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Kombucha A Functional Beverage for Heart, Gut, Mind and Healthier Lifestyle



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ABSTRACT

Objective: This article aims to explore the structural, microbiological, biochemical, and therapeutic aspects of Kombucha as a functional fermented beverage that promotes heart, gut, and mental health and contributes to an overall healthier lifestyle.

Methods and Materials: This review-based study synthesizes current scientific literature on Kombucha tea, emphasizing its microbial composition, fermentation mechanisms, and resulting bioactive compounds. Data from multiple in vitro, in vivo, and experimental studies are analyzed to assess Kombucha's health-promoting functions, its application in sports nutrition, its therapeutic effects, and its role in environmental detoxification. Furthermore, this study details the standard preparation processes, the influence of fermentation variables, and alternative substrates for enhanced production.

Findings: Kombucha exhibits significant antioxidant, antimicrobial, and probiotic properties. It contributes to post-exercise recovery, gut microbiome balance, detoxification, and cardiovascular health. Its bioactive compounds, including polyphenols, organic acids, and B vitamins, provide potential benefits in managing diabetes, hyperlipidemia, digestive disorders, and certain types of cancer. Additionally, Kombucha demonstrates hepatoprotective and anti-inflammatory effects. The beverage also shows promise as a bioabsorbent for removing heavy metals (e.g., Pb²+, Ni²+, Cd²+) from wastewater. Kombucha's bacterial cellulose layer has potential in medical and cosmetic applications, including burn treatment and skincare. The quality and efficacy of Kombucha are influenced by variables such as fermentation time, temperature, pH, substrate type, and oxygen availability.

Conclusion: Its bioactive profile, adaptability to different substrates, and potential for therapeutic and environmental applications make it a valuable functional beverage.

Keywords: Kombucha, fermented drink, Nutritional and Chemical Characteristics, Health aspects, process variables, production

1. Introduction

1.1 History of Kombucha

Month is a sweet-and-sour fermented drink that originated in Manchuria and spread across China (Ancient Beginnings 500+ BCE). Chinese introduced fermented guava drink to Koreans and Russians. By 414 AD, a Korean physician named Kambo reportedly used a fermented mushroom remedy for Japan's Emperor Inigiwa, leading to its Japanese name, kombucha ("tea from the sea" or "Kambo's tea"). Japanese soldiers drank it for energy and carried it into battle. In Asia, kombucha was brewed in homes and sacred spaces, shared as gifts (not sold) due to religious taboos. European adoption surged during tea/sugar shortages, leading to mass production. Medical validation spurred global demand, with factories emerging in Europe/America.

Like yogurt and vinegar, kombucha is a symbiotic culture of bacteria and yeast (SCOBY) fermenting sweet tea, enhancing flavor and digestibility. It has been called "tea mushroom," "Manchurian tea," or "magic mushroom" worldwide. Gifting kombucha symbolized affection and well-being (1-9).

1.2 Kombucha as a recommended drink for athletes

1.2.1 Antioxidant Properties and Oxidative Stress Reduction

Kombucha and kefir as fermented beverages are gaining attention in sports nutrition due to their potential benefits for athletes, including enhanced recovery, gut health, and antioxidant properties. This section possesses a detailed analysis of their roles in sports performance Kombucha, especially when made with green tea, contains high levels of polyphenols (e.g., catechins) and flavonoids, which scavenge free radicals generated during intense exercise, reducing oxidative stress and muscle damage. Antioxidant activity increases during fermentation compared to unfermented tea, aiding post-workout recovery by mitigating inflammation and delaying muscle fatigue. Kefir also exhibits antioxidant effects.

Gut Health

The live probiotics in kombucha (e.g., Lactobacillus, Bifidobacterium) may improve gut microbiome balance, which is often disrupted by intense exercise. A healthier gut enhances nutrient absorption, immunity, and reduces gastrointestinal distress during training. Probiotics in fermented drinks may also modulate the gut-brain axis, potentially improving mood and motivation for training.

Post-Workout Recovery

Organic acids (e.g., glucuronic, acetic, lactic) in kombucha help detoxify lactic acid buildup, reducing DOMS (Delayed Onset Muscle Soreness) and speeding recovery. Also, B vitamins (e.g., B_1 , B_6 , B_{12}) in kombucha support energy metabolism. On the other hand, electrolytes e.g., sodium, potassium of kombucha are less tha sports drinks but contribute to rehydration when combined with water.

Energy and Endurance

Kombucha's low sugar content (if unsweetened) makes it a healthier alternative to sugary sports drinks, avoiding energy crashes. Kombucha's caffeine (from tea) and theobromine may provide mild stimulant effects, though evidence is limited.

Risks and Considerations

Besides all health-beneficial properties, overconsumption may cause bloating or acidity due to probiotics and acetic acid. Alcohol content (up to 0.5% ABV in commercial kombucha) could be a concern for athletes avoiding alcohol. Dental erosion due to acidity is another risk; so rinsing with water post-consumption is advised (10, 11).

2. Properties and constituents of Kombucha

Kombucha is a non-dairy probiotic beverage which is partially carbonated due to fermentation process. It is traditionally made by fermenting sweetened tea utilizing a symbiotic culture of bacteria and yeasts. The symbiotic culture of bacteria and yeast (SCOBY) known as Kombucha, or tea mushroom, is composed of two distinct components: a cellulose layer that floats on the surface and a fermented, sour liquid beneath it (Figure 1).







Figure 1. Kombucha solution (Kombucha liquid below and cellulose layer floating on top of the solution)

2.1 Cellulose layer of kombucha and its formation steps

The cellulose layer of kombucha is composed of a hemopolysaccharide known as cellulose, which is made up of beta-D-glucose monomers with beta 1 to 4 glycosidic bonds. Cellulose represents the predominant polymer in the natural environment. This naturally occurring polymer is insoluble in the most of solvents. The tensile strength of cellulose fiber is comparable to that of a steel thread with the same diameter. Kombucha tea contains a significant amount of cellulose, the principal substance produced by aerobic acetic acid bacteria in the process of fermentation. The predominant bacterium found in Kombucha is Acetobacter xylinum, a subspecies of Acetobacter stearothermophilus. The bacterium synthesizes cellulose sugar polymer through a series of reactions as depicted in Figure (2) and subsequently excretes it into the extracellular environment via gaps in its cell wall. The acetic acid bacteria are situated within the suspended layer of cellulose tissue in Kombucha, protecting the bacteria and yeasts within this consortium (12, 13) from the highly acidic environment produced by acetic acid and elevated concentrations of ethanol. Due to the low volume mass and light nature of cellulose fibers, they exhibit upward movement within the Kombucha liquid upon secretion, subsequently coalescing at the liquid's surface to form a cellulose disk. The microbial activities of these bacteria are characterized by the synthesis of microbial cellulose and the formation of biofilm on the liquid surface as a consequence of their metabolic processes. The procedure involves the formation of uridine diphosphoglucose, a key precursor for cellulose synthesis. Subsequently, each unit of Acetobacter cell catalyze the

polymerization of over 200,000 remaining glucose molecules per second, yielding beta chains comprised of 1 to 4 glucans. One of the primary benefits of this method of cellulose production lies in the rapid growth of the bacterium under carefully regulated environmental conditions, enabling the production of cellulose from a range of carbon sources, including glucose, ethanol, sucrose, and glycerol. The extracellular production of a suspended layer of microbial cellulose occurs via the attachment of fibrils to the bacterial cell. Each cellular unit contains 50-80 complex terminals (CTs) with a 3.5 nm diameter, to remove cellulose from the cell membrane. The chains are subsequently joined together to produce larger fibrils known as macrofibrils, which possess a three-dimensional configuration comprised of approximately 1000 individual glucan chains and can retain up to 200 times more water. During the process of fermentation, the membrane exhibits increased thickness due to the proliferation of new generations on its surface. This results in the formation of a suspended structure in the culture medium, as well as the expansion of the biofilm, facilitated by the presence of hydrogen and C-H bonds. Subsequently, the bacteria inhabiting this inactive layer experience oxygen deprivation, leading to their inactivation. The residual bacteria in the liquid phase remain in a dormant state which can be applied as next inoculaa. To enhance the efficiency of the process, several factors must be carefully evaluated. Podolich (2016) identified multiple factors that may influence the production of Kombucha mushrooms, including the volume of the inoculated starter, the duration of incubation, and the surface area and thickness of the cellulose layer in the supernatant (14). Furthermore, Kombucha's cellulose layer contains a small amount of sphingolipid, in addition to polysaccharides.





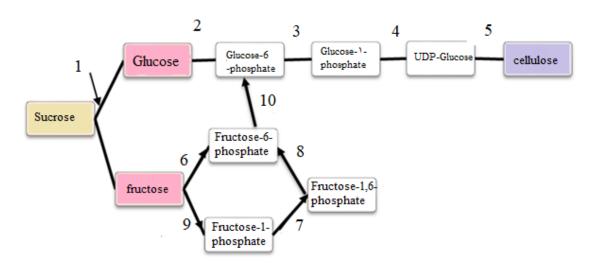


Figure 2. The process of cellulose production from glucose and fructose sugar units using Acetobacter xylinium bacteria. The enzymes involved in the catalysis of the steps indicated in the diagram are as follows: 1 - Invertase, 2 - Hexokinase, 3 - Mutase, 4 - Transferase, 5 - Cellulose synthetase, 6 - Hexokinase, 7 - Phosphofructokinase, 8 - Phosphatase, 9 - Fructokinase, 10 - Isomerase.

2.2 Benefits of Kombucha Cellulose Layer

The human digestive system lacks the enzyme for catalusis of cellulose, rendering cellulose nutritionally insignificant for humans. The consumption of Kombucha cellulose discs, while not inherently appetizing, is nonharmful and potentially beneficial in alleviating constipation. Additionally, the study demonstrates that the well-known bacteria A. xylinium, commonly found in Kombucha and vinegar, can produce different polysaccharide comprised of glucose acetylglucosamine units. The cellulose produced by Acetobacter cilitum bacteria exhibits a high level of purity. The substantial level of purity exhibited by the surface disc cellulose has stimulated interest in its potential application as an artificial skin for treating burns within the medical field. Due to its capacity to retain 148 times its weight in water, the cellulose layer has been suggested as a viable candidate for the development of artificial skin in burn treatments or post-surgical reconstruction. It is anticipated that in forthcoming studies, Kombucha will be explored for its potential applications in fabric, paper, and synthetic leather production. Figure 3 presents a close-up image of the Kombucha cellulose plate, while Figure 4 depicts a microscopic photograph of the cellulose layer.

The cellulose layer in Kombucha is crucial for microorganisms like yeasts and bacteria to form colonies on its lower surface. Thus, one of the primary functions of the disc or cellulose matrix is to facilitate aeration for the microbes, ensuring they have access to the necessary oxygen. The cellulose disk promotes bacterial growth by helping them form microbial biomass and giving them a competitive edge in acquiring nutrients over other microorganisms. The cellulose disc's lack of transparency acts as a shield against the harmful effects of ultraviolet rays, protecting the microorganisms beneath it from mutationThe cellulose matrix present in the liquid phase of Kombucha serves a vital function in protecting against mutations induced by ultraviolet radiation and supporting the viability of the microorganisms below. The utilization of Kombucha cellulose layer yeasts to derive amino acids from the nitrogen present in tea is significant for providing a valuable source of protein, energy, and B vitamins for monogastric animals and ruminants. The utilization of Kombucha biofilm extends beyond its application as an animal feed supplement. Temporary skin substitutes are utilized in various applications such as food packaging, biotechnology, and medical fields for the treatment of skin wounds, including burns, tissue transplants, and as a supplementary treatment for skin injuries. The utilization of dried tea mushrooms as an absorbent to remove heavy metals has been demonstrated. The use of Kombucha, whether consumed or applied topically, has been shown to promote healthy skin and improve its appearance, specifically through enhancing freshness. One method of incorporating Kombucha into skincare involves blending a cellulose disc into a skin cream, followed by application onto the skin and subsequent rinsing



with lukewarm water after a brief period. Cellulose discs have the potential to be utilized for the treatment of burns and other dermal injuries. Live and active yeast is a popular component in expensive and top-notch skincare formulas. Hence, it is plausible that Kombucha yeast exerts a superior and more efficacious influence on the rejuvenation and robustness of the skin (15).



Figure 3. Cellulose plate floating on Kombucha tea

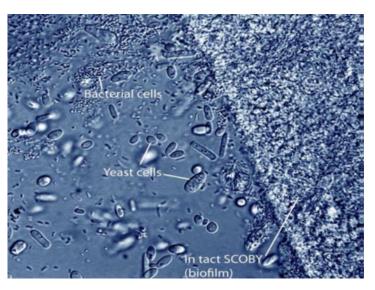


Figure 4. The image shows the Kombucha cellulose layer and demonstrates the cohabitation of yeasts and bacteria within the symbiotic culture of bacteria and yeast (SCOBY).

3. Microbiology of Kombucha

The fermenting of sweetened tea by a symbiotic culture of bacteria and yeasts results in the creation of Kombucha, a fermented beverage. This non-dairy, partially carbonated drink is known for its probiotic and beneficial properties. The metabolic processes and microbial interactions of Kombucha are depicted in figure 5. The Kombucha mushroom is a symbiotic aggregate consisting of smophilic

yeasts and bacteria, predominantly of the genus *Stobacter*, manifesting as a flat, smooth, and slimy structure. Through the fermentation process, successive layers are formed on the plate, each of which can be differentiated from the preceding layer. The initial placement of the mushroom on the tea's surface is in the form of a thin sheet, after which it undergoes a transformation to assume a thicker consistency.



3.1 Bacteria in Kombucha

Various types of bacteria, including Asetobacter xylinium, Asetobacter gelnoides, Asetobacter Asetei, Asetobacter pastorianum, Bacterium gluconicum, and yeasts such as Zygosaccharomyces bieli, Saccharomyces cerevisiae, Schizosaccharomyces pombe, Saccharomyces ludoigii, Brettanomyces brusselsensis, Candida kefir, and Pichia species, have been identified and isolated from the biomass.

Also, the presence of *Acetobacter aceti*, a bacterium that thrives in alcoholic environments and utilizes simple sugars such as glucose as its primary energy source, has been recognized as crucial in the fermentation process of Kombucha. Furthermore, it can derive energy from ethanol in the presence of acetic acid. The essential B vitamins supplied by yeast are required for proper physiological functioning. The microorganism *Acetobacter xylinum* plays a crucial role in the formation of the predominant layer of the Kombucha surface disc. The bacterium exhibits an oval to elongated shape and thrives in an environment rich in

ethanol. Furthermore, it can transform ethanol into acetic acid and metabolize acetate and lactate into carbon dioxide and water. The cellulose disc in both vinegar and Kombucha is synthesized by identical strains of bacteria. Bacterium gluconicum is a bacterium characterized by its spherical to elongated shape and typically possessing three to eight terminal flagella. At the temperature range of 25-30 °C, the conversion of ethanol to acetic acid occurs optimally. Furthermore, it plays a significant role in the conversion of glucose to glucuronic acid (16). The conversion of glucose into glucuronic acid is facilitated by both a specific bacterium and the yeast known as Schizosaccharomyces pombe. Weissella confusa has demonstrated the capability to thrive in an environment with a temperature of 45° C. Weissella is a bacterium with a Gram-positive cell wall structure belonging to the family Leuconostocaceae. Various strains of Wisella, such as Weissella confusa, possess probiotic properties. The cell-free supernatant of Weissella confusa demonstrates a multitude of advantageous qualities, including antibacterial attributes and antiinflammatory efficacy.

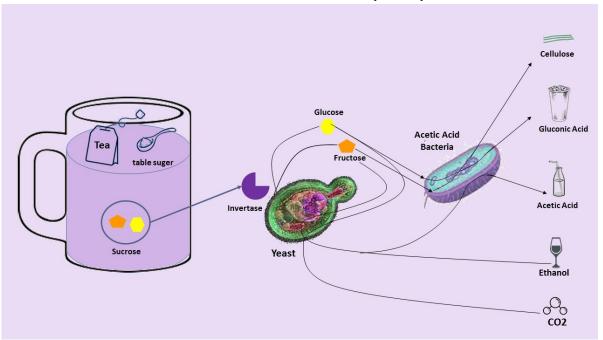


Figure 5. Kombucha metabolism and microbial interactions

3.2 Yeasts in Kombucha

A key area of focus in Kombucha research is the role of yeast in fermentation and its impact on metabolic processes and microbial interactions. Figure 6 displays a microscopic image of a selection of yeast present in Kombucha. Several yeast species found in Kombucha and their respective characteristics are as follows: *Pichia fermentans*, which exhibits false hyphae formation and produces hat-shaped spores. The rapid fermentation of glucose by this organism is accompanied by an infrequent occurrence of its growth





and multiplication in acidic environments with a pH of 1.5. Additionally, this strain of yeast is capable of lactic acid production. Various food items such as Italian cheese, spoiled oranges, California olives, kefir, and Indonesian fermented cocaine are additional sources of this particular yeast. The yeast Saccharomyces loidigii is characterized by the presence of lemon-shaped or elongated cells. The mycelium of these yeasts exhibits minimal growth, thereby indicating a false appearance. These phenomena are responsible for the formation of deposits and rings. The yeast demonstrates the capacity to ferment glucose, raffinose, and sucrose. The micro-organisms exhibit a high sensitivity to sunlight, and consequently, the presence of solar rays significantly inhibits their growth. The yeast strain Saccharomyces pombe is characterized by its spherical, oval, or cylindrical cellular morphology, and its ability to produce sediment. The microorganisms engage in fermentation of the sugars glucose, sucrose, maltose, and raffinose. Most members of this genus can break down complex sugars, particularly larger oligosaccharides. Other potential sources of this yeast include molasses, grape juice, sugar cane, and

apple (17, 18). The yeast Schizochromyces pombe is distinguished by its high fermentation capacity, enzymatic conversion of malic acid to ethanol, and substantial polysaccharide production. Saccharomyces cerevisiae exhibits notable resistance to ethanol. This yeast exhibits rapid fermentation, as well as being unaffected by variations in temperature and substrate concentration. Brettanomyces brusselsensis demonstrates notable resistance to osmotic pressure and ethanol, as well as a higher nitrogen fixation capacity in comparison to Saccharomyces cerevisiae. Additionally, the yeast has a proclivity for consuming sucrose and generating elevated levels of acetic acid in aerobic environments. Hanseniaspora ovarum is frequently observed in mature fruits, notably in grapes, and within the fermentation microbiome. Subsequent investigations have indicated that Hanispora ovarum has been detected in fermented beverages, including apple cider vinegar and kombucha. Hanseniaspora ovarum is recognized for its potential as a biocontrol agent in mitigating the impact of plant pathogenic fungal molds

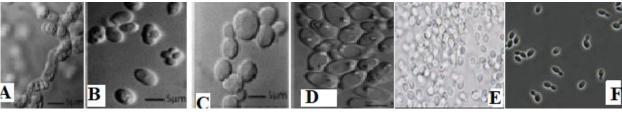


Figure 6. The photomicrograph depicts a selection of yeast cells present in Kombucha. The following microorganisms have been identified as significant in the context of Kombucha production: A: Schizosaccharomyces pombe; B: Saccharomyces cerevisiae; C: Zygosaccharomyces roxiei; D: Brettanomyces brusselsensis; and E: Starmerella basilaris; F: Hanseniaspora ovarum.

4. Biochemistry of Kombucha

Understanding the composition and attributes of Kombucha tea is extremely important. The composition and concentration of metabolites in this mushroom are influenced by various factors, including the source of inoculum, sugar concentration, fermentation time, temperature, pH of fermentation, oxygen and CO₂ levels in the fermentation solution, system performance, and supply of precursors (19). Alterations in these variables may have an impact on the fermentation rate as well as various physicochemical and sensory characteristics of the product. The analyses were conducted on Kombucha and the biochemical tests were performed under controlled and

stable conditions. There is variability in the biochemical composition of Kombucha mushrooms as reported in different research studies. The findings suggest that variations exist in the microbial and fungal compositions across distinct sources and countries. The diversity of metabolites produced by various bacterial and yeast species within the microbial consortium of this mushroom is indicative of the chemical composition of Kombucha tea. Figure 7 illustrates the schematic representation of Kombucha biochemical compounds (20).

4.1 Chemical composition of Kombucha

The primary ingredient employed in Kombucha production typically consists of tea infused with sugar.



Sucrose serves as the primary source of carbon in this particular process. The hydrolysis of sucrose in tea occurs through enzymatic action facilitated by invertase, an enzyme produced by the yeasts present in the Kombucha consortium, resulting in the formation of glucose and fructose (21). Initially, fructose undergoes a conversion process facilitated by yeasts resulting in the production of ethanol and carbon

dioxide. The subsequent procedural stage involves the conversion of ethanol into acetic acid by the action of Stobacter. The principal fermentation pathway of Kombucha results in the production of acetic acid, ethanol, and gluconic acid, all of which are significant constituents of Kombucha tea.

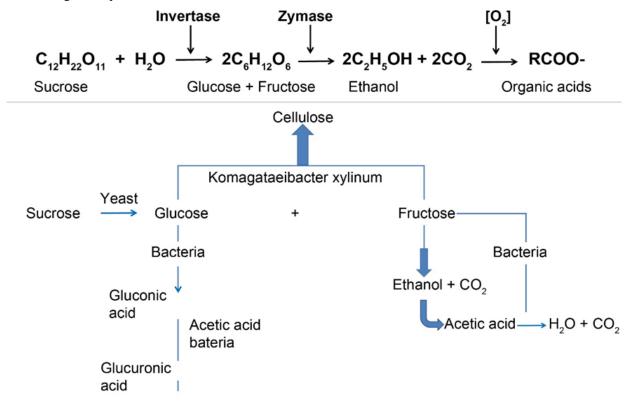


Figure 7. The main metabolic activity of Kombucha tea (source: Villarreal Soto et al., 2018)

Extensive research has been conducted on the chemical makeup of Kombucha, resulting in the identification of several compounds such as acetic acid, carbonic acid, folic acid, gluconic acid, glucuronic acid, lactic acid, oxalic acid, citric acid, malic acid, vitamin C, B vitamins (B_6 , B_3 , B_2 , B_1 , and B_{12}), and tea components like catechins, thiaflavin, and flavonol. Additionally, metabolic enzymes such as invertase, amylase, and other oxidative enzymes have been identified in Kombucha (21). 5. Beneficial Effects of Kombucha

4.2 Therapeutic and Health Effects of Kombucha

The consumption of Kombucha is connected to many protective and healing properties. - The positive effect of this product on public health has increased its usage. Numerous studies have indicated a range of potential health-benefits associated with the consumption of Kombucha. These

benefits include the reduction of risk factors for heart disease and diabetes, lowering of blood pressure, detoxification of the liver, alleviation of arthritis symptoms, reduction of cholesterol levels, reinforcement of the immune system, weight loss, increased longevity, potential treatment of prostate cancer and AIDS, and the prevention of cancer development. The advantageous impacts of Kombucha are attributable to its antioxidant properties as well as the presence of tea polyphenols, gluconic acid, glucuronic acid, lactic acid, vitamins, amino acids, antibiotics, and various micronutrients, which are all associated with the fermentation process (22-27).

4.3 Kombucha and antioxidant activity

It's important to include antioxidant compounds in your diet to improve the body's ability to combat free radicals.



in a beverage with elevated levels of antioxidant activity

acidic environment, or the release of enzymes by bacteria

and yeasts in mushroom tea, leads to the degradation and

decomposition of complex molecules into simpler

molecules, thereby increasing the abundance of these

compounds. The measurement of total phenolic content in

Kombucha tea indicates that fermentation leads to an

The mushroom exhibits robust antioxidant properties,

effectively mitigating lipid oxidation and averting DNA

increase in phenolic compounds (22-27, 30, 33-54).

The presence of complex phenolic compounds in an

During the process of Kombucha tea fermentation, numerous bioactive compounds with antioxidant properties are liberated from the tea leaves, capable of scavenging free radicals. Polyphenols and catechins are the main compounds found in tea and are considered as flavanols. Polyphenols exhibit a substantial antioxidant capacity. Due to their capacity to eliminate free radicals and reactive oxygen species, these molecules demonstrate efficacy in oxidative mitigation (28-32).Polyphenols approximately 30% of the total dry weight of tea leaves, with epigallocatechins, epigallocatechin 3-gallate, epicatechin 3gallate, and epicatechin being the predominant polyphenolic compounds present in tea leaves (Figure 8). The infusion of green tea leaves in the preparation of Kombucha tea results

(22).

fragmentation.

Figure 8. Chemical structure of some polyphenols in Kombucha tea

4.4 Kombucha and antimicrobial activity

Numerous studies have found that Kombucha tea possesses antimicrobial properties that can target a wide range of microorganisms (51, 55-58). Numerous studies have been conducted on the subject, demonstrating that Kombucha tea exhibits inhibitory effects against a wide range of pathogenic microorganisms, including both grampositive and gram-negative strains. Kombucha tea has been found to exhibit inhibitory effects on the growth of various pathogenic microorganisms, including Helicobacter pylori, Escherichia Entamoeba cloacae. Pseudomonas Staphylococcus aureus, Staphylococcus aeruginosa, epidermis, Agrobacterium tumefaciens, Bacillus cereus, Aeromonas hydrophila, Salmonella typhimurium, Salmonella intertidis, Shigella sonae, Leuconostoc

monocytogenes, Yersinia enterocolitica, Campylobacter *jejuni*, and *Candida albicans*. The antimicrobial properties of the liquid at low pH levels are primarily ascribed to the presence of acetic acid, various organic acids, catechins, and proteins generated during the fermentation process. Acetic acid and catechins possess the capacity to inhibit certain and Gram-negative Gram-positive microorganisms. Research has substantiated the presence of antibiotic compounds, including nisin, in this liquid, which exhibits antimicrobial efficacy. Kombucha contains eosinic acid, a naturally occurring antibiotic with demonstrated antibacterial and antiviral properties. It is efficacious against gram-positive bacteria, including Staphylococcus aureus, Enterococcus faecalis, and Enterococcus faecium. Nisin, another compound discovered in Kombucha, has been identified for its antibiotic properties.





Kombucha tea has been found to possess antibacterial and antifungal properties in recent research, with its production of acetic acid being the cause of the latter (59).

4.5 Kombucha and probiotic effects

Probiotics are viable microorganisms that, when consumed in effective dosages, elicit beneficial health effects. The predominant bacterial composition of a probiotic formulation is typically derived from cultures of Lactobacillus or Bifidobacterium, or a combination thereof. In addition to bacterial species, a variety of yeast species, including Saccharomyces cerevisiae and Saccharomyces boulardii, may also be found in this amalgamation. Probiotic microorganisms are integral factors in maintaining human health. Microorganisms play a crucial role in maintaining the equilibrium of essential microflora within the human body, enhancing the body's immune response, facilitating digestion, combating the proliferation of pathogenic bacteria, promoting mental well-being, and mitigating physiological issues such as anxiety and depression. The microorganisms found in Kombucha possess beneficial properties and exhibit high resistance, particularly in acidic environments. As a result, these microorganisms have the potential to supplant harmful microorganisms within the gastrointestinal tract, thereby positioning Kombucha as a product with probiotic attributes. The aforementioned effects serve to heighten interest in the consumption of Kombucha. Numerous studies have demonstrated that Kombucha beverages function as probiotics and can also serve as a consortium of probiotics and prebiotics. Prebiotics are defined as non-digestible food ingredients that are metabolized by beneficial bacteria in the gut, thereby promoting their growth and functionality. microorganisms in this drink, such as bacteria and yeast, have prebiotic properties and can help beneficial microflora grow in the intestines due to the presence of micro cellulose. A prebiotic agent, through selective mechanisms, facilitates the proliferation and activity of advantageous microbial populations within the human gastrointestinal tract. The microorganisms, including bacteria and yeast, found in this beverage exhibit prebiotic properties, while the presence of micro cellulose may facilitate the proliferation of advantageous microflora within the intestinal tract (60). Presently, there is a growing trend in the consumption and adoption of this beverage as a synbiotic. The researchers have discovered that the consumption of this beverage has the potential to serve as a source of essential nutrients,

thereby promoting the maintenance of health and overall well-being among individuals, particularly those employed in hazardous occupational settings, such as mine workers. Extended exposure to challenging conditions, like abnormal environmental factors, emotional stress, and substantial dietary changes, disrupts the gut's native microbial community. This phenomenon may lead to the depletion of commensal microorganisms in the gastrointestinal tract and consequently facilitate the proliferation of pathogenic secondary infections by opportunistic microorganisms. The alteration of the microbial population in the gastrointestinal tract can give rise to a range of health issues, including allergies, autoimmune disorders, multiple sclerosis, and stemming from organ infections transplantation. Consumption of Kombucha tea has the potential to bring about equilibrium in the microbial population (61).

4.6 Kombucha and anti-cancer properties

The key to cellular homeostasis lies in the controlled regulation of healthy and normal cell growth and proliferation. The proliferation of cancer cells is characterized by uncontrolled multiplication. The development of cancer in a genetically predisposed population is facilitated by exogenous factors, including chemical toxins (such as poisons and certain medications), physical agents (gamma, X, alpha, beta, and ultraviolet radiation), and biological agents (certain viruses and potentially some bacteria).

Previous research has recognized plant-based foods as having potential anti-carcinogenic properties (62). In recent years, there has been a burgeoning interest in the utilization of plant-derived products containing high levels of bioactive compounds. Studies conducted by researchers have demonstrated the anticancer properties of Kombucha.

Research findings have indicated that the anti-cancer properties of this beverage are attributed to the presence of tea polyphenols and secondary metabolites that are generated during the fermentation process. Numerous scientific investigations have demonstrated the capacity of tea polyphenols within fermented tea to impede gene mutation, suppress the proliferation of cancer cells, stimulate the apoptosis of cancer cells, and hinder metastasis. These mechanisms are acknowledged as contributing factors to the anti-carcinogenic attributes of the beverage (28, 34, 38, 46, 53, 63-66).

It has been highlighted that drinking Kombucha tea can help cancer patients lower their blood pH levels, which often





exceed 7.56 during the illness. The absence of lactic acid in the connective tissues of cancer patients may be linked to the generation of lactic acid as a result of Kombucha fermentation. Numerous elements found in Kombucha tea, such as polyphenols, gluconic acid, glucuronic acid, lactic acid, and vitamin C, have the potential to decrease the occurrence of cancer. Furthermore, Kombucha also includes disaccharide 1-4-lactone, which hinders the functioning of glucuronidase, an enzyme associated with cancer. Glucuronidase is capable of breaking down glucuronide and creating cancer-causing substances. Studies have shown that the polyphenols found in Kombucha tea have been found to have anti-tumor properties, making them effective in preventing and blocking cancer. Another research study has discovered the existence of dimethyl 2-malonate (2hydroxy-2-methoxypropylidene) and vitexin in the ethyl acetate portion of Kombucha tea, which demonstrates cytotoxic properties when present at a concentration of 100 μg/ml. Although Kombucha is not a cure for cancer on its own, its beneficial compounds can aid in the faster recovery, management, and prevention of cancer when combined with other treatment methods. The beneficial compounds lactic acid, glucuronic acid, and acetic acid found in Kombucha tea may have a role in regulating and potentially preventing cancer (28, 34, 38, 46, 53, 63-66).

4.7 Kombucha and detoxification and its hepatoprotective effects

Detoxification involves a complex set of biological processes designed to remove toxins from a living organism's body. This process may manifest physiologically or medicinally. The physiological functions discussed are primarily executed by the hepatic organ within the human body. The process of detoxification contributes to the preservation of liver function and the maintenance of overall health, thus playing a preventive role in the development of cancer. During the fermentation process of Kombucha tea, microorganisms produce enzymes, bacterial acids, and other secondary metabolites that possess the capacity to aid in the detoxification of the body. Moreover, a substantial number of enzymes and bacterial acids present in Kombucha tea exhibit striking similarities to the endogenous chemicals synthesized by the body for detoxification purposes. Consequently, the addition of Kombucha tea to an individual's dietary regimen is shown to alleviate the burden on the liver for detoxification. Studies have indicated that this capability is primarily attributed to the ability of glucuronic acid to effectively bind with toxic molecules,

thus elevating the excretion of these molecules from the body through the renal and intestinal routes. Glucuronic acid is generated as a byproduct of the oxidative transformation of glucose in the course of Kombucha fermentation. This acid exhibits potent detoxification properties within the human body, specifically by binding to toxins within the liver and effectively competing with them for elimination from the body. Malic acid, similar to glucuronic acid, is a metabolic byproduct produced during fermentation processes and is known to facilitate hepatic detoxification. Furthermore, consumption of Kombucha tea has been found to aid in liver detoxification and the elimination of heavy metals and environmental toxins from the body through renal excretion. Additionally, it plays a useful role in the biotransportation of hazardous metabolites, including bilirubin and surplus steroid hormones, out of the body. Kombucha tea's detoxifying effects have been shown to alleviate gout, vomiting, arthritis, and kidney stones, which are all associated with the accumulation of toxins in the body (4, 67-69).

4.8 Kombucha and diabetes

Diabetes, commonly understood to have a hereditary component, arises from insufficient production of the hormone insulin by the pancreas. Insulin is a hormonal mediator that promotes the uptake of glucose from the bloodstream into cells. Insufficient insulin production and secretion due to pancreatic dysfunction will cause a rise in the concentration of glucose in the bloodstream. Consequently, despite the abundance of extracellular glucose, intracellular glucose deprivation will ensue, resulting in a state of cellular "famine" and hunger. Complications of diabetes include weight loss, excessive thirst, vascular damage, and in severe cases, blindness. Diabetes typically has a low fatality rate, however, its complications significantly contribute to a reduced life expectancy. There are typically two distinct types of diabetes. In the initial or acute onset of diabetes, commonly manifesting before 25 years of age, there is a disruption in the production of insulin by the pancreas. The potential for amelioration of this complication lies within the application of insulin injections, regular exercise, and modification of dietary habits. The second type of diabetes, frequently observed in advanced age, is characterized by a reduction in tissue sensitivity to insulin or deficiencies in pancreatic function. In cases where weight gain (obesity) occurs, this condition may be exacerbated. The implementation of





exercise, dietary modifications, and weight loss is efficacious in ameliorating this form of diabetes. Individuals with diabetes are typically advised to prioritize the consumption of foods that are low in sugar. The findings from laboratory analyses indicate that the sugar content in Kombucha is at a minimal level, ranging from 4 to 5%. Hence, it is preferable to incorporate Kombucha and other fermented foods into one's diet, as these products undergo fermentation which results in the conversion of sugar content into alternative compounds. Kombucha is not a definitive cure for diabetes, however, it may potentially aid in stimulating insulin production in the body, thereby potentially reducing the necessity for insulin injections or oral medication. There is a belief among some people that drinking Kombucha can result in a swift decrease in blood sugar levels for those with diabetes (70).

4.9 Kombucha and digestive problems

The composition of gastric juice includes hydrochloric acid with a pH ranging from 1 to 1.5. The acidic solution exhibits a strong antimicrobial activity, making it suitable for food sterilization, while also aiding in the digestion of proteins. The stomach's acidic conditions help break down complex protein structures, preparing them for enzymatic hydrolysis. The pepsin enzyme initiates protein digestion within the stomach by hydrolyzing the proteins into smaller peptide fragments. Subsequently, the further breakdown of these peptides into individual amino acid units is facilitated by intestinal enzymes. Peptic ulcer disease represents a prevalent acute digestive ailment with diverse etiological factors documented in the literature. There is a divergence of opinion regarding the etiology of stomach ulcers, with some attributing their development to the persistent occurrence of indigestion, while others posit that mental strain is a contributing factor. Recent studies have provided ample evidence to support the identification of the bacterium Helicobacter pylori as the primary cause of gastric ulcers. In instances of heightened emotional states, there is a notable increase in the production of gastric acid within the stomach. In such circumstances, the aforementioned bacteria establishes itself and undergoes reproduction within the gastric environment. Given the acidic nature of Kombucha, with a pH ranging between 2 and 3, individuals experiencing indigestion and stomach ulcers commonly opt not to consume this beverage. After comparing the pH levels of Kombucha and gastric juice, it was found that the acidity of Kombucha is ten times lower than that of gastric juice. There

is a prevailing belief that Kombucha enhances the efficacy of food digestion and facilitates the restoration of the stomach to its physiological equilibrium. Under these circumstances, Hilkobacter's growth and reproductive capacity will be limited. Research has demonstrated that Kombucha elicits the activation of intestinal and digestive functions while also protecting against certain intestinal complications. Butyric acid, a byproduct of yeast metabolism in Kombucha tea, contributes to the protection of human cell membranes. When combined with gluconic acid, it enhances the resilience of the stomach wall, particularly in conditions associated with candidiasis. -Lactic acid is produced as one of the organic acids during the fermentation of Kombucha. Studies have shown that lactic acid has the potential to stimulate blood flow and alleviate symptoms of constipation.

4.10 Kombucha and lowering blood cholesterol

Coronary heart disease often stems from the blockage of coronary arteries caused by the buildup of plaque. There exists a well-established association between plasma lipid disorders and the increased risk of developing coronary artery disease. Furthermore, qualitative disorders of plasma lipids are known to significantly contribute to the pathogenesis of atherosclerosis, in addition to quantitative disorders such as elevated cholesterol levels. Higher levels of cholesterol and, in certain instances, triglycerides are identified as potential risk factors in the progression of atherosclerosis. This condition is known to be a major risk factor for the development of cardiovascular diseases. Research indicates that there is a direct correlation between low-density lipoprotein cholesterol (LDL-C) and the prevalence of coronary heart disease, and an inverse relationship between high-density lipoprotein cholesterol (HDL-C) and the incidence of coronary heart disease. Elevated plasma cholesterol levels lead to heightened oxidative stress within the vasculature, along with alterations in vascular and platelet function. Elevated cholesterol levels contribute to the initiation atherosclerosis lesions and endothelial dysfunction, involving the presence of oxygen radicals and oxidized lowdensity lipoprotein (Ox-LDL). The accumulation of Oxidized Low-Density Lipoprotein (Ox-LDL) within macrophages has been implicated in the development of foamy cells and, ultimately, atherosclerosis. One of the contributing factors to cardiovascular diseases is the elevation of apolipoprotein levels. Apolipoproteins, as





crucial components of lipoproteins, serve as significant cardiovascular diseases. predictors concentrations of apolipoprotein B (ApoB) and very lowdensity lipoproteins (VLDL), low-density lipoproteins (LDL), and lipoprotein (a) (Lp(a)) are significant contributors to the processes of inflammation and atherosclerosis. Apolipoprotein (ApoA)A functions as an anti-atherogenic agent by enhancing antioxidant activity, inhibiting oxidation, exerting anti-inflammatory effects, preventing LDL oxidation, suppressing the expression of adhesion genes, and inhibiting lipoxygenase activity, thereby preventing lipid oxidation. Research findings have demonstrated that Apolipoprotein A (ApoA) can decrease lipid levels and macrophage accumulation within blood vessels, thereby mitigating the development of fatty streaks. Hence, it is imperative to identify compounds capable of halting the advancement of atherosclerosis and managing it through the mitigation of oxidative stress. Currently, synthetic drugs such as lovastatin, LDL, clofibrate, and cholestyramine are employed to lower LDL levels and enhance HDL levels. However, there exists a multitude of debates regarding the utilization of these pharmaceutical substances. Numerous studies have demonstrated that certain medicinal plants can alter the plasma concentration of blood lipids. For instance, Kombucha has been found to decrease the levels of serum lipids, inflammatory markers, the thickness of atherosclerotic plaque hypercholesterolemic rabbits.

4.11 Kombucha and gout complications

Hyperuricemia is a defining feature of gout, indicated by elevated uric acid levels in the body. In this pathological state, sodium urate crystals are deposited in the joints, leading to inflammation and pain in the affected areas. Moreover, the kidneys and urinary tracts are susceptible to the deposition of insoluble crystals of sodium urate. The accumulation of sodium urate in the renal system results in the development of a specific variety of renal calculi. There have been assertions regarding the efficacy of Kombucha in the treatment of gout. If this theory is correct, it suggests that glucuronic acid is the most effective compound in improving gout symptoms because it can both detoxify and remove waste metabolites, as well as help eliminate uric acid from the body. The formation of uric acid occurs as a result of the degradation of purine bases, integral components of nucleic acids. This compound is excreted in considerable quantities on a daily basis through the process of urination.

4.12 Kombucha and hemophilia

Hemophiliacs face difficulties with blood clotting due to low levels of specific coagulation factors, such as factors 7 and 8, in their blood. Due to the presence of heparin in Kombucha, it is not advised for individuals with hemophilia to consume this beverage. Pregnant women and lactating mothers should refrain from consuming Kombucha due to the presence of this compound.

4.13 Kombucha and AIDS treatment

The etiology of acquired immunodeficiency syndrome (AIDS) is attributed to the presence of the RNA virus known as human immunodeficiency virus (HIV). The elevated glucuronic acid content found in Kombucha has been observed to stimulate the long-term production of interferon in the body. This finding raises the possibility that this fermented beverage may hold potential for use in the treatment of AIDS.

4.14 Kombucha and energy

The energy-boosting properties of Kombucha are widely recognized and endorsed by the majority of consumers, constituting a clear and unmistakable effect of its consumption. This fermented beverage provides a substantial boost in energy for individuals, making it a viable option as an energizing supplement during vigorous physical activities and strenuous athletic pursuits. A portion of the aforementioned energy arises from the consumption of compounds, including acetic acid and unfermented sugars present in Kombucha. Kombucha is believed to boost energy levels by improving digestion and absorption, which in turn provides the body with more fuel for cellular functions. The main reason Kombucha is thought to provide energy is that it improves the way the body digests and absorbs nutrients, resulting in a greater supply of fuel for cells. Kombucha energy-boosting properties are thought to be due to its ability to enhance digestion and absorption, leading to increased fuel for cellular function. Oxalic acid, a byproduct generated in the course of Kombucha fermentation, has potential utility in the synthesis of adenosine triphosphate.

4.15 Kombucha and obesity treatment

Kombucha helps improve cellular function and overall health by assisting the body in removing toxins and waste compounds. Kombucha is given to horses in specific parts of Europe and to camels in certain Arab countries because of





the factors mentioned earlier. There is a prevailing belief that Kombucha may also contribute to the mobilization and utilization of adipose tissue reserves in the human body. So, the drinking of Kombucha has been identified as a possible factor in helping overweight individuals lose weight (71).

4.16 Kombucha and skin and hair health

The condition of the skin is seen as the most important factor in determining a person's overall health. Therefore, the appearance and feel of the skin can be a direct indication of the overall health of the body. The outermost stratum of the integumentary system is identified as the epidermis, while the subdermal layer is referred to as the dermis. The dermis contains a network of blood vessels, nerves, sweat glands, and hair follicles, and serves as the site of skin renewal. The epidermis, the outermost layer of the skin, undergoes a process of regular turnover, wherein it is replaced by a new layer generated within the dermis. To achieve healthy, soft, and flexible skin, the various components of the body must collaborate. If an individual's state of health is not optimal, the resulting new tissue formation may lack pliability and flexibility. The circulation of blood, cardiovascular well-being, pulmonary function, digestive health, renal and hepatic function, and proper nervous system activity collectively contribute to the maintenance of healthy skin. The consumption of Kombucha has been associated with enhanced digestive efficiency, detoxification, and liver health, as well as the provision of essential vitamins, free amino acids, and beneficial compounds. Therefore, it is logical to assume that Kombucha could contribute to maintaining skin health. Furthermore, aside from its oral consumption, the application of Kombucha topically on the skin has been shown to contribute to the rejuvenation and overall health of the skin. In addition, it is worth noting that Kombucha cellulose may serve as a facial mask to revitalize the complexion. The composition of a hair strand primarily consists of alpha-keratin, a filamentous protein. The protein strands of hair are encapsulated by a protective layer known as the cuticle, whose color is determined by the presence of pigments within the hair cuticle. The dermal layer of the skin contains bulbous structures known as hair follicles, from which hair growth originates and subsequently emerges from the skin's surface. The manifestation of gray or white hair is attributable to a disruption in the production of hair pigment. Research studies have indicated that Kombucha may possess the potential to restore the pigmentation of

white and gray hair to its natural color. The efficacy of this beverage in addressing the progression of graying hair and enhancing ocular luminosity has been the subject of thorough research (72, 73).

4.17 Other therapeutic effects of Kombucha tea

Many claims have been made about the health-benefits and possible uses of this drink. The bacterial cellulose created in Kombucha fermentation could potentially be used in the development of bio-pharmaceuticals, showcasing its adaptability and effectiveness across different industries. The utilization of bacterial cellulose in this particular field derives from its exceptional purity and physicochemical characteristics inherent in the fermented beverage. Furthermore, bacterial cellulose is favored in scenarios where plant-derived cellulose is not suitable for use. Bacterial-derived cellulose is utilized within the food industry for its multifunctional properties as a food matrix, bulking agent, dietary fiber, stabilizer, and binder. The microorganisms present in Kombucha possess the ability to biosynthesize gluconic acid through the enzymatic degradation of caprenylic acid. This property has been shown to have the potential to inhibit the growth of various pathogens and in preventing candidiasis. Administering Kombucha tea orally at a dosage of 5 mg per kilogram of body weight in alloxan-induced diabetic rats leads to significant inhibition of alpha-amylase and lipase enzyme activity, as well as suppression of elevated blood glucose levels. Hence, it can be inferred that Kombucha tea exhibits promising hypoglycemic properties demonstrates beneficial anti-lipid activity (74).

4.18 Environmental Effects of Kombucha

4.18.1 The Effect of Kombucha as a Bioabsorbent and the Removal of Heavy Metals From Various Types of Wastewater

The presence of heavy metals in the environment is a significant concern due to their profound toxicity and capacity to bioaccumulate and biomagnify in the food chain, even at relatively low concentrations. In contemporary times, pollution caused by heavy metals has emerged as a prominent environmental concern.

During the early 20th century, the biological treatment method was developed, and it has since become a fundamental approach to treatment globally. This process is a rudimentary technique that occurs due to the proliferation of bacteria and fungi in elevated quantities within tanks.





Microorganisms, including bacteria, protozoa, and other microbes, collectively constitute activated sludge. The underlying principle of this approach is characterized by its simplicity. Bacteria derive sustenance from the consumption of organic carbon molecules. The proliferation of bacteria leads to the purification of sewage, enabling its subsequent discharge into receiving bodies of water such as rivers and oceans.

The presence of diverse microorganisms in Kombucha renders it a valuable and efficacious approach to remove heavy metals from various forms of wastewater. The beverage known as Kombucha is formed as a result of the symbiotic relationship between various strains of bacteria and yeasts. It is also commonly referred to as a Kombucha mushroom. The beverage known as Scooby Kombucha, characterized by its floating and solid component in the liquid phase, and also commonly referred to as "tea mushroom", is comprised of a symbiotic relationship of acetic acid bacteria, various fungi, and a cellulose membrane. The symbiosis of acetic acid bacteria and osmotic yeasts results in the formation of a cellulose membrane that remains buoyant on the surface of the fermented liquid, rendering it suitable for a wide range of potential applications. The potential of Scooby Kombucha as a bioabsorbent for the separation and heavy metals removal. Several studies have been conducted to investigate the potential of Kombucha in removing various metals, and the findings of these studies have been evaluated. In 2018, Mousavi et al conducted a study to examine the effectiveness of Scooby Kombucha and graphene oxide/Fe3O4 in removing Pb(II) from artificial wastewater (75). Moreover, the distinctive properties of Scooby Kombucha, including its anti-toxic and antimicrobial capabilities, make it suitable for the treatment of wastewater and the heavy metals removal. The present study examined the effectiveness of iron oxide nanoparticles Fe₃O₄ and Scooby Kombucha in the Pb(II) removal from synthetic wastewater, with a particular focus on their comparative performance. The Langmore model demonstrated a maximum absorption capacity approximately 114. 9 mg/g for iron oxide nanoparticles Fe₃O₄, while an absorption capacity of approximately 126. 6 mg/g was observed for Scooby Kombucha. The results obtained indicated that Scooby Kombucha, characterized by its cost-effectiveness, exhibits superior efficacy in lead (II) ions removal from water when compared to Fe₃O₄ iron oxide nanoparticles. In the year 2020, Kemanuik et al utilized wine effluent as a substrate in the process of fermenting Kombucha. Typically, the wastewater is discharged into the

municipal sewage system. The process of utilizing and managing wastewater has undeniably yielded environmental advantages. The primary growth medium was prepared by diluting winery effluent to a concentration of 70 g/L of total sugars with boiled water. The Kombucha starter was prepared by utilizing black tea and sucrose, and subsequently, 10% (v/v) of the starter culture was added to the initial substrate.

In the year 2019, Mousavi and colleagues conducted a study on the use of scoby Kombucha, a symbiotic colony of bacteria and yeasts, for the separation of nickel (II) from industrial wastewater (76). The process of decomposing nickel (II) ions from an aqueous solution was conducted by employing Scooby Kombucha within a reactor. The study also investigated and analyzed the impact of different parameters, including contact time, dose, adsorption agent, pH, and initial concentration, on nickel (II) removal. The utilization of the aforementioned material offered numerous benefits, including the production of high-quality purified water, reduced sediment formation, and enhanced durability, as well as the potential for adaptation to facilitate future advancements. The objective of this study was to examine critical elements of a viable approach for the extraction of heavy metals utilizing a Kombucha fungal reactor. The objective of this investigation was to demonstrate the efficacy of utilizing Kombucha as a microorganism for the nickel (II) removal from wastewater. Factors such as pH, time, temperature, electrolyte solution, and the volume and type of buffer were considered in this study. The adsorption experiments revealed that the peak absorption capacity of nickel (II) was observed at a pH of 7, a contact time of 15 minutes, and a temperature of 25 °C. Under ideal circumstances, a 94. 5% nickel (II) removal from the solution was observed, indicating the significant impact of Scooby Kombucha on the remediation of heavy metal pollution. Furthermore, the equilibrium experiments demonstrated a strong fit to the Langmuir isotherm model, resulting in the determination of Scooby Kombucha's high maximum absorption capacity of 452.54 mg/g at 25 °C. The research was conducted by Najaf Pour and colleagues. A study in 2020 aimed to investigate the capacity of Kombucha to remove heavy metals from environmental samples. This study aimed to investigate and optimize the impact of four parameters, namely the initial amount of tea fungus, tea content, sugar content, and water hardness, on the biological absorption of metals. The central composite design (CCD) under the response surface method (RSM) was employed for this purpose. The ANOVA analysis of variance of quadratic





models revealed that each of the models exhibited statistical significance. The most favorable conditions for the initial amount of tea mushroom, amount of tea, amount of sugar, and water hardness were determined to be 35. 85 grams, 443 grams, 1842 grams per 500 ml of drink, and 0 mg/L, respectively. The optimal water hardness conditions resulted in a removal efficiency of 93. 3% for Hg²⁺, 76. 7% for As³⁺, 76. 1% for Pb²⁺, 84. 3% for Cd²⁺, and 75. 4% for Cr⁶⁺ in the trial. The experimental data indicated a positive biological absorption. Ultimately, it is evident that Kombucha presents itself as a valuable and effective means of eliminating heavy metal contaminants, including nickel (II), Hg²⁺, As³⁺, Pb²⁺, Cd²⁺, and Cr⁶⁺, from diverse wastewater sources, owing to the rich diversity of microorganisms it contains. "The utilization of Kombucha offers numerous benefits such as improved water purification, reduced sediment in the final product, and flexibility for potential future advancements.

5. Kombucha tea preparation method and effective factors in optimizing taste, color, gas, and removing alcohol from Kombucha drinks

5.1 How to prepare Kombucha tea

The sweetened, sucrose-containing black tea is considered an optimal medium for the fermentation of Kombucha beverages. Green tea is a frequently selected substitute for black tea. The quantity and dimensions of the symbiotic population of bacteria and yeasts (SCOBY) introduced may vary. The diagram in question is represented in Figure 3. In summary, a quantity of 8 grams of tea is solubilized in 1 liter of boiling water. The tea leaves are subsequently filtered and removed from the infusion after 5 min. Tea bags may be utilized for convenience and simplicity of use. Sucrose is introduced into the solution at a concentration of 100 grams per liter. During the subsequent stage, the liquid from preceding Kombucha cultures and a section of cellulose layer are introduced into the cooled solution to initiate the fermentation process.

5.2 Effective factors in Kombucha fermentation

The process of fermentation is subject to various influencing factors including temperature, pH, oxygen levels, dissolved CO₂, system efficiency, availability of precursor materials, and the characteristics and content of the culture medium. Alterations in these variables can potentially impact the rate of fermentation and various physicochemical characteristics of the final product.

5.2.1 Substrates

Kombucha beverage is typically derived from the fermentation of sweetened green or black tea. Several researchers have noted that alternative sources, including cherry juice, blueberry, and date syrup (77). Also, various herbal extracts such as lemongrass tea and borage flower, may also have potential benefits. It can also be regarded as a viable substitute for the production of Kombucha syrup. Wilkanski et al demonstrated that the use of echinacea and savory plants as nitrogen sources can effectively reduce the fermentation time and bring about alterations in the texture of the conventional Kombucha beverage. Watavana et al. utilized coconut water as a carbon source for Kombucha fermentation and elucidated noteworthy instances of the biological activity of this symbiotic culture of bacteria and yeast in the production of novel compounds.

5.2.2 The effect of time

The fermentation duration of Kombucha tea typically ranges from 7 to 60 days, during which biological activities may intensify. Research indicates that the optimal results are commonly achieved within an average timeframe of 15 days. Furthermore, the presence of CO₂ in the environment has been shown to disrupt the interactions between microorganisms within the biofilm and their surrounding solution, leading to a loss of nutrient transfer to the microorganisms. The determination of the duration of the fermentation period is contingent upon the anticipated sensory attributes. Rice's study indicated that the process of fermenting fruit for a duration of 6 to 10 days produces a palatable beverage with a distinct flavor profile that develops gradually over an extended period, distinguishing it from the taste of vinegar. Based on the regulations set forth by the governing body and pharmaceutical authority, it is advised that Kombucha intended for human consumption undergoes fermentation for a maximum of 10 days. Kuten and colleagues (insert publication year) suggested that. The research investigated the microbial population evolution of Kombucha tea over a period of 2, 4, and 8 days in an industrial production setting. Evaluation of polyphenol content and antioxidant activity of Kombucha tea at different interval of fermentation (0, 7, 14, and 21 days) indicated an increase in the concentration of these compounds after 7 days, which may be attributed to the higher microbial diversity achieved at this stage.





5.2.3 Effect of temperature

Optimizing temperature control during Kombucha fermentation supports the growth of microbes and enhances enzyme activity, ultimately improving the fermentation quality. Furthermore, fluctuations temperature can influence the antioxidant and antibacterial properties, including phenolic compounds, of plant-based foods. The typical range for Kombucha fermentation temperatures falls between 22 and 30 °C. In 2013, Vitas and colleagues conducted a study on the fermentation of milk products with Kombucha, employing optimization models to investigate temperature values of 37, 40, and 43 °C. The findings indicated that the optimization of temperature significantly influenced the fermentation period of Kombucha, with the most favorable conditions for antioxidant activity occurring at temperatures ranging from 37 to 42 °C. Lone Kar et al found that samples treated with Kombucha obtained at higher temperatures exhibited elevated levels of acids, metabolites, and vitamin C production.

5.2.4 Effect of pH

The pH of the environment is a critical factor in influencing the fermentation process of Kombucha, as it significantly impacts the production of key acids such as acetic and gluconic acids, which are known to be pivotal contributors to the biological activities exhibited in the resultant beverages. The phenomenon of interest is also intricately linked to the proliferation of microorganisms and the alterations in the structure of phytochemical compounds, which may have an impact on their antioxidant properties. In order to achieve optimal activity, it is imperative that the pH value for this mushroom not fall below 3, as doing so may result in detrimental effects on the human digestive system.

5.2.5 Oxygen transfer rate and oxidation process

Glucose oxidation may be done as follows: $C_6H_{12}O_6 + 6O_2 \longrightarrow 6H_2O + 6CO_2$

The total combustion of 180 g of glucose requires the existence of 192 g of oxygen. However, both components must be soluble to be accessible to a microorganism. It should be emphasized that oxygen's solubility in water is about 6000 times lower than that of glucose. Hence, it is unfeasible to establish an adequate microbial environment to facilitate the oxidation of glucose or any other carbon source due to insufficient oxygen availability in the process. The

oxidation of ethanol to acetic acid involves using 32 grams of oxygen, or one mole, to oxidize 46 g of ethanol. In light of this consideration, in order to cater to the biological requirements of organisms, it is imperative that a microbial environment be supplied with adequate oxygen during the process of growth. The production of Kombucha beverage involves deliberate aeration of the cultivation environment to facilitate oxygen influx, thus mitigating alcohol formation while promoting water production. The designation of Kombucha as alcohol-free is warranted due to its low alcohol content, typically ranging from 0.5% to 1% However, the byproduct of the process of alcoholic fermentation or anaerobic growth is carbon dioxide and alcohol. In order to produce industrial alcohol from Kombucha, it is necessary to minimize the presence of oxygen in the surrounding environment.

6. Processing Kombucha syrup using other sugar sources

The standard method for fermenting Kombucha includes brewing black or green tea mixed with 5-8% sucrose. Furthermore, in addition to conventional substrates, there is potential to utilize alternative substrates in a variety of research investigations. Various types of renowned teas, including black, green, white, and oolong teas, are commonly used in the production of Kombucha. It is noteworthy that black and oolong teas undergo oxidation and exhibit elevated levels of polyphenol oxidase activity, resulting in their characteristic dark coloration. The study team administered a survey and discovered that the majority of participants expressed a preference for utilizing public transportation as opposed to using their private vehicles. Green and white tea undergo minimal oxidation and as a result, yield a beverage with a lighter color. The process of oxidation in tea is initiated by exposing the leaves to ambient air, leading to the transformation of polyphenols, particularly catechins, within the leaves. Moreover, it is important to take into account the variety of tea and the sugar concentration employed in the cultivation of Kombucha. Various sucrose sources, including white sugar, sugar beet, sugar cane, date syrup (77) and molasses, are commonly utilized as a primary carbon source for yeast and symbiotic bacteria. The selection of an appropriate sweetener that possesses the ability to effectively substitute sugar, preserve product quality, and maintain product integrity throughout the storage duration is of paramount significance. Research has demonstrated that the fermentation of lactose by Kombucha results in the production of small quantities of



ethanol, in comparison to sucrose fermentation. The microorganisms present in Kombucha have the capacity to utilize alternative sources of sugar for fermentation. The subsequent section provides a literature review of various scholarly studies on alternative sources of sucrose for the production of Kombucha and their corresponding impacts (77).

Feasibility of substituting sugar with grape and blackberry juices in Kombucha production and its impact on various properties, including pH, acidity, polyphenol, flavonoid (78), antioxidant activity, glucose levels, and sensory characteristics over a 15-day fermentation period. The item has been borrowed or loaned out for use. The findings indicated a substantial decrease in the pH of the samples as a result of the fermentation process. The acidity, antioxidant activity, polyphenol and flavonoids content exhibited a notable increase in all samples throughout the fermentation process. The samples containing berry juice, mixed juices, grape juice, and the control demonstrated the highest levels of these compounds, with the respective order of abundance mentioned. The glucose levels in the samples exhibited a reduction over the course of the fermentation process, with the most significant decrease observed in the samples containing natural juices. Results from the sensory evaluation indicated that the Kombucha beverages derived from fruit juices exhibited a slightly darker and sourer taste profile compared to the control sample, while also appearing lighter in color. However, evaluators considered the Kombucha containing berry juice to be on par with the control sample. - The findings suggest that replacing half of the sugar with berry juice in Kombucha could improve its nutritional profile by lowering glucose levels and boosting antioxidants and organic acids.

In the study conducted by Bolverdi et al (2012), inulin extracted from pickled potato tubers or artichoke was employed as a substrate for the cultivation of Kombucha (79). This study utilized inulin and oligofructose extracted from pickled potato tuber at concentrations ranging from 0% to 10% as substrates for Kombucha fermentation. The research also investigated the chemical changes occurring during the fermentation process and the resultant Kombucha syrup. An examination was conducted to compare the effects of sucrose (sugar) at equivalent concentrations. The findings indicated that the rate of consumption of inulin was observed to be higher than that of sugar during the process of Kombucha cultivation. The Kombucha beverage formulated with inulin demonstrated a resulting higher pH level. The quantification of acetic acid and lactic acid via the HPLC

method revealed that the concentration of acetic acid present in the product synthesized with sucrose was found to be greater than the concentration of lactic acid in the product synthesized with inulin. The findings revealed that Kombucha derived from inulin exhibited a greater concentration of soluble protein and antioxidant activity in comparison to Kombucha produced using sucrose. The research findings indicate that the utilization of inulin as a substrate for Kombucha fermentation can lead to a reduction in fermentation duration and result in a product with an elevated pH level.

Dodgar et al. (2016) employed fig juice as a replacement for sugar in the fermentation process of Kombucha in their study. This study aims to investigate the potential of enhancing the nutritional properties, taste, sensory and rheological properties, and marketability of a product through the substitution of sugar with fig juice. Additionally, the study explores the possibility of introducing greater product variety as a result of the substitution. The investigation involved quantifying the production of glucuronic acid in the samples, given its known health-promoting properties. The analysis of the fig juice sample revealed a substantial increase in glucuronic acid levels on the 30th day compared to the 1st and 15th day. Furthermore, there was no significant difference in the glucuronic acid levels between the 1st and 15th day samples.

7. Conclusion

In comparison with Kefir, kombucha is less in protein and probiotics but it non-dairy-based, which make it suit lactoseintolerant consumer, including athletes and others. Apple cider vinegar lacks probiotics but may aid metabolism. Diluted drink is a solution to avoid acidity. Practical recommendation dosage kombucha for athletes is 4-12 oz/day (post-workout). Those brands with live cultures, low sugar (<8g/serving), and no additives are also recommended. Post-exercise consumption lead to detoxification and antiinflammatory effects. While kombucha and other fermented beverages offer promising benefits (antioxidants, probiotics, recovery aid), their effects are not yet fully validated by large-scale human studies. Athletes should integrate them as part of a balanced diet rather than relying solely on them for performance gains. For further details, refer to the cited studies on kombucha's composition and mechanisms.

Authors' Contributions

All authors equally contributed to this study.





Declaration

In order to correct and improve the academic writing of our paper, we have used the language model ChatGPT.

Transparency Statement

Data are available for research purposes upon reasonable request to the corresponding author.

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Declaration of Interest

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