J Food Sci Technol (August 2018) 55(8):3001–3007
https://doi.org/10.1007/s13197-018-3218-7

ORIGINAL ARTICLE

Inhibitory effect of chemical and natural anti-browning agents on polyphenol oxidase from ginger (*Zingiber officinale* Roscoe)

Win Yee Lim[1] · **Chen Wai Wong**[1]

Revised: 3 May 2018 / Accepted: 9 May 2018 / Published online: 19 May 2018

Abstract Food browning is undesirable as it causes deterioration in food quality and appearance. This phenomenon was related to polyphenol oxidase (PPO), which catalyzes conversion of phenolic compounds into *o*-quinones. The present work evaluated the use of chemical and natural anti-browning agents to prevent the browning of ginger PPO. Sodium metabisulfite (5 mM) is a better chemical inhibitor compared to L-cysteine and sodium chloride as 55.00% of ginger PPO inhibition was achieved. The percentage of inhibition increased as the concentration of anti-browning agents increases. The addition of heated onion, chili pepper and pineapple extracts exhibited a stronger inhibitory effect on ginger PPO than unheated extracts. Heated chili pepper extract was the best natural inhibitor found in this study and it inhibited the ginger PPO (47.97%) mixed-competitively. Natural anti-browning agents have potential to be used to control the browning of ginger as well as other vegetables and fruits.

Keywords Polyphenol oxidase · Ginger · Anti-browning agents · Inhibition

Introduction

Ginger, scientifically named as *Zingiber officinale* Roscoe, belongs to the *Zingiberaceae* family which is a large group consisting of many well-known perennial herbs (Singh et al. 2012). The plant is grown mainly for its rhizome which was widely used for cosmetic, flavouring, medicinal and ornamental purpose. It was found that ginger exhibits antioxidant, antimicrobial, antiemetic anti-tumour and anti-inflammatory properties. It also helps in enhancing gastrointestinal motility, treating digestion problem, reducing cholesterol and reducing nausea (Ramachandran 2008). Therefore, ginger was widely used as a culinary spice and herbal remedy due to its high nutritional and medicinal value.

In food industry, food quality assurance during food processing and storage was critical. Food browning is currently a major concern in food processing and storage sector. It is defined as the phenomenon where brownish black pigment formed on the surface of food under chemical reactions that occurred within (Corzo-Martinez et al. 2012). Normally, browning reaction occurs in fruits, vegetables, seafoods and cereals. Browning effect can deteriorate the marketability, food nutritional value, appearance and consumers' acceptance on the food, causing reduction in shelf life of food and affect economic situation of a country.

Polyphenol oxidase, PPO (EC 1.14.18.1) is a copper-containing oxidoreductase which is universally found in plants (Othman 2014). It is also known as tyrosinase, catechol oxidase, and monooxygenase. It catalyses the hydroxylation of monophenol to diphenol and subsequently oxidation of diphenol into quinone compound, which will then polymerize and form dark pigment melanin (Araji et al. 2014). Polyphenol oxidase is a plastid-localized enzyme which normally being separated from its substrate, phenolic compounds by cell compartments (Escobar et al. 2008). Mechanical injuries and tissue damage such as slicing, peeling and cutting on plant cells cause the

✉ Chen Wai Wong
wongcw@ucsiuniversity.edu.my

[1] Department of Biotechnology, Faculty of Applied Sciences, UCSI University, No. 1, Jalan Menara Gading, UCSI Heights, 56000 Kuala Lumpur, Malaysia

interaction between PPO and phenolic compounds, thus browning reaction occur.

Control of enzymatic browning was essential as to maintain food quality and to ingratiate consumers' needs. Food industry are currently trying to eliminate the usage of chemical in food due to the awareness of the side effects that may brought by the chemicals. The PPO inhibitors occurring in natural resources have been studied in several plants such as potato (Lee et al. 2002), pear (Kim et al. 2005) and banana (Lee 2007). However, research on the natural anti-browning agents on ginger is yet to be established. There is limited scientific information on how the natural inhibitors may influence the PPO activity for ginger. Thus, the inhibitory effects of natural inhibitors (honey, onion, chili pepper and pineapple extracts) on ginger PPO were examined in this study.

Materials and methods

Plant material and natural inhibitors

Ginger (*Zingiber officinale* Roscoe) originated from Bentong, Pahang, Malaysia was used in this study. It was purchased from a local morning market (Pandan Perdana, Kuala Lumpur). Red onion (*Allium* cepa) and chili pepper (*Capsicum annuum*), were also bought from the local morning market (Pandan Perdana, Kuala Lumpur). Red onion and chili pepper were originated from China and Malaysia, respectively. Honey (Capilano Manuka honey Australia) and pineapple (*Ananas comosus*) from Malaysia were bought from Giant supermarket (Taman Connaught, Cheras, Kuala Lumpur). All chemicals used in this study were of analytical grade and were used as obtained.

PPO extraction

The extraction method was adapted from Lin et al. (2016) with slight modification. 100 g of ginger was first washed, peeled and cut into small pieces. The ginger was then homogenized with 2 g of Polyvinylpolypyrrolidone (Sigma Aldrich, St. Louis, MO, USA) in 200 mL of pre-chilled (4 °C) 0.1 M phosphate buffer at pH 6.8 under 22,000 rpm for 1 min using a LB-8011ES industrial blender (Waring Laboratory, Torrington, CA, USA). The homogenate was centrifuged at 7000 rpm for 20 min at 4 °C (Centrifuge Universal 320R, Hettich Zentrifugen, Tuttlingen, Germany). The supernatant obtained was filtered under vacuum with Buchner filtering system (WP6211560 Vacuum-pressure pump, Millipore Sigma, Burlington, MA, USA). The filtrate was used as the crude PPO extract.

PPO assay

PPO activities were assayed with 4-methylcatechol and pyrocatechol at 410 nm as the substrates by using spectrophotometric method with slight modifications (Joseph et al. 2012). The reaction mixture included 0.1 mL enzyme solution, 1.0 mL of 0.1 M substrate solution in 1.9 mL 0.1 M phosphate buffer at pH 6.8. Then, the mixture was transferred into a cuvette and absorbance readings were measured at 15 s intervals for 5 min using a spectrophotometer (Secomam UviLine 9400, Champigny-sur-Marne, France) at room temperature. The blank consisted of 1 mL substrate solution in 2 mL of phosphate buffer. The initial velocity was calculated from the slope of the absorbance value against time curve. One unit (U) of PPO activity was defined as amount of enzyme that caused a 0.001 absorbance change per min (Kim and Kim 2013).

Effect of anti-browning agents

Chemical inhibitors used were L-cysteine, sodium chloride and sodium metabisulfite. PPO activities were assayed with inhibitor at three different concentrations (1, 3 and 5 mM) spectrophotometrically for 4-methylcatechol and pyrocatechol, respectively. Reaction mixture included 0.1 mL crude PPO, 1.9 mL substrate in 0.1 M buffer at pH 6.8, and 1 mL inhibitor. The percentage inhibition (%) values for each inhibitors were calculated using the PPO activity without inhibitor as the initial PPO activity. I_{50} values, which is the concentration of inhibitor needed to show 50% inhibitory effect on enzyme, was obtained from the percentage inhibition. Michaelis–Menten constant (K_m), maximum velocity (V_{max}), dissociation constant (K_i) and type of inhibition for each inhibitor were also determined.

Effect of natural inhibitors

Honey, red onion extract, red chili pepper extract and pineapple juice were used as natural inhibitors. Honey was tested with three different concentrations, which were 1, 3 and 5% (v/v) (Gacche et al. 2009). Onion, chili pepper and pineapple extracts were used freshly and after heating with a concentration of 10 mg/mL.

Extraction of onion, chili pepper and pineapple were carried out according to the method of Yapi et al. (2015) with slight modifications. 90 g of each natural inhibitor was first homogenized with 90 mL pre-chilled (4 °C) distilled water at maximum speed (22,000 rpm) for 1 min. The homogenate was filtered through cloth. The filtrate was centrifuged at 7000 rpm for 20 min at 4 °C. The supernatant obtained was filtered through Buchner funnel with filter paper and the filtrate was used as the fresh inhibitor. Heated inhibitors were prepared by incubating the fresh

extracts of inhibitors at 95 °C for 15 min. The inhibitors were subjected to centrifugation at 10,000 rpm for 60 s if there is precipitation. 50 μL of heated inhibitors were added into 4.95 mL distilled water to prepare 5 mL of 10 mg/mL inhibitors.

PPO activity was assayed and expressed as percentage inhibition (%). Same as the study of inhibitory effect of anti-browning agents, Michaelis–Menten constant (K_m), maximum velocity (V_{max}) and dissociation constant (K_i) were also determined.

Statistical analysis

Statistical analyses of all assays were performed with Microsoft Office Excel. All experiment was performed with triplicate measurements. The data collected were presented as mean ± standard deviation (SD) and also as percentage relative activity.

Results and discussion

PPO extraction

Enzymes are pH dependent as changes in pH may cause impairment of enzyme-catalyzed biochemical reactions (Buchholz et al. 2012). Buffer, which can maintain pH of a solution at or near to certain value, was used in the extraction method to maintain a suitable pH for PPO. As referred to Chikezie et al. (2013), most PPO extracted from food source were found to have maximum activity near neutral pH value, so phosphate buffer of pH 6.8 was used as extraction buffer in this study. Enzymes are also temperature dependent. Hence, pre-chilled Waring blender and phosphate buffer at 4 °C were used in the extraction process to maintain temperature as heat may be produced during homogenization process. All studies of PPO were also carried out at 4 °C to avoid denaturation of enzyme.

Polyvinylpolypyrrolidone (PVPP) was added during homogenization process into the buffer solution. PVPP is a protective agent which it can bind to phenolic compounds, avoid the browning reaction between PPO and its substrates, phenolic compounds (Rojas-Grau et al. 2008).

PPO activity assay

Ginger PPO activities determined in this study were 13,450 EU/min/mL for 4-methylcatechol and 9040 EU/min/mL for pyrocatechol, respectively. This indicates that ginger PPO exhibits a higher catalytic reaction towards 4-methylcatechol.

Effect of anti-browning agents

Table 1 shows the inhibitory effect of various anti-browning agents on ginger PPO with 4-methylcatechol and pyrocatechol as substrates. It can be seen that the inhibition percentage increased as concentration of inhibitors increased from 1.0 to 5.0 mM. Sodium metabisulfite shows the greatest effect in inhibiting ginger PPO as it exhibited 46.36% inhibition at 1.0 mM. Furthermore, sodium metabisulfite has the smallest I_{50} and K_i value as compared to sodium chloride and L-cysteine, which signifies its strongest binding affinity to enzymes (Sharma 2012). L-cysteine and sodium chloride might not be good inhibitors for ginger PPO as low inhibition percentage was obtained (6.63–28.18%). These further prove that sodium metabisulfite has greater efficiency in inhibiting ginger PPO.

From Table 1, Ginger PPO with sodium chloride and L-cysteine exhibited the lowest K_m value by using 4-methylcatechol and pyrocatechol, respectively. V_{max} values were decreased from the initial V_{max} value of control for all anti-browning agents used which indicates the number of substrates used by PPO to produce products per unit time were reduced (Egelhofer 2016).

L-cysteine is a thiol containing compound which it can react with copper in active site of PPO, inactivate catalytic ability of the enzyme (Saeidian 2016). L-cysteine also able to inhibit browning effect by interacting with quinone compound, make it unable to undergo polymerization and produce melanin pigment (Gacche et al. 2006). In this study, mixed-type inhibition and un-competitive-type inhibition of L-cysteine on ginger PPO using 4-methylcatechol and pyrocatechol, respectively were observed (Table 1). Inhibition of polyphenol oxidase activity by L-cysteine has been reported in browning control of acanthus PPO (Saeidian 2016) and wild mushroom PPO (Dedeoglu and Guler 2009).

Sodium chloride was sodium salt, which act as agent of firmness to inhibit browning effects. It is reported by Ayala-Zavala and Gonzalez-Aguilar (2010) that chloride was a weak inhibitor. Sodium chloride was found to be un-competitive inhibitors for ginger PPO (Table 1). This result was similar to those reported by Yong and Hye (2007) on potato PPO, which stated that NaCl may interfere with enzyme–substrate complex and cause inactivation of PPO.

Sodium metabisulfite ($Na_2S_2O_5$) is a type of sulfites which is reported to be highly effective in control of browning effect. Sulfites are reducing agent which it can react with *o*-quinone and form diphenols which can help to prevent the polymerization of *o*-quinone into coloured pigment (Kilic-Akyilmaz and Gulsunoglu 2015). In this study, sodium metabisulfite non-competitively inhibited ginger PPO using both 4-methylcatechol and pyrocatechol

Table 1 Effect of chemical inhibitors on *Zingiber officinale* R. PPO

Substrates	Inhibitors	[I] (mM)	I_{50} (mM)	Inhibition (%)	V_{max} (EU/min/mL)	K_m (mM)	K_i (mM)	Type of inhibition
4-Methylcatechol	Control	–	–	–	12,903.23	7.37	–	–
	L-cysteine	1	8.82	12.27	10,752.69	7.53	5.00/4.42	Mixed
		3		20.90	10,204.08	7.85	11.34/8.65	
		5		28.18	9803.92	7.89	15.82/12.23	
	Sodium chloride	1	8.74	10.91	11,764.71	6.84	10.33	Un-competitive
		3		20.91	10,000.00	6.12	10.33	
		5		28.18	9259.26	5.56	12.70	
	Sodium metabisulfite	1	3.62	46.36	6896.55	7.16	1.14	Non-competitive
		3		50.00	6666.67	7.08	3.21	
		5		55.00	5970.15	7.16	4.31	
Pyrocatechol	Control	–	–	–	11,299.44	4.24	–	–
	L-cysteine	1	10.44	6.63	10,416.67	3.75	11.80	Un-competitive
		3		16.84	9569.38	3.32	16.59	
		5		23.47	8771.93	3.07	17.35	
	Sodium chloride	1	10.68	11.22	10,000.00	4.00	7.70	Un-competitive
		3		18.37	9478.67	3.63	15.62	
		5		23.47	8403.36	3.36	14.51	
	Sodium metabisulfite	1	10.28	16.84	9345.79	4.21	4.78	Non-competitive
		3		21.94	8771.93	4.21	10.42	
		5		24.49	8196.72	4.22	13.21	

as substrates (Table 1). Inhibitory effect of sodium metabisulfite has been studied in the browning control of grape PPO (Sanchez-Ferrer and Garcia-Carmona 2010) and apple PPO (Cortez-Vega et al. 2008).

Effect of honey on PPO activity of ginger

Inhibition parameters of honey on ginger PPO were reported in Table 2. 5% (w/v) honey exhibited a lower percentage of inhibition for 4-methylcatechol (1.4%) than that of pyrocatechol (42.57%). This indicates that honey exhibits a greater inhibition towards ginger PPO using pyrocatechol compared to 4-methylcatechol. There was an increase in the inhibition percentage with the increase of honey concentrations for both substrates used.

The V_{max} value was unchanged and K_m value was increased for PPO with honey using 4-methylcatechol. It can be concluded that honey was a competitive inhibitor (Table 2) and able to diminish the rate of catalysis by reducing the proportion of enzymes molecules bound to a substrate (Wong and Lee 2014). However, honey exhibited non-competitively inhibition towards ginger PPO when

Table 2 Effect of honey which used as natural inhibitor on *Zingiber officinale* R. PPO activity

Substrates	Inhibitors	Concentration (%) (w/v)	Inhibition (%)	V_{max} (EU/min/mL)	K_m (mM)	Type of inhibition
4-Methylcatechol	Control	–	–	8547.01	4.53	–
	Honey	1	1.40	8264.46	5.69	Competitive
		3	2.10	8000.00	6.2	
		5	2.10	8000.00	7.2	
Pyrocatechol	Control	–	–	8333.33	4.39	–
	Honey	1	28.38	5847.95	4.37	Non-competitive
		3	32.43	5633.80	4.39	
		5	42.57	4938.27	4.42	

pyrocatechol was used as the substrate, whereby the K_m value was unchanged (Table 2). Honey did not compete with substrate for enzyme but bound with enzyme at a site other than active site (Engelking 2015).

Mechanism of honey in inhibiting PPO varies depends on the variety of honey used, source of PPO and substrates used (Lozano 2006). Haard and Simpson (2000) have suggested that honey acts as chelating agent to inhibit PPO. A small peptide with molecular weight of 600 found in honey may chelate copper of PPO active site thus inactivate the enzyme. Moreover, high amount of components such as phenolic acids, flavonoids and vitamins found in honey has implied its antioxidant properties (Lee 2013).

Effect of onion, chili pepper and pineapple extracts on PPO activity of ginger

Table 3 shows the inhibitory effect of onion, chili and pineapple extracts on PPO activity of ginger. All heated extracts exhibited stronger inhibition percentage than non-heated extracts for both 4-methylcatechol and pyrocatechol. It can be concluded that heated extracts could have reduced the capability of the binding of enzyme to substrates greater than the fresh extracts. This could probably due to the occurrence of non-enzymatic browning, Maillard reaction in the extracts. Maillard reaction products (MRPs) were found to exert great antioxidant properties which can control browning reaction in food (Phisut and Jiraporn 2013). Shaimaa et al. (2016) has also reported that heating process has increased flavonoids and phenolics content in chili pepper extract as well as increased in its antioxidant capability, which would cause an increase in inhibition percentage as well. Similar results have been found by Wong and Lee (2014) on cassava leaves PPO and Kim et al. (2007) on peach juice PPO whereby heated onion extracts inhibited PPO activity markedly.

According to Table 3, chili pepper extract shows the greatest inhibition percentage (22.90–47.97%) on ginger PPO for both 4-methylcatechol and pyrocatechol as compared to onion and pineapple extracts. This was in agreement with those reported by Mercimek et al. (2015) in which 44% inhibition on potato PPO was obtained with chili pepper extract. Chili pepper extract was an un-competitive and mixed inhibitor for ginger by using 4-methylcatechol and pyrocatechol, respectively (Table 3). A large amount of phenolic and flavonoid compounds as well as high ascorbic acid content were found to be the causes of antioxidant properties in chili pepper, which inhibit the formation of undesired brown pigment (Shaimaa et al. 2016).

Ginger PPO inhibited by chili pepper extract exhibited the lowest K_m and V_{max} values for both non-heated and heated extracts by using 4-methylcatechol. This suggesting a stronger affinity of PPO towards 4-methylcatechol, which prevent the dissociation of enzyme–substrate complex. In contrast, chili pepper extract inhibited ginger PPO exhibited the highest K_m and lowest V_{max} value by using pyrocatechol for both non-heated and heated extracts. The reduction in binding affinity of ginger PPO and pyrocatechol was occurred in the presence of chili pepper extract.

From Table 3, it was found that both heated and non-heated onion extract exhibited similar type of inhibition, which was mixed-type inhibition. However, onion was found to competitively inhibited yam PPO (Yapi et al. 2015) and non-competitively inhibited cassava leaves PPO (Wong and Lee 2014). Onion possesses sulfhydryl or thiol

Table 3 Effect of 10 mg/mL onion, chili and pineapple extract which act as natural inhibitor to *Zingiber officinale* R. PPO activity

Substrates	Inhibitors	Heated/non-heated	Inhibition (%)	V_{max} (EU/min/mL)	K_m (mM)	Type of inhibition
4-Methylcatechol	Control	–	–	12,658.23	6.35	–
	Onion	Non-heated	14.02	11,494.25	6.80	Mixed
		Heated	15.89	11,111.11	6.90	
	Chili	Non-heated	22.90	9615.38	4.44	Un-competitive
		Heated	28.50	9009.01	4.12	
	Pineapple	Non-heated	17.76	10,000	6.35	Non-competitive
		Heated	21.50	9259.26	6.29	
Pyrocatechol	Control	–	–	8928.27	4.39	–
	Onion	Non-heated	28.38	6250	4.59	Mixed
		Heated	33.11	5882.35	4.62	
	Chili	Non-heated	40.54	5128.21	5.95	Mixed
		Heated	47.97	4597.70	6.10	
	Pineapple	Non-heated	25.00	6389.78	4.31	Non-competitive
		Heated	27.03	5882.35	4.37	

(–SH) group which able to bind copper at PPO active site, interfere with catalytic activity of PPO. Flavonoid compound and thiol groups found in onion have identified its antioxidant ability which can also help to control browning reaction (Akhtar 2015).

Non-competitive inhibition of pineapple juice on ginger PPO was obtained for both substrates used as shown in Table 3. Unchanged K_m values and reduced V_{max} values indicates the non-competitive inhibitors did not reacted with the enzyme at the active site. Pineapple contains ascorbic acid, reviewing the antioxidant properties of pineapple which can control PPO activity (Antoniolli et al. 2012). Chaisakdanugull et al. (2007) has suggested that main organic acids like malic and citric acids also played an important role in inhibiting banana puree PPO. Cysteine and other sulfhydryl compounds present in pineapple extract also contribute to PPO inhibition. The effectiveness of pineapple juice in controlling browning reaction has been tested on rose apple fruit PPO by Supapvanich et al. (2012).

Conclusion

This study concluded that heated chili pepper extract was the best natural inhibitor to inhibit ginger PPO. Sodium metabisulfite showed a stronger inhibition on ginger PPO in this study. However, the use of natural inhibitors such as honey, onion, chili pepper and pineapple extract can be used to replace the sulphite-containing agents which could be harmful to human health.

Acknowledgements This research work was financially supported by the Centre of Excellence for Research, Value Innovation and Entrepreneurship (CERVIE) under Pioneer Scientist Incentive Fund (PSIF) with the code of Proj-In-FAS-052, UCSI University, Malaysia.

References

Akhtar S (2015) Advances in conventional breeding approaches for postharvest quality improvement in vegetables. In: Siddiqui MW (ed) Postharvest biology and technology of horticultural crops: principles and practices for quality maintenance. CRC Press, Boca Raton, pp 141–192

Antoniolli LR, Benedetti BC, de Souza Filho MSM, Garruti DS, de Borges MF (2012) Shelf life of minimally processed pineapples treated with ascorbic and citric acids. Bragantia 71(3):447–453

Araji S, Grammer TA, Gertzen R, Anderson SD, Mikulic-Petkovsek M, Veberic R, Phu ML, Solar A, Leslie CA, Dandekar AM, Escobar MA (2014) Novel roles for the polyphenol oxidase enzyme in secondary metabolism and the regulation of cell death in walnut. Plant Physiol 164(3):1191–1203

Ayala-Zavala JF, Gonzalez-Aguilar GA (2010) Use of additives to preserve the quality of fresh-cut fruits and vegetables. In: Martin-Belloso O (eds) Advances in fresh-cut fruits and vegetables processing. CRC Press, Boca Raton, pp 231–254

Buchholz K, Kasche V, Bornscheuer UT (2012) Biocatalysts and enzyme technology. Wiley, Weinheim

Chaisakdanugull C, Theerakulkait C, Wrolstad RE (2007) Pineapple juice and its fractions in enzymatic browning inhibition of Banana [*Musa* (AAA Group) Gros Michel]. J Agric Food Chem 55(10):4252–4257

Chikezie PC, Akuwudike AR, Chikezie CM (2013) Thermal and pH stabilities of partially purified polyphenol oxidase extracted from *Solanum melongenas* and *Musa sapientum* fruits. Afr J Biotechnol 12(38):5688–5694

Cortez-Vega WR, Becerra-Prado AM, Soares JM, Fonseca GG (2008) Effect of L-ascorbic acid and sodium metabisulfite in the inhibition of the enzymatic browning of minimally processed apple. Int J Agric Res 3(3):196–201

Corzo-Martinez M, Corzo N, Villamiel M, Castillo MD (2012) Browning reactions. In: Simpson BK (ed) Food biochemistry and food processing. Wiley, Hoboken, pp 56–83

Dedeoglu N, Guler OO (2009) Differential in vitro inhibition of polyphenoloxidase from a wild edible mushroom Lactarius salmonicolor. J Enzyme Inhib Med Chem 24(2):464–470

Egelhofer V (2016) Textbook bioinformatics. BoD-Books on Demand, Norderstedt

Engelking LR (2015) Textbook of veterinary physiological chemistry, 3rd edn. Academic Press Inc., USA, pp 32–38

Escobar MA, Shilling A, Higgins P, Uratsu SL, Dandekar AM (2008) Characterization of polyphenol oxidase from walnut. J Am Soc Hortic Sci 133(6):852–858

Gacche RN, Schete AM, Dhole NA, Ghole VS (2006) Reversible inhibition of polyphenol oxidase from apple using L-cysteine. Indian J Chem Technol 13:459–463

Gacche RN, Shinde BT, Dhole NA, Pund MM, Jadhav AD (2009) Evaluation of floral honey for inhibition of polyphenol oxidase-mediated browning, antioxidant and antimicrobial activities. J Food Biochem 33(5):693–706

Haard NF, Simpson BK (2000) Seafood enzymes: utilization and influence on postharvest seafood quality. CRC Press, Boca Raton

Joseph A, Thayumanavan B, Panicker PR (2012) Characterization of polyphenol oxidase in ginger (*Zingiber officinale* R.). J Spices Aromat Crops 21(1):33–41

Kilic-Akyilmaz M, Gulsunoglu Z (2015) Additives and preservatives. In: Hui YH (eds) Handbook of vegetable preservation and processing, 2nd edn. CRC Press, Boca Raton, pp 301–318

Kim JJ, Kim WY (2013) Purification and characterization of polyphenol oxidase from fresh ginseng. J Ginseng Res 37(1):117–123

Kim MJ, Kim CY, Park IS (2005) Prevention of enzymatic browning of pear by onion extract. Food Chem 89(2):181–184

Kim CY, Kim MJ, Lee MY, Park IS (2007) Inhibition of polyphenol oxidase and peach juice browning by onion extract. J Food Sci Biotechnol 16(3):421–425

Lee MK (2007) Inhibitory effect of banana polyphenol oxidase during ripening of banana by onion extract and Maillard reaction products. Food Chem 102(1):146–149

Lee MC (2013) United States Patent Application No. 13/336,180, Publication No. 20130164233 A1

Lee MK, Kim YM, Kim NY, Kim KN, Kim SH, Bang KS (2002) Prevention of browning in potato with a heat-treated onion extract. Biosci Biotechnol Biochem 66(4):856–858

Lin H, Ng AWR, Wong CW (2016) Partial purification and characterization of polyphenol oxidase from Chinese parsley (*Coriandrum sativum*). Food Sci Biotechnol 25(1):91–96

Lozano JE (2006) Fruit manufacturing: scientific basis, engineering properties, and deteriorative reactions of technological importance. Springer Science, New York

Mercimek HA, Güzeldağ G, Uçan F, Karayılan R (2015) Inhibition of polyphenol oxidase purified from potato (*Solanum tuberosum*). Rom Biotechnol Lett 20(6):10961–10968

Othman OC (2014) Polyphenoloxidase and perioxidase activity during open air ripening storage of pineapple (*Ananas comosus* L.), mango (*Mangifera indica*) and papaya (*Carica papaya*) fruits grown in Dar es Salaam, Tanzania. Tanzan J Sci 38(3):84–94

Phisut N, Jiraporn B (2013) Characteristics and antioxidant activity of Maillard reaction products derived from chitosan-sugar solution. Int Food Res J 20(3):1077–1085

Ramachandran A (2008) Grains, greens, and grated coconuts: recipes and remembrances of a vegetarian legacy. iUniverse Inc, Bloomington

Rojas-Grau MA, Soliva-Fortuny R, Martin-Belloso O (2008) Effect of natural antibrowning agents on color and related enzymes in fresh-cut Fuji apples as an alternative to the use of ascorbic acid. J Food Sci 73(6):267–272

Saeidian S (2016) Difference of diphenolase and monophenolase activity of partial purified polyphenol oxidase of *Gundelia Tournefortii* using some amino acids. Int J Basic Sci 5(1):62–70

Sanchez-Ferrer A, Garcia-Carmona F (2010) Activation and inhibition of grape polyphenol oxidase. Biochem Educ 20(4):235–237

Shaimaa GA, Mahmoud MS, Mohamed MR, Emam AA (2016) Effect of heat treatment on phenolic and flavonoid compounds and antioxidant activities of some egyptian sweet and chilli pepper. Nat Prod Chem Res 4(3):1–6

Sharma RR (2012) Enzyme inhibition: mechanisms and scope. In: Sharma RR (ed) Enzyme inhibition and bioapplications. InTech, New York, pp 3–37

Singh HP, Parthasarathy VA, Kandiannan K, Krishnamurthy KS (eds) (2012) *Zingiberaceae* crops: present and future: cardamom ginger turmeric and others. Westville Publishing House, New Delhi

Supapvanich S, Prathaan P, Tepsorn R (2012) Browning inhibition in fresh-cut rose apple fruit Cv. taaptimjaan using konjac glucomannan coating incorporated with pineapple fruit extract. Postharvest Biol Technol 73:46–49

Wong CW, Lee APL (2014) Inhibitory effect of onion extract on cassava leaf (*Manihot esculenta* Crantz) polyphenol oxidase. Int Food Res J 21(2):755–758

Yapi JC, Gnangui SN, Dabonné S (2015) Inhibitory effect of onions and garlic extract on the enzymatic browning of an edible yam (*Dioscorea cayenensis-rotundata* cv. Kponan) cultivated in Côte d Ivoire. Int J Curr Res Acad Rev 3(1):219–231

Yong KC, Hye KA (2007) Purification and characterization of polyphenol oxidase from potato: inhibition and catalytic mechanism. J Food Biochem 23(6):593–605