

Effect of different accelerators and inoculums used in fermentation on quality of dead chicken silage flour as feed ingredient for catfish

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Received October 06, 2016; Accepted January 24, 2017

ABSTRAK

Penelitian ini bertujuan untuk mempelajari pengaruh penggunaan bahan aditif berupa molases dan tepung jagung serta inokulum berupa bakteri *Lactobacillus* sp. dan *Streptomyces* sp., terhadap kualitas tepung silase berbahan baku ayam mati. Penelitian dilakukan menggunakan rancangan acak lengkap pola faktorial, terdiri atas 2 faktor dengan 5 ulangan. Faktor pertama adalah jenis bahan aditif dan faktor kedua adalah jenis inokulum yang digunakan pada saat fermentasi. Bahan yang digunakan berupa ayam mati segar dan utuh yang telah digiling dan semua bahan dicampur secara merata, lalu dimasukkan ke dalam kotak plastik berukuran 5 liter, kemudian ditutup rapat dan diberi plaster/lakban untuk menciptakan suasana anaerob. Pengamatan dilakukan setelah difermentasikan selama 3 minggu, meliputi: a) Karakteristik fisik, b) Kandungan mikroba, dan c) Kandungan gizi. Data yang diperoleh diolah dengan analisis ragam menggunakan program komputer SPSS versi 21.0. Diperoleh hasil bahwa berdasarkan kandungan protein maka *Lactobacillus* sp. (19.0%) lebih baik daripada *Streptomyces* sp. jika digunakan bersamaan dengan molases dan tepung jagung. Akan tetapi penggunaan kedua jenis inokulum tersebut akan menghasilkan kandungan lemak yang relatif sama (rata-rata 37,8%). Disimpulkan bahwa untuk memperoleh hasil fermentasi yang baik berdasarkan kandungan protein dan lemak, maka sebaiknya ayam mati difermentasikan menggunakan molases dan tepung jagung sebagai akselerator serta *Lactobacillus* sp. sebagai inokulum.

Kata kunci: akselerator, ayam mati, fermentasi, inokulum, tepung silase

ABSTRACT

This research aimed at investigating the effect using molasses and cornmeal as additives with *Lactobacillus* sp. and *Streptomyces* sp. as inoculums during fermentation on the quality of silage flour made from dead chickens. The study was conducted using a completely randomized factorial design, consisting of 2 factors with 5 replications. The materials used were the newly dead chickens which were chopped and mixed thoroughly with all ingredients; then transferred into a 5 liters plastic box for fermentation. Observations were made after 3 weeks fermentation, including: a) physical characteristics, b) microbial contents, and c) nutritional contents. The data were calculated using variance analysis utilizing computer program of SPSS version 21.0. It was found that based on the protein contents the *Lactobacillus* sp. (19.0%) was better than the *Streptomyces* sp. (17.8%) if combined with molasses and corn meal as the accelerators. However, the fat contents produced were relatively similar for both of the inoculums (mean of 37.8%). It can be concluded that in order to obtain a best fermented product in terms of the protein and fat content, the dead chicken should be fermented using molasses and cornmeal as the accelerator and *Lactobacillus* sp. as the inoculum.

Keywords: accelerator, dead chickens, fermentation, inoculum, silage flour

INTRODUCTION

The average number of live broiler chicken brought to Jakarta in the last 5 years is about 430,648 head per day (Ditjen PKH, 2015). However, during the days of religious festivals such as Eid al-Fitr, Eid al-Adha, Christmas and New Year, as the level of consumption of chicken meat increased, then the number of chicken brought from outside the city is up to 1 million head per day. The birds were mainly come from provinces of West Java, Banten, Central Java, Yogyakarta and Lampung, and usually come through the main entrance of Tangerang, Depok, Bogor, Karawang and Bekasi.

Transporting chickens from the farm to the market/collecting point or to the slaughterhouse could cause the death of the chickens. The death of the chicken was primarily due to exhaustion during the journey, due to long traveling, weather, shock and cramped in the transporting basket. Results of several studies reported that the death rate of chickens' transportation ranged between 0.12 to 1.64% (Nijdam *et al.*, 2006; Haslam *et al.*, 2008; Chauvin *et al.*, 2011). Mileage and long trips can affect chicken mortality rate, which increased from 0.59% on a trip with a distance of less than 50 km to 1.64% if the distance reached 300 km (Voslarova *et al.*, 2007). Travel for 3.5 hours resulting in the death of chickens as much as 0.24% and with the ride of over 5 hours led to an increased number of chickens died almost two-fold to 0.45% (Petracci *et al.*, 2005).

Results of study or report on the death rate of chicken due to travel to the slaughterhouse in Jakarta or other areas in Indonesia had never been available. If it is assumed that the average mortality rate of chickens transported to the area of Jakarta is approximately of 1% per day, then the number of dead chickens may be traded as dead chicken is as much as 4.000 to 10.000 birds per day.

The dead chicken is a chicken that does not cut perfectly, so there are still many blood clots in the flesh. Blood clots are good growth media for microorganisms, especially *Salmonella* sp. and *Escherichia coli* (*E. coli*), which is very dangerous for the health of people who consume it. Nearly 10% of the 1.4 million cases of food poisoning per year have been reported in several countries due to Salmonellosis (Core, 2005; Kinley *et al.*, 2010).

The usual way of handling a dead chicken is by combustion or incineration, burial, composting

(for fertilizer) and processing as raw materials for animal or fish (Shata and Farid, 2013). In addition, a technology that is also quite appropriate for the handling of dead chickens is the fermentation technology. The principle of the implementation of the fermentation is to stop contact between the fermented material with air or oxygen. It is necessary to ensure that during the fermentation process an acidic condition is reached, i.e. through the addition of soluble carbohydrates, such as molasses, cassava, rice bran, cornmeal, etc. (Jamarun *et al.*, 2014). In addition, it is also necessary to add lactic acid bacteria (LAB), such as *Lactobacillus* sp., *Pediococcus* sp, or *Streptococcus* sp. The addition of LAB will lead to increased production of lactic acid, which will create an acidic condition characterized by a decrease in pH. Low pH levels will inhibit the growth of viruses, pathogenic bacteria (especially *Clostridium* and *Enterobacterium*), yeasts and fungi that cause decay (Heinritz, 2011; Utomo *et al.*, 2013).

In relation to the dead chickens which were fermented in the form of a whole chicken, consists of meat, bones, heads, feet, viscera and feather, then the possibility that other materials that can help to degrade these materials should also be added. Chicken feathers containing a high level of crude protein, around 80-91%, but it have a very low digestibility because most of the structure of the feathers consists of keratin proteins belonging to the fiber protein (Manivasagan *et al.*, 2013). One type of bacteria that can produce keratinase to degrade keratin is *Streptomyces* sp. (Jani *et al.*, 2014).

The objective of this study was to investigate the role of carbohydrate as fermentation accelerator during the fermentation process of the whole dead chicken. Furthermore, this investigation was also made to determine the proper inoculum (*Streptomyces* sp. or *Lactobacillus* sp.) degrading the keratin contained in chicken feathers.

MATERIALS AND METHODS

Experimental Design and Treatments

This study was conducted using a completely randomized factorial design consisting of 2 factors with 5 replicates. The first factor was 2 types of fermentation accelerators, namely: a) 20% molasses (Accelerator-1), b) a mixture of 20% molasses and 20% cornmeal (Accelerator-2). The second factor was three types of bacteria as

inoculums, namely: a) 10^6 cfu/g *Lactobacillus* sp. (Inoculum-1), b) 10^6 cfu/g *Streptomyces* sp. (Inoculum-2), c) a mixture of 10^6 cfu/g *Lactobacillus* sp. and 10^6 cfu/g *Streptomyces* sp. (Inoculum-3). Thus, there were six treatments: a) Accelerator-1 + Inoculum-1, b) Accelerator-1 + Inoculum-2, c) Accelerator-1 + Inoculum-3, d) Accelerator-2 + Inoculum-1, e) Accelerator-2 + Inoculum-2, f) Accelerator-2 + Inoculum-3.

Inoculums Preparation

The propagation of isolates *Lactobacillus* sp. and *Streptomyces* sp. was carried out in two stages. The first stage was re-growing them in an agar medium slant, while the second stage was propagation in a liquid medium. The media used for this purpose were the trypticase soy agar (TSA) for the *Lactobacillus* sp. and the potatoes dextrose agar (PDA) for the *Streptomyces* sp. The TSA media consisted of 30 g of trypticase soy broth (TSB) and 15 g agar, while the PDA media comprised of 100 ml of potato extract, 20 g of sucrose and 20 grams of agar. Each media was then added with distilled water up to 1000 ml volume, heated to boiling and sterilized in an autoclave at a pressure of 1 atmosphere for 15 minutes. Each inoculum was grown on an agar medium slant by scraping a wire loop containing bacterial cells on the surface of agar medium and it was finally incubated in the incubator for 5 days (Steubing, 1993; Bujalance *et al.*, 2006).

The propagation of inoculum *Lactobacillus* sp. and *Streptomyces* sp. in the liquid medium was carried out in a 500 ml erlenmeyer flask which contained 200 ml of TSB and Potatoes Dextrose Liquid (PDL). This media had been previously sterilized in an autoclave at a pressure of 1 atmosphere for 15 minutes and allowed to stand for 24 hours. One loop of each isolate which have had been re-grown in the agar medium slant was inoculated in the liquid medium and subsequently incubated on a shaker at the speed of 100 rotary per minute for 5 days (Steubing 1993). Number of cells that are formed was then counted at the end of incubation period, using a haemocytometer, and it was finally diluted to reach the density of 10^6 cells per ml, prior to the fermentation time.

Fermentation Processes

Number of dead chickens used in this study was 150 pieces which were collected in the morning, around 4:00 to 6:00 pm, from several collecting points and slaughterhouse located in

East Jakarta. The dead chicken collected was the freshly dead chicken, with an average weight of 1.0 kg. Chicken deaths were not due to disease but due to exhaustion during the journey from the farm to the slaughterhouse. The dead chickens were immediately taken to the Laboratory of the Jakarta Assessment Institute for Agricultural Technology (Jakarta AIAT) in South Jakarta, to be weighed and immediately chopped/milled using a chopper/grinder into a size of 2.5 cm or smaller.

Then the ground chickens were thoroughly mixed with additives and inoculums in accordance with the type of the treatment that has been designated. Furthermore, each 5 kg of the mixture was placed into a plastic box with a capacity of 5 liters, and then it was sealed using plaster/tape to ensure an anaerobic atmosphere. All plastic boxes were placed in a room temperature for a three weeks period of fermentation. After that, the fermented materials were then dried using an oven with a temperature of 60 °C until it reaches the water content of about 15%. It was finally ground using a grinding machine with a 100 mesh sieve size (Despal *et al.*, 2011; Utomo *et al.*, 2013).

Chemical and Data Analysis

The parameters observed were: a) physical characteristics (aroma, color and texture), b) microbial content (TPC, *Salmonella spp.* and *E. coli*), and c) chemical analysis (protein, fat and energy). Microbial content analysis was conducted in the Microbiology Laboratory of Faculty of Veterinary Medicine, Bogor Agricultural University in Dramaga, Bogor, while the chemical analysis carried out in the Chemistry Laboratory of Livestock Research Institute in Ciawi, Bogor. The microbial analysis is conducted according to SNI No.: 2897-2008 (BSN, 2008), while the chemical analysis done according to AOAC (2000). The protein content was measured through destruction with sulfuric acid and the subsequent measurement of Kjeldahl Nitrogen using auto analyzer. Fat content was measured by extraction with petroleum spirit, whereas the gross energy content was measured using a Bomb Calorimeter.

Analysis of the data from all the measured parameters was done using analysis of variance (ANOVA), using the computer program of SPSS (Statistical Package for Social Sciences) version 21.0. If the results of statistical analysis showed significant differences among treatments, then the analysis was continued with Duncan's multiple

range test (Mattjik and Sumertajaya, 2006).

RESULTS AND DISCUSSION

Physical Characteristics

The dry matter (DM) content of the minced chickens in this study was 47.4%, which is much higher than the DM content of chicken or broiler meat which is only about 25.6% (Dewi, 2013). The high DM content on the ground dead chickens was likely due to that it was not only consist of chicken meat alone but it was in the form of whole chickens which consisted of feathers, bones, offal and so forth.

As it is known that, to ensure the best fermentation process, the DM content in the fermented materials should be at least about 35% (Utomo *et al.*, 2013). The DM content of the minced chicken used in this study was quite high (47.4%), which is already eligible for the implementation of fermentation. However, the DM content decreased after being mixed with 20% molasses to 43.1%, this is possibly due to the addition of the water content derived from the molasses. But on the contrary, the DM content increased to 52.4% after it was mixed with 20% molasses and 20% cornmeal, which might be due to the addition of the DM derived from the cornmeal.

The minced chicken had a fishy smell that is

typical for the minced chicken meat (Table 1). The addition of molasses caused a slight change of the minced chicken aroma, which still smelled fishy, but there is an additional distinctive smell of the molasses. However, there was a slightly fishy odor reduction with the addition of molasses and cornmeal; nonetheless it was still smell a distinctive aroma of the molasses.

The minced chicken had a reddish color which is also typical color of the minced chicken meat, but those mixed with molasses the color was changed to dark red/black. When the minced chicken was mixed with molasses and cornmeal the color was changed to slightly lighter compare to if it was only mixed with molasses. There were no differences on the texture of the minced chicken at before and after mixed with molasses alone or with the addition of cornmeal. There were also no differences in the flavor, color and texture of the minced chicken at the time before and after mixed with all kinds of inoculums used in this study.

Based on the physical characteristics of the dead chickens after fermented, it seems that the fermentation process was successful resulting good quality silage. The silage resulted in this study had a fragrant aroma, dark in color, moist in texture, solid, did not clot and not slimy (Table 1). As it has been reported that a good silage has a) a fragrant aroma as it contains lactic acid, b) does

Table 1. Physical Characteristics of the Dead Chickens with Different Treatments at Before and After Fermentation for a 3 Weeks Period

Treatments / Fermentation Period	Molasses			Molasses + Cornmeal		
	Aroma	Color	Texture	Aroma	Color	Texture
A. Before Fermentation						
• <i>Lactobacillus</i> sp.	Fishy	Reddish Black	Soft	Slightly Fishy	Light Reddish Black	Soft
• <i>Streptomyces</i> sp.	Fishy	Reddish Black	Soft	Slightly Fishy	Light Reddish Black	Soft
• <i>Lactobacillus</i> sp.+ <i>Streptomyces</i> sp.	Fishy	Reddish Black	Soft	Slightly Fishy	Light Reddish Black	Soft
B. After Fermentation						
• <i>Lactobacillus</i> sp.	Fresh & Sour	Brown ++	Watery ++	Fresh & Sour ++	Light Brown	Watery
• <i>Streptomyces</i> sp.	Fresh & Sour	Brown +++	Watery +	Fresh & Sour ++	Light Brown	Watery
• <i>Lactobacillus</i> sp.+ <i>Streptomyces</i> sp.	Fresh & Sour	Brown ++	Watery ++	Fresh & Sour	Light Brown	Watery

not have a pungent odor, c) has an intact texture, smooth, not slimy and not agglomerate (Saun and Heinrichs, 2008; Despal *et al.*, 2011). Results of this study indicate that there was no significant effect on the physical characteristics of the fermented chicken given different types of treatments.

Microbial Contents

To ensure that the fermentation of dead chickens were safe to be used as a feed material, it is necessary to measure the contaminated microbes. In fact, the chicken meat is perishable food which is very easily contaminated by microorganisms, as it is a very good medium for microbial growth. The types of pathogenic microbes often found in animal-based food ingredients are: *Champhylobacter* sp., *Clostridium* sp., *Coliform*, *E. coli*, *Enterococci*, *Listeria* sp., *Salmonella* sp., and *Staphylococcus aureus*. However, most types of microbes often contaminate the chicken meat and are often used as a reference for food safety are *Salmonella* sp. and *E. coli* (Gustiani, 2009).

In order to ensure food security, the maximum limit of the number of microbes or the total plate count (TPC) value allowed for carcass and meat of chickens are as much as 1×10^6 cfu/g or equivalent to 6 log cfu/g (BSN, 2009). The TPC value in all treatments used in this study at the time before and after fermentation is higher than that maximum limit. The TPC value at the time before fermentation ranged between 7.30 to 9.18 log cfu/g, while the value of the TPC after fermentation ranged from 7.00 to 8.49 log cfu/g (Table 2). The lowest TPC value at before fermentation was the minced chickens mixed with molasses and *Lactobacillus* sp., while the highest TPC value when it was mixed with molasses, cornmeal, *Lactobacillus* sp. and *Streptomyces* sp. The high content of the TPC in the minced chickens was likely due to the addition of inoculums *Lactobacillus* sp. and *Streptomyces* sp. respectively amounted of 6 log cfu/g of fermented material. Another possibility was due to that the chickens had died about 2-3 hours before they were ground, so there had been some contaminations from the environment and microbial growth both on the outside and inside of the chicken body.

It seemed that the high number of the TPC on a fermented minced chicken mixture was not directly related to the insecurity of the material to be used as a food ingredient. This product was not

to be intended for direct human consumption, but it will be used as a source of protein in the manufacture of feed for animal or catfish. As it had been noted earlier that the high content of the TPC on the fermented materials is was due to the addition of 10^6 cfu/g each of inoculums in the form of *Lactobacillus* sp. and *Streptomyces* sp. Both types of inoculums were a test material to determine its effectiveness in helping the fermentation process.

Therefore, the maximum limit set by BSN (2009) for the TPC value which is 6 log cfu/g is only for the chicken meat or carcass that will be consumed directly by humans. It appears that the value of TPC on all materials fermented in this study tended to have a lower value at the time after fermentation compared to before fermentation (Table 2). The use of Inoculum-2 tended to increase the content of TPC at the time before fermentation, especially if it was used in conjunction with the Inoculum-1. However, the numbers of TPC at the time after fermentation were not significantly differences ($P > 0.05$) among all treatments. This suggests that the microbe could not be evolved or died during the fermentation process.

It was shown that there was a positive interaction on the content of the TPC at the time before fermentation between the uses Accelerator-1 with the type of Inoculum used. The use of Inoculum-2 caused a higher content of TPC (8.38 cfu/g) compared to the Inoculum-1 (7.30 cfu/g). The content of TPC was also likely to be increased (8.88 cfu/g), if the Inoculum-3 was used. However, after the fermentation period, a significant interaction occurred only between the use of Accelerator-2 with the type of inoculum used. It was shown that the use of Inoculum-1 simultaneously with the Accelerator-2 produced the lowest content of TPC, amounting 7.30 cfu/g.

Although the content of the TPC on the materials tested in this study exceeded the maximum limit specified, but in fact the material is safe from the danger of the pathogenic bacteria. The *Salmonella* sp. content in the material before and after fermentation was negative per 25 gram (Table 2). This is in accordance with the recommended limit of *Salmonella* sp. contamination in chicken meat by BSN (2009). In addition, the *E. coli* content in all of the material was also very low, which only amounted < 3 cfu/g, a figure much lower than the maximum allowed limit, which is 10 cfu/g. Therefore, materials used in this study are very safe to be used as an

Table 2. Microbial Contents of the Dead Chickens with Different Treatments at Before and After Fermentation for a 3 Weeks Period

Accelerator	Inoculum	Before Fermentation			After 3 Weeks Fermentation		
		TPC (Log cfu/g)	<i>Salmonella</i> (per 25 g)	<i>E. coli</i> (MPN/g)	TPC (Log cfu/g)	<i>Salmonella</i> (per 25 g)	<i>E. coli</i> (MPN/g)
Acc-1		8.19	Negative	< 3	7.63	Negative	< 3
Acc-2		8.72	Negative	< 3	8.14	Negative	< 3
	Mean	8.46	Negative	< 3	7.89	Negative	< 3
	Ino-1	7.86 ^a	Negative	< 3	7.15	Negative	< 3
	Ino-2	8.48 ^{ab}	Negative	< 3	8.45	Negative	< 3
	Ino-3	9.02 ^b	Negative	< 3	8.04	Negative	< 3
	Mean	8.45	Negative	< 3	7.88	Negative	< 3
Acc-1	Ino-1	7.30 ^a	Negative	< 3	7.00	Negative	< 3
	Ino-2	8.38 ^b	Negative	< 3	8.28	Negative	< 3
	Ino-3	8.88 ^b	Negative	< 3	7.60	Negative	< 3
	Mean	8.19	Negative	< 3	7.63	Negative	< 3
Acc-2	Ino-1	8.42	Negative	< 3	7.30 ^a	Negative	< 3
	Ino-2	8.59	Negative	< 3	8.62 ^{ab}	Negative	< 3
	Ino-3	9.18	Negative	< 3	8.49 ^b	Negative	< 3
	Mean	8.73	Negative	< 3	8.14	Negative	< 3
Main Effect :							
Accelerator		NS	NS	NS	NS	NS	NS
Inoculum		<0.01	NS	NS	NS	NS	NS
Accelerator x Inoculum		<0.01	NS	NS	<0.01	NS	NS

^{a-b} Means within a row with no common superscript differ significantly (P<0.05). Acc-1 = Accelerator - 1: Molasses; Acc-2 = Accelerator - 2: Molasses + Cornmeal; Ino-1 = Inoculum - 1: *Lactobacillus* sp.; Ino-2 = Inoculum - 2: *Streptomyces* sp.; Ino-3 = Inoculum -3: *Lactobacillus* sp. + *Streptomyces* sp.

ingredient for the formulation of feed for animal or catfish.

The low content of *Salmonella* sp. and *E. coli* bacteria in the tested material showed that the processing and handling of materials in this study were quite hygienic with a fairly good level of sanitation. Contamination by the *Salmonella* sp. can only occur if there is contamination from fecal of infected chickens during processing or handling. Contamination is also easily occurring if the process of cutting and handling of chickens conducted at the same site or adjacent to the raising of the contaminated chicken. The *Salmonella* sp. will also breed well if the

environment is hot and humid (Utari *et al.*, 2016). The *E. coli* is a normal micro flora present in the digestive tract and is often found in water contaminated by animal or human feces. The existence of the *E. coli* bacteria is often used as an indicator to show that the level of poor sanitation and non hygienic handling process (Kornacki and Johnson, 2001).

In this research, the process for handling chicken was not similar to the commonly carried out for broiler chicken, such as slaughtering, plucking, offal cleaning, cutting, packaging etc., so it was less likely to be contaminated by microbes. The chances of contamination by

microbes was also minimum as the chicken used was the freshly dead chickens and their death was not due to diseases or illness but because of fatigue caused by transportation from farm to the market/collecting points or slaughterhouse. The chicken was also never washed with water, but immediately milled and blended with accelerators and inoculums for fermentation. So it is unlikely to be contaminated by microbes contained in the water, but instead the possibility of microbes will be dead or stunted due to the present of the materials used as the fermentation accelerators and inoculums.

Nutritional Contents

As it was shown in Table 3 that the protein content of the fermented chicken was significantly higher ($P < 0.05$) when using the Accelerator-1 (18.7%) compared to the Accelerator-2 (16.5%). Additionally, the use of Inoculum-1 tended to produce a higher protein content (19.5%) compared to the Inoculum-2 (17.8%) or Inoculum-3 (16.1%).

There was a positive interaction on the protein content of the fermented chicken between the types of Accelerator with the type of Inoculum used. The use of Accelerator-1 with Inoculum-1 or Inoculum-2 produced a higher ($P < 0.05$) protein content (mean of 19.5%) than the use of Inoculum-3 (17.3%). When using the Accelerator-2, the protein was better produced if the Inoculum-1 was used (18.7%). However, when using the Inoculum-2 (16.0%) and Inoculum-3 (14.8%) resulted in a lower protein content.

There were no significant differences ($P > 0.05$) in the fat content of the chicken after fermentation period when using different types of Inoculum (Table 3). But there was a positive interaction in fat content between the use of Accelerator-2 and the type of inoculum used. The use of Accelerator-2 with Inoculum-1 or Inoculum-2 produced a fat content which was not significantly different ($P > 0.05$) with a value of 3.31%. However, with the use of Inoculum-3 resulted in a fairly high fat content (36.2%).

There was no difference in the gross energy contents in chickens which were fermented using various types of Accelerator and Inoculum. There was also no interaction of the value of gross energy content between all types of Accelerator and Inoculum used. The average gross energy content of the chicken after fermented was 2308 kcal/kg (Table 3).

The protein content of the chicken meat

generally ranges between 16-23% and the fat content of between 1.5 to 13.0% (Aberle *et al.*, 2001; Rosyidi *et al.*, 2009). The protein content of the fermented dead chickens in this study was between 14.8 to 19.6%, which is almost equal to the protein content of the chicken meat; however, it has a very high fat content of between 31.8 to 43.5%. It is seen that the whole chickens which were fermented with molasses and cornmeal together was not cause any significant effect on the protein content, but the fat content increased almost two-fold.

Based on the data of the protein content obtained from this study, the dead chickens was best fermented using molasses as the accelerator and *Lactobacillus* sp. or *Streptomyces* sp. as the inoculums. The use of *Lactobacillus* sp. may also be combined with molasses and cornmeal as the accelerator because it produces a protein that is was not significantly different (19.3% vs. 18.7%). But the use of *Streptomyces* sp. as inoculum along with molasses and cornmeal should not be done because it would lead to a decrease in protein content (19.6% to 16.0%). However, based on the fat content, the fermentation of the dead chickens could be done using molasses and cornmeal as the accelerator and the *Lactobacillus* sp. or *Streptomyces* sp. as the inoculum, because the fat contents of the fermented product were relatively similar (mean of 37.8%). Therefore, in order to obtain a best fermentation product in terms of protein and fat content, the dead chicken should be fermented using molasses and cornmeal as the accelerator and *Lactobacillus* sp. as the inoculum. The poultry by product meal which is usually used as the catfish feed in the United States contains approximately 58-65% protein and 10-14% crude fat, and it consists of only heads, feet, underdeveloped eggs and visceral organs, but it does not contain feather (Li and Robinson, 2013). This means that the protein content of the fermented dead chicken in this study (mean of 19.0%) was far too low, and the fat (mean of 37.8%) was far too high compared to the poultry by product meal used as catfish feed. The protein content of the dead chickens fermentation obtained in this study was also still very low compared to the protein content of the materials commonly used as ingredients in the manufacture of animal feed or catfish, such as fish meal, blood meal, mustard oil cake and soybean meal which usually contain protein between 41.9 to 90.0%. However, the fat content is too high compared to those materials which contain fat between 1.40-

Table 3. Nutrient Contents of the Dead Chickens with Different Treatments After Fermentation for a 3 Weeks Period

Accelerator	Inoculum	Protein (%)	Fat (%)	Gross Energy (kcal/kg)
Acc-1		18.7 ^a	42.3 ^a	2258
Acc-2		16.5 ^b	34.1 ^b	2357
	Ino-1	19.5 ^a	38.0	2263
	Ino-2	17.8 ^{ab}	37.7	2335
	Ino-3	16.1 ^b	39.0	2325
Acc-1	Ino-1	19.3 ^a	41.6	2210
	Ino-2	19.6 ^{ab}	43.5	2290
	Ino-3	17.3 ^b	41.7	2275
Acc-2	Ino-1	18.7 ^a	34.4 ^{ab}	2315
	Ino-2	16.0 ^b	31.8 ^a	2380
	Ino-3	14.8 ^b	36.2 ^b	2375
Main Effect :				
Accelerator		<0.01	<0.01	NS
Inoculum		<0.01	NS	NS
Accelerator x Inoculum		<0.001	<0.01	NS

^{a-b} Means within a row with no common superscript differ significantly ($P < 0.05$). Acc-1 = Accelerator - 1: Molasses; Acc-2 = Accelerator - 2: Molasses + Cornmeal; Ino-1 = Inoculum - 1: *Lactobacillus* sp.; Ino-2 = Inoculum - 2: *Streptomyces* sp.; Ino-3 = Inoculum -3: *Lactobacillus* sp. + *Streptomyces* sp.

21.0% (Nahar *et al.*, 2000).

Due to its high fat content, the fermented dead chicken will be difficult to pellet if it is to be used as an ingredient in the manufacture of animal or catfish feed. The fat content in the different types of pellet for catfish feed on the market generally only have a fat content of about 2-10% (Robinson *et al.*, 2006). Therefore, before it can be used as feed material in the manufacture of pellets for animal or catfish feed, the fermented dead chicken should be first mixed with other materials that can reduce its fat content, such as cassava pomace flour or calcium carbonate.

CONCLUSION

The fermentation process in this study was successful as it produced a good quality silage which had a fragrant aroma, dark in color, moist in texture, solid, did not clot and not slimy. The dead chicken and its mixture used for the fermentation had a slightly high in the TPC

content but it was safe from the danger of the pathogenic bacteria. In order to obtain a best fermented product in terms of the protein and fat content, the dead chicken should be fermented using molasses and cornmeal as the accelerator and *Lactobacillus* sp. as the inoculum.

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