10

KOMBUCHA: A PROMISING FUNCTIONAL BEVERAGE PREPARED FROM TEA

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10.1 Introduction

All living forms require food for their existence. Water, which constitutes ~70% of our earth's mass, is indispensable for sustenance of life on earth (USGS, 2016a,b). Hence, water which constitutes ~60% of the body mass of an average adult is the most consumed beverage of the world (USGS, 2016a,b). With the advancement of civilization, man has formulated innumerable diverse types of beverages. The importance of beverages in our society is so vast that no gathering can be imagined without the presence of at least one item to drink. The huge diversity of drinks across the globe, however, is more than often not supplemented with suitable scientific literature. Under the circumstance, this series will be a valuable addition to the society at large.

In this chapter, we discuss a very popular oriental, fermented beverage called Kombucha tea (KT). Our chapter has dealt with the ancient history of this beverage along with its microbiology, chemical composition, and beneficial aspects. Since one of the major components for making this beverage is tea itself, we have included a brief section on history, types, composition, and benefits of tea as well. Moreover, KT being a fermented product, we have included a concise section on the history of fermentation and tried to discuss some of the most popular fermented products across the globe.

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Non-alcoholic Beverages. https://doi.org/10.1016/B978-0-12-815270-6.00010-4

10.2 Tea, An Age-Old Concoction

Tea (*Camellia sinensis*) is the most drunk beverage in the world, after water. The beverage is produced from the leaves of the plant that are grown extensively in the tropical and subtropical regions of the world. It is consumed in six broad varieties depending upon the level of oxidation and fermentation employed (Chang, 2015). Of these six varieties, black tea and green tea are most popular across the world. While black tea is produced by nearly complete oxidation of the leaves, green tea is not oxidized at all (Sharma et al., 2007). There is a marked difference of consumption patterns across the globe which is evident from the fact that, while green tea is more popular in Japan and China, black tea has acquired more patrons in South East Asia and the West (Noguchi-Shinohara et al., 2014).

The 2016 worldwide market share of the beverage stands at a staggering US$38.84 billion and is expected to go up to US$47.20 billion by 2020 (Transparency Market Research, 2014–2020). Close to 35 countries have the geographical and climate conditions that allow tea plantations to thrive. Geographically, regions between 42′N (Georgia) and 35′S (Argentina) latitude are known to have favorable conditions for the plant to thrive. The four major countries that produce and export tea are China, India, Sri Lanka, and Kenya. The total amount of tea produced and consumed worldwide, in 2015 were 5305 million kg and around 4999 million kg, respectively. Although, India is the second largest producer of tea in the world, its market share in exports is only 13%. This is attributed to the fact that the country consumes majority of its produce. As far as export is concerned, Kenya (25%) is the market leader with China (18%) and Sri Lanka (17%) as close seconds (Tea Board of India, 2015–2016).

10.2.1 History of Tea

The hot simmering beverage that is known today as tea, have had a complex and long history that spread across multiple cultures and land boundaries, spanning thousands of years. The first written reference of tree can be traced back to around 2700 BCE in China (Chang, 2015). It is believed that the ability to use fire to cook was pivotal in prompting man to boil the abundant wild tea plants along with host of other tree barks and fruits with water and drink the stew. The concoction was widely consumed as a medicinal drink in the Yunnan Province of China during the Shang Dynasty (1766–1050 BCE). However, it was not until the Zhou Dynasty (1122–256 BCE), when people from the neighboring Sichuan province, started to boil tea alone, free from the other vegetative “contaminants” (Heiss and Heiss, 2011). Hence, the journey of tea started as a “romantic” stimulant beyond the mundane guise of a medicinal potion.

The entire history of tea, although fascinating, is too big and not the focus of this chapter. However, it is imperative to chronicle the major events that resulted in the spread of beverage across the globe. Tea, in its current form of consumption, is a result of extensive experimentation by the Chinese. The celebrated Tang Dynasty (618–907 CE) can be attributed for bringing in refinement and sophistication to how tea was prepared and drank across China. The active patronage of the royal family turned the bitter medicinal concoction into a healthful stimulant celebrated by the elites and intellectuals of the Chinese society. Thus, tea which before this period was a common drink of the masses now became a symbol of aristocracy and saw the advent of expensive and rare breeds of the plant. This classic upliftment of tea in the political strata of the country eventually led to the export of the beverage beyond the borders of China. The Tibetans first came in touch with tea in and around 641 CE through a matrimonial exchange between the royal families. It is during this period that strong trade relationships were established between the two countries. This trade continued well into the Yuan Dynasty (1271–1368) and tea formed an important commodity of trade. However, to ease trade, the export version of tea was mostly in the form of leftover twigs that were obtained from refining the more exquisite version of the plant. It is during this same period that the Japanese first started to encounter the beverage through the interactions between Zen priests and Buddhist monks. Stories indicate that the priest Saichō upon returning from China in 814 CE boiled tea leaves for the Emperor Saga. Although, tea did become an important beverage of the royal courts of Japan, its cultivation was not prevalent among common people. The beverage acquired popularity in Japan around 1191 when another Zen priest, Myoan Eisai brought back tea seeds from China and started sharing it with his friends and family. Hence, Eisai can be attributed to be pivotal in popularizing the crop in Japan during the Kamakura period (1185–1333) (Benn, 2015; Heiss and Heiss, 2011).

It was not until the Ming Dynasty (1368–1644) that the preparation of the beverage was done by steeping the tea leaves in hot water, as it is done today. Previously, the tea leaves were compressed into brick like cakes instead of leaves being dried and then roasted in iron woks, a technique started in the Ming era (Benn, 2015). However, this oxidation of tea to produce “black tea” along with the use of mare’s milk, was not Chinese customs. These modifications were the results of the Mongol influence on the Ming emperors. This oxidation of green tea to produce black tea had a considerable economic reward. Previously, the green tea cakes would often decompose before reaching the export destinations due to temperature fluctuations. The advent of black tea meant that the product could be exported without any significant loss (Heiss and Heiss, 2011).

The Ming Dynasty was followed by the Manchu Dynasty (1644–1911). This period saw massive expansion of trade of the west with the Chinese. It is during this period that the Europeans first started to import tea. However, the west was not completely oblivious to this drink before. The beverage was introduced in Europe by Portuguese priests returning from China (Weinberg and Bealer, 2004). Although, all the major European powers were “intoxicated” by the whiff and flavor of the tea, it was the British who took their liking of the concoction to a whole new level. By the early 1700s, the British East India Company established a monopoly of tea trade with China. However, till the 1800s, China was the only producer of tea that could satiate the western demand. To end the Chinese monopoly of tea production, the British introduced Chinese tea seeds in Assam, India. This impetus received major boost when in 1823, British Army Major Robert Bruce stumbled upon wild varieties of indigenous tea plants in Assam. The British were quick to assimilate the Chinese techniques of tea cultivation and soon the crop spread to the Himalayan foothill regions of Bengal. The growth of tea plantations in India ended the Chinese monopoly over tea and further consolidated the colonial power of Great Britain (Day, 2017). Ironically, the introduction of tea in India started a chain of events that would ultimately “rout” the British tea industry and reduce the former colonial champion to a minion in the global tea business of the modern world. The nail in the coffin was further hammered down when in 2000, Tata Tea (now Tata Global Beverages), acquired the British tea behemoth Tetley for £271 million (BBC, 2000). The tea planters of the erstwhile British Raj never imagined that a crop planted to exploit the hapless populace of an impoverished country, would one-day spell doom for their beloved “tea economy.”

10.2.2 Types of Tea

The leaves from the plant *C. sinensis* are the only raw material for production of tea. However, there are varied postharvesting processing and oxidation that gives rise to a diverse type of teas across the globe (Fig. 10.1).

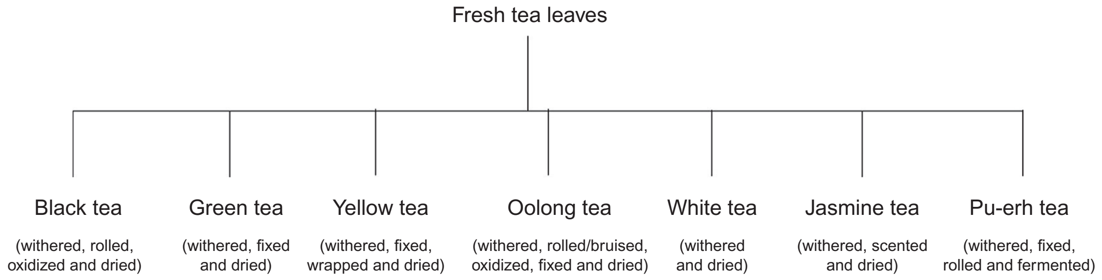

Fig. 10.1 Types of tea.

What is striking is that each variety has its unique taste and aroma. The change in seasonal conditions, climate, and rainfall along with the method of harvesting can have a profound effect on the flavor and the quality. The general gold standard of premium quality tea is ensured during plucking following a simple diktat in the form of, “two leaves and a bud.” The trees usually mature all year round and hence are harvested accordingly. This variation results in different names in various parts of the world. First harvesting of tea is referred to as First flush in India and Nepal, Pre-Qing Ming in China, Sincha in Japan, and Ujeon in South Korea. Each flushes of tea itself have variations on appearances, flavor, and aroma. Most of the countries have four harvests around the year. Tea can be classified into six main types: black tea, green tea, yellow tea, oolong tea, white tea, jasmine tea, and pu-erh tea.

10.2.2.1 Black Tea

This tea originated from Mongolia/China, and is currently cultivated in several countries worldwide. Best qualities of black tea now come from the countries like India and Sri Lanka. It has the highest consumption volume among its relatives (Nie et al., 2013). The leaves are fully oxidized after harvesting and thus are stronger than green tea, and tastes ranges from savory to sweet depending on the degree of oxidation and processing (Roberts et al., 2005).

10.2.2.2 Green Tea

The distinction of the second most consumed type of tea goes to this variety. Green tea is an evergreen plant growing in countries like China, India, Sri Lanka, and Japan. *C. sinensis* and *C. sinensis assamica* are the two primary varieties. Green tea production is mainly associated with inactivating the polyphenol oxidase enzyme in the fresh leaves by heating or applying steam which ensures catechins to remain unoxidized. The tea leaves are then rolled, dried, and finally packaged (Namal Senanayake, 2013). As this variety is not oxidized, the leaves retain their vital color. They have high concentration of epigallocatechin gallate (EGCG) and have been a focus of investigation as EGCG is potent against several human health disorders (Afzal et al., 2015).

10.2.2.3 Yellow Tea

This is a rare type tea that is produced in China. The preparation of this tea resembles that of green tea. The leaves get the characteristic yellow color after the drying phase, mainly due to the slow oxidation of catechin (Wang et al., 2013). Yellow tea is smooth and toasty (Heiss and Heiss, 2011).

10.2.2.4 Oolong Tea

This type of tea is prepared by a partial oxidation of the harvested leaves. Like other teas, this too originated from China. Today, Taiwan also contributes to its production. There is a broad range of oxidation spanning between 10% and 80% and this variation gives rise to a diverse spectrum of flavors from light, sweet, and fruity to rich, dark, and smoky flavor (Heiss and Heiss, 2011).

10.2.2.5 White Tea

Once cultivated only in parts of China, white tea is now grown in India and Sri Lanka as well. The tea leaves are subjected to minimum processing while preparing this type of tea. Chinese define white tea according to the subspecies it is manufactured from, that is, *C. sinensis* var. khenghe bai hao and *C. sinensis* var. fudin bai hao. The rest of the world, however, defines white tea by plucking standards. The process is a very delicate one in which only the bud or first leaves are plucked. Minimal processing retains the white leaf hairs leading to the name: white tea (Hilal and Engelhardt, 2007). White tea has a very light and delicate taste (Rusak et al., 2008).

10.2.2.6 Jasmine Tea

Jasmine tea is a scented tea prepared by amalgamation of base tea with *Jasminum sambac* flowers, which transmits its fragrance into the tea. Aroma is the unique selling proposition of this tea, and the cost here is directly proportional to the number of cycles repeated during the scenting processes. Tea scenting processes involve plucking and drying of base tea, using the flowers and layering of the base tea with flowers, then separating the flowers from the tea, and finally heating the tea to complete dryness before consumption (Chen et al., 2017).

10.2.2.7 Pu-erh Tea

This is a fermented, strong, and earthy tasting tea from Yunnan province in China. Processing of teas includes aging and fermentation of larger leaves in the presence of bacteria to generate a dark drink (Heiss and Heiss, 2011).

10.2.3 Chemical Composition

Tea is a reservoir of large variety of chemical constituents and has been under the scanner over the years (Harbowy et al., 1997). An exhaustive discussion on this topic can often be a bit dragging for the readers; therefore, we are attempting to convey all relevant information, albeit in a simple and concise manner. Tea constituents are broadly classified as follows.

10.2.3.1 Polyphenols, Flavonols, and Catechins

Polyphenols in tea are present in the form of flavonols, known as catechins among which (−)-epicatechin, (−)-epicatechin-3-gallate, (−)-epigallocatechin, (−)-epigallocatechin-3-gallate, and (−)-gallocatechin are the major catechins. Theaflavin and thearubigin present in black tea can also be classified under polyphenols. (−)-Epigallocatechin-3-gallate has been reported to have excellent antioxidant properties (Yousaf et al., 2014).

10.2.3.2 Vitamins and Minerals

Vitamin B1, vitamin B6, vitamin C, carotene, and folic acids are the predominant vitamins, whereas potassium, manganese, calcium, and fluoride are among the minerals that are found naturally in tea (Harbowy et al., 1997).

10.2.3.3 Amino Acids and Enzymes

Although tea contains several amino acids, theanine is the most abundant, and induces brain relaxation by promoting alpha brain wave activity (Nobre et al., 2008). Polyphenol oxidase on the other hand is a major enzyme found in tea extracts, and its deactivation is necessary during green tea production (Harbowy et al., 1997).

10.2.3.4 Methylxanthines and Pigments

Alkaloids mainly include caffeine, theobromine, and theophylline (Harbowy et al., 1997). Caffeine acts as a strong stimulant and enhances mental alertness when consumed (Fisone et al., 2004). Tea contains chlorophylls and carotenoids which are responsible for imparting color. Oxidation converts chlorophyll to black colored pheophytins, which is a characteristic color of the black tea (Harbowy et al., 1997).

10.2.3.5 Carbohydrates and Organic Acids

Majority of the carbohydrate fractions in tea are in the form of cellulose and 2-*O*-(β-ʟ-arabinopyranosyl)-myo-inositol. Tea is also a rich source of diverse types of organic acids such as malic, oxalic, succinic, citric, isocitric, shikimic, quinic acids, etc. (Harbowy et al., 1997).

10.2.3.6 Lipids and Volatile Components

Lipids, saponins, terpenoids, spinasterol, and triacontanol are also present in tea extracts along with other volatile molecules like methyl salicylate, benzaldehyde, phenyl ethanol, etc. The volatile molecules are mainly responsible for tea's flavor and aroma (Harbowy et al., 1997).

10.2.4 Functional Properties of Tea

The health benefits of both green tea and black tea are due to presence of the antioxidant and their ability to scavenge free radicals by its components like polyphenols and flavonoids (Hayat et al., 2015). Various evidences support that tea consumption inhibits lipid peroxidation and decreases oxidative stress, thereby reducing the risk of cardiovascular disease and other chronic diseases (Peluso and Serafini, 2017). Tea consumption decreases the risk of getting heart disease (Bingham et al., 1997) by its cholesterol lowering action and by decreasing systolic blood pressure and increasing the antioxidant levels of blood plasma. Tea has anticancerous effect as well. Studies revealed that intake of tea protected against pancreatic and prostate cancer development (Lynn-Cook et al., 1999). EGCG, the main component of green tea, along with other flavonoids has been found to be advantageous in treating brain, prostate, cervical, and bladder cancers (Yang and Wang, 2016). EGCG has also been shown to bind and hinder the antiapoptotic protein Bcl-2 (Leone et al., 2003), which has been implicated in both cancer cell and normal cell survival (Ge et al., 2011). EGCG also has putative antiinflammatory and antioxidant properties in multiple cell types and thus are beneficial for numerous inflammatory diseases like atherosclerosis, arthritis, etc. (Oz, 2017; Cavet et al., 2011). Green tea also contains an array of other polyphenolic compounds such as epicatechin-3-gallate (ECG), epigallocatechin (EGC), epicatechin, and catechins, which might have antiobesity effects. Rains et al. (2011) demonstrated the antiobesity effect of catechins present in green tea. These catechins were found to affect sympathetic nervous system (SNS) activity and increase energy expenditure, promoting the oxidation of fat. Caffeine, another tea component, also influences SNS activity and increases energy expenditure and fat oxidation by acting synergistically with catechins present in green tea. These compounds could also modify appetite; upregulate enzymes involved in hepatic fat oxidation and decrease nutrient absorption (Rains et al., 2011). The antioxidants present in tea also delay aging processes by scavenging the free radicals (Lee et al., 2014). Green and black tea extracts can also decrease the blood glucose level significantly by reducing the glucose absorption and uptake in diverse ways (Fu et al., 2017). Tea flavonoids additionally have been demonstrated to have antimicrobial activity against Gram-positive and Gram-negative bacteria, viruses, and fungi which can be harmful to humans (Friedman, 2007). Black and green tea extracts have been reported to be able to reduce the risk of *Helicobacter pylori* infection associated with gastric, peptic, and duodenal ulcer diseases (Boyanova et al., 2015). Moreover, daily consumption of tea is also beneficial for other health problems. It can prevent kidney stone formation and constipation, facilitate digestion by helping the stomach muscles and body's digestive juice to work better, and maintain kidney function normal via its diuretic action. In addition, tea also replaces lost body fluids and provides nourishment after exercise (Sharma et al., 2014).

10.3 History of Fermentation

10.3.1 Fermentation an Ancient Story

A standard English lexicon defines fermentation as, "to experience a chemical change because of the action of yeast or bacteria, often changing sugar to alcohol; to make something change in this way" (Oxford University Press, 2015). Biologically speaking fermentation does not come down to production of alcohol alone. It means a slow conversion of an organic substrate to a product by the action of microbes and their enzymes (Swain et al., 2014). Natural fermentation precedes human history. The advent of human harnessing of fermentation is lost in the mist of antiquity. Archeological findings point out to the fact that the advent of ethanol fermentation prompted primitive people to settle down and start cultivating (Battcock and Ali, 1998). The oldest reference of an alcoholic beverage, made from fruit, rice, and honey, dates to 7000–6600 BCE, in the Neolithic Chinese village of Jiahu, and winemaking dates to around 6000 BCE, in Georgia, in the Caucasus area (McGovern, 2003; McGovern et al., 2004). Fermented products like bread, cheese, and wine have been prepared and consumed for thousands of years and almost form important backbones of human culture in various parts of the world (Battcock and Ali, 1998). Fermentation has helped our ancestors of the temperate and arctic weathers to survive the harsh winters. Similarly, the process has helped the prehistoric humans to brave the drought periods of the tropics (Swain et al., 2014). Fermentation is also one of the oldest and widely used food preservation methods across the globe in households, small-scale food industries as well as in large enterprises (Law et al., 2011). Microbial fermentations have also played a huge role in many aspects of human civilization and as a result every human society at every level of its growth and complexity has discovered means of utilizing localized sugar sources for preparation of various fermented beverages and foods. Moreover, fermentation in addition to preservation are also often known to result in pleasant flavor, aroma, texture, enhanced nutritive values, and good keeping quality under ambient conditions (Law et al., 2011). The first fermented food probably was prepared from fruits. Prehistoric hunter-gatherers were naturally used to consuming fresh fruits but at times of scarcity, they might have been forced to eat rotten and fermented fruits. Repeated consumption might have led to the development of the taste for fermented fruits. China is believed to be birthplace of fermented vegetables and the use of *Aspergillus* sp. and *Rhizopus* sp. molds for food preparation (Battcock and Ali, 1998).

Fermented food and beverages are considerably unique and vary considerably according to the social patterns, climate, and consumption practices (Nout and Motarjemi, 1997). The local substrates and raw materials available in a region play a pivotal role on the fermentation process that has evolved with a civilization (Law et al., 2011). The Asia-Pacific region having a tropical climate is a haven for cereal production and thus many of its traditional fermented products originate from cereals (Swain et al., 2014). Similarly using grapes for fermentation mainly originated in Persia and Georgia and later spread west and south and became intertwined with the history of Western civilization (McGovern, 2003). Malt-based fermentations also originated in Iraq and Egypt around 3500–3100 BCE and were later propagated into Europe by Germanic and Celtic tribes (Encyclopedia Britannica, 2017).

10.3.2 Diversity of Fermented Food

Every nation and continent has their own set of fermented food and beverages. Many of these fermented products were results of accidental discoveries and hence the methodology for their production is still unknown. Although the most popular beverages prepared from fruits and cereals are wine and spirits, prepared by yeasts, many beverages and food are prepared by bacterial fermentations viz. by the lactic acid bacteria (LAB) and acetic acid bacteria (AAB). Table 10.1 contains examples of some of the major fermented products using fruits, vegetables, and cereals from across the globe.

The fermented products described in Table 10.1 are mostly regional food and beverages which are almost unknown outside the region where they are popular. Wines, however, are known and consumed worldwide and rank among the most popular fermented products (WHO, 2014). Of the traditional nonalcoholic fermented products, KT is an extremely popular beverage (Mo et al., 2008). The beverage has acquired significant interest all over the world because of its claimed beneficial effects which range from weight loss to having therapeutic potential as an anticancer drug (Dufresne and Farnworth, 2000; Greenwalt et al., 2000). However, many such claims are scientifically unsubstantiated and significant research is required into the matter. We will discuss in detail, the beneficial properties of KT later in the chapter.

Table 10.1 Some Fermented Products of the World (Battcock and Ali, 1998; Haard et al., 1999; Mo et al., 2008; Law et al., 2011; Swain et al., 2014)

Fermented Food Product	Country/Continent	Main Ingredient
Acar, Achar, Tandalachar, Garamnimbooachar, Lemon pickle, Lime pickle, and Mango pickle	Indian subcontinent	Pickled fruit and vegetables
Goyang, Gundruk, Inziangsang, Khalpi, Sinki, and Soidon	Do	Fermented vegetable
Bangla mod, Cholai, Tari, Ara, Arrack, DesiSharab, Tharra, Toddy, Fenny, Raksi, Tadi, and Chayang Tomb	Do	Fermented cereal
Asinan, Burongmangga, Ca muoi, Dakguadong, Dalok, Dhamuoi, Duamuoi, Jeruk, Kiam-chai, Kiam-cheyi, Kong-chai, Naw-mai-dong, Pak-Gard-Dong, Pak-siam-dong, Paw-tsay, Phak-dong, Phonlami-dong, Sajurasin, Sambal tempo-jak, Santol, Si-sek-chai, Sunki, Tang-chai, Tempoyak, and Vanilla	South east Asia/Pacific rim	Fermented fruit and vegetables
Palm wine, Oou, Namtanmao, SarthaWaark, Sato, Krachae, Tuak, Tapai Sabah, Arrack, Samsu, Tapuy, and Ruou nep than	Do	Fermented cereal
Bai-ming, Leppet-so, Miang, Puer tea, and Fuzhuan brick-tea	Do	Fermented tea leaves
Nata de coco, Nata de pina	Do	Fermented fruit juice
Bossam-kimchi, Chonggak-kimchi, Dan moogi, Dongchimi, Kachdookigactuki, Kakduggi, Kimchi, Mootsanji, Muchung-kimchi, Oigee, Oiji, Oisobaegi, Tongbaechu-kimchi, Tongkimchi, and TotkalKimchi	East/Far East Asia	Fermented in brine
Cha-ts'ai, Hiroshimana, Jangagee, Jiang-gua, Nara senkei, Narazuke, Nozawana, Nukamiso-zuke, Omizuke, Paocai, Pobuzihi, Powtsai, Red in snow, Seokbakji, Shiozuke, Suan-tsai, Sunki, Szechwan cabbage, Tai-tan tsoi, Takana, Takuan, TsaTzai, Tsu, Umeboshi, Wasabi-zuke, Yan-dong-gua, Yan-jiang, Yan-taozih, and Yen-tsai-shin	Do	Fermented fruits and vegetable
Kushuk, Lamounmakbouss, Mekhalel, Olives, Torshi, and Tursu	Middle East	Do
Kombucha	International	Fermented tea
Oilseeds, Ogili, Ogiri, Hibiscus seed, Lamounmakbouss, Mauoloh, Msir, Mslalla, Olive, and Wine	Africans	Fermented fruits, vegetables, and seeds

Table 10.1 Some Fermented Products of the World (FAO, 1998; FAO, 1999; Mo et al., 2008; Law et al., 2011; Swain et al., 2014)—cont'd

Fermented Food Product	Country/Continent	Main Ingredient
Ogi, BogobeKoko, Kenkey, Mawe, Mahewu (magou), Uji, Kisra, Enjara, Burukutu, PitoKaffir beer, Busaa (maize beer), Malawa beer, Zambian opaquemaize beer, Merissa, Seketeh, Bouza, Talla, and Kishk	Do	Fermented cereal
Cucumber pickles, Dill pickles, Olives, Sauerkraut, Vanilla, and Wines	Americas	Fermented fruits and vegetables
Abati, Acupe, Agua-agria, Cachiri, Champuz, Chica, Charagua, Fubá, Jamin-bang, Napú, Ostoche, Pozol, Quebrantahuesos, Sendechó, Sora, Tepache, Tesgüino, Tocos, Zarzaparrilla bark wine, and Zambumbia	Latin America	Fermented cereal
Olives, Sauerkohl, and Sauerruben	Europe	Pickled fruits and vegetables
Grape vinegar, Wine vinegar	Do	Vinegar
Wines, Citron	Do	Fermented fruits

10.4 Tea and Kombucha

KT (Fig. 10.2) is a marginally sweet, slightly acidic fermented non-alcoholic beverage consumed worldwide and has gained significant popularity among the increasingly health conscious world populace (Chakravorty et al., 2016). The beverage can be prepared using both black and green tea. However, KT prepared from black tea is the most popular practice and considered as an ingredient of choice for the beverage. Similarly, white sugar or sucrose is the preferred choice of carbon source (Jayabalan et al., 2014). The fermentation is product of microbial activity by a consortium of both yeast and bacteria (Chakravorty et al., 2015). The yeast present in the system first converts the available carbon source to ethanol, which is subsequently converted to acids by the bacteria (Jayabalan et al., 2014). Thus, the Kombucha biofilm, formed on a liquid-air interface due to the activity of both bacteria and yeast, has generated significant scientific interest down the ages (Chakravorty et al., 2016).

The origin of this beverage is somewhat lost in the mist of time. It is believed that KT originated in northeast China (Manchuria) during the Tsin Dynasty (Ling Chi) in about 220 BCE and it had

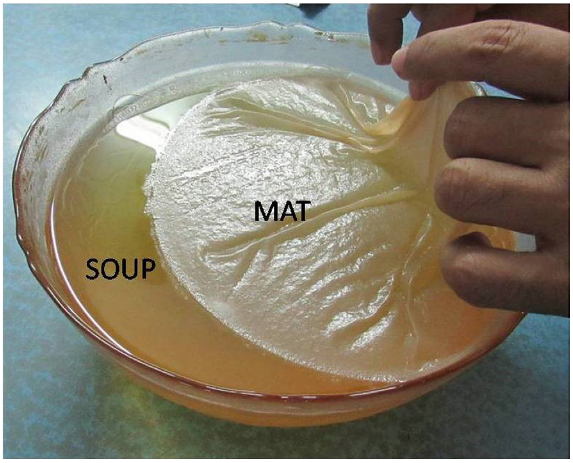

Fig. 10.2 Kombucha tea (soup and mat).

significant popularity for its detoxifying and energizing properties. It is believed that the beverage had reached Japan during 414 CE when the physician Kombu used the beverage to cure the digestive problems of the Emperor Inkyo. It is believed that the name Kombucha originated from the name of the physician as the current name Kombucha is a Japanese term for slightly fermented tea. As the world started expanding its trading routes with the orient, KT, which has acquired the trade name Mo-Gu, quickly found its way among the Russian trading class. The Russians called this beverage as Cainiigrib, Cainii kvass, Japonskigrib, Kambucha, and Jsakvasska. With the dawn of the 20th century, the beverage gained popularity among the Eastern European nations and finally into Germany. The beverage in Germany gained another set of metaphors viz. Heldenpilz and Kombuchaschwamm. The culture of KT however was lost in Europe due to dire shortage of tea leaves and sugar and was reintroduced in the continent after the World War II. In the 1950s, the beverage reached France and also gained popularity in the French colonies of North Africa. When KT reached Italy postwar with yet another metaphor Funkochinese, it quickly became hugely popular as a mysterious oriental health drink. The Swiss researchers in 1960 reported that drinking KT had similar beneficial effects as eating yogurt and its popularity soared. KT today is sold through retail shops in various flavors and the biofilm is sold to enthusiasts through various online vendors (Jayabalan et al., 2014).

KT is prepared by adding a small portion of the biofilm into sweetened black tea. The broth also contains 10%–15% of previously fermented beverage (called as old soup for convenience). The old soup provides the pH balance in the unfermented broth and keeps away undesired microorganisms from spoiling it. The fermentation is usually done in a glass vessel covered with clean cloth and incubated at room temperature (28–30°C) for 7–12 days. No separate incubator is required unless the room temperature is too high or low or damp (Chakravorty et al., 2016). The amount of tea and sugar added in the fermentation differs in different KT (Marsh et al., 2014).

10.5 Microbiology of Kombucha Fermentation

The microbial population of KT is an active field of research. Some inquisitions into the population have been undertaken. The bacterial population is believed to be dominated by the members of AAB (Chakravorty et al., 2015, 2016; Jayabalan et al., 2014; Marsh et al., 2014). However, the yeast population is comparatively more diverse among different KT systems (Marsh et al., 2014; Chakravorty et al., 2016; Coton et al., 2017). Although, most of the publications about KT have exclusively described laboratory-scale fermentations, a recent study by Coton et al. (2017), has dealt with the industrial-scale fermentation of the beverage.

The microbial community of KT is divided over two mutually non-exclusive compartments: the soup or the beverage and the biofilm floating on it. Majority of the studies of the community in the past have been done by traditional culture-dependent methods (Chen and Liu, 2000; Dutta and Gachhui, 2006, 2007; El-Salam, 2012; Hesseltine, 1965; Jankovic and Stojanovic, 1994; Liu et al., 1996; Mayser et al., 1995; Sievers et al., 1995; Teoh et al., 2004). With the advent of cheap modern next-generation sequencing platforms, increasing number of exhaustive community analysis are being published (Coton et al., 2017; Chakravorty et al., 2016; Marsh et al., 2014). Both culture-dependent and independent techniques are powerful tools of microbial community analysis. However, both techniques have fair share of pros and cons. While a culture-dependent method enables the investigator to obtain pure cultures of the identified microorganism, a major drawback of the method is the fact that in most biomes, <1% of the microbes are culturable. Furthermore, the properties of the microbes are often significantly different from those in the actual environment. Culture-independent methods on the other hand, provide a far more comprehensive and penetrative view of the microbiome. But they are again plagued by the fact that identifications are often done based on small fragments of genetic data. As a result, researchers may end up with over stretching the actual diversity of a microbial community (Chakravorty et al., 2016). Thus, increasing number of researchers is complementing both techniques to obtain a comprehensive picture of the various microbiomes and KT is no different.

10.5.1 Bacterial Community

KT fermentation is similar to the vinegar fermentation as the major bacteria found in both the biofilm and the soup belongs to the group of AAB (Coton et al., 2017). Culture-dependent methods have identified the AAB and LAB as the only bacteria present in the system, the major two genera being *Acetobacter* and *Gluconobacter* (Marsh et al., 2014). *Acetobacter xylinum*, now renamed as *Komagataeibacter xylinus* was the most isolated bacteria from different KT variants (Danielova, 1954; Konovalov and Semenova, 1955; Sievers et al., 1995; Roussin, 1996). Two novel bacteria, *Acetobacter nitrogenifigens* RG1T and *Gluconacetobacter kombuchae* RG3T were also isolated from KT (Dutta and Gachhui, 2006, 2007). As discussed previously, this limited bacterial diversity remained a benchmark of publications well into the first decade of the 21st century. Our lab at that period was also exploring the KT biofilm following traditional culture-dependent methods. Our study resulted in the isolation of 152 cultures spread over 15 phylogenetic groups as determined by a limited biochemical screening based on carbon source utilization (Table 10.2). Molecular analysis through 16S rRNA gene sequence revealed the presence of three species of the genus *Enterobacter* (Table 10.3). This was an important observation as it indicated the presence of bacteria other than those capable of withstanding very low pH. It was around the same time that Marsh et al. (2014) published the first next-generation sequencing-based report on KT population. This report, for the first time showed an in-depth diversity of the microbial community structure of KT. The major bacteria identified by this study are being described in Table 10.4. In this study, *Gluconacetobacter* was identified to be the most prevalent bacteria among five several types of KT studied. Chakravorty et al. (2015), used traditional culture-independent methods to explore the bacterial community of the biofilm and reported that *Komagataeibacter* sp. and *Acetobacter* sp. as the major bacterial genera of the microbiome. Although, Marsh et al. (2014) did provide an extensive picture of the KT system, it lacked different time points of the fermentation. Chakravorty et al. (2016) described for the first time the microbial community structure of both the Kombucha biofilm and the soup at four different time points of the fermentation, 3, 7, 14, and 21 days. The bacterial community was first studied by terminal restriction fragment length polymorphism (T-RFLP) for all 4 days. It was observed that the bacterial community showed maximum diversity on the seventh day of fermentation. Next-generation sequencing of the seventh day sample revealed that *Komagateibacter* was the most

Table 10.2 Biochemical Characterization of the Culture-Dependent Samples

		Biochemical Tests									
Group	Members (CD-Number)	GYC	EYC	30% Glucose	Sucrose	Mannitol	Sorbitol	Ethanol	Fructose	Maltose	Accession Numbers
1	1, 2, 4, 5, 6, 8, 9, 10, 11, 12, 13, 14, 16, 17, 18, 19, 20, 21, 22, 23, 28, 29, 30, 31, 32, 33, 35, 37, 38, 39, 40, 41, 42, 45, 46, 49, 51, 52, 53, 55, 56, 58, 59, 60, 61, 63, 64, 65, 66, 67, 68, 69, 73, 74, 76, 77, 78, 80, 81, 82, 83, 84, 85, 86, 87, 88, 90, 92, 95, 97, 98, 99, 100, 101, 103, 107, 109, 110, 114, 115, 116, 117, 118, 119, 121, 124, 126, 127, 131, 132, 133, 135, 136, 137, 138, 143, 146, 147, 149, 150, 151, 154, 155, 156	+	+	++	++	+++	+	+++	+++	+++	KC763592 (CD1), KC763593 (CD2), KC763602 (CD29), KC763604 (CD35), KC763605 (CD41), KC763607 (CD45), KC763610 (CD63), KC763611 (CD64), KC763612 (CD65), KC763618 (CD86), KC763619 (CD95), KC763622 (CD117), KC763623 (CD121), KC763627 (CD147)
2	3, 122	−	+	++	++	+++	+++	+++	+++	+++	KC763594 (CD3), KC763624 (CD122)
3	6, 76	−	+	++	++	+	+	+++	+++	+++	KC763595 (CD6), KC763617 (CD76)

4	15, 25, 50, 57, 62, 79, 91, 94, 96, 104, 105, 108, 111, 113, 120, 123, 125, 145	−	+	−	++	+	+	+++	+++	+++	KC763596 (CD15), KC763598 (CD25), KC763599 (CD57)
5	24, 43, 128, 134	+	+	−	++	+++	+++	+++	+++	+++	KC763597 (CD24), KC763606 (CD43), KC763625 (CD128)
6	26	−	+	+/−	++	++	+	+	+++	+++	KC763600 (CD26)
7	27, 71, 75, 89	−	+	−	++	+	−	+++	+++	+++	KC763601 (CD27), KC763614 (CD71), KC763616 (CD75)
8	34, 36, 44	+	+	+/−	++	+++	++	+++	+++	+++	KC763603 (CD34)
9	47, 54	+	+	++	++	+	−	+++	+++	+++	KC763608 (CD47)
10	48	+	+	+/−	++	+++	−	+	+++	+++	KC763609 (CD48)
11	70	−	+	++	++	++	+++	+++	+++	+++	KC763613 (CD70)
12	72	+	+	+	++	+++	−	+++	+++	+++	KC763615 (CD72)
13	102	+	+	−	++	+	+	+++	−	−	KC763620 (CD102)
14	93, 106, 112, 129, 130, 141, 142, 152	+	+	−	++	+++	+	+++	+++	+++	KC763621 (CD106), KC763626 (CD141)
15	153	+	+	+++	++	++	+++	+++	−	+++	KC763628 (CD153)

For GYC (*G*lucose, *Y*east extract and *C*alcium carbonate) and EYC (*E*thanol, *Y*east extract, and *C*alcium carbonate) +,− represents positive or negative results, respectively. For the rest: +, slight growth; ++, good growth; +++, luxuriant growth; −, no growth; +/−, weak growth.

Table 10.3 Species Variation Among the Pure Cultures as Determined by NCBI BLAST and EzTaxon Server

Culture Names	Number of Cultures	Corresponding Organism (Accession Number)
CD1, CD2, CD6, CD25, CD27, CD29, CD34, CD35, CD41, CD63, CD64, CD65, CD71, CD72, CD76, CD86, CD95, CD102, CD106, CD121, CD147, CD153	22	*Acetobacter nitrogenifigens* strain NBRC 105050 (AB682235)
CD15, CD45, CD47, CD57, CD75, CD117, CD141	7	*Acetobacter nitrogenifigens* strain RG1^T(AY669513)
CD3	1	*Enterobacter cancerogenus* LMG 2693 (Z96078)
CD24, CD26 CD43, CD122	4	*Enterobacter ludwigii* strain DSM16888^T (AJ853891)
CD48, CD70, CD128	3	*Enterobacter cloacae* subsp. *cloacae*^{T} (CP001918)

abundant genus found in both the biofilm and the soup (Table 10.5). A very important observation from this report was that the bacterial community of the soup was significantly more diverse than that of the biofilm. The explorations described till now were from laboratory-scale preparations of KT. There has been a dearth of knowledge about the fermentation in an industrial setting. This problem has been addressed by Coton et al. (2017), where they have described the microbial community structure of both green tea and black tea KT from industrial samples. *Gluconacetobacter europaeus*, *Gluconobacter oxydans*, *Gluconacetobacter saccharivorans*, and *Acetobacter peroxydans* were the major AAB genera identified in this study. The main genus of the LAB, *Oenococcus oeni* was found to be associated largely with the green tea fermentations.

10.5.2 Yeast

The yeast community of KT, like its bacterial counterpart, has mostly been studied by culture-dependent methods. A broad spectrum of yeast has been previously reported through such methods. The major genera being: *Brettanomyces/Dekkera*, *Candida*, *Koleckera*,

Table 10.4 List of Bacterial and Yeast Genera Identified by Marsh et al. (2014)

Bacteria			Yeast		
Soup			Soup		
Day 3	**Day 10**	**Biofilm**	**Day 3**	**Day 10**	**Biofilm**
Gluconacetobacter	*Gluconacetobacter*	*Gluconacetobacter*	*Zygosaccharomyces*	*Zygosaccharomyces*	*Zygosaccharomyces*
Acetobacter	*Acetobacter*	*Acetobacter*	*Dekkera*	*Dekkera*	*Dekkera*
Lactobacillus	*Lactobacillus*	*Lactobacillus*	*Kazachstania*	*Kazachstania*	*Davidiella*
Lactococcus	*Lactococcus*	*Lactococcus*			*Pichia*
Leuconostoc	*Leuconostoc*	*Leuconostoc*			*Wallemia*
Bifidobacterium	*Bifidobacterium*	*Bifidobacterium*			*Lachancea*
Thermus	*Thermus*	*Thermus*			*Leucosporidiella*
Allobaculum	*Allobaculum*	*Allobaculum*			*Kazachstania*
Ruminococcaceae	*Ruminococcaceae*	*Ruminococcaceae*			*Kluyveromyces*
Incertae Sedis	*Incertae Sedis*	*Incertae Sedis*			*Naumovozyma*
Propionibacterium		*Enterococcus*			
		Propionibacterium			

Table 10.5 Relative Distribution of Taxa in the Biofilm and Soup (% Abundance)

	Sample			Sample		Genus	Sample	
Phylum	**Biofilm**	**Soup**	**Family**	**Biofilm**	**Soup**		**Biofilm**	**Soup**
Proteobacteria	88.5	65.7	Acetobacteraceae	88.5	63.5	*Komagataeibacter*	50.3	49.9
Firmicutes	–	11.2	Oscillatoriaceae	–	5.5	*Gluconobacter*	16.8	2.5
Cyanobacteria	–	7.1	Bifidobacteriaceae	–	2.3	*Lyngbya*	–	4.4
Actinobacteria	–	4.1	Ruminococcaceae	–	2.1	*Bifidobacterium*	–	2.3
Unclassified	11.4	11.4	Peptostreptococcaceae	–	1.7	*Collinsella*	–	0.7
Others	0.1	0.5	Coriobacteriaceae	–	1.3	*Enterobacter*	–	0.6
X	X	X	Lachnospiraceae	–	1.2	*Weissella*	–	0.5
X	X	X	Enterobacteriaceae	–	1.1	*Lactobacillus*	–	0.3
X	X	X	Leuconostocaceae	–	0.5	Unclassified	32.8	34.6
X	X	X	Erysipelotrichaceae	–	0.4	Others	0.1	4.2
X	X	X	Rhodobacteraceae	–	0.4	X	X	X
X	X	X	Lactobacillaceae	–	0.3	X	X	X
X	X	X	Unclassified	11.4	17.5	X	X	X
X	X	X	Others	0.1	2.2	X	X	X

Note: "–" signifies below 0.1% or absent. "X" signifies not applicable.
From Chakravorty, S., Bhattacharya, S., Chatzinotas, A., Chakraborty, W., Bhattacharya, D., Gachhui, R., 2016. Kombucha tea fermentation: microbial and biochemical dynamics. Int. J. Food Microbiol. 220, 63–72 (Elsevier article).

Mycoderma, *Mycotorula*, *Pichia*, *Saccharomyces*, *Saccharomycodes*, *Schizosaccharomyces*, *Torulospora*, and *Zygosaccharomyces* (Jayabalan et al., 2014). Similar to the bacterial population, the first extensive structure of the yeast community was provided by Marsh et al. (2014). In the study, the dominating yeast genus in three diverse types of Kombucha was identified to be *Zygosaccharomyces* (Table 10.4). Chakravorty et al. (2016) showed the community structure over four different time points of the fermentation. The paper reported *Candida* sp. as the dominant yeast genus in the biofilm and the soup on the 3rd, 14th, and 21st days of fermentation (Table 10.6). Interestingly, on the seventh day of fermentation, *Lachancea* sp. was the dominant yeast in the soup. The difference in yeast community structure was accompanied by increase in bioactivity of the beverage between the 7th and 14th day of fermentation. Summing up the diversity of bacterial and yeast communities after 7 days of fermentation, we may infer that the variation of the microbial community had direct manifestation on the bioactivity of the beverage (Chakravorty et al., 2016). This observation will have important impacts on future studies of the beverage. Yeast community of industrial-scale KT made using green tea and black tea, did not seem to differ based on type of tea used (Coton et al., 2017). *Dekkera bruxellensis*, *Dekkera anomala*, *Zygosaccharomyces bailii*, and *Hanseniaspora valbyensis* were the dominant yeast identified in the report.

The microbial community studies performed till date are considerably exhaustive and valuable information regarding the impact of these microbes on the bioactivity has been obtained (Chakravorty et al., 2016; Coton et al., 2017). An important roadblock in getting a consensus on the community structure, however, comes from the fact that most of the fermentations are done without following a specific methodology and no two microbial inoculums are the same (Malbaša et al., 2011). Thus, significant research in the diverse microbial communities of KT available across the globe, still needs to be done before we start to devise custom made inoculum that will provide optimum bioactivity of the beverage.

10.6 Chemical Composition of KT

Having discussed on the microbial community of the beverage, it is imperative that we shift our focus toward its chemical composition. It has been reported that, during the fermentation process of KT, tea ingredients and sucrose were modified by the microbes' present in the system. Sucrose added to the fermentation as carbon source is first hydrolyzed to glucose and fructose by the action of the invertase enzyme produced by the yeast. While the yeast converts the available fructose to ethanol, the AAB use the glucose and ethanol to produce gluconic acid and acetic acid, respectively (Jayabalan et al., 2007).

Table 10.6 Species Distribution of Yeast in Kombucha Tea

	Sample							
Species	**Biofilm-Day-3**	**Biofilm-Day-7**	**Biofilm-Day-14**	**Biofilm-Day-21**	**Soup-Day-3**	**Soup-Day-7**	**Soup-Day-14**	**Soup-Day-21**
Candida stellimalicola	59	72.2	71.3	65.3	61.5	17	68.5	56.5
Candida tropicalis	11.9	6.8	8.4	11.8	8.4	6	11.4	7.2
Candida parapsilosis	2	4	2.6	2.5	2.9	3.3	6.5	6.5
Lachancea thermotolerans	7.2	–	–	2.5	1.3	4.8	0.2	1
Lachancea fermentati	2.3	2.5	1.9	4.2	15.1	51.1	3.6	20
Lachancea kluyveri	–	0.3		0.1	–	1.5	–	–
Eremothecium cymbalariae	2.4	0.8	2.9	0.9	0.1	–	0.2	0.6
Eremothecium ashbyii	–	–	–	–	–	0.1	–	–
Kluyveromyces marxianus	2.3	2.6	2.5	2.5	2.1	4.8	2	0.9
Debaryomyces hansenii	0.6	2	0.9	1.5	1.5	0.6	2.2	0.6
Pichia mexicana	0.7	0.3	0.7	0.9	0.9	0.6	0.7	0.7
Meyerozyma caribbica	1.5	0.7	0.3	0.7	0.8	0.9	0.8	0.5
Meyerozyma guilliermondii	0.3	–	–	–	–	–	–	–
Zygowilliopsis californica	1	0.3	0.4	0.2	0.4	0.9	0.3	–
Saccharomyces cerevisiae	0.8	0.4	0.2	–	0.2	0.1	–	–
Saccharomycopsis fibuligera	0.7	0.6	0.4	0.6	0.6	0.7	0.6	0.7
Hanseniaspora uvarum	0.5	0.7	2.5	0.3	0.8	0.4	0.2	0.5
Hanseniaspora meyeri	–	–	–	0.1	–	–	0.8	–
Hanseniaspora vineae	0.1	0.1	–	0.3	0.2	1.2	–	0.5
Merimblaingelheimense	–	1.2	1	–	–	–	–	–
Sporopachydermialactativora	–	0.8	1	0.3	–	0.2	0.8	0.6
Kazachstania telluris	0.5	–	0.6	0.4	–	1.7	–	0.2
Kazachstania exigua			–	–	–	0.3	–	–
Starmera amethionina	0.3	1.4	–	2.6	1	1.8	0.4	–
Starmera caribaea		0.7	–	–	–	–	–	–
Unclassified	3.5	0.3	1.4	0.8	1.0	1.5	0.2	1.8
Others	2.4	1.3	1	1.5	1.2	0.5	0.6	1.2

Adapted from Chakravorty, S., Bhattacharya, S., Chatzinotas, A., Chakraborty, W., Bhattacharya, D., Gachhui, R., 2016. Kombucha tea fermentation: microbial and biochemical dynamics. Int. J. Food Microbiol. 220, 63–72 (Elsevier article).

Gluconoacetobacter xylinum has been reported to synthesize the floating cellulose network which enhances the association formed between bacteria and fungi. Caffeine and related xanthines of the tea infusion stimulate the cellulose synthesis by the bacteria. Thus, the principle ingredients of KT are the organic acids, glucose, fructose, ethanol, different vitamins, minerals, amino acids, etc. (Dufresne and Farnworth, 2000). However, the exact chemical composition of KT is not yet known completely and is still under investigation.

The composition and the concentration of different components of KT vary depending on the source of the tea fungus, type of sugar and its concentration, fermentation duration, and tea content. When sucrose is used as the carbon source, acetic acid is produced as the main metabolite. The other major organic acids that are produced during Kombucha fermentation are gluconic and glucuronic acids. The concentrations of the acids increase with fermentation time, resulting in the lowering of the pH of the fermented tea to as low as 2 (Chen and Liu, 2000; Jayabalan et al., 2007; Chakravorty et al., 2016). In some studies, the presence of lactic acid and citric acid was also reported although these are not the characteristic metabolic products of traditional Kombucha beverage (Jayabalan et al., 2007; Malbaša et al., 2008a,b, 2011). KT is also found to contain sugars like glucose, fructose, and sucrose indicating that they were not utilized entirely even after 30 days of fermentation (Chen and Liu, 2000). In addition, inulo-oligosaccharides were also found to be present in Kombucha (Malbaša et al., 2002).

The tea polyphenols also contribute a major part in the composition of KT. They undergo progressive modifications during Kombucha fermentation. Recent studies have shown that the black tea polyphenol, theaflavin was found to be increased by about 88.63% whereas thearubigin was decreased by about 47.02% after 21 days of fermentation (Chakravorty et al., 2016; Kallel et al., 2012). It is therefore possible that a part of thearubigin was converted to theaflavin during the fermentation. This decrease in thearubigin might be responsible for the change in color of KT from reddish brown to light brown with fermentation time. However, Jayabalan et al. (2007) observed no significant change in theaflavin and thearubigin concentration during Kombucha fermentation. They reported that catechins in tea were degraded during KT fermentation and the concentration of epigallocatechin (EGC) and epicatechin (EC) exceeded the initial concentration on the 12th day of Kombucha fermentation while it was not observed for epigallocatechin-3-gallate (EGCG) and epicatechin-3-gallate (ECG). Therefore, it might be that EGCG was converted to EGC and ECG was converted to EC. There is also a reduction of caffeine in black tea by about 32.7% during the first 3 weeks of Kombucha fermentation (Chakravorty et al., 2016). The microorganisms of Kombucha culture, therefore, might use caffeine as a source of nitrogen, thereby degrading this purine alkaloid.

KT is found to contain water-soluble vitamins like vitamin B1, vitamin B6, vitamin B12, and vitamin C. The presence of manganese, iron, nickel, copper, zinc, and others was also detected in trace amounts in this fermented beverage (Bauer-Petrovska and Petrushevska-Tozi, 2000). However, the essential minerals were found to be increased because of the metabolic activity of KT. Kumar et al. (2008) showed that ions like fluoride, chloride, bromide, iodide, nitrate, phosphate, and sulfate were present in the beverage after 7 days of fermentation.

Fermentation of sugared black tea by Kombucha culture results in the production of certain metabolites which were not present in black tea. One such molecule is D-saccharic acid-1,4-lactone which is reported in KT by many researchers but with varying concentration (Chakravorty et al., 2016; Wang et al., 2010; Yang et al., 2009, 2010). Chakravorty et al. (2016) also showed that DSL (D-saccharic acid 1,4-lactone) concentration increased gradually with fermentation time. *Gluconacetobacter* sp. A4 which is assumed to be the key functional bacterial species has a strong ability to produce DSL (Yang et al., 2010). Bhattacharya et al. (2016) detected isorhamnetin as one of the antibacterial constituents present in the KT polyphenolic fraction. Isorhamnetin is an *O*-methylated flavonol and one of the constituents of Gingko biloba (Qiu et al., 2017), cocoa (Sánchez-Rabaneda et al., 2003), and *Hippophae rhamnoides* (Xie et al., 2015) which exerts potent antimicrobial activity (Nenaah, 2013). Since isorhamnetin is not reported to be present in tea, it can be suggested that the fermentation of black tea by the microbial population of KT may play some role in the production of isorhamnetin in KT.

These are some of the components of KT reported till date. Many other ingredients might be present in KT which has not been elucidated yet.

10.7 Beneficial Aspects of Kombucha

10.7.1 Therapeutic Applications

Kombucha beverage is known to possess many prophylactic and therapeutic benefits which have been reported based on personal observation and testimonials (Greenwalt et al., 2000; Vīna et al., 2014). Although, some of them have been demonstrated by scientific and experimental studies (as depicted in Fig. 10.3), scientific evidences based on human models are still lacking. Such an endeavor can only be undertaken when we have a clear understanding of majority of the beneficial properties of the beverage. The current available knowledge about the biological activities of KT has been discussed in the subsequent sections of this chapter.

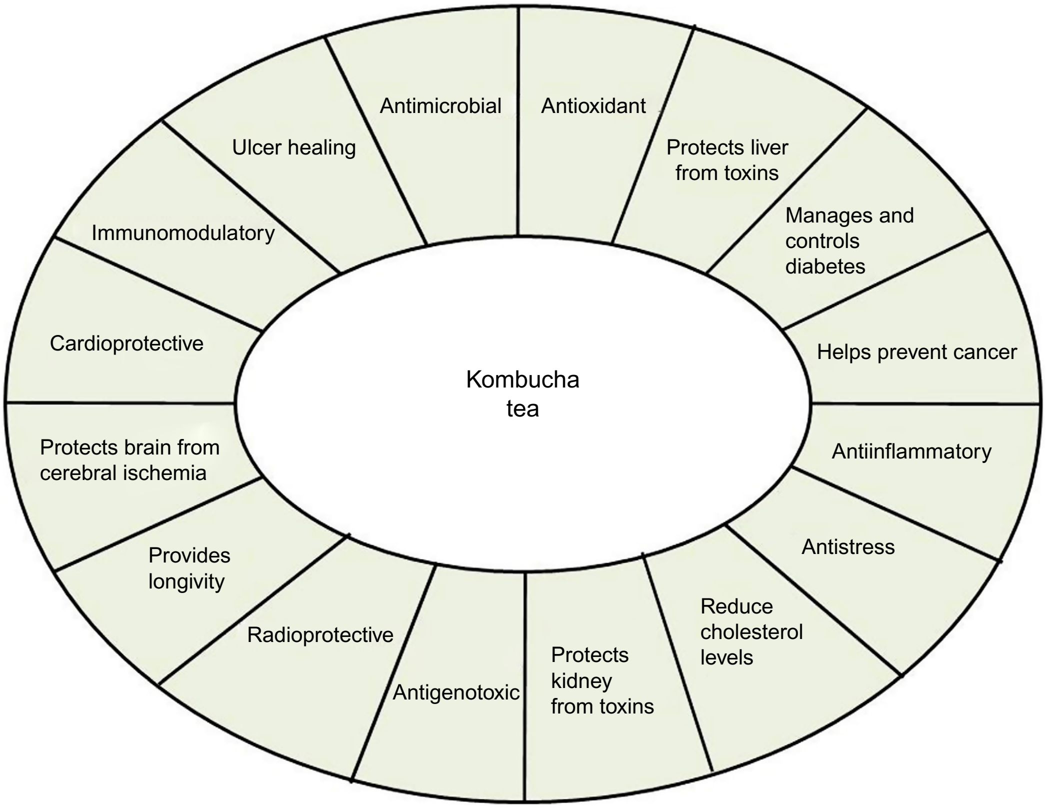

Fig. 10.3 Health benefits of Kombucha tea.

10.7.1.1 Antioxidant

Oxidative stress is an important contributor to the pathophysiology of a variety of pathological conditions (Rashid et al., 2013; Araújo et al., 2016). As a result, molecules with antioxidative properties are gaining increasing interest as therapeutic agents for oxidative stress-related diseases. Recently, much attention has been paid to replacing synthetic antioxidants with natural alternatives like traditional foods and medicines derived from natural sources containing antioxidant molecules (Kumari et al., 2011). The antioxidant property of KT has been investigated by many researchers. Jayabalan et al. (2008a) reported that KT prepared from green tea, black tea, and tea waste material possesses potent free radical scavenging activities. They have demonstrated the scavenging activity of KT on DPPH radical, superoxide radical, and hydroxyl radical. Recently, Gramza-Michałowska et al. (2016) studied the antiradical capacity of Kombucha beverage prepared from white, green, yellow, and black tea and the highest total phenolics content and DPPH radical scavenging ability were detected in yellow tea samples. They have also evaluated the sensory value of KT and noticed that the consumers willingly accepted the fermented drink. Malbaša et al. (2011) studied the effect of three starter cultures on the antioxidant activities of green tea and black tea Kombucha beverage against hydroxyl and DPPH radicals. They were a mixed culture of acetic bacteria and *Zygosaccharomyces* sp., a mixed culture of acetic bacteria and *Saccharomyces cerevisiae*, and native local Kombucha. The highest antioxidant activity for the black tea KT was observed when a combination of AAB and *Zygosaccharomyces* sp. was used as the inoculum for the fermentation. The native Kombucha community on the other hand imparted maximum bioactivity when it fermented the green tea variant. Chakravorty et al. (2016) showed the NO (nitric oxide) scavenging activity of KT suggesting its role in combating nitrosative stress as well. The beverage also exhibited potent antioxidant activity against lead (Dipti et al., 2003) and chromate (Sai Ram et al., 2000) induced oxidative stress. In most of the cases, the antioxidant activity of KT was found to be higher than the unfermented tea. It is assumed that some low molecular weight components are produced, and the tea polyphenols are structurally modified by enzymes of Kombucha consortium during fermentation. The activity of KT was also found to increase with fermentation time. Thus, the extent of activity mainly depended upon the type of tea material, the microbial composition of the Kombucha culture, and the fermentation time which in turn, determined the nature of the metabolites. However, prolonged fermentation leads to accumulation of organic acids, which might be harmful when consumed directly. Moreover, a decrease in free radical scavenging properties was also found during KT storage for 90 days (Jayabalan et al., 2008b). Therefore, this fermented beverage could be used as a source of antioxidants for many pathophysiological conditions provided it is prepared and preserved properly.

10.7.1.2 Antidiabetic

Diabetes mellitus is the most common endocrine disorder that as of 2014, affects a whopping 422 million people worldwide (WHO, 2017). It is characterized by hyperglycemia resulting from defects in insulin secretion, action, or both. Oxidative stress plays an important role in diabetes, contributing to the progress of diabetic complications and different organ damage (Ullah et al., 2016). Many antioxidants or antioxidant containing foods have been investigated as protection against diabetic oxidative stress, and it has been reported that some antioxidants play an important role in the mitigation of oxidative stress in diabetes mellitus (Pal et al., 2014; Ghosh et al., 2015; Manna and Sil, 2012).

KT, being a traditional fermented beverage and containing many antioxidant molecules, has been found to possess antidiabetic activity (Dashti and Morshedi, 2000; Shenoy, 2000). Oral administration of

KT reduced the blood glucose level in both alloxan and streptozotocin (STZ)-induced diabetic rats (Aloulou et al., 2012; Bhattacharya et al., 2013; Srihari et al., 2013b). Moreover, it also prevents weight loss in diabetic rats (Morshedi et al., 2006). Srihari et al. (2013b) reported that Kombucha supplementation significantly decreased glycosylated hemoglobin (HbA1c) and increased the levels of plasma insulin, hemoglobin, and tissue glycogen which was decreased on STZ treatment and significantly reversed the altered activities of gluconeogenic enzymes such as glucose-6-phosphatase, fructose-1,6-bisphosphatase, and glycolytic enzymes such as hexokinase in the tissues of experimental rats. KT was also found to effectively restore alloxan-induced changes of the parameters related to oxidative stress like lipid peroxidation end products, protein carbonyl content, glutathione content, and antioxidant enzyme activities in the pancreatic, renal, cardiac, and hepatic tissues of diabetic animals (Bhattacharya et al., 2013). They also showed that it could ameliorate DNA fragmentation and inhibit caspase-3 activation in the pancreatic tissue of alloxan injected diabetic rats. In all the cases, KT showed more efficiency than black tea. The antiglycation activity of KT was also found to increase with fermentation time (Chakravorty et al., 2016). This may be due to the production of some antioxidant compounds that are very likely to be metabolites produced by the bacteria and/or yeasts during the fermentation period. Hosseini et al. (2016) performed a comparative study of the effect of Kombucha prepared from green and black teas on the level of blood glucose and lipid profile of diabetic rats and showed KT prepared from green tea was better than black tea KT. All these studies suggest that KT may be considered as a potential functional food for the treatment and prevention of diabetes and its secondary complications.

10.7.1.3 Hepatoprotective

Liver is an important organ of all vertebrates that plays a major role in detoxification. Hepatocytes, the most abundant cells of liver, play an important role in the exogenous chemicals and toxins metabolism, and thus make liver a target for toxic substances (Ghosh et al., 2016; Saha et al., 2016; Pal et al., 2015). Hepatoprotection is the ability to prevent the damage occurring to the liver by toxic substances (Adewusi and Afolayan, 2010). Many studies have been carried out on cell lines and animal models to show the hepatoprotective activity of KT against various environmental pollutants and toxins such as carbon tetrachloride (Murugesan et al., 2009), cadmium chloride (Ibrahim, 2011), TBHP (tertiary butyl hydroperoxide) (Bhattacharya et al., 2011a), thioacetamide (Kabiri et al., 2014), trichloroethylene (Gharib and Gharib, 2008), acetaminophen (Abshenas et al., 2012; Wang et al., 2014), aflatoxin B1 (Jayabalan et al., 2010), and paracetamol (Pauline et al., 2001).

In all these studies, it has been demonstrated that KT can ameliorate the toxic effects of these liver toxicants efficiently. Recent study showed that KT prevents obese mice to develop hepatic steatosis and subsequent liver damage (Hyun et al., 2016). However, in all the abovementioned studies, the volume of Kombucha, number of doses, treatment period, and the method of administration were different. Hepatoprotective efficacy of KT was studied by measuring markers associated with liver toxicity like serum glutamic pyruvate transaminase, serum glutamic oxaloacetic transaminase, alkaline phosphatase, etc., reduced glutathione, antioxidant enzymes activities, nitric oxide levels, and histopathological analysis of liver tissue. Results showed that KT could normalize all these parameters more efficiently than black tea. The mechanism of hepatoprotection provided by KT might be attributed to its ability to facilitate both antioxidant and detoxification processes in the liver. Moreover, exploring the detailed molecular mechanisms involved in the hepatoprotective effect of KT revealed its antiapoptotic ability via suppression of mitochondria-dependent pathway (Bhattacharya et al., 2011b). The enzymes, bacterial acids, and other secondary metabolites produced by the microbes during fermentation of Kombucha have displayed the ability to detoxify body (Dufresne and Farnworth, 2000). Many scientific studies have reported that the detoxifying ability of KT is mainly attributed to the presence of glucuronic acid which can bind to the toxins in the liver and encourage them to flush out of the body (Nguyen et al., 2014). Thus, it can be concluded that KT could be used as a preventive as well as curative agent against oxidative stress-mediated hepatotoxicity.

10.7.1.4 Antimicrobial

Antibacterial

KT has been considered as a potential antimicrobial source and its inhibitory activity against different pathogenic microorganisms has been studied by many researchers. In 1996, Steinkraus et al. determined that the fermented tea beverage which contains dry tea (4.36 g/L) and 10% sucrose produced no antibacterial effect beyond that caused by acetic acid which was a main product of fermentation. Greenwalt et al. (1998) determined that KT sample fermented to contain 7 g/L acetic acid in a total acid content of 33 g/L, showed greater antimicrobial activity against Gram-positive organisms like *Staphylococcus aureus*, *Bacillus cereus* and Gram-negative organisms like *Escherichia coli*, *Salmonella choleraesuis* serotype typhimurium, and *Agrobacterium tumefaciens*. From the studies of Sreeramulu et al. (2000, 2001), it was found that KT exerted inhibitory activity against other different pathogenic Gram-negative organisms like *Enterobacter cloacae*, *Pseudomonas aeruginosa*, *Aeromonas hydrophila*, *E. coli*, *Salmonella enteritidis*, *Salmonella typhimurium*, *Yersinia enterocolitica*, *Shigella sonnei*, *Campylobacter jejuni*, *H. pylori* and Gram-positive organisms like *B. cereus*, *Staphylococcus epidermis*, and *S. aureus*. According to Talawat et al. (2006), KT prepared from black tea showed highest antibacterial activity when compared to that prepared from Japanese green tea, mulberry tea, jasmine tea, and oolong tea. *Vibrio parahaemolytica* was found to be the most susceptible among several human and shrimp pathogens such as *Vibrio cholerae*, *Salmonella typhi*, *P. aeruginosa*, *Vibrio harveyi*, *Vibrio alginolyticus,* and *Vibrio vulnificus* (Talawat et al., 2006).

Furthermore, KT prepared from fermenting both green and black teas for 21 days showed antimicrobial efficacy against various human pathogenic microorganisms such as the Gram-positive *Staphylococcus epidermidis*, *S. aureus*, *Micrococcus luteus*, and *Listeria monocytogenes* and Gram-negative *E. coli*, *P. aeruginosa*, *S. Typhimurium* (LT2), and KT from green tea exerted most antimicrobial potential (Battikh et al., 2012a,b). Also, KT prepared from *Melissa officinalis* L. (lemon balm tea) fermentation was found to exhibit strong antimicrobial activity against both Gram-positive prokaryotes such as *S. aureus* and *B. cereus* and Gram-negative prokaryotes such as *S. enteritidis*, *E. coli*, *Proteus mirabilis*, *P. aeruginosa*, and *Erwinia carotovora* (Četojević-Simin et al., 2012; Velićanski et al., 2007). Afsharmanesh and Sadaghi (2013) suggested that KT could be used as an alternative source of promoters to antibiotic growth in the diets of broiler chickens, thus increasing their body weight, feed intake, and protein digestibility. Significant antibacterial activity was also observed in KT prepared from fermentation of both green tea and black tea for 12 days against the Gram-negative bacilli such as *E. coli*, *P aeruginosa*, and *Salmonella enterica* serovar typhimurium and Gram-positive bacilli such as *Enterococcus faecalis*, *M. luteus*, *S. aureus*, and *S. epidermidis* (Deghrigue et al., 2013). Recently, it has been reported that black tea KT, fermented for 14 days, showed maximum antibacterial activity against enterotoxigenic *E. coli*, *V. cholerae*, *Shigella flexneri*, and *S. typhimurium*. While *V. cholerae* was found to be most susceptible to KT treatment, *S. typhimurium* was identified to be most resistant (Bhattacharya et al., 2016).

Antifungal

Sreeramulu et al. (2000, 2001) reported inhibitory activity of KT prepared from black tea against *Candida albicans* but not against *Z. bailii*. The 21-day fermented KT from both green and black teas inhibited the growth of *C. albicans*, *Candida tropicalis*, *Candida parapsilosis*, *Candida glabrata*, *Candida dubliniensis*, and *Candida sake* excepting *Candida krusei* (Battikh et al., 2012a,b).

Thus, KT has been found to demonstrate potent antimicrobial activity against different Gram-positive and Gram-negative pathogenic microorganisms. According to Greenwalt et al. (1998), the antimicrobial activity of Kombucha was contributed by acetic acid, a major antimicrobial compound of KT. But Sreeramulu et al. (2000) and Battikh et al. (2012a) have suggested that besides acetic acid or other organic acids, other bioactive components such as polyphenols, bacteriocins, proteins, enzymes, etc. may be responsible for the antimicrobial activity of KT. Moreover, a recent study by Bhattacharya et al. (2016) reported a polyphenolic fraction of ethyl acetate extract of 14-day-old KT that showed significant antimicrobial activity. This fraction has been identified to be composed of cathechin and isorhamnetin. This finding is an indicative of the fact that the antimicrobial activity of KT may be contributed to the ample polyphenols present in the beverage.

10.7.1.5 Antiinflammatory

Many researchers have investigated the antiinflammatory activity of KT using different experimental model systems. KT prepared from fermenting black tea was found to possess significant activity against oxidative stress induced by chromate (VI) treatment in male Sprague–Dawley albino rats. Chromate treatment significantly increased the plasma and tissue levels of malondialdehyde (MDA), glutathione peroxidase, catalase activities, and decreased delayed type of hypersensitivity (DTH) response. However, a complete reversion of these changes was observed in KT-fed rats (Sai Ram et al., 2000). Black tea KT also produced antiinflammatory activity against lead-induced oxidative stress in male Sprague-Dawley albino rats by decreasing lipid peroxidation (MDA levels) and DNA damage, increasing the levels of antioxidant enzymes associated with an increase in reduced glutathione and glutathione peroxidase activity (Dipti et al., 2003). KT from black tea also showed protective activity against trichloroethylene-induced nephrotoxicity in male albino rats (Gharib, 2009).

In 2010, histopathological and biochemical studies by Banerjee et al. established that KT prepared from black tea fermentation could produce healing effect against gastric ulceration induced by the nonsteroidal antiinflammatory drug (NSAID) indomethacin male Swiss albino mice. They inferred that the antioxidant activity, the ability to protect gastric mucin and reduction of gastric acid secretion by the beverage might be pivotal aspects behind its ulcer healing capabilities. Furthermore, black tea KT ameliorated experimental autoimmune encephalomyelitis model for multiple sclerosis in female C57BL/6 mice (Marzban et al., 2015). Also, black tea KT and green tea KT, both fermented for 12 days showed protective effects against a hypercholesterolemic diet in adult male Wistar rats by means of its antioxidant activity (Bellassoued et al., 2015). A recent study showed that KT prepared from oak leaves produced potent antiinflammatory activity in lipopolysaccharide (LPS)—stimulated macrophages by decreasing the levels of NO, tumor necrosis factor (TNF)-α, and interleukin-6 (IL-6) (Vázquez-Cabral et al., 2017). The antiinflammatory activity of KT is mainly attributed to its various phenolic compounds and flavonoids (Banerjee et al., 2010; Bellassoued et al., 2015).

10.7.1.6 Anticancer

KT has been claimed to possess anticancer activity for many years based on personal observations and testimonials. A population study conducted in Russia, in 1951 by the Central Oncological Research Unit and the Russian Academy of Sciences, Moscow claimed anticancer properties of Kombucha (Dufresne and Farnworth, 2000; Jayabalan et al., 2014). Cetojević-Simin et al. (2008) studied the antiproliferative activity of several KT samples prepared from traditional black tea and winter savory tea (*Satureja montana* L.) on different cell lines such as HT-29 (colon adenocarcinoma), HeLa cells (cervix epithelial carcinoma), and MCF-7 (breast adenocarcinoma) using the sulforhodamine B colorimetric assay. The authors stated that the antiproliferative properties of both winter savory tea KT and traditional black tea KT were comparable and further concluded that KT from winter savory tea might be consisting of more active antiproliferative components than that of the water extracts of winter savory tea. Furthermore, an ethyl acetate extract of KT prepared from black tea containing dimethyl 2-(2-hydroxy-2-methoxypropylidene) malonate and vitexin at a concentration of 100 μg/mL each, produced significant cytotoxic effects on 786-O (human renal carcinoma) and U2OS (human osteosarcoma) cells. The treatment additionally resulted in reduction in cell invasion and cell motility in A549 (human lung carcinoma), U2OS, and 786-O cells, followed by reduction in the effects of matrix metalloproteinase-2 (MMP-2) and MMP-9 in 786-O cells and MMP-2 in A549 cells (Jayabalan et al., 2011). A recent study also showed that lyophilized extract of KT prepared from fermenting black tea decreased significantly the survival of prostate cancer cell line PC-3 by downregulating the expression of angiogenesis stimulators like cyclooxygenase-2, matrix metalloproteinase, endothelial growth factor, interleukin-8, and human inducible factor-1α. Therefore, KT can alter the expression of different angiogenic stimulators, resulting in inhibition of angiogenesis (Srihari et al., 2013). KT prepared from green tea showed significant cytotoxic activity against the human cancer cell lines A549 and Hep-2 (epidermoid carcinoma) whereas KT from black tea was found to be effective only against Hep-2 (Deghrigue et al., 2013). The possible mechanisms of anticancer activity of KT as agreed by most researchers are mainly (1) inhibition of cancer-cell proliferation; (2) inhibition of gene mutation; (3) termination of metastasis; and (4) induction of cancer-cell apoptosis (Jayabalan et al., 2014). The presence of a variety of compounds such as polyphenols, glucuronic acid, gluconic acid, lactic acid, vitamins like (vitamin C), and D-saccharic acid-1,4-lactone (DSL) might contribute to the anticancer properties of KT (Deghrigue et al., 2013).

10.7.1.7 Other Therapeutic Properties of KT

Apart from the above-mentioned health benefits of KT, the fermented beverage is also known to possess other therapeutic activities. The beverage has been reported to possess antistress activity (Pauline et al., 2001). The authors exposed albino rats to cold and hypoxia followed by evaluating the levels of MDA and reduced glutathione in plasma/blood. The effect of KT on physical stress-like restraint, etc., was additionally studied by recording the fecal output of the animal. KT was also found to have hypocholesterolaemic effect, as an evident from its capability in lowering the total cholesterol and low-density lipoprotein (LDL) cholesterol in high-cholesterol fed mice (Yang et al., 2009; Bellassoued et al., 2015). These effects were largely attributed to DSL (Yang et al., 2009). Another study showed that oral supplementation of KT at a dose of 5 mg/kg body weight in alloxan-induced diabetes rats, inhibited α-amylase, and lipase enzyme activity in the plasma and pancreas of the experimental animals, thereby suggesting its antilipidemic activity (Aloulou et al., 2012). As KT could inhibit pancreatic alpha-amylase in the small intestine, it helps in starch digestion and net absorption of glucose (Kallel et al., 2012). KT also exhibited nephroprotective effect against thrichloroethylene-induced oxidative stress in rats (Gharib, 2009). Maghsoudi and Mohammadi (2009) showed the effect of Kombucha on intraabdominal adhesion formation after operation in rats. Peritoneal adhesions are fibrous bands of tissues formed between organs that are normally separated and/or between organs and the internal body wall after peritoneal injury. Intraperitoneal administration of KT was found to be useful in preventing such peritoneal adhesions. The fermented beverage also exerted protective role on phenol-induced cytotoxicity in the erythrocytes of albino mice. The study showed KT supplemented mice had a lower micronucleus frequency than erythrocytes in only phenol-treated group (Yapar et al., 2010). Again, antigenotoxic effect of lemon balm Kombucha was confirmed on mitomycin-C-damaged Chinese hamster cells CHO-K1 using chromosome aberration assay (Četojević-Simin et al., 2012). Cavusoglu and Guler (2010) showed the potential radioprotective effect of KT on gamma radiation-induced chromosomal aberrations in human peripheral blood lymphocytes in vitro. Pretreatment of peripheral blood lymphocytes with KT resulted in a significant reduction in the frequency of mitotic index and the numbers of aberrant metaphases before radiation exposure. KT was also found to ameliorate gamma radiation-induced cellular and DNA damage in human blood lymphocytes (Mondal et al., 2015). A pilot study on the effect of chronic Kombucha ingestion by c57-bl/6 mice showed that both male and female mice that drank Kombucha lived longer than controls (Hartmann et al., 2000). KT was further found to have a protective role against the effects of electromagnetic radiation in rat hearts and lungs (Gharib, 2011). Treatment with KT furthermore attenuated the increase in different trace elements levels like copper, zinc, and iron in the brain, spleen, and intestine of male albino rats exposed to a 950 MHz electromagnetic field (Gharib, 2013). It was additionally found to have protective effect on brain damage induced by transient cerebral ischemia and reperfusion in rats (Kabiri and Setorki, 2016). The fermented beverage was found to detoxify patulin which is a toxic chemical contaminant produced by several species of fungi. Exposure to this mycotoxin is associated with immunological, neurological, and gastrointestinal outcomes (Puel et al., 2010). Ismaiel et al. (2016) showed that KT significantly inhibited patulin production by three toxigenic fungal strains (*Penicillium expansum* LC015096, *Talaromyces purpureogenus* LC015095, and *Acremonium implicatum* LC015097) in liquid medium and apple fruit. Recent study showed the myocardial potency of KT against isoproterenol-induced myocardial damage in rats by preventing membrane destabilization (Lobo and Shenoy, 2015). Induction of myocardial infarction using isoproterenol resulted in a significant decrease in tissue antioxidants and an increase in the levels of total, ester and free cholesterol, triglycerides, free fatty acids, and glycoprotein components in plasma and heart. The phospholipid content showed an increase in plasma and a simultaneous decrease in the heart tissue, while the Na+/K+ ATPase activity decreased and Ca^{+2} ATPase and Mg^{+2} ATPase activities increased, resulting in destabilization of the membranes. Pretreatment with KT could bring these components to near normal, indicating its reactive oxygen species scavenging, lipid-lowering, membrane-stabilizing, and glycoprotein-modulating effects (Lobo et al., 2017). Very recently, KT has been found to have antidepressant effect against reserpine-induced depression in mice (Rabiei et al., 2017). Another very recent study reported that KT zoogloea has positive effects on intestine microbiome of rats during the experimental antibiotic-associated dysbiosis (Ivanovna et al., 2017). They showed that animals receiving gentamin sulfate for induction of intestinal dysbiosis, had a significant decrease in the total bacterial counts, as well as the decrease of *Bifidobacterium* spp., *Lactobacillus* spp., and *E. coli* levels and increase of *Candida* spp. levels. However, group of animals treated with KT after discontinuation of gentamicin administration showed rapid disappearance of dysbiosis symptoms and the number of microflora started to improve significantly.

All these evidences supported the health promoting properties of KT and established it as a functional food.

10.7.2 Industrial Applications

Apart from various health benefits of KT, this fermented beverage also has other beneficial aspects. Mamisahebei et al. (2007) showed that arsenic(V) could be effectively removed from aqueous solution using Kombucha biomass and the pretreatment of biomass with $FeCl_3$ was found to improve the biosorption efficiency. The maximum uptake capacity (qm) of tea fungal biomass for As(V) was found to be 3.98 μmol/g in the pH ranges between 6 and 8 and the equilibrium was reached in 90 min. Very recently, Hopfe et al. (2017) reported the leaching of rare earth elements by the microbial community of Kombucha from fluorescent powder. The highest leaching rates were observed in shake cultures using the entire Kombucha-consortium or its supernatant as leaching agent. However, when *Zygosaccharomyces lentus* and *Komagataeibacter hansenii*, isolated from the Kombucha community were used as leaching agents, the results were not so encouraging. The tea fungus is also used to produce some compounds with medicinal significances. Glucuronic acid is the key component in human health due to its detoxifying action through conjugation to the xenobiotic metabolisms in liver. Yavari et al. (2011) have optimized glucuronic acid production using Kombucha biomass on grape juice with the help of response surface methodology. Another group of investigators prepared and characterized Kombucha-synthesized bacterial cellulose (KBC) and further evaluated the biocompatibility of KBC with peripheral nerve cells and tissues in vitro and in vivo (Zhu et al., 2014). They showed that KBC possessed good biocompatibility with Schwann cells cultured primarily and did not exert hematological and histological toxic effects on nerve tissues when performed in vivo.

10.8 Conclusion

This chapter has had the aim of impressing upon its readers that Kombucha is not just a tea extract. Anybody who is aware of the process of making tea might be confused because for tea, fermentation usually refers to the degree of oxidation that the leaves are subjected to. As we have discussed before, in case of green tea, the leaves are not at all oxidized, while complete oxidation is done for black tea. What happens in KT is that a consortium of yeast and bacteria biotransforms the tea into a fermented food beverage. The beauty of this fermentation is that despite being a sugar-based yeast fermentation, the final beverage after 21 days, is left with <1% of ethanol due to the activity of the bacteria.

The central player of KT is the AAB; AAB are very special kind of bacteria on several counts: to name a few, (i) they are highly aerobic, (ii) consume lots of carbohydrate without making much of biomass (low growth yield), but successfully keep the niche to themselves, (iii) a few of them produce bacterial cellulose, (iv) a few of them are nitrogen fixing, but most importantly, and (v) they thrive in high sugar and low pH environments like yeast. Thus, KT is a natural cradle where AAB and yeast live together. Yeast could be either aerobic, facultative to anaerobic (bottom dweller).

Just as the knowledge of fermentation is not unknown to mankind, KT, also is far from being a secret. It has been consumed for thousands of years and its popularity is only increasing by the day. Apart from having several health benefit effects, another very important reason behind its ever-increasing popularity is its ease of production under household conditions. The beverage more than often only requires normal clean environments, and hence large-scale production facilities are also growing as we delve upon the topic. The only major constraints in producing the beverage are to keep away from (i) flies, as they are attracted to ethyl acetate ester produced, (ii) excess moisture that often result in an invasion by unwanted fungi, and (iii) any metal surface as that could produce toxicity in the acidic environment.

During the last two decades or so KT has come of age with several scientific explorations which have slowly but steadily established most of the claimed effects on several health conditions. To summarize, KT has been reported to have positive impact in prevention and/or cure of diseases such as: diabetes, ulcer, hepatotoxicity, cancer, inflammation, cardiac problems, etc. In view of the topics that we have touched in this chapter about this very cheap yet effective functional beverage, we hope that soon Kombucha would become the beverage of choice of every household.

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