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Fungi Level Analysis of Cocoa Beans Based on Fermentation Box Type and Duration

Retno Utami Hatmi^a*, Mahargono Kobarsih^a and Nurdeana Cahyaningrum^a

^aAssessment Institute for Agricultural Technology Yogyakarta Jl. Stadion Maguwoharjo No. 22 Ngemplak Sleman Yogyakarta - 55584

Abstract

In certain location, it is undeniable that there are qualities variations of fermented cocoa beans depend on processing techniques. In addition, its cocoa bean's production central at Yogyakarta have their specific characteristic own regarding the use of fermentation box (box type). The purpose of this research was to analyze the effects of three types of fermentation box and duration to fungi level of fermented cocoa beans. CRD (completely randomized design) with two factors and three replications is applied as assessment method. The first factor is the type of fermentation box (basket, storey box and single box with a hand crank) and the second factor is the length of fermentation time (3 and 5 days). The result of the research explained that the lowest levels of the fungi achieve within 5 days of fermentation processes. Meanwhile, a single box with a hand crank provide the lowest of fungi's level. Furthermore, the combination of those two results may give the most favorably expected condition of fungi level.

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Keywords: fermentation box types, duration, fungi levels, basket, storey box and single box with a hand crank.

INTRODUCTION

Fermentation is the core of processing cocoa beans [20]. The purpose fermentation process is remove the pulps from cocoa beans and turn off the cocoa beans, improve and establish odor and distinctive flavor of chocolate and reduce bitter taste. The fermentation process can be done in three ways, namely using a box, in a pile, and in the

^{*} Corresponding author. *E-mail address:* tamibptp@yahoo.co.id

basket [9].

The factors that influence the fermentation process include length of fermentation, speed of fermentation, size piles of cocoa beans, the reversal, aeration and weather [5]. The determination of cocoa bean fermentation distinguished on several things, among others varieties of cocoa, size of the cocoa beans, quantity fermented cocoa beans. In some countries, the fermentation is divided into two groups, i.e. groups with a long fermentation 2-3 days and 6-8 days. Generally, varieties of *Lindak* (the cocoa beans from cocoa plants Criolo and Trinitario types and the results of its cross) that fermented for 5-7 days, whereas for the cocoa varieties *Mulia* (the cocoa beans from cocoa plant Forastero type) requires shorter fermentation time, which is 3 days.

In addition to duration of fermentation, fermentation box type affecting the fermentation process itself and to the quality of fermented cocoa beans are produced. The fermentation box type influence fermentation aeration during the lasted. Aeration is the processing that encourages the changes that will lead to the death of cocoa beans. Good aeration will provide a more well fermentation. Mass amounts of fermented cocoa beans, container shape and size, surface area, thick pile of cocoa beans, container ventilation, and number of reversals determines good or not aeration

This research purposes to identify the fungi level on some fermentation box type and duration (three and five days). Gunungkidul and Kulonprogo is the highest cocoa producing areas in Yogyakarta. Each region has different habits in using fermentation box. The fermentation box types are the basket (the fermentation box that being use commonly in Gunungkidul, storey box (Kulonprogo) and single box with a hand crank (introduction fermentation box). This is important, because each fermentation box type drive a quality range of cocoa beans. These results are expected to identify which boxes and duration providing the lowest fungi level contaminant. Whereas, the benefit of this study is produces an efficient box design for better fermentation process with fungi levels low.

MATERIALS AND METHODS

The raw material that being used in this research are *Lindak* Varieties which supplied by designated targeted farmer from Kulonprogo and Gunungkidul area. The number of cocoa beans will be based on the capacity of fermentation containers. This study carried out within one year (January – December 2012) in cocoa central of Yogyakarta.

Primary equipment in the fermentation is fermentation box. There are three types of fermentation box are used in this study, namely basket, storey box, and a single box with a hand crank. While fermentation support tools used are plastic sacks, thermometer, bucket, *rigen*, and drying racks. The capacity of each box is 40 kg (single box with a hand crank), 20 kg (basket), and 60 kg (storey box). The process of cocoa fermentation refers to the Research Centre of Coffee and Cocoa (*Puslit KoKa*) procedure [Fig 1.].

This study was conducted using a completely randomized design, two factors and three replications. The first factor is the type of fermentation box (basket, storey box and single box with a hand crank) and the second

factor is duration of fermentation (3 and 5 days). Observation material was fungi level on fermented cocoa beans. Determination of fungi's level refers to the Indonesia National Standard of cocoa bean: SNI 2323-2008 procedure [2]. Observations were processed using descriptive and statistical analysis with one way ANOVA test and DMRT.

RESULTS AND DISCUSSION

Design and a working systems of fermentation container Basket (traditional container)

The basket is one of the traditional fermentation containers [Fig 2.]. The basket is made of woven bamboo materials. It is also preferable to family holders with small production of cocoa beans. The basket has been used commonly to farmers in Gunungkidul. The basket has capacity ranging 15 - 30 kg. In the fermentation process required two units basket for reversal of cocoa beans. Reversal of cocoa beans using a greater manpower to lift and spilling cocoa beans from one basket to another basket. The liquid that formed during fermentation will come out through the bottom and sides of the woven bamboo.



Figure 2. Traditional design of fermentation container (basket)

Storey Box

Storey box is a modification of single box [Fig 3.]. They are consisting of two or more wooden boxes in a fermentation box system. The box number one is placed on top and another on bottom. This type of box has been developed in used commonly at Kulonprogo for more than five years ago. It has capacity of 40 - 100 kg which is larger, the basket's capacity.

The advantages of this storey box are the farmers do not need separated boxes to reversal during fermentation. The storey box automatically has two boxes. Another advantage of storey box is the ease of reverse the cocoa bean. The reversal of cocoa beans is done by moving the cocoa beans from the top to the bottom, through the front side of the box. The cacao beans will be reversed automatically. Number of levels in the storey box is adjusted for the number of reversal of cocoa beans is done. The front side of storey box can be used as a door, which can be opened and closed, up and down, so it can be used as a way to move the cocoa beans from the top to the bottom of the box.

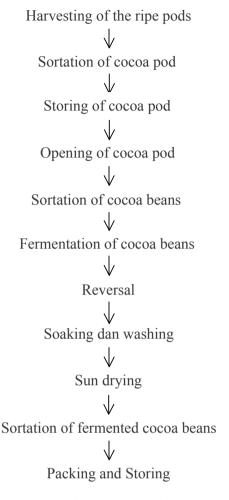


Figure 1. Flow diagram of cocoa bean fermentation

The dimensions of storey box as presented in Fig 3. Aeration of this box type passed through the hole. The holes have a diameter of 1 cm and the spacing between holes of 10 cm. There are nine holes in every sides of the box. This hole function as the place of turnover of air and as the discharge of waste products of fermentation. The disadvantages of the storey box is requires a lot of materials and space, but it provide more reversal, so the cocoa beans fermentation become complete.

Introduction box (Single box with a hand Crank)

Single box with a hand crank is a modification of the storey box [Fig 4.]. It has been introduced to targeted farmer in Kulonprogo. This box does not require a lot of material and space as a storey box. It has a capacity of 40 kg of cocoa beans. Cocoa bean's reversal technique is also different with storey box. Reversal of cocoa beans in this

box is done by turning the crank as much as 3 times. Every side of the box is also given hole. The holes have a diameter of 1 cm and the spacing between holes of 5 cm. There are 40 holes to every side of the box, because the distance of the holes are shorter.



Figure 3. Design of storey box



Figure 4. Design of single box with a hand crank

Temperature in The Fermentation Container

The fermentation process is divided into two phase, namely the first phase (0-48 h) and advance phase (48-120 h) [1]. In an exothermic reaction, microbial activity will lead to an increase in temperature in the fermentation box. Cocoa bean heap temperature increases, due to the heat that is formed of several reactions, namely (1) Sugar Ethanol + CO2 + 18 calorie, (2) Ethanol Acetate Acid + H2O + 235 calorie, (3) Acetate Acid H2O + CO2 + 419 calorie) [17].

The temperature rise in the fermentation process takes place periodically. In the first 12 hours, the temperature reached 25°-27°C. At that time there is a process of fermentation by yeasts pulp, converted into ethanol and organic acids. The second day, the temperature reached 32°-36°C. Ethanol and dead yeast converted into acid by bacteria. This process continues until the third day and thereafter in a temperature of 42°-48°C. On the third day and thereafter, the success factor of the fermentation process is air circulation well. Cocoa beans were converted into

lactic acid [anaerobic] or to acetate acid [aerobic] [4].

Temperature becomes important in the fermentation process. Observations of temperature on the fermentation box are used to determine the success of the fermentation. The fermentation process will go well, if at the end of fermentation temperature over 40° C and less than 50° C. Cocoa beans will die (not germinate) when the temperature over 40° C and will decompose when the temperature over 50° C. The temperature can be achieved when the piles of certain cocoa beans. Small scale fermentation (<100 kg) requires a pile of cocoa beans between 30-40 cm (Sime method - Cadbury). If the pile of cocoa beans more than 40 cm, the temperature of middle side is high. This causes air aeration is reduced so that the activities of organisms disrupted [12, 17].

This study collects the temperature data during the fermentation process takes place. Temperature of the fermentation container observed is the top, middle and bottom for each type of fermentation boxes.

Temperature in the basket

Fermentation of cocoa beans for three and five days in the basket produce temperature an average of top $(37^{\circ}\text{C} \text{ and } 36.47^{\circ}\text{C})$, middle $(38^{\circ}\text{C} \text{ and } 37.53^{\circ}\text{C})$ and bottom $(36.32^{\circ}\text{C} \text{ and } 35^{\circ}\text{C})$ [Fig 5.]. Basket middle part has a higher temperature than the top and bottom. The data shows that during the process of fermentation, the temperature in the basket never reached 40°C . This resulted in a fermentation of microbiological and enzymatic imperfect and the quality of cocoa beans will be lower. There are several causes of fermentation temperature can't be achieved, i.e. the thickness of cocoa beans in the basket and the basket aeration. Base area of the basket is too wide causing the expected piles of cocoa beans is not achieved (30-40 cm), so that the heat is reduced. Well fermentation can only take place at temperatures of more than 40° C and less than 50° C. The second cause is the basket aeration. Basket is a type of fermentation container that has many small holes. The holes are causing air circulation takes place very quickly, so the aerobic process (acetic acid) and anaerobic (lactic acid) inhibited. However, the use of baskets as fermentation container can be further optimized, i.e. put banana leaves on all sides except the bottom of the basket to limit the air circulation.

Temperature in the storey box

In the storey box shows a slight rise of temperature differences in the upper, middle and bottom. The top and middle of the box generate an average rise of temperature is quite stable, either by fermentation for three or five days (top: 42.33° C and 42.42° C) (the middle: 42° C and 44.17° C). In the storey box has an average temperature of more than 40° C, except at the bottom (39.17° C and 38.17° C) (Fig 5.). The bottom of the storey box has the lowest temperature due to melting pulp, so that the temperature decreases. The storey box produces a temperature rise higher than the basket. It showed that the fermentation process at the storey box takes place better than the basket.

Temperature in the introduction box (single box with a hand crank)

The temperature of a single box with a hand crank is not significantly different than storey box temperature. The temperature of the top, middle and bottom of this box has the same trend with the storey box temperature in three and five day fermentation (top: 42.17° C and 43.33° C) (the middle: 42.33° C and 43.53° C) (bottom: $37,67^{\circ}$ C and 39.33° C). Fermentation duration affects the temperature, the longer the fermentation duration,

the higher the temperature. In the fermentation process of cocoa bean, box capacity, aeration and reversal techniques in this box has produced the same temperature of the storey box. Temperature has a critical role in the success of the fermentation process. This introduction fermentation boxes can be recommended as an option fermentation container.

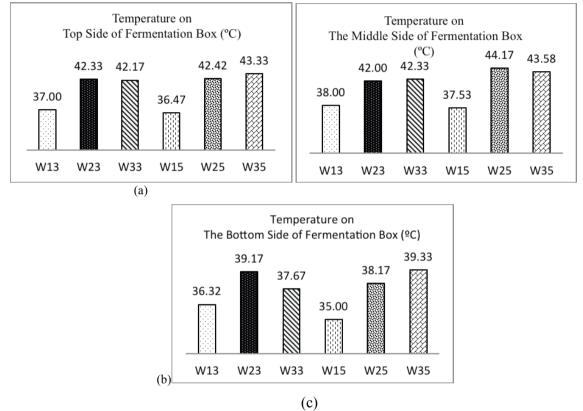


Figure 5. Temperature of fermentation box type and duration

Explanation:

W13: three days fermentation with basket, W23: three days fermentation with storey box, W33: three days fermentation with single box with a hand crank, W15: five days fermentation with the basket, W25: five days fermentation with storey box W35: five days fermentation with single box with a hand crank.

Moisture Content of Fermented Cocoa Bean

Moisture content is the amount of water contained in a material expressed in percent (%) [7]. Moisture content of fermented cocoa beans according to the general requirements of a maximum of 7.5% [2]. High moisture content causes a short shelf life and is vulnerable to pests and diseases. Moisture content above 8% is almost certain to fungi growth [5]. The critical point of moisture content suggested to prevent the proliferation of fungal and bacterial pathogens in fermented cocoa beans is 6-8% [13, 10].

The high temperature causes uneven drying. The outer part is dry and the inside still contains a lot of water [7]. The drying should not be too fast or too slow [9]. The drying of cocoa beans recommends two stages, namely the beginning and further drying. The beginning drying should be done immediately after the fermentation is complete (1-3 days) and under the sun [6]. It is necessary to achieve the required moisture content of 15% prior to the further process of drying. The initial drying could prevent fungal attack. The best way and cheap of drying is using a drying floor with a capacity of 15 kg per m2 for 7-10 days. As long the drying process, the reversal of cocoa beans is done at least every 1-2 hours. In the area with a high rainfall suggested using a mechanical dryer to achieve the requirements of the cocoa beans moisture content [9]. In this research, the farmers use the sun drying. Farmer drying technique is highly dependent on the weather. The drying time needs 7-10 days. The results showed that the moisture content of the fermented cocoa beans in each type of the fermentation container and duration meet the requirements of ISO 2323: 2008 (less than 7.5%). This moisture content can prevent fungal attack during storage.

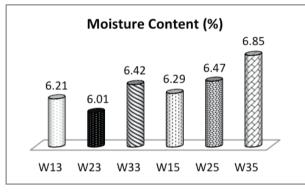


Figure 6. Moisture content of fermented cocoa bean

Explanation:

 W^{13} : three days fermentation with basket, W^{23} : three days fermentation with storey box, W^{33} : three days fermentation with single box with a hand crank, W^{15} : five days fermentation with the basket, W^{25} : five days fermentation with storey box W^{35} : five days fermentation with single box with a hand crank.

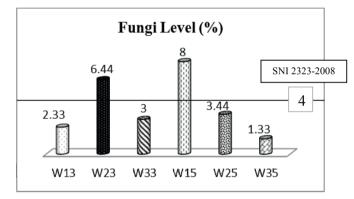
Fungi Level

Fermentation of cocoa beans is a complex process. This encourages the growth of undesirable microorganisms namely micotoxin [11]. Micotoxin can damage the flavor of cocoa [18]. There are four categories of fungi that contaminate food products, namely field fungi, storage fungi, invasive fungi and contaminant fungi [8]. This study focuses on invansive and contaminant fungi, the fungi that contaminate during processing and attack seeds. The imperfection fermentation or slow drying causes these type of fungus grow rapidly [18].

The factors that influence the fungi growth and aflatoxin production is a container that does not have good aeration, the influence of the atmosphere (CO₂ and O₂), temperature (25^{0} C is the optimum temperature to produce a toxin) and the influence of humidity (RH > 80% is ideal humidity of fermentation) [4]. Furthermore, the reversal also influences the quality of cocoa beans. The reversal aims to ferment cocoa beans evenly. It is usually done 2 or 3 times depending on the seed layer thickness. The reversal once will cause the fungi to grow on the top, thus causing slaty. Whereas, if excessive reversal will cause the fragile cocoa bean [12, 17].

Fungi level in cocoa beans can be detected quantitatively [laboratory analysis] and qualitatively. Qualitative detection can be done visually from the appearance of white color on the surface of the seed coat. Low fungi level, if the fungi only grow on the seed surface and can be removed by washing. While the fungi levels is severe, if the color white had entered into pieces of seed [nib] accompanied by a pungent odor [foul] and less savory [18]. In the cocoa trading, one of the provisions of cocoa bean is fungi level. A maximum limit is 3% fungi levels (grade I) and 4% (grade II) [10, 2].

The results of the analysis using one way ANOVA followed by DMRT, the fungi level in some fermentation box type and duration showed no significant difference. The storey box type in three days of fermentation and the basket in five days of fermentation have fungi level higher than the requirements of SNI 2323-2008 (> 4%). While introduction box (single box with a hand crank) with three and five days fermentation have met the requirements of SNI 2323-2008. Fungi level was produced under the threshold of SNI requirements (<4%).



Explanation:

W13: three days fermentation with basket, W23: three days fermentation with storey box, W33: three days fermentation with single box with a hand crank, W15: five days fermentation with the basket, W25: five days fermentation with storey box W35: five days fermentation with single box with a hand crank.

Fungi level on the basket fermentation

There are several causes of fungi level is high in the basket fermentation for five days. The basket fermentation with a hole that is very much inter-woven. The many hole in the basket cause greater aeration process and faster than a wooden box with a limited holes. The increase of temperature and acidity are very influenced by cocoa mass aeration [5]. pile capacity is smaller, the aeration is better. The pile capacity reduction affects the fermentation rate and uniformity of the speed of fermentation. However, the capacity of cocoa beans should not be less than 40 kg for each fermentation process [20].

In the fermentation using baskets, the circulation of air (aeration) takes place very quickly. Fermentation of cocoa beans for five days will be over fermented or otherwise, because the heat that is used to fermentation is not formed. Over fermented of cocoa bean can lead to the loss of the distinctive flavor of cocoa beans and the cocoa beans surface invaded by fungi [5]. While the basket capacity of less than 40 kg will inhibit the fermentation

process. The heat released during fermentation will disappear around a lot. The heat that is formed is not enough for fermentation, so that the temperature of the mass of cocoa beans cannot achieve the desired fermentation temperature [5]. The fermentation process that produces heat which is not enough causes the growth of fungi is rapid. The longer fermentation will increase fungal contaminant. Within five days, the levels of fungi increased more than three times higher than fermented cocoa beans for three days. Fermentation using a basket for three days will provides fungi level is lower, ranging 2.33% below the requirement threshold of ISO 2323-2008.

Fungi level on the storey box fermentation

On the storey box in 3 days fermentation produce high fungi level and exceeding the requirement SNI 2323-2008. However, the cause of the fungi on storey box and the basket is different. The capacity of storey box, the number and size of holes, and material have been meet the suggestion from the existing research. The thickness of container is not less than 3 cm and the fermentation capacity is not less than 40 kg [20]. This temperature sufficient for the purposes of the fermentation process. On the fermentation of cocoa bean that use the box, the depth or the maximum height of the box requires setting up.

One of the causes of fermented cocoa beans in the storey box for three days has high fungi level (6:44%) is the cocoa bean fermentation process is not perfect. The cocoa bean fermentation process takes place in two stages. At the beginning of fermentation, fermented cocoa beans within 0-48 hours, after that further fermentation within 48-120 hours. In further fermentation take place aerobic and anaerobic processes. The success of the fermentation process at this stage is highly dependent on air circulation smoothly. The large enough capacity (60 kg) with a total number of aeration holes 45 can not produce a smooth aeration. So the fermentation for three days still produces high fungi level. Meanwhile, the fermentation for five days had fungi level is lower (3.44%). The fermentation process has run perfectly. However, the aeration in storey box is considered still not smooth, because the fungi level is still quite high.

Fungi level on the introduction box fermentation (single box with a hand crank)

Fungi level in the fermentation that using single box with a hand crank for three and five days show to meet the requirements of SNI 2323-2008 [2]. However, five days fermentation gives a lower fungal level than three days fermentation. The advantages of this box, it has an efficiency of time and energy when reversal, moreover it has sufficient aeration than storey box. Single box with a hand crank has 40 aeration holes on each side, while the storey box only has 9 holes on each side. The number of holes on the sides of the box provides better aeration [16, 5]. High temperatures can be achieved at the end of fermentation.

Aeration mentioned above would be inefficient, if there is no reversal. Reversal in a single box with a hand crank more than the storey box. Reversal is done in two periods. Each period reversal, single box with a hand cranks turning a crank three times. So the total of reversal is six times. Single box with a hand crank to ferment cocoa beans is more perfect than the storey box or basket.

CONCLUSION

Each type of fermentation containers has different characteristics (material, capacity, number of aeration holes and the amount of the reversal). These characteristics affect the fermentation time and aeration. It determines the success of the fermentation process. One of the characteristics of successful fermentation is having low fungi levels (<3%). The introduction fermentation box (single box with a hand crank design) requires material and space more efficient, having better aeration and optimal temperature ($>40^{\circ}$ C) and produces the lowest fungi level (1.33%) in five days fermentation.

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