Eco. Env. & Cons. 24 (1) : 2018; pp. (132-139)
Copyright@ EM International
ISSN 0971–765X

Identification and isolation of fungi indigenus on spontaneous fermentation corn flour Bisi 18

Andi Sukainah[1], Eva Johannes[2] and Reski Praja Putra[3]

[1,3]Faculty of Engineering, Universitas Negeri Makassar, Indonesia
[2]Faculty of Mathematics and Science, Universitas Hasanuddin

(Received 10 September, 2017; accepted 30 October, 2017)

ABSTRACT

Modification of corn flour using spontaneous fermentation methods were deemed to be flawed in that type of microbe that lives can be varied and difficult to control. The fermentation process needs to be controlled so that the resulting higher quality flour, one of the efforts is the utilization of microbial isolates indigenus as starter cultures. This study aims to determine the microbes involved in spontaneous fermentation of corn as well as the isolation and identification of fungi indigenus. Corn milling was added water that has been cooked in the ratio 1: 2 (b / v) spontaneously fermented in a fermentation tank clean for 48 hours in microaerophilic.The calculation of the number of microbes performed to determine the indigenus microbes (fungi, yeasts, bacteria and lactic acid bacteria) involved during the process of spontaneous fermentation using media PCA, PDA, NA, and MRSA. In addition, the total acid titration (%) and the pH of the fermentation liquid was also measured. Calculations and analysis was conducted on fermentation time interval of 0, 3, 6, 24, 27, 30, and 48 hours.The results showed that at the beginning of fermentation, which is 0 to 3 hours (adaptation phase), a microbe that was instrumental is mold, after fermentation entered the exponential phase and stationary, i.e. 6 to 48 hours, the fermentation process is dominated by the growth of yeasts, bacteria, and bacteria lactic acid which causes increased levels of total acid titration until 0.38%and the pH value becomes 3.970. In this phase, the mold growth decreased. The result of the isolation and identification of fungi using slide culture techniques are found 7 species of mold that are involved in the process of spontaneous fermentation of corn Bisi 18, namely *Aspergillus fumigatus*, *A. flavus*, 3 types of classified *Aspergillus*sp, C*unninghamella elegans*, dan *Dendryphio psisatra.*

***Key words** : Corn Bisi 18, Spontaneous fermentation, Molds, Corn flour, Aspergillus*

Introduction

Corn is one crop that has the prospect to be developed in all regions of Indonesia. One type of corn that is grown in Indonesia is a type of hybrid maize. Corn Bisi-18 is a derivative of corn hybrids Bisi-2 and have physicochemical characteristics that resemble corn hybrids Bisi-2 is its ability to adapt to the environment so it can be grown easily in all regions of Indonesia, ranging from rice fields in Java to the dry plains in East Nusa Tenggara. Until now, the use of corn into food products is still lacking. Conditions were hard corn seed to form large seeds require a longer processing time. Therefore, required processing into maize flour products. Compared with corn shaped grains, corn flour will be more easily applied to food products, despite the application corn flour is highly dependent on the physicochemical properties.

The physicochemical properties is one of the properties related to viscosity and gelatinization of corn starch during the heating process, the peak vis-

***Corresponding author's email:** andisukainah@yahoo.com

cosity and hot paste viscosity, where this parameter describes the ability of granule undergo maximum development upon heating (Zhang *et al.,* 2013). *Breakdown viscosity*or changes in the hot pasta is physico chemical properties that describe the resistance of a granule of the process of heating and mechanical treatment during processing. Parameter cold paste viscosity and setback is physico chemical properties that describe the ability granules do retrogradation during cooling (Alvanidkk *et al.,* 2012). In addition, the starch gelatinization profile is also an important physicochemical properties that describe the initial temperature, peak temperature, and the final temperature gelatinization, as well as the value of the enthalpy which reflects the degree of crystallization of a starch.

Corn flour as a natural starch still has a viscosity gel that is not uniform, is not resistant to high temperatures, is not resistant to acidic conditions, can not stand the mechanical treatment, have limited solubility, and still prone to siniresis. This led to the application corn flour into food products is still very limited, so it required an effort to modify the corn flour that is expected to modify the physical properties corn flour so that the potential applications become larger.

Modification of flour with fermentedis very potential because low operational costs. The fermentation process is defined as the decomposition of starch by enzymes produced by the microorganisms on a substrate. Sukainah (2014) research three types of corn flour (corn Bisi-2, pop) are modified using spontaneous fermentation, pregelatinisasi and the combination of spontaneous fermentation and pregelatinisasi show preferential treatment to fermentation capable of lowering the value of peak viscosity, reverse viscosity, the initial temperature and the peak temperature and enthalpy value of corn flour. However, modification of corn flour using spontaneous fermentation method has the disadvantage that it is still kind of living microbes can vary and depends on the conditions and environment so it is difficult to control. The fermentation process needs to be controlled to improve the quality of modified corn starch produced by utilizing isolates indigenus as starter cultures. Therefore, this study will focus on the observation of microbial strains involved in the fermentation of corn flour spontaneously as well as the isolation and identification of microbial indigenus, particularly the identification of types of mold that are involved in the process of spontaneous fermentation of corn.

Materials and Tool

The main materials used are Bisi-18 corn hybrids derived from Jeneponto. Microbial growth medium used was PCA media Oxoid Basingstoke Hampshire England, media NA Merck Darmstadt Germany, media PDA Oxoid Basingstoke Hampshire England, and media MRSA Oxoid Basingstoke Hampshire England. The materials used for the analysis is water and distilled water, NaOH 0.1 N pa, PP indicator, iodine solution, filter paper, paper filter "Whatman", oil immersion, solution of crystal violet, 70% alcohol, 95% alcohol, spirits, cotton, aluminum foil, and a solution of safranin.

Processing equipment used is discmill machine blender type 9FZ -23 and corn seed milling machine type PPK N 70. Equipment analysis covering, test tube, tube rack vortek, hot plate, beaker glass, flask, clamps, analytical balance, measuring cups, volume pipette, a pipette, pH meter, water bath, Erlenmeyer, petri dishes, incubator, autoclave, burette, microscopes and glass objects

Research Methods

Equipment analysis covering, test tube, tube rack vortek, hot plate, glass beaker, flask, clamps, analytical balance, measuring cups, volume pipette, a pipette, pH meter, water bath, Erlenmeyer, petri dishes, incubator, autoclave, burette, microscopes and glass objects.

First Stage: Assessment of Microbial indigenus Involved during Spontaneous Fermentation

Milling the Seed Corn

Bisi-18 corn seed that have been cleaned of impurities and defects seeds soaked for 1 hour at room temperature with a ratio of corn with the amount of water 1: 2 (b/v). Furthermore, the milling corn seed using milling machine type PPK N 70.

Indigenous microbial Involved in Rice Corn basis Spontaneous Fermentation

Rice corn added water that has been cooked in the ratio 1: 2 (b/v) spontaneously fermented in a fermentation tank clean for 48 hours using microaerophilic. During fermentation calculation of the amount of microbes to determine the microbial indigenus involved during the process of spontaneous fermentation, total acid titration (%) and pH.

Calculations and analysis was conducted on fermentation time interval of 0, 3, 6, 24, 27, 30 and 48 hours. Microbial count (total plate count) were grown using media PCA (CFU/mL), the number of bacteria using media NA (CFU/mL), the number of fungi and yeasts using a PDA (CFU/mL), and the amount of lactic acid bacteria using media MRSA (CFU/mL).

Total Microbial, total bacteria, total fungi and yeasts, and total lactic acid bacteria during fermentation was measured by the method of fertilization on media PCA (calculation of the total microbial or total plate count), PDA (calculation of the total fungi yeasts), media MRSA (counting total bacteria lactic acid), and media NA (counting total bacteria). A total of 10 mL of liquid sample is inserted in a physiological solution NaCl 0.85% 90 mL and divortex for dilution 10^{-1}. Dilution is done to10^{-3} and 10^{-9} in the same way. Fertilization is done in duplicate at dilutions 10^{2}-10^{-3} and 10^{-7}-10^{-8} using PCA, PDA, MRSA, and NA in a petri dish. Petri dishes were incubated at 37 ^{o}C in an inverted position. Colony counts for total lactic acid bacteria and total bacteria were performed after 48 hours based on ISO methods in (CFU/mL).

Second Stage: Isolation and Identification Kapangindigenus

Fungi that grows on PCA and PDA media were isolated using the technique of scratching quadrant. Having obtained isolates with separate colony to be identified using a microscope with a slide culture techniques. Petri dish was given of filter paper so that the board pad inside a closed cup. U-shaped rod placed in it, and above was placed alongside a glass object with a glass cover. Grail sterilized in an autoclave. Once cool, glass objects by a drop of sterile media that has been thawed. Furthermore, the media leveled using a sterile glass rod to form a thin layer with an area not exceeding extensive cover glass and allowed to solidify in a closed cup. After a solid medium, the agar surface inoculated with a bit of mold spores using a needle ose, then covered with a sterile cover glasses. Glycerol 10% sterile as much as 5-7 mL dripped on the paper filter to provide the optimum moisture for fungi growth. Incu-

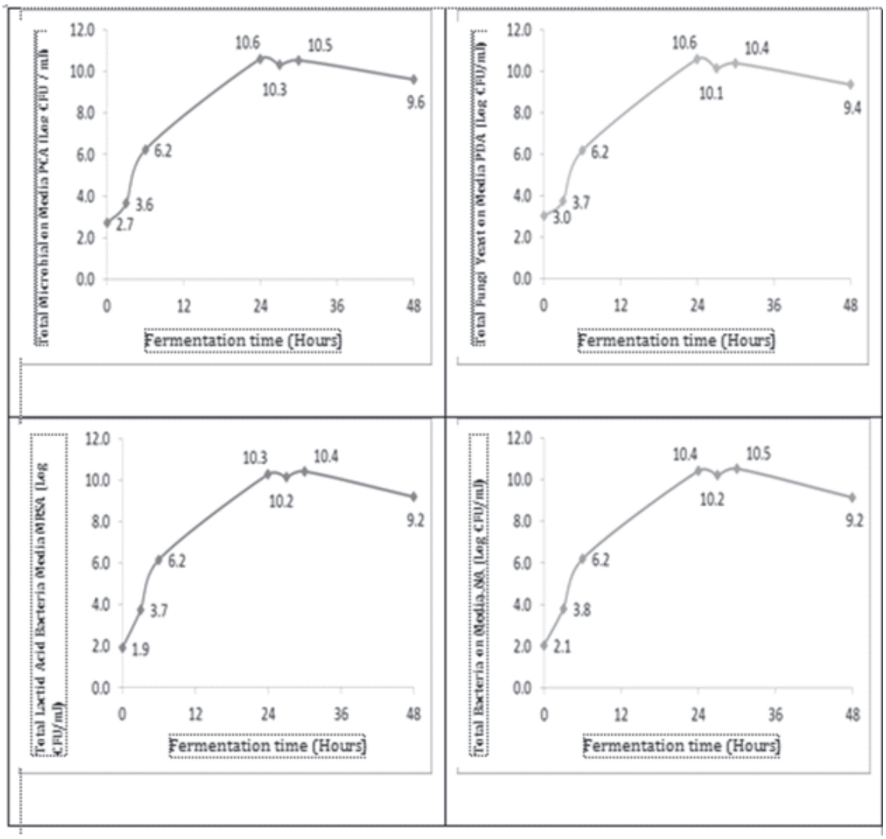


Fig. 1. Microbial Biomass indigenus Involved during Spontaneous Fermentation of Corn Starch in Different Media

bation was performed at room temperature for 3-5 days. Then the fungi structure was observed under a microscope.

Data processing was performed using Descriptive Analysis. The data were processed using SPSS 17.0 software.

Results and Discussion

Microbes indigenus Involved during Spontaneous Fermentation of Corn Bisi-18

Changes in the number of microbesindigenus during spontaneous fermentation of corn Bisi 18 is presented in Figure 1. At the beginning of fermentation, produced the highest number of microbes on PDA with the total amount of yeast fungi, i.e. 3.0 log CFU /mL and produced the lowest number of microbes on the media by the number of MRSA bacteria lactic acid 1.9 log CFU/mL. These results indicate that the initial fermentation, molds and yeasts was instrumental in the process of fermentation.The role of the mold during the initial process of fermentation is suspected as the microbes that contribute to elaborate a complex carbohydrate compound from corn Bisi-18 into simpler compounds, so that the compound can be utilized by other indigenus microbes for growth. In the adaptation phase, ie 0 to 3 hours, mold growth have increased, but the growth is decreased when the fermentation process enters the exponential phase, which is 6 to 24 hours, and the stationary phase, which is 24 to 48 hours.

Growth of total microbial mesophilic (PCA), yeast (PDA), total bacteria (NA), and total lactic acid bacteria (MRSA) has increased in the fermentation 6 hours until the end of fermentation, 48 hours. The number of microbes that are involved during the process of spontaneous fermentation of corn Bisi 18 at the end of fermentation ranges from ± 9.2-9.6 log CFU/mL. Microbial growth trends of the three types of microbial growth media (PDA, NA, and MRSA) used has similarities with trend growth of microbes on media PCA. These results indicate that the yeasts, bacteria and lactic acid bacteria involved in the fermentation process ismesophilic. Mesophilic microorganisms is a microorganism that has an optimum growth temperature between 20-40 pC (Fardiaz, 1992).

Levels of Total Acid titration

Levels of total acid titration corn liquor Bisi 18 during spontaneous fermentation increased (Figure 2). Total acid fermentation began to increase at 6 hours until the end of fermentation, ie 0.04 to 0.38%. At the beginning of fermentation, which is 0 to 3 hours, total acid has not increased. This is due to the amount of lactic acid bacteria is only around 1.9-3.7 log CFU /mL (adaptation phase).

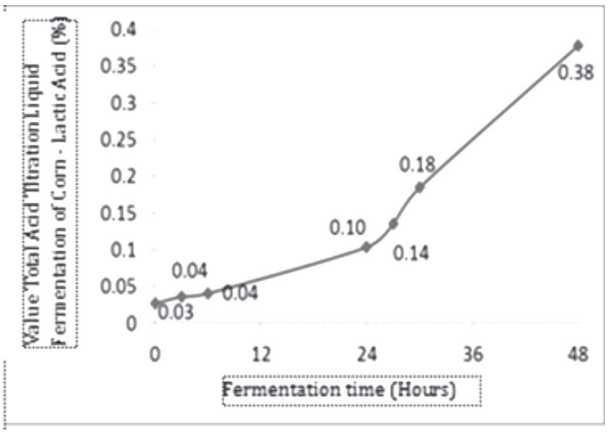


Fig. 2. Levels of Total Acid Titration Liquid Corn Bisi 18 during Spontaneous Fermentation

The increase in total acid fermentation began to increase at 24 hours, which is 0:10%. Exponential phase of lactic acid bacteria fermentation started 6 hours. The growth of lactic acid bacteria fermentation increased significantly after 6 hours, ie from 6.2 to 10.3 log CFU/mL, causing levels of total tertitrasi acid also increased. Lactic acid bacteria are bacteria that produce lactic acid as a major metabolite product. According to Axelsson (2004) metabolism of lactic acid bacteria during the fermentation result in a change of carbohydrates (simple sugars) into the substrate phosphorylation. This group of bacteria

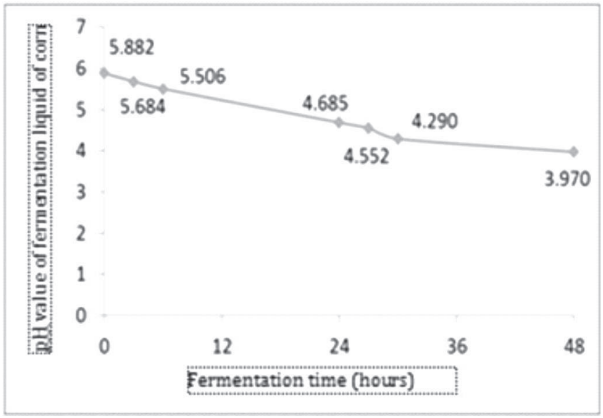


Fig. 3. Changes in pH Value Fluid of Corn Bisi 18 during Spontaneous Fermentation

has the capacity to degrade carbohydrates (simple sugars), the metabolism of sugar depending on the substrate available, but generally the final product is lactic acid.

The pH Value

The pH value (acidity) of corn liquor Bisi 18 decreased during spontaneous fermentation process (Fig. 3). At the beginning of the fermentation, the liquid corn Bisi 18 had a pH of 5.882. The pH value has decreased significantly after 6 hours of fermentation, the pH value of 5.506 into 4.685. A decrease in fluid pH decreased until the end of fermentation.

A decrease in the pH value was positively correlated with increased levels of total tertitrasi acid which is the major metabolic product produced by lactic acid bacteria during fermentation of corn spontaneous Bisi 18. In addition, the fermentation time also affects the pH value. The longer the fermentation time, more and more number of microorganisms that will grow to produce acid, so that the pH value will decrease with increasing concentration of dissolved acid (Silins, 2014; Arroyo-López, 2009)

Isolation and Identification of Microorganisms Growing on Fermentation of Corn Bisi 18

The results of microbial isolation indigenus involved in the process of spontaneous fermentation of corn Bisi 18 can be seen in Table 1. Microbial indigenus involved during spontaneous fermentation of corn Bisi 18 consists of molds, yeasts, bacteria and lactic acid bacteria. The results of microbial isolation on PDA showed at the beginning of fermentation up to 6 hours of fermentation found 7 isolates of mold that are further identified using a slide culture techniques.

Results of simple identification of bacterial isolates from media NA and MRSA in the form of the Gram stain, catalase test, and staining endospores showed the bacteria involved in the spontaneous fermentation of corn Bisi 18 not only from the group of lactic acid bacteria, but the bacteria in addition to the group of lactic acid bacteria are also involved (test data not shown). Isolates of yeasts, bacteria and lactic acid bacteria that have different colony morphology are found in fermented 24-48 hours. This shows the fermentation hours 24 to 48 hours (final

Table 1. Isolation of microorganisms on Spontaneous Fermentation of Corn Bisi 18

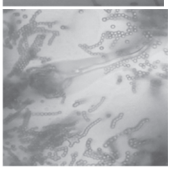
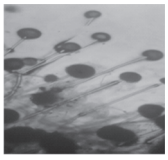
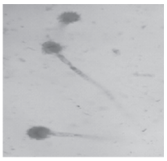
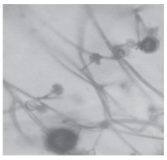
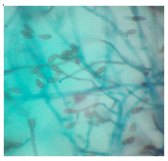
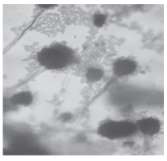
Isolates code	Media type	Hour to -	Characteristics colonies	Microorganisms type
A	PDA	0	Misty green colonies	Fungi
B		0	Dark green colonies	Fungi
C		3	White colonies	Fungi
D		3	Cream-colored colonies with white mycelia	Fungi
E		3	Colonies of dark gray with a texture resembling mounting cotton	Fungi
F		3	Colonies of black with flat velvety texture.	Fungi
G		6	White colonies, has a greenish-black mycelia	Fungi
H			Colonies of spherical, white and slimy	Yeast
I		27	White colonies and have irregular shapes	Yeast
J			Colonies of spherical and white	Yeast
K			Colonies of spherical and beige	Yeast
M			Colonies of spherical, slimy, and form a transparent layer on the surface of the colony	Yeast
G	NA	6	Colonies ofspherical, slimy, around (surface) transparent	Bacteria
H			Colonies of spherical, inside the colony and not transparent	Bacteria
C		24	Colonies of white, small, spherical, and slimy	Bacteria
A	MRSA	24	Small spherical colonies, in the middle, produces mucus	Bacteria
C		27	Spherical colonies with large size and do not produce mucus	Bacteria
D		27	Spherical Colonies of with large size and produces mucus	Bacteria
E		30	Colonies ofspherical, cream-colored, and does not produce mucus	Bacteria
F		48	Colonies ofbeige, spherical, and situated in the middle	Bacteria
G		48	Colonies ofspherical, located in the middle, and the edges of slimy colonies	Bacteria
H		48	Colonies ofspherical, large, situated at the base, beige	Bacteria

fermentation), the dominant microbes that play a role in spontaneous fermentation derived from yeast, bacteria and lactic acid bacteria.

The identification of isolates found fungii ndigenus presented in Table 2. The results show the identification of fungi involved in spontaneous fermentation of corn Bisi 18 dominated from the genus *Aspergillus*, 7 isolates produced fungi, five of which are from the genus *Aspergillus*. Results slide culture techniques showed that three out of five isolates *Apergillus* species has not been identified, so it is reported as *Aspergillus* sp.

Some studies have reported that the genus *Aspergillus* are fungi indigenus of the corn including *Aspergillus* sp isolated from maize kernels in Nigeria (Atehnkeng *et al.*, 2008), isolation of *A. niger* of fermented foods Maza (Adegbehingbe, 2014) and Ogi of corn (Adegbehingbe, 2013; Ojokoh, 2009), isola-

Table 2. Identification Results of Fungiindigenus on Spontaneous Fermentation of Corn Flour Bisi 18 with Culture Technique Slide

Isolates code	Picture	Description	Fungi Identified
A		Conidia on columnar shape (elongated) green to dark green. Cup-shaped vesicles, smooth-walled conidiophores are generally green, Conidia glubusa, ekinulat green. Misty green coloredColonies of with a texture like velvet.	*Aspergillus fumigatus*
B		Mushrooms in this group often cause food spoilage. Conidia this group colored yellow to green and may form skerotia (Heroine, 1989). Conidiophores colorless, rough top roundish to columnar, slightly rounded vesicles to the rod-shaped heads, while the large head shape globusa. Dark green Colonies of with average grained texture	*Aspergillus flavus*
C		Mushrooms grow forming colonies of filamentous fungi, smooth, convex and compact colony. Color white colored colonies with a texture resembling velvet tight and flat.	*Aspergillus sp*
D		Forming long branched filaments, which produce septate conidio phores or not that produces konidium-konidium are colored beige and gives color to the fungus. beige Colonies of with white mycelia, texture resembles velvet	*Aspergillus sp*
E		White mycelium, spread in culture, not insulated, single conidiophores, with tips enlarge heads of conidia bearing, form globusa, generally saprophytic in soil. Colonies of dark gray cotton with a texture resembling mounting.	C*unninghamella elegans*
F		Conidiophores dark, stout, upright, branched dendritically, branches producing conidia solitary, 4 or more cells, cylindrical, straight or curved; saprofit on wood. Colonies of black with flat velvety texture.	*Dendryphiopsisatra*
G		Mushrooms grow forming colonies of filamentous mold, smooth, convex and compact colony. colonycolor is influenced by the color of the spores. Mycelium colored white and greenish-black spores.	*Aspergillus sp*

tion *A. parasiticus* and *A. tamarii* as fungiindigenus of corn and beans in Kenya (Okun *et al*., 2015)

Aspergillusfumigatus found as one indigenus fungi found on corn Bisi 18. *A. fumigatus* is a fungi pathogenic to humans because it can cause allergic aspergillosis. This fungiColonies of misty green with a velvety texture. It has also been reported by Nyogesa *et al.* (2015) that the color green colonies of *A. fumigatus* foggy on PDA, rod-shaped conidia and mycelia less colorful. Fungi is also reported to have been isolated from maize in France (Seung-Beom *et al*., 2005) dan Kenya (Odhiambo *et al*., 2013).

Aspergillusflavus also found as corn fungiindigenus Bisi 18 during spontaneous fermentation. This Fungiare pathogen because it can produce aflatoxin is carcinogenic, teratogenic and mutagenic for humans. *A. flavus*fungi has also been reported as indigenus the spontaneous fermentation of corn by Rahmawati *et al.,* (2013). Other studies have also reported the isolation of *A. flavus*on corn in Iran (Houshyar-Fard *et al.,* 2014) and a kernel of corn in Italy(Mauro *et al*., 2013).

C*unninghamella elegans* found as a corn fungiindigenus Bisi 18. Several studies have reported that *C. elegans* has been isolated from the raw materials of food products in Lithuania (Lugauskas *et al.,* 2006) and sorghum (Soliman, 2003). Dendryphiopsisatra also found as a fungi isolate indigenus. *D. atra*fungiColonies of are black and conidiophores are solitary (Prasher and Verma, 2016). Both types of these fungi (*C. elegans* and *D. atra*) have not been reported as indigenusfungi on corn. This may be due Bisi 18 corn are corn that has undergone genetic engineering processes are constantly being developed, so that the potential difference microfloraindigenus on corn has the potential to change.

Conclusion

At the beginning of fermentation, which is 0 to 3 hours (adaptation phase), a microbe that was instrumental in the process of fermentation are fungi, after fermentation entered the exponential phase and stationary, ie 6 to 48 hours, the fermentation process is dominated by the growth of yeasts, bacteria and acid bacteria which lead to increased levels of lactic acid total tertitrasi until 0.38% and a pH value of 3.970. In this phase, the fungi growth decreased. The result of the isolation and identification fungi using slide culture techniques are found 7 species of mold that are involved in the process of spontaneous fermentation of corn Bisi 18, namely *Aspergillus fumigatus*, *A. flavus*, 3 types of classified *Aspergillus* sp, C*unninghamella elegans*, dan *Dendryphiopsisatra*

References

Adegbehingbe, KH. 2013. Fermented Sprouted and Unsprouted Maize for Ogi Production. *International Journal of Advanced Research*. 1 : 428-434.

Adegbehingbe, KH. 2014. Production of Masa Using Maize-Sorghum Blends. *International Journal of Science and Research (IJSR)*. 3 : 484-489.

Alvani, K., X. Qi., R.F Tester (2012). Gelatinisation Properties of Native and Annealed Potato Starches. *Starch - Stärke.* 64: 297-303.

Arroyo-López, F. Noé. 2009. Effects of temperature, pH and sugar concentration on the growth parameters of *Saccharomyces cerevisiae, S. kudriavzevii* and their interspecific hybrid. *International Journal of Food Microbiology.* 131 : 120–127.

Atehnkeng J, *et al*. 2008. Distribution and toxigenicity of Aspergillus species isolated from maize kernels from three agro-ecological zones in Nigeria. *International Journal of Food Microbiology.* 122 : 74–84.

Axelsson L. 2004. Lactic acid bacteria : classification and physiology. Di dalam :Salminen S, Wright AV, Ouwehand A, editor. *Lactic Acid Bacteria Microbiological and Functional Aspects*. New York. Marcel Dekker, INC. hlm 1-66.

Fardiaz S. 1992. Mikrobiologi Pangan 1. Gramedia Pustaka Utama, Jakarta.

Houshyar-Fard, M. *et al*. 2014. Studies on *Aspergillus flavus* Link. isolated from maize in Iran. *Journal of Plant Protection Research.* 5 (3) : 218-224.

Lugauskas A, *et al*. 2006. Micromycetes as toxin producers detected on raw material of plant origin grown under various conditions in Lithuania. *Ekologija.* 3: 1–13.

Mauro A, *et al.* 2013. Structure of an Aspergillusflavus population from maize kernels in northern Italy. *International Journal of Food Microbiology*. 162 : 1–7.

Nyongesa BW, S. Okoth, V. Ayugi. 2015. Identification Key for *Aspergillus*Species Isolated from Maize and Soil of Nandi County, Kenya. *Advances in Microbiology,* 5 : 205-229.

Odhiambo, B.O., Murage, H. and Wagara, I.N. 2013. Isolation and characterisation of aflatoxigenic *Aspergillus* species from maize and soil samples from selected counties of Kenya. *African Journal of Microbiology Research.* 7(34) : 4379-4388.

Ojokoh AO. 2009. A comparative study on the effect of traditional and improved methods of fermentation in the production of Ogi food. *Oriental Journal of Chemistry.* 25 (3) : 471-476.

Okun, D.O. *et al*. 2015. Distribution of indigenous strains of atoxigenic and toxigenic *Aspergillus flavus* and *Aspergillus parasiticus* in maize and peanuts agro.ecological zones of Kenya. *Agriculture and Food Security.* 4 : 14.

Prasher, I.B. and Verma, R.K. 2016. Hyphomycetes diversity of Himachal Pradesh-I. *Journal on New Biological Reports.* 5 (1) 52–58.

Rahmawati, R. Dewanti-Hariyadi, Hariyadi, P., Fardiaz, D. and Richana, N. 2013. Isolation and Identification of Microorganisms During Spontaneous Fermentation of Maize. J. Teknol. *Dan Industri Pangan.* 24 (1): 33-39.

Seung-Beom Hong, Seung-Joo Go, Hyeon-Dong Shin, Jens Frisvad, C. and Robert A. Samson. 2005. Polyphasic taxonomy of *Aspergillus fumigatus* and related species. *Mycologia.* 97 (6) : 1316–1329.

Silins, Indulis, 2014. The Effect of pH, aw, and lactic Acid Bacteria on Listeria Monocytogenes in Fermented //llufb.llu.lv/conference/foodbalt/2014/FoodBalt_Proceedings_2014-55-60

Soliman, H.M. 2003. Mycoflora and Mycotoxins of Cereal Grains in Delta, Egypt. *Mycobiology*. 31(4): 183-190

Zhang, X., Tong, Q., Zhu, W. and Ren, F. 2013. Pasting, rheological properties and gelatinization kinetics of tapioca starch with sucrose or glucose. *Journal of Food Engineering.* 114 : 255-261.