

The effect of capsaicin on the Butyrate production pathway and taxonomical distribution in a gut environment

Aim of study

In this study it is investigated if consumption of chili will increase the relative abundance of genes producing the short chain fatty acid butyrate, as well as affects the relative abundance of firmicutes.

Introduction

Over the past decades it has become increasingly evident that the microbiome has a major effect on the health of the host. One of the common bacterial phyla found in the human gut, is Firmicutes. One of the positive contributions from Firmicutes is their production of short chain fatty acids, of which butyrate is especially important. Butyrate has a positive effect on human health, as it has key role in maintaining gut homeostasis. Furthermore butyrate is the main energy source for colonocytes. The positive effect of butyrate becomes evident as it is associated with prevention of the diseases ulcerative colitis and diabetes II [1].

Butyrate is synthesized via four pathways, however only the pathway using Acetyl Coenzyme A (Acetyl-CoA) as substrate is acknowledged as active in the gut (shown in orange in figure 1) [1].

Chili, and its active ingredient Capsaicin (CAP), is thought to have several health benefits. In a recent study performed in china, it was proven that CAP increased the Firmicutes/Bacteroidetes ratio [2]. In this study the focus lies on the relative abundance of Firmicutes.

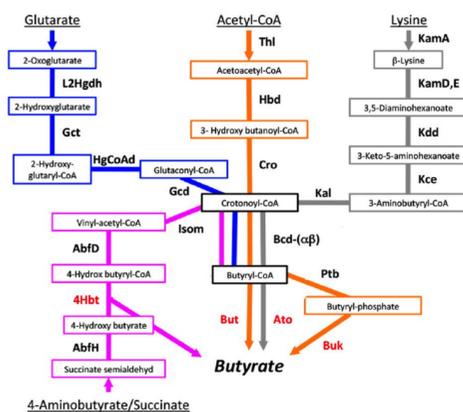


Figure 1. Butyrate production pathways as found primarily in distinct families within the Firmicutes. Corresponding genes (coding for enzymes) are also shown, Only the pathway using Acetyl-CoA (shown in orange) is thought to be active in the gut [1].

Methods and Workflow

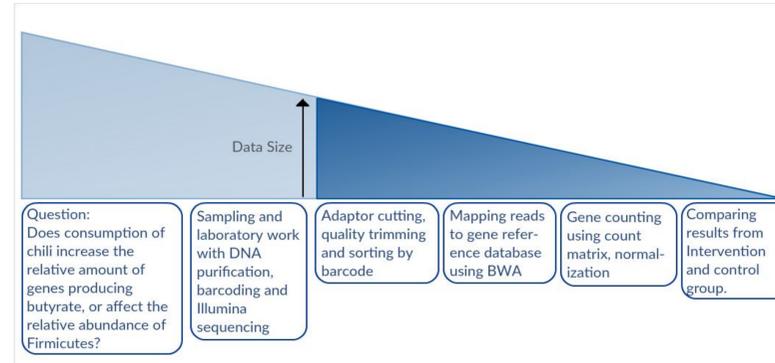


Figure 2. Flowsheet of the methods applied for this intervention study. Cohort of 21 individuals split into an intervention (10 subjects) and a control group (11 subjects). The intervention group had one ingestion of chili in between the two samplings. Illumina paired-end was applied for the sequencing. The raw reads were filtered using the Prinseq software (<http://prinseq.sourceforge.net/manual.html>) with a quality cutoff of 20 and reads shorter than 35 bp discarded based on a fastqc report. Reads from each sample were mapped against a database, containing 3.9 million genes that may be present in a gut environment, using the Burrows-Wheeler Aligner (BWA) [3]. Genes required for butyrate production were identified by KEGG Orthologous (KOs). The taxonomical references were based solely on reads mapping to genes related to 741 defined Metagenomic Species (MGSs) obtained by Nielsen et al. [3].

Results

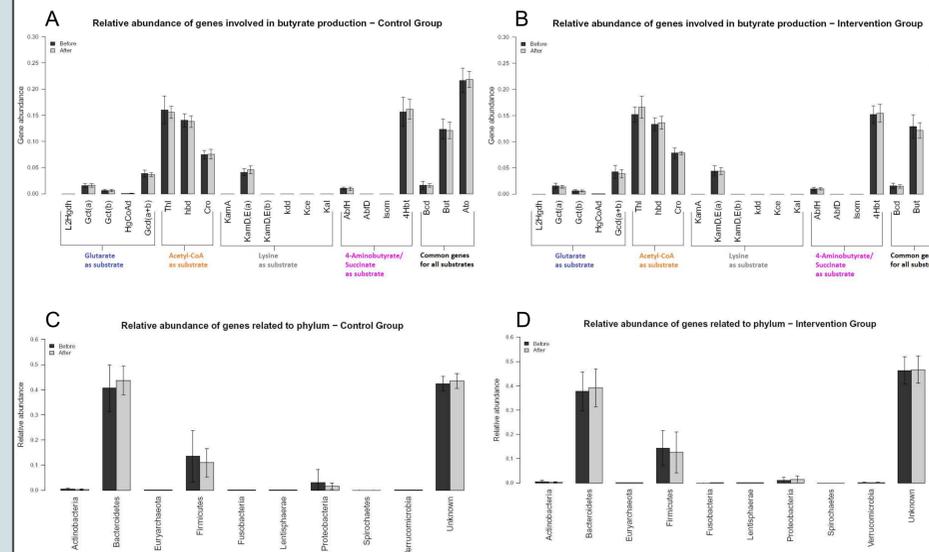


Figure 3. Graphic representation of the relative abundance of genes involved in butyrate production in the control group (A) and in the intervention group (B). Graphic representation of the relative abundance of genes related to phylum in the control group (C) and the intervention group (D).

Results and Discussion

Butyrate Production:

An increase in the amount of the genes from the Acetyl-CoA pathway is observed. From this it can be deduced that chili may have an effect in butyrate production via the Acetyl-CoA pathway. However these results are not consistent enough in order to demonstrate that chili has a direct effect on the butyrate production pathway. A possible reason could be the short intervention period and the size of the two groups

Firmicutes:

There is no clear evidence of increase in the relative abundance of genes related to Firmicutes or other phyla. Therefore it can be assumed that chili does not have any effect on the abundance of species in the gut.

Bray Curtis:

In order to determine the dissimilarity between the relative abundance of genes between the before and after samples, the Bray-Curtis index has been calculated for both the control and intervention group:

Bray-Curtis Index	
Control	0.012
Intervention	0.019

These results show that the dissimilarity between genes is more pronounced in the intervention group than in the control group. This is consistent with the results shown in figure 3(A+B), which show an increase of the relative abundance of genes related to the butyrate production via the Acetyl-CoA pathway.

Conclusion

- The abundance of bacteria producing butyrate through the Acetyl-CoA pathway may increase or the genes related to the Acetyl-CoA pathway may be promoted by eating chili.
- The abundance of different phyla does not change after the intervention.
- There is no evident effect on the production of short chain fatty acid while eating chili.



References

1. Vital M., Howe A. C., and Tiedje J. M., Revealing the Bacterial Butyrate Synthesis Pathway by Analyzing (Meta)genomic Data, *American society for microbiology*, 2014
2. Kang, C. et al., Healthy Subjects differently respond to dietary capsaicin correlating with the specific gut enterotypes, *The Endocrine Society*, 2016
3. Nielsen B. H. et al., Identification and assembly of genomes and genetic elements in complex metagenomic samples without using reference genomes, *Nature Biotechnology*, 2014

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All group members contributed equally to the poster.
 Morten performed most of the analytical work in UNIX.