COLOR DEVELOPMENT IN COMPLEX MODEL SYSTEM ON VARIOUS TIME AND TEMPERATURE

PEMBENTUKAN WARNA PADA SISTIM MODEL KOMPLEK PADA BERBAGAI SUHU DAN WAKTU PEMANASAN

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Abstract

The research was intended to study the development of color during heat treatment in a model real system consisted of sucrose, sweetened condensed milk, egg and margarine. Research applied was factorial completely randomized design with steaming time consist of 1,2,3 and 4 hours and temperatures consist of 100ºC, and 110ºC as treatment with 3 replication. The changes of color was investigated by using Munsell color system consist of value/lightness, hue angle, and chroma. Browning index was measured by using the absorbance of ethanol extracts at 420 nm in 40.0 mm silica. Amino acid content was observed by using HPLC. Research showed that there were negative correlation between steaming time and temperature with lightness, hue angle, chroma, and amino acid. While for TCD, and browning index indicated positive correlation.

Keywords : Browning, Heat processing, color development, Maillard reaction, real system.

Abstrak

Penelitian ini bertujuan untuk mempelajari pembentukan warna selama pemanasan pada sistim pangan yang terdiri dari gula sukrosa, susu kental manis, telur ayam dan margarin. Penelitian menggunakan rancangan acak lengkap yang disusun secara faktorial. Perlakuan yang digunakan adalah suhu pengukusan terdiri 100ºC, dan 110ºC dan waktu pengukusan terdiri dari 1jam, 2 jam, 3 jam dan 4 jam dengan 3 replikasi. Perubahan warna diamati menggunakan *Munsell color system* yang terdiri dari *value* (*lightness*), *hue angle*, dan *chroma*. Pengukuran *browning index* diamati menggunakan absorbansi pada etanol pada panjang gelombang 420 nm. Pengujian asam amino menggunakan metode *HPLC*. Hasil penelitian menunjukaan adanya korelasi negatif antara suhu dan waktu pengukusan dengan *value* (*lightness*), *hue angle*, *chroma*, dan asam amino, sedangkan untuk *TCD*, dan indeks pencoklatan menunjukkan korelasi positif.

Kata kunci: browning, pemanasan, pembentukan warna, reaksi *Maillard*,

INTRODUCTION

Indonesia's traditional foods generally dominated by carbohydrate, protein and fat which are processed by using heat treatment at periode of time. The application of heat treatment on food processing lead to physical change such as browning. In certain foods like kue delapan jam the acceptability and quality are determined based on the level of browning. In addition color also could indicate chemical changing or chemical reaction which are exposured on foods (Agustini *et al.*, 2014a). Hidalgo and Zamora (2000) stated that the color were used to indicate the sensories quality of foods.

One of the most important reaction which occur during processing and storaging of foods is browning reaction (Simpson, 2012). Basically browning reaction could be classified into enzymatic browning and non enzymatic browning. Enzymatic browning reaction are caused by enzyme such as phenol oxidase, phenolase, monophenol oxidase, diphenol oxidase and tyrosinase.

Non ezymatic browning in foods is divided into four types namely Maillard reaction, lipids peroxidation, caramellization and ascorbic acid degradation (Nursten, 2005 and Hidalgo and Zamora, 2000). Browning reaction in foods especially Maillad reaction is influenced by chemical factor and physical factor. Chemical factors including the type and amount of reactant, pH, moisture content, water activity, and the presence of other compound such as humectans and buffer. While physical factors including processing and storage temperature, oxygen and packaging (Saltmarch and Labuza, 1982; Ajandouz *et al.,* 2001; Nursten, 2005; Kim and Lee, 2008 and Simpson, 2012).

Maillard Reaction is important for food scientist and food industry, since it can affecting foods quality mainly on sensories properties such as color, flavour and taste of foods (Martins and van Boekel, 2005; Matiacevich *et al*., 2005). During Maillard reaction there are various of product formed those affecting foods quality significantly. Maillard Reaction could decline foods nutrition due to decreasing of digestability and possible formation of mutagenic and poissoning substance. It also could improve nutritional value due to formation of antioxidant product (Martins *et al.*, 2001). Food systems that could potentially to undergo Maillard reaction is traditional foods which are rich of reactant for Maillard reaction such as kue delapan jam, kue senting (kue ambtenar) and srikaya. The traditional cake containing protein (source of amino acids), sugar (source of carbonyl compounds) and steamed in a periode of time.

This paper will discuss the changes and the formation of color and a decrease in the amino acid content in a complex model system consisting of sugar, milk, eggs and margarine which are heated (steamed) at various temperatures and times.

MATERIAL AND METHODS

Materials

Materials used were eggs, sucrose, margarine, sweetened condensed milk and reagent for analysis.

The Food system was made by mixing 1,500 g of eggs, 600 g of sugar, 400 g of sweetened condensed milk, and 100 g of margarine by using hand mixer to obtain homogeneous mixture. The mixture was then poured into baking pan that has smeared with margarine. Then it was covered with aluminium foil to prevent water dropplet on the surface. Steaming was then performed according to treatment at 100ºC and 110 ºC for 1h, 2h, 3h, and 4h. The cake was taken out and air cooled immediately after steaming time reached to stop further reaction.

Equipment

The apparatus used were autoclave (Hirayama type Hiclave HVE 50), mixer, baking pan, desiccators, glasswares, incubator, analytical balance, Konica color reader (Minolta, Italy), *UV-Vis Spektrofotometer (Hitachi spectrofluorophotometer F-2500),* and HPLC,

Method

Research appliled was factorial completely randomized design with 3 replications. The treatment was steaming time and temperature. Steaming temperature consist of 100ºC and 110ºC, while steaming time consist of 0 h (t_0) 1 h (t_1), 2 h (t_2), 3 h (t_3), dan 4 h (t_4).

Test on amino acid using *HPLC*. Color measurement using *Konica Minolta color Reader*. Color measurement using Munsell color system consisting of V (*value/lightness*), c (*chroma*), and h (*hue*). The value of L_0, h_0, and c_0 were measured after the dough steamed for 5 minutes so the dough became solid.

As the color index were used Total Color Difference (TCD) calculated using the following equation :

TDC= $(\Delta L^2+\Delta c^2+\Delta h^2)^{1/2}$ .... (1)

Data were presented as a mean and standard deviation of three measurement. Statistical analysis were performed using SAS 9.13 software. Data were statistically analyzed using one-way ANOVA and Duncan tests. Differences were considered significant at 95% ($p<0.05$).

RESULT AND DISCUSSION

Value (Lightness/L)

Changing in color during heat treatment can be seen in figure 1a-d. As shown in figure 1 a-d, the lightness (L), *chroma* (c) and *hue* (h) decreases during heating. Increasing in steaming temperature resulting declining in lightness, chroma and hue angle.

Declining in lightness were caused by Maillard reaction during steaming. Maillard reaction occuring between reduction sugar with amines and amino acid, lipid oxidation and interaction between lipid oxidation product with amino acid to form brown color polymer known as melanoidin (Agustini, 2015). This result consistent with previous studies which stated that a reaction between reducing sugars with amino acids lead to the formation of polymer containing brown pigment compound (Ames, 1990; Batos *et al*., 2012; Nursten, 2005; Saltmarch and Labuza, 1982; Yu and Zang, 2010). Brown polymer which were formed during heating process accumulated so that the color of system become darker so the lightness become lowered. Decreasing in the lightness of food during processing and storage are also reported by Tan *et al.* (2012), Matsuo *et al.*(2012), Gamli (2011), Mohammadi *et al.*(2008), Bosch *et al.*(2007); Coghe *et al.* (2006), Tosun (2004), and Ramirez-Jimenez *et al.* (2000).

Increasing the temperature causes the kinetic energy of the material to be increased so that more molecules reach the activation energy. As a result the oxidation reaction and browning reactions more easily occur. This is in accordance with the Ajandouz *et al.* (2001), Kim and Lee (2008), Nursten (2005), Saltmarch and Labuza (1982), and Simpson (2012) who stated that Maillard reaction on foods affected by chemical and physical factors, such as temperature and time.

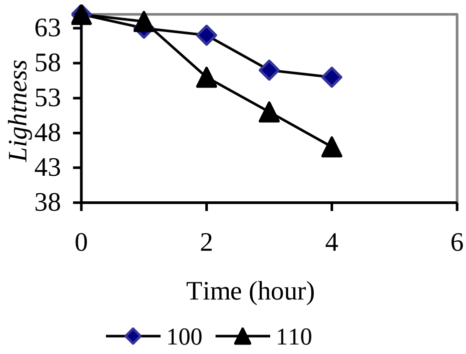

Fig. 1a Changing in Lightness

Analysis of variance showed that steaming temperature, steaming time, and their interaction have significant effect on the lightness ($P<0.01$). The coefficient of determination (R^2) on the effect of temperature, steaming time and the combination on the change in lightness was 0.978. This may imply that the lightness decreases with increasing temperature and steaming time.

Hue angle/h

Hue angle is attribute of a color by which it distinguish wether the object is red, yellow, blue, green, or purple. Munsell color system defines hue as the quality to distinguish one color with other colors. There are five basic colors: red, yellow, green, blue and purple, and there are five intermediate colors are: yellow-red, green-yellow, blue-green, purple-blue and red-purple, arranged in a circle with zero angle **(**0^o, 360^o) is red.

The value of hue angle (h) of the system during heating was ranging from 43 – 84^o which represent red color (0^o-18^o) and yellow color (72^o-90^o). At the beginning of process prior to heating the hue angle was on 84^o which meant that the system on the yellow area. This is consistent with other studies that also reported a decrease in the value of hue angle during processing and storage of food (Ke ren *et al.*, 2005, Guine and Barocca, 2014).

Declining in hue agle were caused by degradation on the pigment carotenoid contained in the raw material mainly in yolk. According to Von-Elbe and Schwartz (2007), carotenoid pigment reflect the colors of yellow, orange or red. Carotenoid easily isomerized by acids, heat, and light. Oxidation causes the carotenoid loss of color which is the color degradation mechanisms.

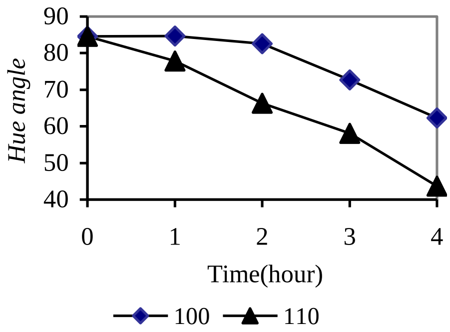

Fig. 1b Changing in Hue angle

Analysis of variance showed that steaming temperature, steaming time, and their interaction have significant effect on the hue angle (P<0.01). The coefficient of determination (R^2) on the effect of temperature, steaming time and the combination on the change in hue angle was 0.99. This may imply that the hue angle decreases with increasing temperature and steaming time.

Chroma (Saturation/c)

Chroma is the saturation of color. Colors of low chroma are sometimes called “weak,” while those of high chroma are said to be “highly saturated,” “strong,” or “vivid.” The scale start from zero, for neutral colors. The chroma scale for normal reflecting materials extends beyond 20 in some cases. Fluorescent materials may have chromas as high as 30 (www.munsell.com)

At the beginning of heating, chroma was located on 27.2 scale, which mean that the system is on strong yellow color. During steaming chroma decreased. This result is consistent with other studies that also reported a decrease in chroma scale during processing and storage of food (Gamli, 2011; Jaiswal and Abu-Ghannam, 2013; Reyes *et al.*, 2007).

Changing in chroma during heating due to carotenoid pigments degradation and the formation of brown pigments due to the Maillard reaction between carbonyl compounds present in milk and amino acids of eggs. In addition there were oxidation of lipids derived from egg yolks and margarine. Degradation of carotenoid pigment causes the yellow color becomes faded, followed by the formation of brown color shifting the chroma scale.

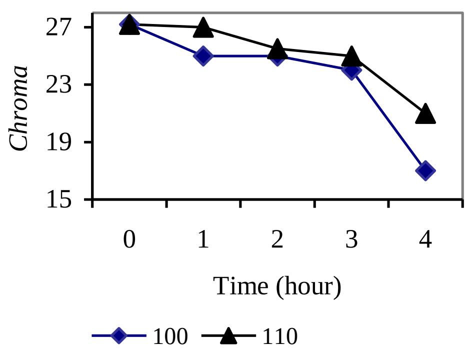

Fig. 1c Changing in Chroma

Analysis of variance showed that steaming time, and their interaction have significant effect on the chroma (P<0.01). The coefficient of determination (R^2) on the effect of temperature, steaming time and the combination on the change in chroma was 0.91. This may imply that the increase in temperature and steaming time will decrease the chroma.

Table 1. Effect of Temperature and Time on Color

Waktu	L		H		C		TCD	
	100ºC	110ºC	100ºC	110ºC	100ºC	110ºC	100ºC	110ºC
1 jam	63±0,21a	64±0,81a	84,7±0,19a	77,9±0,21a	25±0,21a	27±1,18a	3±0,37a	7±,91a
2 jam	62±0,75a	56±2,0b	82,5±0,11a	66,3±0,85c	25±0,58a	25,5±1,3a	4±0,88a	20±0,61b
3 jam	57±1,25b	51±0,51b	72,7±0,59b	58,1±2,74c	24±0,89a	25±1,45a	14±1,53b	30±1,73c
4 jam	56±0,95b	46±0,97c	62,3±0,42c	43,7±1,94d	17±1,37b	21±1,08a	26±2,0c	45±1,18d

Data were expressed as mean ± standard deviation (n=3).
[a-b] Values in each column with different superscripts are significantly different (P<0.05).

Table 2. Effect of Temperature and Time on browning index and amino acid.

Waktu	IB		Amino Acid.	
	100ºC	110ºC	110ºC	100ºC
1 jam	0,21±0,007a	0,25±0,004b	8,67±0,49a	7,92±0,30b
2 jam	0,26±0,003b	0,31±0,010c	8,42±0,43a	7,75±0,31b
3 jam	0,26±0,003b	0,33±0,006c	8,26±0,59a	7,59±0,34b
4 jam	0,27±0,003b	0,34±0,004c	7,63±0,45b	6,70±0,20c

Data were expressed as mean ± standard deviation (n=3).
[a-b] Values in each column with different superscripts are significantly different (P<0.05).

Total Color Difference (TCD)

Total color difference (TCD) is one of the best parameter to describe the color change, because it is a combination of the parameters of lightness, chroma and hue angle (Ibarz *et al.,* 2000). Figure 1d shows that TCD increases linearly during steaming at various temperatures. This suggests that the formation of a brown color is closely related to changes in TCD. Changes in the value of TCD is a function of changes in temperature and time.

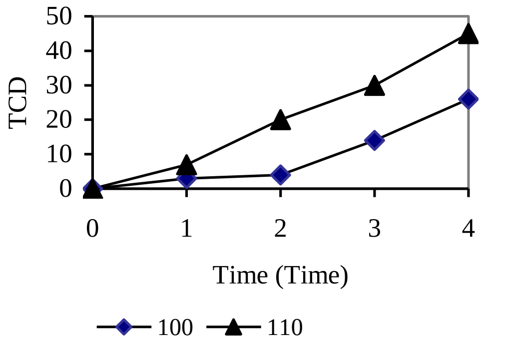

Fig. 1d. Changing in TCD

Analysis of variance showed that steaming time, steaming temperature and their interaction have significant effect on the TCD (P<0.01). The coefficient of determination (R^2) on the effect of temperature, steaming time and the combination on the TCD was 0.99. This may imply that increase in the temperature and steaming time will increase the TCD.

Browning Index(IB)

Tablel 2 indicates that the steaming time and temperature causes increase in browning index. This was seen by the increase in absorbance values during steaming. The increase in steaming time cause a rise in absorbance values, as well as an increase in steaming temperatures cause a rise in absorbance values. The longer the steaming time the higher the browning index. Increasing in absorbance value indicated that the more brown polymer (melanoidin) was formed. Melanoidin formation showed that the browning reaction occurs primarily during the steaming was Maillard reaction (Agustini *et al.,* 2014b). This result is accordance with various studies that suggest the browning reaction is affected by the length of the heating (Ames, 1998).

Increasing in steaming temperature also resulted in an increase in the absorbance value. This means that the rate of browning reaction increases with increasing temperature and steaming time. During steaming reaction occurs between lactose with amino acids derived from proteins to form Amadori compounds which in turn form brown polymers. In addition the fat content in the materials undergo auto oxidation reaction due to the heat treatment to form a brown polymeric compound (Agustini *et al.,* 2014b). Brown color can also be formed from auto oxidation of lipids into hydroperoxide, which are then degraded to aldehydes and hydro carbons. Hydro peroxides and aldehydes polymerize into a stable product and the product is brown (Nawar, 1996; Hidalgo and Zamora, 2000; Kerler *et al.*, 2010). In the food processing, reaction of lipid oxidation products with amines, amino acids and proteins cause browning (Van Boekel, 2006; Nursten, 2005; Zamora and Hidalgo, 2005; Hidalgo and Zamora, 2000).

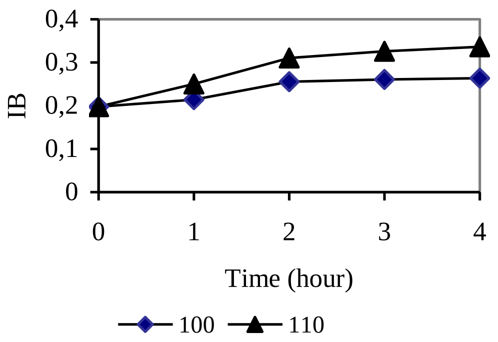

Fig. 2. Changing Browning Index

Analysis of variance showed that steaming time, steaming temperature and their interaction have significant effect on the browning index (P<0.01). The coefficient of determination (R^2) on the effect of temperature, steaming time and the combination on the browning index was 0.977. This may imply that increasing in temperature and steaming time will increasing the browning index. Duncan test indicated that steaming at 100ºC significantly different with at 110ºC for each steaming time. Steaming for 1 hours differ to 2 hours, 3 hours and 4 hours.

Amino Acid

The amino acid in this study derived from egg whites and sweetened condensed milk. The test results showed that during steaming there were changes in amino acids content. Declining in amino acid due to amino acid reacts with the carbonyl compounds in the system mainly glucose and lactose. The reaction between amino acids with carbonyl compounds will produce a brown polymeric compounds known as Maillard reaction (Ames, 1990; Batos *et al*., 2012; Nursten, 2005; Saltmarch and Labuza, 1982; Yu and Zang, 2010). The decreasing in the amino acid, lightness, hue angle and chroma as well as increasing of TCD and browning index indicated there are Maillard reaction during steaming.

The increasing in steaming temperatures also resulted in decreasing the levels of the amino acid. Similarly the increasing in steaming time also cause a decreasing in amino acid content. This means that the temperature and steaming time will affect on the amino acid content. The longer the steaming, the lower of amino acid content.

Maillard reaction starts from the condensation reaction between the carbonyl group of aldose and free amino group of the amino acid to form N - substituted aldosylamin. Eggs contain all kinds of amino acids, such as amino acid lysine. The reaction of lysine with glucose provides Ɛ-N-deoxifructolysin. Reaction of lactose with lysine will produce residue lactulosillysine, which further degraded during heating to produces N-carboxymethyllysine (CML), pentosidine, pyrraline, or oxalic acid monolysinylamide/OMA (Ditrich *et al.,* 2006).

Heat treatment will also produce protein - crosslinker lysinoalanine (LAL). LAL is formed from the condensation of lysine with dehydroalanin. LAL is found in mozzarella cheese, baby food, UHT milk and sterilized milk (Cattaneo *et al*., 2008; van Boekel, 1998).

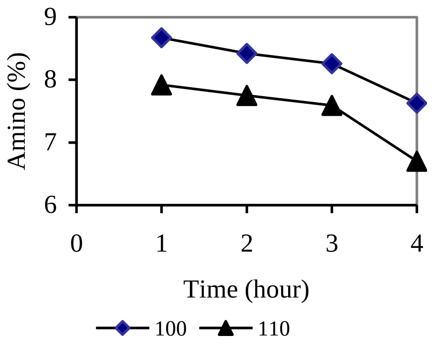

Fig. 3. Changing in Amino Acid

The longer the steaming, the more amino acid lysine which react to form melanoidin and LAL thereby reducing the amino acid residue lysine available in the system. This is consistent with the literature that states that a decrease in the concentration of lysine is the most significant consequence of the Maillard reaction (Cattaneo *et al*., 2008; van Boekel, 1998). Ramirez- Jimenez *et al*. (2001) stated that the decline in lysine in bread baked until brown was about 40 %. The decline of lysine in commercial dairy which stored for a year were maximum of 10% at a temperature of 32ºC and 23% at 55ºC (Ramirez - Jimenez *et al.,*2004). Fernandez- Artigas *et al.* (1999) reported that the loss of lysine during toasting of bread ranges between 14-29%. Processing using a roller dryer at rice-corn-soy causes a loss of 53% lysine.

Analysis of variance showed that steaming time and temperature have significant effect on the amino acid ($P<0.01$), while their interaction have not. This may imply that increasing in temperature and steaming time will decrease amino acid content. Duncan test indicated that amino acid in the system which were steamed at 100ºC significantly different with at 110ºC. The system which were steamed at 110ºC as long as 4 hours significantly different with those were steamed for 1 h, 2 h and 3 h.

CONCLUSION

The reseach showed that the temperature and steaming time significantly affect the formation of the color and amino acid content. During steaming there were color formation and decreasing the amino acid content. Formation of brown color in the system during steaming indicated by the increase of TCD, and browning index, as well the decrease of the amino acid. This means that in the food system which contains proteins, fats, and sugars such as *bolu senting*, *srikaya* and *kue delapan jam*, the higher the temperature applied the faster lossing in nutrient content (amino acid) and color formation. Heating in the long periode of time lead to changes in food color where the color becomes darker.

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