The relationship between alpha-defensin 1 and gut microbiome composition in a population of Bhutanese refugee adults with a high prevalence of type 2 diabetes

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INTRODUCTION

Alpha-Defensin 1

- Alpha-defensins are **antimicrobial peptides** released by neutrophils in response to inflammation (Figure 1)
  - Target bacterial cell wall, causing bacterial cell lysis and neutralization of bacterial toxins
- There are 2 categories of alpha-defensins:
  - Human neutrophil peptides (HNPs): alpha-defensins 1-4
  - Human enteric defensins (HDs): alpha-defensins 5 and 6
- Alpha-defensins have been implicated in **glucose homeostasis**
- **Alpha-defensin 1 is the focus of this study as it can be quantified in plasma samples**

Interleukin 6 (IL-6)

- Interleukin 6 (IL-6) is a **pro-inflammatory cytokin**e that is secreted in response to increased lipopolysaccharide in the bloodstream
- Induces the production of acute phase proteins and is **associated with inflammation**

Human Gut Microbiome

- Gut microbiome **richness** and **diversity** are **inversely** associated with **diabetes**

Study Population

- Bhutanese refugee adults who reside in New Hampshire
  - Refugee populations experience an increased risk for developing metabolic diseases due to the dietary and lifestyle changes that are caused by immigration to the United States

OBJECTIVE

- To determine if the relationship between alpha-defensin 1 and gut microbiome diversity is affected by type 2 diabetes (T2D) status as well as determine if IL-6 has any correlation with gut microbiome richness and diversity
  - **Previous findings**: IL-6 was previously found to have a significant positive correlation with alpha-defensin 1

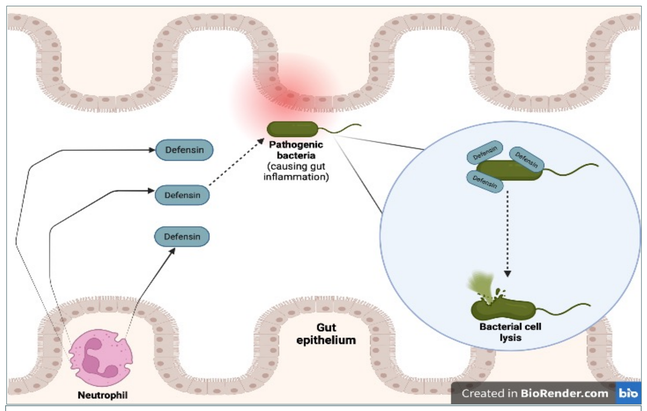


Figure 1. Antimicrobial function of alpha-defensin 1

METHODS

- Plasma and fecal samples were previously obtained from a convenience sample of New Hampshire Bhutanese refugee adults (N = 50) in a previous study led by Dr. Sherman Bigornia
- T2D status was self-reported by participants and data was grouped according to T2D status
- Immunoassays were conducted plasma samples using the MESO Quickplex SQ120 to quantify biomarkers of interest
- Fecal samples were analyzed via shotgun sequencing to obtain gut microbiome richness and diversity measures
- R Studio was used to visualize results and analyze them using Spearman correlations and Wilcoxon test
- Excel was used to create tables and graphs to visualize results

RESULTS

Table 1. Population Characteristics (stratified by diabetic status).

	All				No T2D				T2D				Wilcoxon Test
	n	Median	Min	Max	n	Median	Min	Max	n	Median	Min	Max	p-value
Age	50	18	49.5	72	29	45	18	71	21	58	31	72	**0.01**
Sex													0.38
Male	9 (18%)				4 (14%)				5 (24%)				
Female	41 (82%)				25 (86%)				16 (76%)				
Alpha-Defensin 1	50	113338	18289	1050346	29	129070	18289	1050346	21	81456	25663	476689	0.10
BMI	50	27.1	20	38.9	29	27.1	20	38.9	21	27	20.1	35.9	0.89
Hba1c	50	5.9	4.9	10.4	29	5.6	4.9	6.1	21	6.9	5.6	10.4	**<0.0001**
IL-6	50	2	0	6.9	29	2	0	6.8	21	1.9	0	6.9	0.74
LBP	50	419705	1250758	8394710	29	4023756	1524137	5860338	21	4616820	1250758	8394710	0.40
Observed Richness	50	140	35	252	29	154	35	237	21	128	75	252	**0.05**
Shannon	50	4.9	3.5	5.6	29	5	3.5	5.4	21	4.8	4.2	5.6	**0.05**

Table 2. Relationship between alpha-defensin 1 and fecal microbial richness and diversity. Spearman correlation analysis.

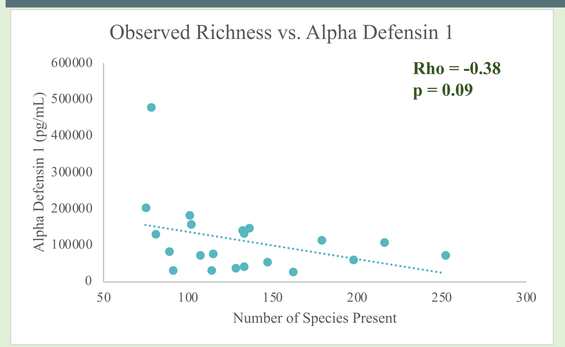
	All			No T2D			T2D		
	n	Rho	p-value	n	Rho	p-value	n	Rho	p-value
Alpha-Defensin 1 vs.									
Observed Richness	50	0.06	0.70	29	0.21	0.26	21	-0.38	**0.09**
Shannon	50	0.07	0.65	29	0.19	0.33	21	-0.39	**0.08**
InvSimpson	50	0.08	0.60	29	0.23	0.24	21	-0.34	0.13
Fisher	50	0.07	0.63	29	0.15	0.43	21	-0.39	**0.08**

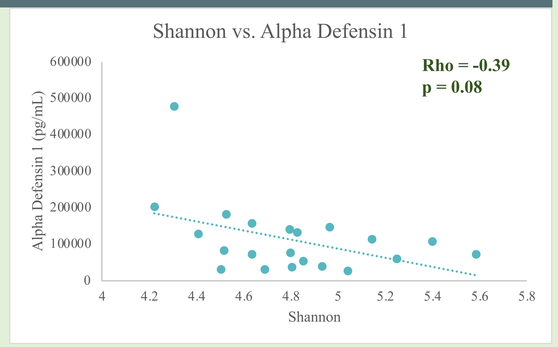
Table 3. Relationship between IL-6 and fecal microbial richness and diversity. Spearman correlations analysis.

	All			No T2D			T2D		
	n	Rho	p-value	n	Rho	p-value	n	Rho	p-value
IL-6 vs.									
Observed Richness	50	-0.26	**0.07**	29	-0.18	0.36	21	-0.37	0.11
Shannon	50	-0.25	**0.08**	29	-0.14	0.48	21	-0.37	0.11
InvSimpson	50	-0.22	0.14	29	-0.07	0.74	21	-0.38	0.10
Fisher	50	-0.26	**0.07**	29	-0.12	0.53	21	-0.43	**0.06**

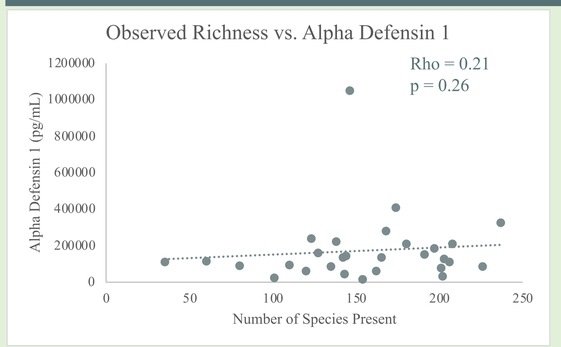
Figure 2. Correlation plots of observed species richness vs. alpha-defensin 1 in participants with & without self-reported T2D.

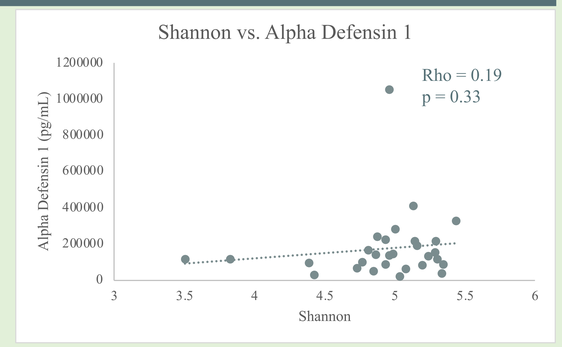
T2D





No T2D





CONCLUSIONS

Correlation Results

Alpha-defensin 1

- Trends for alpha-defensin 1 to be **negatively correlated** with **observed species richness** (Rho = -0.38, p = 0.09) as well as **Shannon** and **Fisher** (Rho = 0.39, p = 0.08 for both) in individuals with T2D
  - This trend is not observed in the group without T2D

IL-6

- Trend for IL-6 to be **negatively correlated** with **Fisher** in individuals with T2D (Rho = -0.43, p = 0.06)
  - This trend is not observed in the group without T2D
- Trends for IL-6 to be **negatively correlated** with **observed species richness** (Rho = -0.26, p = 0.07), **Shannon** (Rho. = 0.25, p = 0.08), and **Fisher** (Rho = -0.26, p = 0.07) in all study participants

Limitations

- Relatively small study population
- Uneven distribution of sex (82% female)
- High prevalence of pre-diabetes in the group without T2D

Future steps

- Investigate the relationship between alpha-defensins and abundance of different fecal taxonomic groups
- Repeat analysis in other populations

Project Significance

- Contributes to the knowledge of the roles of alpha-defensins, IL-6, and gut microbiome richness and diversity in inflammation associated with T2D
- Continued research is important to produce effective interventions to lower the disproportionate risk of developing metabolic diseases that refugee populations experience upon immigration to the United States

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