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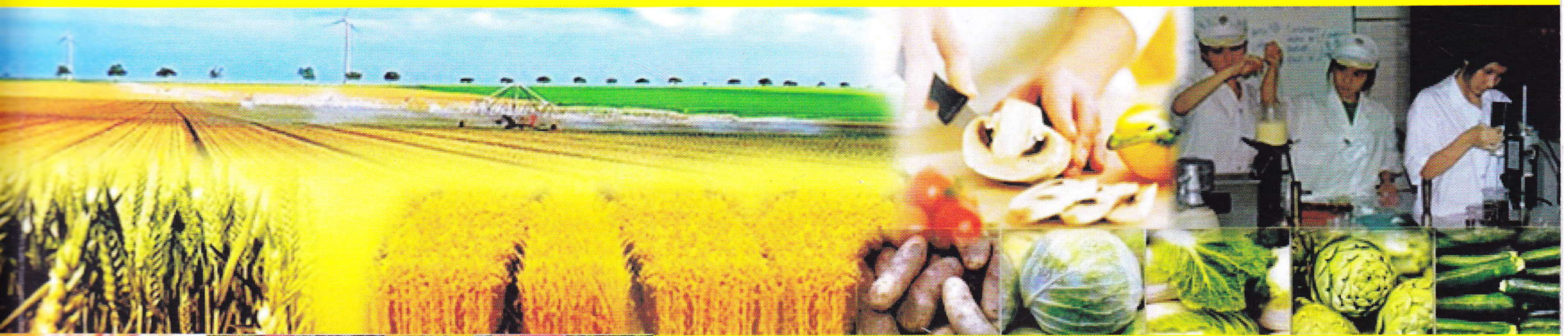
"Life Improvement through Food Technology"
Surabaya, October 28th - 29th, 2011



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LIST OF PAPER

PAPER OF ORAL PRESENTATION

PAPER OF ORAL PRESENTATION (Cont'd)

NITROGEN GAS APPLICATION FOR PACKAGING OF *NILA* CHIPS ON VARIOUS STORAGE TIMES AND TEMPERATURES

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ABSTRACT

Nila is a kind of traditional fish usually used as foodstuff at Indonesian society especially at South Sumatera area. The objective of this research was to study the nitrogen gas application for nila chips packaging. Polypropilene plastic with thickness of 0.6 mm was used as the packaging material, filled out with nitrogen gas at full volume, sealed and stored under various times (0, 20, 40 and 60 days) and temperatures (30 ^{0}C and 40 ^{0}C). Control, without filling with nitrogen gas, was also performed to compare the result. The nila chips were analyzed for the moisture content, rancidity values, browning index and total microbes. The results showed that moisture content, peroxide value and browning index increased during storage, with linier correlation model of $Y_i = 0.0264\ X_i + 5.198$ ($R^2 = 0.99$), $P_{21} = 0.2277 + 0.2856t$ ($R^2 = 0.97$) and $B_{21} = 0.73 + 0.0033t$ ($R^2 = 0.56$), respectively. The total microbes were also increased of about 1 log cycle during 60 days of storage. Nitrogen gas package could be able to decrease deterioration of nila chips during storage at ambient temperature. The rate of quality changes was decreased to 82-89 % when nitrogen applied in the package. The value of Q_{10} for all of quality parameters was higher when nitrogen applied in the package compare to control. It was found nitrogen in the package not only to be preservation agent but also increase temperature sensitivity of nila chips quality during storage.

***Key words**: nitrogen, nila chips, storage*

INTRODUCTION

Fishery product, such as *nila* fish, is important protein resources for most of civil society at Sout Sumatera. It was traditionally culture in the village, but it was easy to deteriorate or decrease the quality to be perishable foods. Microbiological and chemical reaction was leading potential factor to destroy the quality. Postharvest technology was needed to solve that problem in order to save the economic and social value (Saptanto, Suryawati dan Koeshendrajana, 2005).

Nila fish was given name from Directorat General of Fishery- Indonesia at 1972 for species member of *Nilotika*. The fish has spesific taste and easy to be cultured, but the consumption was ristricted because of stinking-smelling fish. *Nila* chips is one of many ways for enhancing *nila* utilization with no stinking-smelling fish, but it was need carefully treatment to store any times and temperatures. Chips deterioration was signed by rancid smelling, uncrackling, abnormal texture, bacterial spot etc. Most of chips manufacturer was managed those quality by suitable packaging, but for *nila* chips it has not yet been observed which the best one. Kadoya (1990), reported that there are some advantages when plastic was used as packaging, for example it was easy performed, relatively cheaper, posible any design, not rusty and relatively lower water permeability. Murniyati (1999) had used poliethilene plastic for snack fish stored at ambient temperature, and showed that although it was packed by plastics the product was increased water activity value, rancidity and microbiological number. Therefore, it needed to find other method to pursue the quality decrease, at least decrease the rate of quality changes during storage.

Modified atmosphere storage has ben recomended to extent shelf life of fish product (Torry Research Station, 2009). Atmosphere of surounding material in the package, without treatment for changing, has same composition with outer space of package at initial start of packaging. It was naturally consited of gas such as nitrogen, oxygen, carbon dioxide, and water vapour.. The oxygen gas was not major component but it could pursue any chemical reactions (e.g. oxidation). It was important to restrict oxygen and water vapour contact to extend shelf life product in the package. Koseki and Itoh (2002) have reported the positive effect of nitrogen gas packaging on quality of vegetable. Modified atmosphere storage is one of many ways to implement that concept, and it was usually maintain by increasing proportion of nitrogen to decrease the portion of other gas in the package. Kadoya (1990) had reported the case of peanut butter packaging method, when it was increased nitrogen portion in the package it would extended shelf life product by smaller increasing of moisture content and peroxide value. It was recommended to maintain the oxygen less then one percent of atmospheric gas in the package.

Temperature had been noted as signifcant factors influenced the product quality during storage (Potter and Hotchkiss, 1995; Bridge, 1992). The rate of chemical reaction in the foodstuff usually was increased when temperature was increased. Syarief and Halid (1993) recommended it to maintain the stabilization of temperature when product quality was needed. Temperature sensitivity of product in the package was varied depend on packaging method. It was need to be considered when we need to design packaging system and storage (Krochta, 2007). Arrhenius model approximation was widely used for expressing the dependency of rate on temperatures and characterized by activation energy. Q_{10} concept is another way to express the rate dependency on temperature, usually counted by ratio of rate value at temperatures difference 10 °C or relatively shelf life when it was stored at difference temperature at 10 °C.

Quality changes of food product during storage had been succesfully expressed by kinetics model approximation with various parameters, such misture content (Saguy, 1983), browing index (Priyanto, 1997), texture, phisical and biochemical characteristics (Labuza, 1980), etc. It is important thing to know the rate constant (*k*) of the quality changes during storage (Priyanto, 2009). There are no information the value of nila chips during storage when nitrogen was applied to modified atmosphere in the package. Therefore, research on this field is needed to conduct for studying the application of nitrogen gas for packaging of nila chips during storage at various times and temperatures.

MATERIALS AND METHOD

Main material of *Nila* fish was prepared from local market at Palembang, South Sumatera. Other material such as tapioca, salt, eggs, *food grade* oil and spices was also purchased from local market. Chemical reagent for analysis was bought from local chemical store, such as alcohol (96 %), kloroform, KI, $Na_2S_2O_3$ and aquadest. The apparatus for processing and analysis was available at the Agricultural Product Laboratory, Faculty of Agriculture, Sriwijaya University.

Nila chips was prepared from fresh fillet *nila* fish that was cleaned and separeted with others part usually omitted. Fillet was store at the freezer, and then it was chopped to be thin and small fillet. At about 600 g fillet was added 420 g tapioca, 25 g salt, 50 g onions, 25 g red onion, 5 g pepper, 15 g ginger, 50 g margarine and 70 eggs. Those are mixed and performed thin layer (c.a. 0.30 cm) to be raw *nila* chips. It was then to be fried by deep frying method at about three minutes, and made leak to ready *nila* chips for packaging.

Nila chips ready for packaging, at amount of 20 g packed in the polipropilene packaging and strighly sealed for the first treatment A_1 (none nitrogen modified), the other at the same amount of nila chips was packed in the polipropilene packaging inserting by nitrogen gas in the package and sealed as second treatment labelled by A_2 code. Many unit samples were made for both two treatment (A_1 and A_2). Some samples from both treatment was then stored at first storage temperature treatment labelled by B_1 for ambient temperature (30 ± 2 ^{0}C) and the others was stored at 40 ± 2 ^{0}C and lebelled by B_2 code. The quality was observed during storage times at 0, 20, 40, dan 60 days.

The quality of *nila* chips was expressed by quality parameters consisted of moisture content, peroxide value, browning index and total microbiological number. Moisture content was measured by gravimetric method (Sudarmadji *et al.*,1997), peroxide valued was measured by titimetric method (Sudarmadji *et al*,1997), browning index was measured by spectrofotometric method (Cohen *et al.*, 1994) and microbiological number by microbial total count spread plate method (Fardiaz, 1989). Experiment was conducted on completly factorial block design. Colected data was then used to performed kinetic models to find the rate value of deterioration and to evaluate the efectiveness of nitrogen gas application compare to normal condition for preserving *nila* chips on packaging during storage. Determination coeficient ( ) was used as criteria for valid model (Ganjloo *et.al.*, 2009) for expressing the realtionship between variables.

RESULTS AND DISCUSSION

The result show that nitrogen gas package could be able to decrease deterioration of *nila* chips during storage at ambient temperature. The rate of quality changes was decreased to 82-89 % when nitrogen applied in the package. The value of Q_{10} for all of quality parameters was higher when nitrogen applied in the package compare to control. It was found nitrogen in the package not only to be preservation agent but also increase temperature sensitivity of nila chips quality during storage. The phenomenon was slightly difference when product was stored at higher temperature (40 ^{o}C). It was shown the rate of changes of moisture content and microbe number of *nila* chips in nitrogen gas package was higher than normally package (no nitrogen treatment). The other quality parameters i.e. peroxide value and browning index was still same as shown phenomena by storage at ambient temperatures.

Moisture Content

Moisture content of *nila* chips was ranged from 4.3 percent to 8.85 percent, with average value of 6.54 percent. It was increased during storage. At the end of storage (60 days) at ambient temperatures, the average moisture content of *nila* chips in nitrogen gas packaging was 6.78 percent, while control product was 6.99 percent. When the product stored at higher temperature (40 ^{o}C), the moisture content about 8.48 percent for nila chips in nitrogen packaging, while the other one was 8.29 percent.

The moisture content of *nila* chips during storage could be expressed by linier model approximated by polynomial orthogonal method, where moisture content and storage time was represented, respectively, by Y and X. The linier equation founded on ambient temperature storage is $Y_i = 0.0264 X_i + 5.198$, $R^2 = 0.99$, for *nila* chips on nitrogen gas packaging and $Y_j = 0.0305X_i$

+ 5.330, R^2 = 0.91, for control *nila* chips. While for higher temperature storage (40 °C) for those treatment, respectively, Y_k = 0.0418x + 5.785, R^2 = 0.93 and Y_l = 0.0531x + 5.296, R^2 = 0.99. The index, i and j, was respectively indicated for *nila* chip stored at ambient temperature with nitrogen gas packaging and control (none nitrogen gas treatment), while k and l was respectively indicated for *nila* chip stored at higher temperature (40 °C) with nitrogen gas packaging and control (none nitrogen gas treatment). Those models tell us that moisture content of *nila* chips was increased during storage. It means there are mass transfer from outer side package to the inner one, and addition of amount water to the product. The increasing moisture content would increase the water activity value that indicated more water could be used by microbial substance and also pursue hidrolysis process. Therefore, it leads to deteriorate or decrease the product quality.

The above models was justified that moisture content of *nila* fish was changes following the zeroth order kinetics model, which was expressed by linier model supporting by higher value of determination coeficient Based on zeroth order kinetics model as dQ/dt = k, by spesific integration method was found out linier model as $Q=Q_o + k(t)$ where Q: quality, t: storage time and k: rate constant of quality changes (van Boekel, 1996). Those above equation then shown us the rate value of moisture content change as stated in the table and also ratio of the rate value when temperatures storage was at difference of 10 °C that usually called Q_{10}. It was shown that nitrogen application increased the sensitivity moisture change the temperature, where the change of temperature would change the rate constant significantly.

Table 1. The rate of moisture content changes of *nila* chips during storage

Nila chip treatment packaging	Rate (k) at 30 °C	Rate (k) at 40 °C	Q_{10}
Nitrogen gas application	0.0264	0.0531	2.011
Control (no treatment)	0.0305	0.0418	1.371

Peroxide Value

Peroxide value was measured for expressing the quality of *nila* chips especially in relation with rancidity, taste and role of oil or fat component of product during storage. At start of storage time (zero days), the rancidity was undetectable given by zero peroxide value. This fact was tell us that product was healty and in good contition after packaging. Futher analysis at another storage time showed that peroxide value was increased during storage. *Nila* chips treatment by nitrogen gas packaging consistenly showed lower peroxide value than control at ambient temperature storage, but it was not happenned on higher temperature (40 °C) storage. The average of peroxide value at the end of storage (60 days) in ambient temperatures was 16.61 for *nila* chip with nitrogen gas packaging where as control about 18.59, and at higher temperature (40 °C) storage, respectively, 19.55 and 19.94 meq/kg fat.

Futher analysis data by polynomial orthogonal method was founded that the relationship of peroxide value of *nila* chip with storage time, as shown in above figure, was valid and significant described by linier model, otherwise not significant for square or kuadratic models. Based on those facts, by adopting the kinetics models for chemical reaction, the zeroth order reaction kinetics model was the most suitable for describing deterioration process of *nila* fish. When P_{ij} was defined as peroxide value of *nila* chip with packaging method i (i=1: control, no treatment; I=2 :

with nitrogen gas packaging) and storage temperature j (j=1: ambient temperature; j=2: 40 °C), and t was storage time (days), the kinetics model for peroxide value of *nila* chips are: P_{11} =0.3833+ 0.3186t, R^2 = 0.92; P_{12} =-1.3357+ 0.3323t, R^2 = 0.98; P_{21} =0.2277+ 0.2856t, R^2 = 0.97; P_{22} =0.6473+ 0.3150t, R^2 = 0.97. The slope value of those linier equation, reffered to zeroth order kinetic model in linier format Q=Q + k(t), was represented the rate constant for peroxide value (k_p) changes during storage as shown in Table 2. It was founded that rate constant on peroxide value for *nila* chips with nitrogen gas in packaging was consistently lower than control where as at ambient temperature storage about 89 percent and at 40 °C storage about 95 persen. This fact indicated that nitrogen application was able to decrease significantly below 90 percent the rate of *nila* chips deterioration based on peroxide value parameters during storage at ambient temperature. The lower value when storage temperature was higher indicated that nitrogen application was not effective on higher temperature. The Q_{10} of peroxide value of *nila* chips with nitrogen gas application was calculated about 1.103, that is higher than control about 1.043 as shown at Table 2. Based on this phenomenon, it is recommended that nitrogen application in packaging of *nila* chips was done at ambient temperature or the lower one.

Table 2. The rate of peroxide value changes of *nila* chips during storage

Ni*la* chip treatment packaging	Constant rate (k_p) at 30 °C	Constant rate (k_p) at 40 °C	Q_{10}
Nitrogen gas application	0.2856	0.3150	1.103
Control (no treatment)	0.3186	0.3323	1.043

Browning Index

Nila chips after packaging at initialy start of storage (zero day storage) has been measured the average value of browning index about 0.215 A_{420nm}. At the end of storage (60 days), browning index of *nila* chips with nitrogen gas packaging was 0.39 and 0.48 for storage ambient temperature and 40 °C, respectively, while the control was 0.42 and 0.57 A_{420nm}. It was observed that browning index of *nila* chips was increased during storage both with nitrogen gas packaging or control under ambient temperature or higher.

Futher analysis data by polynomial orthogonal method was found that the relationship of brwoning index of *nila* chip with storage time, as shown in above figure, was significantly valid described in a linier model, otherwise not significant for others. This analysis was lead to adopt the zeroth order kinetics models, in linier format Q=Q_o + k(t), that was applied before for describing the change of moisture content and peroxide value of *nila* chip during storage. It was supported by many researcher previously as reported by Priyanto (1997) that zeroth order kinetic models had been used and valid for describing browning index development on foodstuf during storage.

Based on zeroth order kinetic model, the formula of browning index development could be set as B= B_o + k_b(t), where k_b is rate constant of browning development. When B_{ij} was defined as browning index of *nila* chip with packaging method i (i=1: control, no treatment; I=2 : with nitrogen gas packaging) and storage temperature j (j=1: ambient temperature; j=2: 40 °C), and t was storage time (days), the kinetics model for browning index of *nila* chips are: B_{11} =0.190+ 0.0039t, R^2 = 0.92; B_{12} =0.211+ 0.0059t, R^2 = 0.93; B_{21} =0.73+ 0.0033t, R^2 = 0.56; B_{22} =0.195+ 0.0047t, R^2 = 0.94. The rate constant of browning index development (k_b) was shown as slope of the equation of those kinetic models and presented on Table 3.

Table 3. The rate of browning index development of *nila* chips during storage

Nila chip treatment packaging	rate constant (k_b) at 30 °C	rate constant (k_b) at 40 °C	Q_{10}
Nitrogen gas application	0,0033	0,0047	1.405
Control (no treatment)	0.0039	0.0059	1.509

The rate contant of browning index development for *nila* chips with nitrogen gas packaging, as shown in above table which consistently lower than control, both at ambient temperature and 40 °C storage. Based on comparison of that rate constant, it is shown that aplication of nitrogen gas for *nila* chip packaging was able to decrease browning index development to 85 percent for ambient temperature and 80 percent at higher temperature (40 °C). Nitrogen gas in this case was kicked the role of water pavour and oxygen to promote browning precusor to be unusable. When atmosphere in the packaging were filled in full size by nitrogen gas, there are no space for water vapor and oxygen, and prevent accelerated reaction of browning formation. Browning reaction, especially in processed food product, during storage at ambient temperatures had been reported by many researcher that it was dominated by Maillard reaction. This reaction was not only contributed to color of product but also perform the odor and taste as whole quality of product. Browning reaction should be managed carefully in foodstuf to submit the best product performance.

Nitrogen gas application in packaging of *nila* chips to prevent browning reaction was faunded not more sensitive than control. It was shown by calculated Q_{10} where presented on Table 3 above, that the Q_{10} value for *nila* chips with nitrogen gas in packaging is 1.405 which is lower than Q_{10} control as 1.509. *Nila* chip with nitrogen gas in packaging was more tolerable to the temperature change. However this pehenomena was one of the advatage where there are no quaranty of stable temperature during storage such as on traditional transportation from village to town and vice versa.

Microbial Plate Count

Microbiological aspect is an important factor of product quality such as *nila* chip because of its original fish composition where potentially resource for microbial life. It was investigated that after packaging when ready to store treatment, average microbial count of *nila* chip is still about of 10^5 cfu/mL. The microbial number on 60 days storage was about 10^6 cfu/mL During the storage, both on ambient temperature or at higher temperature (40 °C), the microbial count of *nila* chip was increased. The incrreasing of microbial count during storage was aproximated by logaritmic model as usually used by microbiologist to describe microbial growth stated by common equation: $N= N_o.10^{dt}$ or in linier formula as $\text{Log N} = \text{Log}(N_o) + dt$ where d is the rate constant of microbial count increasing in log microbial count per time unit. Based on the obtained data, the model of microbial count increasing for *nila* chips during storage are: $\text{Log } N_{11} =0.190+ 0.000745t$, $R^2 = 0.89$; $\text{Log } N_{12} =0.211+ 0.000903t$, $R^2 = 0.74$; $\text{Log } N_{21} =0.73+ 0.000613t$, $R^2 = 0.82$; $\text{Log } N_{22} =0.195+ 0.00096t$, $R^2 = 0.77$. Those equations show that *nila* chip with nitrogen gas in packaging come to be slower than control at ambient temperature, but at higher temperature (40 °C) the slope is greater indicated rapid growth than control. Calculated Q_{10} was founded that the Q_{10} of microbial count of *nila* chip was about 1.564, which was greater than control with Q_{10} about 1.213. This indicated that application of nitrogen gas in the packaging would increase the sensitivity of shelf life product during storage especially in point of view microbial count.

CONCLUSION

The application of nitrogen gas promised many advantages, *nila* chip would be more satisfied in quality during storage. Nila chip stored under nitrogen treatment packaging was potentially to extend of its shelf life. Moisture content, peroxide value and browning index was changed during storage based on linier relationship model between its quality parameter with storage times, while microbial plate count number was suitable expressed by logarithmic model. Nitrogen gas package could be able to decrease deterioration of nila chips during storage at ambient temperature. The rate of quality changes was decreased to 82-89 % when nitrogen applied in the package. The value of Q_{10} for all of quality parameters was higher when nitrogen applied in the package compare to control. Nitrogen in the package was not only to be preservation agent but also increase temperature sensitivity of nila chips quality during storage.

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