

# Proximate Analysis and Digestibility of Modified Corn Flour



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**Abstract: Introduction:** The quality of corn flour can improve through a modification process. The modification method that can be done to improve the characteristics of corn flour is through a controlled fermentation process accompanied by pregelatinization. This study aims to determine the effect modification of corn flour by spontaneous fermentation, controlled fermentation using *Lactobacillus fabifermentans* or *Aspergillus* sp culture and mixed culture of *L. fabifermentans* and *Aspergillus* sp (1:3) followed by pregelatinization at 80°C for 15 minutes.

**Methods:** The treatment variables in this study were spontaneous fermentation, controlled fermentation using single cultures of *L. fabifermentans* or *Aspergillus* sp., and mixed cultures of these microbes with a ratio of 1:3. This study consisted of 4 treatments with three groups each. This research is an experimental study using Randomized Block Design (RBD). Observation variables in this study were proximates tests (moisture content, ash, protein, fat, and carbohydrates) and starch digestibility. The data were processed using the SPSS version 22 program, with the method analysis of variance (ANOVA), and continued with Duncan's Multiple Range Test (DMRT).

**Results:** The results of variance analysis from spontaneous fermentation treatments, single culture fermentation of *L. fabifermentans* or *Aspergillus* sp, and mixed cultures of *L. fabifermentans* and *Aspergillus* sp (1:3) gave significant effect on moisture content, fat, carbohydrates, and starch digestibility of modified corn flour.

**Conclusion:** The best-modified corn flour was obtained by controlled fermentation treatment using *L. fabifermentans* followed by pregelatinization with characteristics of moisture content 14.46%, ash 0.13%, protein 7.41%, fat 0.65%, carbohydrates 80.06%, and starch digestibility 87.15%.

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## 1. INTRODUCTION

Corn (*Zea mays* L.) is a plant that has an important role in human life. Corn is the second food crop commodity after rice. One type of corn that is widely grown in Indonesia is a type of hybrid corn, including Bisi-18 corn. Bisi-18 corn has advantages that benefit farmers, namely the cob size which is large and very uniform. Currently, corn has been processed into various products ranging from semi-finished products to ready-to-eat processed products. One of the products produced from corn is corn flour whereas in corn flour there is starch which is the largest component of carbohydrates in the form of granules. Physically, starch can be distinguished from flour, including whiter and finer starch [1]. Maize production in Indonesia is very high so corn has the potential to be developed as a raw material for producing high-quality food products. The high content of carbohydrates and protein in corn plants makes this commodity very well processed into corn flour and becomes a local alternative resource to

replace wheat flour. Corn flour has a high carbohydrate (starch). Juniawati stated that [2] corn flour contains quite high starch, which is 60.07% with a composition of 22.88% amylose and 37.19% amylopectin. Corn flour has a lower fat content compared to wheat flour, but has a higher fiber content. The low fat in corn flour can make corn flour last longer because it doesn't go rancid easily due to fat oxidation. Therefore, corn flour can be used as a substitute for wheat flour in the manufacture of food products. Corn has good prospects to be developed as a food product and industrial raw material. The utilization of corn as an industrial raw material will provide added value to the commodity farming business. The use of corn starch as a basic ingredient in the manufacture of food products is still limited. This is caused by several problems related to retrogradation, low stability, and low paste resistance. In addition, corn starch also has disadvantages such as poor storage and flow rates, and cannot expand in cold water [3]. Modification of the process of processing corn into flour is one of the efforts to improve the natural characteristics of starch contained in corn. The modification process of corn flour that has been studied and re-

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ported in several scientific articles is a modification using fermentation and pregelatinization methods.

Modification of corn flour can also be initiated through a fermentation process which is then followed by the pregelatinization of starch. Fermentation is one of the processing engineering techniques by utilizing certain microorganisms in producing products. During the fermentation process, the decomposition of organic matter will occur due to the decomposition of complex compounds into simpler compounds by the microbes involved [4]. In addition, fermentation technology is also known to be a processing method that can improve the nutritional value characteristics of food ingredients to be of higher quality because the fermentation process affects the taste, aroma, texture, and shelf life of food. Fermentation method engineering is thought to play an important role in improving the characteristics of the resulting corn flour. Modification of corn flour by fermentation method can be carried out spontaneously or controlled by adding certain microbial cultures. Lactic acid bacteria are a group of bacteria that produce lactic acid as the main product of carbohydrate fermentation [5]. Sukainah *et al.* [4] succeeded in isolating *Lactobacillus fabifermentans* and *Aspergillus* sp cultures as indigenous microbes involved in the spontaneous fermentation process of Bisi-16 corn. In this study, after the fermentation process, the modification will be continued by pregelatinizing the starch. The starch pregelatinization process can improve the physicochemical characteristics of corn flour, especially the resistance and stability of corn starch to high-temperature processing.

Pregelatinization is a physical flour modification technique that uses the heating method, where the temperature used is above the range of starch pregelatinization [6]. The use of a single culture of *L. fabifermentans* in modifying corn flour has been reported by Angraini [7], the characteristics of the modified corn flour produced through this treatment are amylose content of 29.45% and starch content of 48.73%, while the characteristics of corn flour fermented by *Aspergillus* sp indigenous have high levels of amylose 26.45% and starch content of 45.22% [8]. The use of mixed cultures of *L. fabifermentans* and *Aspergillus* sp (1:3) in modifying corn flour has also been studied by Sukainah *et al.* [9], the characteristics of the resulting modified corn flour are 2.12% water content, liquid pH of 3.60, amylose content 17.42%, starch content 65.31%, viscosity 1825 cps with rheological properties belonging to the non-Newtonian category (pseudoplastic flow). The results of previous studies only reported the physicochemical characteristics of the modified corn flour produced. Studies on the content proximate quality and digestibility of starch-modified corn flour have not been carried out. Therefore, research on the characteristics of proximate quality and digestibility of modified corn flour, whether obtained from the spontaneous fermentation method or fermentation using a single culture of *L. fabifermentans* or *Aspergillus* sp and mixed cultures followed by the pregelatinization process needs to be studied. The purpose of this study was to examine the effect of the fermentation modification method (spontaneous and controlled using *L. fabifermentans* culture or *Aspergillus* sp culture and mixed culture

*L. fabifermentans* and *Aspergillus* sp (1:3) followed by pregelatinization treatment of the chemical content and digestibility of corn flour which generated

## 2. MATERIALS AND METHODS

### 2.1. Modified Corn Flour

This research was carried out in two stages; the preparation and implementation stages. The preparation stage consisted of making corn flours and rejuvenation of cultures, while the implementation stage involved making starters, applying the starters, and pregelatinization (heat-moisture treatments).

### 2.2. Preparation Stages

#### 2.2.1. Making Corn flour

Corn kernels were grated and soaked in water for 24 hr, till their pericarp and germ were separated, before draining. The kernels were then ground (disk mill; MDM-30; Andaro, Malang, Indonesia) and dried (dryer room; PT ATMI Kreasi Agro, Jakarta, Indonesia) at 50 – 60°C for 48 hr., before sieving with an 80-size mesh.

#### 2.2.2. Rejuvenation of *Aspergillus* sp.

Pure culture of *Aspergillus* sp., from spontaneous fermentation of Bisi-18 corn flour [10], was used. The culture was aseptically transferred, using the scratching method, and incubated (120 hr., 300C) in a potato dextrose agar (PDA).

#### 2.2.3. Rejuvenation of *L. Fabifermentans*

Pure culture of *L. fabifermentans*, from spontaneous fermentation of Bisi-18 corn flour [10], was used. The culture was aseptically transferred, using the injection method, and incubated (48 hr., 300C) in a De Man, Rogosa, and Sharpe broth (MRSB).

### 2.3. Implementation Stages

#### 2.3.1. Making *Aspergillus* sp. (AS) and *L. fabifermentans* (LF) Starters

Each incubated culture was diluted (1 mL culture; 10 mL water) before it was aseptically added to a sterile dispersion (1:2 w/v) of the corn flour and fermented microaerophilically for 48 hr.

#### 2.3.2. Application of Starters

Four treatments (*L. fabifermentans*, *Aspergillus* sp., mixed *L. fabifermentans* and *Aspergillus* sp 1:3) of the starters and spontaneous fermentation were investigated with each ratio being applied to the corn flour dispersion (1:2, w/v). The mixed-culture microaerophilic fermentation was for 24 hr., after which each fermentate was removed and dried (48 hr., 500C) before blending (food disc mill grinder IC-10B).

### 2.3.3. Pregelatinization

Water was added (70% w/v) to the fermented corn flour and steamed at 80°C for 15 min., before drying for 48 hr as before, blended and stored for analysis; proximate (moisture content, ash, protein, fat, and carbohydrates) [11] and starch digestibility testing [12].

### 2.4. Data Analysis

The data obtained were statistically analyzed using SPSS version 22. The requirements test used the normality test and homogeneity test. The results which showed that the research data were normally distributed and homogeneous were continued for analysis of variance (ANOVA). If the treatment test showed a significant effect between treatments, it was continued with the Duncan’s Multiple Range Test (DMRT) further tests with a 5% confidence level.

## 3. RESULTS AND DISCUSSION

### 3.1. Moisture Content

Moisture content is the amount of water contained in food. The moisture content of food determines its freshness of food and shelf life. In addition, high moisture content makes it easy for bacteria, molds, and yeast to multiply, causing changes in the food ingredients [13]. Moisture content shows the total amount of water contained in the material, either in the form of free water or bound water compared to the weight of the material. The moisture content of modified corn flour can be seen in Table 1 [14].

The moisture content of the modified corn flour obtained was in the range of 14.46%-16.57%. The highest moisture content of modified corn flour resulted from spontaneous fermentation followed by pregelatinization, at 16.57%. The results of DMRT analysis showed that the modified corn flour obtained from the fermentation treatment used the sin-

gle culture of *L. fabifermentans* and mixed culture of *L. fabifermentans*: *Aspergillus* sp (1:3) with continued pregelatinization produces lower moisture content than the spontaneous fermentation method. This is closely related to the concentration of *L. fabifermentans* culture used. The decrease in moisture content is due to evaporation of bound water. Before fermentation, some water molecules form hydrates with other molecules containing oxygen atoms, nitrogen, carbohydrates, proteins, salts, and other organic compounds, so water content is difficult to evaporate. During fermentation, microbial enzymes break down carbohydrates, so bound water turns into free water.

*L. fabifermentans*, which are indigenous bacteria with high concentrations when applied at the beginning of fermentation, make it easier for these bacteria to grow and reduce the adaptation time at the beginning of fermentation. Faster adaptation time causes the logarithmic growth time of *L. fabifermentans* culture to have a positive correlation with the metabolic processes. During the logarithmic growth phase, the metabolic rate will be significantly faster. Kumar *et al.* stated that [15] *Lactobacillus* can adapt well so that the metabolic processes that occur during fermentation will increase more quickly.

The fermentation process causes the change of complex compounds into simple compounds to become faster than the spontaneous fermentation treatment. One of the changes that occur is the hydrolysis of corn starch amylose into simple compounds by the amylase enzyme produced by *L. fabifermentans*. Songré-Ouattara *et al.* [16] stated that *L. fabifermentans* is an amylolytic lactic acid bacterium that can utilize starch as a substrate. Hydrolysis of amylose into simpler compounds by the amylase enzyme will be accompanied by a reaction to release chemically bound water into water that is easily released (physically bound water or free water). Drying at the same time and temperature, the water content of modified corn flour treated with *L. fabifermentans* culture,

Table 1. Proximate Analysis of Modified Corn Flour.

Parameter	Previous Research		Methods of Modified Corn Flour			
	Corn Flour (without Fermentation)	Spontaneous Fermentation	Spontaneous Fermentation Followed by Pregelatinization	Controlled Fermentation with Culture <i>L. Fabifermentans</i> Followed by Pregelatinization	Controlled Fermentation with Culture <i>Aspergillus</i> sp. Followed by Pregelatinization	Controlled Fermentation with Mixed Culture <i>L. Fabifermentans</i> : <i>Aspergillus</i> sp. (1:3) Followed by Pregelatinization
Moisture content (%)	16.19 ± 0.50	13.60 ± 0.19	16.57 ± 0.01 <sup>a</sup>	14.46 ± 1.31 <sup>b</sup>	15.28 ± 0.36 <sup>ab</sup>	14.87 ± 0.21 <sup>b</sup>
Ash (%)	0.25 ± 0.01	0.17 ± 0.01	0.13 ± 0.02	0.13 ± 0.03	0.12 ± 0.03	0.14 ± 0.02
Protein (%)	7.75 ± 0.09	9.73 ± 0.09	7.57 ± 0.22	7.41 ± 0.17	7.37 ± 0.03	7.45 ± 0.10
Fat (%)	0.32 ± 0.02	0.20 ± 0.02	0.71 ± 0.14 <sup>a</sup>	0.65 ± 0.13 <sup>ab</sup>	0.54 ± 0.10 <sup>b</sup>	0.34 ± 0.07 <sup>c</sup>
Carbohydrates (%)	71.10 ± 0.36	69.97 ± 0.36	79.20 ± 0.39 <sup>a</sup>	80.06 ± 0.41 <sup>b</sup>	79.97 ± 0.23 <sup>b</sup>	79.84 ± 0.22 <sup>b</sup>

Note: Remarks: mean values in a row followed by different letters (a, b, c, ab) are significantly different ( $p < 0.05$ ).

\*[14].

both single and mixed cultures, was the lowest because the moisture content of the material was evaporated and removed from the materials.

The higher concentration of *L. fabifermentans* in a given mixed culture, the greater the components of the material are broken down, resulting in large amounts of bound water being released and causing the texture of the material to become softer and more porous. This situation causes during the drying process, water evaporation becomes easier, thereby reducing the water content of flour [9].

*L. fabifermentans* is a type of lactic acid bacteria that produces lactic acid as its metabolite. *Lactobacillus* is a bacterium capable of producing lactic acid because this bacterium is a genus of Gram-positive bacteria that form lactic acid bacteria. Lactic acid produced by lactic acid bacteria during fermentation also affects the hydrolysis of complex compounds [17, 18]. Corn flour treated with *L. fabifermentans* had higher total acid than the spontaneous fermentation treatment. Hidayat [6] stated that the fermentation of *L. fabifermentans* culture produces higher lactic acid. In spontaneous fermentation, there are many other types of microbes that are active in the fermentation process so the total acid produced is lower. Lactic acid is known to also play an important role in the hydrolysis of complex compounds in foodstuffs into simple compounds, this hydrolysis reaction also causes a higher release of water so that it is easily removed during the drying process. The decrease in moisture content was caused by the fermentation process using *L. fabifermentans* culture which is capable of producing lactic acid so that during the fermentation process the temperature of the material will increase. With the formation of heat during the fermentation process, the temperature of the material will increase and the water produced during the fermentation process will evaporate resulting in a decrease in water content. So, it is suspected that the longer the fermentation, the heat as a result of metabolism increases and causes the water content to decrease [10].

Spontaneous fermentation treatment is fermentation in food where the manufacturing process does not add a starter, but indigenous microbes that play an active role in the fermentation process. During the spontaneous fermentation process, indigenous microbes also produce amylase and acid enzymes. However, during the spontaneous fermentation process, the types of microbes involved are very diverse with low concentrations. During the beginning of spontaneous fermentation, competition between types of microbes is also high, so the dominance of certain microbial species is also inhibited. Competition for microbial growth involved in the early spontaneous fermentation resulted in a slower adaptation and logarithmic growth phase of indigenous microbes during the fermentation process. The process of metabolism by microbes also runs slower, so the release of water formed during the hydrolysis process of complex compounds into simple compounds is also lower. Suprihatin stated [19] that spontaneous fermentation is a fermentation that utilizes indigenous microorganisms which are present in the slow fermentation process and experience adaptation and metabolic

processes that occur more slowly resulting in a high yield of moisture content.

The moisture content of modified corn flour ranges from 14.46%-16.57%. Winarno stated [20] that food products with a moisture content of less than 14% are safe enough to prevent mold growth. A food ingredient must have a low moisture content (moisture content below 10%) so that it can be stored for a long time. Richana *et al.* stated that [21] wheat flour which has a moisture content of 13 to 15% has a shelf life of one year.

### 3.2. Ash

Ash content indicates the amount of mineral content contained in food. The amount of ash content in food products depends on the amount of mineral content of the ingredients used. A comparison of ash on modified corn flour can be seen in Table 1. The results of the research on the ash content of modified corn flour, both the spontaneous and controlled fermentation treatments were in the range of 0.12% - 0.14%, this value range still met the Indonesian National Standard (SNI) for corn flour ash content, namely, 1.5%.

Modification of corn flour had a similar ash content, which was in the range of 0.12%- 0.14% (DMRT analysis). The treatment of the four types of flour was lower than that of Aini *et al.* [5], namely with a value of 0.27%. The low level of the four types of corn flour is due to the release of minerals during the fermentation process during soaking. Ash is an inorganic residue from the process of burning or organic oxidation of foodstuffs. Total ash content is part of the proximate analysis which aims to evaluate the nutritional value of a food product, especially the total minerals. The ash content of a food ingredient shows the total minerals contained in the food.

### 3.3. Protein

Protein is a high molecular weight complex organic compound which is a polymer of amino acid monomers linked to each other by peptide bonds. The results showed that the protein content of modified corn flour was in the range of 7.37%-7.57% (Table 1). Based on the results of research on the modification of corn flour with various treatments (spontaneous fermentation, single culture fermentation of *L. fabifermentans* or *Aspergillus* sp, and mixed fermentation of the two microbes namely *L. fabifermentans* and *Aspergillus* sp (1:3) followed by pregelatinization) has an average index value almost the same average in the range of 7.37%-7.57% (DMRT analysis).

The protein content of modified corn flour produced in this study was higher than the protein content of corn flour reported by Richana *et al.* [21] namely 5.07-6.84%; however, lower if compared with the protein content of corn flour reported by Aini *et al.* [5], namely 8.7-10%. The protein content of modified corn flour produced in this study had almost the same protein content for all treatments. The microbes involved during the fermentation process can produce protease enzymes so that dissolved proteins are also reduced during the fermentation process. Michodjehoun *et al.* [22] that

protease enzymes break down peptide bonds to produce amino acids. The release of protein content bonds is also due to the role of bacteria which are proteolytic and will degrade proteins during the fermentation process. The longer the fermentation process lasts, the more opportunities for bacteria to degrade protein so that the protein content is released. The modified corn flour produced in this study has a protein content that is almost equivalent to low protein flour, namely 8 to 11%, so it can be applied to several food products that require wheat flour with low protein content and does not experience swelling.

### 3.4. Fat

The fat content of the modified corn flour obtained is shown in Table 1. The results showed differences in the fat content of corn flour. Modified corn flour obtained from the controlled fermentation method using a mixed culture of *L. fabifermentans* and *Aspergillus* sp (1:3) followed by pregelatinization produced the lowest fat content, that is 0.34%. The highest fat content of modified corn flour was produced by spontaneous fermentation followed by pregelatinization, that is 0.71%.

The results of DMRT analysis show that corn flour modified by controlled fermentation using a mixed culture of *L. fabifermentans* and *Aspergillus* sp (1:3) followed by pregelatinization produced the lowest fat content. The cultures of *L. fabifermentans* and *Aspergillus* sp were able to produce fat-breaking enzymes, but the culture that was thought to play the most role in reducing the fat content of corn flour was the culture of *Aspergillus* sp. This can be seen in the fat content of the modified corn flour treatment using a single culture of *Aspergillus* sp. The resulting value of the corn flour fat content is lower than that of the spontaneous fermentation (control) corn flour treatment. *Aspergillus* sp. culture can produce fat-breaking enzymes, namely lipase. The lipase enzyme has the function of breaking down fat into simpler compounds such as fatty acids and glycerol. This is supported by Koswara [23] who stated that the more concentration of *Aspergillus* sp. in corn flour, the more fat is broken down. Winarno [20] that lipase is a group of hydrolytic enzymes that can catalyze the process of hydrolysis or the breakdown of fats into their constituent fatty acids.

The highest fat content was obtained from corn flour which was modified by spontaneous fermentation followed by the pregelatinization process, that is 0.71%. This is because during the spontaneous fermentation process, the growth of indigenous molds, including *Aspergillus* sp. which produces lipase, was only found to grow dominantly at the beginning of the fermentation up to 6 hours of fermentation. After the spontaneous fermentation process enters the logarithmic phase, the dominant microbial growth involved in the spontaneous fermentation process comes from yeast and bacteria groups. Therefore, the use of *Aspergillus* in controlled fermentation is very possible because *Aspergillus* can produce the enzymes endoglucanase, exoglucanase, and  $\beta$ -glycosidase. Thus, these enzymes have an important role in the breakdown of starch into simpler ones so that the struc-

ture of corn starch will change, which will affect the physicochemical properties of corn flour [4].

Fat is a food substance that is important for maintaining human health. In addition, fat is also a more effective source of energy compared to carbohydrates and protein [20]. The low-fat content also determines the shelf life of these food-stuffs, the parts of corn that contain fat content are endosperm 0.8%, seed 33.2%, skin 1.0%, and tip cap 5.3%. In this study, the part of corn used to make corn flour was the endosperm, pericarp, or corn husk so the modified corn flour fat content was minimal. The fat content of the four types of modified corn flour produced in this study was lower than the fat content of corn flour reported in the study by Richana *et al.* [21], that is 1.42%. The fat content of modified corn flour produced in this study was low. Before the corn kernels are floured, the corn kernels are milled first, that is, the corn rice is separated from the corn kernels and the pericarp which contains high fat and fiber. This separation aims to lower the fat content of the modified corn flour and to have a longer shelf life because the modified corn flour does not go rancid during storage. Kulp *et al.* stated that [24] the fat content of corn kernels reaches 32-35%. The low-fat content in corn flour causes during the long-term storage process, corn flour will not go rancid easily due to the oxidation of fatty acids.

### 3.5. Carbohydrates

Carbohydrates have an important role in determining the characteristics of food ingredients, such as taste, color, texture, and others. The results showed that the carbohydrate content of modified corn flour was in the range of 79.20% - 80.06% (Table 1). The lowest carbohydrate content was shown in the modified spontaneous fermentation and pregelatinized corn flour treatment as a control with a value of 79.20%.

The results of the DMRT analysis showed that modified corn flour using controlled fermentation has the highest carbohydrate content compared to modified corn flour using spontaneous fermentation. During the controlled fermentation process, the bacterial culture of *L. fabifermentans* or the culture of *Aspergillus* sp. was allegedly more modifying the starch content of corn flour by the amylase enzyme produced. However, these two cultures were considered less than optimal in breaking down or modifying the dietary fiber content in corn flour, so the carbohydrate content of these treatments was higher than that of spontaneous fermentation. *L. fabifermentans* and *Aspergillus* sp. is a microbe that is capable of producing amylase, so during the fermentation process, the amylose content in corn flour is broken down into simpler compounds.

Lactic Acid Bacteria (LAB) are a group of bacteria capable of converting carbohydrates (glucose) into lactic acid by fermenting carbohydrates. LAB that can utilize starch as a substrate is known as amylase-producing LAB. *Lactobacillus* that can produce amylase enzymes can break down the amylose content into simpler compounds and can utilize starch as a substrate [25]. Alpha amylase enzyme is an en-

zyme that hydrolyzes linear  $\alpha$ -1,4 glycosidic bonds in amylose randomly to produce a mixture of dextrin, maltose, and glucose.

Modification of corn flour with spontaneous fermentation treatment resulted in the lowest carbohydrate content. During the spontaneous fermentation process, the indigenous microbes involved are very diverse. Indigenous microbes involved during the corn flour fermentation process are thought to not only modify the starch component in corn flour but also to overhaul or modify the dietary fiber components available in corn flour. Generally, during the spontaneous fermentation process, the indigenous microbes involved are very diverse, including indigenous microbes that are capable of producing food fiber-breaking enzymes such as cellulase, hemicellulase, and pectin. Suprihatin stated [19] that the microorganisms involved in the spontaneous fermentation process are capable of producing cellulase during fermentation which is found in foodstuffs. The cellulase enzyme is a protein found in living cells that functions as a catalyst in biochemical reactions so that the enzyme has specific properties to hydrolyze the  $\alpha$  1-4 glucoside bonds of cellulose to produce cellobiose which is then converted into glucose monomers [26]. The growing microbes are capable of producing pectinolytic and cellulolytic enzymes that can destroy cell walls, resulting in the release of starch granules. This will also cause changes in the characteristics of the flour produced, including a decrease in the total carbohydrate content of the modified corn flour produced.

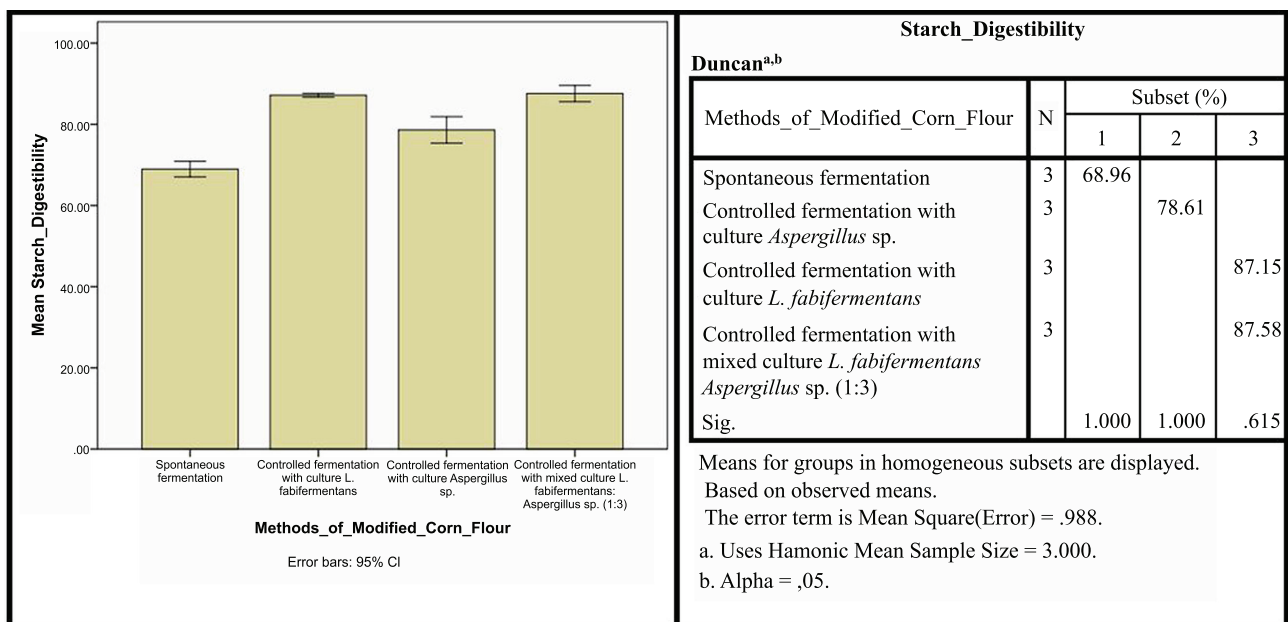
The carbohydrate content test conducted in this study was by a different method. The modified corn flour carbohydrate content obtained in this study was in the range of 79.20%-80.06%. Carbohydrate levels calculated differently are influenced by other nutritional components, the lower the other nutritional components, the higher the carbohydrate content. *Vice versa*, the higher the other nutritional compo-

nents, the lower the carbohydrate content. Nutritional components that affect the amount of carbohydrate content include protein, fat, water, and ash content.

### 3.6. Starch Digestibility

Starch digestibility is the ease with which a type of starch can be hydrolyzed by enzymes that break down starch into simpler units. Factors that can affect the low digestibility of starch are the presence of anti-nutrients and anti-amylose (dietary fiber and tannins) and the chemical structure of starch [27]. The results of the study of starch digestibility of corn flour are presented in Fig. (1). The digestibility of modified corn flour starch was different for each treatment. The digestibility of modified corn flour starch was in the range of 68.96%-87.58%. The lowest digestibility of starch was shown in the treatment of modified corn flour using the spontaneous fermentation method and pregelatinization as a control with a value of 68.96%.

Based on the results of the DMRT analysis test on starch digestibility parameters, modified corn flour using *L. fabifermentans* single-culture fermentation and *L. fabifermentans* and *Aspergillus* sp mixed cultures produced corn flour with higher starch digestibility. The culture of *L. fabifermentans* belongs to the amylolytic lactic acid bacteria, which are bacteria capable of producing  $\alpha$  amylase and lactic acid enzymes during fermentation. *Aspergillus* sp culture is classified as capable of producing  $\alpha$  amylase enzymes, these bacteria can produce and isolate glucoamylase enzymes from *Aspergillus oryzae* [28]. The  $\alpha$ -amylase enzyme can hydrolyze the amylose component in modified corn flour starch into simpler compounds so that the digestibility of the resulting corn flour starch increases. The amylase and pullulanase enzymes produced by lactic acid bacteria cultures cause changes in amylose into short-chain amylose, oligosaccharides, maltose, maltotriose, and glucose which are easier to



**Fig. (1).** Starch digestibility of modified corn flour.

digest with a higher glycemic index. Herawati stated that [29] the hydrolysis of the  $\alpha$ -amylase enzyme goes through two stages. The first stage is the degradation of amylose into maltose and maltotriose which occurs randomly. The next stage, namely the formation of maltose, as a result, is not random and runs more slowly. Lactic acid which is also produced by *L. fabifermentans* can also affect the digestibility of modified corn flour starch. Lactic acid is also able to hydrolyze amylose into simpler compounds, so that the digestibility of corn starch modified by *L. fabifermentans* treatment, both single culture and mixed culture, will increase. *L. fabifermentans* bacteria are lactic acid which is amylolytic in nature, namely lactic acid bacteria that can hydrolyze amylose into simpler ones.

Starch digestibility is also affected by the composition of amylose and amylopectin. Amylose is more slowly digested than amylopectin because amylose is a polymer of simple sugars with straight, unbranched chains. This straight chain forms a solid amylose bond so it is not easily gelatinized. Therefore, amylose is more difficult to digest than amylopectin which is a simple, branched, and open-structured sugar polymer [30]. Amylopectin has a branched structure, where the branch points are connected by  $\alpha$ -1,6 glycosidic bonds. Karim *et al.* [31] reported that the number of  $\alpha$ -D-glucose that makes up the branching point in amylopectin is 20-30 anhydroglucose units. Amylopectin has a higher molecular weight when compared to amylose, which is around  $10^6$ - $10^9$ , which causes amylopectin to have a wider surface area per molecule so that it is more easily broken down by  $\alpha$ -amylase enzymes into simpler forms [32].

The starch digestibility of modified corn flour using spontaneous fermentation treatment was lower than that of modified corn flour with other treatments. The low digestibility of modified corn flour starch produced by spontaneous fermentation treatment is caused by the amylase enzyme produced by indigenous microorganisms that are not optimal so that not much amylose is converted into a simpler structure. Starch digestibility is the ease with which a type of starch can be hydrolyzed by enzymes that break down starch into simpler units. Herawati stated that [29] starch with lower digestibility, the less starch is hydrolyzed at a certain time. The main cause of the amylase enzyme produced not working optimally during the spontaneous fermentation process is thought to be due to competition for the growth of indigenous microbes involved during the fermentation process. This competition causes indigenous microbes that are amylolytic not to grow optimally so the amylase enzyme produced is also not optimal. Spontaneous fermentation has a weakness, namely the microbes that grow in the fermentation process vary so that they are less able to produce enzymes during the fermentation process. The digestibility of starch-modified corn flour by spontaneous fermentation followed by pregelatinization was also the lowest compared to the other treatments. This is related to the carbohydrate content of modified corn flour which has the lowest value compared to other treatments. Several factors can reduce starch digestibility, namely anti-nutrition and anti-amylase, and high re-

sistant starch content. Modified starch has lower digestibility because it may contain higher resistant starch [27].

## CONCLUSION

Modification of corn flour using spontaneous and controlled fermentation methods, controlled fermentation using a single culture of *Lactobacillus fabifermentans*, *Aspergillus* sp., or mixed cultures of *L. fabifermentans* and *Aspergillus* sp followed by pregelatinization affected moisture content, fat, carbohydrates, and starch digestibility. However, this modified treatment did not affect the ash and protein content of corn flour. The treatment of *L. fabifermentans* produced corn flour with the best physicochemical properties and starch digestibility with 14.46% water content, 0.13% ash, 7.41% protein, 0.65% fat, 80.06% carbohydrates, and 87.15% starch digestibility.

## LIST OF ABBREVIATIONS

ANOVA	=	Analysis of Variance
DMRT	=	Duncan's Multiple Range Test
PDA	=	Potato Dextrose Agar
MRSB	=	De Man, Rogosa, and Sharpe broth
LAB	=	Lactic Acid Bacteria

## ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Not applicable.

## HUMAN AND ANIMAL RIGHTS

Not applicable.

## CONSENT FOR PUBLICATION

Not applicable.

## AVAILABILITY OF DATA AND MATERIALS

Not applicable.

## FUNDING

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## CONFLICT OF INTEREST

The authors declare no conflicts of interest, financial or otherwise.

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