

PROLONGED WATER-FASTING AND THE GUT MICROBIOTA: PRELIMINARY RESULTS OF AN OBSERVATIONAL STUDY

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Objectives: Water-only fasting (“WF”) is defined as a dietary intervention with absence of food intake and ad libitum water consumption. WF, complete or partial, has anti-inflammatory and anti-oxidative effects, as well as positive effects on lipid metabolism, the endocrine system and emotional well-being. WF alters the gut microbiota composition and functionality, which could be a plausible explanation for the observed effects. To the best of our knowledge, this study is the first to evaluate the impact of prolonged WF on the gut microbiota.

Methods: Healthy male volunteers (n=3, age 25-40 years, BMI 20-27 kg/m²) participated in a medically supervised WF, consuming solely water for 7 consecutive days. Body composition, bowel movements and basal metabolic rate were monitored before, during and after WF. Prior and post intervention stool samples were obtained and 16s rDNA-Seq was conducted utilizing the Intest.pro stool kit (BIOMES NGS GmbH, Germany). Descriptive data analysis was performed.

Results: The WF induced statistically significant changes in the composition of the gut microbiota on all taxonomic level as well as its functionality. The taxonomic changes were not consistent among subjects and depended on baseline gut microbiota composition. On phylum level, a regulation of *Proteobacteria* was observed as well statistically insignificant increases in *Actinobacteria* and *Verrucomicrobia*. Regarding relative abundances of taxa, a significant decrease in *Ruminococcus* was noted, besides insignificant, but consistent decreases of *Fusicatenibacter*, *Anaerostipes*, *Coprococcus*. It should be noted that an increase in *Lactobacillus-Group* was seen. Function-wise, pathways involved in food degradation, especially sugar- and fiber-degrading pathways and fermentation were reduced, as well as pathways involved in the biosynthesis of purine and pyrimidine bases.

Conclusion: These preliminary findings suggest that WF significantly impacts the gut microbiota by altering its composition and dominant pathways. The observed pathway alterations can be attributed to the complete substrate deprivation by WF. The resulting regulation of potentially pathogenic and pro-inflammatory *Proteobacteria* and increase in anti-inflammatory *Lactobacillus-Group* could potentially mediate the anti-inflammatory and anti-oxidative effects of WF. Although based on a small study sample, the results of this study suggest that prolonged WF may have a potential therapeutic effect on the gut microbiota.

Key words: anti-inflammatory, prolonged fasting, water-only fasting, gut microbiota, pathways