Food additives and microbiota

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ABSTRACT
The usage of food additives in food production is inevitable in this modern world. Although only safe amount of them are approved, their safety has always been questioned. The effects of food additives on microbiota have not been investigated in detailed manner up to know. In this review, the effects of artificial sweeteners, sugar alcohols, emulsifiers, food colorants, flavor enhancers, thickeners, anticaking agents, and preservatives on microbiota were reviewed. Even though, most of the results illustrated negative outcomes, few of them showed positive effects of food additives on microbiota. Although it is difficult to obtain exact results due to differences in experimental animals and models, it can be said that nonnutritive synthetic sweeteners lead to glucose intolerance by affecting microbiota and a part of sugar alcohols show similar effects like probiotics.

Keywords: Anticaking agents; emulsifiers; food additives; food colorants microbiota; flavor enhancers; glucose intolerance; synthetic sweeteners; sugar alcohols; preservatives; thickeners.

Food additives are used in food processing industry to improve color, taste, smell, nutritional value, and shelf-life of food products, which are indicated in labels as an “ingredients”. Several toxicologic investigations have been done before determining permission limits to use food additives in food products. Depending on investigations, safe quantities for animals are determined, and then, these results divided by 100 to obtain safety level for human, which is called acceptable daily intake (ADI). According to the ADI, the usage amounts of food additives are calculated. There is also a follow up part to evaluate ADI in case of adverse effects of food additives on humans. Based on this periodical follow up, ADI values of the additives can be reduced or the additives can be banned if there are any serious negative side effects. Therefore, food additives with permission are under medium safety level. Although approved food additives are assumed as safe, emerging of new techniques and research subjects indicate that some of them may have still health concerns.

In recent years, microbiota is getting attention of researchers. However, the relationship between microbiota and food additives is new subject to search. Human is a superorganism with 10% human and 90% microbial cells [1]. In the mean time, human and microbial genome develops together, thereby their metabolisms and sustainability mixes and becomes inseparable. Microbiota occurs by combination of bacteria, viruses, and some unicellular eucaryotes.

The gut microbiota genome codes more than 3.3 million genes and it is almost 150 times of human genome. In gut microbiota, there has been several species based on these 6 phylum of bacteria: Firmicutes, Bacteroidetes, Actinobacteria, Proteobacteria, Fusobacteria, and Verrucomicrobia. The intestinal microbiota has highly dynamic and variable structure depending on genotypes, geography,
life style, and age. These changes started at the first year since birth, reached to adults at age 2.5, and remained constant until senescence [2].

Gut microbiota is very important for human health. It regulates most of the physiological events. Gut microbiota locates in mucosa layer of intestine and gives shape. It plays an important role by helping digestion of pulp in food, synthesing of vitamins and amino acids, helping in energy metabolism and storage, regulating immune system, growing and developing nerve system, even regulating our behaviors [2].

In this article, food additives, which are grouped as artificial sweeteners, sugar alcohols, emulsifiers, food colorants, flavor enhancers, thickeners, anticaking agents, and preservatives, are reviewed based on their effects on microbiota.

**Sweeteners**

**A) Artificial sweeteners:**

Artificial sweeteners or non-nutritive sweeteners are known as low calorie and sweet taste compounds. Generally preferred by people who are on low calorie diet or have diabetes. In this part, artificial sweeteners such as saccharine, sucralose, aspartame, acesulfame-K, neosperidine DC, and splenda were evaluated. There have been few studies regarding with artificial sweeteners and mostly combined effect of two or more of them on microbiota are demonstrated. Therefore, instead of each artificial sweetener, studies are explained under separate titles.

**Saccharine (E954), Sucralose (E955), and Aspartame (E951)**

Sucralose, also called trichlorogalactosucrose, tastes 600–650 times sweeter compared to sugar. It has been used as a sweetener in energy reduced soups, jams, jellies, marmelades, breakfast cereals, and fruit concentrates. Saccharine is an artificial sweetener, which tastes 350 times sweeter than tea sugar. It has been used in food products such as chips, instant salep, carbonated drinks, flavored fruit juices, fruit nectars, and different diet food products. Basically, aspartame consists of two amino acids and one dipeptide and tastes 150–200 times sweeter than sugar. It is not only used in diet food products, but also nonalcoholic beverages, gelatin desserts, low calorie foods, gums and hot chocolates.

Suez et al. (2014) demonstrated that artificial sweeteners, including saccharine, sucralose and aspartame, induced glucose intolerance in rats, which was related to increase the number of *Bacteroides* spp. and bacteria in *Clostridiales* phylum in intestine. Thereby, alterations in intestinal microbiota resulted in glucose intolerance induction; non-caloric artificial sweeteners (NAS) did not affect glucose intolerance in germ-free mice and antibiotic – treated rats. In addition, their results indicated that NAS-mediated harmful metabolic effects can be removed with antibiotic therapy and there is a possibility to transfer harmful effects by transplantation of feces from mice given NAS or by administering anaerobically incubated microbiota under effect of NAS to microorganism free mice [3]. However, researchers reported that saccharine might affect gut microbiota negatively, and thereby, cause liver inflammation in mice [4]. Result of recent studies investigating the effect of sucralose on glucose hemostasis in humans are controversial. While Grotz et al showed that sucralose has no effect on glucose metabolism, Romo-Romo et al. revealed that sucralose affects negatively [5, 6].

**Aspartame (E951) and Acesulfame-K (E950)**

Acesulfame-K is an artificial sweetener, which tastes 200 times sweeter than sucrose. Generally it has been used in diet foods including bakery products, gums, desserts, and non-alcoholic beverages.

Researchers found that rats under low-dose aspartame diet gained less weight compared to rats under high fat and further into aslibitum water diet, however low-dose aspartame diet increased fasting glucose level and affected glucose accumulation based on insulin tolerance tests. Metabolites of aspartame into the short chain fatty acid propionate, a bacterial end product and highly gluconeogenic substrate might be the reason of insulin tolerance. Low-dose aspartame consumption increased total bacteria including Enterobacteriaceae and Clostridium leptum. Besides, combined high fat and aspartame diet consumption increased not only the amount of Roseburia ssp. but also Firmicutes/Bacteroidetes ratio [7].

In another study, the high intensity sweeteners – aspartame and acesulfame-K – were investigated to determine the modulation in gut absorption of sugars. Aspartame and acesulfame-K were provided to healthy volunteers for 4 days and bacterial community in fecal samples was analyzed by using multitag pyrosequencing on 5th day. Results indicated that consumption of aspartame and acesulfame-K did not increase bacterial abundance profiles and predicted gene function but they changed bacterial diversity [8]. Moreover, eight weeks of aspartame consumption changed the gut microbiota of
mice, thereby, increased the blood glucose level and affected the insulin resistance [9]. According to the study in Canada (2014), increase in blood glucose level and insulin resistance were determined in mice fed with aspartame due to changes in gut microbiota, which may cause type 2 diabetes and other illnesses [10]. Recently, there has been an increase in researches investigating relationship between artificial sweeteners and gut microbiota. According to the study, changes in gut microbiota and accordingly weight gain was observed in mice fed with acesulfame potassium for a month [11].

**Saccharine (E954) + Neosperidine DC (E959)**

Neosperidine DC is an artificial sweetener and 1000–1800 times sweeter than sucrose. It increases the efficacy of other artificial sweeteners when it is used together.

Feeding piglet with Sakkarine (E954) + Neosperidine DC (E959) increased the amount of *Lactobacillus* in fecal sample and lactic acid concentration in gut lumen, which indicated artificial sweetener may affect the gut microbiota as prebiotics [12].

**Splenda**

Splenda is a non-nutritive sweetener, consists of 1% w/w sucralose with glucose (1% w/w) and maltodextrin (94% w/w) as fillers. Research showed that it altered gut microbiota and increased weight gain in rats after 12 weeks of exposure [13].

**B) Sugar alcohols**

Sugar alcohols are organic compounds, group of polyols, and typically produced from sugars. The main characteristics of them are less digestible because they are not totally digested in small intestine and some of them are fermented in colon. Some of the sugar alcohols are used as food additives. In this part, maltitol, xylitol, sorbitol, and erithritol are evaluated.

**Maltitol (E965)**

Maltitol is 10–25% less sweet than sucrose. Since mouth bacteria are not fed by maltitol, it does not affect teeth rotting negatively and gives cool mouth feel. Energy value of maltitol is half of the sugar. Maltitol is used as a sweetener, humectant, tissue agent, bulking agent and stabilizer in gums, delights and halva products.

Addition of maltitol (E965) 22.8 g/day to the chocolate products increased the amount of *Bifidobacteria*. Combination of maltitol and polydextrose increased both *Bifidobacteria* and *Lactobacillus* concentration. Besides, it enhanced propionate and butyrate. Results indicated that maltitol was a fermentable product for this kind of microorganisms [14, 15].

**Xylitol (E967)**

Xylitol is a natural sugar alcohol that found in many plants. It is a low energy sweetener. Even though it has lower energy compared to sugar, it tastes similar. However, it gives cool mouthfeel. Bacteria in mouth do not use xylitol as an energy source, therefore xylitol do not cause teeth rotting and mostly preferred to use in gum production [16, 17]. It has been reported that xylitol is an eligible component of a diabetic diet [18]. and intake of it may be useful in preventing the development of obesity and metabolic disturbances in diet-induced obesity [19].

It has been used in desserts, candies, reduced sugar jams and marmelades, and some bakery products as a sweetener and a humectant. In addition, it has been preferred to use in gum because it reduces teeth rots [16].

Xylitol (E967) is affecting intestinal microbiota. Xylitol consumption is shifted rodent intestinal microbial population from gram negative to gram positive bacteria [20]. The effect of xylitol on isoflavonoid of daidzein metabolism and mice intestinal microbiota was observed. Addition of xylitol to daidzeine decreased plasma cholesterol level, increased equol in urine and fecal lipids. Researchers found that the amount of *Bacteroides* was higher in groups feeded by xylitol compared to xylitol and daidzene. As a result, there has been potential effect of xylitol on daidzeine metabolism via changing the metabolism activity of intestinal microbiota [21].

**Sorbitol (E420)**

Sorbitol is a sugar alcohol, which naturally exists in fruits, has a similar structure to sugars. It is obtained from glucose and fructose after several chemical treatments and its taste is at least half less sweet compared to regular sugar. It is used in confectionaries, bakery products and low calorie foods and gums as a humectant, sweetener, texturizer, bulking agent, and binder. Bacteria in the mouth are not able to use sorbitol as a nutrient source, for this reason, it is used as a sweetener and preventing growth of bacteria, which is essential to mouth and teeth health.

Sorbitol is used by some *Lactobacillus* species [22] and used as carbon source by human intestinal *Bifidobacteria* [23]. For this reason, some researchers illustrated that sorbitol is prebiotic [24]. In addition, few in vivo studies indicated that sorbitol has a potential prebiotic effect. Microbial population in rat fed by sorbitol shifted...
from Gram negative to Gram positive [25].

Sarmiento-Rubiano et al. reported that sorbitol increased number of Lactobacillus reuteri and helped Lactobacillus sp. AD102 survival. Rats fed with sorbitol had high butyrate level, however acetate/propionate level was low in colon and caecum. Total, HDL, and LDL cholesterol levels were lower in those who consumed sorbitol, and researchers suggested that this may be due to the low ratio of acetate/propionate [26].

**Erythritol (E968)**

Erythritol is a sugar alcohol and widely found in nature. Commercially, it is obtained from glucose by using osmophilic yeasts [27]. It can be used in cheese products, milk powder, desserts made with milk, ice cream, breakfast cereals, processed meat products, desserts made with egg, sauces as a sweetener.

Oral microorganisms do not metabolize erythritol. Ninety percent of erythritol is absorbed in small intestine by passive diffusion and distributed to other tissues. It is minimally metabolized in the body and most of them excreted with urine [28]. It does not affect glucose and insulin levels [29, 30].

Less amount of erythritol is not absorbed thereby passes through colons and affected by microbiota fermentation. One of the study indicated that only 10% erythritol was suitable for fermentation on rats [31].

Arrigoni et al. (2005) searched erythritol metabolism in human microbiota in vitro conditions. Fresh human intestine microbiotas from three volunteers were incubated with erythritol for 24 h. They evaluated total gas production, hydrogen gas accumulation, pH changes, short chain fatty acid production, and erythritol degradation. No gas or fatty acid production was observed. After fermentation, polyol was regained. With these results researchers have concluded that erythritol is not fermented [32].

**C) Emulsifiers**

Emulsifiers have similar effects with detergents due to chemical structure, which consists of homogeneous mixture of fat and water based materials. There have been several emulgators produced with natural and artificial ways. In this section, carboxymethyl cellulose and polysorbate 80 are evaluated.

**Carboxymethyl cellulose (E466) and Polysorbate 80 (E433)**

Carboxymethyl cellulose is modified cellulose. It is obtained from its reaction with acetic acid derivatives. It is used as a stabilizator, thickener, and suspension agent in powder form of drinks, fruit yogurts, whipped creams, sauces, diet food products, and ice creams. Polysorbate 80 is a synthetic emulgator, produced by using fatty acids and ethylene oxide. It is used as an emulgator in candies, desserts, dairy products, soups, gums and special diet products. Chassaing et al. (2015) illustrated excessive increase in Ruminococcus gnavus and decrease in Bacteroidales in rats fed with carboxymethyl cellulose (E466) and polysorbate 80 (E433) for 12 weeks. Besides microbial changes, there were metabolic syndrome symptoms like intestinal mucus density, low level of inflammation and fat deposition and disorder in glucose metabolism. Emulgator has no negative effects on germ free mice. These changes occurred by transferring microbiota from mice fed with emulgators to germ free mice. On the other hand, emulgators increased resistant colitis in IL-10-deficient and Toll-like receptor 5 (TLR5)-deficient rats. According to this result, researchers found emulgator enhanced colitis and induced low level inflammation whom has emulgator intolerance [33].

Singh et al. found that glicemic tolerance disorder occurred, blood insulin level increased, hepatic enzyme level enhanced, hepatic mitochondria and gall bladder increased in rats fed with polysorbate 80. Acetate, propionate and butyrate level was low in rat fecal. High level of DCA and low level of Muc2 RNA expression were occurred in intestinal mucus, also decrease in mucus thickness and increament in intinstine permability were observed. In addition, intestinal bacteria were in deep part of the mucus and close to intestinal epithel thereby bioactive LPS, flagellin levels and LCN2 expression enhanced. Results indicated that there is a relationship between emulgator such as polysorbate 80 and obesity related intestinal inflammation and liver disfunction thereby supported changes in gut microbiota [34].

Other study showed that polysorbate 80 changed bile acid level and then affected microbiota composition [35].

**D) Food colorants**

There has been several food colorants with oil based and water based properties. In this section, two food colorants are evaluated in nanoparticle structures: silver and titanium dioxide.

**Silver (E174) and Titanium dioxide (E171)**

Silver is a grey color natural metal and obtained from silver gem. It is used as a colorant in candy and chocolate surface coating. Titanium dioxide is a natural white min-
eral. It is produced from natural sources by using chlorine and sulphuric acid in chemical reactions. It is used as colorant in gums, chocolates, candies, flavored fruit juices, and some dishes made with yogurt (e.g. haydari).

The effect of silver on gut microbiota was discovered for experimental models with different sizes of silver particles. When zebrafish was fed with 60 nm and 500 mg/kg silver containing food, richness and variety of microbiota didn’t change. In other study, mice fed with size of 14 nm and 4.5 or 9 mg/kg for 28 days, however there was no change in amount of Firmicutes or Bacteroidetes in caecum. Similar result was determined in mice fed with 110 nm and 10 mg/kg for 28 days. There was no change in membership, structure, and diversity of microbiota. In all particles with 10, 75, 110 nm and 9, 18, 36 mg/kg silver increased gram negative bacteria, and in 10 nm particle decreased Firmicutes (Lactobacillus) in ileum. Twenty eight days of exposure to silver (46, 460 and 4600 µg/L) caused reduction of bacterial richness, increase in dose dependent Firmicutes and decrease in Bacteroidetes. Faecal mixture from 33 healthy people were on anaerobic fermentation in vitro in silver nanoparticles environment and researchers observed that change in bacteria population such as fatty acids [36].

There have been several studies related to titanium dioxide in vitro. These are applied in dark environment while titanium dioxide nanoparticles are activated with UV light to kill bacteria. In acidic environment without UV light, titanium dioxide affects the bacteria surface; thereby, electrostatic interaction occurs and then inhibits E. coli cell division [37].

Gut microbiota from healthy donors were treated with 3 mg/L titanium dioxide for 5 days in dark environment of colon model. Researchers observed phenotypic changes in short fatty acid production such as butyric acid, cell hydrophobicity, sugar ingredient of extracellular polimeric substrate, cell enlargement and electrophoretic mobility of microbiota [38]. Treatment of E. coli with titanium dioxide in dark environment damaged lypopolysaccharides and decreased membrane fluidity [39]. In other study, with same conditions, reactive oxygen variety production and glutation level were decreased, thereby oxidative stress resulted with lipid peroxydation and DNA damage [40].

E) Flavor enhancers

Flavor enhancers are used in a wide range of food types in order to reveal the flavor in foods. Monosodium glutamate is a well-known common used flavor enhancer and evaluated in this section.

Monosodium glutamate (E621)

Monosodium glutamate (MSG) is used as a flavor enhancer in some food products such as meatball mixtures, chicken pane mixtures, and meat bouillon. There are still continuing debates about MSG consumption and obesity [41].

Feng et al. investigated effects of MSG and/or fat on gut microbiota. They fed 32 growing pigs with 3% MSG basal food for 30 days and then evaluated jejunium, ileum, cekum and colon ingredients. MSG modified gut microbiota diversits specifically in colons and increased gut microbiota variety. MSG and fat promoted the colonization of microbes related to energy extraction in gastrointestinal tract. MSG helped colonization of microbes such as Faecalibacterium prausnitzi and Roseburia.

MSG and fat consumption increased fat accumulation at the back muscle called longissimus dorsi. Consumption of both MSG and fat synergistically enhanced fat accumulation. MSG helped colonisation of microbes thereby it is consistent with fat deposition in muscles [41].

F) Thickeners

Thickeners are substances, which can increase viscosity of liquids without substantially modifying their other properties. In this part, thickeners -pectin, polydextrose and alginic acid- are evaluated.

Pectin (E440)

Pectin is a plant based natural thickener and commercialy produced from orange peels, apple sediments, and beet pulps. It is used as a thickener and emulgator in food products such as ice cream, jams, marmalades, candies, some beverages, cheese, salep, and yogurt.

Apple derived pectin (E440) decreased weight gain and total cholesterol level when consumed with fatty foods. Fatty foods reduce amount of Bacteroidetes phylum and increase amount of Firmicutes phylum, however pectin addition normalizes these variables. Results indicated that consumption of pectin with fatty foods caused remission of intestinal inflammation, and then, improved intestinal barrier functions [42].

Polydextrose (E1200) (PDX)

Polydextrose (E1200) produced synthetically from glucose and sorbitole (E420(i)) by heating with citric acid (E330). It consists of 90% glucose, 10% sorbitole, 1% citric acid and 0.1% phosphoric acid. Generally it is used as
bulking agent, viscosity enhancer, humectant, and stabilizer in cookies and halva products.

Polydextrose has prebiotic function due to change of composition and activity of gut microbiota and it improves intestine functions [43, 44]. Polydextrose is slowly fermented, probably used in distal colon, which will have positive effect for distal colon illnesses.

Twelve gram of polydextrose affected fecal anaerobes. Species of Bacteroides (B. fragilis, B. vulgatus, and B. intermedius) decreased, Lactobacillus and Bifidobacterium species increased. Fecal weight (wet and dry) and short chain fatty acids (especially: butirate, iso-butirate and acetate) increased but pH decreased [43].

When amount of polydextrose is 8 g, there was no change observed in fecal weight, short chain fatty acid concentrations such as propionate and acetate, fecal lactic acid bacteria and bifidobacteria ingredients. Orofecal transit time shortened, pH decreased, gall bladder acidity and neutral sterol existence has been varied [44].

Alginic acid (E400)
Alginic acid is hydrophilic, colloidal and naturally oc- curred polysaccharides. It is obtained from some types of sea weeds (Phaeophyceae). It has been used as stabilizer, viscosity enhancer, gelator, and emulgater in food products such as jam and jelly.

Gut microbiotas from 6 Chinese volunteers were completely fermented by using different concentrations of Alginic acid. When compared to control group, pH value of fermentation with alginic acid decreased. Fermentation bacterias are Bacteroides ovatus, Bacteroides xylanisolvens, and Bacteroides thetaiotaomicron. During this process, free fatty acids such as acetic acid, propionic acid, and butyric acid are increased [45].

G) Anticaking agents
Anticaking agents prevent powdered or crystallized form of foods such as flour and salt from aggregating and agglomerating to maintain a free flow. One of the anticaking agents, bentonite, is evaluated in this part.

Bentonite (E558)
Bentonite, a colloidal and hydrated aluminium ciliate, is obtained from natural clay varieties. It can contain different amounts of iron and some alcalic materials in commercial forms. It is used in food industry to prevent agglomaration.

In pet food, it is used for stabilizator, lubricant, etc. It extends travel time in intestine of fowl thereby increase effectiveness of feed [46]. It decreases negative effects of aflatoxin [47].

When chickens were fed with bentonite, it increased egg number and size. Food additives did not affect the microorganism populations negatively in terms of richness and variety. Food additives reduced potential pathogenic bacteria and missing parts were determined in order Campylobacterales [48].

H) Preservatives
Preservatives are used to maintain an existing condi-
### TABLE 1. The effects of some food additives on microbiota

<table>
<thead>
<tr>
<th>Sweeteners</th>
<th>Effect</th>
<th>Species</th>
<th>Increase/Decrease</th>
</tr>
</thead>
<tbody>
<tr>
<td>Artificial sweeteners</td>
<td></td>
<td>Rats</td>
<td>increased the number of Bacteroides spp. and bacteria in Clostridiales phylum [4]</td>
</tr>
<tr>
<td>Saccharine (E954), Sucralose (E955), Aspartame (E951)</td>
<td>induced glucose intolerance</td>
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<tr>
<td>Aspartame (E951)</td>
<td>increased fasting glucose level and affected glucose accumulation</td>
<td>Rats</td>
<td>increased total bacteria including Enterobacteriaceae and Clostridium leptum [5]</td>
</tr>
<tr>
<td>Aspartame (E951)</td>
<td>determine the modulation in gut absorption of sugars as prebiotics</td>
<td>Healthy volunteers</td>
<td>Not increase bacterial abundance profiles and predicted gene function</td>
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<tr>
<td>Neosperidine DC (E959)</td>
<td>increased weight gain</td>
<td>Piglet</td>
<td>increase the amount of Lactobacillus</td>
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<tr>
<td>Splenda</td>
<td></td>
<td>Rats</td>
<td>increase lactic acid concentration [6]</td>
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<tr>
<td>Sugar alcohols</td>
<td></td>
<td>Rats</td>
<td>altered gut microbiota [7]</td>
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<tr>
<td>Maltitol (E965)</td>
<td>was associated with a decrease of dry matter amount</td>
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<td>Xylitol (E967)</td>
<td>(Addition of xylitol to daidzeine) decreased plasma cholesterol level, increased equol in urine and fecal lipids</td>
<td>Mice, Rats</td>
<td>shifted rodent intestinal microbial population from gram negative to gram positive bacteria [12, 13]</td>
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<td>Sorbitol (E420)</td>
<td>had high butyrate level, however acetate/propyionate level was low in colon and caecum</td>
<td>Rats</td>
<td>shifted from Gram negative to Gram positive [17]</td>
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<td>Erythritol (E968)</td>
<td>does not affect glucose and insulin levels</td>
<td>Healthy volunteers</td>
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<td>Emulsifiers</td>
<td></td>
<td>Rats</td>
<td>excessive increase in Ruminococcus gnarus and decrease in Bacteroidales [25]</td>
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<td>Carboxymethyl cellulose (E466) and Polysorbate 80 (E433)</td>
<td>low level inflammation and fat deposition and disorder in glucose metabolism</td>
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<td>Food colorants</td>
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<td>Silver (E174) and Titanium dioxide (E171)</td>
<td>damaged lypopolysaccharides and decreased membrane fluidity</td>
<td>Healthy volunteers</td>
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<td>Flavor enhancers</td>
<td></td>
<td>Pigs</td>
<td>helped colonization of microbes such as Faecalibacterium prausnitzii and Roseburia [33]</td>
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<td>Monosodium glutamate (E621)</td>
<td>increased fat accumulation</td>
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<td>Thickeners</td>
<td></td>
<td>Rats</td>
<td>Fatty foods reduce amount of Bacteroides phylum and increase amount of Firmicutes phylum</td>
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<td>Pectin (E440)</td>
<td>decreased weight gain and total cholesterol level when consumed with fatty foods</td>
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<td>however pectin addition normalizes these variables [34]</td>
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<tr>
<td>Polydextrose (E1200)</td>
<td>short chain fatty acids (especially: butirate, iso-butirae and acetate) increased but pH decreased</td>
<td>Healthy volunteers</td>
<td>affected fecal anaerobes. Species of Bacteroides (B. fragilis, B. vulgatus, and B. intermedius) decreased, Lactobacillus and Bifidobacterium species increased [35, 36]</td>
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<td>Alginic acid (E400)</td>
<td>free fatty acids such as acetic acid, propionic acid, and butyric acid are increased, pH value of fermentation with alginic acid decreased</td>
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<td>Bentonite (E558)</td>
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<td>Preservatives</td>
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<td>Pigs</td>
<td>reduced coliform and lactic acid bacteria and changed microbiota in</td>
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<td>Benzoic acid (E210)</td>
<td>total and branched chain fatty acids were decreased</td>
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tion or prevent damage likely to be brought by chemical (oxidation), physical (temperature, light) or biological (microorganisms) factors. Benzoic acid is one of the preservative types, considered in this part.

**Benzoic acid (E210)**

Benzoic acid, is a bacteriostatic agent, syntetically obtained and used as an antimicrobial preservative. It is preferred in a wide range of foods such as sauces, pickles, acidic fruit juices, dried fruits, salty margarine, fruit and vegetables salads, sugary creams, and gums.

Yousaf et al. showed that benzoic acid reduced coliform and lactic acid bacteria and changed microbiota in pigs. The amount of E. coli in young and gram negative bacteria in adult decreased. High amounts of benzoic acid decreased growth rate (0.5–0.75%) in fowl, also ileal coliforms reduced but caecal lactic acid bacteria increased. 0.1% ratio increased growth performance however 0.2% ratio decreased. Same study also indicated that 0.2% ratio increased lactic acid bacteria in ileum and both 0.1 and 0.2% ratio decreased coliform bacteria. In different parts of intestine different short chain fatty acid profile was observed. Benzoate did not affect the pH. Lactate in craw, D-lactate in jejunium was enhanced. Caecal total was observed. Benzoate did not affect the pH. Lactate in craw, D-lactate in jejunium was enhanced. Caecal total was observed. Benzoate affected the gut microbiota positively by increasing lactic acid bacteria [49].

The effects of food additives on microbiota are summarized in Figure 1 and Table 1 below.

**Conclusion**

In literature, there are not many studies related to the effect of food additives on microbiota. Even though, most of the results illustrated negative outcomes, few of them showed positive effects of food additives on microbiota. Besides that artificial sweeteners destroy glucose tolerance and support weight gaining by affecting microbiota negatively. Most of the sugar alcohols are fermentable by bacteria and may show similar properties with probiotics. Due to differences in experimental animals and models, there is not exact result obtained. For this reason, more studies are needed to evaluate the effect of food additives on gut microbiota.

**Conflict of Interest:** No conflict of interest was declared by the authors.

**Financial Disclosure:** The authors declared that this study has received no financial support.

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**Authorship Contributions:** Concept – FG, HBS; Design – FG, BD; Supervision – FG, MEO; Materials – MEO, HBS; Data collection and/or processing – BD, MEO; Analysis and/or interpretation – FG, HBS; Literature review – MEO, HBS; Writing – BD, HBS, MEO, FG; Critical review – FG, BD.

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