Food Research 8 (Suppl. 5) : 70 - 83 (2024)
Journal homepage: https://www.myfoodresearch.com

FOOD RESEARCH

REVIEW

Review of the benefits of kombucha tea: chemical components and pharmacological activities

[1]Md Farid, D.F., [2]Abdullah, S. and [1,*]Abu Bakar, M.F.

[1]Department of Technology and Natural Resources, Faculty of Applied sciences and Technology, Universiti Tun Hussein Onn Malaysia (UTHM) Kampus Cawangan Pagoh, Hab Pendidikan Tinggi Pagoh, KM 1, Jalan Panchor, 84600 Panchor, Johor

[2]Department of Physics and Chemistry, Faculty of Applied Sciences and Technology, Universiti Tun Hussein Onn Malaysia (UTHM) Kampus Cawangan Pagoh, Hab Pendidikan Tinggi Pagoh, KM 1, Jalan Panchor, 84600 Panchor, Johor

Article history:
Received: 5 December 2023
Received in revised form: 10 October 2024
Accepted: 17 October 2024
Available Online: 20 November 2024

Keywords:
Kombucha tea,
SCOBY,
Fermentation,
Health benefits,
Chemical,
Pharmacological

DOI:
https://doi.org/10.26656/fr.2017.8(S5).8

Abstract

Kombucha tea is a type of tea that undergoes fermentation through the inclusion of tea leaves, sugar, and a symbiotic culture of bacteria and yeast (SCOBY). The fermentation process typically spans a period of around 10 to 15 days. The utilization of this substance dates back to ancient times, mostly for medicinal purposes. The review has demonstrated that kombucha tea has distinctive characteristics, including anticancer, antioxidant, antimicrobial, and antifungal activities. The chemical component of kombucha plays an important role in kombucha tea which enhances the pharmacological activities such as acetic acid, lactic acid, phenolics and flavonoids. This paper presents an analysis of recent findings pertaining to the potential health advantages of kombucha, along with an examination of the chemical constituents and metabolites generated during the fermentation process. Additionally, further recommendations are provided for future investigations in this field.

1. Introduction

Since the early years of human civilization, fermented food has been known in the human diet and also in medication (Admassie, 2018). It is believed that the fermentation process could enhance flavour and shelf -life and this process involves microorganism and their metabolite which will change the environment of the product to the point which is favourable for good bacteria. The fermentation process can be categorized by the microorganism involved which is mainly lactic acid bacteria (LAB) and also the present of food substrate. Through those explanations, fermented food is mostly defined as food which is produced through the involvement of microbes and enzymatic conversion of food components which is beneficial to human health when consumed (Marco *et al*., 2017; Dimidi *et al*., 2019). Many foods can undergo a fermentation process including perishable and non-perishable food which contain high concentrations of substrate such as monosaccharide or disaccharide which will undergo enzymatic action (Marco *et al.,* 2017). As the outcome of these various methods of fermentation, there are thousands of different kinds of fermented food that have been known nearly in every culture world-wide which has been used for thousands of years and remain relevant until now due to the benefit of the food product towards consumer. Plus, there were several groups which recommended that fermented food should recognized as a national dietary recommendation (Chilton *et al.,* 2015). The fermentation process can be done by two different methods which are natural and or spontaneous fermentation, for example, kimchi, tapai and other fermented products whereby the microbes are already present naturally in the product. Vice versa the second fermentation process involves the addition of starter culture which is produced by the previous fermented batch and is included in the formulation of the product which we call “backslopping” (Yann and Pauline, 2014; Marco *et al*., 2017; Rezac *et al*., 2018; Dimidi *et al*., 2019). The present of microorganisms in the product not only improves the sensory characteristic and shelf-life but also improves gastrointestinal health, antibody and anticancer properties. There are numerous studies have shown that microorganisms that exist in fermented products can reach until gastrointestinal tract to exert a physiological benefit against pathological bacteria and produce immune-regulatory benefits in the tract (Derrien

*Corresponding author.
Email: *fadzelly@uthm.edu.my*

and Vlieg, 2015). For example, with the present of lactic acid bacteria, the production of bioactive peptides and polyamides will increase which leads to potential beneficial towards cardiovascular, immune and metabolic health. Also, the presence of lactic acid through fermentation can lead to the conversion of phenolic compounds to biologically active compounds which is beneficial towards free radical scavengers (Filannino *et al*., 2015; Pessione and Cirrincione, 2016).

Another food product which has been known for thousands of years is tea. Tea is the second most known liquid after water and has already been consumed since 3000 BC (Sharangi, 2009). The study about tea is still ongoing to show the optimum result for certain points for potential health benefits and medicinal properties (Abu Bakar *et al*., 2006). The cultivation of tea in India from 1818 until 1834 can be considered the origin of tea and it has been one of the most important agriculture products throughout the world (Harbowy *et al.,* 1997). All of the type of tea leaves comes from the same species which is *Camellia sinensis* L. but differ in their appearance, flavour and chemical component due to the exposure for fermentation and hydration process (Sharangi, 2009). Green tea contains a high concentration of monomeric polyphenols from the catechin group due to the hydration process. The preparation of green tea is by exposing the freshly picked tea leaves to the air until the moisture in the leaves is evaporated. The second step is to roast and stir continuously until it becomes moist and flaccid which eventually improves the appearance of the product before being passed to the rolling table and being shaped into balls to extract more moisture. Next, the leaves are roasted until the leaves assume dull green colour (Poonam *et al.,* 2018). Most of the steps in processing green tea leaves which involve heat are to extract most of the moisture present and also lead to inactivation of enzymes (Marco *et al.,* 2017). The preparation for black tea is a bit different compared to green tea. The first step in producing black tea is withering which results in the disruption of the cell structure which triggers the fermentation process (Łuczaj and Skrzydlewska, 2005; Marco *et al*., 2017). In the production of black tea, enzymatic transformation does occur which involves oxidation and polymerization resulting in 75% of catechins (Bokuchava *et al*., 1980; Katiyar and Mukhtar, 1996; Marco *et al*., 2017).

Tea product which undergoes the fermentation process is known as Kombucha tea. This product is originally known in northeast China during the Tsin Dynasty in Manchuria (Laureys *et al.,* 2020). Originally kombucha tea was used as medicine due to its health-promoting properties and there are still studies being conducted to know its full potential towards consumers' health. Kombucha tea is a sweetened tea which being fermented in time by the presence of a symbiotic culture of bacteria and yeast (SCOBY) creating a cellulose layer on top of the solution during the fermentation process (Laureys *et al.,* 2020). This beverage has become more popular recently in Western culture and the retail market has shown an increase in value growth over the past 2 years (Laureys *et al.,* 2020). Kombucha tea usually consists of tea leaves, solute which is water, sucrose (5-8%) and SCOBY. Sucrose acts as a substrate for SCOBY which consists of acetic acid bacteria (AAB) and osmophilic yeast which trigger the natural fermentation process which leads to the production of beneficial acid and alcohol (Gaggìa *et al.,* 2019; Jarrell *et al*., 2000). In the past 10 years, the market interest in kombucha tea has shown a positive increase towards the world and also, self-brewing for the production of kombucha tea has been quite common these days (Gaggìa *et al*., 2019; Kapp and Sumner, 2019). Kombucha tea commonly being consumed due to the availability of beneficial chemical compounds and antioxidant value such as organic acid, vitamins and tea polyphenols, and the low pH to avoid bacterial contamination (Gaggìa *et al.,* 2019). Based on the quality of tea, the tea beverage is usually based on species, season, age of tea leaves, position of plucking, climate, and horizontal practice which highly affects the composition of antioxidants and polyphenols which eventually affect the composition of fermented tea beverage (Lin *et al*., 1996; Lin *et al*., 1998). Currently, kombucha can be claimed as of probiotic product due to the presence of beneficial microbe but also claimed as an "unsafe medicinal tea" (Hartmann *et al.,* 2000). Due to the insufficiency of clinical proof and support, there are a lot of misconceptions and false information about the idea of kombucha tea's chemical benefits. This study will analyze the benefits of kombucha tea in terms of its pharmacological properties and chemical components, such as antioxidants and polyphenols.

2. Definition of Kombucha tea

Kombucha is a beverage made from the fermentation of tea, primarily black tea, but other varieties such as green and oolong tea, can be used as a base for its manufacture. It is also possible to find variations which can be infused into the formulation such as mint, lemon balm or jasmine. Some metabolic products of Symbiotic Culture of Microorganisms and Yeast (SCOBY), like acetic acid and other organic acids, have antibacterial properties and prevent pathogenic bacteria from contaminating the drink (Xia *et al.,* 2019). Due to the presence of beneficial microbes, a cellulose mat will appear on top of the solution which is called SCOBY daughter which also contains beneficial microbes and

https://doi.org/10.26656/fr.2017.8(S5).8

can be used for another batch. As mentioned above, kombucha has been consumed as a nutraceutical in Asia for thousands of years (Kaewkod *et al.,* 2019b) It is said that kombucha is possibly made by accident like many other fermented foods. Due to its promising potential towards health which involves functional capabilities such as anti-inflammatory potential and antioxidant activity, reduction of cholesterol level and blood pressure, reduction of cancer propagation, the improvement of the liver, the improvement of the immune system and gastrointestinal function, Kombucha tea can also be claimed as a functional food (Kaewkod *et al.,* 2019b). In terms of analysis for this product, mainly it focuses on its chemical component which is nutritional and antioxidant value. A study has recorded that kombucha tea contains rich organic acids such as acetic acid, glucuronic acid, and gluconic acid, vitamins and polyphenols (Rasu Jayabalan *et al*., 2014; Gaggìa *et al*., 2019). Another study also has been conducted which involves animal study to analyze the beneficial effects of kombucha tea which include antimicrobial activity, hepatic detoxification, anti-inflammatory, hypocholesterolaemia, anti-proliferative, and hypoglycaemic activities (Dufresne and Farnworth, 2000; Villarreal-Soto *et al*., 2018; Gaggìa et al., 2019). But kombucha is not yet universal and well known throughout the world because it is a live fermenting drink, requiring constant precaution and refrigeration making it more difficult to transport and distribute.

3. Preparation of Kombucha tea

The aerobic and static fermentation process is vital in the process of Kombucha tea production and it usually includes sucrose-sweetened black, green, or blue (Oolong) tea with a symbiotic culture of bacteria and yeast (SCOBY) which will trigger the fermentation process (Xia *et al.,* 2019). The process starts with a combination of boiling water with 50-100 g of sugar or sucrose. Following that, tea leaves, either loose or in tea bags, are steeped for a brief time before being extracted. After that, the solution is cooled to 20 degrees Celsius and infected with a piece of SCOBY mat, which contains a lot of native microbes (Figure 1). The purpose of the addition of the previous batch of Kombucha tea is to lower the pH of the Kombucha tea which will inhibit the growth of bad pathogens such as *Clostridium botulinum*, *C. perfringens*, and *Bacillus cereus*. These microbes do not grow in an environment below pH 4.7. The fermentation process usually takes place under aerobic conditions and at a temperature of 18-28°Cfor 8-14 days (Mousavi *et al.,* 2020).

The fermentation process in the making of kombucha can be considered natural due to the addition of a certain amount of a previous kombucha fermentation which can be called back-slopping. Back slopping is a technique that is also utilised in the production of sourdough, fermented meats and cereals, water kefir, and milk kefir, among other natural food fermentation processes (Rasu Jayabalan *et al.,* 2011). The fermentation process for kombucha normally takes 8 to 14 days at room temperature, during which time the characteristics of the sweetened tea evolve significantly. The fermentation medium is aerobic, rich in sugar and sucrose, and somewhat acidic due to the inclusion of a previously fermented kombucha solution. The oxygenation of the liquor gradually reduces due to the creation of a cellulose layer on top of the fermenting liquor and the consumption of oxygen by the microorganisms in the SCOBY mat and the liquor. The substrate concentrations eventually decrease due to microbial metabolism and the formation of the cellulose layer. A more intense anaerobic condition with a low amount of substrate may arise at the end of the process.

Figure 1. Kombucha tea.

4. From tea to kombucha: the fermentation process

An extensive study about the microbial diversity in the kombucha fermentation process has been established and it shows that during the fermentation process of kombucha tea yeast and acetic acid bacteria (AAB) are both present. The main components in the production of kombucha tea are tea leaves, water, sugar which act as a substrate and SCOBY as mentioned in the previous chapter. Sweetened black tea is one of the best substrates in kombucha tea production compared to green tea but the preparation for both teas is the same (Dufresne and Farnworth, 2000). The identification of microbiological components in kombucha tea takes place in the early study of kombucha tea. There were studies which proved that one of the main microbiological components in the beverage is acetic acid bacteria which is Acetobacter *xylinum*, *A. xylinoides*, Bacterium *gluconicum, A. aceti* and *A. pasteurianus* (Reiss, 1994; Balentine, 1997). For fungi, the main component found in kombucha tea is *Schizosaccharomyces pombe, Kloeckera apiculate, Saccharomycodes ludwigii, Saccharomyces cerevisiae,*

https://doi.org/10.26656/fr.2017.8(S5).8

Zygosaccharomyces bailii, Brettanomyces bruxellensis, B. lambicus, B. clusters, *Candida* and *Pichia* species (Reiss, 1994; Mayser *et al*., 1995; Balentine, 1997). One of the characteristics that contribute to the popularity of kombucha tea is the presence of symbiotic interactions between bacteria and yeast, which are capable of limiting pathogen growth in the product (Villarreal-Soto *et al*., 2018, Villarreal-Soto *et al*., 2019). The presence of beneficial microbes in the product creates an aerobic condition which converts substrate that is present in the formulation for the fermentation process which takes place approximately 7-10 days. The main substrate in the formulation of kombucha tea is sugar also known as sucrose and in the fermentation process, an invertase enzyme in the periplasm of yeast cells hydrolyzes sucrose into glucose and fructose, which is the monomer form of sucrose, increasing the quantity of glucose and fructose. (Dufresne and Farnworth, 2000; Manach *et al*., 2004). The process continues with glucose and fructose being fermented by yeast into ethanol, carbon dioxide and glycerol which affect the alcohol content in the tea. The by-product of the yeast which is ethanol will be used by AAB to produce acetic acid (Ahmed and Dirar, 2005). There is evidence which proves the presence of carbon dioxide in the fermentation process which is due to the aerobic process of the microbes but it will partly escape into the atmosphere.

As explained earlier, the main by-products of the fermentation process are ethanol, glycerol, acetic acid, gluconic acid, glucuronic acid D-saccharic acid 1,4-lactone, and lactic acid and those components reach their maximum value due to the fermentation process after 6-10 days and reaches its equilibrium state (Figure 2) (Sievers and Swings, 2015; Gomes *et al*., 2018). But this information is still under development due to incomplete analysis and more detailed analyses are needed to be done. During the fermentation process, there are many selective pressures happening that control the variety of microbial species in kombucha, and hence influence substrate consumption and metabolite synthesis (Rasu Jayabalan *et al.,* 2014). For example, the acidic stress caused by organic acid synthesis which also produced by yeast (acetic acid), and AAB (lactic acid and acetic acid).

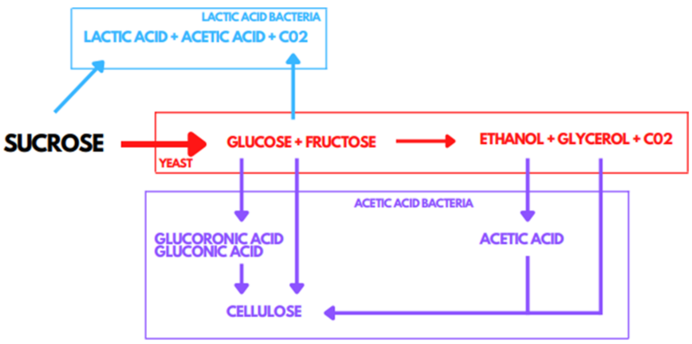

Figure 2. Main metabolic activities of SCOBY during the fermentation of kombucha tea (Ahmed and Dirar, 2005).

This process affects the pH value of the product which might drop to 2.0-4.0 after 7-10 days of incubation. There is also evidence that the restricted availability of essential vitamins and nitrogenous compounds in the mixture might be a significant pressure factor for the microbial population. It argues that the availability of the tea leaves employed in the recipe is the only source of (micro)nutrients (Gholami, 2017).

In the fermentation process for the production of kombucha, all of the microbes present in the mixture will undergo aerobic respiration which supports that the existence of oxygen and its concentration is considered as an important property which will affect the growth of AAB (Laureys *et al.,* 2020). In the initial phase of fermentation, the environment of the mixture is highly oxygenated and favourable by the AAB but the concentration of oxygen will be decreased due to the formulation of growing cellulose pellicle on top of the mixture of we can call SCOBY daughter (Rodrigues *et al*., 2006). It is crucial to optimize the parameters during the kombucha tea fermentation process since they have a major influence on the final microbial composition and, as a result, the metabolite composition of kombucha. The measures of the fermentation vessel, for example, and the surface area to volume ratio, might have an effect on the quantity of oxygenation during a fermentation. This is less of a concern when the mixture is aggressively stirred or aerated throughout the fermentation process, as some producers have done effectively. A pH above 4 could be linked to low oxygenation and hence low acid generation by AAB, which can sometimes stimulate the growth of LAB (lactic acid bacteria) and so lactic acid production. The fermentation period will also have a serious influence on the microbial ecosystem's acidic stress and, as a result, the end product's microbial and chemical composition. Higher fermentation temperatures tend to favour LAB, which is consistent with their higher optimum development temperatures as compared to yeasts and AAB. (Mousavi *et al.,* 2020).

5. Chemical component of kombucha tea

When it comes to the active compound of kombucha tea, it typically reveals the presence of multiple compounds including organic acid, sugar, water-soluble vitamins, amino acid, biogenic amines, purines, pigments, lipids, proteins, hydrolytic enzymes, ethanol, acetic acid bacteria and lactic acid bacteria, carbon dioxide, poly-phenols, minerals, anions, D-saccharic acid -1,4- lactone (DSL), and metabolic product of yeast and bacteria (Yang *et al.,* 2010; Rasu Jayabalan *et al.,* 2014). The amount of production of chemical content in kombucha tea mainly depends on multiple factors such as the symbiotic culture used for the fermentation et al. (2007)

process, duration of fermentation, sucrose content and type of tea used. These chemical compounds are originally produced by SCOBY but it still depends on the type of substrate used in the formulation of the tea which can alter the structure to create a new compound during the fermentation process (De Miranda *et al.,* 2021). Table 1 shows the compounds which have been identified in the fermentation process of black tea and green tea leaves.

The pH value of Kombucha tea steadily falls throughout the alcoholic fermentation due to the brewing process, which results in the creation of organic acid, as proved by Jayabalan *et al*. (2007). This demonstrates a rise in acetic acid production on the 15th day of fermentation (De Miranda *et al.,* 2021). Another study also proved the increase in acetic acid and gluconic acid concentration in green tea kombucha which reached the level of 16.57 and 7.36 g/L after 21 days of fermentation duration (Chakravorty *et al.,* 2016). Another interesting study done by Malbasa *et al*., (2018) uses sugar beet molasses as the source of carbon for the fermentation process. It demonstrates a higher drop in pH and an increase in organic acid output when compared to the identical manufacturing of kombucha tea that solely uses sugar as a substrate. These changes can be explained by the presence of molasses itself which includes other

Table 1. Identified compounds in the fermentation process of black tea and green tea leaves.

Substrate	Organic acid	Amount (g/L)	Vitamins and minerals	Amount (g/L)	Fermentation duration (days)	References
Black tea	Acetic acid	4.69	-	-	18	Jayabalan *et al.* (2007)
	Glucuronic acid	1.71				
	Lactic acid	0.18				
	Acetic acid	8.0	-	-	15	Kallel *et al.* (2012)
	Acetic acid	16.57	-	-	21	Chakravorty *et al*. (2016)
	Glucuronic acid	7.36				
	Acetic acid	1.83	-	-	14	Ismaiel *et al.* (2016)
	Glucuronic acid	1.86				
	Lactic acid	0.702				
	Citric acid	2.77				
	Acetic acid	9.18	-	-	14	Gaggìa *et al.* (2019)
	Glucuronic acid	3.23				
	Acetic acid	6-15.1	-	-	21	Villarreal- Soto *et al.* (2019)
	Acetic acid	11.15	Vitamin C	0.70	15	Kaewkod *et al.* (2019)
	Glucuronic acid	1.58				
	Succinic acid	3.05				
	Acetic acid	3.6	-	-	10	Cardoso *et al.* (2020)
	Glucoronic acid	1.17				
	Lactic acid	0.015				
	Acetic acid	1.55	Iron	-	7	Ivanišová *et al.* (2020)
	Tartaric acid	0.23	Manganese			
	Citric acid	0.05	Zinc			
	Acetic acid	9.08	-	-	14	Jakubczyk *et al.* (2020)
Green tea	Acetic acid	8.36	-	-		Jayabalan *et al*. (2007)
	Glucuronic acid	1.73				
	Lactic acid	0.12				
	Acetic acid	5.4	-	-	15	Kallel *et al.* (2012)
	Acetic acid	7.65	-	-	14	Gaggìa *et al.* (2019)
	Glucuronic acid	1.96				
	Acetic acid	10.42	Vitamin C	0.61	15	Kaewkod *et al.* (2019)
	Glucuronic acid	1.37				
	Acetic acid	3.2	-	-	10	De Miranda *et al.* (2021)
	Glucuronic acid	0.47				
	Lactic acid	0.02				
	Acetic acid	9.14	-	-	14	Jakubczyk *et al.* (2020)

https://doi.org/10.26656/fr.2017.8(S5).8

components such as fatty acid, lactic acid and non-volatile acids (Malbaša *et al.,* 2008). However, this study shows the production of acetic is lower compared to others and it may also be due to the presence of molasses which decreases the activity of acetic acid bacteria. Some investigations have demonstrated the presence of vitamins in kombucha tea, such as water-soluble vitamins detected in kombucha brewed with 0.7% sugar and 5 g/L black tea by Bauer-Petrovska and Petrushevska-Tozi (2000). Vitamin B1 was 74 mg/100 mL, vitamin B6 was 52 mg/100 mL, vitamin B12 was 84 mg/100 mL, and vitamin C was 151 mg/100 mL (Bauer-Petrovska and Petrushevska-Tozi, 2000). Another study shows the maximum content of vitamin B2 in a kombucha formulation of 7% sucrose in 8% of tea leaves. In that investigation, vitamin C was proven to be present in the formulation and reached the highest value on the 10th day of fermentation with 28.98 mg/mL. Another research used the RSM approach in fermented milk products, including earlier kombucha batches grown on winter savoury and stinging nettle extract. According to this study, the value of vitamin C in this product is higher than in standard kombucha products, with a value of 15.19 mg/mL. (Vitas *et al.,* 2013).

There has been only a little research done on the mineral content of kombucha tea. Minerals are inorganic compounds that are required in trace amounts for normal biological function and development. Research was conducted to demonstrate the presence of copper, iron, manganese, nickel, and zinc. These minerals rose as a result of the kombucha's metabolic rate throughout the fermenting process. Lead also has been proven to be present in the mixture but only in a small amount (Bauer-Petrovska and Petrushevska-Tozi, 2000). Toxic blood lead levels for adults are 20 g/dL and for children are 10 g/dL, which is approximately 0.2 and 0.1 g/mL, respectively, according to the Agency for Toxic Substances and Disease Registry (ATSDR). Because kombucha tea has substantially lower amounts, it does not pose a health hazard. A Review paper also reported the amount of minerals which are copper, iron, magnesium, nickel, and zinc with a composition of 0.1-0.4 mg/mL with an initial sucrose concentration of 70 g/mL with a fermentation duration of 15 days (Villarreal-Soto *et al.,* 2018). Polyphenols are active substances that include one or more phenol structural units per molecule and are a class of phytochemicals that are the most prevalent antioxidants in the diet (Leal *et al.,* 2018; Abu Bakar *et al.,* 2010). Polyphenols are also one of the wonderful compounds present in kombucha which are able to scavenge free radicals which may resulting kombucha tea as a potent antioxidant agent (Mousavi *et al.,* 2020). During the production of kombucha tea, the overall polyphenol content of kombucha increases linearly and takes around 15 days to stabilize (Chu and Chen, 2006). Also, one of the main ingredients in making kombucha tea is tea leaves. Both epicatechin (EC) and epigallocatechin (EGC) are present in the product (Manach *et al.,* 2004). There is a study of kombucha tea which uses green tea and black tea at the same time. For green tea, a higher level of EC (150%) was found on the 12th day of fermentation and for black tea, 115% of EGC was present on the same day (Jayabalan *et al.,* 2007). As mentioned before, temperature is one of the main factors which will affect the quality of the product. There was a study which included exposure to high temperatures approximately 76^{o}C- 100^{o}C during the fermentation process. For 6 and 8 days, the antioxidant activity is reduced by 36.42 and 35.95%, respectively (Ahmed *et al.,* 2020).

D-saccharic acid-1,4-lactone (DSL) is one of the chemical components in kombucha tea it has the potential to inhibit glucosidase activity which facilitates glucuronic acid to repel toxicants which include carcinogens, certain tumour promoters and hepatotoxin (Yang *et al*., 2010; Kaewkod *et al*., 2019). According to Yang and others (2010), *Glunacetobacter* sp. A4 has a strong ability to produce DSL and was the key functional bacterial species isolated from preserved kombucha. From the study, they recorded the highest amount of DSL during the 8th day of the fermentation process and gradually decreased until the end of the process. Another study has shown the content of DSL in kombucha with a range of 57.99-132.72 mg/mL (Wang *et al.,* 2010). Also, a study shows the presence of DSL as well as other organic acids which are glucuronic, gluconic, ascorbic, acetic, and succinic acids by using the HPLC system (Kaewkod *et al.,* 2019). During the fermentation process, sucrose is hydrolysed by yeast into glucose and fructose which is a monomer for sucrose and carbon dioxide which leads to a carbonation sensation towards the product. Yeast also produces ethanol in the process which is oxidised into acetaldehyde. The combination of acetic acid and ethanol results in an increased rate of ethanol production via yeast (Mousavi *et al.,* 2020). The study shows the amount of ethanol (5.5 g/L) with an initial sucrose content of 100 g/L with a fermentation duration of 20 days. There is also a study which involves different types of kombucha which involve tea, rice and barley and all those samples show the presence of ethanol. All of the sample shows a high amount of ethanol from the 1st until the 4th day and gradually decreases (Ahmed *et al.,* 2020). Chen and Liu (2000) established that the content of ethanol increased with time and reached the highest value at around 5.5 g/L, followed by a slow decline (Chen and Liu, 2000). As mentioned before, the relationship between acetic acid and ethanol is vital in the production of kombucha tea.

The dual relationship can act as an antimicrobial agent against pathogenic bacteria (Mousavi *et al.,* 2020). This component is also responsible for vinegar's acidic odour and flavour. There was research that showed acetic acid progressively increasing to 11 g/L at 30 days of fermentation and gradually decreasing to 8 g/L after 60 days of fermentation. This occurs owing to the use of acetic acid as a carbon source for bacteria in kombucha when the substrate in the combination, sucrose, has been depleted, or due to a reduction in ethanol metabolism by yeast due to low pH (Chen and Liu, 2000).

6. Alcohol content

Generally, Muslims are required to follow specific dietary laws and practices which say which foods are permissible or prohibited for consumption based on the Quran and Sunnah. Malaysia was one of the pioneers in creating halal food laws in the early 1980s which has become the baseline for other countries in establishing halal food laws (Riaz and Chaudry, 2003). Among these prohibited foods and drinks, the use of alcohol is one of the most controversial and arguable issues on various platforms (Al-Mazeedi *et al*., 2013; Ahmad *et al*., 2014). As mentioned in the above content, alcohol is one of the products in the fermentation process even during the second phase of fermentation which involves yeast and more specifically ethyl alcohol or ethanol which is colourless, highly volatile and has a density lower than water (Rasu Jayabalan *et al*., 2014; Hendriks, 2020; Laureys *et al.,* 2020). Ethanol is a hygroscopic liquid which usually not 100% pure and has a chemical structure of two carbon atoms which bond with the hydroxyl group. As mentioned above, the production of the chemical component in Kombucha tea mostly depends on multiple factors such as the symbiotic culture used for the fermentation process, the duration of fermentation, sucrose content and the type of tea used. There is specific guidance on the manufacturing of kombucha tea due to excessive production of ethanol which may be hazardous for consumption. This is due to incomplete fermentation of residual sugars and ethanol, the presence of flavouring agents which contain sugar which will interrupt the fermentation stage and the absence of pasteurization of the final fermented product which allows the survival of yeast (Mcintyre and Jang, 2020). One of the methods to avoid over-production of alcohol is by adding inhibitors such as sodium benzoate or potassium sorbate before bottling Nevertheless, there is still a lack of scientific information related towards the concentration of ethanol throughout the years.

Most of the studies that have been done to prove the availability of alcohol in kombucha tea mostly contain 0.5-0.7%. Based on the discussion between the expert of Halal Products Research Institute, Universiti Putra Malaysia (UPM), with Fatwa Committee of the National Council for Islamic Affairs Malaysia, has reached a new agreement related to alcoholic beverages. From that discussion, it stated that beverages which are produced without the intention of producing alcohol are still permissible to drink if the alcohol level is below 1%(v/v) and foods or drinks containing natural alcohol such as fruits, nuts or grains and its juice, or alcohol produced as a by-product during the manufacturing process of food or drink is not najs and permissible to be eaten or drink (Arshad and Mokhtar, 2018). From that agreement, it shows that most Kombucha tea production is still permissible for consumption if the level of ethanol does not exceed 0.5% ABV and any food or drinks which contain natural alcohol as a by-product during the production of the product is considered as not najs and permissible to consume (Arshad and Mokhtar, 2018). Few kinds of research have been conducted to evaluate the quantity of ethanol in Kombucha drinks, and the majority of them reveal an average of 0.5% ABV. One research found that the ethanol content in green and black tea Kombucha tea was roughly 0.2-0.3% on the 15th day (Kallel *et al.,* 2012). Another study has been conducted by Chakravorty which focuses on black tea kombucha. That study shows an increase in ethanol concentration with a maximum value of 0.028% on the 7th day, followed by a decrease to 0.0073% on the 21st day and this phenomenon may be due to the presence of acetic bacteria thus the production of acetic acid (Chakravorty *et al.,* 2016). The analysis to investigate the concentration of ethanol in kombucha tea also involves different temperature conditions during the production period and it shows higher ethanol content which reached 1.1% at 25°C (Neffe-Skocińska *et al.,* 2017). There is one study which analyses the ethanol concentration after bottling or packaging. From the study, it shows an increase in ethanol concentration from 7-14 days with a maximum amount of 1.57% and it may be due to the production of carbon dioxide which is one of the main by-products of the fermentation process in kombucha tea production which accumulate thus inhibit the conversion of ethanol to acetic acid (Nummer, 2013). In other conditions in which the tea is being stored at 4°C temperature, the ethanol level will increase and remain unchanged on the 14th day which supports the hypothesis that refrigeration does help to stabilize alcohol production and microbial metabolism (Bamforth, 2014). The concentration of ethanol in kombucha tea may be low and may not be aware by the consumer. Nevertheless, this still can be considered as dangerous to people who are pregnant and have younger children. Children particularly smaller-weight infants and toddlers have a lower tolerance to alcohol which eliminates alcohol from their body system due to their liver not metabolising ethanol as efficiently as an adult which can cause rapid alcohol intoxication which leads to hypoglycemia. For pregnant women, it is medically advised to abstain from consuming alcohol to avoid fetal alcohol spectrum disorder which can harm the baby (Chudley and Longstaffe, 2010; Mcintyre and Jang, 2020). In term of method to determine the concentration of alcohol in kombucha tea is different compared to other alcoholic beverages due to the low concentration of ethanol which is less than 5% ABV. The refractometer method and hydrometer method is one of the common methods used in the beer and wine industry but it only can provide an approximate of alcohol in kombucha tea and it is not recommended. The method uses headspace gas chromatography (GC) with flame ionization detection (HS-GCFID) or mass spectrometry (HS-GCMS) which is a more accurate detection due to its ability to eliminate other compounds (Mcintyre and Jang, 2020).

7. Health benefits of kombucha tea

Kombucha's protective ability mainly comes from the presence of organic acid and antioxidant compounds found during microbial fermentation which effectively fight against free radical scavengers (Chakravorty *et al*., 2016; De Miranda *et al.*, 2021). High concentration of phytochemical components mainly phenolic compounds in Kombucha tea plays an important role in the protective characteristic of the beverage towards the consumers. Furthermore, they serve critical roles in the prevention of various major diseases associated with oxidative stress, including cardiovascular disease, cancer, and neurological disorders (Manach *et al*., 2004; Leal *et al*., 2018). Generally, it assists in the production and activity of most enzyme and cell receptors which resulting a defense mechanism against oxidative stress caused by reactive oxygen species (Tsao, 2010; Bakar *et al*., 2018; Leal *et al*., 2018). An *in vivo* study has been done which proves the reduction in glycosylate haemoglobin (HbAIc) and increase in plasma insulin, haemoglobin, and tissue glycogen levels in diabetic rats which have been induced with lyophilized black tea kombucha over 45 days of treatment (Srihari, Karthikesan, Ashokkumar *et al*., 2013; De Miranda *et al*., 2021). A noticeable drop in enzymatic activities such as glucose-6-phosphatase, fructose-1, 6-bisphosphatase, and hexokinase, which are the main enzymes responsible for the control of glycolysis and gluconeogenesis in liver and muscle, was also observed, implying that kombucha has the potential to act at multiple points in the glucose regulatory pathways (Srihari, Karthikesan, Ashokkumar *et al*., 2013; De Miranda *et al*., 2021). Another study shows a potential antidiabetic effect of kombucha tea in diabetic rats which was reduced by approximately 56.4% after 14 days of treatment with kombucha extract (Bhattacharya *et al.,* 2013). Another recent *in vivo* study which involves rats and mice proved that kombucha tea may consist of healthy prophylactic and recovery effects through multiple processes such as immune stimulation, detoxification, antimicrobial activity and antioxidant component (Baschali *et al.,* 2017). One study compared black tea kombucha and enzyme-processed tea with fungus against CCl4-induced hepatotoxicity in rats due to the antioxidant produced during the fermentation process and the result shows that black tea kombucha was more efficient (Baschali *et al.,* 2017).

Cancer is one of the most common causes of today's worldwide death which is still being managed by anticancer drugs. A new alternative to reduce the chance of getting cancer would be beneficial (Abu Bakar *et al*., 2015; Gholami, 2017). The Russian Academy of Science in Moscow and the Central Oncological Research Unit in Russia have done research and they have proven that this beverage contains beneficial components to inhibit cancer cells (Kapp and Sumner, 2019). Complications related to more severe diseases mostly diabetes and cancer mainly related to oxidative stress and radical scavengers which in turn can be suppressed or minimized with the presence of bioactive compounds (Srihari, Arunkumar, Arunakaran *et al.,* 2013; De Miranda *et al.,* 2021). A study which involves diabetic rats induced by streptozotocin shows a reduction in glycosylated haemoglobin, and tissue glycogen levels when being treated with black tea kombucha after 43 days of treatment (Srihari, Karthikesan, Ashokkumar *et al*., 2013). It shows a great reduction of enzymes which are mainly responsible for glycolysis and gluconeogenesis in the liver and muscle. Another result shows that kombucha tea exerts an antidiabetic effect in alloxan-induced diabetic rats when it shows a significant reduction in blood glucose by 54.6% after being treated with kombucha extract for 14 days (Srihari, Karthikesan, Ashokkumar *et al*., 2013). A study compared black tea kombucha tea and green tea kombucha tea towards cancer cells and it favours green tea kombucha tea due to reducing abilities towards GI50 values mostly it can be explained by a higher amount of catechins compound and the presence of verbascoside (Cardoso *et al.,* 2020).

Catechins compound has already known for their great potential for protection against the development of specific cancer cells by disrupting the process of the development of cancer cells by inhibiting the enzymatic process (Yang and Wang, 2016; Ali *et al*., 2020) while verbascoside have a potential to inhibiting cell growth and promote apoptosis in in vitro and in vivo models which also have the potential as a therapeutic agent in human (Zhang *et al.,* 2018).

Tea polyphenols also have been proven to be the most impact towards anticancers by inhibiting the propagation of cancer cells (Rasu Jayabalan *et al*., 2011, 2014). The other beneficial chemical component such as vitamin C, glucuronic acid, gluconic acid, and lactic acid also helps in diminishing the incidence of stomach cancer (Xia *et al.,* 2019). The US Food and Drug Administration confirmed that there is no harmful effect of this product by conducting an *in vivo* study in mice for 3 months (Mousavi *et al.,* 2020). The ability of kombucha tea as an antiproliferation agent has been proven in several human tumour cells such as HeLa cells (epithelial carcinoma of the cervix), HT-29 (adenocarcinoma of the colon), MCF-7 (breast adenocarcinoma), Hep-2 (human laryngeal epidermoid carcinoma), PC-3 (prostate cancer) (Srihari, Arunkumar, Arunakaran *et al.,* 2013), RD (human rhabdomyosarcoma-derived cell line), Hep2c (HeLa-derived human cervical carcinoma- derived cell line), A549 (pulmonary adenocarcinoma), HCT8 (human ileocecal colorectal adenocarcinoma), and Caco-2 (colorectal adenocarcinoma) (Uțoiu *et al*., 2018; Vitas *et al*., 2018; Cardoso *et al*., 2020; De Miranda *et al*., 2021).

Even though kombucha fermentation does not undergo a complicated pathway, it does produce several beneficial compounds which in this issue are catechin and polyphenols. Polyphenols have the ability to scavenge free radical's reactive oxygen species (ROS) which resulting kombucha tea as a potent antioxidant agent. There is a student who has proven that the presence of complex phenolic compounds in an acidic environment can lead to the splitting of large and complex molecules into smaller molecules which increases the amount of phenolic compounds in kombucha tea (Srihari and Satyanarayana, 2012; Mousavi *et al*., 2020)

Even though the presence of yeast and bacteria is one of the primary ingredients of kombucha tea, it is also has been proven that kombucha tea has a supreme antimicrobial effect on a wide range of microorganisms including Gram-negative bacteria and Gram-positive bacteria (Vohra et al., 2019; Dufresne and Farnworth, 2000). *Pseudomonas aeruginosa, Agrobacterium tumefaciens, Helicobacter pylori, Enterobacter cloacae, Salmonella enterica* serovar Enteritidis*, Escherichia coli, Yersinia enterocolitica, Candida albicans, Shigella sonnei, Campylobacter jejuni,* and *Staphylococcus aureus* are only a few of the microbes which already being proven to effected by kombucha tea regarding their growth rate (Ahmed and Dirar, 2005). Protein and acetic acid found in kombucha have antibiotics that lead to an increase in antimicrobial performance which can also lead to utilized as an agent to reduce the pathogens associated with human illness (St-Pierre, 2019).

8. Conclusion

In terms of its various uses, it has been suggested that Kombucha tea consists and can be a source of bioactive compounds which can be beneficial towards consumers including lowering blood pressure, improving digestion, and managing arthritis. It can also be used for treating various serious illnesses such as cancer and diabetes. It is also claimed that this drink can help maintain a healthy weight loss and reduce the risk of various diseases. It can also help in the recovery of haemorrhoids and reduce the symptoms of various infections. Despite the numerous articles claiming that kombucha has miraculous effects, further studies are still needed to confirm these claims. Also, its main fermentation inoculum, SCOBY, is not well understood to date. An excessive amount of toxins, such as cyanogenic glycosides and phytates, can occur during the fermentation process of Kombucha. Although many countries have regulations for the production of high-volume products, such as Kombucha, they do not seem to be sufficient for the development of novel compounds. Various approaches have been suggested for organizing an assembly of experts. However, these approaches are still not yet fully explored and several methods can be implied to improve the outcome.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgements

The study received support from Universiti Tun Hussein Onn Malaysia (UTHM) through the GPPS, with the grant code H701. We would also like to recognize the work of Mohd Mirza bin Mohd Basyir in the design of several figures and the overall editing process.

References

Abu Bakar, M.F., Ahmad, N.E., Suleiman, M., Rahmat, A. and Isha, A. (2015) *Garcinia dulcis* fruit extract induced cytotoxicity and apoptosis in HepG2 liver cancer cell line. *BioMed Research International* 2015, 916902. https://doi.org/10.1155/2015/916902

Abu Bakar, M.F., Mohamed, M., Rahmat, A., Burr, S.A. and Fry, J.R. (2010). Cytotoxicity and polyphenol diversity in selected parts of *Mangifera pajang* and *Artocarpus odoratissimus* fruits. *Nutrition and Food Science* 40(1), 29–38. https://

doi.org/10.1108/00346651011015890

Abu Bakar, M.F., Teh, A.H., Rahmat, A., Hashim, N., Othman, F. and Fakurazi, S. (2006). Antiproliferative properties and antioxidant activity of various types of *Strobilanthes crispus* tea. International *Journal of Cancer Research*, 2(2), 152158. http://doi.org/10.3923/ijcr.2006.152.158

Admassie, M. (2018). A Review on Food Fermentation and the Biotechnology of Lactic Acid Bacteria. *World Journal of Food Science and Technology*, 2 (1), 19-24. https://doi.org/10.11648/j.wjfst.20180201.13

Ahmad, A.N., Yang, T.A. and Abdullah, W.N.W. (2014). Perceived versus actual knowledge of alcohol and halal food among food technology undergraduate students in a Malaysian university Anis Najiha Ahmad Tajul A. Yang Wan Nadiah Wan Abdullah. *Journal of Islamic Marketing*, 6(3), 294–313. https://doi.org/10.1108/JIMA-10-2013-0069

Ahmed, R.F., Hikal, M.S. and Abou-taleb, K.A. (2020). Annals of Agricultural Sciences Biological, chemical and antioxidant activities of different types Kombucha. *Annals of Agricultural Sciences*, 65, 35-41. https://doi.org/10.1016/j.aoas.2020.04.001

Ahmed, S.E. and Dirar, H.A. (2005). Studies on the Microbiology of Kombucha (Tea Fungus). *University of Khartoum Journal of Agricultural Sciences*, 13(1), 68–84.

Ali, H., Abu Bakar, M.F., Majid, M., Muhammad, N. and Lim, S.Y. (2020). In vitro anti-diabetic activity of stingless bee honey from different botanical origins. *Food Research,* 4(5), 1421–1426. https://doi.org/10.26656/fr.2017.4(5).411

Al-Mazeedi, H. M., Regenstein, J.M. and Riaz, M.N. (2013). The issue of undeclared ingredients in halal and kosher food production: A focus on processing aids. *Comprehensive Reviews in Food Science and Food Safety*, 12(2), 228–233. https://doi.org/10.1111/1541-4337.12002

Arshad, S. and Mokhtar, N.F.K. (2018). Alcohol: Halal or Haram? What Is Alcohol and Khamar ? Retrieved from website: https://halal.upm.edu.my/artikel/alcohol halal_or_haram_what_is_alcohol_and_khama r_-42087

Bakar, F.I.A., Bakar, M.F.A., Abdullah, N., Endrini, S., Rahmat, A.A. (2018). Review of Malaysian Medicinal Plants with Potential Anti-Inflammatory Activity. *Advances in Pharmacological Sciences*, 2018, 8603602. https://doi.org/10.1155/2018/8603602

Balentine, D.A. (1997). Antioxidants in tea. *Critical Reviews in Food Science and Nutrition*, 37(8), 691–692. https://doi.org/10.1080/10408399709527796

Bamforth, C.W. (2014). Fermented Beverages. In Encyclopedia of Agriculture and Food Systems, p. 124–136. Elsevier Online Reference Module in Food Science. https://doi.org/10.1016/B978-0-444-52512-3.00154-6

Baschali, A., Tsakalidou, E., Kyriacou, A., Karavasiloglou, N. and Matalas, A.L. (2017). Traditional low-alcoholic and non-alcoholic fermented beverages consumed in European countries: A neglected food group. *Nutrition Research Reviews,* 30(1), 1 – 24. https://doi.org/10.1017/S0954422416000202

Bauer-Petrovska, B. and Petrushevska-Tozi, L. (2000). Mineral and water soluble vitamin content in the Kombucha drink. *International Journal of Food Science and Technology*, 35(2), 201–205. https://doi.org/10.1046/j.1365-2621.2000.00342.x

Bhattacharya, S., Gachhui, R. and Sil, P.C. (2013). Effect of Kombucha, a fermented black tea in attenuating oxidative stress mediated tissue damage in alloxan induced diabetic rats. *Food and Chemical Toxicology*, 60, 328–340. https://doi.org/10.1016/j.fct.2013.07.051

Bokuchava, M.A., Skobeleva, N.I. and Sanderson, G.W. (1980). The biochemistry and technology of tea manufacture. *Critical Reviews in Food Science and Nutrition*, 12(4), 303-370. https://doi.org/10.1080/10408398009527280

Cardoso, R.R., Neto, R.O., dos Santos D’Almeida, C.T., do Nascimento, T.P., Pressete, C.G., Azevedo, L., Martino, H.S.D., Cameron, L.C., Ferreira, M.S.L. and Barros, F.A.R.D. (2020). Kombuchas from green and black teas have different phenolic profile, which impacts their antioxidant capacities, antibacterial and antiproliferative activities. *Food Research International*, 128, 108782. https://doi.org/10.1016/j.foodres.2019.108782

Chakravorty, S., Bhattacharya, S., Chatzinotas, A., Chakraborty, W., Bhattacharya, D. and Gachhui, R. (2016). Kombucha tea fermentation: Microbial and biochemical dynamics. *International Journal of Food Microbiology*, 220, 63–72. https://doi.org/10.1016/j.ijfoodmicro.2015.12.015

Chen, C. and Liu, B.Y. (2000). Changes in major components of tea fungus metabolites during prolonged fermentation. *Journal of Applied Microbiology*, 89(5), 834–839. https://doi.org/10.1046/j.1365-2672.2000.01188.x

Chilton, S.N., Burton, J.P. and Reid, G. (2015). Inclusion of fermented foods in food guides around the world.

Nutrients, 7(1), 390–404. https://doi.org/10.3390/nu7010390

Chu, S.C. and Chen, C. (2006). Effects of origins and fermentation time on the antioxidant activities of kombucha. *Food Chemistry*, 98(3), 502–507. https://doi.org/10.1016/j.foodchem.2005.05.080

Chudley, A.E. and Longstaffe, S.E. (2010). Fetal Alcohol Syndrome and Fetal Alcohol Spectrum Disorder. In Cassidy, S.B. and Allanson, J.E. (Eds.) Management of Genetic Syndromes. UK: Wiley Online. https://doi.org/10.1002/9780470893159.ch25

De Miranda, J.F., Ruiz, L.F., Silva, C.B., Uekane, T.M., Silva, K.A., Gonzalez, A.G.M., Fernandes, F.F. and Lima, A.R. (2021). Kombucha: A review of substrates, regulations, composition, and biological properties. *Journal of Food Science*, 87(2), 503-527. https://doi.org/10.1111/1750-3841.16029

Derrien, M. and van Hylckama Vlieg, J.E.T. (2015). Fate, activity, and impact of ingested bacteria within the human gut microbiota. *Trends in Microbiology*, 23(6), 354–366. https://doi.org/10.1016/j.tim.2015.03.002

Dimidi, E., Cox, S., Rossi, M. and Whelan, K. (2019). Fermented Foods: Definitions and Characteristics, Gastrointestinal Health and Disease. *Nutrients*, 11, 1806.

Dufresne, C. and Farnworth, E. (2000). Tea, Kombucha, and health: A review. *Food Research International*, 33(6), 409–421. https://doi.org/10.1016/S0963-9969(00)00067-3

Filannino, P., Bai, Y., Di Cagno, R., Gobbetti, M. and Gänzle, M.G. (2015). Metabolism of phenolic compounds by *Lactobacillus* spp. during fermentation of cherry juice and broccoli puree. *Food Microbiology*, 46, 272–279. https://doi.org/10.1016/j.fm.2014.08.018

Gaggìa, F., Baffoni, L., Galiano, M., Nielsen, D. S., Jakobsen, R.R., Castro-Mejía, J.L., Bosi, S., Truzzi, F., Musumeci, F., Dinelli, G. and Di Gioia, D. (2019). Kombucha beverage from green, black and rooibos teas: A comparative study looking at microbiology, chemistry and antioxidant activity. *Nutrients*, 11, 1. https://doi.org/10.3390/nu11010001

Gholami, A. (2017). Suicide gene therapy: A special focus on progress and concerns about cancer treatment. *Trends in Pharmaceutical Sciences*, 3(4), 221-236.

Gomes, R.J., Borges, M.D.F., Rosa, M.D.F., Castro-Gómez, R.J.H. and Spinosa, W.A. (2018). Acetic acid bacteria in the food industry: Systematics, characteristics and applications. *Food Technology and Biotechnology*, 56(2), 139–151. https://doi.org/10.17113/ftb.56.02.18.5593

Harbowy, M.E., Balentine, D.A., Davies, A.P. and Cai, Y. (1997). Tea Chemistry. *Critical Reviews in Plant Sciences*, 16(5), 415–480. https://doi.org/10.1080/07352689709701956

Hartmann, A.M., Burleson, L.E., Holmes, A.K. and Geist, C.R. (2000). Effects of chronic 60 kombucha ingestion on open-field behaviors, longevity, appetitive behaviors, and organs 605 in C57-BL/6 mice: A pilot study. *Nutrition*, 16(9), 755–761. https://doi.org/10.1016/S0899-9007(00)00380-4

Hendriks, H.F.J. (2020). Alcohol and Human Health: What Is the Evidence? In *Annual Review of Food Science and Technology*, 11, 1-21. https://doi.org/10.1146/annurev-food-032519-051827

Ismaiel, A.A., Bassyouni, R.H., Kamel, Z. and Gabr, S.M. (2016). Detoxification of Patulin by Kombucha tea culture. *CYTA - Journal of Food*, 14(2), 271–279. https://doi.org/10.1080/19476337.2015.1096828

Ivanišová, E., Meňhartová, K., Terentjeva, M., Harangozo, Ľ., Kántor, A. and Kačániová, M. (2020). The evaluation of chemical, antioxidant, antimicrobial and sensory properties of 612 kombucha tea beverage. *Journal of Food Science and Technology*, 57(5), 1840–1846. https://doi.org/10.1007/s13197-019-04217-3

Jakubczyk, K., Kałduńska, J., Kochman, J. and Janda, K. (2020). Chemical profile and antioxidant activity of the kombucha beverage derived from white, green, black and red tea. *Antioxidants*, 9(5), 447. https://doi.org/10.3390/antiox9050447

Jarrell, J., Cal, T. and Bennett, J.W. (2000). The Kombucha consortia of yeasts and bacteria. *Mycologist*, 14(4), 166–170. https://doi.org/10.1016/S0269-915X(00)80034-8

Jayabalan, R., Marimuthu, S. and Swaminathan, K. (2007). Changes in content of organic 619 acids and tea polyphenols during kombucha tea fermentation. *Food Chemistry*, 102(1), 392–398. https://doi.org/10.1016/j.foodchem.2006.05.032

Jayabalan, Rasu, Chen, P.N., Hsieh, Y.S., Prabhakaran, K., Pitchai, P., Marimuthu, S., Thangaraj, P., Swaminathan, K. and Yun, S.E. (2011). Effect of solvent fractions of kombucha tea on viability and invasiveness of cancer cells-characterization of dimethyl 2-(2-hydroxy-2-methoxypropylidine) malonate and vitexin. *Indian Journal of Biotechnology*, 10(1), 75–82.

Jayabalan, Rasu, Malbaša, R.V., Lončar, E.S., Vitas, J.S. and Sathishkumar, M. (2014). A review on kombucha tea-microbiology, composition, fermentation, beneficial effects, 628 toxicity, and tea

REVIEW

fungus. *Comprehensive Reviews in Food Science and Food Safety*,13(4), 538–550. https://doi.org/10.1111/1541-4337.12073

Kaewkod, T., Bovonsombut, S. and Tragoolpua, Y. (2019). Efficacy of kombucha obtained from green, oolong and black teas on inhibition of pathogenic bacteria, antioxidation, and toxicity on colorectal cancer cell line. *Microorganisms*, 7(12), 1–18. https://doi.org/10.3390/microorganisms7120700

Kallel, L., Desseaux, V., Hamdi, M., Stocker, P. and Ajandouz, E.H. (2012). Insights into the fermentation biochemistry of Kombucha teas and potential impacts of Kombucha drinking on starch digestion. *Food Research International*, 49(1), 226–232. https://doi.org/10.1016/j.foodres.2012.08.018

Kapp, J.M. and Sumner, W. (2019). Kombucha: a systematic review of the empirical evidence of human health benefit. *Annals of Epidemiology*, 30, 66–70. https://doi.org/10.1016/j.annepidem.2018.11.001

Katiyar, S.K. and Mukhtar, K. (1996). Tea in chemoprevention of cancer: Epidemiologic and experimental studies (Review). *International Journal of Oncology*, 8(2), 221–238. https://doi.org/10.3892/ijo.8.2.221

Laureys, D., Britton, S. J. and De Clippeleer, J. (2020). Kombucha Tea Fermentation: A Review. *Journal of the American Society of Brewing Chemists*, 78(3), 165–174. https://doi.org/10.1080/03610470.2020.1734150

Leal, J.M., Suárez, L.V., Jayabalan, R., Oros, J.H. and Escalante-Aburto, A. (2018). A review on health benefits of kombucha nutritional compounds and metabolites. *CYTA-Journal of Food*, 16(1), 390–399. https://doi.org/10.1080/19476337.2017.1410499

Lin, J.K., Lin, C.L., Liang, Y.C., Lin-Shiau, S.Y. and Juan, I.M. (1998). Survey of Catechins, Gallic Acid, and Methylxanthines in Green, Oolong, Pu-erh, and Black Teas. *Journal of Agricultural and Food Chemistry*, 46(9), 3635–3642. https://doi.org/10.1021/jf980223x

Lin, Y.L., Juan, I.M., Chen, Y.L., Liang, Y.C. and Lin, J.K. (1996). Composition of polyphenols in fresh tea leaves and associations of their oxygen-radical-absorbing 657 capacity with antiproliferative actions in fibroblast cells. *Journal of Agricultural and Food Chemistry*, 44(6), 1387–1394. https://doi.org/10.1021/jf950652k

Łuczaj, W. and Skrzydlewska, E. (2005). Antioxidative properties of black tea. *Preventive Medicine*, 40 (6), 910–918. https://doi.org/10.1016/j.ypmed.2004.10.014

Malbaša, R., Lončar, E. and Djurić, M. (2008). Comparison of the products of Kombucha fermentation on sucrose and molasses. *Food Chemistry*, 106(3), 1039–1045. https://doi.org/10.1016/j.foodchem.2007.07.020

Manach, C., Scalbert, A., Morand, C., Rémésy, C. and Jiménez, L. (2004). Polyphenols: Food sources and bioavailability. *American Journal of Clinical Nutrition*, 79(5), 727–747. https://doi.org/10.1093/ajcn/79.5.727

Marco, M.L., Heeney, D., Binda, S., Cifelli, C.J., Cotter, P.D., Foligné, B., Gänzle, M., Kort, R., Pasin, G., Pihlanto, A., Smid, E.J. and Hutkins, R. (2017). Health benefits of fermented foods: microbiota and beyond. *Current Opinion in Biotechnology*, 44, 94-102. https://doi.org/10.1016/j.copbio.2016.11.010

Mayser, P., Fromme, S., Leitzmann, G. and Gründer, K. (1995). The yeast spectrum of the 'tea fungus Kombucha': Das Hefespektrum des 'Teepilzes Kombucha.' *Mycoses*, 38(7–8), 289–295. https://doi.org/10.1111/j.1439-0507.1995.tb00410.x

Mcintyre, L. and Jang, S.S. (2020). A study of Alcohol Levels in Kombucha Products in British Columbia. British Columbia: BC Centre for Disease Control.

Mousavi, S.M., Hashemi, S.A., Zarei, M., Gholami, A., Lai, C.W., Chiang, W.H., Omidifar, N., Bahrani, S. and Mazraedoost, S. (2020). Recent Progress in Chemical Composition, Production, and Pharmaceutical Effects of Kombucha Beverage: A Complementary and Alternative Medicine. *Evidence -Based Complementary and Alternative Medicine*, 2020(1), 4397543. https://doi.org/10.1155/2020/4397543

Neffe-Skocińska, K., Sionek, B., Ścibisz, I. and Kołożyn-Krajewska, D. (2017). Acid contents and the effect of fermentation condition of Kombucha tea beverages on physicochemical, microbiological and sensory properties. *CYTA - Journal of Food*, 15(4), 601–607. https://doi.org/10.1080/19476337.2017.1321588

Nummer, B.A. (2013). Kombucha brewing under the Food and Drug Administration model Food Code: risk analysis and processing guidance. *Journal of Environmental Health*, 76(4), 8–11.

Pessione, E. and Cirrincione, S. (2016). Bioactive molecules released in food by lactic acid bacteria: Encrypted peptides and biogenic amines. *Frontiers in Microbiology*, 7, 876. https://doi.org/10.3389/fmicb.2016.00876

Poonam, V., Archita, M., Deepali, S., Hemant, G. and Himanshu, S.K. (2018). A review on green tea: A miraculous drink. *International Journal of Pharmaceutical Sciences Review and Research*, 51 (2), 26–34.

Reiss, J. (1994). Influence of different sugars on the metabolism of the tea fungus. *Zeitschrift Für Lebensmittel-Untersuchung Und-Forschung*, 198(3), 258–261. https://doi.org/10.1007/BF01192606

Rezac, S., Kok, C.R., Heermann, M. and Hutkins, R. (2018). Fermented foods as a dietary source of live organisms. *Frontiers in Microbiology*, 9, 1785. https://doi.org/10.3389/fmicb.2018.01785

Riaz, M.N. and Chaudry, M.M. (2003). Halal Food Production. Boca Raton, USA: CRC Press. https://doi.org/10.1201/9780203490082

Rodrigues, F., Ludovico, P. and Leão, C. (2006). Sugar Metabolism in Yeast: an Overview of Aerobic and Anaerobic Glucose Catabolism. In Péter, G. and Rosa, C. (Eds.) Biodiversity and Ecophysiology of Yeasts. Heidelberg, Germany: Springer Berlin.

Sharangi, A.B. (2009). Medicinal and therapeutic potentialities of tea (*Camellia sinensis* L.) - A review. *Food Research International*, 42(5–6), 529–535. https://doi.org/10.1016/j.foodres.2009.01.007

Sievers, M. and Swings, J. (2015). *Acetobacteraceae*. In Bergey's Manual of Systematics of Archaea and Bacteria. Reference Work Online. UK: John Wiley and Sons, Inc. in association with Bergey's Manual Trust. https://doi.org/10.1002/9781118960608.fbm00174

Srihari, T. and Satyanarayana, U. (2012). Changes in free radical scavenging activity of Kombucha during fermentation. *Journal of Pharmaceutical Sciences and Research*, 4(11), 1978–1981.

Srihari, T., Arunkumar, R., Arunakaran, J. and Satyanarayana, U. (2013). Downregulation of signalling molecules involved in angiogenesis of prostate cancer cell line (PC-3) by kombucha (lyophilized). *Biomedicine and Preventive Nutrition*, *3*(1), 53–58. https://doi.org/10.1016/j.bionut.2012.08.001

Srihari, T., Karthikesan, K., Ashokkumar, N. and Satyanarayana, U. (2013). Antihyperglycaemic efficacy of kombucha in streptozotocin-induced rats. *Journal of Functional Foods*, 5(4), 1794–1802. https://doi.org/10.1016/j.jff.2013.08.008

St-Pierre, D.L. (2019). Microbial Diversity of the Symbiotic Colony of Bacteria and Yeast (SCOBY) and its Impact on the Organoleptic Properties of Kombucha. USA: University of Maine, MSc. Thesis.

Tsao, R. (2010). Chemistry and biochemistry of dietary polyphenols. *Nutrients*, 2(12), 1231–1246. https://doi.org/10.3390/nu2121231

Uțoiu, E., Matei, F., Toma, A., Diguță, C.F., Ștefan, L.M., Mănoiu, S., Vrăjmașu, V.V., Moraru, I., Oancea, A., Israel-Roming, F., Cornea, C.P., Constantinescu-Aruxandei, D., Moraru, A. and Oancea, F. (2018). Bee collected pollen with enhanced health benefits, produced by fermentation with a Kombucha Consortium. *Nutrients*, 10(10), 1365. https://doi.org/10.3390/nu10101365

Villarreal-Soto, S.A., Beaufort, S., Bouajila, J., Souchard, J.P., Renard, T., Rollan, S. and Taillandier, P. (2019). Impact of fermentation conditions on the production of bioactive compounds with anticancer, anti-inflammatory and antioxidant properties in kombucha tea extracts. *Process Biochemistry*, 83, 44-54. https://doi.org/10.1016/j.procbio.2019.05.004

Villarreal-Soto, S.A., Beaufort, S., Bouajila, J., Souchard, J.P. and Taillandier, P. (2018). Understanding Kombucha Tea Fermentation: A Review. *Journal of Food Science*, 83(3), 580–588. https://doi.org/10.1111/1750-3841.14068

Vitas, J.S., Cvetanović, A.D., Mašković, P.Z., Švarc-Gajić, J.V. and Malbaša, R.V. (2018). Chemical composition and biological activity of novel types of kombucha beverages with yarrow. *Journal of Functional Foods*, 44, 95–102. https://doi.org/10.1016/j.jff.2018.02.019

Vitas, J.S., Malbaša, R.V., Grahovac, J.A. and Lončar, E.S. (2013). The Antioxidant Activity of Kombucha Fermented Milk Products with Stinging Nettle and Winter Savory. *Chemical Industry and Chemical Engineering Quarterly*, 19(1), 129–139. https://doi.org/10.2298/CICEQ120205048V

Vohra, B., Fazry, S., Sairi, F. and Othman, B.A. (2019). Effects of medium variation and fermentation time towards the pH level and ethanol content of Kombucha. *AIP Conference Proceedings*, 2111, 040008. https://doi.org/10.1063/1.5111247

Wang, K., Gan, X., Tang, X., Wang, S. and Tan, H. (2010). Determination of D-saccharic acid-1,4-lactone from brewed kombucha broth by high-performance capillary electrophoresis. *Journal of Chromatography B*, 878(3–4), 371–374. https://doi.org/10.1016/j.jchromb.2009.12.003

Xia, X., Dai, Y., Wu, H., Liu, X., Wang, Y., Yin, L., Wang, Z., Li, X. and Zhou, J. (2019). Kombucha fermentation enhances the health-promoting properties of soymilk beverage. *Journal of Functional Foods*, 62, 103549. https://doi.org/10.1016/j.jff.2019.103549

Yang, C.S. and Wang, H. (2016). Cancer preventive activities of tea catechins. *Molecules*, 21(12), 1679. https://doi.org/10.3390/molecules21121679

Yang, Z., Zhou, F., Ji, B., Li, B., Luo, Y., Yang, L. and Li, T. (2010). Symbiosis between microorganisms from kombucha and kefir: Potential significance to the enhancement of kombucha function. *Applied Biochemistry and Biotechnology*, 160(2), 446–455. https://doi.org/10.1007/s12010-008-8361-6

Yann, D. and Pauline, G. (2014). Usefulness of Natural Starters in Food Industry: The Example of Cheeses and Bread. *Food and Nutrition Sciences*, 5(17), 1679–1691. https://doi.org/10.4236/fns.2014.517181

Zhang, Y., Yuan, Y., Wu, H., Xie, Z., Wu, Y., Song, X., Wang, J., Shu, W., Xu, J., Liu, B., Wan, L., Yan, Y., Ding, X., Shi, X., Pan, Y., Li, X., Yang, J., Zhao, X. and Wang, L. (2018). Effect of verbascoside on apoptosis and metastasis in human oral squamous cell carcinoma. *International Journal of Cancer*, 143(4), 980–991. https://doi.org/10.1002/ijc.31378

