



## Studies on Processing and Storage of Tender Coconut Water

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### ABSTRACT

Tender coconut water (*Cocos nucifera* L.) is a delicate and refreshing natural beverage whose original properties have drawn to highlight as a natural functional drink. Thermal treatments combined with chemical additives are used by the industry but other technologies such as micro and ultrafiltration are yet to be used on an industrial scale. MF and UF offer excellent potential in food industry for clarification and pasteurization of liquid foods to replace conventional processing techniques. A continuous cross flow flat sheet membrane module was used to process by microfiltration (MF) and ultrafiltration (UF) technology. In the first treatment, the tender coconut water (TCW) was passed through MF of 0.2  $\mu\text{m}$  pore size at a pressure of 5.06  $\text{kg}/\text{cm}^2$  to remove microbes and suspended particles. In the second treatment, TCW was passed through UF of molecular weight cut off (MWCO) 40 kDa at pressures about 5.06  $\text{kg}/\text{cm}^2$  to remove enzymes such as polyphenoloxidase (PPO) and peroxidase (POD). In the third treatment, TCW was bottled and pasteurized at 85°C for 10 min in a water bath. In the fourth treatment, TCW was filtered through a MF membrane and chemical preservative nisin was added to coconut water in two concentrations of 5000 IU and 2500 IU. The TCW filtered through muslin cloth was bottled and refrigerated as control sample. All the samples using different treatments were bottled using crown corking machine and refrigerated at 4 °C to assess the quality during storage. The samples were taken out for every four days interval to assess physico-chemical, microbiological and sensory characteristics upto 20 days of storage. Finally among all the treatments, pasteurized, microfiltered and ultrafiltered treatments gave quality bottled TCW.

Key words: *Membrane processing, Microfiltration, Ultrafiltration.*

Coconut water (*Cocos nucifera* L.) is a tropical beverage whose original properties have drawn the attention of processors as a natural functional drink (Alexia *et al.*, 2011). The tender coconut water (TCW) technically the liquid endosperm, is the most nutritious wholesome beverage that the nature has provided for the people of the tropics. TCW is rich in essential minerals such as potassium, sodium and natural nutrients like polyphenols (Yong *et al.*, 2009). This refreshing drink is filled with many healthy natural nutrients which can enhance the body's metabolism and immunity and is used more as a health supplement. The important significant and useful components in coconut water are cytokinins.

Currently, pasteurization is the main technique used for preservation and microbiological safety. Some studies have been conducted to evaluate the effect of pasteurization on the inactivation of oxidative enzymes, peroxidase (POD) and polyphenoloxidase (PPO) in coconut

water. But pasteurization affects some of the heat sensitive aroma and other sensory attributes. Therefore, it is important to establish a preservation process that does not cause a negative effect on the composition and sensory characteristics of coconut water (Nakano *et al.*, 2012). Preservation by concentration to reduce water activity is one of the important food processing techniques. However, concentration using thermal processing is one of the most energy intensive processes among unit operations in the food industry. So, membrane separation processes such as Microfiltration (MF), Ultrafiltration (UF), Nanofiltration (NF) and Reverse Osmosis (RO) are promising novel alternative non-thermal and non-chemical methods that are relatively less energy intensive and retain heat labile components.

In the present study, the objective is to develop process technology for preservation TCW using membrane processing, pasteurization and chemical additive to produce a shelf stable bottled TCW.

## MATERIAL AND METHODS

### Raw materials

The tender coconuts were obtained from a local market Bapatla town, Guntur dist, Andhra Pradesh. Nisin (Bacteriocin), chemical preservative was obtained from Sigma Aldrich, USA. The coconut variety used locally for retail sale of tender coconut water was used as raw material. The nuts were procured from Eluru region (East coast tall) of Andhra Pradesh. Tender coconuts with good quality and without any pest or disease infestation were selected. The water is extracted fresh just before use for experiment.

### Membranes

For microfiltration, a 0.2  $\mu\text{m}$  membrane of poly acrylo nitrile has been used. Ultrafiltration was conducted using 40 kDa MWCO membrane of Polysulfone has been used. These membranes were supplied by IIT, Kharagpur.

### Experimental set up

Experimental setup is available at College of Food Science and Technology, Bapatla, Acharya NG Ranga Agricultural University, Guntur.. The cross flow cell membrane module setup developed by Prof. Sirshendu De, Department of Chemical Engineering, IIT, Kharagpur was used for filtration of tender coconut water. A water bath was used to pasteurization technique.

### Cross flow set up

Cross flow cell membrane set up made of stainless steel was used for membrane processing of coconut water. Two neoprene rubber gaskets were placed over the membrane forming the flow channel. The channel height after tightening the two flanges was found to be  $3.0 \times 10^{-3}$  m. The effective dimension of the membrane was 14.5 cm in length and 5.5 cm in width. The cell consisted of two rectangular matching flanges. The inner surface of the top flange was mirror polished. The bottom flange was grooved, forming the channels for the permeate flow. A porous stainless steel plate was placed on the lower flange that provides mechanical support to the membrane. Two flanges were tightened to create a leak proof channel for conducting experiments in cross flow-mode.

The feed was pumped by a high pressure reciprocating pump from the stainless steel feed

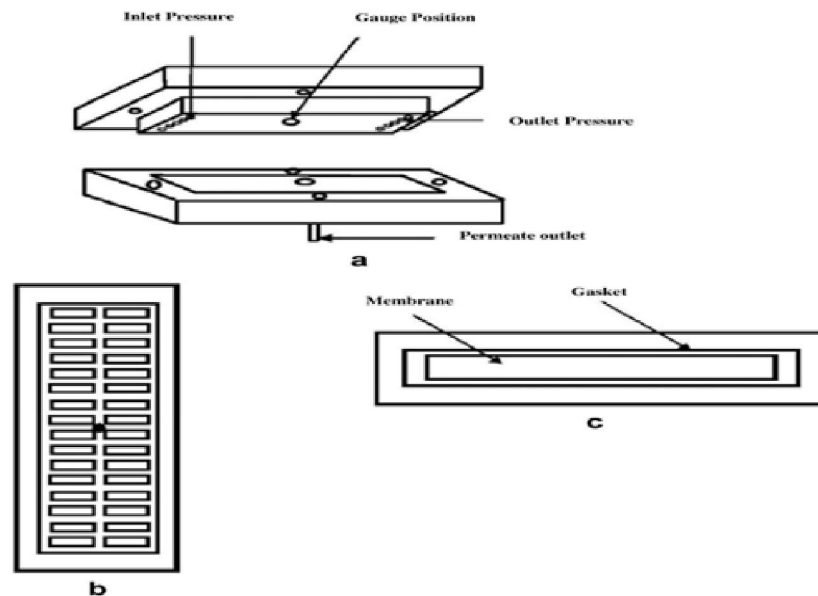
tank to the cross flow cell (Fig.1). The retentate stream was recycled to the feed tank routed through a rotameter. The pressure and the cross flow rate inside the membrane channel were independently set by operating the valves in the bypass line and that at the outlet of the membrane cell. Permeate samples were collected from the bottom of the cell and were analyzed for physico-chemical and microbial tests.

### Methods

Five different treatments were investigated to develop process technology and extend shelf life during storage of TCW. In the first treatment, the coconut water was passed through a microfiltration membrane of 0.2  $\mu\text{m}$  pore size at a pressure of 5.06 kg/cm<sup>2</sup> to remove microbes and suspended particles. In the second treatment, coconut water was passed through ultrafiltration membranes of 40 kDa MWCO at a pressure of 5.06 kg/cm<sup>2</sup> to remove enzymes such as polyphenoloxidase (PPO) and peroxidase (POD). In the third treatment, the coconut water was bottled and pasteurized at 85 °C for 10 min. In the fourth and fifth treatments, the coconut water was filtered through a MF membrane and chemical preservative nisin was added in two concentrations of 5000 I.U and 2500 I.U. The TCW filtered through muslin cloth was taken as control sample. The control as well as all the treated samples were bottled and stored at 4 °C. The samples were taken out at every four days interval and their physico-chemical, microbiological and sensory characteristics were determined upto 20 days of storage.

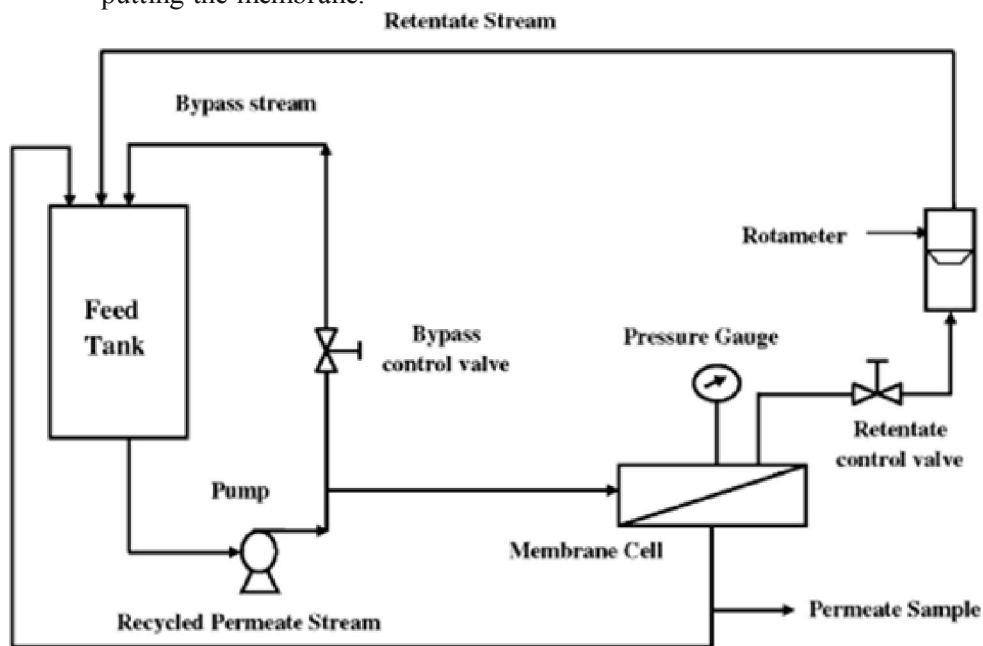
### Operation of membrane plant for conducting experiments

A fresh membrane was compacted at a pressure higher than the maximum operating pressure for 3 h using distilled water and then its permeability was measured. The raw material was placed in a feed tank of 5 L capacity. A high pressure reciprocating pump was used to feed the TCW into the cross-flow membrane cell. A bypass line was provided from the pump delivery to the feed tank. Retentate and bypass control valves were used to vary the pressure and flow rate accordingly. The permeate stream after collecting required amount of sample was recycled to the feed tank to maintain a constant concentration in the feed tank under total



**Fig.1** Various parts of the membrane cell.

(a) Two flanges, (b) the top view of grooved bottom flange, (c) top view of bottom flange after putting the membrane.



**Fig.2.** Schematic diagram of cross flow experimental set up.

recycle mode. The recirculation flowrate of TCW was kept constant at 100 lph. The corresponding cross flow velocity of the retentate is 1.68 m/s. Once an experimental run was over, the membrane was thoroughly washed, in situ, with distilled water for 30 min applying a maximum pressure of 30 psi. The cell was dismantled and the membrane was rinsed with distilled water and was dipped in 2% sodium dodecyl sulphate solution overnight. On the next day, the

membrane was washed carefully with distilled water to remove traces of surfactant. After that, the set up was ready for the next experiment product. All the experiments were conducted at a room temperature of  $33 \pm 2^\circ\text{C}$ . All the experiments were triplicated and averages were presented. The holdup volume in the plant is 1.5 L. Minimum volume of feed required to conduct experiments is 4 L. The schematic diagram of cross flow experimental setup is given in Fig. 2.

### Physico-chemical parameters and Sensory

Samples of fresh, filtered tender coconut water coming from MF and UF experiments were collected and stored at refrigeration condition at 4 °C. Total soluble solids (TSS) were determined by Pocket Refractometer (ATAGO make, range 0-93%). pH was measured by a digital pH meter (Systronics  $\mu$  pH system, Model 362). Turbidity was measured by a Systronics Spectrophotometer 166 at transmittance of 610 nm using a transparent cuvette. Reducing sugars when heated with alkaline copper tartrate, the copper converts from cupric to cuprous state and thus cuprous oxide was formed, when cuprous oxide is treated with arsenomolybdic acid, the reduction of molybdic acid to molybdic blue takes place. The blue colour absorbs maximally at 540 nm (Sadashivam and Manikam, 1992). Sensory evaluation of treatments was carried out by 10 untrained panelists selected at random. Appearance, flavour, overall acceptability of the samples were rated using a 9 point Hedonic scale where 9 and 1 represent like extremely and dislike extremely respectively. Sensory evaluation was carried out at ambient conditions in a comfortable and quiet area without disturbance under fluorescent lighting. Water was supplied to cleanse palate between samples.

### Microbial load

The presence of microorganisms viz bacteria, fungi and E.coli were determined by performing standard dilution technique using suitable media (Fernanda *et al.*, 2009).

## RESULTS AND DISCUSSION

### Total soluble solids (TSS):

Total Soluble Solids (TSS) were found to be in the range of 4.95 to 5.29 % initially, then the values gradually decreased with time in all treatments. There was no significant change observed in TSS of treatment T<sub>3</sub>. Similar observation was made by Chowdhury *et al.* (2009) for heat treated coconut water samples at 85°C for 10 minutes. Both microfiltered and ultrafiltered samples were observed low TSS perhaps due to removal of some suspended particles from the tender coconut water. It was observed that TSS generally decreased on storage for all the treatments (Fig.3).

The tender coconut water comprises of sugars, minerals and other nutrients. The initial TSS of the treatments T<sub>1</sub>, & T<sub>2</sub> were found to be slightly

low because some of the complex sugars and cloud forming solids may have been retained in filtration because of their higher molecular size. TSS in all treatments decreased during storage due to fermentation process. Similar observation was made by Rosa *et al.* (2012). Treatment T<sub>3</sub> exhibited less change in TSS because of resistance to fermentation process due to thermal treatment (Chowdhury *et al.*, 2009). Among all treatments, the TSS of untreated TCW reduced more due to the conversion of carbohydrates into sugars, organic acids and other soluble materials by metabolic processes during storage (Manashi *et al.* 2012).

### pH:

The pH of tender coconut water decreased during storage (Fig. 4). The pH in treatments T<sub>1</sub> and T<sub>2</sub> decreased from 4.864 to 4.562 and 4.951 to 4.64 respectively. The pH was observed high initially after four days of storage in the treatments because of removal of most of the colloidal particles which might cause acidity in TCW and then the pH decreased during storage probably due to fermentation process as it was completely non-thermal process. The pH of treatment T<sub>6</sub> decreased from 4.921 to 4.25. The decrease in pH in control treatment may be due to microbial growth. It was very low on 20<sup>th</sup> day storage as compared to all other samples. The pH of treatment T<sub>3</sub> decreased relatively less from 4.81 to 4.563 perhaps due to thermal treatment by which fermentation process might have been delayed. The pH of all treatments decreased during storage which could be due to the production of free acids by microbial growth or by polysaccharides (Manashi *et al.* 2012).

### Reducing Sugars

It was observed from the Fig.5, the reducing sugars increased with increasing storage period. The reducing sugars for treatment T<sub>6</sub> were observed from 3.2 to 3.58 % during storage. The reducing sugars were found low for treatment T<sub>3</sub> i.e., 3.1 % and increased to 3.254 % upon storage. The values of reducing sugars for all other treatments T<sub>1</sub>, T<sub>2</sub>, T<sub>4</sub> and T<sub>5</sub>, increased from 3.2 to 3.43%, 3.1 to 3.29%, 3.25 to 3.39% and 3.2 to 3.38%, respectively. The reducing sugars found to be increasing in all the treatment including control because of the breakdown of total sugars into reducing sugars. The reducing sugars might have also increased because of the hydrolysis of non-

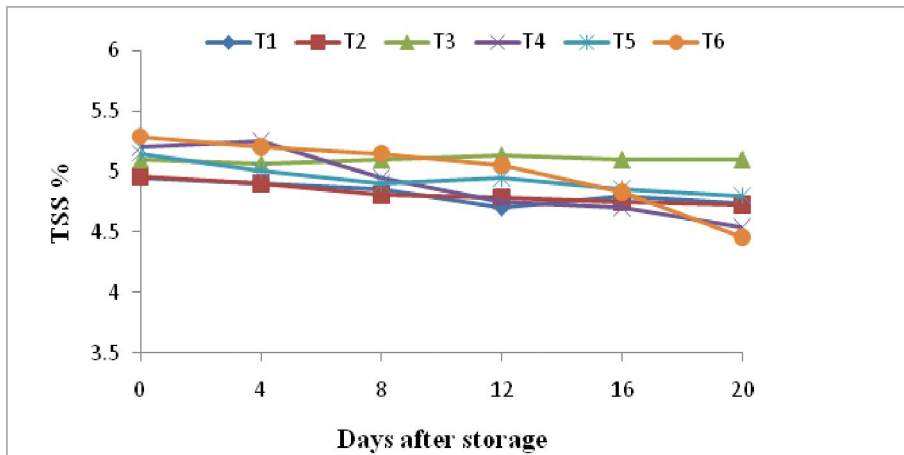


Fig.3. Variation of TSS for different treatments of tender coconut water during storage.

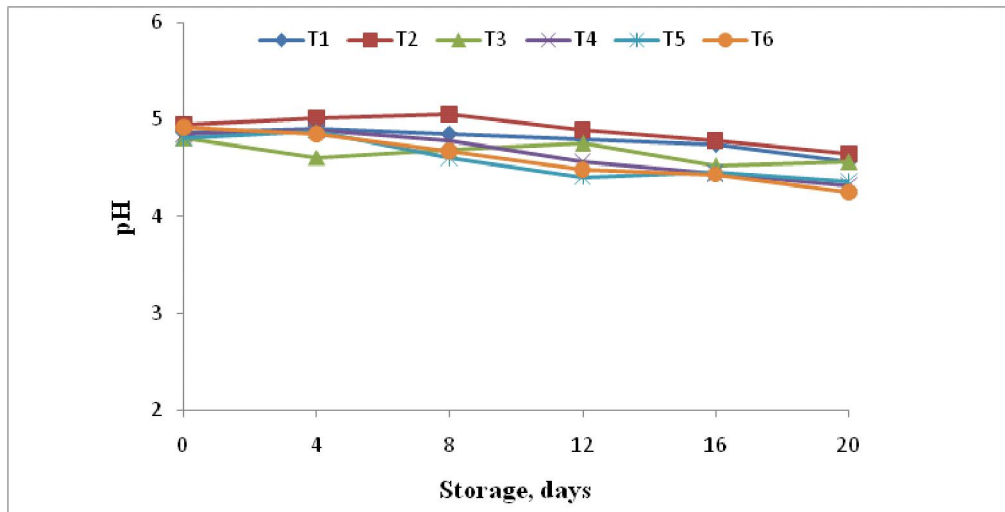


Fig.4. Variation of pH of different treatments of tender coconut water during storage.

reducing sugars to reducing sugars. Similar observations were made by Manashi *et al.* 2012 and Nikhil *et al.* 2014. The trend in increase of reducing sugars for treatment T<sub>3</sub> was relatively low perhaps due to thermal treatment that delayed the fermentation process of conversion of total sugars to reducing sugars.

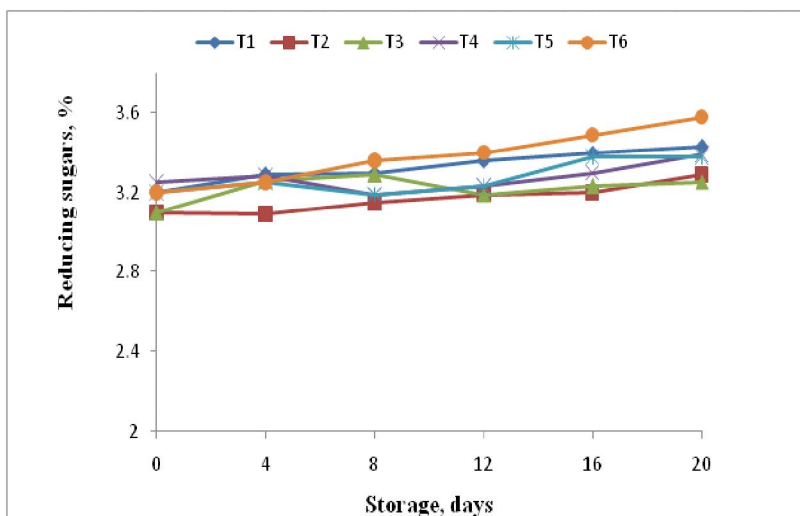
#### Turbidity

The turbidity of tender coconut water samples was found to increase during storage (Fig. 6). The turbidity was found to be more for sample T<sub>6</sub>. Faster fermentation process would have taken place and breakdown of sugars would have led to high turbidity in control sample. It was observed from the graph, the transmittance values decreased with increasing storage period. The turbidity of stored samples can be attributed to the increasing

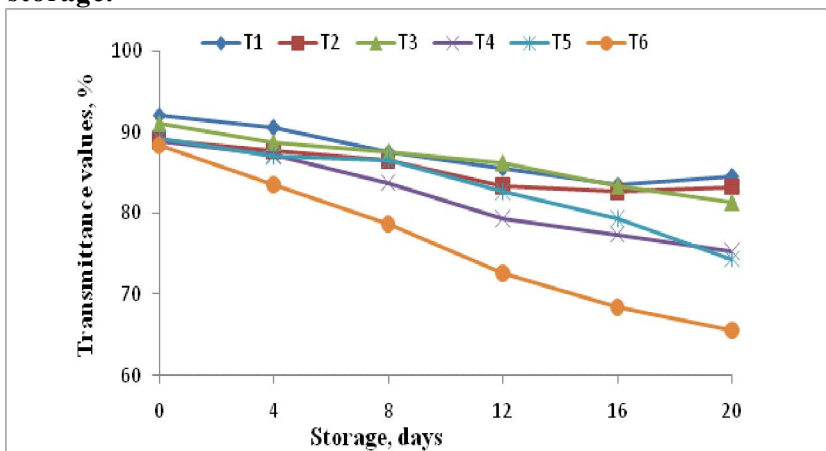
microbial load and other cell debris (Manashi *et al.*, 2012). The treatment T<sub>1</sub> had very high transmittance value because of removal of colloidal particles by microfiltration. Treatment T<sub>2</sub> also recorded high transmittance value when compared to treatment T<sub>6</sub> because of ultrafiltration by which most of solids were removed and the samples were less turbid. The experiments using MF and UF suggest that turbidity could be reduced and a clear TCW is possible using membrane filtration process.

#### Microbial load

There was no E.coli count found initially in all the treatments. In treatment T<sub>1</sub>, E.coli count increased to  $1.02 \times 10^6$  CFU / 10 ml during storage. There was no E.coli in treatment T<sub>3</sub> even on 20<sup>th</sup> day of storage period. However, in treatment T<sub>6</sub> higher colonies were observed. The fungal count



**Fig. 5.** Variation of reducing sugars of different treatments of tender coconut water during storage.



**Fig.6.** Variation of transmittance (%) of different treatments of tender coconut water during storage.

indicated no growth in all treated samples but in treatment T<sub>6</sub>, increased to  $2.73 \times 10^6$  CFU/10 ml on 20<sup>th</sup> day. Similar observations were made by Fernanda *et al.* (2009). The bacteria count other than coliform was observed to be more in treatment T<sub>6</sub> during storage and less for treatments T<sub>2</sub> and T<sub>3</sub>. Initially, there was no growth in treatment T<sub>2</sub>, then increased from  $0.01 \times 10^6$  CFU / 10 ml on 8<sup>th</sup> day to  $0.19 \times 10^6$  CFU / 10 ml on 20<sup>th</sup> day. The treatment T<sub>3</sub> had bacterial count of  $0.02 \times 10^6$  CFU / 10 ml on 20<sup>th</sup> day. The treatment T<sub>4</sub> had bacterial count of  $0.06 \times 10^6$  CFU / 10 ml on 4<sup>th</sup> day to  $2.74 \times 10^6$  CFU / 10 ml on 20<sup>th</sup> day. The treatment T<sub>5</sub> had bacterial count of  $0.18 \times 10^6$  CFU / 10 ml on 4<sup>th</sup> day to  $2.89 \times 10^6$  CFU / 10 ml on 20<sup>th</sup> day. Fernanda *et al.* (2009) reported similar microbial changes during storage in reconstituted stored coconut water.

### Sensory Evaluation

The membrane filtered, Pasteurized and untreated (control) tender coconut water samples were evaluated by panelists. The evaluated data assessed by panelists by filling a form. Appearance rating has been compiled. The treatments T<sub>1</sub>, microfiltered and T<sub>3</sub>, pasteurized tender coconut water scored highest rating on hedonic scale than other treatments. The sensory quality assessed by panel of experts for establishing flavour rating was also compiled. The treatment T<sub>3</sub> pasteurized TCW scored highest rating on hedonic scale than other samples. Similarly, the sensory quality assessed by panelists for establishing overall acceptability rating has been compiled. The treatments T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> scored higher rating on hedonic scale than other treatments.

## CONCLUSION

Experiments using a continuous cross flow flat sheet membrane module to process tender coconut water by microfiltration (MF) and ultrafiltration (UF) demonstrated the potential of membrane technology as one of the food preservation methods. Different treatments of TCW such as MF through 0.2 µm pore size, UF through a membrane of molecular weight cut off, 40 kDa, pasteurization at 85°C for 10 min and combination of MF filtered, addition of chemical preservative nisin revealed that pasteurization, MF and UF treatments result in better quality bottled TCW in that order, the first being the best among all the treatments upto a storage period of 20 days.

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