

Ant It Sweet? Analyzing the Effects of Aspartame, Sucralose, Acesulfame K, and Saccharin on the Feeding, Mortality, and Gut Microbiota of *Camponotus floridanus*

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Globally, over 18% of men and 20% of women have been diagnosed with obesity as of 2025. While approved artificial sweeteners like aspartame, sucralose, acesulfame K, and saccharin are common sugar substitutes to solve this, most consumers are unaware about their possible effects on gut microbiota. This examined sweetener impact on gut bacterial composition and mortality using *Camponotus floridanus*, a model for feeding and microbial health. It was hypothesized that sucralose would be most detrimental. 375 *Ca. floridanus* ants were exposed to one of four artificial sweeteners over seven days. Feeding preferences were recorded, mortality tracked daily, and gut microbiota analyzed through culturing and Gram staining. Colony-forming units (CFUs) were quantified, and the Firmicutes-to-Bacteroidetes ratios and general gut dysbiosis were quantified. Sucralose was the most consumed sweetener, with over 20% higher preference than others. However, saccharin showed the highest mortality at 23%. ANOVA and pairwise comparisons revealed significant bacterial shifts across sweeteners ($F(4, 95) = 23.67$, $\eta^2 = 0.726$), with saccharin reducing Bacteroidetes from 29% to 25% ($F(12, 380) = 15.89$, $r^2 = 0.332$). Visual analyses, illustrated microbial shifts, while Cox regression and Kaplan-Meier survival curves confirmed saccharin's negative impact. Brays-Curtis dissimilarity heatmaps also displayed general gut bacterial shifts. These findings depict saccharin, not sucralose, as the most harmful sugar substitute to survival and gut microbiota in *Ca. floridanus*. The Firmicutes-to-Bacteroidetes ratio and general bacterial shifts highlight potential metabolic risks, emphasizing the need for further research and consumer awareness about artificial sweeteners' health effects.

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