Potential Application of Cashew Apple (Anacardium Occidentale L.) as a Probiotic Beverage Fermented with Lactobacillus

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ABSTRACT
Cashew nut is a tremendous income generating plantation crop. It is popularly called as Dollar earning crop. There has been substantial area expansion of the crop with commercial exploitation of the kernel, however a paramount quantity of the crop is wasted on account of poor awareness and technical knowhow on its utilization pattern. Application of cashew apple into a Probiotic beverage is a perfect example of converting waste to wealth. Probiotic foods are the fastest growing sectors of functional food production. The increased health consciousness among consumers have contributed to rapid market expansion of probiotic beverages. Probiotics are mostly prepared from dairy-based products, however the problem of lactose intolerance and milk protein allergy among many consumers have created a surge for non-dairy substitutes for pro-biotic formulations for which cashew apple serves as a suitable choice. Cashew apple provide immense nutritive and health benefiting attributes. Potential utilization of this agricultural waste into a nutritive drink can provide tremendous scope to add new dimensions to income earning pattern of cashew growers and the industries engaged in cashew processing. Such initiatives which establishes the efficient utilization of existing underutilized resources into value-added functional products can play significant role in sustainable development of Indian agriculture.
INTRODUCTION

Cashew nut (*Anacardium occidentale* L.) is a popular plantation crop. The crop was first introduced from Brazil in Goa and thereafter it has spread all over the country and received wide popularity for its economic value. India is the first country to commercially utilize the crop for its extremely nutritious kernels. The Commercial cultivation of cashew began in the early 1960s and gradually, over the years, it became a crop with immense economic significance and acquired the status of an export-oriented commodity, earning substantial foreign exchange for the country. Presently, India is the largest processor, exporter, importer and consumer of cashew nuts. The crop has been commonly referred to as a wasteland crop and mostly grown in a larger scale in degraded soils to prevent erosion. The crop is very popular among the farmers on account of its profitability. The crop performs well under hot humid conditions and bear profusely with abundant nuts. The nut is the economic part, which has a profound preference amongst the Indian consumers and also tremendous export potential. The export of cashew kernel is mediated by Cashew Export Promotion Council of India (CEPCI) who works to promote the export of cashew kernels and cashew nut shell liquid (CNSL). However, a larger part of the fruit i.e cashew apple, the apical part is often left unutilized and is considered as an ‘agricultural waste’. It has been observed that the weight of the unused cashew apple is about 10 times of the harvested nuts. The fruit of cashew or cashew apple is consumed fresh, but it often tastes astringent due to high content of tannins. It is usually processed into several fermented and non-fermented beverages. Among the fermented beverages, *feni* or fenny is a popular alcoholic beverage, extensively prepared and consumed in India. Fenny contains about 40-42 % alcohol.

The utilization of cashew apple as a potent agricultural waste to develop some fortified drink has now attracted attention. As a result, probiotic beverage, prepared from cashew apple juice fermented with *Lactobacillus* is an excellent alternative to utilize the potent waste emerging from the crop. Thus, diverting it into a functional drink fortified with microorganism which prove beneficial for human health. The term functional food was first used and evolved in Japan during the 1980s, which refers to a food designed to be medically beneficial to the consumers. Functional food is thus defined as food or dietary components that deliver a health benefit beyond the basic purpose of providing nutrients. Probiotic foods are the fastest growing sectors of functional food production. Several food products viz., dairy, meats, beverages, cereals, vegetables, and fruit have been utilized as conveyance for probiotics production. The development of probiotic drink from cashew apple proves to be promising to diversify the market and add employment opportunities for several farm women & rural youths. However, production of health drinks from cashew apple appears to be challenging as it quite astringent due to high content of tannin and growth of the bacteria *Lactobacillus* sps is again a crucial aspect as the bacterial population requires sufficient amount of sucrose to sustain its growth and load. The *pH* management is another critical factor for successful growth of bacteria along with standardization of working temperature for optimum bacterial load i.e CFU (colonies formed per unit). The increased health consciousness among consumers have contributed to rapid market expansion of probiotic beverages. Cashew apple appears to be as efficient as dairy products for *Lactobacillus casei* growth and hence, Excellent variation in *Cashew* apple and kernel characteristics of some popular varieties under Odisha condition

![Figure 1: Variation in Cashew apple and kernel characteristics of some popular varieties under Odisha condition](source.png)
serves as an excellent medium for probiotic drink preparation.

**Nutritive value of Cashew apple:**

Cashew apple is a pseudo fruit. It is highly juicy in nature, enriched with numerous nutritive values. It is rich in sugar, amino acids, tannin, vitamin C and crude fibre. The ascorbic acid content (Vitamin C) is 240 mg/100g, which is about six folds higher than citrus fruits (40 mg/100 g). Cashew apple is also abundant in free soluble sugar, mostly reducing sugars (glucose, galactose and fructose). It is also rich in dietary fibre and crude fibre (15 to 18 %, on dry weight basis). It also serves as an excellent source of anti-oxidants viz., phenols, tannin and flavonols. These natural antioxidants play a major role in destroying free radicals.

Consumption of cashew apple could assist in overcoming the Vitamin C deficiency and also constipation.

**Table 1: Bio-chemical constituents in Cashew apple:**

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Constituents</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Moisture</td>
<td>86.10 g/100g</td>
</tr>
<tr>
<td>2</td>
<td>Protein</td>
<td>0.80 g/100g</td>
</tr>
<tr>
<td>3</td>
<td>Fat</td>
<td>0.20 g/100g</td>
</tr>
<tr>
<td>4</td>
<td>Carbohydrate</td>
<td>12.60 g/100g</td>
</tr>
<tr>
<td>5</td>
<td>Fibre</td>
<td>0.60 g/100g</td>
</tr>
<tr>
<td>6</td>
<td>Ash</td>
<td>0.30 g/100g</td>
</tr>
<tr>
<td>7</td>
<td>Calcium</td>
<td>0.20 mg/100g</td>
</tr>
<tr>
<td>8</td>
<td>Phosphorus</td>
<td>19.00 mg/100g</td>
</tr>
<tr>
<td>9</td>
<td>Iron</td>
<td>0.40 mg/100g</td>
</tr>
<tr>
<td>10</td>
<td>Vitamin B1</td>
<td>0.20 mg/100g</td>
</tr>
<tr>
<td>11</td>
<td>Vitamin B2</td>
<td>0.20 mg/100g</td>
</tr>
<tr>
<td>12</td>
<td>Niacin</td>
<td>0.50 mg/100g</td>
</tr>
<tr>
<td>13</td>
<td>Vitamin C</td>
<td>200.00 mg/100g</td>
</tr>
</tbody>
</table>

(Source: NRC for Cashew, ICAR)

**Processed products from Cashew apple:**

Cashew apple has immense potential to be utilized either for fresh consumption, processed items, fermented beverages, industrial application and medicinal uses. It is highly juicy and also preferred for fresh consumption. In many areas it is also preferred as a popular curry in many Indian households. Cashew apples have the potential to be processed into various value-added products viz., juice, syrup, jam, ice cream, candy, chutney, pickle, and other products. It can also be fermented into different products, such as alcoholic beverages like fenny, wine, bioethanol, enzymes, biosurfactants, probiotic beverages, lactic acid and oligosaccharides.

It also possesses immense health promoting and medicinal Uses. Cashew apple juice, without removal of tannin, is usually prescribed as a remedy for sore throat and chronic dysentery. Fresh or distilled juice also serve as a potent diuretic and is reported to possess sudorific properties. The brandy prepared from it is applied as an ointment to relieve the pain of rheumatism and neuralgia. Cashew apple is reported to contain high levels of lutein and zeaxanthin, which act as antioxidants and can protect the eyes from light damage when consumed regularly. These compounds can even reduce the instances of cataract. The apple possesses anti-bacterial properties and has been beneficial in treating stomach ulcers and gastritis. The proteins in cashew apple helps in rejuvenating skin and hair. Traditionally the juice is used as a potent ingredient in scalp and hair treatment. The presence of negligible amount of sugar and absence of cholesterol makes them safe for diabetic patients. It even helps in lowering the risk of type 2 diabetes. The abundance of anti-oxidants helps in combating premature aging. According to a study reported in 2013, published in the Journal of the International Society of Sports Nutrition, men who drank cashew apple juice during high intensity exercise had enhanced fat utilisation.

Cashew apple juice also exhibits moderate anti-tumour and anti-inflammatory properties and is effective against breast cancer and cervical cancer. Antioxidants like anacardic acids, cardanols and cardols in cashews make them effective for people undergoing treatments for tumours and cancer. The juice
has tremendous industrial uses, cashew juice is extensively utilized in the cosmetic industry and is used in preparation of various creams, shampoos and conditioners. In some places, cashew apple seeds are used in preparation of snake bite venom.

Probiotic beverage from cashew apple juice:

Probiotics are classified as live microorganisms which confer a health benefit to host when it is administered in sufficient amounts (FAO/WHO, 2001). These products have enhanced the stability of microbiota in the human body and give rise to physiological benefits. In recent years, probiotic products have acquired extensive acceptance in the entire world and are predominantly successful in United States, Europe, Japan and Australia. In market, mostly dairy-based products are the most important products within the probiotic products and account for around 74% of probiotic product market shares. However, the problem of lactose intolerance and milk protein allergy among many consumers have created a surge for non-dairy substitutes for pro-biotic formulations. In order to combat the cholesterol problems and to lower the levels of saturated fatty acids, which is the major problem with traditional milk-based pro-biotics, focus is now driven in formulating fruit & vegetable juices, cereal based pro-biotics. The most common bacteria used in pro-biotic preparation are *Lactobacillus acidophilus* and *bifidobacterium* species. Probiotics presently available in the market contain *Lactobacilli*, *Streptococci*, *Enterococci* or *Bifidobacteria*. The production of probiotics from cashew apple involves the following steps.

**Fig 2: Utilization of Cashew Apple**

**Probiotic beverage from cashew apple juice:**

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**Fig 3: Flowchart of Probiotic cashew apple beverage preparation**

* SMS-Sodium Metabisulphite
1. Cashew apple collection and juice extraction: Cashew apple collected from cashew plantation should be clean, disease and dirt free. After collection, it is pre-cooled and washed thoroughly in water to cleanse its surface from adhering dirt and inert material. The cashew apple can also be initially decontaminated by washing in 100 ppm chlorine water before juice extraction. The juice extraction is carried out with the help of a clean pulper/juicer. The juice from cashew apple is extracted aseptically by straining to get clarified juice, after which it is subjected to pre-fermentation treatment.

2. Pre-fermentation treatment: Pre-fermentation treatment include removal of tannins from the fruit juice. In general, the cashew apple contains about 0.6 mg tannins/100 g juice. Tannins are classified as a group of phenolic compounds. It usually has a strong tendency to form complexes with proteins and other macromolecules. The astringency due to presence of tannins is due to the fact that it forms complexes with salivary protein and glycoprotein. Even the ingested tannin could prevent action of digestive enzymes and consequently affect the utilization of nutrients. The removal of tannins from cashew apples is carried out by adding proteins (viz., gelatin) or starch (viz., starch, rice gruel, cassava, sago), after which it is subjected to filtration and siphoning. Out of several tannin precipitating agents, gelatin has been reported to be most effective and is most commonly used. It is usually used in the range of 0.3 to 1.0% w/v. After the gelatin dissolution, the juice was allowed to stand at 4 °c for precipitation of the tannins and suspended solids, which were thereafter removed by filtration. In order to increase the extraction yield and clarification of fruit juice, pectinase enzyme is also added to the fruit juice. Pectinase is a group of enzymes comprising of pectin lyase, pectin esterase and polygalacturonase. pH adjustment is another critical factor and is adjusted according to the microbial strain used for fermentation. The Ph is adjusted to the optimum pH for the growth of the microorganisms in the clarified and pre-treated fruit juice. The optimum initial pH for probiotic cashew apple juice production is 6.4

3. Preparation of Lactobacillus inoculum, its culture and inoculation in cashew apple juice: The inoculums are prepared by transferring a glycerol stock culture tube containing Lactobacillus casei to a 250 ml Erlenmeyer flask containing 100 ml of sterile MRS broth. MRS Broth is a medium for the cultivation and enumeration of Lactobacillus spp. This product has the same formulation as LAB093 MRS Agar with the omission of agar. It was originally developed by de Man, Rogosa & Sharpe in 1960, the medium can be used for confirmatory tests on organisms isolated on MRS Agar. The bacterial cell cultivation is carried out in an incubator set at a temperature of 37 °c. The cell density is spectrophotometrically determined at 590 nm. The incubation period is extended until the cell density reaches 0.600 which corresponds to 9.00 Log CFU/ml of L. casei. Cashew apple juice (100 ml of samples) are inoculated with 7.00-7.48 Log CFU/mL of L. casei, which corresponds to adding 1-3 ml of activated inoculums prepared. This concentration was chosen on the basis of the recommendations for probiotic foods, which states that the minimal counts of 7.00 Log CFU/mL is highly essential for improved efficacy in regulating beneficial effects. This culture was used as an inoculum and added to the cashew apple juice followed by fermentation.

4. Fermentation and frequent monitoring: The optimum fermentation conditions is highly essential for successful bacterial growth. It has been reported in many studies that the initial pH does not significantly affect the biomass. The increase in temperature causes an increase...
in biomass of *L. casei* followed by a decrease at temperature above 35⁰ c. Temperatures higher than 35⁰ c also causes loss of viability of the inoculated bacterial cells. Hence, the optimum temperature for *L. casei* growth in cashew apple juice is around 35⁰ c. Optimum pH is also another crucial factor for successful growth and viability of bacterial cell. The highest (optimum) microbial viability was obtained at pH 6.4 and fermentation temperature of 30⁰ c. The Erlenmeyers flasks containing 100 ml of clarified cashew apple juice inoculated with 7.00 - 7.48 log CFU/mL (1-3 ml of MRS broth containing 9.00 CFU/ml) are then subjected to fermentation under optimum conditions in an incubator. The optimum fermentation time is 16 h. Towards the end of the process, the biomass, viable counts and pH are analyzed. The fermented cashew apple juice containing probiotics is then dispensed into sterile Erlenmeyers flasks closed with cotton plugs and stored at 4°c. The physico-chemical and biological properties viz., biomass, viable counts, colour and pH of cashew apple juice are recorded prior to cold storage and at an interval of 7 days, during 42 days shelf life period for constant monitoring of the product efficacy and quality.

Fermented cashew apple juice can serve as an inexpensive source of beneficial probiotics. There has been a sincere attention in the development of fruit-juice based functional beverages fortified with probiotics because fruit juices have excellent flavour, taste & aroma and are preferred by all age groups. Fruit juices are always refreshing and healthy. There is a target to serve a functional drink which is rich in vitamins, minerals, anti-oxidants along with beneficial microorganism, hence fruit-based probiotics have tremendous scope in near future. It has been reported that fruit juices have been recommended as an ideal media for probiotic growth because they intrinsically contain essential nutrients, look attractive and have excellent taste. However, the stability and ultimate survival of probiotics in fruit-based medium is more complex than in dairy products, because the bacteria need protection from the acidic conditions in these media.

**CONCLUSION:**

Probiotic beverage from cashew apple juice is an excellent alternative to dairy-based probiotics. It is a potent functional drink and the best example of adding value to waste. The scope of utilization of this agricultural waste into a nutritious drink has tremendous potential to add new dimensions to income earning pattern to cashew growers and to industries engaged in cashew processing. The farmers growing cashew are now realizing the commercial significance of cashew apples. Production of the probiotic juice will be an important source of supplementary income in cashew plantations. However, in a developing country like India, cashew apple is still not exploited to its full potential. Farmers’ awareness in this aspect, followed with optimum quality research with aid from institutional policies and Govt. schemes & subsidies can enable in popularizing the technology for mass adoption by the farming community. To sustain the agricultural productivity, there is a need for building agro-based industries which can effectively utilize farm waste and generate healthier and more prosperous livelihood options for the farming community. To strengthen the cashew apple value chain there is a need to strengthen the post-harvest infrastructure facilities at village level along with capacity building of the farmers. Beverages prepared from cashew apple preferably fenny is quite popular among consumers but the healthy functional drink, ‘Probiotic Cashew beverage’ is positioning as an emerging arena for cashew growers, entrepreneurs, manufacturers, dealers and distributors in various wholesale and retail markets. The consequences for value addition of cashew apple has the potential to mitigate nutritional insecurity, health security, livelihood sustainability, employment
opportunities for landless farmers and hence can bring prosperity and inclusive development in cashew producing states of India. Such initiatives which establishes the efficient utilization of existing underutilized resources into value added functional products can play immense role in sustainable development of agriculture in a developing country like India.

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