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Abstract

The purpose of this research is to quantitatively and qualitatively examine the fermentation parameters of dry cocoa beans to be more effective in determining the role and function of microorganisms. The three fermentation methods are as follows: control (without inoculums), Saccharomyces cerevisiae (FNCC inoculums 3056), Lactobacillus lactis (FNC 0086), and Acetobacter aceti (FNCC 0016), all at a concentration of approximately 108 CFU/g were added simultaneously at the start of the fermentation (A), and all three inoculums were added in stages (B). The results indicated that during fermentation, all three treatments decreased total sugar content, pH, and total polyphenols. In all treatments, the concentrations of ethanol, lactic acid, and acetic acid peaked at 24, 60, and 108 hours of fermentation, respectively. At 24, 48, and 72 hours of fermentation, the highest populations of Saccharomyces cerevisiae, Lactobacillus lactis, and Acetobacter aceti were obtained from the three treatments.

Keywords

Microbes, Saccharomyces Cerevisiae, Lactobacillus Lactis, Acetobacter Aceti, Fermentation and Dried Cocoa Beans

1. Introduction

Cocoa is one of Indonesia's primary plantation commodities, as it generates foreign exchange, creates jobs, and stimulates agribusiness and agro-industry growth [1, 2]. In general, the production of dried cocoa beans from community plantations does not involve fermentation, either naturally or through the addition of inoculums. Generally, cocoa farmers soak fresh cocoa beans in water to aid in pulp removal and then expose them to the sun [3, 4]. Dried cocoa beans with an unknown water content are sold without regard for their quality, both in terms of moisture content and condition. This is because there are a small number of farmers producing cocoa and the fermentation process takes an excessive amount of time [5, 6]. Fermentation is a critical step in the processing of cocoa beans, particularly for the formation of flavour precursor compounds. Fermentation of fresh cocoa beans occurs in two stages: the first stage involves the removal of pulp from the seeds' surfaces, and the second stage involves a hydrolytic reaction within the seed cotyledons [7, 8]. Microbial succession refers to the changes in the type and number of microbes that occur during the fermentation of cocoa beans. The initial stages of fermentation are dominated by yeast, followed by lactic acid bacteria, and finally by acetic acid bacteria.

Cacao, a precursor compound for the flavour of seeds, can be formed through fermentation. When fermentation is partitioned into two stages. The pulp is removed first, followed by a hydrolytic reaction in the seed cotyledon. The second step involves the microbial succession process [9, 10]. The fermentation of cocoa takes 120 hours or six days and is reversed on days 2, 3, 4, and 5. During the early stages of S. cerevisiae fermentation, a microbial colony plays a role in the start of fermentation [11, 12]. This is because S. Cerevisiae can grow well in the presence of ethanol and tolerate a low pH, which is required for pectinolytic activity. Lactobacillus Lactis, which is tolerant of acidic environ-

ments with a high oxygen concentration. At the end of fermentation, Acetobacter aceti predominates due to its ability to grow at pH 3.5 and low ethanol concentrations. Following that, ethanol is oxidized to form acetic acid, which is then converted to CO2 and water [13]. As the ambient temperature rises, alcohol and acetic acid will diffuse into the seeds, resulting in seed death [14, 15].

2. Methods

Fruit matures to a length of 16 cm and a diameter of 9 cm. The method refers to research [16] with minor modifications. Fermentation is accomplished in three distinct ways. Technique as a control, I fermented spontaneously (without inoculum). The second technique (A) involves the simultaneous addition of S. Cerevisiae pure culture (FNCC 3056), Lactobacillus lactis (FNC 0086), and A. aceti (FNCC 0016) at the start. The third technique (B) incorporates pure cultures gradually, beginning with S. Cerevisiae pure culture (FNCC 3056) at the start of hour 0 fermentation, followed by L. lactis (FNC 0086) after 24 hours, and finally A. aceti after 72 hours [17]. The success of fermentation is determined by changes in total sugar content, pH, total polyphenols, S. cerevisiae (FNCC 3056), Lactobacillus lactis (FNC 0086), and Acetobacter aceti (FNCC0016), total polyphenols, and ethanol, lactic acid, and acetic acid concentrations [10].

3. Results

The results indicated that the initial temperatures of the control treatments II and III were 28, 29 and 29.5°C, respectively. The highest fermentation temperatures were 38, 42, and 51°C for control treatments II and III, respectively. At 120 hours, the fermentation temperatures for control treatment, II, and III were 35, 38, and 39°C, respectively. The sugar content for control treatment, II, and III were 3.07 ± 0.67, 2.60 ± 0.65 and 2.70 ± 0.7. pH for control treatment, II, and III were 4.45 ± 0.17, 4.28 ± 0.2 and 4.20 ± 0.2. The Reduction sugar content for control treatment, II, and III were 10.53 ± 0.55, 10.63 ± 0.53 and 10.57 ± 0.52. S. cerevisiae (log cfu/g) for control treatment, II, and III were 3.55 ± 0.25, 2.30 ± 0.5 and 2.22 ± 0.5. L. lactis (log cfu/g) for control treatment, II, and III were 5.42 ± 0.17, 6.42± 0.6 and 6.23± 0.6. A. aceti (log cfu/g) for control treatment, II, and III were 4.87 ± 0.47, 5.30 ± 0.8 and 9.22 ± 0.8. Etanol (%) for control treatment, II, and III were 0.40 ± 0.30, 0.33 ± 0.7 and 0.20 ± 0.5a. Lactic acid (%) for control treatment, II, and III were 2.00 ± 0.17, 1.82 ± 0.9 and 1.70 ± 0.3a. Asetic acid (%) for control treatment, II, and III were 5.90± 0.28, 6.32 ± 0.2, and 6.80 ± 0.6. Pholipenol (mg asam galat/g) for control treatment, II, and III were 0.079 ± 0.14, 0.07 ± 0.05 and 0.068 ± 0.2 (Table 1).

Table 1. Results of statistical analysis of changes in total sugar content, pH, total polyphenols, ethanol, lactic acid, acetic acid, S. Cerevisiae (FNCC 3056), L. lactis (FNC 0086) and A. aceti (FNCC0016)

Parameter	Treatment		
	control	II	III
Sugar content (%)	3.07 ± 0.67a	2.60 ± 0.65a	2.70 ± 0.7a
Reduction sugar (%)	10.53 ± 0.55a	10.63 ± 0.53a	10.57 ± 0.52a
pH	4.45 ± 0.17a	4.28 ± 0.2a	4.20 ± 0.2a
S. cerevisiae (log cfu/g)	3.55 ± 0.25a	2.30 ± 0.5a	2.22 ± 0.5a
L. lactis (log cfu/g)	5.42 ± 0.17a	6.42± 0.6b	6.23± 0.6b
A. aceti (log cfu/g)	4.87 ± 0.47a	5.30 ± 0.8b	9.22 ± 0.8b
Etanol (%)	0.40 ± 0.30a	0.33 ± 0.7a	0.20 ± 0.5a
Lactic acid (%)	2.00 ± 0.17a	1.82 ± 0.9a	1.70 ± 0.3a
Asetic acid (%)	5.90± 0.28a	6.32 ± 0.2a	6.80 ± 0.6a
Pholipenol (mg asam galat/g)	0.079 ± 0.14a	0.07 ± 0.05a	0.068 ± 0.2a

Notes: Different letters behind show significant differences p <0.05. Results are of an average of 3 replications of the analysis.

4. Discussion

The change in fermentation temperature observed in this study is consistent with the findings of [18], who investigated ways to improve the fermentation process of dried cocoa beans. In the gradual treatment, S. cerevisiae can remodel more pulp sugar, increasing the amount of ethanol produced, and at the appropriate time, ethanol is converted to acetic acid by [20]. A. aceti, where the reaction to convert ethanol to acetic acid also generates heat, increasing the fermentation temperature [20]. When the inoculum is added concurrently with the seeds, and the seed death temperature is

not reached, competition between S. cerevisiae and L. lactis is expected. Throughout the treatment, the total sugar content of the dried cocoa beans decreased by up to 120 hours. Sucrose, fructose, and glucose are the sugars found in cocoa beans [21], with sucrose accounting for 90% of total sugar and fructose and glucose accounting for 6% (0.9 and 0.7 per cent) of total sugar, respectively, and including mannitol and inositol at less than 0.50 mg/g [22]. The total sugar content of dry cocoa beans at the start of fermentation treatment control, A, and B was 8.45; 8.45; and 8.5 per cent, respectively, and then decreased to 3.07; 2.60, and 2.70 per cent [22].

5. Conclusion

The quality of dried cocoa beans after fermentation results can be improved. Treatment B, namely the addition of the inoculum gradually at the end of the fermentation showed better quality than the other treatments.

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