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The Effect of Fermentation Time and Mass Variation of Saccharomyces Cerevisiae on The Characteristics of Virgin Coconut Oil from The Coconut Milk Fermentation Process

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A B S T R A C T

Virgin coconut oil is one of the processed coconut products that has many benefits. Virgin coconut oil can be produced through extraction, centrifugation, and fermentation processes. In this study, virgin coconut oil was made by fermentation using baker's yeast (Saccharomyces cerevisiae). This study aims to determine the effect of fermentation time and mass variation of Saccharomyces cerevisiae used on the characteristics of virgin coconut oil produced. The process of making virgin coconut oil in this study is to separate skimmed water from coconut milk. Skimmed steering is mixed with Saccharomyces cerevisiae with variations of 0, 1, 1.5, and 2 grams, with fermentation durations of 12, 18 and 24 hours. The resulting virgin coconut oil is then analyzed to determine the iodine, peroxide, and acid numbers. From the results of the study, it is known that there is no relationship between the duration of fermentation and the mass of yeast Saccharomyces cerevisiae in the manufacture of virgin coconut oil from coconut milk against iodine number. The peroxide number for all variables of fermentation duration and yeast mass is 2 meq/kg. The acid number tends to increase with the length of fermentation, with the highest value being 0.6% at 24 hours fermentation time and the yeast mass of Saccharomyces cerevisiae 1.5 grams.

INTRODUCTION

Coconut (*Cocos nucifera L.*) is a plant that can be used by humans, from the tops of the leaves to the roots. Coconut belongs to the palm tribe (aracaceae), is a plant that has many benefits, which thrives in tropical regions such as in Indonesia. Coconut palms grow up to 30 meters, producing 75 fruits per year with favorable environmental conditions (da Silva Lima &; Block, 2019).

In the city of Sorong, Southwest Papua Province, coconuts thrive along coastlines as well as plantations. Coconut production in West Papua Province in 2019 was 16,169 yon, an increase of 2.62% compared to 2018. So that diversification of coconut products is needed to increase the usefulness of coconuts in the community.

At the beginning of 2022, Indonesia experienced a scarcity of palm oil products, which are the people's main choice in processing fried foods. This causes people to start looking for other alternatives in the use of goring oil, one of which is by using coconut oil. Coconut oil can be made on a household scale by cooking coconut milk into coconut oil and Blondo. However, this process requires a long time and a lot of fuel (Karouw & Santosa, 2013).

Besides being able to be obtained into cooking oil, old coconut milk can be processed into *Virgin Coconut Oil* (VCO) which does not require high temperatures in the manufacturing process, otherwise known as *the wet coconut process*. Unlike in the manufacture of coconut oil, VCO maintains the distinctive aroma of

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coconut to produce more fragrant dishes. VCO contains higher saturated fatty acids than palm oil, so it tends to be more stable in structure (Mujdalipah, 2016).

VCO is also considered healthier, with antioxidant content, acts as a probiotic, and can play a role in weight loss. In addition, VCO can also be used in skin and hair care. The process of making VCO can be done in several ways, namely: enzymatic, microbial fermentation, acidification, centrifugation and fishing methods (Mujdalipah, 2016).

Virgin coconut oil is extracted from fresh coconut flesh or dried coconut meat. The oil content in copra is generally 60-65%, while the oil content in fresh coconut flesh is around 43% (Andaka &; Sentani, 2016). The centrifugation method is a mechanical method, which is done by breaking the fat-protein bonds in coconut milk by rotating at high speed. After centrifugation, water and oil will separate by themselves due to differences in the specific gravity of water and oil (Sherliana et al., 2021).

In this study, a fermentation process was used for making VCO by using baker's yeast (*Saccharomyces cerevisiae*) as a starter in the fermentation process of old coconut milk to become VCO. Baker's yeast is used because in the form of emulsions it can produce enzymes that play a role in converting glucose into alcohol. The resulting alcohol plays a role in breaking down the coconut milk emulsion to produce oil (Khazalina, 2020).

The advantages of fermentation demoted are energy saving, easy to take the oil produced and the quality of the oil produced can be maintained by adjusting the ratio of raw materials with *Saccharomyces cerevisiae* used (Erika et al., 2014).

METHOD

The necessary ingredients are aged coconut meat, water, baker's yeast (*Saccharomyces cerevisiae*), chloroform, Wijs solution, Potassium Iodide, NaOH, Sodium Thiosulfate, amylum indicator, phenolphthalein indicator, 95% ethanol. Old coconut meat obtained at the market in Sorong City is shredded and squeezed by adding enough water to produce coconut milk. Coconut milk is then allowed to stand for 2 hours to get coconut milk cream. The coconut milk cream is then separated from the water. Baker's yeast starter as much as 1 gram, 1.5 grams, 2 grams added to 100 grams of coconut milk cream. After that, the fermentation process is carried out anaerobically for 12 hours, 18 hours and 24 hours.

IOD Number Test

The iodine number was measured by weighing 5 grams of coconut oil sample and then put into a lided Erlenmeyer. Then added 10 ml chloroform and 25 ml of Wijs reagent, then stored in a dark room for 30 minutes and occasionally shaken. Then added 10 ml of 15% KI solution, 50 ml of water and 2 ml of

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amylum solution. Then continue by titrating with 0.05 N sodium thiosulfate until the blue color disappears (Sinurat &; Silaban, 2021)

The iodine number is expressed as grams of iodine absorbed per 100 g using the formula:

$$Bilangan\ iod = \frac{12{,}69\ \times T\ (V_3 - V_4)}{m}$$

Information:

T = Normality of standard solutions of sodium thiosulfate

V_3 = Volume of sodium thiosulfate solution required for blank titration (ml)

V_4 = Volume of natrium thiosulfate solution required for sample titration

m = sample weight (grams)

Peroxide Number Test

The peroxide number was measured by weighing 0.3 - 5.0 grams of oil samples into a 300 ml Erlenmeyer, adding 30 ml of a solution mixture of 20 ml of glacial acetic acid, 25 ml of 95% ethanol and 55 ml of chloroform. Add I gr KI and keep in a dark place for 50 ml, then add water, titrate with a standard solution of 0.02 N sodium thiosulfate with starch solution as an indicator. Determine blanks and calculate the peroxide number in the sample (*Indonesian National Standard: How to Test Oil and Fat*, 1998).

The peroxide number is expressed in milligrams equivalent of active oxygen per kg, using the formula:

$$Bilangan\ peroksida\ \left(\frac{mgrek}{kg}\right) = \frac{(V_0\ -\ V_1\ ) \times T}{m} \times 1000$$

Information:

V_0 = Volume of sodium thiosulfate solution for blanks, in ml

V_1 = Volume (ml) of sodium thiosulfate solution for sample

T = Normality of standard solutions of sodium thiosulfate used

M = Sample weight in grams

Acid Number Test

Weigh 2-5 grams of oil sample into Erlenmeyer 250 ml, then add 50 ml of 95% neutral ethanol. Add 3 – 5 indicators of Phenolphthalein and titrate with a standard solution of 0.1 N NaOH until the pink color remains (unchanged for 15 seconds) (*Indonesian National Standard: How to Test Oil and Fat*, 1998).

The acid number is expressed as mg KOH / gram of fat using the formula:

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$$Kadar\ asam\ lemak\ bebas = \frac{M \times V \times T}{10\ m}$$

Information:

V = volume of NaOH required in titration (ml)

T = normality NaOH

m = Example weight, in grams

M = Molecular weight of fatty acids

In coconut oil testing, the acid number is calculated as lauric acid, which has a molecular weight of 200.

RESULT AND DISCUSSION

The result of the physical appearance of virgin coconut oil is that it has a distinctive coconut smell, not rancid, a distinctive taste of coconut oil and is clear in color. The physical appearance of virgin coconut oil produced in accordance with the Indonesian National Standard (SNI), is similar in terms of smell, taste, and color.

Effect of Fermentation Duration and Yeast Mass of Saccharomyces cerevisiae on Iodine Number

The iodine number can indicate the degree of unsaturation of the constituent fatty acids of the fat. Based on linear regression analysis, it showed that the entire treatment, both the addition of yeast and the duration of fermentation, had no noticeable effect ($p > 0.05$). Based on table 1, the average iodine number resulting from the influence of fermentation duration and yeast addition is 4.16 – 4.57, where this value is in accordance with the standard set by SNI for iodine number in virgin coconut oil, which is 4.10 – 11.0 mgrek / kg.

Table 1. Effect of Fermentation Duration and Yeast Mass of *Saccharomyces cerevisiae* on Iodine Number

Fermentation Time	Yeast of *Saccharomyces cerevisiae* mass			
	0 gr	1 gr	1,5 gr	2 gr
12 Hours	4,31	4,48	4,40	4,19
18 Hours	4,42	4,37	4,57	4,26
24 Hours	4,16	4,29	4,29	4,25

A low iodine number indicates that the oil does not contain many unsaturated fatty acids. Unsaturated fatty acids can bind iodine and form saturated compounds or unsaturated bonds contained in oil. The number of iodine tied indicates the number of double bonds (Widjaja &; Anjarsari, 2014).

Effect of Fermentation Time and Yeast Mass of Saccharomyces cerevisiae on Peroxide Number

Peroxide number is the amount of peroxide present in the sample, expressed in terms of milliequivalent active oxygen per kg, which oxidizes potassium iodide under treatment conditions such as in peroxide number testing (SNI). It is associated with rancidity in the oil, as it is associated with a decrease in quality and shelf life. oil (C.C. Kusuma et al., 2022).

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Table 2. Effect of Fermentation Time and Yeast Mass of *Saccharomyces cerevisiae* on Peroxide Number

Fermentation Time	Yeast of *Saccharomyces cerevisiae* mass			
	0 gr	1 gr	1,5 gr	2 gr
12 Hours	2	2	2	2
18 Hours	2	2	2	2
24 Hours	2	2	2	2

In this study, it was found that in all variables, both the length of fermentation time and the mass of yeast Saccharomyces cerevisiae used, produced the same peroxide number, which is 2 meq / kg. The value of this peroxide number is still in accordance with the SNI standard for virgin coconut oil, which is 2 meq / kg. Meanwhile, the results of Patty's research (2015) show that the longer the fermentation time in traditional coconut oil products, the peroxide number will increase. This is because the longer the fermentation time, the unsaturated fatty acids contained in coconut oil will be greater and make the oil in direct contact with oxygen, thus the reaction of free radical formation which is then converted into hydroperoxide will increase (Patty, 2015). Oil oxidation reactions begin with the formation of free radicals caused by factors that can speed up reactions such as light, thermal energy, metal catalysts and enzymes. Free radicals with oxygen will form active peroxides that can form hydroperoxides that are very unstable.

Effect of Fermentation Time and Yeast Mass of Saccharomyces cerevisiae on Acid Number

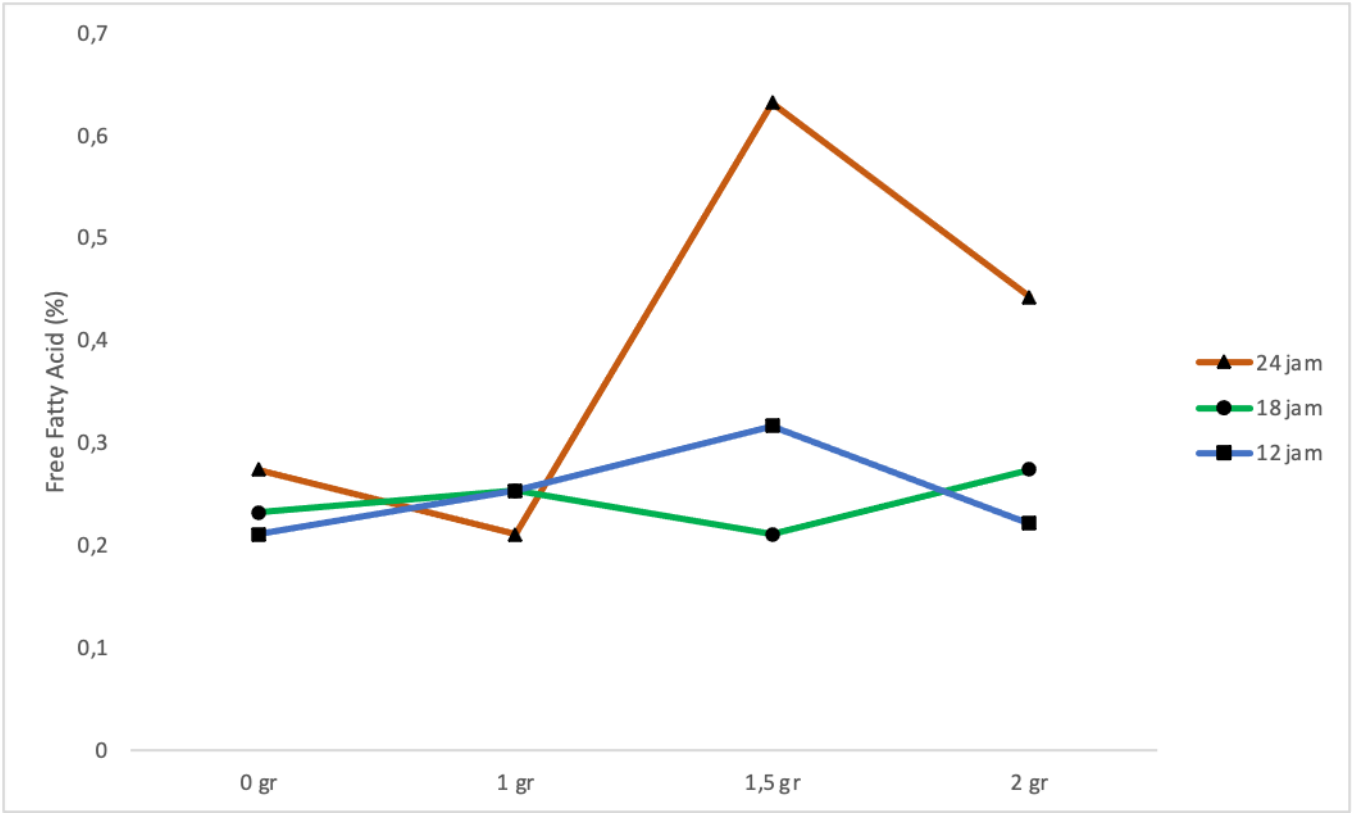

Figure 1. The acid number of virgin coconut oil

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The results of the number of free fatty acids obtained ranged from 0.2 – 0.6%, where the number of free fatty acids at 24 hours fermentation time with the addition of *Saccharomyces cerevisiae* yeast as much as 1.5 gr higher than the time and the addition of other yeasts.

Free fatty acids can be benthic since the oil is still in plant tissues, due to the presence of lipase enzymes that can hydrolyze neutral fats (triglycerides). But in living organisms, enzymes are generally in an inactive state or state because there are still interactions between cells. In organisms that have died, the mechanism of the cells gets damaged so that the lipase enzyme starts working and damages the fat molecules. The speed of hydrolysis of lipase enzymes present in tissues is relatively slower at low temperatures and will be more intensive under suitable conditions. Coconuts that have been shredded, the cell structure has been damaged so that the lipase enzyme begins to work to damage fat molecules (Pontoh, 2008). The longer the fermentation time, the higher the free fatty acids contained in the oil (Sherliana et al., 2021). Free fatty acids are one of the parameters of oil damage due to the hydrolysis process with interaction with water and lipase enzyme activity, so the lower the free fatty acids, the better the quality of the oil produced. The maximum requirement for the number of free fatty acids for virgin coconut oil according to SNI is 0.2%.

CONCLUSION

There is no relationship between the duration of fermentation and the mass of the yeast *Saccharomyces cerevisiae* in the preparation of virgin coconut oil from coconut milk to iodine number. The peroxide number for all variables of fermentation duration and yeast mass is 2 meq/kg. The acid number tends to increase with the length of fermentation, with the highest value being 0.6% at 24 hours of fermentation time and the yeast mass of *Saccharomyces cerevisiae* 1.5 grams.

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