Bittersweet: artificial sweeteners and the gut microbiome

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Standfirst: In a clinical trial, non-nutritive sweeteners – supposedly inert – were shown to disrupt the gut microbiome of healthy people and impair glucose tolerance.

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For many years, the use of artificial (non-nutritive) sweeteners has been widespread across the globe. A source of profit for the food industry, sweeteners have been considered as an essential alternative to the excessive use of sugar — considered harmful on account of its association with cardiometabolic pathologies, cancers and poor dental health[1]. Sweeteners can be consumed directly and are available in a range of options with different sweetening power, but these supposedly inert and calorie-free compounds can also be found in many foodstuffs[2]. The description of the health risks potentially associated with their regular use is the subject of regular controversy, whether for artificial sweeteners (saccharin, sucralose, aspartame) or for natural sweeteners such as steviol glycosides. In a recent issue of Cell, Suez et al.[3] report results of a randomized controlled trial performed in 120 healthy participants, which shows that non-nutritive sweeteners induce perturbations of glucose tolerance in a proportion of healthy individuals, which might be mediated by compositional and functional changes in the gut microbiome[3].

The replacement of sugar-sweetened beverages with non-nutritive sweetened beverages may contribute to a reduction in body weight and body fat[4], but concerns about the toxicity of sweeteners and their impact on the brain were first raised more than a decade ago[5]. However, there is a surprising irony in the fact that these sweeteners are now suspected of being involved in the deterioration of glucose tolerance — despite their supposed neutral impact in metabolic diseases.

Associations between artificial sweeteners and intestinal microbiome composition have been observed over a decade ago[6] and considering the tremendous impact of the microbiota on host metabolism, the hypothesis that sweeteners could impact the host through alterations of the intestinal microbiota composition became plausible. This was highlighted in a previous in vivo study from some of the current authors (published in 2014)[7], in which mice that consumed relatively high doses of three non-caloric artificial sweeteners (aspartame, saccharin and sucralose) developed glucose intolerance, owing to a disturbance of the microbiota and its metabolic functions. Previous *in vitro* exploration using genetically modified *E coli* also revealed that six artificial sweeteners may alter bacterial growth, thereby reinforcing the fact that sweeteners indeed affect the activity of the gut microbiota[8].

In this very well-conducted and data-rich new study, Suez et al.[3] explored the effects of oral supplementation with sweeteners sucralose, saccharin, stevia and aspartame, compared to glucose supplementation for 14 days in 100 healthy participants (5 arms, 20 participants per group). Importantly, sweeteners were used at doses below acceptable daily intake recommendation (from 8 to 75% of ADI). A sixth control group of 20 participants without supplementation was also evaluated. All participants were metabolically healthy people who did not previously consume sweeteners in their regular diet; they were selected after a long screening phase involving 1,325 people, which highlights the prevalence of non-nutritive sweeteners in a regular diet. In the study, the period of sweetener administration was preceded and followed by a 7-day observation phase, during which a series of detailed clinical, anthropometric and biological measurements as well as shotgun oral and fecal metagenomics and blood metabolomics were performed. Glucose tolerance tests and continuous blood glucose measurements were repeatedly performed and data on diet, physical activity and smoking were collected; following these analyses, the causality of the effects of sweeteners on the gut microbiome was explored by fecal transfer experiments.

Despite large but expected inter-individual differences in glucose tolerance changes, the ingestion of saccharin and sucralose caused worsening of glucose tolerance while aspartame and stevia were neutral after a glucose challenge test. However, all sweeteners had distinct effects on oral and fecal microbiome composition and key functions (such as purine or pyrimidine metabolism, glycolysis, amino-acid metabolism). These modifications could explain the variation in glucose tolerance, at least for sucralose which displayed the most prominent effect on microbiome functional potential. Together, these findings confirm the previous observations that some sweeteners are not neutral on the gut microbiome and glucose tolerance — although the intensity of the observed effects (e.g. both on the microbiome and glucose tolerance) is variable from sweetener to sweetener and from person to person.

To confirm the causal link between sweeteners, the microbiome and glucose tolerance, Suez et al.[3] carried out fecal microbiota transfer experiments from selected study participants (representing all sweetener-supplemented and control groups) to germ-free mice. Fecal samples were taken before and after supplementation from the 42 participants whose microbiome was most or least responsive to sweeteners in each group. Almost all of the animals who were colonized with samples from sweetener-supplemented groups had deterioration in glucose tolerance, but those colonized with samples from control groups (glucose supplement or no supplement) did not. Surprisingly, even animals colonized by samples from participants with the lowest microbial response to saccharin had impaired glucose tolerance. Indeed, participants in the saccharin group who showed the greatest functional effects on the microbiome, these effects were corroborated by measurements of systemic metabolites, which in turn were associated with individual glucose variation. The precise mechanism by which these sweeteners may exert host effects via faecal microbiota changes warrant deeper investigation in light of the fact that some of them (sucralose, saccharin and stevia), are partially metabolized in the upper intestinal tract and only a fraction of them reaches the colon where it still significantly impacts microbiota composition and functionality.

The work from Suez et al. demonstrates the molecule-dependent and the individual-dependent impact of non-nutritive sweeteners on the human microbiome and the short-term downstream consequences on host glucose metabolism — at least in metabolically healthy individuals. In a broader context, this study also reinforces the previously described notion of the variability of individual microbiota-food[9] or microbiota-drug[10] interactions and thus the variability of outcomes based on dietary or drug interventions. In particular, the results highlight the need for

robust evaluation of the short-term and long-term impact of all commercially available sweeteners on human health; for example, several generations of stevia glucosides have been launched on the market during recent years and little information has been provided about the differential impact of those glucosides on intestinal microbiota. It will also be important to conduct similar studies evaluating the impact of sweeteners in the people with various diseases such as type 2 diabetes and cardio-vascular diseases.

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