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The Effects of Artificial Sweeteners on the Human Body and Microbiome

Lauren K. Steely
Eastern Kentucky University, lauren_steely3@mymail.eku.edu

Eastern Kentucky University

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Lauren Steely

Faculty Member

Dr. Bill Staddon

Department of Biology

Abstract

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Lauren Steely

Dr. Bill Staddon Department of Biology

While artificial sweeteners are becoming a popular sugar alternative, their negative effects remain understudied and obscure to the general public. Whether people purposely incorporate them into their diet, artificial sweeteners are found in thousands of products and are consumed by all ages. The calories and weight gain associated with refined sugar often cause people to turn to zero-calorie options, thinking they are healthier; however, artificial sweeteners serve just as, if not greater, a threat to one's health. Artificial sweeteners have been associated with weight gain, numerous health risks, type 2 diabetes, and negative effects on the gut microbiome. The goal of this project was to examine artificial sweeteners' effects on the body to see if they are safe for consumption. A literature review was conducted to gather all the negative effects. Also, two experiments were designed; one tested to see if popular artificial sweeteners inhibited bacterial growth. The other investigated if artificial sweeteners were capable of promoting *Escherichia coli* or *Enterococcus faecalis* growth. The literature review concluded that artificial sweeteners can cause weight gain, increase the risks for stroke and cardiovascular disease, and increase the severity of disorders of the gut microbiome. The experiments showed that artificial sweeteners do not prevent the growth of bacteria but enhanced the growth of *E. coli*.

Keywords and phrases*: artificial sweetener, bacteria, sucralose, saccharine, aspartame, microbiome, diet*

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Introduction

Artificial sweeteners, often referred to as non-nutritive sweeteners (NNS), are chemically engineered sugar alternatives that provide consumers with the same sweetness of sugar at a zero- or lower-calorie benefit. To dieters and diabetics trying to manage their sugar intake, artificial sweeteners seem to be an easy way to achieve the sweetness they desire without the consequences that come with refined sugar. According to Harvard T.H. Chan School of Public Health (2023), artificial sweeteners can be found in baked goods, candies, desserts, yogurts, gelatins, puddings, breakfast cereals, sports drinks, sodas, and thousands of other products. As seen in many restaurants, brands like Sweet N' Low®, Equal®, and Splenda® manufacture individual packages so that artificial sweeteners can be added to meals and beverages. In the one-year period between 1999 and 2000, the consumption of artificial sweeteners skyrocketed by 200% in children and adolescents and over 50% in adults (Basson et al., 2021). The argument to whether artificial sweeteners are harmful remains a hot topic of debate. Small amounts may seem harmless; however, with their overwhelming presence in most products, people are likely consuming more artificial sweeteners than they realize. Unfortunately, not only has there been little research on artificial sweeteners, but most of the research has been inconclusive or resulted in minimal findings. Many articles contradict one another, and many find that only certain populations are affected. Too little is known about artificial sweeteners for them to be consumed at the rate they currently are. After reading many studies and examining the data produced from these experiments, artificial sweeteners may serve more as a detriment than a benefit to the human body due to their negative effects on the metabolism and microbiome, their obesogenic effects, and because they increase the risks of diseases and numerous health conditions.

Literature Review

History of Artificial Sweeteners

Three of the most popular artificial sweeteners, saccharin, sucralose, and aspartame, were all discovered accidentally. Saccharine is the oldest and first widely commercialized artificial sweetener. It all started when it was accidentally discovered by a chemist working with benzoic sulfimide in the late 1870s. After noticing a sweet taste on his hand, he named the compound saccharin (American Chemical Society, 2019). Attempting to create a new insecticide, sucralose was created by the combination of sucrose and chlorine molecules, making it 450-600 times sweeter than sucrose (Jolly, 2004). Similarly to saccharine, aspartame was discovered after a scientist licked his finger after performing experiments; to his discovery, he tasted a sweetness, and aspartame was created (Czarnecka et al., 2021).

Not all accidental discoveries are a bad thing; however, scientists were too quick to release artificial sweeteners to the public before truly examining their effects on the body. It was not long before research came out declaring some artificial sweeteners to be harmful. According to the National Cancer Institution (2023), the first banned artificial sweetener was cyclamate and its various forms in 1969 because animal trials suggested they caused cancer. Since then, scientists have found additional data that suggest cyclamate does not cause cancer, but cyclamate has yet to have been approved for public use. Some European countries approve of sucralose use, but only for adults and not foods for young children due to the lack of nutrients that children would suffer from with frequent consumption of artificial sweeteners (Russel, 2023). The question of safety for more popular sweeteners like aspartame, sucralose, and saccharin have been reviewed by the FDA. It deems these artificial sweeteners to be safe; however, this is only if the sweeteners are consumed at the recommended amount. Also, the FDA does not take into consideration the subtle, yet long-term tolls that artificial sweeteners can have on the body.

There have been cases in the past where artificial sweetener companies have misled consumers to believe that some artificial sweeteners are more natural and healthier than other artificial sweeteners. This particular case involved two of the largest artificial sweeteners: aspartame and sucralose. In 2004, the makers of Equal® sued the makers of Splenda® due to Splenda's slogan, "Made from sugar – so it tastes like sugar". The company claimed that the tagline misled millions of consumers by implying that sugar is an ingredient in Splenda®. In an attempt to answer the lawsuit, the company's maker, McNeil Nutritionals, had stated, "The sweetening ingredient in Splenda® is made by a multistep process that starts with cane sugar." He immediately added, "Splenda® is an artificial sweetener that does not contain sugar," which indicated the sugar is lost during the manufacturing process (Browning, 2007). According to CBS News (2007), since the manufacturing of Splenda® involves the chemical alteration of cane sugar, McNeil's attorneys argued that the company could claim that Splenda® is made from sugar. The makers of Equal® counteracted and said that the slogan caused people to think that they were using real, zero-calorie sugar instead of understanding that Splenda® was an artificial sweetener. The final verdict was not released to the public, but talks of settlement hinted to Equal's® maker, Merisant, receiving compensation.

Weight Gain & Obesogenic Effects

With weight loss being one of the hottest topics in the media since the 1980s, people are constantly looking for simple, easy ways to slim down. While some of these weight loss trends include liquid diets, fat burning pills, Ayds candies, or the multitude of diet programs, one of the most common ways people cut out calories in today's time is by using artificial sweeteners. While thousands of products contain FDA-approved artificial sweeteners, it is still questionable as to how they affect the human body. Because they do not contain fiber, vitamins, or minerals, they cannot offer the same health benefits that natural sugars do; however, the use of artificial sweeteners is frequently promoted in dieting plans.

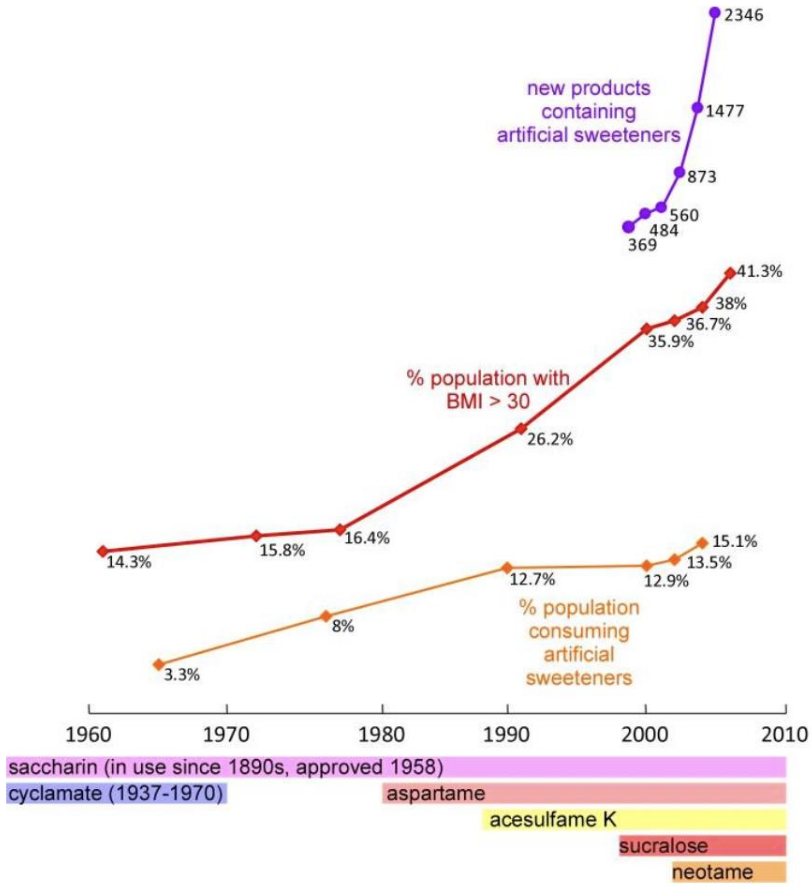

Figure 1: Relationship between products containing artificial sweeteners, percentage of the population with BMIs > 30, and the percentage of the population that consumes artificial sweeteners (Yang, 2010).

The incidence of obesity has tripled since 1975. As of 2021, the World Health Organization estimated that there are nearly 650 million obese adults and 1.9 billion overweight. As the amount of artificial sweeteners in foods and beverages has increased, the obesity rate has not slowed. As seen in Figure 1, there is a positive relationship between the number of products containing artificial sweeteners, the percentage of the population with BMIs > 30, and the percentage of the population that consumes artificial sweeteners (Yang, 2010). A study that supported the findings of Yang measured the relationship between artificial sweetener consumption and long-term weight gain. It concluded, “Overall, adjusted ΔBMIs were 47% greater among artificial sweetener users than nonusers,” (Fowler et al., 2008). The participants were stratified by gender, ethnicity, base weight gain category, or exercise-change category; ΔBMIs were significantly higher in artificial sweetener users in 13 of the tested strata. The findings of the study showed that artificial sweetener consumption may be helping to fuel the obesity epidemic.

In a study performed in 2022, the idea that the artificial sweetener Ace-K induces weight gain was supported when Sprague Dawley rats (5–6-week-old males) that consumed Ace-K-sweetened water showed both weight gain and fat gain. However, these findings were not consistent with juvenile rats (4-week-old males). The findings presented that chronic Ace-K consumption led to suppressed sugar taste responsiveness, reduced lingual sweet taste receptor expression, and an impaired hippocampal-dependent memory (Iizuka, 2022). Aspartame consumption increased weight gain and fat mass, and sucralose behaved similarly at a lesser extent. Sucralose consumption also enhanced a high-fat-diet and increased reactive oxygen species and induced endoplasmic reticulum stress in HepG2 cells. Saccharine intake resulted in weight gain that was unrelated to caloric intake (Iizuka, 2022). Overall, all four of the artificial sweeteners tested resulted in weight gain as well as numerous other health issues amongst the different sweeteners.

The examination of how artificial sweeteners interact with taste receptors helps explain why artificial sweeteners can lead to weight gain. When a person ingests a food or drink, the thousands of taste bud-containing papillae on the tongue receive the signal through taste hairs. This signal is then sent to the thalamus and gustatory complex, so the taste can be determined as sweet, savory, bitter, salty, or sour (Dingman, 2018). When products with artificial sweeteners are consumed, the sweet taste determined by the brain causes the body to prepare for the calories that come along with natural or refined sugar. However, when calories enter the body, the body is unable to shut down the hormonal cues in the brain that control food intake. This can lead to overeating. Frequent consumption of artificial sweeteners was shown to increase appetite for sweet foods, which not only fuel the obesity epidemic, but also increase one's risk for type two diabetes due to a buildup of glucose. More importantly, the relationship between artificial sweeteners and butyric acid also must be taken into account. Butyric acid contains anti-obesogenic effects such as reducing appetite, inducing sustained satiety, activating brown adipose tissue which promotes energy expenditure and fat oxidation, reducing insulin resistance, and improving dyslipidemia (Farup, 2019). Butyric acid levels are mediated by the gut-brain neurocircuits. Due to the imbalance between the sensory and metabolic signals caused by artificial sweeteners, butyric acid levels can be reduced.

Artificial sweeteners were linked to insulin resistance, increased insulin activity, and insulin desensitization which all increase a person's risk for developing type two diabetes. Dryden summarized findings that artificial sweeteners may increase insulin activity after oral glucose administration. In obese, nonconsumers of artificial sweeteners, insulin secretion was higher after sucralose ingestion than water. After the subjects drank a sucralose-sweetened beverage, their blood sugar peaked at a higher level and their insulin levels rose about 20% higher than when the subjects drank water (Dryden, 2016). Another study reported that insulin levels increased after the participants drank diet beverages containing aspartame, Ace-K, and sucralose in relation to water (Sylvetsky, 2016). However, in normal, healthy-weight adults, there was no effect of artificial sweeteners on insulin release, suggesting that body weight may permit certain hormonal responses (Wilk et al., 2022). In a 2018 study, scientists found that with a moderate consumption of sucralose in healthy adults came a decrease in insulin sensitivity (Romo-Romo et al.). A 2019 study found that, “In summary, consumption of artificial sweeteners may activate GPCR signaling pathways that lead to cross-activation of insulin receptors through the neuromedin B receptor (NMBR) and ultimately promote the development of insulin resistance and T2DM,” (Liauchonak et al.). The role of insulin can be compared to how locks and keys work together. Glucose enters the body with the intake of foods or drinks. It enters the bloodstream, but it cannot enter cells without insulin. Insulin serves as a key that can unlock the openings on cells, so glucose can easily move into them. Insulin resistance and insulin desensitivity prevents insulin from properly interacting with the cells, resulting in a buildup of glucose in the bloodstream. With nowhere to go, the glucose is stored as fat. The consumer is left tired and hungry, thus the cycle repeats.

Effects on the Gut Microbiome

Billions of microorganisms come together in the gastrointestinal tract, skin, mucosa, respiratory tract, urogenital transact, and mammary gland to create the human microbiome. Species in the microbiome mutualistically exist with the host by helping humans to maintain general health and wellbeing. As seen in Figure 2, eubiota in the human gut microbiome is responsible for maintaining homeostasis, regulation, and the development of the immune system, host’s nutrition, and colonization resistance (Ogunrinola et al., 2020). Many things can affect the gut microbiome, such as antibiotics, diet, obesity, and pertaining to this study: artificial sweeteners.

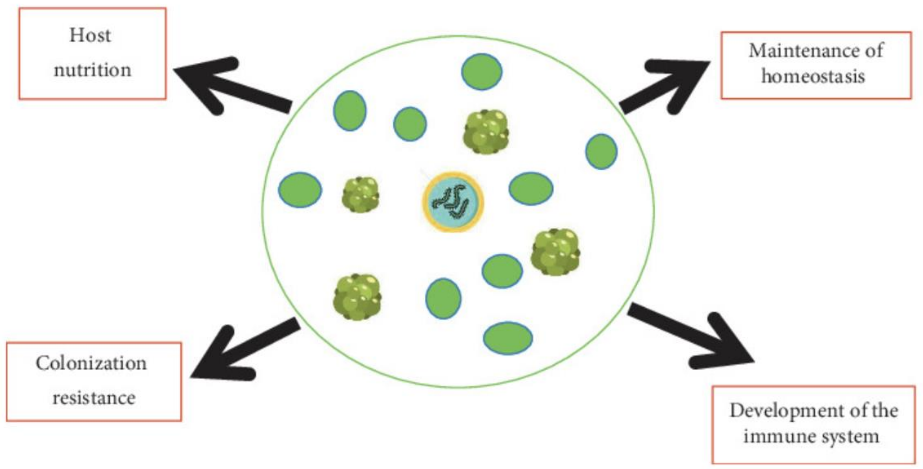

Figure 2: Symbiotic interactions between eubiotic flora and the human body (Ogunrinola et al., 2020).

Artificial sweeteners have been known to alter gut microbiota which can result in the diseases listed above. This is because, with the exception of aspartame, the majority of artificial sweeteners cannot be broken down by the body. They remain intact and pass through the gastrointestinal tract until they are excreted in feces or through urine via the kidneys. Since they are not absorbed or metabolized, they can disrupt the bacteria present in the gut microbiome. Aspartame on the other hand, is broken down into aspartic acid, phenylalanine, and methanol which is toxic. According to the CDC (2011), the by-products of methanol metabolism can lead to a buildup of acid in the blood, blindness, and death. Aspartame also serves as a threat to those with phenylketonuria, a rare genetic disorder that prevents the breakdown of phenylalanine resulting in a buildup in the bloodstream (Mayo Clinic, 2022).

Dysbiosis, or an imbalance of microorganisms in the gut, can lead to or worsen inflammatory diseases like rheumatoid arthritis, atherosclerosis, ulcerative colitis, Crohn's disease, type one and two diabetes, and even cancer (Elias-Oliveira et al., 2020). Organisms in the microbiome are responsible for carrying out biochemical activities that influence carcinogenesis, tumor development, and response to immunotherapy (Thursby & Juge, 2017). As seen in Figure 3, one result of dysbiotic gut flora is the production of carcinogenic metabolic toxins (Ogunrinola et al., 2020). They also cannot properly carry out their typical functions, resulting in an increased risk for colorectal cancer in particular. The presence of higher levels of certain species of bacteria in the gut microbiome were linked to breast cancer, prostate cancer, gastric cancer, and extraintestinal cancer (Ogunrinola et al., 2020).

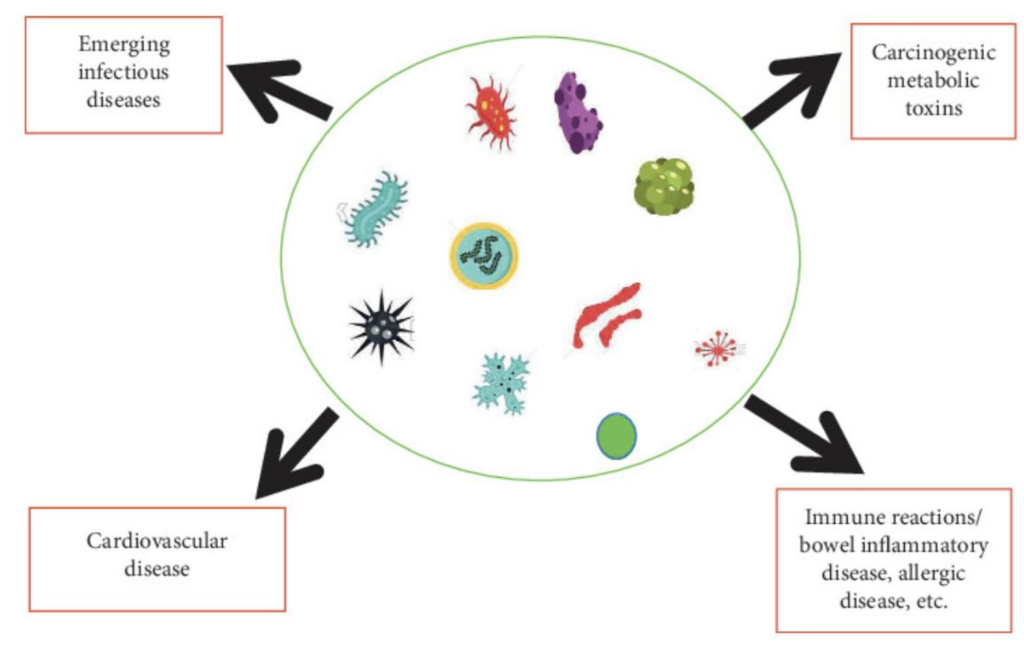

Figure 3: Dysbiotic flora and its impact on human health (Ogunrinola et al., 2020).

Another problem that can result from the accumulation of disease-causing organisms is the triggering of abnormal immune responses against body tissues. This can contribute to autoimmune diseases, bowel inflammatory disease, and other life-threatening conditions (Ipci et al., 2017). Disruptions to the balance between the gut microbiota and the host impair immune responses and can cause inflammatory disorders. According to the National Institute of Environmental and Health Sciences (2023), inflammation can lead to autoimmune diseases, including rheumatoid arthritis. Inflammation of the bowel can result in ulcerative colitis, Crohn's disease, or more common irritable bowel syndrome. Finally, inflammation can cause cardiovascular diseases such as high blood pressure and heart disease.

A 2022 study found that total artificial sweetener intake was associated with increased risks for cardiovascular and cerebrovascular disease. More specifically, aspartame intake was associated with an increased risk of cerebrovascular disease, and Ace-K and sucralose were associated with increased coronary heart disease risk (Debras et al.). There are many different types of cardiovascular disease including: coronary artery disease, valve disease, aneurism, cardiac arrhythmia, cardiomyopathy, pericarditis, and heart failure (Moore, 2021). Cerebrovascular disease refers to conditions that affect blood flow or the blood vessels in the brain. Examples include the three types of strokes: ischemic, hemorrhagic, or transient ischemic (Kauvery Hospital, 2023). A 2023 study on the newer, popular artificial sweetener erythritol found it increased risks for major adverse cardiovascular events which include nonfatal strokes, nonfatal heart attacks, or death due to cardiovascular causes. Scientists observed that when healthy volunteers ingested erythritol, changes were triggered in platelet function that increased the risks for enhanced thrombosis, or the formation of blood clots. This in turn increased the risks for heart attacks and strokes. (Witkowski et al.). Another study concluded that artificially-sweetened beverage consumption was associated with stroke risk and dementia. This included all-cause dementia and dementia because of Alzheimer's Disease. One explanation as to why dementia risks would be increased with artificially-sweetened beverage consumption was that diabetes mellitus was more prevalent in those who frequently consumed artificially-sweetened beverages, and diabetes mellitus is a known risk factor for dementia. While the linkage between artificial sweeteners and stroke risk can easily be explained, the study could not establish whether dementia was caused by diabetes mellitus, or if people with diabetes were simply more likely to consume artificially-sweetened beverages (Pace et al., 2017).

Some studies on artificial sweeteners tested their effects specifically on women. One examined the association between artificially-sweetened beverages (ASB) and strokes, coronary heart disease, and all-cause mortality in post-menopausal women in the United States. Scientists found that those consuming higher levels of artificially-sweetened beverages had significantly greater likelihood of all the previously listed adverse effects, except hemorrhagic stroke, than those who never or rarely (less than one per week) consumed artificially-sweetened beverages. In women with no prior history of diabetes mellitus or cardiovascular disease, there was an association between high ASB consumption and a more than 2-fold increased risk of small artery occlusion ischemic stroke. Finally, the study found that pertaining to obese women, there was an association between high ASB consumption and significantly increased risk for ischemic stroke (Mossavar-Rahmani et al., 2019).

Another study examined the effects of artificial sweeteners on kidney function decline. It concluded that two or more servings per day of artificially-sweetened beverages doubles the likelihood of kidney function decline in women (Lin & Curhan, 2011). The experiment measured the associations with artificially-sweetened beverages and the eGFR, estimated glomerular filtration rate, which measures the kidneys' efficiency. The study also examined the effects of sugar-sweetened beverages, and scientists found no significant associations with eGFR decline or microalbuminuria, the persistent elevation of albumin in the urine, for any category, including ≥2 servings per day (Lin & Curhan, 2011).

Experiments

Introduction

One of the major adverse effects of artificial sweeteners on the gut microbiome is the formation of antibiotic-resistant bacteria through biofilm formation. A biofilm is a slimy, net-like substance that protects bacterial cells and allows them to stick to surfaces such as the lining of the intestines. Biofilms are made of microbial cells and extracellular polymeric substances (Donlan, 2002). Prinzi and Rhode (2023) found that, “Bacteria living in a biofilm can exhibit a 10 to 1,000 fold increase in antibiotic resistance.” Not only will harmful bacteria be harder to eliminate, but alterations to “normal” gut bacteria will be harder to control with antibiotics and can result in other problems. Two examples of prevalent bacteria found in the human gut are *Escherichia coli* and *Enterococcus faecalis.* In normal amounts these bacteria are perfectly healthy; however, if their levels are heightened or alterations are made, side effects could include diarrhea, stomach cramps, fever, chills, fatigue, abdominal pain, and more (Watson, 2023). A 2021 study found that, “Sucralose, saccharin, and aspartame all promote pathogenic changes to *E. coli* and *E. faecalis,*” (Shil & Chichgar, 2021). Two experiments were conducted using the artificial sweeteners aspartame, saccharin, and sucralose, and the bacteria *E. coli* and *E. faecalis.*

Minimum Inhibitory Concentration

The goal of this experiment was to determine if aspartame, saccharin, or sucralose could inhibit the growth of *E. coli* and *E. faecalis*, and if so, at what minimum concentration. Concentrations of each artificial sweetener and ampicillin spanned across 13 concentrations from 512 µg/mL to 0 µg/mL.

Stimulation of Bacteria Growth

The goal of this experiment was to see if saccharine, sucralose, or aspartame could increase *E. coli* or *E. faecalis* growth. Six concentrations from 250 µg/mL to 0 µg/mL of each artificial sweetener were observed. Aa seen in Figure 4, a spectrophotometer was used to

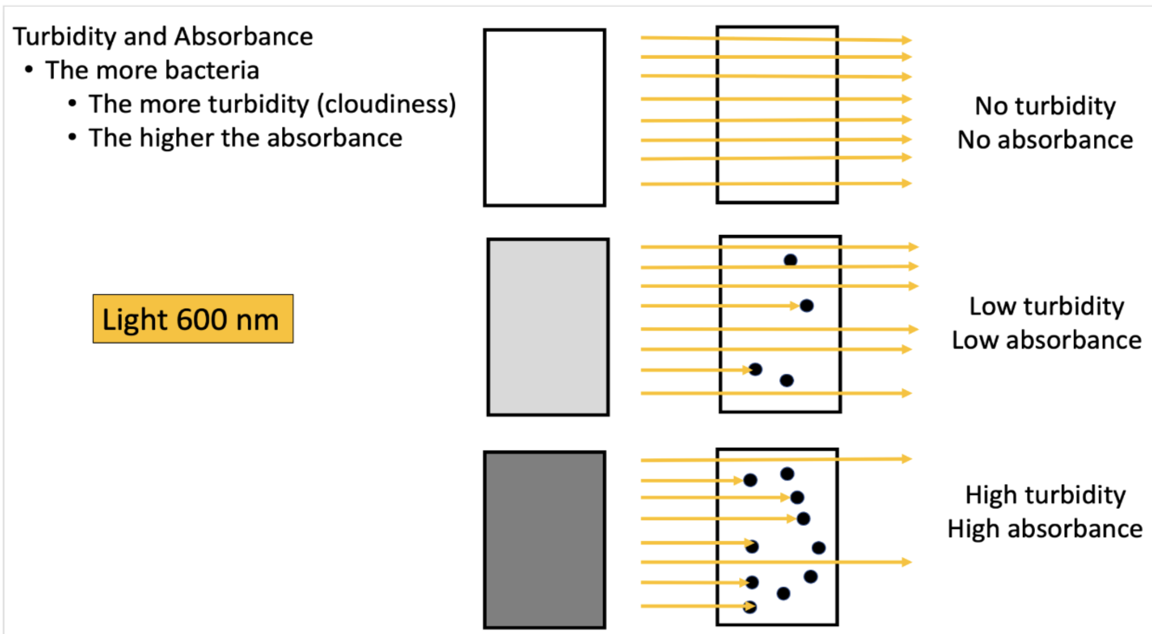

Figure 4: Absorbance readings of a spectrophotometer at different turbidity levels (Staddon, 2023).

measure the opacity of each solution to determine the amount of bacterial growth.

Materials & Methods

The MIC experiment used thirteen concentrations from 512 µg/mL to 0 µg/mL for each artificial sweetener and ampicillin. Sucralose, saccharine, aspartame, or ampicillin was added to a tube with 1 mL of TSB and 10µL of *E. coli*. Three replicas were made for each concentration for each artificial sweetener/antibiotic. The protocol was the same regarding *E. faecalis*. The tubes were incubated at 37°C and shaken at 55 rpm overnight. After, they were checked for cloudiness.

The stimulation growth experiment tested six concentrations from 250 µg/mL to 0 µg/mL for saccharine, sucralose, and aspartame. Each artificial sweetener was added to a tube with 1 mL of M9 media and 10µL of *E. coli*. To achieve the concentrations, various amounts of DI water were added. Four replicas were made for each concentration per artificial sweetener. The samples were read by a spectrophotometer. The protocol was the same regarding *E. faecalis.*

Results

The ampicillin minimum inhibitory growth concentration was 128 µg/mL for *E. coli* and *E. faecalis*. No artificial sweetener prevented the growth of *E. coli* (Table 1). However, sucralose was able to prevent the growth of *E. faecalis* at 512 µg/mL and partially at 256 µg/mL. Saccharine was able to partially prevent *E. faecalis* growth at the 512 and 128 µg/mL concentrations (Table 2).

Table 1: MIC results for *E. coli* (Steely & Staddon 2023).

NNS/Antibiotic	512	256	128	64	32	16	8	4	2	1	0.5	0.25	0
Ampicillin	-	-	-	+	+	+	+	+	+	+	+	+	+
Aspartame	+	+	+	+	+	+	+	+	+	+	+	+	+
Aspartame	+	+	+	+	+	+	+	+	+	+	+	+	+
Sucralose	+	+	+	+	+	+	+	+	+	+	+	+	+
Sucralose	+	+	+	+	+	+	+	+	+	+	+	+	+
Saccharine	+	+	+	+	+	+	+	+	+	+	+	+	+
Saccharine	+	+	+	+	+	+	+	+	+	+	+	+	+

Table 2: MIC results for *E. faecalis* (Steely & Staddon, 2023).

NNS/Antibiotic	512	256	128	64	32	16	8	4	2	1	0.5	0.25	0
Ampicillin	-	-	-	+	+	+	+	+	+	+	+	+	+
Aspartame	+	+	+	+	+	+	+	+	+	+	+	+	+
Aspartame	+	+	+	+	+	+	+	+	+	+	+	+	+
Sucralose	-	+	+	+	+	+	+	+	+	+	+	+	+
Sucralose	-	-	+	+	+	+	+	+	+	+	+	+	+
Saccharine	-	+	-	+	+	+	+	+	+	+	+	+	+
Saccharine	+	+	+	+	+	+	+	+	+	+	+	+	+

The results examining the association between artificial sweetener presence and enhanced *E. faecalis* growth was inconclusive. However, aspartame, sucralose, and saccharine significantly increased the growth of *E. coli* (Figure 5).

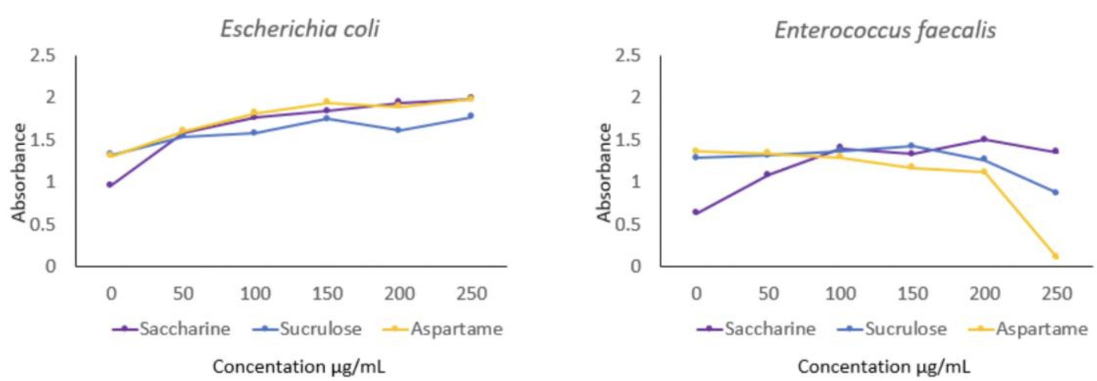

Figure 5: *E. coli* and *E. faecalis* absorbance readings at various concentrations of artificial sweeteners (Steely & Staddon, 2023).

Discussion

Ampicillin prevented the growth of *E. coli* at the 512, 256, and 128 µg/mL concentrations. Ampicillin is known to kill *E. coli* by inhibiting the synthesis of its bacterial cell wall by attaching to penicillin-binding proteins (Li et al., 2019). No artificial sweetener tested was able to prevent the growth of *E. coli* (Table 1). Similarly, many studies have found that artificial sweeteners bear minimal effects on planktonic bacterial growth (Shil & Chichger, 2021). Sucralose and saccharin were able to prevent growth at a few concentrations (Table 2). One possible explanation to the inconclusive results is that *E. faecalis* takes longer than *E. coli* to grow. *E. faecalis* may not have not incubated long enough before being examined for bacterial growth, which could result in the negative abnormalities.

No conclusive results were found for the association between artificial sweetener concentration and *E. faecalis* facilitated growth (Figure 5). Likewise, "Experiments with *E. coli* showed no significant change in normalized growth in response to sucralose or aspartame exposure at any time point or concentration," (Shil & Chichger, 2021). On the other hand, as the concentration of aspartame, saccharin, and sucralose increased, the growth of *E. coli* significantly increased (Figure 5). A 2022 study found that aspartame can significantly impact the growth of bacterial pathogens, such as *E. coli* (de Dios et al.). Other studies concluded that artificial sweeteners had no impact on individual bacterial growth, but many found that artificial sweeteners alter *E. coli* and *E. faecalis* biofilm production, which ultimately increases their presence in the microbiome.

Conclusion

Artificial sweeteners harm the body in a multitude of ways, including their obesogenic effects, disruptions to the gut microbiota, and risks for insulin resistance, cancer, cardiovascular diseases, cerebrovascular diseases, dementia, and kidney disease. Diabetics may use artificial sweeteners to control their sugar intake. Those who are health conscious may choose products with artificial sweeteners to avoid the calories of refined sugars. Nevertheless, many people consume artificial sweeteners daily whether it is intentionally or by accident. Artificial sweeteners are found in thousands of products, making them easily accessible to all ages. Unfortunately, the research that has been conducted on the effects of artificial sweeteners is minimal and often inconclusive. The goal of this study was to examine the results of artificial sweeteners on the body by performing a literature review, a minimum inhibitory growth concentration experiment, and a growth stimulation experiment. Results showed obesogenic effects were linked to artificial sweeteners due to the taste receptor-brain miscommunication, reduction of butyric acid, and increased risk for insulin resistance which could lead to type two diabetes. Next, disruptions to the microbiome conflict homeostasis, host nutrition, and development of the immune system. This results primarily in inflammation, which can increase the risks for heart disease, stroke, cancer, irritable bowl syndrome, rheumatoid arthritis, ulcerative colitis, Crohn’s disease, and other autoimmune diseases. Artificial sweeteners were seen to increase bacterial biofilm production which causes bacteria to have a higher antibiotic resistance and allows them to stick to the cell walls in the gastrointestinal tract. Not only does this fuel the world-wide problem of antibiotic-resistant bacteria, but it also plays a part in the disruption of the gut microbiota. The results of the experiments showed that aspartame, saccharin, and sucralose do not inhibit the growth of *E. coli* or *E. faecalis.* No conclusive results were shown for the effects of artificial sweetener concentration on *E. faecalis* growth, but as artificial sweetener concentration increased, the growth of *E. coli* significantly increased.

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