiber and indigestible short-chain carbohydrates [2]
  - FODMAPs - low–fermentable oligo-, di-, and monosaccharide and polyol
  - Implicated in in food intolerance and IBS
  - Breakdown by gut bacteria can produce gas, bloating, etc.
- Gluten produces an inflammatory response in the gut [3]
- Microbiome composition is shaped by diet, but also by mode of delivery (vaginal or caesarean), lifestyle factors, and antibiotic use (especially during infancy) [4]
- May also be implicated in autoimmunity, mental illness, Alzheimer's, Crohn's, etc.

The study

- n = 78
  - Obese individuals
  - Lasts 12 weeks
- 3 dietary interventions
  - Study A (n = 28) and B (n=24) eats gluten-containing foods; whole grains or low-fiber, refined wheat bread; from Finland
  - Study C (n = 13) consumes 8g inulin and 8g oligofructose daily; from Belgian
  - Study D (n=13) has a one-week run-in diet followed by three weeks each of a resistant-starch-enriched, non-starch-polysaccharide-enriched, and low carb/fat, high pro; from Britain
- Collect fecal samples pre- and post-intervention

The data

- Continuous data representing microarray signal intensities
- Log-transformed and min/max normalized
- Used the 16S rRNA gene to categorize microbiota [4]
- 1037 species or phylotypes
- 130 genus-like group



Hypotheses

H1: Increased gluten intake will affect the relative number of glutenase-producing microbiota

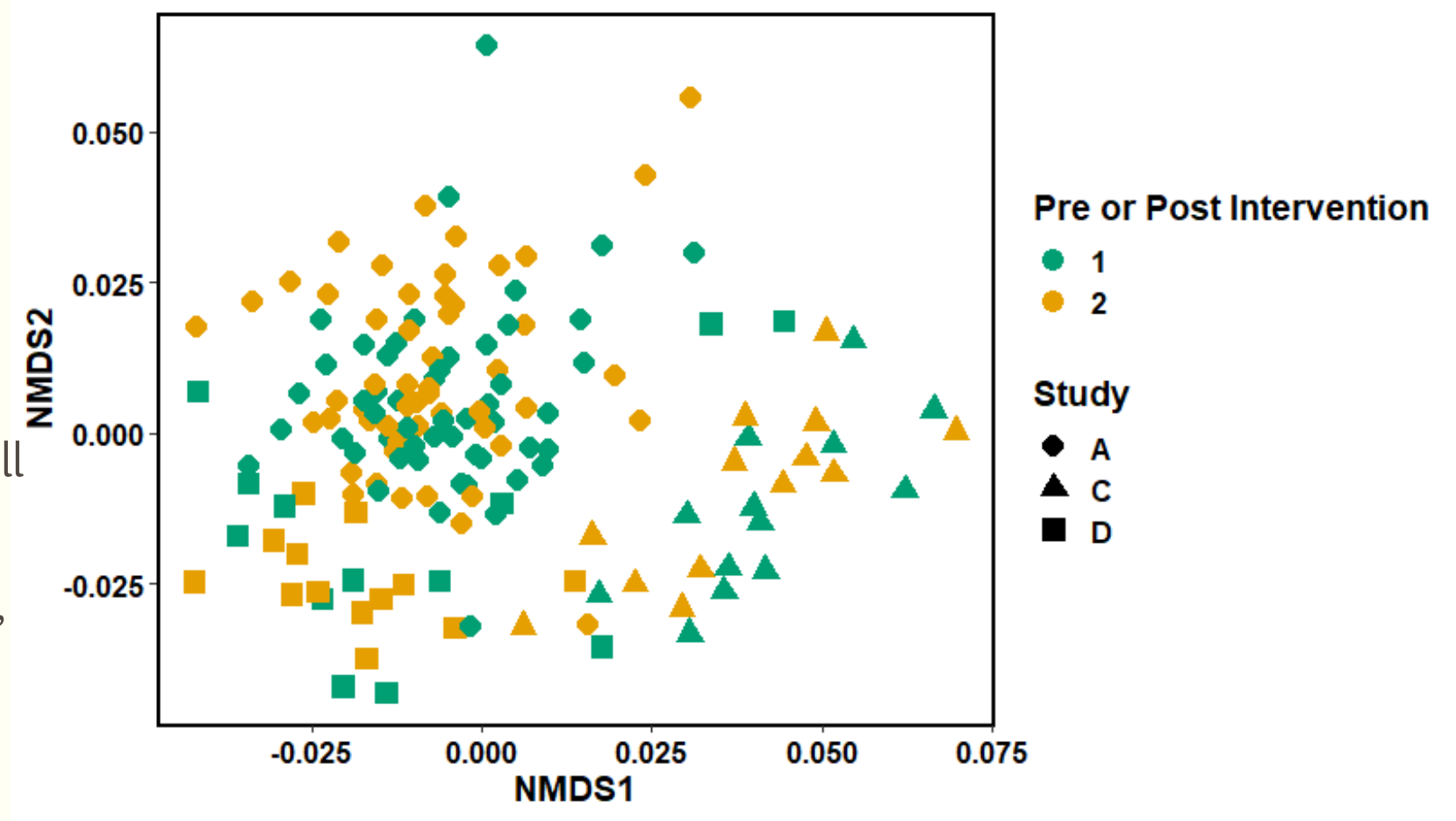
- Microbes may mediate gluten sensitivity
  - *Lactobacilli*
  - *Aspergillus niger*
  - *Flavobacterium meningosepticum*
  - *Myxococcus xanthus*
  - *Sphingomonas capsulata*

H2: Oligofructose consumption will affect gut microbiota composition differently than wheat or rye bread consumption.

- Oligofructose and inulin are both fructans, a group of specific carbohydrates found in wheat and rye.
  - can cause digestive issues for some

Non-metric Multi-dimensional Scaling

- Iterative algorithm
- Rank-based
  - Non-parametric
- uses a distance matrix (Bray-Curtis) as input
- BC considers species presence/absence, as well as abundance
- The closer two points are, the more similar their microbiota composition



ANOSIM Test

- H_0: there is no difference between the microbial communities of the groups
- Used 9999 permutations
- And Bray–Curtis dissimilarity
  - used to quantify the compositional dissimilarity between two different sites
- R(0-1): compares mean ranked dissimilarity between groups to mean ranked dissimilarity within groups.
  - 1.0 = most dissimilar
  - 0 = even distribution of high and low ranks within and between groups

	ANOSIM statistic R	P - value
Pre/post	0.0023	0.3027
Pre - AB/C/D	0.6714	<.0001
Post – AB/C/D	0.6351	<.0001
Pre – AB/C	0.7996	<.0001
Post – AB/C	0.7566	<.0001

Indicator Species Analysis

- Identifies microbial species that are found more often in one group compared to another

- Eubacterium's are associated with opportunistic infections [6]
  - minor clinical importance

Genera Pre-intervention	Stat value	P-value
Roseburia intestinalis	0.222	0.0068
Aeromonas	0.217	0.0057
Micrococcaceae	0.198	0.0149
Novosphingobium	0.177	0.0314
Mitsuokella multiacida	0.177	0.0275
Clostridium leptum	0.163	0.0422
Akkermansia	0.160	0.0474
Haemophilus	0.157	0.0491
Genera Post-intervention		
Eubacterium cylindroides	0.161	0.0436

Discussion

- Pre-intervention groups do not cluster together
- General decrease in diversity
- Proof of concept
  - Diet can alter the microbiome
- Criticisms
  - Did not distinguish the individuals in study A from those in study B
  - Inconsistent data recording; pre/post measurements were not clearly indicated for study D
  - Did not distinguish male vs female subjects
  - No supplementary materials, only explanation for the dataset was a short description on data dryad
  - Non-obese and obese controls were used in their analysis but not included in the dataset
  - Study D only had 1 pre and 1 post measurement
  - Cholesterol, insulin resistance, and inflammation markers were measured but not included
  - Samples were processed differently

References

1. Morgan, X. C. et al. Dysfunction of the intestinal microbiome in inflammatory bowel disease and treatment. Genome Biology 13, R79 (2012).

2. Hill, P., Muir, J. G. & Gibson, P. R. Controversies and Recent Developments of the Low-FODMAP Diet. Gastroenterol Hepatol (N Y) 13, 36–45 (2017).

3. Drago, S. et al. Gliadin, zonulin and gut permeability: Effects on celiac and non-celiac intestinal mucosa and intestinal cell lines. Scandinavian Journal of Gastroenterology 41, 408–419 (2006).

4. Jandhyala, S. M. et al. Role of the normal gut microbiota. World J Gastroenterol 21, 8787–8803 (2015).

5. Rajilić-Stojanović, M. et al. Development and application of the human intestinal tract chip, a phylogenetic microarray: analysis of universally conserved phylotypes in the abundant microbiota of young and elderly adults. Environ Microbiol 11, 1736–1751 (2009).

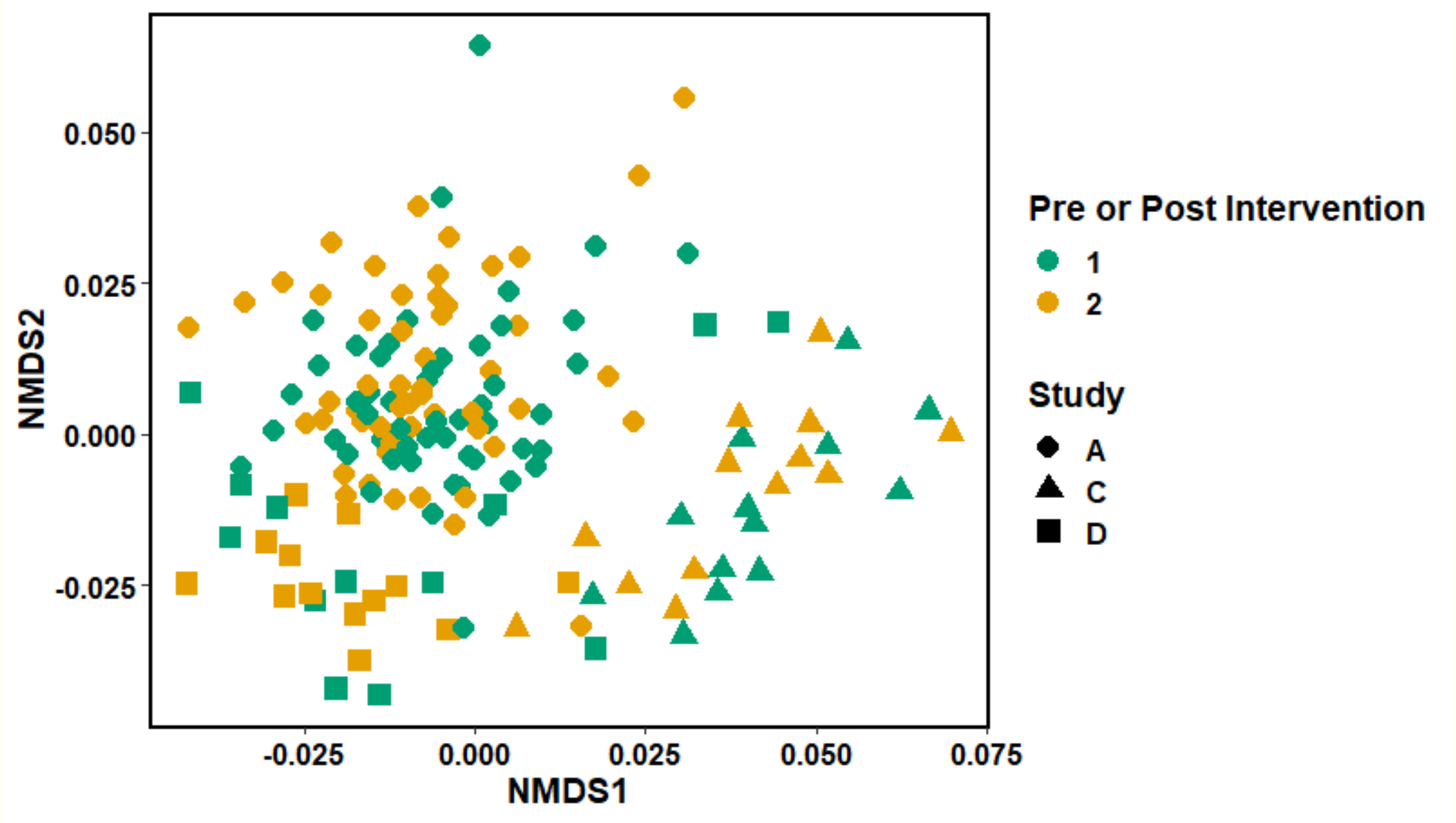
6. Eubacterium - an overview | ScienceDirect Topics. https://www.sciencedirect.com/topics/medicine-and-dentistry/eubacterium.

Image 2: Microarrays - an overview | ScienceDirect Topics.

Study: Korpela, K. et al. Gut Microbiota Signatures Predict Host and Microbiota Responses to Dietary Interventions in Obese Individuals. PLOS ONE 9, e90702 (2014).

Dataset: https://datadryad.org/stash/dataset/doi:10.5061/dryad.bv4k7

Questions?


0.050
0.025
0.000
-0.025
NMDS2
-0.025
0.000
0.025
0.050
0.075
NMDS1
Pre or Post Intervention
1
2
Study
A
C
D