SUNDAY 916

Changes in the gut microbiota and fermentation products associated with enhanced longevity in acarbose-treated mice.

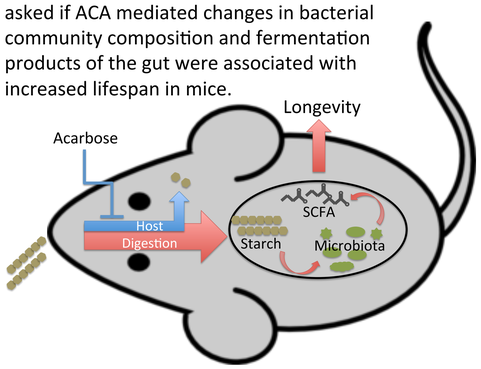
Byron J. Smith[1], Richard A. Miller[1], Aaron C. Ericsson[2], Randy Strong[3], David E. Harrison[4], Thomas M. Schmidt[1]

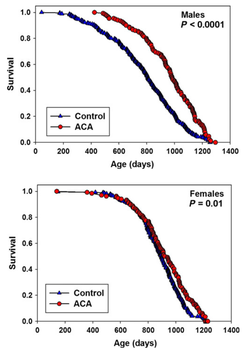
[1]University of Michigan, [2]University of Missouri, [3]The University of Texas Health Science Center at San Antonio, and [4]The Jackson Laboratory

University of Michigan
202-507-9572
bjsm@umich.edu

Introduction

Mice treated with acarbose (ACA) continuously during adulthood, have a median lifespan 22% and 5% longer in males and females, respectively [1]. By inhibiting the host enzymes that normally degrade starch, ACA increases the quantity of polysaccharide entering the colon [2], in humans leading to increased production of short-chain fatty acids (SCFAs) [3]. Given the documented benefits of SCFAs, we asked if ACA mediated changes in bacterial community composition and fermentation products of the gut were associated with increased lifespan in mice.

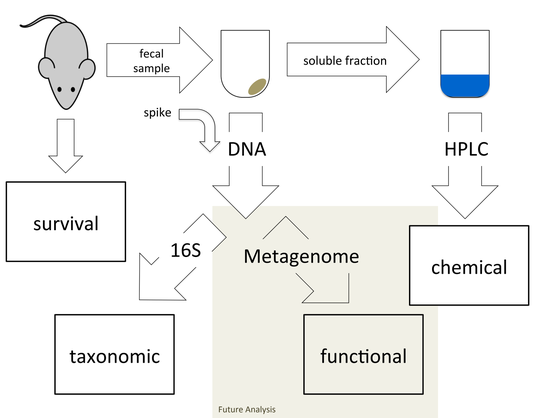




Methods

At three sites, The University of Michigan (UM), University of Texas San Antonio Health Science Center (UT), and The Jackson Labs (JL), mice were either fed a control diet, or the same diet amended with 1000 ppm ACA from 8 months of age onwards. Individual fecal samples were collected from mice between 109 and 137 weeks of age. At each site, samples from 24 control and 24 ACA treated mice were collected, with an equal number from males and females. Matched longevity data were collected for sampled mice from UM and UT.

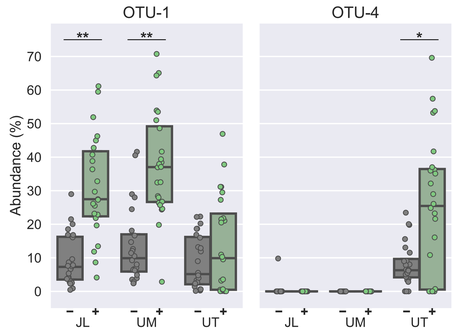
Samples were used for measurement of both fecal metabolites (HPLC) and bacterial community composition (16S rRNA gene survey). Samples were spiked with a constant quantity of *Sphingopyxis alaskensis* stationary phase culture before DNA extraction, allowing for comparison of rRNA gene densities across samples [4]. Operational taxonomic units (OTUs) were defined at a 97% similarity threshold.

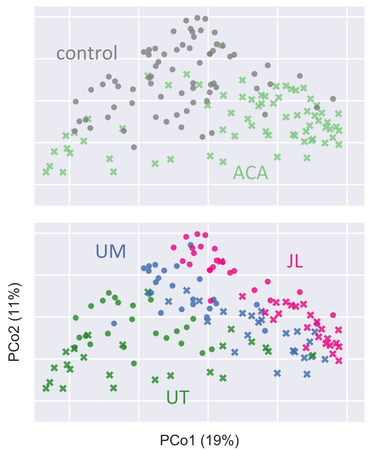


Results

Surveys of the 16S rRNA gene confirm that ACA modulates the composition of the fecal bacterial community. A principle coordinate analysis based on Bray-Curtis dissimilarities (**figure right**) demonstrates the significant separation between control and treated mice at each site, as well as within the pooled data (ANOSIM R = 0.28). Communities at each site were also distinct (R = 0.36).

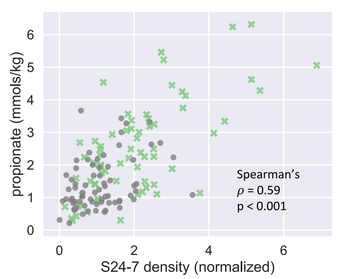
The difference between treatment groups was dominated by large increases in the relative abundance of two OTUs, designated OTU-1 and OTU-4 (**figure below**). Based on spike-and-recovery quantification, the density of the 16S rRNA gene from these taxa was greater in ACA. Both OTUs are classified as members of the largely uncultured family S24-7, which has previously been observed to be common in the gut of mice and to respond to dietary perturbations [5].

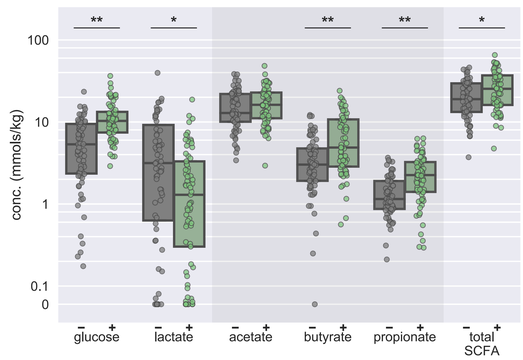




Along with shifts in bacterial populations, concomitant changes were observed in the metabolic profile, in particular increased SCFAs (**figure right**). Treatment effects on fecal metabolites varied between sites. Propionate was found to be subject to an interaction between sex and treatment, resulting in a greater increase in propionate with ACA treatment in males than in females.

Propionate was correlated with the abundance of S24-7. The **figure below** plots the density of 16S rRNA genes from OTUs classified as family S24-7 against the concentration of propionate. Density values shown are normalized to the median density in control mice. Reconstructed genomes have suggested that propionate production is common in members of the S24-7 family [5].





Including concentrations of acetate, butyrate, and propionate in a Cox proportional hazards model (**table below**) resulted in a substantially better fit to the data (ΔAIC = 4.0). Butyrate and propionate were associated with increased survival and acetate with reduced survival.

covariate	log HR	P > |t|
treatment (ACA)	0.078	N.S.
sex (male)	-0.040	N.S.
site (UT)	0.581	0.012
propionate	-0.246	0.015
butyrate	-0.128	0.016
acetate	0.060	0.043

Conclusions

- Fecal metabolite profiles in mice treated with ACA were shifted towards **increased butyrate and propionate** concentrations, SCFAs linked with decreased inflammation and host health.
- Bacterial communities responded with **large changes in composition**, particularly favoring two OTUs in family S24-7. The 16S rRNA density of this family was **associated with higher propionate concentrations**.
- Fecal **propionate, butyrate, and acetate** concentrations were statistically significant **predictors of mouse longevity**.
- **Future work** is needed to (1) explain the high variability between samples, in particular assessing temporal patterns at the scale of days or months, and (2) identify mechanisms (e.g. inflammation) through which SCFAs might affect host longevity.

Citations

[1] Harrison, D. E. *et al.* Acarbose, 17-α-estradiol, and nordihydroguaiaretic acid extend mouse lifespan preferentially in males. *Aging Cell* **13,** 273–282 (2014).

[2] Dehghan-Kooshkghazi, M. & Mathers, J. C. Starch digestion, large-bowel fermentation and intestinal mucosal cell proliferation in rats treated with the α-glucosidase inhibitor acarbose. *Br. J. Nutr.* **91,** 357 (2004).

[3] Holt, P. R. *et al.* Effects of acarbose on fecal nutrients, colonic pH, and short-chain fatty acids and rectal proliferative indices. *Metabolism.* **45,** 1179–1187 (1996).

[4] Stämmler, F. *et al.* Adjusting microbiome profiles for differences in microbial load by spike-in bacteria. *Microbiome* 1–13 (2016). doi:10.1186/s40168-016-0175-0

[5] Ormerod, K. L. *et al.* Genomic characterization of the uncultured Bacteroidales family S24-7 inhabiting the guts of homeothermic animals. *Microbiome* **4,** 36 (2016).

Acknowledgments & Contact

This work was supported by NIA grant AG022303, a Burroughs Wellcome Fund training grant, and the Glenn Foundation for Medical Research.

@ByronJSmith
bsmith89
bjsm@umich.edu
ByronJSmith.com

Digital Poster