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Research Article

Thermal Degradation Kinetics of Capsaicin on Drying of Blanching-Brine-Calcium Pretreatment Red Chili Pepper

Uma Fadzilia Arifin, Mohamad Djaeni*

Department of Chemical Engineering, Faculty of Engineering, Diponegoro University Jl. Prof. Soedarto, SH, Tembalang, Semarang 50275, Indonesia

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Abstract

Post-harvest red chili pepper (*Capsicum frutescens*) has highly capsaicin as bioactive compound and moisture content. However, capsaicin is the responsible bioactive compound in chili for hot sensation that easy to degrade by partial oxidation caused introduction of heat in drying process. The objective of this research was to investigate kinetics of capsaicin degradation in the drying process under blanching-brine-calcium pretreatment and various temperatures. For this purposes, chili provided local farmer was pretreated using blanching-brine-calcium pretreatment. Afterward, they were dried at 40, 50, 60, and 70 °C for 8 hours. Degradation of capsaicin content was observed every 2 hours using Thin Layer Chromatography (TLC). Results showed kinetics of capsaicin degradation was categorized as second order reaction. At the same temperature and time, capsaicin retention of blanching-brine-calcium pretreated chili has highest value. The temperature dependence of the capsaicin degradation rate was analyzed using Arrhenius correlation. The activation energy for degradation rate of capsaicin during drying was around 45.10367 kJ/mol.K. It indicated the degradation rate increased as well as increased the temperature at the same time.

Keywords: Capsaicin; Kinetics; Pretreatment; Red Chili Pepper Drying

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1. Introduction

The red chili pepper is abundantly used in making spice sensation for foods. Certainly, hotness or spice sensation of chili is important characteristics for consumer acceptance. The pungent sensation of red chili pepper is due to existence of bioactive compound called capsaicin and its capsaicinoid analogs [1-3]. Capsaicin (trans-8-methyl-N-vanillyl-6-nonenamide) is fat-alcohol-oil soluble, colorless, crystalline and odorless [4-6]. It also is a major compound of capsaicinoids (alkaloid) that are widely applied in pharmaceutical and clinical product [7]. As one of the natural products, it was reported that capsaicin has therapeutic properties currently employed as antitumor [8], anti-mutagenic [9], antioxidant compounds [7], antimicrobial [3], anticancer [10], cardio-protection [11], anti-obesity [12], anti-diabetic [13], and anti-inflammatory effect [14].

Red chili pepper is highly demanded in the world due to its great benefits. Unfortunately,

* Corresponding Author.
E-mail: moh.djaeni@live.undip.ac.id (M. Djaeni)
Telp: +62-24-7460058, Fax: +62-24-76480675

red chili pepper is perishable vegetable that can easy to rotten and it cannot be stored for long. The post-harvest red chili pepper has relatively high moisture content [15]. It creates favorable conditions for harmful microorganism, yeast, enzymatic and chemical activity that leads deterioration process for products in a few days (around 3 days) after harvest [16]. As a response, drying is one of the completion methods to extend product life during storage by removal moisture content [17].

Drying is preservation method for foods that involves the thermal process to vaporize moisture content [18]. This methods have been reported and can change undesirable physical and chemical properties of product [19,20]. It can be resulted that drying cause the loss of some nutritional and other quality factors [21]. Absolutely, the spice level of dried chili is important characteristics for consumer acceptance. However, capsaicin as bioactive compound that responsible the hotness sensation has been found easily to decrease in greater thermal and oxidative degradation [22]. This is the reason that various pretreatments such as blanching, osmotic dehydration and chemical pretreatment in drying techniques have been adopted to accelerate drying rate, reduce drying time and protect the nutritional properties of fresh vegetables during drying process [23].

Previous study concerned on capsaicin properties and kinetics during storage. There have been limited studies focused on kinetics of capsaicin during drying process with drying pretreatment. Therefore, the main objective of this study was to investigate kinetics of capsaicin degradation in the drying process under blanching-brine-calcium pretreatment and various temperatures. They include order reaction, constant of degradation rate and energy activation in degradation process during drying. The information obtained to establish the best drying condition as well as favorable pretreatment and temperature for red chili pepper to retain higher capsaicin.

2. Materials and Methods

2.1 Materials

The fresh red chili pepper was obtained from a local market in Semarang, Central Java, Indonesia. It has 78.08 ± 3.65 % (wet basis) moisture content by ten replications using gravimetric refers to the AOAC (2000) methods [22]. Distilled water, calcium chloride (food grade with purity 94.0 %, Merck Germany), and sodium chloride (purity 97.99 %, UniChem Candi Indonesia) were purposed as pretreatment materials. Capsaicin standard (purity 97.0 % p.a) was provided by Sigma, USA used for capsaicin content analyzed in red chili pepper. Methanol (purity 95 %, Merck Germany) was used as solvent in extraction. Toluene, chloroform and acetone were used as a mobile phase in TLC. Silica gel 60 F_{254}-precoated TLC plates (Merck, Germany) as adsorbent and then it is known as the stationary phase. The capsaicin in red chili pepper was carried out by TLC dual wavelength chromate-scanner Shimadzu CS-930, Japan.

2.2 Methods

The study was conducted in the schematic diagram as illustrated in Figure 1. Briefly, the methods consisted of several main steps that involved drying pretreatment, experiment for red chili pepper drying, capsaicin analysis, kinetics model development, and validation of model with experimental data. The models were used for estimating kinetics parameter of capsaicin degradation at various drying conditions.

2.2.1 Drying pretreatment of red chili pepper

Drying pretreatment aimed to enhance the drying kinetics, retain the nutritional and physical quality of product, and preserve volatile compound [24,25]. In this case, various methods of drying pretreatment were conducted that involved blanching, osmotic dehydration and chemical pretreatment and combi-

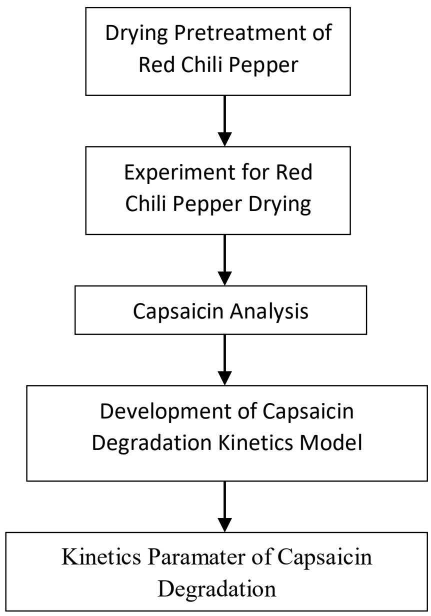

Figure 1. The schematic diagram of research

nation of these methods. The blanching was conducted by dipping the red chili pepper in hot distilled water for 3 minutes at 80-90 °C. It was then quickly cooled in cold water [16,26]. The osmotic dehydration was performed by soaking red chili pepper in hypertonic solution 10 % (w/v) of sodium chloride (brine solution) for 30 minutes [16]. Meanwhile, the chemical pretreatment was conducted by immersing red chili pepper in 1 % $CaCl_2$ solution for 30 min [27]. The combination pretreatments were carried out by combining of blanching-brine-calcium pretreatment, successively. All of pretreated red chili peppers were dried at certain drying temperatures as described in the section 2.2.2. The results were also compared with red chili pepper drying without pretreatment.

2.2.2 Procedure of red chili pepper drying

The red chili pepper drying was carried out in a tray dryer with air as drying medium. The schematic of the dryer system was depicted in Figure 2. The drying process used air with velocity of 0.22 $m.s^{-1}$ at certain temperature. Two hundred gram of sample was dried at drying temperature of 40, 50, 60, and 70 °C for 8 hours, respectively. Every 2 hours, amount of 5 gram of chili product was taken from the dryer. It was then sent to Thin Layer Chromatography (TLC) unit to be analyzed capsaicin content.

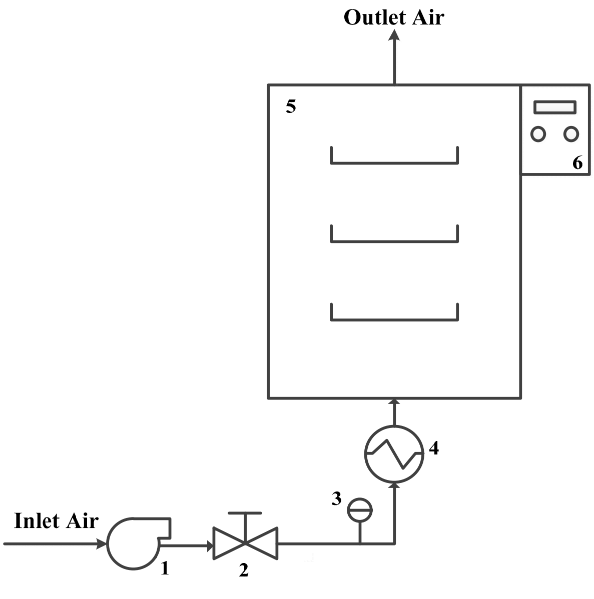

Figure 2. The schematic diagram of the tray dryer system (1. Blower; 2. Valve; 3. Relative humidity and thermal indicator; 4. Heater; 5. Tray column dryer and 6. Automatic thermo regulator)

2.2.3 Determination of capsaicin by Thin Layer Chromatography (TLC)

Thin layer chromatography (TLC) was one of methods that can be executed to quantitative measuring capsaicin content. The analyses were carried out using TLC dual wavelength chromate-scanner Shimadzu CS-930, Japan [28]. For the determination of capsaicin content, around 0.1 grams of the chili sample was put in a 10 mL flask containing 2 mL of methanol. Sample was stirred using vortex for 3 minutes on water bath at temperature 60 °C and was centrifuged for 2 minutes, afterwards. Supernatant was taken and the extraction was replicated 3 times. Supernatant was evaporated using gas nitrogen and then it was then added 200 μL methanol. Furthermore, standard capsaicin and 20 μL supernatant were spotted in silica gel 60 F_{254}-precoated TLC plate. This plate was put into chamber containing saturated mobile phase toluene-chloroform-acetone (45:25:30 v/v). It was eluted to limit mark, lift and drain, afterwards. The sample was measured by densitometry at 228 nm wavelength and retardation factor (R_f) 0.61 using TLC dual wavelength chromate-scanner Shimadzu CS-930 [28]. Quantitative analysis of capsaicin content was done by comparative the peak area with the standards of serial dilutions in 0.125, 0.25, 0.5, 1, 2, and 4 μg capsaicin concentrations. The standard regression curve of capsaicin by TLC can be seen in Figure 3.

2.2.4 Kinetics models of capsaicin degradation

Nutritional degradation kinetics during drying is essential to give information about predicting the losses of nutrient content in the

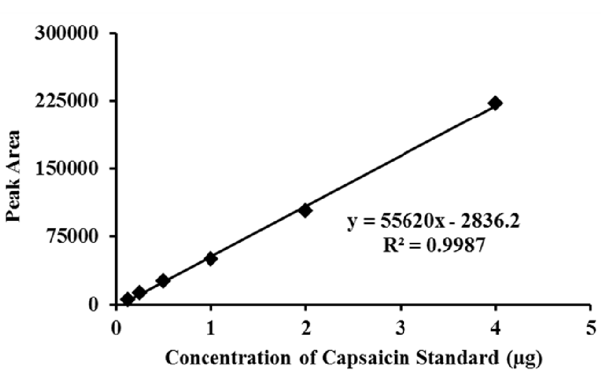

Figure 3. Standard regression curve of capsaicin by Thin Layer Chromatography (TLC) in the range 0.125, 0.25, 0.5, 1, 2, and 4 μg capsaicin concentrations

product [29]. Experimental data of capsaicin concentration versus drying time were plotted in various empirical kinetics order models, respectively. Kinetics parameters were estimated by Polymath Educational 6.0 Software. The plotting of data in various kinetics order models were expressed by linear regression. The various kinetics order models were expressed in mathematical equation as can be seen in Table 1.

Based on Table 1, C is the concentration of capsaicin and t is time. The concentration of the capsaicin at time zero is C_0, n is reaction order and k is the reaction constant (time^{-1}). The slope in linear regression was resulted by plotting data referred the value of rate constant.

Generally, the drying process needs amount of heat provided by hot air [30]. Reis *et al.* [31] reported that capsaicin content degraded by the increase of temperature. The constant rate of capsaicin degradation corresponding to the temperature change can be expressed with the Arrhenius correlation as can be seen in Equation 5.

$$ln\, k = -E_a / RT + k_0 \quad (5)$$

where, k_0 is frequency factor or Arrhenius constant (C.time^{-1}), R is the universal gas constant (8.314 Joule.mol^{-1}.K^{-1}), and T is absolute temperature (K).

3. Results and Discussion

3.1 Degradation of capsaicin under various pretreatments

Capsaicin is the responsible bioactive compound for hot sensation in red chili pepper [2]. Based on quantitative analysis using thin layer chromatographic analysis, post-harvest red chili pepper contains capsaicin around 1491.37 μg/gram red chili pepper. The capsaicin is one of bioactive compound that can be oxidized or degraded due to the introduction of heat during drying process. The longer drying time can decrease capsaicin content in red chili pepper, significantly [22]. The drying pretreatments are the techniques that can be applied to shorten drying time. Thus, it can retain bioactive and other nutrition compound in red chili pepper. The effect of various drying pretreatments on capsaicin content can be seen in Table 2.

The results showed that drying pretreatment can retain capsaicin content in red chili pepper. Perhaps, the pretreatments can inactivate peroxidase enzyme. By combination drying pretreatment, capsaicin retention can be positively improved. As mention in literature, this enzyme can degrade vanillylamine of capsaicin by oxidation process [32].

3.2 Kinetics of capsaicin degradation at blanching-brine-calcium pretreatment under various temperatures

Capsaicin content in red chili pepper can be degraded during drying process. Temperature and drying time influence degradation rate of capsaicin in red chili pepper. In this case, chili was dried at blanching-brine-calcium pretreatment under various temperatures and analyzed using TLC every 2 hours for 8 hours, as depicted in Figure 4.

Based on Figure 4, it can be known that the higher and longer drying time degraded capsaicin content significantly. This result was comparable with previous study pointed by Renate *et al.* [33]. In this research, the kinetic order model was developed to find the best fitting according to Table 1. The best of the kinetics order models was considered by R^2 at highest value [25,34]. Afterwards, kinetics parameters such as constant of degradation rate (k) and activation energy can be predicted by the suitable kinetics models. The result of data fitting based on various kinetics order models can be seen in Table 3.

Table 3 showed that data fitting with n^{th} kinetics order model in various temperatures had highest R^2 value. This implied that n^{th} kinetics order was suitable model to describe behavior of capsaicin degradation during drying. Accord-

Table 1. Mathematical equation for kinetics order models

Kinetics Order	Differential Equation	Linear Regression	
Zero order	$-dC/dt = k$	$C = C_0 - kt$	(1)
First order	$-dC/dt = kC$	$\ln C = \ln C_0 - kt$	(2)
Second order	$-dC/dt = kC^2$	$1/C = 1/C_0 - kt$	(3)
n^{th} order	$-dC/dt = kC^n$	$C^{1-n} = C_0^{1-n} + (1-n)\, kt$	(4)

ing to Table 3, it was known that n^{th} value in kinetics order models under various temperatures close to second order. Meanwhile, R^2 value differences in second and n^{th} orders were not significant. Therefore, the second order can be considered as suitable kinetics model for capsaicin degradation in red chili pepper drying.

The structure of capsaicin (trans-8-methyl-N-vanillyl-6-non-enamide) can change under higher temperature (upper 40 °C) as well as longer drying time. The capsaicin consists of vanylamine and decanoic acid with double bond [33]. Katritzky *et al.* [35] divided the capsaicin molecule into three parts, namely aromatic ring, amide bond and hydrophobic bond. The aromatic ring especially in 3 and 4 position bonds was easily substituted by phenol groups.

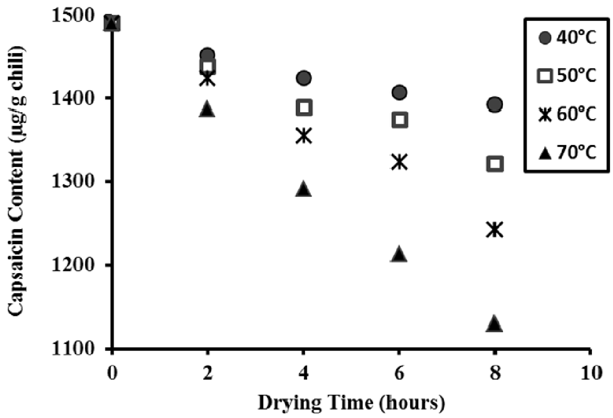

Figure 4. Kinetics of capsaicin degradation at blanching-brine-calcium pretreatment under various temperatures

Table 2. The effect of various drying pretreatments on capsaicin content in the red chili pepper dried at 70 °C during 8 hours

Drying pretreatment	Sample (g)	Spouting of sample (µg)	Peak area	Capsaicin content in sample (µg)	Capsaicin content in dried chili (µg/g)
Non-pretrated dried chili	0.1019	20	95289.1	1.764	865.66
Blanching dried chili	0.1065	20	113144.6	2.085	978.98
Brine dried chili	0.1005	20	120764.6	2.222	1105.59
Calcium dried chili	0.1019	20	101962.6	1.884	924.53
Blanching-Brine-Calcium dried chili	0.1016	20	124797.1	2.295	1129.30

Table 3. Kinetics order models for chili drying using blanching-brine-calcium pretreatment under various temperatures

Kinetics Order (*n*)	Temperature (°C)	*k*	R^2
0	40	13.689	0.9343
	50	21.349	0.9701
	60	30.477	0.9868
	70	46.382	0.9951
1	40	0.0095	0.9405
	50	0.015	0.9737
	60	0.022	0.9806
	70	0.0348	0.9953
2	40	7E-06	0.9485
	50	0.00001	0.9762
	60	0.00002	0.9838
	70	0.00003	0.9982
2.788	40	2.122E-08	0.9533
2.318	50	1.056E-06	0.9781
1.745	60	0.0001	0.9869
1.684	70	0.0002533	0.9993

The hydrophobic bonds especially the octyl and benzyl chains were very easily substituted by the other group function. These changes played a significant role in capsaicinoid activity [35]. The change of capsaicin structure can cause vanillylamin condensation and its fatty acid derivatives that can produce vanillyl nonanoate, as depicted in Figure 5. Introduction of heat during drying process caused the change of capsaicin to be vanillyl nonanoate [35]. The vanillyl nonanoate was analog capsaicin compound that can influence low hot sensation in red chili pepper [36].

3.3 Effect temperature for kinetics of capsaicin degradation

Kinetics of capsaicin degradation depends on temperature. The influence of the constant of degradation rate on various drying temperatures was correlated referring to the Arrhenius, as depicted in Figure 6. The higher temperature increased the constant of degradation rate. Here, the activation energy (E_a) was about 45.10367 kJ/mol K. Hence, the degradation of capsaicin content in red chili pepper at certain operational temperature, can be accurately estimated.

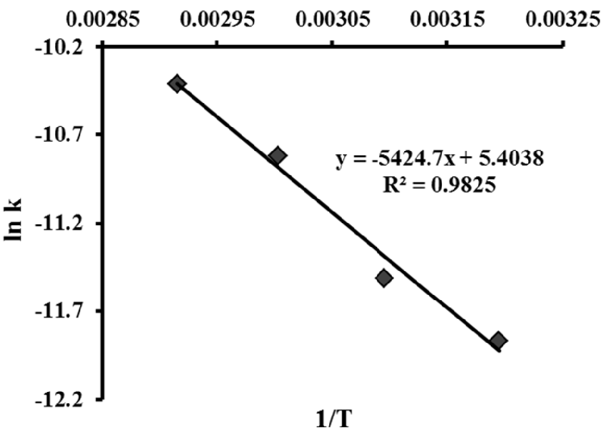

Figure 6. Arrhenius correlation in kinetics of capsaicin degradation

4. Conclusions

Capsaicin is the responsible bioactive compound for hot sensation in red chili pepper. Post-harvest red chili pepper contains capsaicin around 1491.37 μg/gram red chili pepper. Blanching-brine-calcium pretreatment before red chili pepper drying showed the highest capsaicin retention. Kinetics of capsaicin degradation was categorized as second order reaction. Hence, the capsaicin degradation in red chili pepper can be well predicted at various temperatures and pretreatments.

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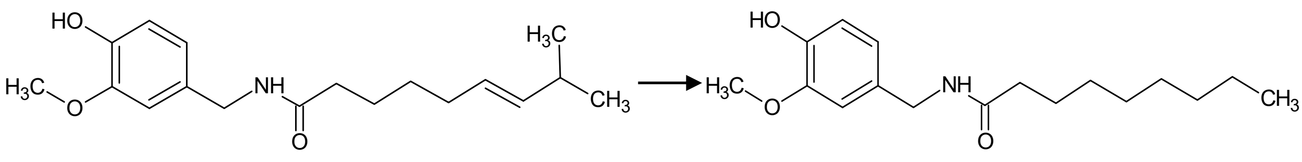

Figure 5. Structure change of capsaicin [35] to vanillyl nonanoate [36] by the drying process

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