3. EFFECT OF PROCESSING ON THE ANTIOXIDANT CONTENT OF CHILLI PEPPERS

Spices and herbs such as chilli peppers are very effective as an antioxidant because they have the excellent antioxidant capacity. Antioxidants have benefit to prevent lipid oxidation. Lipid oxidation can cause rancidity on food that contains oil and fat. Antioxidants are proved to minimize the rate of lipid oxidation in food (Embuscado, 2015). The addition of antioxidant in food industries can protect and stabilize colour, aroma, and nutritional quality especially vitamin (Moure et al., 2001).

When food that contains fat and oil react with the presence of oxygen, it will cause the formation of hydroperoxides and peroxides, which later carbonyl compound will be formed as a secondary product of oxidation. Besides the presence of oxygen, lipid oxidation can be caused by metal ion transition, light, heat, and also moisture. Storing the food in a place at low temperature and protected from light are highly recommended to prevent lipid oxidation. Removing the oxygen and metal catalysts can also be another option to prevent the rate of lipid oxidation (Embuscado, 2015).

Free fatty acid and unsaturated fatty acid are more convenient to be invaded by oxidizing agent obtaining free radical. If unsaturated fatty acid (RH) losses their hydrogen atom (H), then it will be transformed into alkyl radical (R•) and lipid oxidation started (Reaction 1; Figure 5). Lipid oxidation can also initiate by the presence of singlet oxygen, hydroperoxide (ROOH), and other radical compounds (R•). The alkyl radical then can react with oxygen (O_2) which will then form a high energy peroxyl radical (ROO•; Reaction 2; Figure 5). Peroxyl radical can separate a hydrogen atom from another fatty acid and generate a new free alkyl radical and hydroperoxide (ROOH), the main product of lipid oxidation (Reaction 3; Figure 5). This process then enhances hydroperoxide into alkoxy radical (RO•) and hydroxyl radical (HO•; Reaction 4; Figure 5). An antioxidant will obstruct the lipid oxidation reaction by donating the hydrogen atom (H) to free radical species (Brewer, 2011). Lü et al. (2010) reported that hydroxyl radical (OH•) is the most reactive and toxic radical species than other.

(1) R:H + O::O + Initiator → R• + HOO•

(2) R•+ O::O → ROO•

(3) ROO•+ R:H → ROOH + R•

(4) RO:OH → RO• + HO•

Figure 6. Lipid Oxidation Reaction

According to Brewer (2011), there are some mechanisms of antioxidant that can inhibit the establishment of free radicals. Free-radical scavenging is one of the known mechanisms by which antioxidants inhibit lipid oxidation (Bonde et al., 1997). Free radicals are involved in many disorders like neurodegenerative diseases, cancer and AIDS (Pourmorad et al., 2006). The antioxidant compound will donate their single electron that is hydrogen (H) to a free radical species. After electron transfer process, antioxidant radicals are formed, because the radical character is transferred to the antioxidant (Cadenas, 1997). The antioxidant radical is more stable than free radical species in food, caused by resonance delocalization along the structure of the phenolic ring. Bond dissociation among a phenolic hydrogen and oxygen is one of the reasons that can affect antioxidant effectiveness to scavenge free radical in food. If the bond dissociation energy is low, the antioxidant radical is more stable and the hydrogen donors are more effective which means it's a good antioxidant (Choe & Min, 2015).

Besides free radical scavenging activity, Brewer (2011) said some autoxidation can be inhibited by an antioxidant mechanism such as metal chelating, singlet oxygen quenching that can be avoiding the establishment of peroxides, cracking the autoxidative reaction, and diminishing localized oxygen concentrations. Establishment of radicals due to separating hydrogen atoms can be catalyzed by metals. Hydroxyl radical can be generated by catalyzing disintegration of hydroperoxide. Metal chelators reduce oxidation process (Choe & Min, 2015).

Fresh chilli peppers are the best source of antioxidant content compared to processed chilli peppers (Ornelas-Paz et al., 2010). Shobana & Naidu (2000) found that boiling chilli pepper for about 30 minutes not only retain the antioxidant capacity but also increase the antioxidant activity of chilli pepper. The feasibility release of bound antioxidants during heat treatment could lead to the higher antioxidant activity. On the other hand, Hwang et al. (2012) investigated that boiling will significantly reduce antioxidant activity of red chilli peppers. Boiling result in a significant decrease of antioxidant activity (42.0-60.5%) and then followed by steaming (23.5%), roasting (11.6-15.4%), and stir-frying (4.6-15.8%). It might be caused by water thermal processing. Antioxidants were leaching into the water during boiling and result in a higher loss of antioxidant activity. High temperature causes a reduction in the free radical-scavenging activity. Fresh chilli peppers contain the highest amount of antioxidant activity than dried chilli pepper. The lower temperature of drying can result in slower loss of antioxidant activity (Reis et al., 2013). On the other hand, Wangcharoen & Morasuk (2009) reported that drying process at 100 and 121^{o}C can cause increasing of antioxidant capacity.

Ajolo et al. (2005) classified values of antioxidant activity as high (70% inhibition), intermediate (40-70% inhibition), and low (<40% inhibition). Chuah et al. (2008) reported that increasing the boiling time from 5 to 30 minutes can cause considerable loss of antioxidant from six varieties of coloured chilli peppers from *Capsicum annuum* from the initial level. Boiling for 5 minutes might reduce the radical scavenging activity to below 77% from its initial level, while if boiling time is prolonged to 30 minutes, it might result in reducing of scavenging activity to 64%. The authors indicated that antioxidant could have leached into boiling water. Total of the radical scavenging activity values in cooking water and cooked tissue after being boiled for about 5 minutes didn't significantly different from the fresh chilli peppers tissues, while boiling for about 30 minutes showed significant reduction of total radical scavenging activity of cooking water and cooked tissues in orange paprika, yellow paprika, and green paprika compared with the fresh tissues. Nevertheless, in both the tissues and cooking water on red pepper and red paprika when boiled for 30 minutes showed resistance to deterioration in their radical scavenging activity. Interestingly, during 5 minutes and 30 minutes of boiling had shown the increase of radical scavenging activity in the green pepper on a total of both the tissues and cooking water.

According to Amaguchi et al. (2001), the increase of radical scavenging activity values after boiling due to suppression of the oxidation of antioxidants because of thermal inactivation of oxidative enzyme, discharge of greater antioxidant compounds by thermal destruction of cell wall, and can also be caused by liberation a lot of antioxidant compounds during thermal treatment that cause broken cell wall. However, Chuah et al. (2008) investigated that microwave heating for 5 minutes and stir-frying for 5 minutes didn't show significant differences when compared with the fresh chilli peppers. The authors also concluded that red, orange and yellow paprika will retain higher antioxidant during cooking compared with red and green peppers.

Chilli peppers have many antioxidant compounds such as phenolic, flavonoid, capsaicinoid (Loizzo et al., 2015; Li-E et al., 2008), ascorbic acid, and carotenoid (Pugliese et al., 2013). Based on an experiment conducted by Alvarez-Parrilla et al. (2011), the main contributors to the antioxidant activity of processed or fresh chilli peppers are gallic acid and catechin, while ascorbic acid and capsaicinoid have lower values. Total phenolic content and flavonoids are reported as gallic acid and catechin equivalents, respectively. Antioxidant activities in red and green chilli peppers are different depending on carotenoid, flavonoid, and phenolic compounds. Red chilli peppers contain higher pigment compound such as carotenoid, and flavonoid that can contribute to antioxidant activity, while the green chilli pepper has a deficiency of these compounds. It indicates that red chilli peppers have higher antioxidant activity compared with green chilli peppers (Sun et al., 2007). Zhang & Hamauzu (2003) found that in the phenolic extract, green chilli peppers have higher antioxidant activity than yellow and red chilli peppers. In the methanol extract, red chilli peppers have the higher antioxidant activity than yellow and green chilli peppers. They said that green peppers have a higher level of phenolic content and radical scavenging activity of phenolic content is related to phenolic extract, however radical scavenging activity that found in the methanol extract is higher than phenolic extract. This result is due to the greater level of ascorbic acid that left behind in the methanol extract. Ascorbic acid and phenolic compounds contained in chilli peppers are the main antioxidants that contribute to the radical scavenging activity in the methanol extract. Changes of antioxidant capacity of chilli pepper during processing are shown in table 1.

Table 1. Effect of Processing Methods on Antioxidant Capacity

Type	Method	Temperature (°C)	Time (min)	Chilli:water (w/v)	Weight	Size	Extraction	Changes of antioxidant capacity (%)	References
Capsicum chinense Jacqui	Drying	45	NA	NA	NA	NA	Methanol and acetone	-38.528[1]	(Reis et al., 2013)
Capsicum chinense Jacqui	Drying	55	NA	NA	NA	NA	Methanol and acetone	-39.105[1]	(Reis et al., 2013)
Capsicum chinense Jacqui	Drying	65	NA	NA	NA	NA	Methanol and acetone	-40.059[1]	(Reis et al., 2013)
Capsicum annuum L.	Boiling	NA	5-15	1:5	200 g (sample), 1000 L (water)	2x2 cm	Ethanol	(-42.0)~(-60.5)[1]	(Hwang et al., 2012)
Capsicum annuum L.	Boiling	NA	5-15	1:5	200 g (sample), 1000 L (water)	2x2 cm	Ethanol	(-39.8)~(-55.7)[2]	(Hwang et al., 2012)
Capsicum annuum L.	Steaming	95	5-15	NA	200 g (sample)	2x2 cm	Ethanol	(-23.5)~(-30.0)[1]	(Hwang et al., 2012)
Capsicum annuum L.	Steaming	95	5-15	NA	200 g (sample)	2x2 cm	Ethanol	(-21.7)~(-29.8)[2]	(Hwang et al., 2012)

[1] = Use DPPH method to determine the antioxidant capacity
[2] = Use ABTS method to determine the antioxidant capacity
NA= Not Available

Table 1. (Continued)

Type	Method	Temperature (°C)	Time (min)	Chilli:water (w/v)	Weight	Size	Extraction	Changes of antioxidant capacity (%)	References
Capsicum annuum L.	Stir-frying	NA	5-15	NA	200 g (sample) 2 g (oil)	2x2 cm	Ethanol	(-13.2)~(-17.9)[2]	(Hwang et al., 2012)
Capsicum annuum L.	Stir-frying	NA	5-15	NA	200 g (sample) 2 g (oil)	2x2 cm	Ethanol	(-11.6)~(-15.4)[1]	(Hwang et al., 2012)
Capsicum annuum L.	Roasting	190	5-15	NA	200 g (sample)	2x2 cm	Ethanol	(-4.6)~(-15.8)[1]	(Hwang et al., 2012)
Capsicum annuum L.	Roasting	190	5-15	NA	200 g (sample)	2x2 cm	Ethanol	(-4.9)~(-16.1)[2]	(Hwang et al., 2012)

[1] = Use DPPH method to determine the antioxidant capacity
[2] = Use ABTS method to determine the antioxidant capacity
NA= Not Available

3.1.Flavonoid

Flavonoid can act as an antioxidant agent that can eradicate free radicals and also have advantages for human health. The mechanisms of action of flavonoids are through scavenging or chelating process (Pourmorad et al., 2006). Flavonoid can inhibit lipid peroxidation, chelate redox-active metals, and also impair any processes that concerning reactive oxygen species. Flavonoids are derivates of benzo-ɣ-pyrone that consist phenolic and pyrane rings (Heim et al., 2002). Flavonoid consists about 15 carbons which been arranged in three rings. Flavonols, quercetin and kaempferol are the major flavonoids that can be found in fruits or vegetables.

Flavonoids can inhibit enzymes which cause the production of superoxide anion for examples are xanthine oxidase and protein kinase C. Some flavonoids can chelate trace metals such as free iron and copper that play an important role with reactive oxygen species formation. Catechol is part of flavonoid that mainly contributes to metal chelation. Flavonoid can reduce the oxidizing free radicals due to the lower redox potentials by hydrogen donation. The free radicals that can reduce by flavonoid are peroxyl, alkoxyl, superoxide, and hydroxyl radical. Flavonoids that had been donated their hydrogen atom are called aroxyl radical. Aroxyl radical can react with a second free radical and generate a stable quinone structure. However if the aroxyl radical react with a second oxygen, it will produce quinones and superoxide anion instead of discontinuing the chain reaction (Pietta, 2000).

Lee et al. (1995) said that flavonols, kaempferol and quercetin are the main flavonoids contained in vegetables. Levels of flavonoid are varying with the ripening of chilli peppers. Hot chilli peppers have the higher flavonoid concentration rather than mild or semi-hot chilli peppers. According to Miean & Mohamed (2001), myricetin and quercetin are the major flavonoids contained in *capsicum*, while quercetin is not detected in green coloured *capsicum*. According to Sun et al. (2007), red chilli pepper contains highest quercetin content than green and yellow chilli peppers, while it was none in the orange chilli peppers.

Loizzo et al. (2015) said that lower flavonoid was appearing after boiling treatment for about 10 minutes. Fresh chilli peppers contain the highest flavonoid contents, followed with frozen chilli peppers (-20^{o}C for 4 month), and boiled chilli peppers contain the lowest flavonoid contents except for Arlecchino, Idealino, Effix (*Capsicum annuum*), and Campana (*Capsicum baccatum*). According to Shin et al. (2007), flavonoid will decrease after heating at a certain temperature due to the destruction of some flavonoid. Flavonoids contain C- glycoside bonds and present as oligomers and dimmers. Heating and boiling will result in the establishment of monomers by the hydrolysis of C-glycosides bonds. Higher temperature will cause flavonoids degradation due to the structure of particular flavonoids.

Shaimaa et al. (2016) said that boiling and ripening process of chilli peppers from green to red colour can increase total flavonoid contents in both chilli peppers from *Capsicum frutescens* var. sina and *Capsicum annuum* var. goduion. Total flavonoid contents in fresh and boiled chilli peppers ranged from 10.28 until 15.52 QE/g DW and from 11.63 until 17.62 QE/g DW, respectively. Chilli peppers were boiled at 100^{o}C for 15 minutes. Increasing of flavonoid in boiled chilli peppers might be caused by the increase of extractability of flavonoid due to damage of food matrix during boiling. The changes of flavonoid after boiling according to Shaimaa et al. (2016) are shown in table 2.

Table 2. Effect of Boiling on Flavonoid Content (Shaimaa et al., 2016)

Type	colour	Extraction	Changes of flavonoid (%)
Capsicum frutescens var. sina	Red	Ethanol 80%	13.331
Capsicum frutescens var. sina	Green	Ethanol 80%	5.874
Capsicum annuum var. goduion	Red	Ethanol 80%	1.224
Capsicum annuum var. goduion	Green	Ethanol 80%	13.132
Capsicum frutescens var. sina	Red	Aqueous	205.970
Capsicum frutescens var. sina	Green	Aqueous	138.938
Capsicum annuum var. goduion	Red	Aqueous	231.126
Capsicum annuum var. goduion	Green	Aqueous	142.568

Red chilli peppers from var sina have higher changes of flavonoid contents than green chilli pepper from var. sina in both extraction method (aqueous and ethanol 80%). Changes of flavonoid of green chilli peppers from var goduion extracted with ethanol are higher than the red one, while in aqueous extract the green one is lower. Flavonoid changes in aqueous extract also higher rather than ethanol extract, but flavonoid content with aqueous extract is lower than ethanol extract. According to Li-E et al. (2008), ethanol is a solvent that has highest extract efficiency than aqueous extract. Yields of the extract are affected by the concentration of the solvent, and they said that the best concentration of ethanol is 85%.

3.2.Phenolic

Phenolic compounds can act as antioxidant agents because of their ability to donate hydrogen. Food or beverage that contain rich of phenolic compounds have some advantages to human health such as the risk reduction of cancer, inflammation, arthritis, immune-related diseases, neurodegenerative diseases, heart disease, and heart attack (Gurnani et al., 2015). These many biological functions according to Soobrattee et al. (2005), have been attributed due to antioxidant activity and free radical scavenging. Denre (2014) and Li-E et al. (2008) reported that the higher total phenol compound resulted in higher antioxidant activity.

Phenolic compounds or polyphenols are very abundant compounds that distributed as a group of plant secondary metabolites. There are almost 8000 phenolic structures that have been figured. Natural polyphenols can be distinguished from simple molecules to highly polymerized compounds. Phenolic acids, flavonoids and phenylpropanoids can be categorized as simple molecules, while lignins, tannins, and melanins are categorized as highly polymerized molecules. Antioxidants from phenolic compounds are very good hydrogen or electron donors; they also stable due to resonance delocalization and have a limited part of being attacked by oxygen. Phenolic antioxidants inhibit the lipid oxidation by rapid donation of the hydrogen atom to lipid radicals. The effects of antioxidant concentration on autoxidation rates are due to antioxidant structure, oxidation conditions, and the nature of the oxidized sample. High concentration of prooxidants can decrease the antioxidant activity (Shahidi et al., 2009).

$$(1)\ R\bullet + ArOH \rightarrow RH + ArO\bullet$$

$$(2)\ R\bullet + ArOH \rightarrow R^{-} + ArO\bullet^{+}$$

Figure 7. Antioxidant Mechanism of Phenolic

The first mechanism of antioxidant is hydrogen atom from the antioxidant is removed by free radicals and the antioxidant itself later becomes a radical (Reaction 1; Figure 6). The more efficient the antioxidant ArOH, the stability of the radical ArO• is higher, so it is not possible to react with the substrate. The second mechanism is the antioxidant gives an electron to the free radical become a radical cation (Reaction 2; Figure 6). The radical cation which arises from the electron transfer is quite stable and doesn't react with the substrate (Leopoldini et al., 2004).

Temperature, light, hydrolysis and oxidation reaction can affect the phenol content. Enzymatic oxidation of phenolic contents is usually caused by polyphenol oxidases. Injured by the cell membrane will liberate this enzyme and then activated it. The liberation of polyphenol oxidases will oxidize the phenolic content into quinones (Reis et al., 2013).

Shaimaa et al. (2016) explicate that total phenolic compound contained in fresh and heat treated chilli peppers are high but boiled chilli peppers contain more phenolic content than fresh chilli peppers. Increases of phenolic content of vegetables by cooking have been attributed to alteration of food matrix and an improved extractability of phenolics from the food. Boiling can inactivate the polyphenol oxidase enzyme during heating and lead to the inhibition of polyphenolics degradation. Turkmen et al. (2005) investigated that boiling and some cooking method such as microwaving and grilling can increase phenolic content. Ornelas-Paz et al. (2010) also found that phenolic content in chilli peppers can increase by boiling and grilling. Increases of phenolic compounds in chilli peppers by cooking are ranged between 7.4-137% in habanero and serrano pepper (Table. 3). Cooking can inactivate the polyphenol oxidase enzyme and leads to inhibit degradation of polyphenolic (Chuah et al., 2008). In addition, Roy et al. (2014) reported that polyphenolic content on chilli pepper of *Capsicum annuum* has significantly increased during cooking with pressure cooking and microwave heating. Microwaving chilli pepper resulted in highest polyphenolic content than fresh and pressure cooking, especially for red chilli pepper.

On the other hand, Loizzo et al. (2015) reported that boiling will cause lower phenol content in chilli peppers. Same as a flavonoid, fresh chilli peppers and frozen chilli peppers contain a higher amount of phenol than boiled chilli peppers except for Arlecchino, Idealino, Effix (*Capsicum annuum*), and Campana (*Capsicum baccatum*). Ornelas-Paz et al. (2010) also reported that boiling and grilling can decrease total phenolic compounds of Bell pepper approximately 1.6-26.9%. According to Hwang et al. (2012), boiling, steaming, stir-frying, and roasting will decrease total polyphenol, with a range 66.99-95.47, 119.10-127.99, 141.54-145.93, and 141.32-142.61 mg/100g, respectively. Boiling and steaming significantly decrease total polyphenol in red chilli pepper than stir-frying and roasting. The highest loss of total polyphenol is observed in boiling method which is about 35.8-54.9%, while stir-frying results minimum loss of total polyphenol which is about 18-4.8%. The decline of total polyphenol in boiled or steamed chilli peppers caused to the dispersion of phenolic compound into cooking water. Processing time and the size of food also contribute to phenolic compound decrease. The Higher temperature contributes to the reduction of some phenolic compounds in chilli peppers (Li-E et al., 2008).

Chuah et al. (2008) investigated that cooking in water seems to be causing a leakage of antioxidant like phenols into the cooking water. Polyphenol was significantly reduced by 5 minutes and 30 minutes of boiling. Boiling for about 30 minutes will result in greater polyphenol losses than 5 minutes of boiling. Chuah et al. (2008) said that cooking method such as stir-frying and microwave heating didn’t significantly reduce polyphenol compound of chilli peppers. This can happen because of the inactivation of polyphenol oxidase during heating that will cause the inhibition of polyphenol oxidation (Amaguchi et al., 2001). A prolonged time of microwave method might cause degradation of phenol and antioxidant compound. Microwaving might generate the higher total phenolic contents than steaming and boiling method. Boiling and steaming method released them into the water, even if with directly or indirectly hot water during processed.

According to Reis et al. (2013), dried chilli peppers contain lower total phenolic content than fresh chilli peppers. There was no statistical difference between different temperature and phenolic compound in dried chilli peppers, therefore the use of highest temperature for drying chilli peppers are preferred because the time of drying and cost of processing can be reduced. Based on Vega-gálvez et al. (2009) investigation, the higher temperature of air-drying can cause loss of phenolic content. Varietal differences between chilli peppers can influence total phenolic contents sensitivity to the processing condition. Fresh chilli pepper contains higher phenolic compounds than dried chilli peppers but drying chilli peppers at high temperature (90^{o}C) generate higher phenolic content than drying at the lower temperatures. At high temperature, the availability of phenolic molecule's precursors by enzymatic mutual conversion between phenolic molecules will result in the establishment of phenolic contents.

Freeze drying is a popular method of drying food material. Freeze drying process can increase tissue porosity that can result in higher phenolic content in some varieties because of greater extraction efficiency (Materska, 2014). Based on his work, total phenolic contents of freeze-dried chilli pepper var Shanghai were increased about 55% compared with fresh chilli pepper. Besides, total phenolic contents of chilli peppers especially var Capel Hot were decreased approximately 40%. To minimize from the harmful environmental effect, freeze-dried chilli peppers must be stored in the dark and tight containers at a lower temperature (<0^{o}C). Use of high temperatures on processing or during storage of food according to Reis et al. (2013) will lead the phenolic compounds to be reduced due to anthocyanins reduction. In table 3, the changes of phenolic during some processing method of chilli pepper are shown.

Iqbal et al. (2015) reported that total phenolic contents decreased with the increase in storage temperature. Total phenolic contents of chilli peppers have decreased when storage for 5 months. The amount of total phenolic contents reduction for storage in polyethylene with temperature at 20^{o}C, 25^{o}C, and 30^{o}C are 14.1%,16.9%, and 20.5%, respectively, while reduction of total phenolic content for storage in jute bags with temperature at 20^{o}C, 25^{o}C, and 30^{o}C are 15.3%,19.0%, and 22.8%, respectively. Reductions of total phenolic contents are higher with increasing the temperatures. Between polyethylene and jute bag, the stability of total phenolic contents exhibits the same trend. Polyethylene and jute bag preserved similar extent on total phenolic contents of chilli peppers.

Table 3. Effect of Processing Methods on Phenolic Content

Type	Colour	Method	Temperature (°C)	Time (min)	Chilli:water (w/v)	Size	Extraction	Changes of phenolic (%)	References
Capsicum annuum	Green	Pressure cooking	NA	15	1:10	NA	Aqueous	-7.895	(Roy et al., 2014)
Capsicum annuum	Red	Pressure cooking	NA	15	1:10	NA	Aqueous	14.535	(Roy et al., 2014)
Capsicum annuum	Yellow	Pressure cooking	NA	15	1:10	NA	Aqueous	181.818	(Roy et al., 2014)
Capsicum annuum	Green	Microwave heating	160	30	1:10	NA	Aqueous	17.105	(Roy et al., 2014)
Capsicum annuum	Red	Microwave heating	160	30	1:10	NA	Aqueous	22.674	(Roy et al., 2014)
Capsicum annuum	Yellow	Microwave heating	160	30	1:10	NA	Aqueous	416.746	(Roy et al., 2014)
Capsicum frutescens var. sina	Red	Boiling	100	15	1:1	NA	Ethanol 80%	32.395	(Shaimaa et al., 2016)
Capsicum frutescens var. sina	Green	Boiling	100	15	1:1	NA	Ethanol 80%	49.297	(Shaimaa et al., 2016)
Capsicum annuum var. goduion	Red	Boiling	100	15	1:1	NA	Ethanol 80%	0.531	(Shaimaa et al., 2016)

NA= Not Available

Table 3. (Continued)

Type	Colour	Method	Temperature (^{o}C)	Time (min)	Chilli:water (w/v)	Size	Extraction	Changes of phenolic (%)	References
Capsicum annuum var.*goduion*	Green	Boiling	100	15	1:1	NA	Ethanol 80%	6.287	(Shaimaa et al., 2016)
Capsicum frutescens var. sina	Red	Boiling	100	15	1:1	NA	Aqueous	15.473	(Shaimaa et al., 2016)
Capsicum frutescens var. sina	Green	Boiling	100	15	1:1	NA	Aqueous	0.992	(Shaimaa et al., 2016)
Capsicum annuum var. goduion	Red	Boiling	100	15	1:1	NA	Aqueous	51.814	(Shaimaa et al., 2016)
Capsicum annuum var. goduion	Green	Boiling	100	15	1:1	NA	Aqueous	18.120	(Shaimaa et al., 2016)
Capsicum chinense J. var.Habanero	Green	Boiling	96	7.5 ± 0.6	NA	(4.0±0.1) x (3.4± 0.1)	Methanol 80%	110.079	(Ornelas-Paz et al., 2010)

NA= Not Available

Table 3. (Continued)

Type	Colour	Method	Temperature (°C)	Time (min)	Chilli:water (w/v)	Size	Extraction	Changes of phenolic (%)	References
Capsicum annuum L. var bell pepper	Yellow	Boiling	96	13.0 ± 0.8	NA	(8.9±0.1) x (9.1±0.1)	Methanol 80%	-9.842	(Ornelas-Paz et al., 2010)
Capsicum chinense J. var.Habanero	Yellow	Boiling	96	7.0 ± 0.3	NA	(4.4±0.1) x (3.3± 0.1)	Methanol 80%	100.591	(Ornelas-Paz et al., 2010)
Capsicum annuum L. var bell pepper	Red	Boiling	96	11.8 ± 0.5	NA	(9.0±0.1) x (9.7 ±0.1)	Methanol 80%	-8.063	(Ornelas-Paz et al., 2010)
Capsicum annuum L. var bell pepper	Green	Boiling	96	11.5 ± 0.7	NA	(8.9±0.1) x (10.1±0.1)	Methanol 80%	-1.631	(Ornelas-Paz et al., 2010)
Capsicum annuum L. cv. Serrano	Red	Boiling	96	8.5 ± 0.9	NA	(7.0±0.1) x (1.6±0.0)	Methanol 80%	14.354	(Ornelas-Paz et al., 2010)
Capsicum annuum L. cv. Serrano	Green	Boiling	96	11.8 ± 2.2	NA	(8.6± 0.1) x (1.8 ± 0.0)	Methanol 80%	7.422	(Ornelas-Paz et al., 2010)
Capsicum chinense J. var.Habanero	Yellow	Grilling	210	10.0 ± 0.3	NA	(4.4±0.1) x (3.3± 0.1	Methanol 80%	137.288	(Ornelas-Paz et al., 2010)

NA= Not Available

Table 3. (Continued)

Type	Colour	Method	Temperature (°C)	Time (min)	Chilli:water (w/v)	Size	Extraction	Changes of phenolic (%)	References
Capsicum chinense J. var.Habanero	Green	Grilling	210	8.8 ± 0.3	NA	(4.0±0.1) x (3.4± 0.1)	Methanol 80%	133.839	(Ornelas-Paz et al., 2010)
Capsicum annuum L. var bell pepper	Red	Grilling	210	17.0 ± 0.7	NA	(9.0±0.1) x (9.7 ±0.1)	Methanol 80%	-20.918	(Ornelas-Paz et al., 2010)
Capsicum annuum L. var bell pepper	Green	Grilling	210	16.8 ± 0.8	NA	(8.9±0.1) x (10.1±0.1)	Methanol 80%	-7.321	(Ornelas-Paz et al., 2010)
Capsicum annuum L. var bell pepper	Yellow	Grilling	210	19.0 ± 1.4	NA	(8.9±0.1) x (9.1±0.1)	Methanol 80%	-26.866	(Ornelas-Paz et al., 2010)
Capsicum annuum L. cv. Serrano	Red	Grilling	210	9.3 ± 0.3	NA	(7.0±0.1) x (1.6±0.0)	Methanol 80%	21.935	(Ornelas-Paz et al., 2010)

NA= Not Available

Table 3. (Continued)

Type	Colour	Method	Temperature (°C)	Time (min)	Chilli:water (w/v)	Size	Extraction	Changes of phenolic (%)	References
Capsicum annuum L. cv. Serrano	Green	Grilling	210	9.8 ± 0.5	NA	(8.6± 0.1) x (1.8 ± 0.0)	Methanol 80%	39.378	(Ornelas-Paz et al., 2010)
Capsicum chinense Jacqui	NA	Drying	45	NA	NA	NA	Methanol 50% and acetone 70%	-84.809	(Reis et al., 2013)
Capsicum chinense Jacqui	NA	Drying	55	NA	NA	NA	Methanol 50% and acetone 70%	-85.118	(Reis et al., 2013)
Capsicum chinense Jacqui	NA	Drying	65	NA	NA	NA	Methanol 50% and acetone 70%	-85.480	(Reis et al., 2013)

NA= Not Available

3.3.Capsaicinoid

Capsaicinoids according to Lee & Howard (1999) are the most stable antioxidant. Capsaicinoids can help to reduce plasma total cholesterol, increase lipoprotein profile and also reduce aortic plaque in the high cholesterol-fed situation (Srinivasan, 2016). Other than that, capsaicinoids also potential benefits for pain relief, prevent cancer and reduce body weight (Luo, Peng, & Li, 2011). According to Giuffrida et al. (2014) and Topuz et al. (2011), capsaicin and dehydrocapsaicin are two predominant molecules of capsaicinoid. Capsaicin represents the 58.5% of the total capsaicinoids, while dehydrocapsaicin represents the 34% of the total capsaicinoids. Reis et al. (2013) also said that capsaicin is the highest alkaloid found in chilli peppers. The amount of capsaicin contained in chilli peppers are in the range 33 to 77%, followed by dehydrocapsaicin with range 22 to 51%. For about 90% of capsaicinoid compounds are capsaicin and dehydrocapsaicin. A Higher level of capsaicin in chilli peppers indicates the higher antioxidant activity.

(1) $CAP + R\bullet \rightarrow CAP\bullet^{+} + R^{-}$

(2) $CAP + R\bullet \rightarrow CAP\bullet(\text{-}H) + HR^{-}$

(3) $CAP + R\bullet \rightarrow (CAP\text{-}R)\bullet$

Figure 8. Antioxidant Mechanism of Capsaicinoid

According to Galano et al. (2012), there is three mechanisms of capsaicin to react with free radicals, those are hydrogen transfer (Reaction 1; Figure 7), radical adduct formation (Reaction 2; Figure 7), and single electron transfer (Reaction 3; Figure 7). Hydrogen transfer is the major mechanism of capsaicin in the free radical scavenging activity. According to Materska & Perucka (2005), capsaicin that extracted from *Capsicum annuum* L. has greater antioxidant activity than dihydrocapsaicin. They assumed that the double bond in the lipid chain of capsaicin influences antioxidant activity.

Freezing and boiling process can decrease the content of capsaicin and dihydrocapsaicin in chilli peppers except for some cultivars such as yellow Cayenne, Effix, Duemila, Hierro, Acrata, Pelegrino (*Capsicum annuum*), and Capezzolo di Scimmia (*Capsicum chinense*) (Loizzo et al., 2015). Ornelas-Paz et al. (2010) investigated that boiling can reduce capsaicinoid of chilli peppers. Boiling will reduce capsaicinoid content about 1.1-28.1% from the initial concentration. Capsaicinoid loses are varied depending on the type of chilli peppers. Loss of capsaicin during boiling might be because of capsaicin leached into boiling water. In otherwise, grilling increase capsaicinoid of chilli peppers. Increases of capsaicinoid contents on thermal treatment such as grilling can be caused by alteration of the food matrix, inactivation of peroxidase enzymes, the release of conjugated capsaicinoids, and improved extractability of these compounds with cell interference during the thermal process. Alvarez-Parrilla et al. (2011) found that fresh chilli peppers have higher capsaicinoid content than processed chilli peppers.

Schweiggert et al. (2006) investigated that thermal treatment with blanching at 80^{o}C and 90^{o}C for 5 and 10 minutes will decrease the capsaicinoid concentration about 21.7-28.3%. According to Suresh et al., (2007), capsaicin will loss from red chilli pepper in the range 18-36% during heat treatment and pressure cooking caused maximum loss than boiling. Srinivasan, Sambaiah, & Chandrasekhara (1992) also reported that capsaicin decrease in the range 19-33% during boiling depending on the duration of cooking. The duration of boiling makes a difference to the extent of capsaicin loss. Drying with temperature 45^{o}-65^{o}C can also make capsaicin in chilli peppers diminish approximately 28,8-33,5% (Reis et al., 2013). Baking can also reduce capsaicin in chilli peppers. According to Srisajjalertwaja et al. (2012), baking at temperatures 180^{o}C, 210^{o}C, and 250^{o}C for about 5, 10, 20, and 30 minutes decreased capsaicin for about 3.558-67.031%. The highest capsaicin loss was after the chilli pepper being baked at 250^{o}C for about 30 minutes, while the lowest capsaicin loss was after being baked at 180^{o}C for about 5 minutes. The capsaicin content will reduce along with increasing temperatures and prolonged times.

Toontom et al. (2016) found that drying method such as sun drying, freeze drying, and hot air drying didn’t affect the capsaicin content, however, drying method can increase the capsaicin content on chilli peppers (*Capsicum annuum* L. var. Acuminatum Fingerh). Freeze drying method has the highest capsaicin content than sun drying or hot air drying method, while sun drying has the lowest capsaicin content in chilli peppers. That was possible because sun drying method is directly exposed to air and takes a long time to be dried. The authors indicated that increases of capsaicinoid during drying due to the activity of peroxidase enzyme and temperature during drying and blanching. Blanching before drying process might inactivate the peroxidase enzyme, while these enzymes were still active in the fresh chilli peppers. Another explanation was hydrolysis of glycosides in dried chilli pepper from blanching and drying heat. Huffman et al. (1978) also reported that during thermal processing, capsaicin is volatilized and many cells are lysed. This process effects capsaicin to spread and free throughout the fruit. Many complexing agents also might spit off, leave the free capsaicin. Changes of capsaicin and capsaicinoid on chilli pepper because of processing are shown in table 4.

Capsaicin and dihydrocapsaicin content will slightly reduce during storage. Capsaicinoids will decrease when being stored at room temperature, but stable at low temperature (Giuffrida et al., 2014). Storage at ambient temperature at 6 months will result reduce of capsaicinoid contents for about 6.8-11.9%. Capsaicinoid losses in chilli pepper usually due to non-enzymatic oxidation during thermal treatment, not dependent on exposure to light (Schweiggert et al., 2006).

Table 4. Effect of Processing Methods on Capsaicinoid and Capsaicin

Type	Colour	Method	Temperature (°C)	Time (min)	Chilli:Water (w/v)	Size	Extraction	Changes of capsaicin (%)	Changes of capsaicinoid (%)	References
Capsicum chinense Jacqui	NA	Drying	45	NA	NA	2 g	Methanol and acetone	NA	-28.824	(Reis et al., 2013)
Capsicum chinense Jacqui	NA	Drying	55	NA	NA	2 g	Methanol and acetone	NA	-19.706	(Reis et al., 2013)
Capsicum chinense Jacqui	NA	Drying	65	NA	NA	2 g	Methanol and acetone	NA	-33.529	(Reis et al., 2013)
Capsicum annuum L. var. acuminatum Fingerh	Red	Freeze drying[4]	-50°C	NA	NA	(1.5±0.24) x (10.4±0.98)	Acetone	122.414	NA	(Toontom et al., 2016)
Capsicum annuum L. var. acuminatum Fingerh	Red	Air drying[4]	60°C	NA	NA	(1.5±0.24) x (10.4±0.98)	Acetone	101.724	NA	(Toontom et al., 2016)

[3] = Pre-treated with drying
[4] = Pre-treated with water blanching
NA= Not Available

Table 4. (Continued)

Type	Colour	Method	Temperature (°C)	Time (min)	Chilli:Water (w/v)	Size	Extraction	Changes of capsaicin (%)	Changes of capsaicinoid (%)	References
Capsicum annuum L. var. acuminatum Fingerh	Red	Sun Drying[4]	Apx. 37°C	8 x 8 H	NA	(1.5±0.24) x (10.4±0.98)	Acetone	68.966	NA	(Toontom et al., 2016)
Capsicum annuum	Red	Pressure cooking[3]	NA	10	1:20	5 g	Ethyl acetate	-36.187	NA	(Suresh et al., 2007)
Capsicum annuum	Red	Boiling[3]	NA	10	1:20	5 g	Ethyl acetate	-18.315	NA	(Suresh et al., 2007)
Capsicum annuum	Red	Boiling[3]	NA	20	1:20	5 g	Ethyl acetate	-21.896	NA	(Suresh et al., 2007)
Capsicum chinense J. var habanero	Yellow	Boiling	96	7.0 ± 0.3	NA	NA	Methanol	-1.162	NA	(Ornelas-Paz et al., 2010)
Capsicum annuum L. cv serrano	Red	Boiling	96	8.5 ± 0.9	NA	NA	Methanol	-9.469	NA	(Ornelas-Paz et al., 2010)
Capsicum chinense J. var. habanero	Green	Boiling	96	7.5 ± 0.6	NA	NA	Methanol	-4.284	NA	(Ornelas-Paz et al., 2010)

[3] = Pre-treated with drying
[4] = Pre-treated with water blanching
NA= Not Available

Table 4. (Continued)

Type	Colour	Method	Temperature (°C)	Time (min)	Chilli:Water (w/v)	Size	Extraction	Changes of capsaicin (%)	Changes of capsaicinoid (%)	References
Capsicum annuum L. cv serrano	Green	Boiling	96	11.8 ± 2.2	NA	NA	Methanol	19.975	NA	(Ornelas-Paz et al., 2010)
Capsicum chinense J. var habanero	Green	Grilling	210	8.8 ± 0.3	NA	NA	Methanol	51.940	NA	(Ornelas-Paz et al., 2010)
Capsicum chinense J. var habanero	Yellow	Grilling	210	10.0 ± 0.3	NA	NA	Methanol	48.884	NA	(Ornelas-Paz et al., 2010)
Capsicum annuum L. cv serrano	Red	Grilling	210	9.3 ± 0.3	NA	NA	Methanol	6.139	NA	(Ornelas-Paz et al., 2010)
Capsicum annuum L. cv. serrano	Green	Grilling	210	9.8 ± 0.5	NA	NA	Methanol	33.086	NA	(Ornelas-Paz et al., 2010)

[3] = Pre-treated with drying

[4] = Pre-treated with water blanching

NA= Not Available

Table 4. (Continued)

Type	Colour	Method	Temperature (°C)	Time (min)	Chilli:Water (w/v)	Size	Extraction	Changes of capsaicin (%)	Changes of capsaicinoid (%)	References
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	180	5	NA	10	Methanol	-3.558	NA	(Srisajjalertwaja et al., 2012)
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	180	10	NA	10	Methanol	-8.031	NA	(Srisajjalertwaja et al., 2012)
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	180	20	NA	10	Methanol	-30.680	NA	(Srisajjalertwaja et al., 2012)
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	180	30	NA	10	Methanol	-41.951	NA	(Srisajjalertwaja et al., 2012)
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	210	5	NA	10	Methanol	-5.107	NA	(Srisajjalertwaja et al., 2012)

[3] = Pre-treated with drying
[4] = Pre-treated with water blanching
NA= Not Available

Table 4. (Continued)

Type	Colour	Method	Temperature (°C)	Time (min)	Chilli:Water (w/v)	Size	Extraction	Changes of capsaicin (%)	Changes of capsaicinoid (%)	References
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	210	10	NA	10	Methanol	-11.800	NA	(Srisajjalertwaja et al., 2012)
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	210	20	NA	10	Methanol	-41.247	NA	(Srisajjalertwaja et al., 2012)
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	210	30	NA	10	Methanol	-52.061	NA	(Srisajjalertwaja et al., 2012)
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	250	5	NA	10	Methanol	-16.907	NA	(Srisajjalertwaja et al., 2012)
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	250	10	NA	10	Methanol	-31.455	NA	(Srisajjalertwaja et al., 2012)

[3] = Pre-treated with drying
[4] = Pre-treated with water blanching
NA= Not Available

Table 4. (Continued)

Type	Colour	Method	Temperature (°C)	Time (min)	Chilli:Water (w/v)	Size	Extraction	Changes of capsaicin (%)	Changes of capsaicinoid (%)	References
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	250	20	NA	10	Methanol	-47.270	NA	(Srisajjalertwaja et al., 2012)
Capsicum annuum Linn. var. *Jak Ka Pat*	Green	Baking	250	30	NA	10	Methanol	-67.031	NA	(Srisajjalertwaja et al., 2012)

[3] = Pre-treated with drying
[4] = Pre-treated with water blanching
NA= Not Available

3.4.Carotenoid

Carotenoid is abundant and usually can be found in vegetables or fruits. Carotenoid is one of the natural pigments contained in fruit and vegetables that have a contribution in yellow to red colour. Carotenoid also acts as antioxidants. Carotenoid can act as photoprotective agents against the hazardous effect of O_2, light, and photosensitizing pigments. Moreover, it can role as reactive compounds against chemical compounds generated in cells which can cause oxidative damage. Carotenoids are capable to restrict the oxidative damage caused by oxy radical-generating systems. The protection of carotenoid involves both nuclear and lipid molecules (Palozza & Krinsky, 1992).

According to El-agamey et al. (2004), antioxidant characteristics of carotenoid are due to their radical scavenging properties and their singlet oxygen quenching abilities. Some parameters that can affect the type and the rate of carotenoid mechanism with different radicals are the structure of carotenoid, radical reactivity, and medium polarity. There are three possibilities of initial step from carotenoids scavenge radical such as electron transfer (Reaction 1; Figure 8), hydrogen abstraction (Reaction 2; Figure 8), and addition (Reaction 3; Figure 8).

(1) CAR + ROO• → $CAR•^{+}$ +ROO^{-}

(2) CAR + ROO• → CAR• +ROOH

(3) CAR + ROO• → ROOCAR•

Figure 9. Antioxidant Mechanism of Carotenoid

The main carotenoids that usually occur in red chilli peppers are free capsanthin, β-carotene, zeaxanthin, and various carotenoids monoesters or di-esters of capsanthin and zeaxanthin with lauric, myristic and palmitic acids (Giuffrida et al., 2013). Di-ester carotenoid was more stable compared with monoesters and unesterified carotenoid in chilli peppers (Daood & Albrecht, 2006). Capsanthin is one of the predominant carotenoids contains in red chilli peppers with a range from 230 until 848 μg/100g in chilli peppers. Capsanthin, β-carotene and β-cryptoxanthin were found to be very abundant in all red genotypes of *Capsicum annuum* and *Capsicum baccatum* (Pugliese et al., 2013). According to Sun et al. (2007), red and green chilli peppers contain the highest level of β-carotene compared with orange chilli peppers, while yellow chilli peppers have the lowest level of β-carotene.

Daood & Albrecht (2006) and Ornelas-paz et al. (2013) investigated that stability of carotenoid in the pungent chilli peppers was higher than non-pungent chilli peppers at high drying temperature. Capsaicinoid contributes the pungent flavour of chilli peppers, capsaicinoid can affect the chemical interactions of carotenoids in the tissue of chilli peppers. Therefore it can provide substantial protection against destruction caused by heat. They also conduct that high loss of carotenoid content contained on dried chilli peppers at 90 ^{o}C and 100 ^{o}C.

According to Daood & Albrecht (2006), lutein, cryptocapsin, β-cryptoxanthin mono-esters and all of the di-esters found to be more stable during thermal processing than other carotenoid content on pungent or non-pungent chilli peppers. The factors that might be affecting the degradation of carotenoids during thermal drying are thermal destruction by heat and also accelerated lipid oxidation in presence of oxygen. The amount of carotenoid degradation was increased with the increased of drying temperature. On the other hand, Pugliese et al. (2013) found that β-carotene and β-cryptoxanthin were a component of carotenoid that most affected due to processing method that is boiling and freezing. Boiling and freezing result in significant decrease in these two carotenoids of chilli peppers. Red chilli peppers are richer in β-carotene than yellow or green chilli peppers. Boiling and grilling can reduce β-carotene in chilli pepper. Although β-carotene in boiled chilli pepper has lower reduction than a grilled chilli pepper, in general, the result of β-carotene content in boiled and grilled chilli peppers are similar (Ornelas-paz et al., 2013).

Chuah et al. (2008) investigated that cooking method such as microwave heating for 5 minutes, stir-frying for 5 minutes and boiling for 5 and 30 minutes can slightly reduce total carotenoid in chilli peppers (*Capsicum annuum* L.). A significant decrease of carotenoid was found in 5 minutes of boiling for the green pepper and orange paprika. However, after being boiled for about 5 minutes, total carotenoid content constantly stable for up to 30 minutes. Heat treatment did not severely affect the carotenoid content. Besides the method of cooking, amount of carotenoid that can find in chilli peppers depends on heat treatment and varietal differences. The changes of carotenoid according to Chuah et al. (2008) are shown in table 5.

Hwang et al. (2012) also investigated that method cooking such as boiling, steaming, and roasting significantly decrease carotenoid content on chilli peppers. Carotenoid content of boiled and roasted red chilli peppers will decrease as cooking time increases, with the range of 1217.25-1317.86 and 1232.58-1372.06 μg/100g, respectively. Carotenoid content on chilli pepper is decrease during steaming, with a range of 1239.58-1264.86 μg/100g, but there is not significantly decrease as cooking temperature increases. Stir-frying method not significantly affect the carotenoid content in red chilli peppers. Carotenoid content after stir-frying for 5, 10, and 15 minutes are ranged from 1351.75-1441.21 μg/100g, though carotenoid quiet increase after 5 minutes. When compared with boiling, steaming, and roasting, stir-frying is the best method of cooking to retain carotenoid content because it has minimal effect on carotenoid.

Pugliese et al. (2013) said that cooking and processing of may influence the carotenoid content and bioavailability. Cooking and processing can increase carotenoid released from food matrix, however, cooking and processing can also reduce it. The increase of carotenoid compounds might be caused due to the heat treatments that raise the bioavailability and liberation of carotenoids. Carotenoids in fresh vegetables are bound by the protein. Heat treatments will help to destruct binding-proteins and release the carotenoids, so it can be more easily extracted (Howard et al., 1995). The increase of carotenoid compound might be caused due to the heat treatments that raise the bioavailability and liberation of carotenoids.

According to Iqbal et al. (2015), carotenoids are sensitive to the temperature, light, and moisture. Higher temperature storage will increase carotenoid reduction. Losses of carotenoids can also be caused by peeling and development of prooxidative processes during storage. Giuffrida et al. (2014) said that carotenoid on chilli powder will be more stable in freezing condition (-18^{o}C) rather than room temperature (20-24^{o}C). Carotenoid content will decrease up to 20.11% after storage for 12 months. Moisture content and storage temperature are the most important parameters that can affect the stability of red chilli powder during storage. The low temperature will reduce the oxidative degradation processes.

Anjun et al. (2008) found that boiling process for one hour caused β-carotene in chilli peppers lose up to 14.9%. Storage for about one week at room temperature also induces β-carotene losses up to 27.27%. According to Q. Iqbal et al. (2015), during storage for 5 months at temperature 20^{o}C, 25^{o}C, and 30^{o}C, total carotenoid were decreased gradually for about 11.5-20.2% during stored in polyethylene bags, while in jute bags carotenoid will decrease for about 11.7-22.9%. The rate of carotenoid loss increased with storage temperature and tended to be a little higher if chilli peppers were packed with jute bag compared with packed in polyethylene. Carotenoid losses were higher when storage at 30^{o}C than at 20^{o}C and 25^{o}C. Same as phenolic content, polyethylene and jute bag preserved similar extent on total carotenoid contents of chilli peppers.

Table 5. Effect of Processing Methods on Carotenoid (Chuah et al., 2008)

Type	Colour	Method	Time (min)	Weight	Changes of carotenoid (%)
Capsicum annuum L. var. pepper	Red	Microwave heating	5	300-500 g (sample)	-10.112
Capsicum annuum L. var. pepper	Green	Microwave heating	5	300-500 g (sample)	-17.769
Capsicum annuum L var. paprika	Red	Microwave heating	5	300-500 g (sample)	-3.556
Capsicum annuum L var. paprika	Green	Microwave heating	5	300-500 g (sample)	-17.949
Capsicum annuum L var. paprika	Orange	Microwave heating	5	300-500 g (sample)	-3.236
Capsicum annuum L var. paprika	Yellow	Microwave heating	5	300-500 g (sample)	35.912
Capsicum annuum L.var. pepper	Red	Stir-frying	1	300-500 g (sample) 2-3 g (oil)	9.738
Capsicum annuum L. var. pepper	Green	Stir-frying	1	300-500 g (sample) 2-3 g (oil)	0.826
Capsicum annuum L var. paprika	Red	Stir-frying	1	300-500 g (sample) 2-3 g (oil)	13.111

Table 5 (Continued)

Type	Colour	Method	Time (min)	Weight	Changes of carotenoid (%)
Capsicum annuum L var. paprika	Green	Stir-frying	1	300-500 g (sample) 2-3 g (oil)	9.615
Capsicum annuum L var. paprika	Orange	Stir-frying	1	300-500 g (sample) 2-3 g (oil)	-3.698
Capsicum annuum L var. paprika	Yellow	Stir-frying	1	300-500 g (sample) 2-3 g (oil)	35.912
Capsicum annuum L. var. pepper	Red	Boiling	5	300-500 g (sample) 500 ml (water)	-20.412
Capsicum annuum L. var. pepper	Green	Boiling	5	300-500 g (sample) 500 ml (water)	-18.595
Capsicum annuum L var. paprika	Red	Boiling	30	300-500 g (sample) 500 ml (water)	-20.889
Capsicum annuum L var. paprika	Green	Boiling	30	300-500 g (sample) 500 ml (water)	35.897
Capsicum annuum L var. paprika	Red	Boiling	5	300-500 g (sample) 500 ml (water)	16.222
Capsicum annuum L var. paprika	Green	Boiling	5	300-500 g (sample) 500 ml (water)	11.538

Table 5. (Continued)

Type	Colour	Method	Time (min)	Weight	Changes of carotenoid (%)
Capsicum annuum L. var. pepper	Red	Boiling	30	300-500 g (sample) 500 ml (water)	-21.723
Capsicum annuum L. var. pepper	Green	Boiling	30	300-500 g (sample) 500 ml (water)	-26.033
Capsicum annuum L var. paprika	Orange	Boiling	30	300-500 g (sample) 500 ml (water)	40.986
Capsicum annuum L var. paprika	Yellow	Boiling	30	300-500 g (sample) 500 ml (water)	28.729
Capsicum annuum L var. paprika	Orange	Boiling	5	300-500 g (sample) 500 ml (water)	21.880
Capsicum annuum L var. paprika	Yellow	Boiling	5	300-500 g (sample) 500 ml (water)	35.359

3.5.Ascorbic Acid

ascorbic acid plays a role as important nutritional and functional constituent of hot chilli peppers (Iqbal et al., 2015). Ascorbic acid is unstable due to heat, metal, oxygen, and low pH (pH>7). Contact with water during chilli pepper processing is undesirable due to ascorbic acid is water soluble (Lee & Howard, 1999). Howard et al. (1994) found that L-ascorbic acid in *Capsicum annuum* was present in much greater amounts compared with dehydroascorbic acid. Zhang & Hamauzu (2003) investigated that ascorbic acid was a strong radical scavenger. However ascorbic acid was not quite stable because the level of ascorbic acid was reduced by time and temperature of storage.

Ascorbic acid scavenges free radical due to hydrogen donated to the radicals lead to the more stable compound. Loss proton of ascorbic acid will change it into dehydroascorbic acid (Choe & Min, 2015). Antioxidant activities of ascorbic acid according to Frei (1994) are due to its ability to react with some free radicals and oxygen species. Ascorbic acid is also effective to protect low-density lipoprotein (LDL) cholesterol against oxidation. The antioxidant mechanism of ascorbic acid is ascorbic acid oxidized in two steps; the first will become ascorbyl radical, and then become dehydroascorbate. Ascorbate releases two hydrogens of an atom that react with the AAPH-derived peroxyl radical to establish non-radical compound.

Castro et al. (2008) reported that ascorbic acid will progressively decrease during blanching process according to temperature and duration of blanching process. The higher the temperature and the longer the blanching time will result an ascorbic acid loss for about 45% for green pepper and 30% for the red pepper. Pressurized of Bell peppers will also reduce ascorbic acid content for about 15-20% in red Bell pepper, while 10-20% in green Bell pepper. Pressure treatment that applied to red and green Bell peppers results in a lower reduction in ascorbic acid content.

According to Chuah et al. (2008), cooking treatment such as microwave heating, boiling, and stir-frying will result in a reduction of ascorbic acid on chilli peppers (*Capsicum* annuum L.). Loss of ascorbic acid during thermal processing could be caused by accelerating oxidation of ascorbic acid to dehydroascorbic acid, followed by the hydrolysis to 2, 3-diketogluconic acid and at last polymerization to another nutritionally inactive component. Microwave heating didn't cause significant loss of ascorbic acid in chilli pepper. Ascorbic acid will have a higher reduction by the stir-frying method. Ascorbic acid losses during stir-frying is due to high temperatures, long cooking time, enzymatic oxidation during the preparation and cooking processes and frequent stirring that expose the materials to atmospheric oxidation. However, microwave heating and stir-frying method assure a higher retention of ascorbic acid in chilli peppers than boiling. Ascorbic acid was significantly decreased by 5 minutes of boiling and will have a further reduction after 30 minutes of boiling. The total of ascorbic acid of red, orange and yellow paprika in boiling water and cooked tissue by 5 and 30 minutes of boiling were almost the same with ascorbic acid content in fresh tissue. Ascorbic acid will reduced during boiling because the ascorbic acid in chilli peppers is leached because ascorbic acid is water soluble (Howard et al., 1994). However, red and green peppers result in significant reduction in the total of ascorbic acid after boiled for about 5 and 30 minutes. This could be caused by thinner pericarp layer than paprika varieties. Thinner cell membrane was more permeable to heat resulting to rapid leaching of ascorbic acid into the water as the cell die (Chuah et al., 2008).

In addition, Ornelas-paz et al. (2013) said that boiling at 210^{o}C for 8.8-19 minutes and grilling at 96^{o}C for 7-13.5 minutes significantly reduce the content of ascorbic acid content in chilli peppers. Grilling result higher losses of ascorbic acid content than boiling, however the differences the ascorbic acid content between both processes are small. According to Ornelas-paz et al. (2013), leaching of ascorbic acid into boiling water has a higher detrimental effect than degradation of ascorbic acid caused by high temperature.

Masrizal et al. (1996) also said that microwave steaming and stir-frying have affected to the higher retention level of ascorbic acid than boiling. Howard et al. (1994) reported that lower levels of ascorbic acid have been found due to losses during water blanching and thermal processing. According to Roy et al. (2014), pressure cooking and microwave heating decrease ascorbic acid contents of red chilli pepper. However, different from red chilli peppers, ascorbic acid increase during pressure cooking and microwave heating on the yellow and green chilli pepper. Yellow chilli pepper contains the highest ascorbic acid content compared with green and red chilli pepper, while green chilli pepper has the lowest ascorbic acid content. Microwave heating generates higher ascorbic acid content than pressure cooking in all chilli peppers. It might be caused due to the temperature used in microwave heating is lower than pressure cooking.

In their report, Hwang et al. (2012) found that ascorbic acid will decrease as cooking time increase. The amount of ascorbic acid content decrease as an increase of cooking time for boiled, stir-fried, steamed, and roasted chilli peppers are 55.80-75.87, 105.82-119.95, 88.51-98.66, and 103.46-109 mg/100g, respectively. Boiling method yield highest decrease of ascorbic acid compared with steaming, stir-frying, and roasting. After cooking for 15 minutes, boiled chilli peppers will loss ascorbic acid content for about 66.5%, while steamed, stir-fried, and roasted chilli peppers will result in loss of ascorbic acid for about 34.2%, 14.0%, and 25.9%, respectively.

The air-drying process can also decrease ascorbic acid content in chilli peppers. The Higher temperature of air-drying can lead to huge loss of ascorbic acid. Air-drying process with high temperature (90 ^{o}C), can lead to maximum loss of ascorbic acid until 98.2% (Vega-gálvez et al., 2009). According to Tunde-Akintunde (2010), drying can reduce ascorbic acid content on chilli peppers. On his work, he said that pretreatment method such as steam and water blanching before the chilli peppers are being dried can decrease the ascorbic acid content higher than chilli peppers without pretreatment method. Loss of ascorbic acid is because of application of heat on chilli peppers. Degradation of ascorbic acid was increased with the increased of drying temperature (Reis et al., 2013).

Several factors that can usually lead to reduced ascorbic acid are water solubility and mass transfer, heat sensitivity, and enzymic oxidation (Rumm-Kreuter & Demmel, 1990). Ascorbic acid is easily too oxidized even if it was stored in the refrigerated storage. Decrease rate of ascorbic acids during storage are different for each. It depends on plant matrix, the condition of pre-harvest, the condition of postharvest, and initial ascorbic acids concentration (Howard et al., 1995). Lee & Howard (1999) also complement that differences amount of vitamin C retention can be caused due to differences in genetic, blanching method, the temperature used, time used, maturity stage, brine composition, and cultivar (Howard et al., 1994). In table 6, the changes of ascorbic acid during processing of chilli pepper are shown.

Bhattacharya et al. (2010) reported that there is some correlation between some variables capsaicin, carotene, ascorbic acid, phenol, peroxidases, and catalase. The increasing of carotene, capsaicin, and catalase activity will decrease the contents of ascorbic acid, phenol, and peroxidase activity. There is no association between carotene and ascorbic acid because carotene is existed in the lipid phase and ascorbic acid in the aqueous phase. Ascorbic acid also has an inverse relationship with capsaicin. The increasing of phenol would associate with the increasing of peroxidase activity.

Iqbal et al. (2015) reported that ascorbic acid of chilli peppers will significantly decrease during 5 months of storage. Higher temperature will cause a higher decrease in ascorbic acid. The amount of ascorbic acid retention during storage with temperature at 20^{o}C, 25^{o}C, and 30^{o}C is 87.8%, 77.9% and 72.4%, respectively in polyethylene bags, while in jute bags, total amount of ascorbic acid during storage with temperature at 20^{o}C, 25^{o}C, and 30^{o}C are 85.3%, 73.8% and 66.8%, respectively.

Table 6. Effect of Processing Methods on Ascorbic Acid

Species	colour	Method	Temperature	Time	Weight (g)	Size (cm)	Changes of ascorbic acid (%)	References
Capsicum frutescens	NA	Oven drying[4]	60°C	25 hour	30	9x5x3	-96.875	(Tunde-Akintunde, 2010)
Capsicum frutescens	NA	Oven drying[5]	60°C	25 hour	30	9x5x3	-97.656	(Tunde-Akintunde, 2010)
Capsicum frutescens	NA	Oven drying	60°C	25 hour	30	9x5x3	-84.375	(Tunde-Akintunde, 2010)
Capsicum frutescens	NA	Solar drying[4]	45°C	45 hour	30	9x5x3	-95.313	(Tunde-Akintunde, 2010)
Capsicum frutescens	NA	Solar drying[5]	45°C	45 hour	30	9x5x3	-96.875	(Tunde-Akintunde, 2010)
Capsicum frutescens	NA	Solar drying	45°C	45 hour	30	9x5x3	-84.375	(Tunde-Akintunde, 2010)
Capsicum frutescens	NA	Sun drying[4]	37°C	50 hour	30	9x5x3	-96.875	(Tunde-Akintunde, 2010)

[4] = Pre-treated with water blanching
[5] = Pre-treated with steam blanching
NA= Not Available

Table 6. (Continued)

Species	colour	Method	Temperature	Time	Weight (g)	Size (cm)	Changes of ascorbic Acid (%)	References
Capsicum frutescens	NA	Sun drying[5]	37°C	50 hour	30	9x5x3	-96.875	(Tunde-Akintunde, 2010)
Capsicum frutescens	NA	Sun drying	37°C	50 hour	2 (sample) 20(water)	9x5x3	-89.063	(Tunde-Akintunde, 2010)
Capsicum annuum	Green	Pressure cooking	NA	15 min	2 (sample) 20 (water)	NA	10.309	(Roy et al., 2014)
Capsicum annuum	Red	Pressure cooking	NA	15 min	2 (sample) 20 (water)	NA	-18.826	(Roy et al., 2014)
Capsicum annuum	Yellow	Pressure cooking	NA	15 min	2 (sample) 20 (water)	NA	11.647	(Roy et al., 2014)
Capsicum annuum	Green	Microwave heating	160°C	30 min	2 (sample) 20 (water)	NA	23.093	(Roy et al., 2014)
Capsicum annuum	Red	Microwave heating	160°C	30 min	2 (sample) 20 (water)	NA	-5.885	(Roy et al., 2014)
Capsicum annuum	Yellow	Microwave heating	160°C	30 min	2 (sample) 20 (water)	NA	24.933	(Roy et al., 2014)

[4] = Pre-treated with water blanching
[5] = Pre-treated with steam blanching
NA= Not Available