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Effect of yeast concentration and fermentation time on the characteristics of *tuak* from coconut sap

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Abstract

Tuak is a traditional fermented drink made from the sap of the Palmae family that is favored by rural communities. The quality of *tuak* varies greatly and depends on the type of microbe that contaminates the sap raw material. This research aimed to get the best yeast concentration and fermentation time, to get the best characteristics of *tuak* from coconut sap. The first factor was yeast concentration (0, 2, 4, 6, and 8%). The second factor was fermentation time (24, 48, 72, 96, and 120 hours). The research was grouped based on the processing time and got 50 units of the samples. The experiment was carried out at room temperature ($\pm 30^{\circ}\text{C}$). The parameter of this research was sensory evaluation test, alcohol content, pH, total dissolved solids, and total sugar. The data of this research were analyzed with analysis of variance, followed by the Duncan's test. The results showed that yeast concentration 4% w/v and fermentation time of 72 hours produced the best characteristics of *tuak* from coconut sap with the characteristics of sensory test for the alcohol flavor was between like to really like (4.30), the sour flavour was neutral to like (3.59), the sweet flavour was like to really like (4.85), and overall acceptance was like to really like (4.95) with alcohol content (9.38%), pH (4.2), total dissolved solids (7.52 mg/L) and total sugar (1.54%). The addition of starter culture can speed up the fermentation process and at the same time improve the quality of *tuak* from coconut sap.

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1. Introduction

Coconut plants (*Cocos nucifera* L.) are natural resources with great potential in Indonesia. Every part of the coconut palm can be utilized by humans in some way (1). One part of the coconut plant that can be used is the sap. The clear liquid that comes out of coconut flowers whose shoots have not yet opened is the main source of coconut sap. Coconut sap is tapped from the inflorescences of coconut trees that have not yet bloomed and are ripe. Tapping of coconut sap is done by cutting the inflorescence heads (2–4). Xia et al. (1) and Trisnamurti (5) reported that good quality and fresh coconut sap has a sweet taste, smells good, colorless, pH ranges from 6–7, has high nutrition, and is easy to digest. The composition of the sap of a plant species is influenced by several factors, including plant variety, plant age, plant health, soil conditions, climate, fertilization, and irrigation. Likewise, each plant has a different composition of sap and generally consists of water, sucrose, reducing sugar, other organic materials, and inorganic materials. The water in the sap is the largest part, which is between 75–90%. Sucrose is the largest part of solids ranging from

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12.30–17.40% (3). While Hebbar et al. (6) reported that the total sugar of fresh coconut sap (*Coconut inflorescence* sap) ranged from 9.2–16.2%. Reducing sugars are between 0.50–1.00% and the rest are organic and inorganic compounds. Reducing sugars may consist of hexose, glucose, and fructose, as well as very low amounts of mannose. Organic matter consists of carbohydrates (excluding sugars), proteins, organic acids, amino acids, dyes, and fats. Inorganic materials consist of mineral salts (7).

Fresh coconut sap is usually consumed as juice by local people in Southeast Asia and is also used as a raw material for producing sugar, alcoholic beverages, vinegar, and acetic acid (8,9). The milky white fermented sap is sold as an alcoholic beverage called 'toddy' or 'palm wine'; it is referred to as 'tuba' in the Philippines and 'tuak' in Indonesia (10). *Tuak* is a traditional drink produced from the fermentation of *Palmae* family sap (11–13). *Tuak* contains 4% alcohol (3,14). This product when distilled can produce *arak*, with an alcohol content ranging from 20–40% (15). The better the quality of the *tuak*, the better the *arak* produced.

Fermentation is influenced by temperature, pH, fermentation time, oxygen content, type and concentration of yeast (16). Indonesian people generally produce *tuak* using ingredients that come from nature and are fermented spontaneously without the use of starter cultures, such as dry yeast. The quality and yield of *tuak* can be increased by fermenting coconut sap with added yeast. Yeast is a microbe that can trigger or initiate fermentation in the processing of certain foodstuffs. The yeast that is often used in the process of making alcoholic beverages is *Saccharomyces cerevisiae* (17), and this microbe is naturally dominant in sap (12). These microbes will convert glucose on a substrate into CO₂ and ethanol. This microbe grows well at 25–30°C with a maximum growth temperature of 35–40°C. It is necessary to pay attention to the concentration of yeast in the manufacture of *tuak*. If the concentration is too little, it will reduce the speed of fermentation because there is little mass that will decompose glucose into ethanol, whereas if it is too much, more substrate will be needed because there is not enough substrate (18). Likewise with the fermentation time, the longer the fermentation time, the higher the ethanol content produced. However, if the fermentation time is too long, the ethanol that has been produced will be converted by bacteria into acetic acid (19). For this reason, it is necessary to research the concentration of yeast and fermentation time to produce the best characteristics of *tuak* from coconut sap.

2. Material and Methods

2.1. Materials

The materials used in this study were: coconut sap obtained from Jungutan Village, Karangasem Bali, Indonesia. The sap used is the sap that has just been taken down from a coconut tree and is stored in a sterile container. *Saccharomyces cerevisiae* ATCC 9763 was obtained from the Bogor Agricultural Institute Culture Collection (IPBCC). Materials for media manufacture: CaCl₂·2H₂O (Pudak Scientific brand), MgSO₄·7H₂O (KgaA brand), K₂HPO₄ (KgaA brand), KH₂PO₄ (KgaA brand), NaHCO₃ (KgaA brand), NaCl (KgaA brand), peptone and yeast extract (Himedia brand), glucose (Brataco brand), distilled water and materials for analysis were obtained from the Laboratory of Bioindustry, Agroindustrial Technology Study Program, Faculty of Agricultural Technology, Udayana University.

2.2. Experiment Design

The experiment design used was a factorial Randomized Block Design (RBD) consisting of two factors. The first factor was the concentration of yeast which consisted of 0, 2, 4, 6,

and 8% (v/v). The second factor is the fermentation time which consists of 24, 48, 72, 96, and 120 hours. All treatments were grouped into two groups based on the processing time so that 50 experimental units were obtained.

2.3. Preparation of Media, Culture, and Coconut Sap Fermentation

Cultures of *S. cerevisiae* were taken from culture stock, then rejuvenated on PDA media. After growing, 2-3 oses were taken to be re-grown in 50 mL of media containing yeast extract, peptone, and glucose (YPG). YPG media was prepared by taking 5 g/L yeast extract, 5 g/L peptone, and 10 g/L glucose dissolved in 50 mL distilled water and then sterilized at 121°C for 15 minutes. Cultures of *S. cerevisiae* were transferred aseptically into YPG media which had been cooled and incubated at 30°C for 24 hours and shaken at 100 rpm (20,21). After multiplication of yeast cells, then centrifuged at 5000 rpm at 4°C, then the pellet cells were washed with saline solution twice and finally the cell concentration was adjusted to OD660 5 before being used in the fermentation of coconut sap.

Sample preparation includes material preparation, i.e. freshly harvested coconut sap in a sterile container in a closed condition. The composition of fresh coconut sap before fermentation includes 8.11% total sugar, 2.75% reducing sugar, 14.07% total dissolved solids Brix, 0.06% total protein, 84.89% water content, 0.44 ash content, and pH 4.82. The sap is put into one container and then pasteurized at 72°C for 15 minutes, then used as a fermentation medium in the next stage.

The fermentation media was prepared by pouring 400 mL of sap into a bottle with a volume of 500 mL, then the yeast culture of *S. cerevisiae* was inoculated with a concentration of 0, 2, 4, 6, and 8% (v/v). The bottle that already contains the media and culture is closed, the bottle cap is given a hose as a distributor of CO₂ gas produced from the fermentation process, and the other end of the hose is placed in a 4% sodium metabisulfite solution.

The fermentation process was carried out for five days at room temperature ($\pm 30^\circ\text{C}$) at the Bioindustry Laboratory of the Faculty of Agricultural Technology (22). Observations were made daily, i.e. sensory tests, alcohol content, pH, and total dissolved solids.

2.4. Sensory Evaluation

Sensory evaluation was carried out to know the level of preference of the panelists for a food product. The acceptability of a product was always determined by the consumer of point of view (23). A total of 20 people were used as trained panelists to conduct sensory tests on *tuak* products from coconut sap. This sensory evaluation includes a hedonic test to assess the acceptability of product quality attributes such as aroma (alcohol, sour and sweet), color, taste, and overall acceptance of the *tuak*. The scores used are: 5 = really like, 4 = like, 3 = neutral, 2 = less like, and 1 = very dislike. The test was continued with a preference test by giving a score of 1 to the sample that had poor quality (very dislike) to a score of 5 for the best quality (really like) (24).

2.5. Alcohol Level Test

The alcohol produced during fermentation was determined using an alcoholmeter. The distillate resulting from the distillation of *tuak* as much as 100 mL was used as a sample, then the alcoholmeter was dipped into the distillate. The immersed boundary on the surface of the distillate indicates the alcohol content of the sample being tested (25).

2.6. Measurement of pH

The measurement of degree of acidity was determined using a pH meter. The pH meter electrode was inserted into each *tuak* sample solution. Measurement of pH was carried out on each sample at the same time every day (26,27).

2.7. Total Dissolved Solids

The measurement of total dissolved solids uses a refractometer and before being used, calibration was carried out using distilled water first. A total of 1 drop of the sample was put in the prism of the refractometer and the amount of dissolved solids content is expressed as °Brix. The value shown on the instrument was recorded and observations were made on the process before fermentation and after fermentation for each treatment variation. The difference between observations before and after fermentation in each treatment variation was compared (28).

2.8. Data Analysis

The data of psychochemical analysis were analyzed with analysis of variance using the SPSS application. If a treatment has a significant effect, then Duncan's test was performed. Sensory evaluations were analyzed using quantitative description.

3. Results and Discussion

3.1. Sensory Evaluation

3.1.1. Alcohol Aroma Sensory Evaluation

The analysis of variance showed that the interaction between yeast concentration and fermentation time had a very significant effect ($P < 0.01$) on the aroma of alcohol of *tuak* from coconut sap. The average value of the sensory test for the aroma of *tuak* from coconut sap can be seen in Table 1.

Table 1. The average value of sensory evaluation of the aroma of *tuak* from coconut sap.

Concentration starter culture (% v/v)	Fermentation time (hours)				
	24	48	72	96	120
0	4.60 ± 0.070 ^{ab}	4.73 ± 0.035 ^{ab}	4.82 ± 0.035 ^{ab}	4.22 ± 0.035 ^b	4.17 ± 0.035 ^b
2	4.83 ± 0.035 ^{ab}	4.93 ± 0.035 ^a	4.05 ± 0.070 ^{bc}	4.55 ± 0.070 ^{ab}	4.30 ± 0.070 ^{ab}
4	4.95 ± 0.070 ^a	4.17 ± 0.035 ^b	4.30 ± 0.070 ^{ab}	4.52 ± 0.035 ^{da}	4.47 ± 0.035 ^{ab}
6	4.18 ± 0.035 ^b	4.38 ± 0.035 ^b	4.55 ± 0.070 ^{ab}	3.87 ± 0.035 ^c	3.60 ± 0.000 ^c
8	3.88 ± 0.035 ^c	3.97 ± 0.035 ^{bc}	4.10 ± 0.070 ^b	3.57 ± 0.106 ^c	3.36 ± 0.160 ^d

Different letters behind the mean value indicate a very significant difference at the 5% error level ($p < 0.05$).

Based on Table 1, the average value of sensory evaluation of the aroma of alcohol ranged from 3.36 (neutral-like) to 4.95 (really like). *Tuak* from coconut sap treated with 8% yeast concentration and 120 hours of fermentation time had a neutral or normal preference level and *tuak* from coconut sap with 4% yeast concentration and 48 hours of fermentation time had a very favorable level of preference (4.17±0.035) and not different from 2% yeast treatment and 24 hours fermentation time (4.93±0.035). The alcohol aroma of *tuak* from coconut sap is influenced by the yeast concentration and fermentation duration.

3.1.2. Sour Aroma Sensory Evaluation Test

The analysis of variance showed that the interaction between yeast concentration and fermentation time had a very significant effect ($P < 0.01$) on the sour aroma of *tuak* from coconut sap. The average value of the sensory evaluation of the aroma of *tuak* from coconut sap can be seen in Table 2.

Table 2. The average value of sensory evaluation of sour *tuak* aroma from coconut sap.

Concentration starter culture (% v/v)	Fermentation time (hours)				
	24	48	72	96	120
0	3.57 ± 0.042 ^{ab}	3.7 ± 0.070 ^{aa}	3.67 ± 0.035 ^{ab}	3.21 ± 0.049 ^b	3.05 ± 0.035 ^{bc}
2	3.67 ± 0.035 ^{ab}	3.82 ± 0.035 ^{ab}	3.95 ± 0.007 ^a	3.51 ± 0.091 ^{ab}	3.17 ± 0.106 ^{bc}
4	3.80 ± 0.070 ^{ab}	3.87 ± 0.035 ^{ab}	3.95 ± 0.000 ^a	3.55 ± 0.106 ^{ab}	3.20 ± 0.045 ^b
6	3.25 ± 0.063 ^b	3.42 ± 0.035 ^{ab}	3.06 ± 0.084 ^{ab}	2.47 ± 0.035 ^c	3.02 ± 0.035 ^{bc}
8	2.89 ± 0.077 ^c	2.95 ± 0.035 ^c	2.85 ± 0.007 ^c	2.50 ± 0.070 ^c	2.59 ± 0.318 ^c

Different letters behind the mean value indicate a very significant difference at the 5% error level ($p < 0.05$).

The average value of the panelists' preference for the sour aroma of coconut sap ranged from 2.47 (less like-neutral) to 3.95 (like). *Tuak* from coconut sap treated with yeast concentration of 6% and fermentation time of 96 hours had a dislike level of preference, while the treatment with yeast concentration of 4% and fermentation time of 72 hours had a preference level of 3.95 (like) and was not different from the treatment with yeast 2% and 72 hours of fermentation.

3.1.3. Sweet Aroma Sensory Evaluation

The analysis of variance showed that the interaction between yeast concentration and fermentation time had a very significant effect ($P < 0.01$) on the sweet aroma of *tuak* from coconut sap. The average value of the sensory evaluation of the sweet aroma of coconut sap can be seen in Table 3.

Table 3. The average value of sensory evaluation of the sweet aroma of palm wine from coconut sap.

Concentration starter culture (% v/v)	Fermentation time (hours)				
	24	48	72	96	120
0	3.93 ± 0.021 ^{bc}	4.12 ± 0.035 ^{bc}	4.24 ± 0.055 ^b	4.35 ± 0.035 ^{3b}	3.79 ± 0.014 ^{bc}
2	4.37 ± 0.035 ^b	4.65 ± 0.141 ^{bb}	4.63 ± 0.021 ^{ab}	4.70 ± 0.077 ^{ab}	4.21 ± 0.049 ^b
4	4.50 ± 0.070 ^{ab}	4.74 ± 0.056 ^{ba}	4.85 ± 0.035 ^a	4.57 ± 0.035 ^b	4.29 ± 0.014 ^b
6	3.57 ± 0.035 ^c	3.85 ± 0.035 ^{bc}	3.92 ± 0.035 ^{ac}	3.37 ± 0.098 ^{cd}	3.14 ± 0.056 ^d
8	3.45 ± 0.035 ^c	3.51 ± 0.070 ^c	3.67 ± 0.106 ^c	3.14 ± 0.056 ^d	2.85 ± 0.033 ^d

Different letters behind the mean value indicate a very significant difference at the 5% error level ($p < 0.05$).

Based on Table 3, the average sensory test value for the sweet aroma of *tuak* from coconut sap ranges from 2.85 (neutral) to 4.85 (really like). *Tuak* from coconut sap treated with 8% yeast concentration and fermentation time of 120 hours had a neutral or ordinary preference level (2.85) and *tuak* from coconut sap with 4% yeast treatment and 72 hours fermentation time had a very high preference level (4.85). Panelists on the sensory test of the sweet aroma of *tuak* from coconut sap preferred *tuak* with 4% yeast concentration treatment and 72 hours of fermentation.

3.1.4. Overall Acceptance Sensory Evaluation

Based on Figure 1, the average sensory evaluation value of the overall acceptance of *tuak* from coconut sap ranged from 3.13 (neutral-like) to 4.95 (really like). *Tuak* from coconut sap treated with yeast concentration of 8% and fermentation time of 120 hours had a preference level of 3.13 (neutral-like). *Tuak* from coconut sap with yeast treatment of 4% and fermentation time of 72 hours had a preference level of 4.95 (really like). Panelists on the sensory test of the overall acceptance of *tuak* from coconut sap preferred *tuak* with 4% yeast concentration treatment and 72 hours of fermentation.

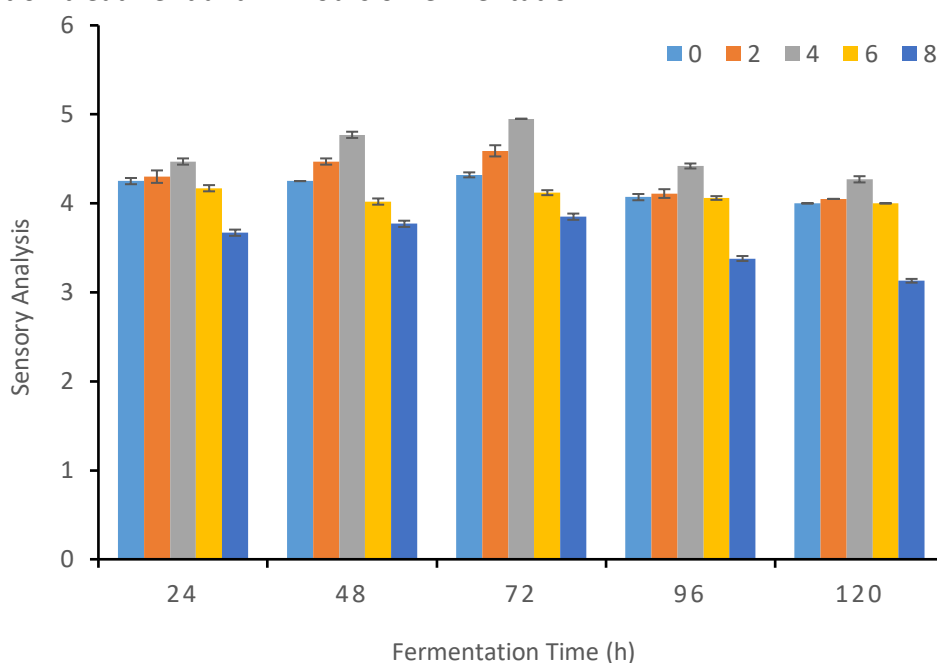


Figure 1. Sensory evaluation of the overall acceptance of *tuak* at different concentrations of yeast during fermentation.

3.2. Physicochemical Analysis

3.2.1. Alcohol Level

In Figure 2 it can be seen that the concentration of 8% starter culture with a fermentation time of 120 hours produced *tuak* with the highest alcohol content of 10.37%, but it was not significantly different from the starter culture treatment of 4% and the fermentation time of 72 hours (9.38%), starter culture 6% and fermentation time 72 hours (9.81%), starter culture 8% and fermentation time 72 hours (9.87%) and 96 hours (9.88%). While the starter culture concentration of 2% with 24 hours of fermentation resulted in the lowest average value of 3.88% alcohol content. This indicates that the higher the yeast concentration and the longer the fermentation time will increase the alcohol content of the *tuak* produced. This change can occur because yeast can break down glucose into alcohol and carbon dioxide. *S. cerevisiae* can form two molecules of ethanol and carbon dioxide from one glucose molecule. Fermentation time affects the alcohol content of *tuak*, longer the fermentation time, the higher the alcohol content will be. Similar results have been reported by Asngad et al. (29), the longer the fermentation process and the more doses of yeast given, the higher the alcohol content in papaya peel fermentation. The longer the fermentation, the more glucose is converted into alcohol, so the resulting alcohol content is higher (30).

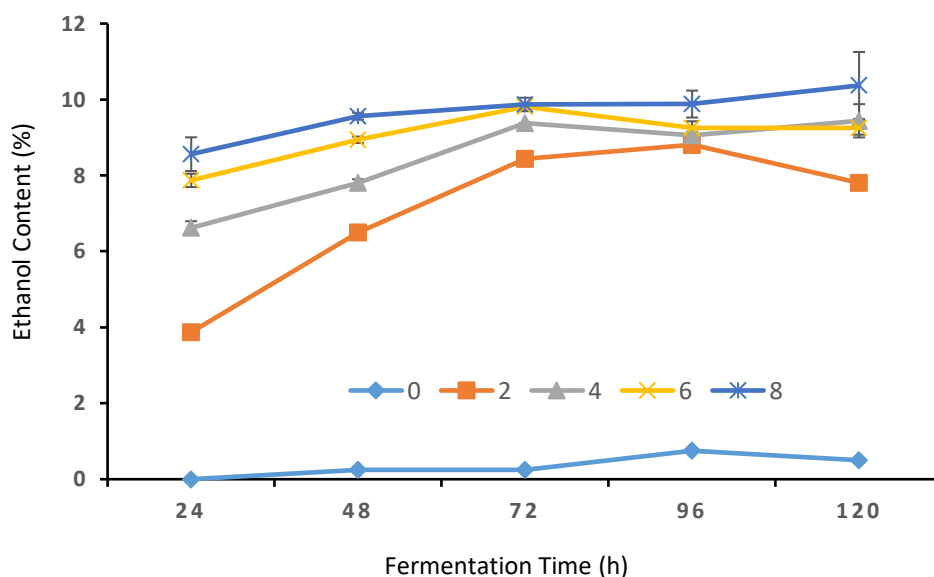


Figure 2. Changes in the alcohol content of *tuak* with different concentrations of yeast during fermentation.

3.2.2. pH

The analysis of variance showed that the yeast concentration treatment and fermentation time had a very significant effect ($P < 0.01$) on the pH of *tuak* from coconut sap. The average pH value of *tuak* can be seen in Table 4.

Table 4. The average pH value of *tuak* from coconut sap at different starter concentrations during storage.

Concentration starter culture (% v/v)	Fermentation time (hour)				
	24	48	72	96	120
0	4.9 ± 0.070 ^a	4.4 ± 0.141 ^{ab}	3.9 ± 0.000 ^b	3.8 ± 0.000 ^{bc}	4.0 ± 0.144 ^b
2	4.2 ± 0.212 ^{ab}	3.9 ± 0.070 ^b	3.9 ± 0.000 ^b	3.8 ± 0.144 ^{bc}	3.6 ± 0.000 ^c
4	4.2 ± 0.212 ^{ab}	4.2 ± 0.000 ^{ab}	4.2 ± 0.000 ^{ab}	4.0 ± 0.070 ^b	4.1 ± 0.000 ^{ab}
6	4.1 ± 0.070 ^{ab}	4.2 ± 0.000 ^{ab}	4.2 ± 0.000 ^{ab}	4.1 ± 0.070 ^{ab}	4.2 ± 0.141 ^{ab}
8	4.1 ± 0.000 ^{ab}	4.2 ± 0.000 ^{ab}	4.2 ± 0.000 ^{ab}	4.2 ± 0.140 ^{ab}	4.2 ± 0.000 ^{ab}

Different letters behind the mean value indicate a very significant difference at the 5% error level ($P < 0.05$).

Table 4 shows a decrease in the pH value with the increasing yeast concentration and the longer the fermentation time. The decrease in the pH of *tuak* from coconut sap was caused by fermentation products in the form of alcohol and carbon dioxide and the metabolism of *S. cerevisiae*. The formed CO₂ gas will react with water molecules to form H₂CO₃ as a carbonation reaction characterized by the formation by gas bubbles. H₂CO₃ will give an acidic atmosphere to *tuak* products so that *tuak* from coconut sap has a low pH (31). The study of Taherzadeh et al. (32) added that acetic acid can diffuse through cell membranes by lowering the internal pH. Thus, when the pH is low (acid), the enzyme activity will be inhibited so that the ability of microbes to break down sugar into bioethanol is lower. In addition, the presence of oxygen will also oxidize lactic acid so that the pH of the medium will decrease (33).

3.2.3. Total Dissolved Solids Content

The analysis of variance showed that the yeast concentration and fermentation time had a very significant effect ($P < 0.01$) on the total solids of *tuak* from coconut sap. The average value of the total dissolved solids content of *tuak* can be seen in Table 5.

Table 5. Average value of total dissolved solids content of coconut sap *tuak* (mg/L).

Concentration starter culture (% v/v)	Fermentation time (hour)				
	24	48	72	96	120
0	14.01 ± 0.000 ^a	14.12 ± 0.000 ^a	14.00 ± 0.000 ^a	13.06 ± 0.000 ^a	14.03 ± 0.000 ^a
2	8.04 ± 0.000 ^{cd}	9.00 ± 0.006 ^{bc}	9.12 ± 0.000 ^{bc}	9.97 ± 0.000 ^b	9.21 ± 0.000 ^{bc}
4	6.50 ± 0.707 ^d	8.34 ± 0.000 ^c	7.52 ± 0.707 ^{cd}	8.06 ± 0.000 ^{cd}	7.53 ± 0.707 ^{cd}
6	6.06 ± 1.414 ^{de}	7.29 ± 0.000 ^d	7.47 ± 0.770 ^{cd}	7.00 ± 0.000 ^d	7.50 ± 0.707 ^{cd}
8	5.00 ± 1.414 ^e	6.50 ± 0.711 ^d	6.50 ± 0.704 ^d	6.55 ± 0.707 ^d	6.50 ± 0.707 ^d

Different letters behind the mean value indicate a very significant difference at the 5% error level ($P < 0.05$).

During the fermentation process, sugar will be metabolized by *S. cerevisiae* into alcohol and CO₂, so that the total dissolved solids become low. Sintasari (34) reported that the low total dissolved solids were thought to be because, during the fermentation process, sugar which was the dominant solid component in the medium was metabolized by yeast into alcohol, and CO₂ was then utilized by acidic bacteria as a carbon source so that the total dissolved solids became low. Mulyawanti et al. (35) reported that the decrease in total dissolved solids during storage is due to the sugar contained will experience changes to alcohol, aldehydes, and amino acids. The remnants of organic acids, sucrose, and lactose dissolved in water will be counted as total dissolved solids (34). The decrease in total dissolved solids was also caused by the activity of yeast in breaking down sugar to produce alcohol during the fermentation process. Yeasts need substrates and nutrients for their survival. Substrate and nutrients will be reduced, causing the total amount of dissolved solids in the medium to be reduced (36). At an initial pH of 4.5 total dissolved solids were lowest at the end of fermentation, but with the highest total ethanol. According to research by Mulyawanti et al. (35), during the fermentation process, there was a decrease in total dissolved solids, due to the activity of yeast and bacteria in the sap. The lower the total dissolved solids content at the end of the fermentation, the better and the higher the ethanol produced, on the contrary, the higher the total solids content, the lower the ethanol produced and the less good quality of palm wine.

3.2.4. Total Sugar

The results of variance showed that the interaction between yeast concentration and fermentation time had a very significant effect ($P < 0.01$) on the total palm sugar from coconut sap. The average value of total sugar from coconut sap can be seen in Table 6.

Table 6. The average value of the total sugar of *tuak* from coconut sap (%).

Concentration starter culture (% v/v)	Fermentation time (hours)				
	24	48	72	96	120
0	8.53 ± 0.028 ^a	8.13 ± 0.007 ^{ab}	7.56 ± 0.084 ^{ab}	7.15 ± 0.063 ^{ab}	6.94 ± 0.036 ^b
2	6.20 ± 0.035 ^{bc}	4.15 ± 0.056 ^c	1.73 ± 0.035 ^d	0.46 ± 0.014 ^e	0.19 ± 0.007 ^e
4	6.00 ± 0.021 ^{bc}	3.57 ± 0.011 ^c	1.54 ± 0.033 ^{de}	0.35 ± 0.007 ^e	0.16 ± 0.007 ^e
6	5.69 ± 0.120 ^{bc}	3.11 ± 0.014 ^c	1.52 ± 0.063 ^{de}	0.27 ± 0.014 ^e	0.14 ± 0.014 ^e
8	5.22 ± 0.022 ^{bc}	3.21 ± 0.267 ^c	1.25 ± 0.021 ^{de}	0.24 ± 0.021 ^e	0.15 ± 0.007 ^e

Different letters behind the mean value indicate a very significant difference at the 5% error level ($p < 0.05$).

In Table 6 it can be seen that the highest total sugar was obtained from the treatment with 0% yeast concentration and 24-hour fermentation time, i.e., $8.53 \pm 0.028\%$. The lowest total sugar was obtained from the treatment with a 6% yeast concentration and 120 hours of fermentation time, i.e., $0.14 \pm 0.014\%$. At the time of increasing the concentration of yeast and fermentation time resulted in a decrease in total sugar. This decrease in total sugar was due to the use of glucose by *S. cerevisiae* for metabolism. Pandiselvam et al. (37) reported that during fermentation there was a rapid decrease in total sugar at room temperature compared to under refrigerated conditions. *S. cerevisiae* has a high ability to ferment glucose, fructose, galactose, maltose and has resistance in the environment at relatively high alcohol levels and is resistant to other microbes. *S. cerevisiae* is a genus of yeast that can convert glucose into ethanol and CO₂. During fermentation, sugar will be consumed as a carbon source and converted into alcohol and CO₂ due to yeast activity (15).

4. Conclusions

From the results of the study, it can be concluded as follows: the interaction between yeast concentration treatment and fermentation time has a very significant effect on sensory tests, alcohol content, pH, total dissolved solids, and total palm sugar from coconut sap. The characteristics of the best *tuak* from coconut sap were obtained from the treatment of 4% yeast concentration and 72 hours of fermentation time producing characteristics based on the results of sensory evaluation, i.e. the panelists' preference level for alcohol aroma 4.30 (between like-really like), sour aroma 3.95 (like), sweet aroma 4.85 (really like), and overall acceptance was 4.95 (really like), with physicochemical characteristics, i.e. alcohol content (9.38%), pH (4.2), total solids (7.52 mg/L) and total dissolved sugar (1.54%).

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Author Contributions

I.B.W.G. and N.P.S. were responsible for experiments design and coordinating all the research processes; T.E.B.K. performed the experiments and analyzed the data; I.B.W.G. dan T.E.B.K. wrote the paper.

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Data Availability Statement

Not applicable.

Conflicts of Interest

Authors may declare no conflict of interest.

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