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REVIEW ARTICLE

Palm Sap—Quality Profiles, Fermentation Chemistry, and Preservation Methods

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Abstract Palm sap is quite nutritious and highly prone to fermentation. The unfermented juice could be an ideal health drink. Palm sap's quality profile and fermentation chemistry help to predict its shelf life and potential safety. There is demand from farmer–producer companies and food processing industries to develop bottling technology and a transportation/distribution protocol for palm sap similar to common soft drinks. Different techniques were followed for bottling palm sap, but none proved successful at the pilot level or commercial scale. To develop a systemic preservation technique, it is crucial to understand the biochemical composition, fermentation chemistry, and existing preservation methods and their disadvantages. This review mainly focuses on the chemical, microbial, volatiles, and flavor changes of palm sap. In addition to a detailed discussion on contemporary sap preservation techniques, this paper also addresses the effect of pasteurization in combination with preservatives, such as nisin, sodium benzoate, chitosan, potassium sorbate, sorbic acid, citric acid, and sodium metabisulfite, on the shelf life of sap, challenges to preserving palm sap, and future directions for preservation methods.

Keywords Palm sap · Inflorescence · Fermentation · Nisin · Pasteurization · Neera

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Introduction

Palm trees encompass perennial lianas, shrubs, and trees. They are part of the Arecaceae family and can be found from the 44° northern latitude to the 44° southern latitude (Wood 2002). Nearly 2600 species of palm trees have been found in tropical and warm temperate zones. One of the world's oldest flowering plants is the palm (Redhead 1989). Many common products and foods originate from palms. These include traditional fresh juices and fermented beverages such as toddy, wine, and arak, concentrated syrup such as honey, and brown sugar (jaggery powder). These are all produced by tapping sap from various palm trees. In Table 1, some of the common palm trees used for sap extraction, their habitat, and products are explained. One of the fermented drinks enjoyed by the Asian and African population is palm toddy (Jirovetz et al. 2001). The annual production of palm juice and toddy was estimated at 9×10^6 L per annum (Davis and Johnson 1987).

Arenga pinnata palm was believed to be the first source of palm sugar used by humans. *Borassus flabellifer* palm sugar was used in India during the fourth century BC. Similarly, in Sri Lanka, *Caryota urens* palm jaggery was used as sugar in the distant past. The palm sap is known as '*Neera*' and is boiled for 3 to 4 h above 100 °C to produce brown sugar with a sweet taste (Apriyantono et al. 2002; Ho et al. 2007). These sugars are used for making chocolate, cake, sweet soy sauce, food coating, ice cream, and typical Bengali sweets. The results of earlier research studies have confirmed that coconut and palm sugar consist mainly of sugars (glucose, fructose, and sucrose) at amounts of 91.4 and 89.2%, respectively (Apriyantono et al. 2002). Due to the presence of sugars, palm sap is subjected to the Maillard reaction and caramelization while heating. Pyrazines, furans, ketones, fatty acids, and organic

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Table 1 Scientific and common name of palm trees, their habitat, products, and other information

Scientific and common name	Native distribution and habitat	Major products	Minor products	Comments and selected references
Amiga pinnata sugar palm	S. and SE Asia south tropical rain forest into dry forest, to 1200 m	Sap used to make sugar, wine, alcohol, and vinegar. Sap yield 3–6 L/tree/day, starch from stem yield 75 kg/tree	Leaf sheath fiber; edible heart; etc.	Solitary, terminal flowering feather palm; traditional multipurpose palm with a history of cultivation; strong candidate for domestication; agroforestry potential (Mogea et al. 1991)
Borassus flabellifer, R. aethiopum palmyra, ron	S. and SE. Asia; Africa tropical dry forest into savanna, to 750 m	Sap to make sugar, wine, alcohol, vinegar, sap yield 11–20 L/tree/day	Leaf stalk fiber; leaves for thatching and basketry; edible immature fruit	Solitary fan palms; multipurpose species of major utility to local peoples: incipient management already in practice in S. and SE. Asia: candidate for domestication, agroforestry (Davis and Johnson 1987)
Caryota urens *Fish tail palm*	S. and SE. Asia tropical rain forest to 1500 m	Sap to make sugar, wine, alcohol, vinegar, sap yield 20–27 L/tree/day; starch from stem, yield 100–150 kg/tree	Leaf sheath fiber; edible heart; etc.	Solitary, terminal flowering feather palm; numerous products; informal cultivation practiced; domestication potential in agroforestry systems (De Zoysa 1992)
Raphia spp. *raffia*	West Africa tropical rain forest, seasonally flooded lowland sites	Commercial leaf base fiber (African bass fiber) for brushes and brooms; sap for wine. Alcohol	Petioles as poles, leaves for thatching and weaving; etc.	Suckering (most spp.) terminal flowering feather palm; *R. hookeri* and *R. palma-pinus* are main brush fiber sources. Also, tapped for sap; one or more spp could be managed for multiple products (Tuley 1994)
Corypha umbraculifera *C. utan* *Talipot buri*	S. and SE. Asia tropical rain forest to 600 m	Sap to make sugar. wine alcohol, vinegar, sap yield 20 L/tree/day for 3–4 months for *C. utan*; starch from stem petiole to make hats; leaf midrib used to make furniture	Leaves for thatching and weaving various products, edible heart; etc.	Solitary, terminal flowering fan palm multipurpose palm with good mix of commercial and subsistence products; strong candidate for management or domestication, also agroforestry potential (Madulid 1991)
Phoenix sylvestris wild date	S. Asia tropical rain forest to dry forest, to 1500 m	Sap used to make sugar and wine. Sugar yield 40 kg/tree/year; edible fruit	Leaves for weaving and to make brooms; stem wood for fuel; etc.	Solitary feather palm; already under management and informal cultivation; good multipurpose palm with domestication potential within agroforestry systems (Davis 1972)
Hyphaene spp. doum	Africa semideserts and deserts, to 600 m	Edible fruit; sap for wine and alcohol	Leaves for thatching and weaving	Solitary branched fan palm; management of wild stands would provide sustainable sources of commercial and subsistence products in dry areas (Tuley 1994)
Nypa fruticans nipa	S. and SE Asia tropical rain forest, brackish water swamps of tidal rivers	Sap for sugar, alcohol, sugar yield 3000 kg/ha/year; leaves for thatching (atap)	Edible fruit: etc.	Suckering feather palm; incipient management in practice. could benefit from improved practices and broader utilization of products, especially in Papua, New Guinea (Hamilton and Murphy 1988)
Cocos nucifera (*coconut*)	Spread across much of the tropics and grown in 90 countries	Every product is useful. Important product is nut, sap, and oil	All parts are useful	Inflorescence is used for tapping. Tapping coconut inflorescence for sap and its sale as health drink or value-added products like coconut sugar would improve the returns by at least 8–10-fold: (Hebbar et al. 2015a)

Table 1 continued

Scientific and common name	Native distribution and habitat	Major products	Minor products	Comments and selected references
Phoenix sylvestris thakil (sugar date palm)	India (common) Nepal	Multipurpose palm: sap from stem as beverage and to make sugar	Edible fruit: leaves made into brooms or woven into baskets and mats; stem wood for fuel	A graceful palm 10–16 m tall with a large crown and rough trunk covered with persistent leaf bases. Fruiting spadix about 90-cm-long, bearing oblong-ellipsoid berries (Madulid 1981)
Nypa fruticans (monotypic) golpata	India: Orissa, West Bengal. Andaman islands; Bangladesh; Sri Lanka	Sap from inflorescence for beverage or sugar	Leaves for thatching, mature seeds suitable for vegetable ivory	The yellow inflorescences are on long, sturdy stalks arising from the base of the plant. The female inflorescence is a densely packed, spherical head of flowers. The male inflorescence is a club-shaped spike of closely arranged flowers emerging from lateral stalks below the female inflorescence. The large spherical fruiting body is 30–45 cm in diameter (Bonde et al. 1990)
Arenga pinnata Gomuti (sugar palm)	India : Eastern Andaman Islands Bangladesh, Sri Lanka	Multipurpose palmi sap for sugar and other products	Edible immature seed (fresh mesocarp of ripe fruit is filled with irritant needle crystals); edible starch from stem; edible palm heart; leaf base fiber for fish nets, etc., leaflets for weaving baskets, etc., stem wood for various uses	Sweet sap (toddy, tuba, sugar) is obtained by tapping the male inflorescences. An inflorescence can be tapped for 1–2 months, producing about 5–12 L of sap per day. This sap can be used as beverage or processed into vinegar, sugar, alcohol, or animal feed. Sugar yields may be about 70 kg/day per ha or 25 t/ha per year (Bonde et al. 1990)

acids constitute volatile components of coconut and palm sugar, and they are responsible for their sweet, toasty, and nutty caramel-like aroma (Sohn and Ho 1995). Pyrazines are released when palm sap is heated above 110 °C (Barbara and Michael 2004; Ho et al. 2007). The initial quality of palm sap, boiling temperature, and heating time affects the volatile components of palm sap (Martins et al. 2001).

The quality or nutrition and flavor of palm sap are appealing features for consumers. The appearance and flavor of palm sap indicate its stage of fermentation. Due to the development of convenient packaging and preservation techniques, the demand for palm sap is high. In this report, the tapping, collection, and quality profile of palm sap are reviewed and the present scientific status of fermentation chemistry and preservation methods of palm sap is outlined.

Palm Sap Tapping and Collection Process

Palm sap is tapped from the matured unopened inflorescence of the palm. The palm sap is collected by cutting the head of the inflorescence. In rural area, palm sap is traditionally collected from palmyra and coconut tree by organized practice for its local consumers (Nathanael 1966). The inflorescence of matured stage is tapped (Redhead 1989; Borse et al. 2007). The development of swelling at the base of the inflorescence is considered as the appropriate tapping stage. The identified coconut inflorescence is beaten uniformly and gently by traditional tools (bones/wooden sticks) during morning and evening for 7 days to simulate the sap flow. The selected spathe should be tied with a strong coconut fiber or coir rope in order to arrest bursting or opening of inflorescence. Then, 7–10 cm front portion of inflorescence is removed by sharp sickle. In the traditional method, the inflorescence to be tapped is inserted into an earthenware pot for sap collection. As the sap oozes drop by drop and over a period of time, it undergoes fermentation in the open container. It also gets contaminated by pollen, ants, insects, and other pollutants.

Thus, freshly collected sap is oyster white in color, sweet in taste, and has opaque appearance with acidic pH (Gupta et al. 1980). In India, to prevent fermentation of coconut inflorescence sap the inner surface of earthen pot is coated with lime. This practice prevents fermentation only to a certain extent. In Thailand, Kiam wood (*Cotylelolbium melanoxylon Pierse*) is added in the bamboo tube during the collecting process to slow down the growth of the microorganisms (Naknean et al. 2014).

Hygienic and Unfermented Collection of Palm Sap

ICAR-CPCRI (Central Plantation Crops Research Institute) developed the coco-sap chiller to collect the hygienic (contaminant-free) and unfermented (non-alcoholic) sap. It is made of a hollow PVC pipe, weighs 2.6 kg, and is, thus, portable and economically feasible to farmers (Rs. 2000/-). Top opening of the device is enlarged to insert a collection bag and receive the collected sap. The lateral opening is used for the insertion of a cut inflorescence (Fig. 1). The collection bag is usually a 100-micron-thick low-density polyethylene (LDPE) film that can hold two to three of liters of sap. Along with ice cubes, the collection box maintains the inside temperature at 2–3 °C for 10–12 h to prevent the sap from fermenting. Thus, the collected sap is fresh, unfermented, hygienic, sweet in taste, and delicious. It is called *Kalparasa*™ and registered the Trademarks Registry as class 32 "Mineral and aerated waters and non-alcoholic beverages; fruit beverages and fruit juices; syrups and other preparations for making beverages." It can be marketed as a fresh, ready-to-serve, and nutritious cold beverage. It can also be converted into different value-added products such as syrup, squash, concentrate, jaggery, and sugar.

As the sap that the coco-sap chiller collects is non-alcoholic and contaminant-free, the processing cost is minimized. The sap thus collected can be stored for 1–2 h under ambient condition, 1–2 days under refrigerated conditions (5–8 °C), and any length of time under deep freezer or sub-zero temperature. One way of marketing sap as fresh juice in small outlets entails using refrigerated dispensers. Efforts are on to develop tetra-pack or retort-pack technology to facilitate the long-distance transportation of the sap. Studies conducted on *kalparasa* using the 'coco-sap chiller' and on *neera* using traditional methods showed distinct variations in physical and biochemical properties (Tables 2, 3). *Kalparasa* is slightly alkaline (pH 7–8), golden brown in color, sweet, and delicious, while *neera* is oyster white, has a pH of 6 or below, and has an astringent smell. *Kalparasa* in addition to high sugar also contains amino acids, total phenols, flavonoids, and antioxidants which is 2.5, 1.5, 4.6, and 1.8 times, respectively, higher than *Neera*. *Kalparasa* is also rich in ascorbic acids, vitamins, and niacin. Further, the products of *Kalparasa* like sap concentrate and sugar were also found to be rich in amino acids, polyphenols, flavonoids, vitamins, and antioxidants (Hebbar et al. 2015a).

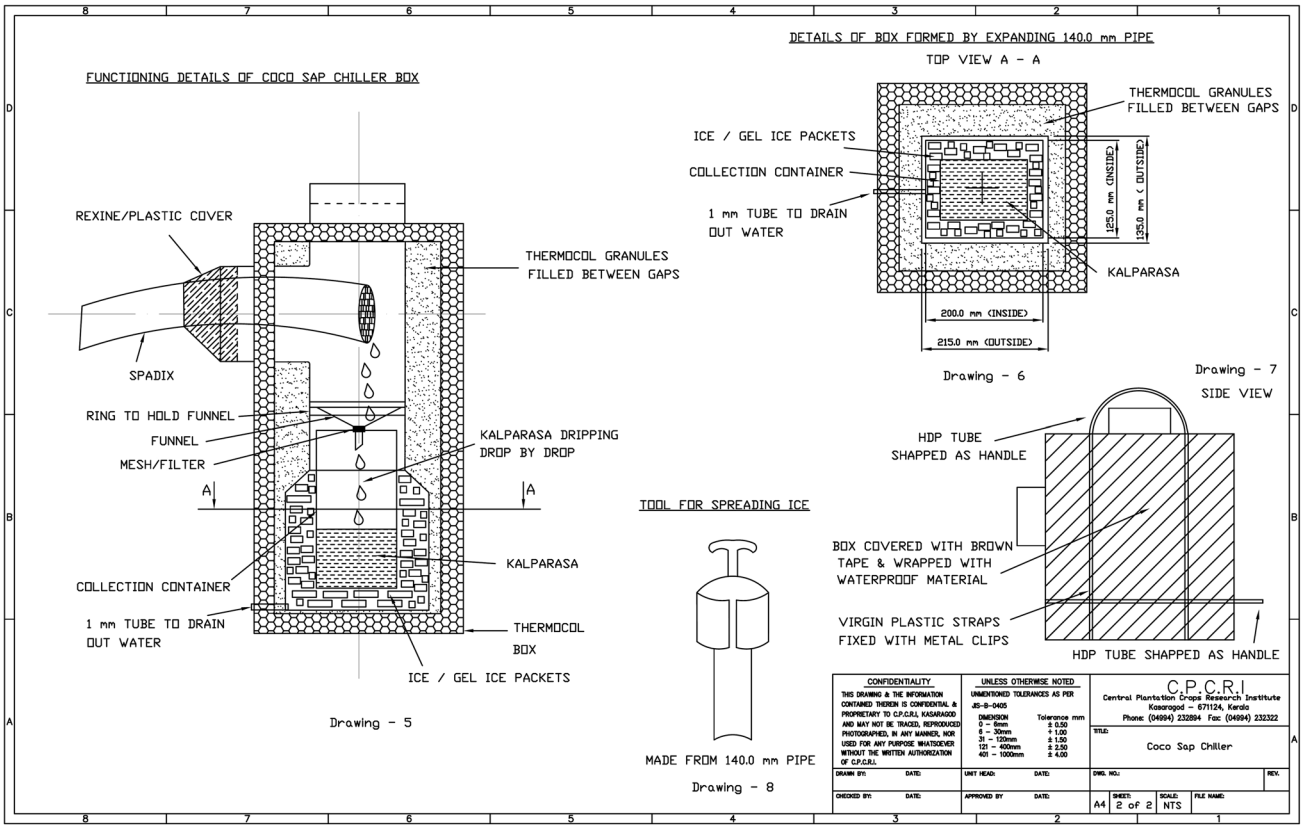

Fig. 1 Coco-sap chiller

Table 2 Quality attributes of *kalparasa and neera*

Attribute	*Kalparasa*	*Neera*
Total soluble solids (°Brix)	15.5–18	13–14
pH	7–8	6 or below
Color	Golden brown or honey	Oyster white
Flavor	Sweet and delicious	Fetid smell
Chemicals and extraneous matter	Absent	Present
Alcohol (%)	0	2.32

Table 3 Biochemical constituents and minerals of sap collected by coco-sap chiller and traditional method

Constituents	Sap collected by	
	Coco-sap chiller	Traditional method
pH	7.5	5.0
Total phenols (mg/100 g)	21.99	14.1
Total flavonoids (mg/100 g)	0.96	0.17
Free amino acids (mg/100 g)	901	350
Total sugars (g/100 g)	15.96	6.52
Alcohol %	Nil	4.7
Vitamins (mg/100 g)		
Vitamin C	13.45	4.73
Niacin	14.86	1.98
Vitamin E	7.9	2.95
Minerals		
Potassium (mg/100 mL)	168	–
Sodium	90.6	–
Iron	53	–
Zinc	20	–

Quality Profiles of Sap

Palm sap is the phloem sap and known to be very rich in nutrients. The composition and quality of sap are varying with the place and duration of tapping (Borse et al. 2007). The nutritious sap secreted from the unopened inflorescences before they converted into fruits is utilized in the preparation of typical, ethnic, and nutritious food products. The sap has been found with low glycemic index of 35 as compared to refined white sugars with glycemic index of 65 (Trinidad et al. 2010). The fresh sap is rich source of phenolics, vitamins (B and ascorbic acid), protein, essential elements (nitrogen, phosphorous, potassium, and magnesium), and micronutrients (zinc, iron, and copper). It helps in regulating high blood pressure and sugar metabolism due to the presence of potassium. It also acts as a potential antioxidant for overall production of immune system, respiratory system, cardiovascular, and reduction of inflammation. It also aids in electrolyte balance and acid/alkaline ratio (Hebbar et al. 2015a). The biochemical parameters and inorganic constituents of fresh coconut inflorescence sap are given in Table 4.

Carbohydrate

Generally, palm sap contains mostly sucrose, followed by glucose and fructose. The Ben Thabet et al. (2009) found 95.27% sucrose, 2.51% glucose, and 1.61% fructose in one of the palm species *Phoenix dactylifera.* Many studies have found that sugars with high sucrose content are known to be good for health. Among the carbohydrate, sucrose causes less fattening effect than glucose or starch (Jentjens and Jeukendrup 2005). Ruzzin et al. (2005) also reported 15% increase in energy consumption by sucrose without increase in weight gain. The total sugar content of coconut sap is high (16.19 g/100 mL), compared to its reducing sugar content (0.68). Further, sucrose- and fructose-rich sugars have low glycemic index (GI) and thus the coconut sap is reported to contain low GI (Trinidad et al. 2010). The extent of the sap's freshness can be easily measured by the composition of total and reducing sugars. If the sap is fresh and unfermented, the reducing sugar content should be less than 1.0 g/100 mL (Hebbar et al. 2013).

Proteins and Amino Acids

Fresh palm sap is rich in amino acids; it is mainly composed of asparagine and glutamine, which are polar side-chain amino acids. It is found that coconut sap contain about 17 amino acids which are the building blocks of proteins and also helps to maintain proper acid/alkaline balance (neutral pH) (Xia et al. 2011). Amino acid content of fresh sap is reported as 2.6 g kg^{-1}. The reduction in amino acid of sap was observed on first day due to the production of microorganism. But, the reason for decrease in amino acid during second day might be due to degrade in protein content of sap. The significant decreases in amino acid was also observed on third day, but later it was insignificant reduction which may be due to the less amount of amino acid by death or autolysis of part of the yeast (Jolly et al. 2006).

The polar side-chain amino acids are considered as essential precursors to the formation of volatiles during the production of sugar from palm sap, especially in the Maillard reaction, by releasing free amino acid groups (Sohn and Ho 1995). The released amino acid substrate catalyzes sucrose into monosaccharides or undergoes retro-

Table 4 Major biochemical and inorganic constituents of coconut inflorescence sap. *Source*: Hebbar et al. (2015b)

Biochemical constituents	Range[a]
pH	6.57–7.50
Total sugar (g)	10.08–16.50
Reducing sugar (g)	0.439–0.647
Amino acids (g)	0.123–0.338
Protein (g)	0.150–0.177
Phenolics (mg GAE)	4.80–5.40
Antioxidant activity[b] (mM TE)	0.299–0.355
Sodium (mg)	69.4–117.5
Potassium (mg)	146.1–182.4
Phosphorus (mg)	2.0–6.4
Manganese (mg)	0.009–0.014
Copper (mg)	0.028–0.035
Zinc (mg)	0.018–0.026
Iron (mg)	0.049–0.058

GAE gallic acid equivalent, *TE* Trolox equivalent

[a]Composition indicated is per 100 mL

[b]Antioxidant activity was measured by cupric ion reducing antioxidant capacity (CUPRAC method)

aldol reactions to produce C_2–C_5 dicarbonyl compounds, depending on cooking temperature and heating time (Martins et al. 2001; Carline and Van Boekel 2003). All these dicarbonyl compounds can react with amino acids to form a-aminoketones and aldehydes. Subsequently, in the later stage the chain reactions take place including dehydrations, cyclization, isomerizations, and retro-aldolizations, which ultimately lead to the formation of volatile compounds. These volatile substances are responsible for the characteristic aroma of palm sugar (Ho et al. 2007).

Minerals

Mineral elements are investigated in the sap originating from various palm species and reported about the predominant presence of potassium and some of the micronutrients. Mineral nutrients have importance in numerous metabolic functions. Indeed, zinc has a fundamental role in the brain, whereas calcium, iron, magnesium, etc., have physiological actions in muscular contraction, blood oxygenation, and coagulation, nervous impulses conduction, blood acid–base balance ensuring, and appropriate immune and heart functions. Some minerals, such as calcium, support the building of bone, teeth, or muscle tissues, while others are main components or activators of enzymes and hormone molecules. Mineral elements represent 4% human body weight and are provided by the food.

N'guessan et al. (2015) reported that inflorescence sap produced from various coconuts (*Cocos nucifera* L.) cultivated in Côte d'Ivoire is rich in minerals and showed the varietal difference. Ash contents ranged between 0.18 and 0.27% (w/w). Saps of MYD (0.26%) and $PB113^+$ (0.27%) were richer in ash compared to WAT and $PB121^+$. Thirteen minerals comprising eight microelements (K, Cl, Si, Na, Mg, P, S, and Ca) and five oligoelements (Fe, Cu, Mn, Zn, and Br) were found in the coconut inflorescence sap. Macroelements in the sap ranged between 1.25 and 90.65 mg/100 g, and oligoelements varied from few traces to 0.70 mg/100 g ($P < .05$). Coconut sap collected from MYD correlated with greatest mineral properties, with a large presence of P, K, Si, Fe, Na, Mg, and S. $PB113^+$ hybrid also revealed highly significant sap minerals. Conversely, the sap of WAT resulted in lower mineral contents.

Vitamins

Palm sap contains a wide range of vitamins such as vitamin C and B. It is found that in palmyrah sweet sap (*Borassus flabellifer),* vitamin C content is as high as 13.25 mg/100 cc, while for fresh coconut sap, it is 12.86 mg/100 cc. The vitamin C was observed to be decreased during first day of storage, but it increased to 20.7 mg/L during third day due to the activities of yeast during fermentation (Bremus et al. 2006).

Phenolics and Flavonoids

Phenolic compounds influence the color, sensory, nutritional, and antioxidant properties of foods. Free radical scavenging activity of the phenol moiety (hydroxyl substituent on the aromatic ring) is responsible for antioxidant property of phenolics. Generally, flavonoids constitute two-third of dietary phenols and phenolic acid occupy remains one-third. These simple phenolic acids have aroused awareness and demand due to the potential antioxidant nature and health advantage. They also have other biological activities, such as blocking the biosynthesis of leukotrienes by caffeic acid (one of the most prominent naturally occurring cinnamic acids) which are involved in immunoregulation diseases, asthma, and allergic reactions (Koshihara et al. 1984).

The total phenolic content of fresh sap is about 0.33 g/L. During fermentation, the phenolic content starts increasing and reached the peak level of 1.24 g/L at 58 h and then there was no significant change (Xia et al. 2011). The reason for increased phenolic content may be due to binding of plant polyphenols with protein, sugar, starch, and cellulose, and formation of glucosidic bonds. The acids

produced during fermentation degrade these glucosidic bonds and resulted in the production of phenolic components (Landbo and Meyer 2004). Phenolic components are also produced due to the metabolism of some microorganism. Xia et al. (2011) reported the presence of caffeic acid, gallic acid, *p*-coumaric acid, protocatechuic acid, and galangin in coconut sap. Fresh sap was reported to have less amount of these five phenolic compounds than fermented sap. Gallic acid was dominant phenolic compound in fresh sap, and its content was 350 µg/L, whereas caffeic acid content (730 µg/L) was higher in fermented sap.

Volatiles

Fresh palm sap contains a non-objectionable odor and it changed into harsh odor, unpalatable in spite of nutritious during fermentation. Acids and volatiles present in the sap are believed to cause the astringency and harsh odor of fermented sap. Due to the abundant presence of polar side-chain amino acids such as glutamine and asparagine and sucrose, volatile compounds are formed and more number of free amino acid groups are released during Maillard reaction in heating palm sap for sugar production (Sohn and Ho 1995). The major flavors such as ethyl lactate, ethyl lactate, 3-hydroxy-2-pentanone, farnesol, phenylethyl alcohol, 2-methyl tetrahydrofuran, and tetradecanone were found in fresh sap. But fermented sap contained 12 volatile compounds which represents more than 95% of the volatiles. Ethyl lactate, phenylethyl alcohol, and farnesol were among the seven compounds which are present in both fresh and fermented sap. The increased amounts of acids such as palmitoleic acid and dodecanoic acid (19.0 mg/L) and higher concentrations of ethyl alcohol and ethyl esters cause to astringency and harsh note to fermented sap (Borse et al. 2007).

Fermentation

Palm sap trickles from the cut end of the spadix in a drop by drop fashion. Hence, during a long collection period, the sap becomes deteriorated. Due to the sugar content (10–15%) and unhygienic open tapping practice, the sap undergoes fermentation and sugar is converted into alcohol (5–8%) (Iwuoha and Eke 1996). Instantaneous fermentation initially leads to alcoholic and acidic fermentation (Iwuoha and Eke 1996; Odunfa 1985). Rapid fermentation is observed under sunlight and other unhygienic environmental conditions. The fermented sap is known as "toddy" in India and Sri Lanka, "tuba" in the Philippines, and "tuak" in Indonesia and is a popular beverage (alcoholic drink) in most Southeastern Asian countries. Sources of the fermenting organisms such as the gourds, tapping implements, and air were reported (Odunfa 1985). Yeasts, particularly *Saccharomyces cerevisiae* in the initial stages dominates the fermentation process (Sanni 1993).

Chemical Changes

The chemical or microbiological compounds of fresh coconut inflorescence sap (FCIS) and naturally fermented coconut inflorescence sap (NCIS) have been studied by many researchers. About 21 compounds representing more than 98% of the volatiles in FCIS and 12 compounds representing more than 95% of the volatiles in NCIS were characterized during fermentation at 30 ± 2 °C for 24 h (Borse et al. 2007). Xia et al. (2011) studied changes on total sugar, ethanol, volatile acid, reducing sugar, amino acid, total phenolic contents, total acidity, and vitamin C of coconut inflorescence sap during fermentation. Increase in volatile acid, total acid, and total phenolic contents and decrease in total sugar contents were observed during natural fermentation. There was a reduction in amino acid for first three days, and then no significant change was recorded. The reduction in vitamin C content was observed on first day and increased to 20 mg/L on third day, and then apparent reduction was observed. NCIS exhibited more amounts of five types of phenolic compounds detected by HPLC than FCIS. Other types of phenolic compounds were also observed in both NCIS and FCIS.

Hebbar et al. (2015a) observed a good relationship between sugar content of sap and pH during fermentation (Figs. 2, 3). During ambient storage, fresh sap is subjected to lactic acid fermentation initially followed by alcoholic fermentation and finally acetic acid fermentation due to the action of microorganism. As the sap gets fermented, it

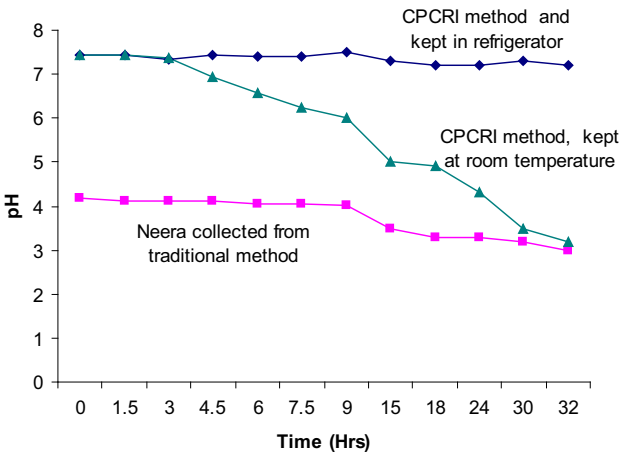

Fig. 2 pH of overnight sap collected by traditional method and CPCRI method

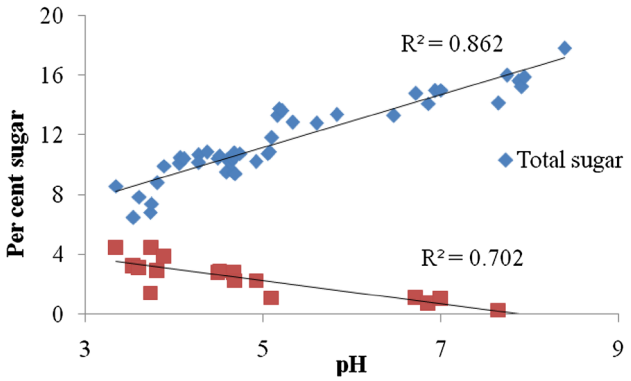

Fig. 3 Relation between the pH and total sugar and reducing sugar content of sap

becomes acidic and the pH reduces. The freshly collected sap starts fermenting within 2–3 h under ambient temperature and the pH starts declining. At the end of fermentation, pH of sap is normally about 3.5. During refrigerated storage at − 1 to − 3 °C, no significant change in pH was observed and the sap maintained its freshness. Freshly tapped sap of pH 7.5 contained 15% sugars and reduced to about 6% at pH 4 during fermentation (Fig. 3). Meanwhile, the increase in reducing sugar to the level of 5% was observed.

During fermentation, there was an initialization of sugar inversion. Initial 3 days of fermentation showed reduction in total sugar and increase in reducing sugar by conversion of sucrose. Then there was a decrease in reducing sugar content of sap due to its consumption by microorganism (Xia et al. 2011). Ethanol content was gradually increased to 90 g/kg during 7 days of fermentation, and then it reduced considerably (Atputharajah et al. 1986).

Microbial Changes

Various types of microorganism's particularly large number of aerobic mesophils were supported by palm sap due to its sugar content. During tapping, fermentation and storage of palm sap, lactic acid bacteria, yeasts, and acetic acid bacteria were predominantly found (Nwachukwu et al. 2006; Ogbulie et al. 2007).

Amoa-Awua et al. (2007) observed multiplication of yeasts dominated by *S. Cerevisiae* immediately after tapping, and there was a substantial increase in alcohol content after third day of fermentation. The presence of different types of microorganisms and quality changes during natural fermentation were investigated by Atputharajah et al. (1986). They isolated 166 isolates of yeasts, identified 39 isolates of bacteria, and reported 17 species of yeasts belonging to eight genera. Faparusi and Bassir (1971) recorded the active presence of *Lactobacillus* spp. and *Leuconostoc* spp. during the initial stages of fermentation. According to Okafor (1975), the domination of lactic acid bacteria *L. plantarum* formed important components of the bacterial populations, while *L. mesenteriodes* acidified the sap after first day of tapping. On third day, after the increase in alcohol concentration, population of Acetobacter and Gluconobacter species of acetic acid bacteria were increased. This increase was associated with increase in acetic acid concentration from 0.42 to 0.48% between third and fourth day and then increased to acceptable level of 0.6%. Two strains of *A. tropicalis* and *A. pasteurianus* with the ability to grow at temperature around 40 °C were isolated by Ndoye et al. (2006). The following flowchart explains the complete sequence of events during fermentation by the microorganisms in palm sap (Fig. 4). However, there was no consistent pattern in the species of microorganisms and it was found to vary with palm sap, storage, season, geographical location. Thus, while the prominent presence of lactic acid bacteria was found in palm sap, the simultaneous growth of other different genus was not observed in the sample (Okafor 1975). Sap sample was not having any single genus. For example, one sample had streptococci throughout the 7 days of incubation period along with the other sample lactobacilli which had persisted during the first 3 days of fermentation. Conversely, another sample had lactic acid bacteria *Leuconostoc* spp. after 2 days of incubation.

Similarly, on the first day of fermentation, palm sap had wine flora groups of *Streptococcus* species by other than lactic acid bacteria. *Serratia* and *Aerobacter (Klebsiella)* species also developed for short period and contributed to acid fermentation, and later they were suppressed by bacteria which are surviving under acid atmosphere. Development of *Acetobacter* was also observed during third day of fermentation. This may be after the formation of sufficient alcohol which assumes as substrate for the synthesis of acetic acid by *Acetobacter* (Amoa-Awua et al. 2007).

Role of Enzymes

During ambient storage and fermentation, the yeasts present in palm sap produced invertase enzyme which broken down the important sugar of palm sap, i.e., sucrose into monosaccharides such as glucose and fructose. These monosaccharides were subjected by glycolysis process and converted into pyruvate. This process was accompanied by a conversion of two ADP molecules into two ATP and water molecules due to the difference in size of two molecules, namely NAD^+ and NADH. With the help of an enzyme pyruvate decarboxylase and cofactor thiamine diphosphate, pyruvate was then converted to acetaldehyde

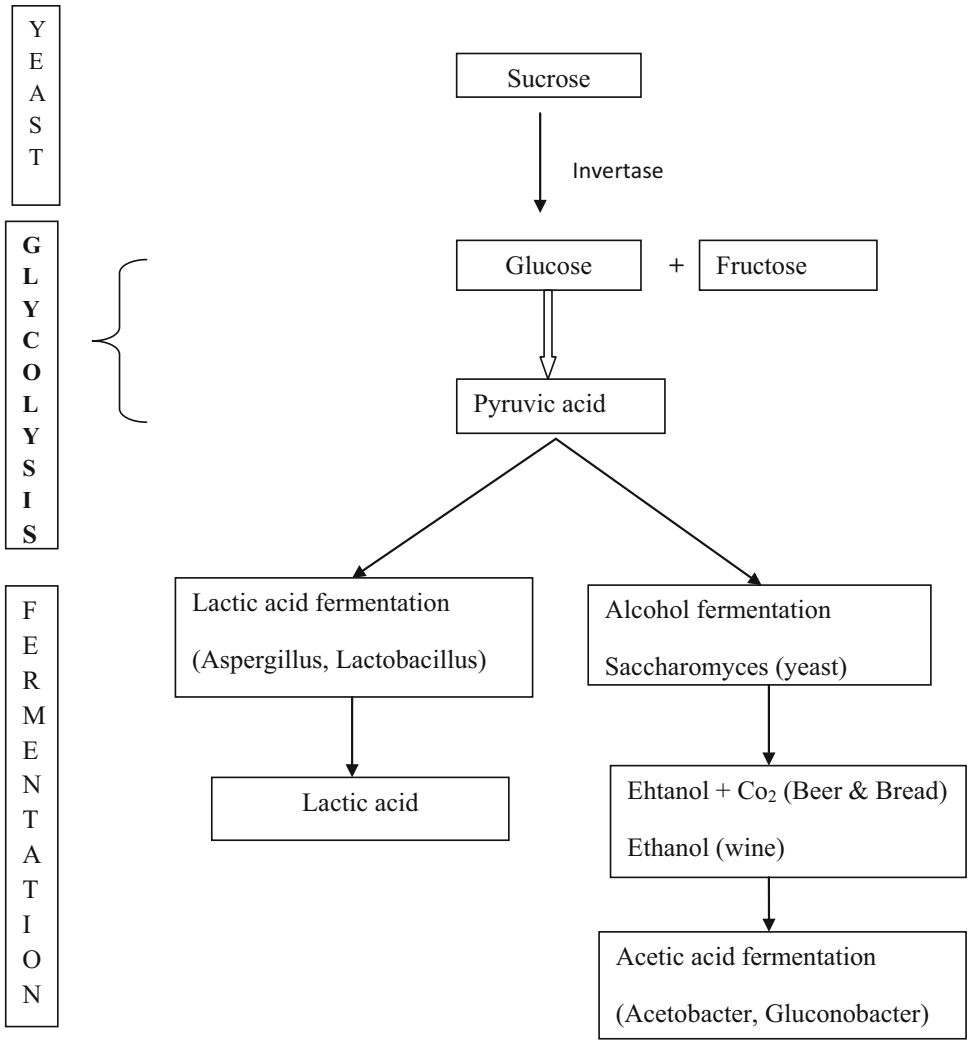

Fig. 4 Flowchart showing the microbial fermentation of palm sap into various products

and carbon dioxide. Then conversion of acetaldehyde into ethanol took place by the action of alcohol dehydrogenase (ADH) enzyme (Dioha et al. 2009). The whole conversion of monosaccharide into ethanol occurred through complex reaction process and catalyzed by collection of enzymes called zymase. The sequences of reactions are shown below. On the other hand, pyruvic acid from the glycolysis is converted to lactic acid either by pyruvate de carboxylase or in some cases by the enzyme lactic acid dehydrogenase.

$$C_{12}H_{22}O_{11} + H_2O \xrightarrow{\text{Invertase}} \underset{\text{(Glucose)}}{C_6H_{12}O_6} + \underset{\text{(Fructose)}}{C_6H_{12}O_6}$$

$$C_6H_{12}O_6 \xrightarrow{\text{Zymase}} \underset{\text{(Alcohol)}}{2C_2H_5OH} + 2CO_2$$

$$CH_3COCOO^- + H^+ \rightarrow CH_3CHO + CO_2$$

$$CH_3CHO + NADH \rightarrow C_2H_5OH + NAD^+$$

Disadvantages of Fermentation

Tiepma et al. (2013) and Ukhum et al. (2005) noticed nutrient degradation, loss of sensory qualities, and development of sour taste due to the production of acid during the fermentation process of palm sap which may be due to the action of microorganisms. It is also found that vitamins C, E, and B complex decreased in fermented sap (Hebbar et al. 2015b). Coconut sap is nutritious and aids in digestion (Devdas et al. 1969; Lata and Kamala 1966). Freshly tapped sap possessed tolerable odor and became unpalatable with harsh odor during fermentation process. This may be due to the product of palmitoleic acid, dodecanoic acid, ethyl alcohol, and ethyl esters during fermentation of palm sap. If the sap is fermented, the quality and quantity of sugar and other nutrients decline and become difficult to crystallize the sugar without the use of chemicals (Hebbar et al. 2015b).

Efforts to Improve the Shelf Life of Palm Sap

Many preservation techniques have been developed for enhancing the shelf life of palm sap. However, they were experimented on the sap collected by a traditional method which is unhygienic and partially fermented. Ogbulie et al. (2007) used an extract from the bark of different trees such as *Saccoglottis gabonensis*, *Vernonia amygdalina*, *Euphobia* sp., *Nauclea* sp., and *Rubiacae* species for the preservation of sap. In Nigeria, the water extract from the bark of *Sucoglottis gabonemis* was used to inhibit the yeasts and bacteria of palm sap. But Okafor (1975) reported that it only inhibited the growth of *Sarcina lutea* at 10% concentration and no other organism was affected by using this extract.

Thermal processes such as pasteurization and sterilization are also normally applied to extend the shelf life of palm sap. In India, palm sap was pasteurized by heating at 90–95 °C for 3–7 min, and in Thailand, pasteurization was normally done by heating at boiling temperature (60 or 70 °C) for 60 min (Naknean et al. 2015b). Table 5 shows the effect of pasteurization and preservatives on the shelf life of sap. Pasteurization of sap above 85 or 87 °C for 4–5 min imparted a cooked flavor (Baliga and Ivy 1961). It has been reported that conventional thermal processing of palm sap influenced the nutritional, color, and flavor characteristic of palm sap. The thermochemical reactions such as Maillard reaction, inversion reaction, and caramelization affected the quality of palm sap (Ho et al. 2007). The time and temperature factor needs to be optimized to decrease the thermal degradation of palm sap. Natural preservatives or antimicrobial agents could also be used as a part of hurdle technology so that heating time and temperature could be reduced in order to reduce the effect of thermal degradation (Potter and Hotchkiss 2012).

Concerted efforts have been made by CPCRI to improve the shelf life of *kalparasa* and thus help to transportation of distant places for marketing. Simple pasteurization of the

Table 5 Effect of different preservatives and pasteurization on shelf life of palm sap/wine

Sl. no.	Preservatives	Optimized treatment	Shelf life	References
1.	Sodium benzoate and citric acid	Pretreatment with 0.05% sodium benzoate and 0.15% citric acid and pasteurization at 170–175° F	–	Baliga and Ivy (1961)
2.	–	Pasteurization at 170–175° F	4–5 days under refrigeration temperature	Baliga and Ivy (1961)
3.	–	Heat sterilization at 80 °C for 25 min or at 90 °C for 20 min	6 months	Mohanadas (1974)
4.	Diethylpyrocarbonate (DEPC), sodium metabisulfite, and sorbic acid	Pasteurization at 70 °C for 30 min combined with sorbic acid (1%)	–	Okafor (1975)
5.	Potassium sorbate and sodium metabisulfate	Pasteurization (60 °C for 30 min) of supernatant of centrifuged palm wine with 0.05% sodium bisulfite.	6 months	Okafor (1977)
6.	Sodium metabisulfite (SM), sodium benzoate (SB) and propionate (P)	0.024 g/L sodium benzoate	3 months	Iwuagwu and Izuagbe (1985)
7.	Sodium benzoate and potassium sorbate	Sodium benzoate (0.08% w/v) with pasteurization (70 °C for 30 min	4 days stored at ambient temperature (27–33 °C)	Efiuvwevwere and Akoma (1997)
8.	*Saccoglottis gabonensis* (pulverized dust of the bitter bark tree)	Treatment with > 0.6% *S. gabonensis*	4 days stored at ambient temperature (27–29 °C)	Ojimelukwe (2000)
9.	Sulfur dioxide and dimethyldicarbonate	10 mg/L sulfur dioxide + 50 mg/L dimethyldicarbonate	60 days (stored at 20 °C)	Threlfall and Morris (2002)
10.	–	Pasteurized at 60 °C for 1 h	6 months	Dioha et al. (2009)
11.	Nisin	Pasteurization at 75 °C for 10 min with the addition of nisin (30 IU/mL)	10 weeks under low-temperature storage (4 °C)	Naknean (2013)
12.	Bentonite, polyvinylpolypyrrolidone (PVPP), gelatin, and chitosan	Addition of chitosan (0.1%), agitation for 1 h and pasteurization at 80 °C for 10 min	25 days (stored under 10 °C)	Naknean et al. (2014)
13.	Chitosan	Addition of 0.50 g/L chitosan, homogenization at 5 min and pasteurization at 80 °C for 10 min	6 weeks	Naknean et al. (2015)

unfermented sap in polypropylene bottles could extend the shelf life of *kalparasa* up to 45 days at 4–6 °C. The pasteurized and bottled sap maintained all the qualities of fresh *kalparasa* (Hebbar et al. 2015a). Pasteurization with zero additives was found to extend the shelf life of Raphia palm sap without much change in taste (Dioha et al. 2009). Sulfite, propionic acid, and benzoate were also used for preservation of palm sap (Levi and Oruchec 1957). Iwuagwu and Izuagbe (1985) could preserve the oyokpo beer using sodium benzoate or combinations of pasteurization and chemical preservatives. But this attempt resulted in change of taste due to the action of fermenting microbes.

Baliga and Ivy (1961) reported that 12–15 min (125° and 130 °F) pasteurization resulted in the reduction of microorganism. Most of the yeasts were killed in the sap by heating at 68° and 70 °C for 25–30 min. The pasteurization process had no significant effect on the concentration of riboflavin, thiamine, and ascorbic acid. Mohanadas (1974) described the method for preserving and bottling of coconut sap. He found that sterilization at 80 °C for 25 min and 90 °C for 20 min in Lanka Glass Co. bottles was most satisfactory for preservation and their characteristics were similar to the original coconut sap. The treated samples could be kept for 6 months without a change of flavor characteristics. He observed that a yellowish discoloration appeared which darkened to a brownish tint after 6 months of storage. Okafor (1975) produced a clear liquid without change in flavor by centrifuging palm sap-based beverages which was unacceptable by consumer due to the lack of characteristic white color. Suppression of non-ethanol producing microorganisms was observed in coconut inflorescence sap by the addition of 200 mg L^{-1} sodium metabisulfite (Samarajeewa et al. 1985). But Faparusi (1969) reported the failure of sulfite in preserving palm sap due to the presence of higher population of microorganisms and an unfavorable pH. Sodium metabisulfite (SMS) treatment contributed in the production of undesirable odor. Okafor (1977) reported the reduction of bacterial load by addition of 0.05% sodium metabisulfite before pasteurizing the supernatant from 200 to 5/mL and no reduction of bacteria load was observed by using 0.05% potassium sorbate.

Pasteurization of fruit juice resulted in 50% more reduction of bacterial population than control (Frazier 1967). Okafor (1975) studied the effect of preservatives (sorbic acid and sodium metabisulfite) and pasteurization on palm wine. He reported that palm sap could be more effectively preserved by pasteurization of palm sap at 70 °C for 30 min than preservatives in reducing the microbial population. He also suggested that combination of sorbic acid (1%) and pasteurization treatment may prove useful for preserving the palm sap. A sharp reduction of bacterial population to 2.63×10^4 cells/mL and 1.03×10^2 cells/mL from initial level of 1.06×10^8 cells/mL was observed using pasteurization along with propionate and sodium benzoate treatments, respectively, during one month storage of fresh beer (Iwuagwu and Izuagbe 1985).

Chitosan and nisin are recognized as safe preservatives for food (Naknean et al. 2015b). They can be applied as food additive in packaging material to retard the growth of microorganism in food (Leceta et al. 2013). Nisin proved to inhibit the growth of gram-positive bacteria and their spore forms (Sanlibaba et al. 2009). Naknean (2013) exhibited the application of nisin for extension of shelf life of pasteurized sap. He reported that pasteurization at 75 °C for 10 min along with 30 IU/mL nisin and low-temperature storage at 4 °C achieved 10 weeks of shelf life of sap as against 2 weeks shelf life for control sample. Concentration of nisin beyond 40 IU/mL proved unacceptable during sensory evaluation due to the development of sourness (Koiso 2010).

Naknaen and Meenune (2015a, b) treated the palm sap with different clarifying agents [gelatin, chitosan, bentonite, and polyvinylpolypyrrolidone (PVPP)] and then concentrated by open pan and vacuum evaporator to yield sugar syrup. They observed that syrups produced by open pan developed more browning and antioxidant activities. They suggested that combination of clarified palm sap and the vacuum evaporation could produce improved palm syrup with respect to clarity and browning reaction. Naknean et al. (2015b) investigated the influence of chitosan on pasteurized sap quality. It was observed that chitosan lowered the activity of PPO and invertase activity minimized the loss of sucrose along with increase in glucose and fructose content of sap during storage. Chitosan also ensured the increase in DPPH radial scavenging activity. Hence, the combination of pasteurization, chitosan (0.50 g/L), and low-temperature storage was recommended for 6-week shelf life of palm sap. Further addition of chitosan (1.00 g/L) may extend the shelf life of pasteurized palm sap which may end with more sedimentation, bitter taste, and consumer unacceptability.

Kapilan (2015) investigated with the different fermentation inhibitor (*Vateria copallifera*, *Careya arborea*, *Azadirachta indica,* and lime) for the preservation of coconut sap. They observed that lime at optimized concentration of 3 g L^{-1} sap proved to inhibit fermentation of sap more effectively. They also found the presence of significant lower concentration of alcohol due to the addition of lime in the sap. A membrane technique developed by National Chemical Laboratory (NCL), Pune, claims to extend the shelf life of 45 days under refrigerated storage condition (4–8 °C) by removing the microorganism present in palm sap without compromise in nutritional quality.

Integrated Approach

Defence Food Research Laboratory (DFRL) Mysore, Central Food Technological Research Institute (CFTRI) Mysore, Defence Research and Development Organisation (DRDO), Kerala Agricultural University (KAU), Coconut Development Board (CDB), and others made concerted efforts to improve the shelf life and palatability of the sap collected by traditional methods. Different techniques like filtration, treatment with clarifying agents and deodorizing using activated carbon/bentonite, centrifugation, pasteurization at 95 °C, the addition of preservatives, carbonation were employed before bottling of sap. All the mentioned techniques yielded a pasteurized sap with change in taste and had not been able to retard the growth of fermenting microbial population. Ozone technology, pulsed light, pulsed electric field, and high-pressure processing have a broad antimicrobial property (Pandiselvam et al. 2017) that may meet the requirements of palm sap processing industries.

Conclusions and Future Directions

This review paper discussed various techniques for the improvement in quality and safety aspects of palm sap without affecting its natural flavor. Nevertheless, the main aims of fermentation chemistry are the identification of potent microbes responsible for pleasant as well as the off-flavors. Such scientific knowledge is required for the quality and safety of palm juice, through technological processes. The quality of palm sap depends on the place and tapping duration poses a preservation problem leading to oyster white color, harsh flavor, and taste, due to the fermentable nature of the sap. This necessitated for the development of processing of palm sap for its shelf life improvement. The major challenge of collecting hygienic and unfermented sap from the palms has been to a certain extent resolved with the development of coco-sap chiller technology at ICAR-Central Plantation Crops Research Institute (CPCRI). The sap thus collected is very healthy and nutritious, and hence, it can be directly sold as fresh juice under the refrigerated condition in the local market or converted into different diversified products such as coconut sugar, jaggery, nectar, or syrup without the addition of any chemicals/preservatives/additives.

The future directions of studies including flavor profile changes of *kalparasa* during honey/sugar production and comparison of quality profile of honey/sugar produced from *kalparasa* by different processing methods (open evaporation and vacuum evaporation) could be useful to produce quality jaggery products. The experience suggests that tapping the sap and selling is eight- to tenfold more profitable than selling matured nuts. However, the shelf life of the sap for long-distance transport is a major issue. Concerted efforts are needed to improve the shelf life without affecting the natural aroma and taste of the sap. A combination of heat and chemical preservatives and low-temperature storage affect the nutritional and sensory qualities of sap which may also be uneconomical. The food industries need to fulfill the consumers demand for convenient, fresh, healthy products without any synthetic preservatives. As an alternative, non-thermal processing methods such as ozonation, high-pressure processing, pulsed electric field, cold plasma technique, UV light treatment, and pulsed light could be used for the preservation of palm sap. Use of non-thermal and minimum use of chemical preservatives for microbial control in palm sap may result into enhanced accessibility to reach the customers of non-traditional areas and attain its commercial potential.

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Conflict of interest The authors declare that they have no conflict of interest.

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