

Properties and Total Initial Activities of Three Oxidative Enzymes Isolated from some Fruits and Vegetables Grown in Egypt

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Abstract: The presence of residual endogenous quality-related fruit and vegetable enzymes in either raw or processed fruit or vegetable products may cause loss of quality during processing or storage. The content and properties of such enzymes in these raw sources vary widely with type, species, and variety of the sources and the nature of the environment in which they grew. This study was carried out to determine the optimum conditions (pH and temperature) and total initial activities of three quality-related oxidative enzymes [peroxidase (POD), polyphenol oxidase (PPO), and lipoxygenase, (LOX)] in three fruit varieties; mango (*Mangifera indica* var. Zebda), banana (*Musa cavendishii* var. Enana), (peel and pulp), olive (*Olea europaea* var. Picual) and three vegetables; green beans (*Phaseolus vulgaris* var. Littel Marvel), tomatoes (*Lycopersicon esculentum* var. Marmand), cucumber (*Cucumis sativus* var. Ria) grown in Egypt. The POD, PPO and LOX activities were detected in the six examined homogenates. The optimum pH and temperature values for high enzyme activities were varied according to the type of fruits and vegetables. The highest amount of POD was found in green beans extract (0.207 unit mg⁻¹ protein of the sample) and the lowest amount was for banana peel (0.087 unit mg⁻¹ protein). However, the green beans extract had the lowest content of PPO. In contrast, the highest content was for banana peel. For LOX activity, the mango extract exhibited the greater extent (0.249 unit mg⁻¹ protein) than cucumber and tomato extracts (0.228 and 0.211 unit mg⁻¹ protein, respectively). The activity of LOX was very low in the crude banana pulp homogenate (0.068 unit mg⁻¹ protein).

Keywords: Fruits, vegetables, peroxidase, polyphenol oxidase, lipoxygenase

INTRODUCTION

Enzymes are biocatalysts that are essential in the physiology and metabolism of plants. However, most enzymes remain active postharvest. Although this may be desired in cases where ripening takes place during postharvest storage (Terefe *et al.*, 2014). It is well known that the presence of residual endogenous enzymes in either raw or processed fruit or vegetable products may cause loss of quality during storage (Anthon and Barrett, 2002). It may also lead to detrimental changes in quality attributes such as color, flavor, texture and nutritional value. The activities of endogenous deteriorative enzymes considerably shorten the shelf life of horticultural products. The peroxidase (POD), polyphenoloxidase (PPO) and lipoxygenase (LOX) enzymes may be responsible for other color and flavor changes (Terefe *et al.*, 2014).

With the consumer's increased awareness of health benefits from consuming fresh fruits and vegetables, there is an increasing demand for higher quality of fresh-cut fruits and vegetables in a convenient form. In order to meet these demands, the food industry has focused on the development of new processing techniques for minimally processed fruit and vegetable products. Processing operations, such as peeling, cutting and shredding, induce enzymatic browning and enhance the ethylene synthesis, respiration, softening and microbial contamination, which all correlate with quality deterioration (Son *et al.*, 2001).

On the other hand, contents and properties of enzymes in raw fruit and vegetables vary widely with type, species, and variety of the sources and the nature of the environment in which they grew in.

The changes in ionization of prototropic groups in the active site of an enzyme at lower acid and higher alkali pH values may prevent proper conformation of

the active site, binding of substrates, and/or catalysis of the reaction (Tipton and Dixon, 1983; Whitaker, 1994). In addition, irreversible denaturation of the protein and/or reduction in stability of the substrate as a function of pH could also influence the catalytic activity of enzymes. Kinetic behavior of some enzymes such as polyphenol oxidase was reported to alter depending on the pH of the assay which may induce conformational changes in the enzyme (Valero and Garcia-Carmona, 1992; 1998).

Temperature is another important factor that significantly influences the catalytic activity of enzymes. It is well known that a decrease in the kinetic energy of the reactant molecules at low temperatures corresponds to a slower reaction (Nelson and Cox, 2008). In addition, integrity of the delicate three-dimensional structure of the enzyme molecule is subjected to disruption and denaturation at high temperatures (Whitaker, 1994).

Valuable information about the initial activities of endogenous enzymes in Egyptian fruits and vegetables is still lacking. So, this study aimed to evaluate the initial activities and the optimum conditions (pH and temperature) of three oxidative enzymes; POD, PPO, and LOX, the most quality related enzymes in three fruits [banana (B:pe and B:pu), mango (M) and olive (O)] and three vegetables [green beans (G), tomatoes (T) and cucumber (C)] grown in Egypt.

MATERIALS AND METHODS

Materials:

Raw materials

Mango (*Mangifera indica* var. Zebda), banana (*Musa cavendishii* var. Enana), olive (*Olea europaea* var. Picual), green beans (*Phaseolus vulgaris* var. Littel Marvel), cucumber (*Cucumis sativus* var. Ria) and

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tomatoes (*Lycopersicon esculentum* var. Marmand) were got at ripe stage from Ismailia Governorate local market, Egypt.

Chemicals and reagents

All fine chemicals used in determination of the enzyme activities were obtained from Sigma–Aldrich chemical company, while the other chemicals (solvents) were of analytical grade.

Methods:

Preparation of enzyme extracts

a. Peroxidase (POD) and lipoxygenase (LOX) extracts

The POD and LOX extracts were prepared according to the method described by Anthon and Barrett (2002) with some modifications. Studied fruits and vegetables raw samples were washed and cut into small pieces. Two hundred grams from each sample was mixed with 200 ml of 0.1 M phosphate buffer (pH 6) using a warring blender (Matsushita, ELEC, IND, CO, LTD, Japan). The slurry was homogenized with ultra Turrax homogenizer at 10,000 rpm for 2 min. The resulted homogenate was filtered through cheese cloth and then centrifuged at 3800 xg for 15 min then the supernatant was filtered (Whatman No 1). This supernatant was used in determination of POD and LOX activities.

b. Polyphenol oxidase (PPO) extracts

PPO was extracted according to Coseteng and Lee (1987) method, about 200 g of studied raw materials were homogenized in 400 ml of cold acetone (-25 °C), using a pre-chilled warring blender for 2 min at maximum speed. The slurry was filtered and the residue was re-extracted with 200 ml of cold acetone. This procedure was repeated until a white powder was obtained. The resultant acetone extract was dried overnight at room temperature and stored at (-25 °C). In order to obtain enzyme extract, a 0.5 g of acetone powder was suspended in 37.5 ml pre-chilled 0.1 M phosphate buffer (pH 6.8), and then stirred for 1 h at 4°C. The suspension was centrifuged at 7500 xg for 30 min at 4 °C. The supernatant was used as a crude PPO.

Protein determination

Protein content of the studied samples was determined spectrophotometrically as described by Janairo *et al.* (2015) using Biuret reagent test. For the test sample, 1 ml of the sample extract was added to 4.5 ml of Biuret reagent. The solutions were shaken using a vortex. The color of the solution was observed to be light blue and the absorbance of the protein samples at 545 nm was determined using the UV-VIS spectrophotometer (6505 UV/ VIS, Jenway LTD, Felsted, Dunmow, UK). A calibration curve ($R^2=9998$) of standard protein solution of bovine albumin (0.0 – 2.0 mg/ ml) was prepared and tested under similar conditions.

The pH and temperature optima of the examined enzymes

The pH optima: The optimum pH for POD, PPO and LOX activities was determined in the pH ranged from 4.5- 8.5 using acetate (4.5 - 5.5), phosphate (6.0 - 8.0) and boric-acid-borax (8.5) buffer adjusted with 0.1M

NaOH or 0.1M HCl, using the standard reaction mixture.

The temperature optima: The effect of temperature on the activity of studied enzymes was tested by heating the standard reactions (buffer and substrate) to the appropriate temperatures before addition of the enzyme extract. The activity was assayed at various reaction temperatures controlled by a circulation water bath. The temperature was varied over the range of 10 – 50 °C. The mixtures of buffer and substrate solution were incubated at the different temperatures. Once temperature equilibrium was reached, the enzyme extract was added to the mixture and the activity was determined spectrophotometrically as rapidly as possible at constant temperature.

Determination of enzyme activities

Peroxidase activity assay: The POD activity was determined according to the method described by Güneş and Bayindirli (1993). The substrate solution was prepared by mixing 0.5 ml guaiacol (99.5%), 0.5 ml hydrogen peroxide (30%), and 99 ml sodium phosphate buffer (pH 6.5). The POD assay was conducted by mixing 0.1 ml enzyme extract with 2.9 ml substrate solution. The increase in absorbance at 470 nm was measured with 10 s intervals. One unit of activity was defined as an initial absorbance (OD) change of 0.001/min under the assay conditions. Standard curve was prepared by plotting the OD values against time. The activity was measured from the linear phase of reaction. One unit of POD activity was defined as the change in OD value per minute per ml of enzyme extract.

Polyphenol oxidase activity assay: The PPO activity was assayed using the procedure of Cano *et al.* (1997). The enzyme activity was determined by measuring the rate of increase in absorbance at 420 nm and 25 °C using spectrophotometer (6505 UV/ VIS, Jenway LTD, Felsted, Dunmow, UK). The reaction mixture contained 2.9 ml of 0.07 M catechol solution in 0.05 M phosphate buffer (pH 7.0) and 100 µl of diluted (1:1, 0.2 M phosphate buffer pH7) enzyme extract. The activity was calculated on the basis of the slop of the linear portion of the curve of ΔA_{420} plotted against time (up to 3 min). The enzyme activity was expressed as $\Delta A_{420} \text{ min}^{-1} \text{ ml}^{-1}$ of enzyme extract.

Lipoxygenase activity assay: The LOX activity was measured by the method described by Bonnet and Croljzet (1977), based on absorption at 234 nm of the conjugated dines formed when linoleic acid (used as substrate) was oxidized in the presence of LOX. The substrate consisted of 10 µl of linoleic acid, 4 ml of H₂O, 1 ml of NaOH (0.1N) and 5 µl of tween 20. The mixture was shaken and diluted to 25 ml with distilled water. Each test contained 2.6 ml of phosphate buffer (0.2 M, pH 6.5) was mixed with 0.3 ml of substrate followed by the addition of 100 µl crude enzyme extract. The absorbance at 234 nm was followed over 7 min using spectrophotometer (6505 UV/ VIS, Jenway LTD, Felsted, Dunmow, UK). For blank, 2.6 ml phosphate buffer and 0.3 ml of substrate were used. One unit of LOX activity was defined as 0.1 increases in OD value at 234 nm/ min under the conditions described above.

Statistical analysis

All experiments were done in triplicates. Data were expressed as means \pm standard deviation (SD) by SPSS (version 17.0 SPSS Inc) program.

RESULTS AND DISCUSSION

The optimum pH and temperature of the POD, PPO and LOX activities:

Determination of the effects of pH value and temperature on the POD, PPO and LOX activities was carried out because of their importance in the control of enzymatic reactions in food processing.

Optimum pH

The effect of pH on the activity of POD enzymes was determined using buffer solutions at pH 4.5 - 8.5 (Figure, 1 A and B). As seen in Figure (1-A), M-POD extract showed maximum activity at pH 5.5 followed by a sharp decrease with increasing the pH value. Khan and Robinson (1994) reported that the optimum pH of purified mango isoperoxidase varied from pH 5.0 - 6.25. On the other hand, the optimum pH for mango soluble POD was 5.0, while ionically bound POD was 4.5 - 5.0. For B:pe-POD and B:pu-POD the optimum activities were observed at pH 6.5 and pH 6, respectively (Figure, 1-A). Similar results were obtained by Nagle and Haard (1975). They found that the maximum activity for ripe banana POD was between pH 5.0 - 6.0. While, MacDonald and Schaschke (2000) studied the POD

activity in banana at pH value of 7.0. T-POD exhibited optimum activity at pH 7.5 (Figure, 1-B). Suha *et al.* (2013) found that, T-POD revealed optimum activity at pH 7.0 - 8.0. From the same figure it was observed that the maximum activity of C-POD was at pH 7.0. Similar results were reported by Zhu *et al.* (2004) who mentioned that, the optimum activity for C-POD enzyme was at pH 7.0. Furthermore, Figure (1-B) demonstrated that, the optimum pH for G-POD activity was at pH 6.5. This behavior was similar to that used by Akyol (2004). Who measured the G-POD activity at optimum pH value of 6.5 during inactivation the enzyme by some methods such as high hydrostatic pressure and blanching at moderate temperatures (20 - 70 °C). Finally it can be seen that, the maximum activity for O-POD was detected at pH 7.0 (Figure, 1-B). The maximum activity of O-POD was found at pH 7.0 and decreased steeply for higher and lower values of pH (Saraiva *et al.*, 2007). In general, POD activity in plants shows variation in pH optima and this is depending on plant variety and substrate. The literature has been reported that, the optimum pH for maximum activity of POD were 5.4 in grape, 4.5 - 5.0 in banana, 4.2 in pineapple, 5.0 - 5.4 in potato, 5.5 in cabbage leaves, 5.2 in strawberries and 4.0 in marula fruit (Billaud *et al.*, 1999; Martínez *et al.*, 2001; Mdluli, 2005; Kharatmol and Pandit, 2012).

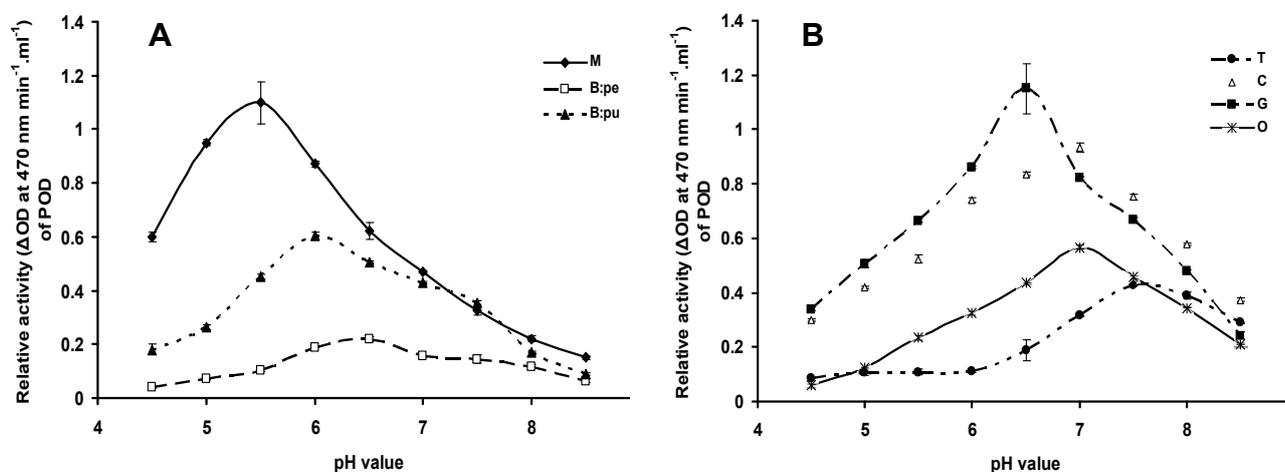


Figure (1): pH profiles of POD activity for investigated samples in 0.1 M acetate buffer (4.5 - 5.5), 0.1 M phosphate buffer (6 - 8) and 0.1 M boric acid–borax buffer (8.5). The reaction medium containing 2.9 ml 129.63 mM guaiacol, 2.9 ml 129.63 mM H₂O₂ and 0.1 ml of enzyme extract.

The optimum pHs for PPO of M, B:pe and B:pu were performed (Figure, 2-A). PPO showed pH optima at 7.0 in both B:pe and B:pu. The optimum pH of PPO activity varies widely with plant source but it is generally in the range of 4.0 - 8.0 (Yoruk and Marshall, 2003). Optimal value of pH 6.0 for M-PPO with catechol as a substrate have also been reported by Labib (1992). However, Yang *et al.* (2000) remarked an optimum pH for PPO banana pulp at pH 6.5 and the enzyme activity was stable in the range of pH 5 - 11 at 5 °C for 48 h. Also, from Figure (2-B) it can be noticed

that the T-PPO, C-PPO and, G-PPO had their maximal activities at pH 7.0. Identical results have been obtained by Lokhandwala and Bora (2014). They declared that, the optimum pH for tomato PPO activity was 7.0 in 0.1 M sodium phosphate buffer. Likewise, they reported that the enzyme activity measured at this pH was 53.54 μ moles/ ml/ min. However, PPO was found more stable between the ranges of pH 8 to pH 10 in 0.1M sodium phosphate buffer after incubation of 8 days. The PPO activity reduced to zero at pH 1, 2 and 3 after incubation for 6, 7 and 8 days, respectively; which indicated that

PPO was more stable at alkaline pH. Guo *et al.* (2009) claimed that the all isoforms of G-PPO activities were stable between pH 6.8 and pH 7.2, but they lost 90% of their original activities when adjusted to a pH below 6.2 or above 8.0. So the change of pH would greatly influence the stability of PPO in green beans. As well as the O-PPO had maximum activity at pH 6.0 (Figure, 2-B). Similar results were obtained by Ortega-Garcia *et al.*

(2008) who reported that the optimum pH of *Olea europaea* was 6.2. Generally, several researchers found that PPOs were stable near neutral pH and were unstable in acidic media, such as for longan fruit and yali pear (pH 7.0) (Zhou and Feng, 1991; Jiang, 1999), artichoke (pH 6.0– 7.0) (Aydemir, 2004), and marula fruit (pH 6.0) (Mdluli, 2005).

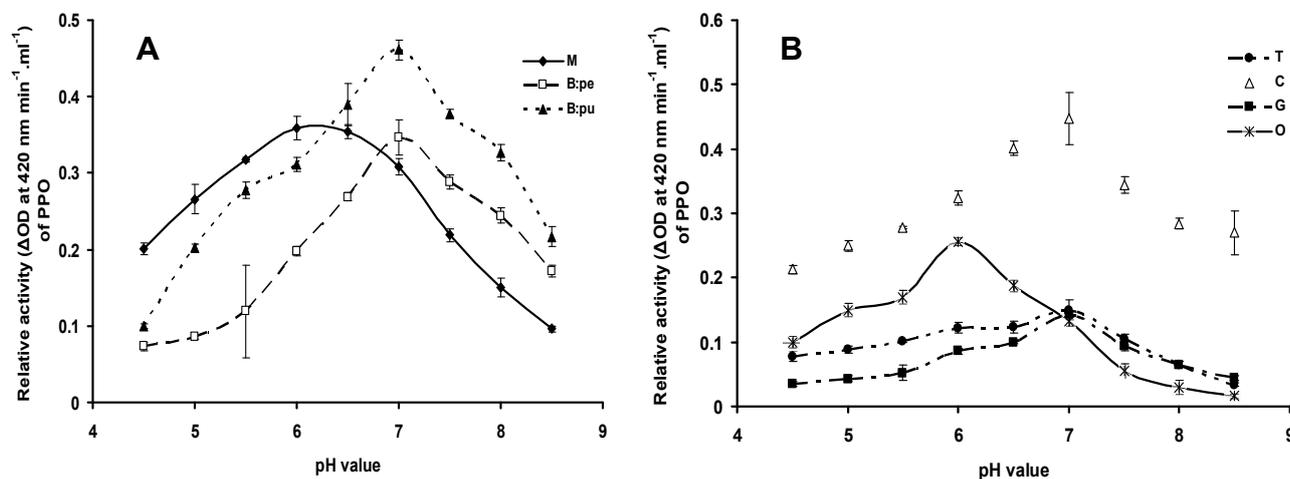


Figure (2): pH profiles of PPO activity for investigated samples in 0.1M acetate buffer (4.5 - 5.5), 0.1 M phosphate buffer (6 - 8) and 0.1 M boric acid–borax buffer (8.5). The reaction medium containing 2.5ml 1.25mM catechol and 0.5 ml of enzyme extract

The effect of pH on LOX activities of the seven plant materials was carried out (Figure, 3 A and B). For M extract, LOX showed maximum activity at pH 6.5. Rayan (2009) found the same result. While, Ding *et al.* (2007) studied the M-Lox activity during storage period at pH value of 7.0. In both B:pe and B:pu, the optimum pH for LOX activity was at 7.0 (Figure, 3-A). Kuo *et al.* (2006) found that the optimal pH of the purified LOX from banana leaf was 6.2 and at pH 5.5, 84% activity

was observed, while only 11% activity at pH 8.0. The T-LOX enzyme demonstrated maximum activity at pH 6.0 (Figure, 3-B). A similar optimum pH was found at pH 6.0 by Bowsher *et al.* (1992). The C-LOX showed height activity at pH 5.5 (Figure, 3-B). For G-LOX and O-LOX (Figure, 3-B) optimum activities were at pH 6.0. Indrawati *et al.* (2000) and Lorenzi *et al.* (2006) found equal figure. They reported that olive LOX activity showed its maximum at pH 6.0.

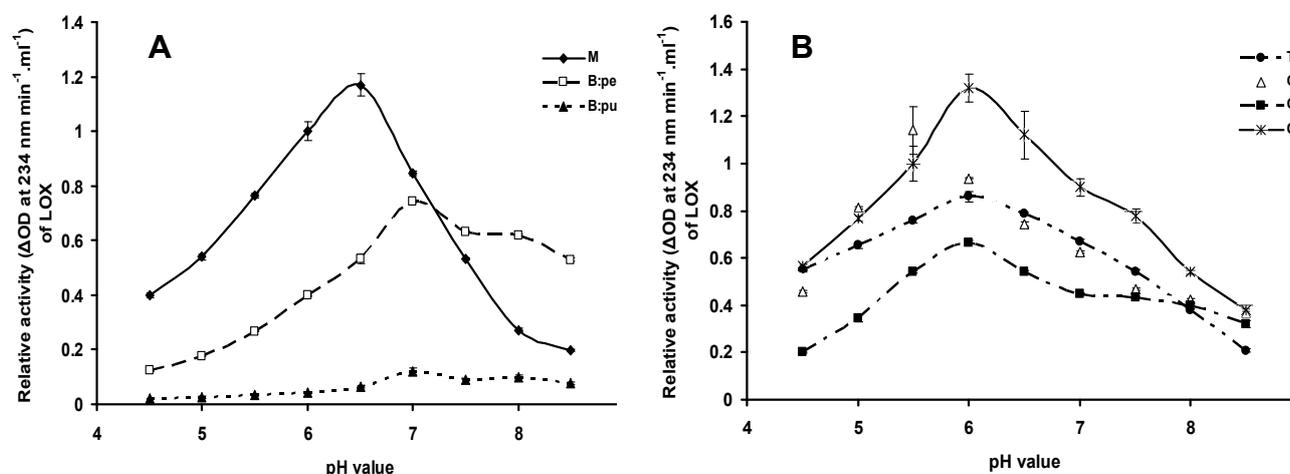


Figure (3): pH profiles of LOX activity for investigated samples in 0.1M acetate buffer (4.5 - 5.5), 0.1 M phosphate buffer (6 - 8) and 0.1 M boric acid–borax buffer (8.5). The reaction medium containing 2.9ml 1.2mM linoleic acid and 0.1 ml of enzyme extract

Optimum temperature

Reaction mixtures were prepared as described in materials and methods and then incubated at different temperatures (10 - 50 °C). Relative POD activities obtained from these tests are shown in (Figure, 4 A and B). Under the used working conditions, the M-POD had optimum temperature at 20 °C and stayed active at temperatures 25 - 30 °C (Figure, 4-A). Sugai and Tadini (2006) found similar results, where they reported that the suitable temperature for determination the POD activity of mango puree was at 25 °C. Rayan (2009) found that, the optimum temperature for M-POD activity was at 40 °C, and high activities at temperature ranged between 20 - 50 °C, were occurred. The B:pe-POD and B:pu-POD displayed a maximum activity at

30 °C (Figure, 4-A). Yadav *et al.* (2012) mentioned that purified POD from banana stem had optimum temperature at 25 °C. Additionally, Figure (4-B) demonstrates that the optimum temperatures for T, C and G-POD activities were at 25 °C. Similar results were announced by Quiroga *et al.* (2000), Akyol (2004) and Battistuzzi *et al.* (2004). They found that the activities of POD extracted from tomato, green beans and cucumber were optimum between 25 – 30 °C. With regard to O-POD its optimum temperature was at 35 °C. Tzika *et al.* (2009) studied the effect of the reaction temperature on the initial rate of olive POD catalyzed ABTS oxidation from 10 to 50 °C and observed that the optimal temperature for O-POD was between 25 and 30 °C.

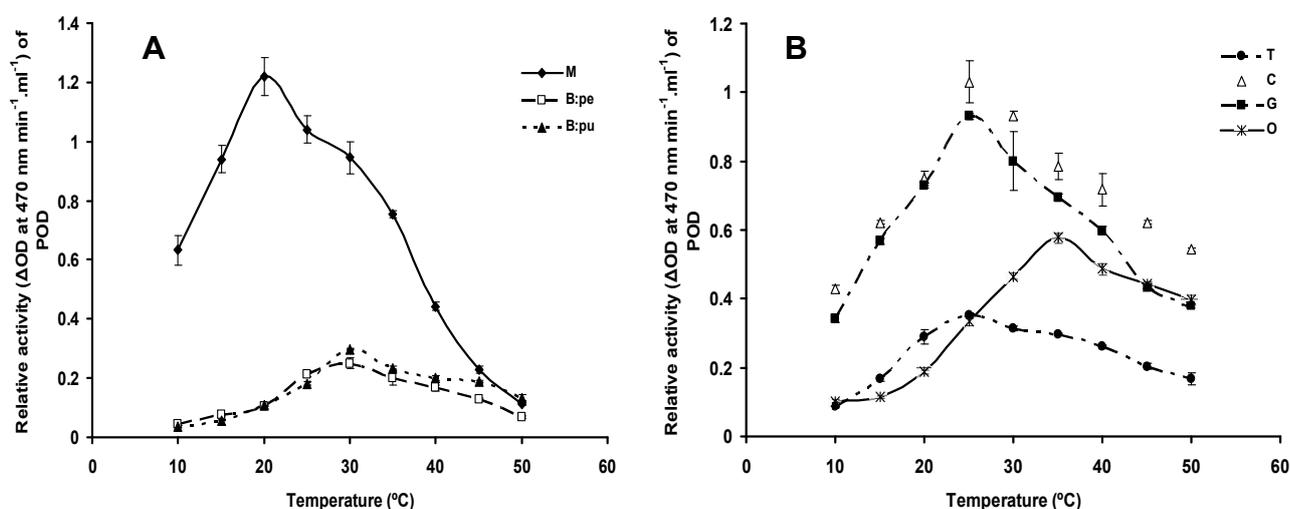


Figure (4): Effect of temperature on POD activity of investigated samples at optimum pH value for each. The reaction medium containing 2.9ml 129.63mM guaiacol, 2.9ml 129.63mM H₂O₂ and 0.1 ml of enzyme extract

Effect of temperature on PPO activity is shown in Figure (5 A and B). M-PPO displayed a maximum activity at 30 °C (Figure, 5-A). This result is in agreement with those obtained by Robison *et al.* (1993) and Rayan (2009). They noted that the optimum temperature of M-PPO was 30 °C using 4-methylcatechol as a substrate. The data also expedited that, the B:pe-PPO and B:pu-PPO had the same optimum temperature (30 °C). Yang *et al.* (2000) found that the optimum temperature of banana pulp PPO was at 30 °C. On the other hand, it can be seen that the maximum activity for T-PPO was achieved at 35 °C (Figure 5-B). Lokhandwala and Bora (2014) mentioned that the optimum temperature for T-PPO activity was at 40 °C and the activity obtained was 43.32 μmoles/ min/ml. For C-PPO, the optimum temperature was 40 °C. Millar *et al.* (1990) revealed that C-PPO had optimum temperature at 50 °C. Further, Figure (5-B) illustrates that G-PPO had maximum activity at 35 °C. Guo *et al.* (2009) reported that there are four isoforms of (PPOs) were characterized in purified extracts of coats (PPOIa and PPOIb) and pods (PPOIIa and PPOIIb) of green beans. The effects of temperatures between 0 and 80 °C

on the PPOs activities showed that optima temperatures for PPOIa, PPOIb and PPOII were 20, 30 and 50 °C, respectively. The O-PPO extract showed maximum activity at 35 °C (Figure 5-B). In a study carried out by Ortega-Garcia *et al.* (2008) olive fruits PPO showed optimum activity at 30 °C.

Figure (6, A and B) is showing the effect of temperature on LOX activity. As seen in Figure (6-A) M-LOX had an optimum temperature at 35 °C. Additionally, at the same Figure verify that LOX of B:pe and B:pu, exhibited their highest activities at 35 and 30 °C, respectively. However, from Figure (6-B) it can be detected that T-LOX, G-LOX and O-LOX possessed optimum temperature at 30 °C. But C-LOX the maximum activity was found at 15 °C. Matsui *et al.* (1999) observed that the optimum temperature for C-LOX was at 25 °C with free fatty acid (linoleic acid) as a substrate. Lorenzi *et al.* (2006) confirmed that the O-LOX was stable up to 60 °C and remained stable at -20 °C for more than 1 year. It was particularly thermostable in comparison to 13-LOX tomato isoforms (Anthon and Barrett, 2003).

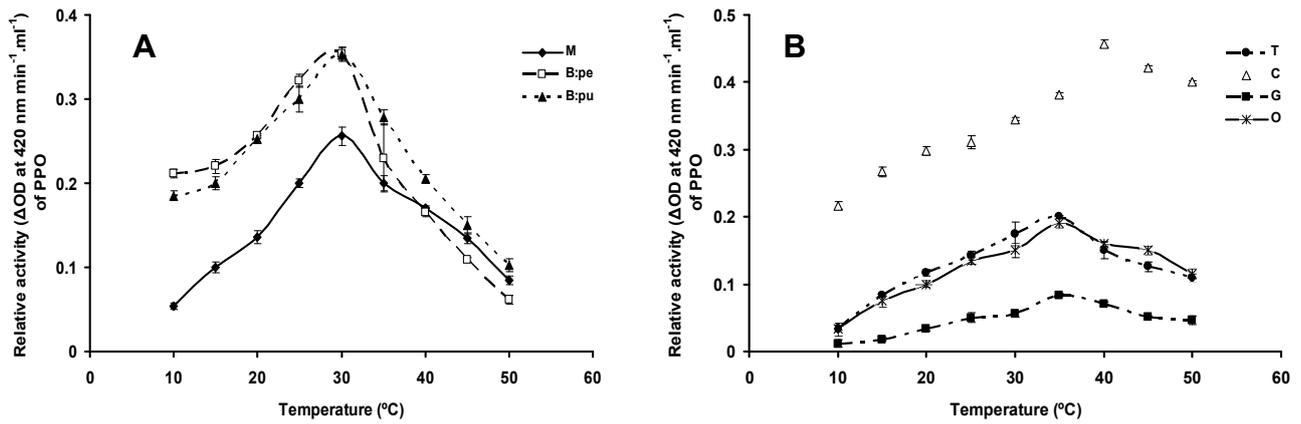


Figure (5): Effect of temperature on PPO activity of investigated samples at optimum pH value for each. The reaction medium containing 2.5ml 1.25mM catechol and 0.5 ml of enzyme extract

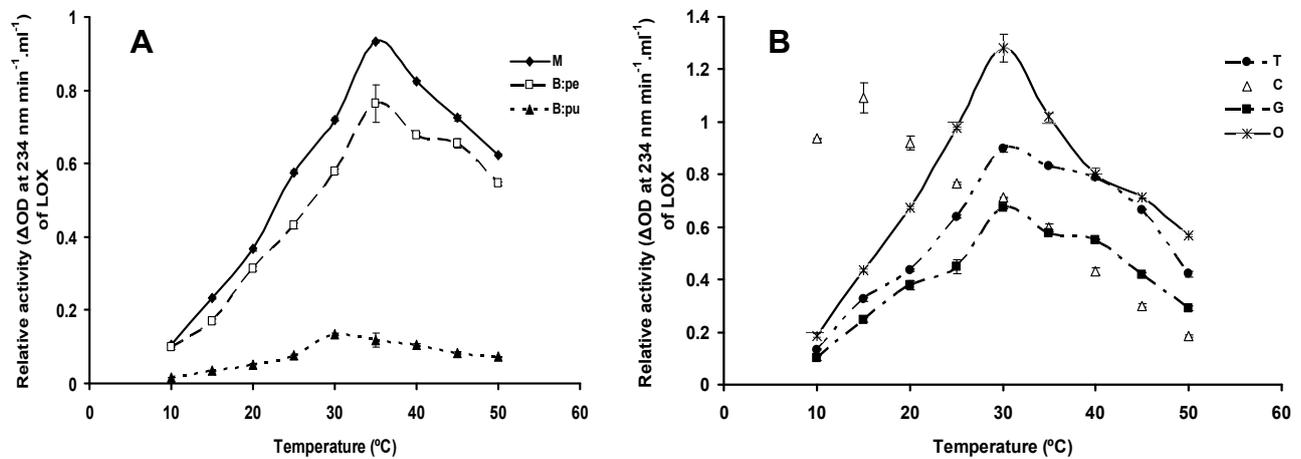


Figure (6): Effect of temperature on LOX activity of investigated samples at optimum pH value for each. The reaction medium containing 2.9ml 1.2mM linoleic acid and 0.1 ml of enzyme extract

Total initial activities of the POD, PPO and LOX enzymes

As seen in Table (1) the highest amount of POD was found in G extract (0.207 unit mg^{-1} protein of the sample). Similar result was found by Fernandes *et al.* (2007). Also, Regalado *et al.* (2004) reported that G-POD is considered to have an empirical relationship with deterioration in flavor, color, texture and nutritional qualities of raw and processed G. However, the other fruit and vegetable extracts were differed in POD activity (Table 1). The POD activity in M extract was greater than that for T or C and the lowest activity was for B:pe (0.087 unit mg^{-1} protein). Sathya (2014) found low amount of Bpe-POD due to that B:pe contains various antioxidant compounds which in turn reflects on the activity of POD.

The B:pu extract (Table 2) contained the greatest content of PPO (0.365 unit mg^{-1} protein) which causes undesirable browning in tissue as reported by Sojo *et al.* (1998). According to Yang *et al.* (2000), in the peel and pulp of B, dopamine was detected in large quantity, and it was strongly oxidized by the crude and/ or partially purified PPO. These results indicated that the enzymatic browning in B:pe or pu appears to be due to the

oxidation of dopamine by the endogenous PPO. While, G-PPO gave the lowest content (0.083 unit mg^{-1} of protein), it was contrast with Guo *et al.* (2009) who found that green beans pods had a high PPO content (4.5 unit mg^{-1} of protein).

With regard to LOX activity (Table 3), the mentioned extracts displayed reasonable activities fall between 0.068 - 0.249 unit mg^{-1} protein. Mango pulp had the highest amount of LOX (0.249 unit mg^{-1} protein) compared with the other fruits and vegetables extracts. Similar results were found by Rayan (2009) who found that the mango pulp had higher LOX activity than that for cauliflower heads. The high content of M-LOX is responsible for loss of yellow color in the M slices which undergo co-oxidation of carotenoid by reacting with hydroperoxides formed by lipoxygenase-mediated oxidation of polyunsaturated fatty acids. Chedea and Jisaka (2013) reported that LOX catalyzes the oxidation of polyunsaturated fatty acids. When LOX oxidizing unsaturated fatty acids co-oxidizes carotenoids many or all of their biological functions will be lose. The B:pu had the lowest activity (0.068 unit mg^{-1} protein) and this may be due to its high content from antioxidants as already mentioned.

Table (1): Summary of POD activities* in the investigated fruits and vegetables

Fruits and vegetables	POD activity**		
	Relative activity ($\Delta OD \text{ min}^{-1} \cdot \text{ml}^{-1}$)	Total activity (Unit 100 g ⁻¹)	Specific activity (unit mg ⁻¹ of protein)
Mango (M)	0.900	504.00	0.206
Banana (B):			
Peel (pe)	0.230	96.60	0.087
Pulp(pu)	0.300	137.00	0.126
Tomato (T)	0.854	515.50	0.176
Cucumber (C)	0.860	526.50	0.193
Green beans (G)	1.480	777.00	0.207
Olive (O)	0.250	126.85	0.089

* Means of three replicates

** All reactions have been carried out under the optimum conditions for the enzyme

- One unit of POD activity was defined as the change in OD value at 470 nm per minute per ml of enzyme

Table (2): Summary of PPO activities* in the investigated fruits and vegetables

Fruits and vegetables	PPO activity**		
	Relative activity ($\Delta OD \text{ min}^{-1} \cdot \text{ml}^{-1}$)	Total activity (Unit 100 g ⁻¹)	Specific activity (unit mg ⁻¹ of protein)
Mango (M)	0.300	47.25	0.210
Banana (B):			
Peel (pe)	0.345	39.35	0.138
Pulp(pu)	0.460	38.60	0.365
Tomato (T)	0.140	14.40	0.089
Cucumber (C)	0.420	57.37	0.317
Green beans (G)	0.100	13.80	0.083
Olive (O)	0.206	22.95	0.112

* Means of three replicates

** All reactions have been carried out under the optimum conditions for the enzyme

- One unit of PPO activity was defined as the change in OD value at 420 nm per minute per ml of enzyme

Table (3): Summary of LOX activities* in the investigated fruits and vegetables

Fruits and vegetables	LOX activity**		
	Relative activity ($\Delta OD \text{ min}^{-1} \cdot \text{ml}^{-1}$)	Total activity (Unit 100 g ⁻¹)	Specific activity (unit mg ⁻¹ of protein)
Mango (M)	1.146	7.161	0.249
Banana (B):			
Peel (pe)	0.744	3.013	0.177
Pulp(pu)	0.120	0.387	0.068
Tomato (T)	0.926	5.000	0.211
Cucumber (C)	1.140	6.327	0.228
Green beans (G)	0.664	3.237	0.132
Olive (O)	1.060	5.155	0.231

* Means of three replicates

** All reactions have been carried out under the optimum conditions for the enzyme

- One unit of LOX activity was defined as 0.1 increase of OD at 234 nm per ml of enzyme

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الخصائص والنشاطات الأولية الكلية لثلاث إنزيمات مؤكسدة تم فصلها من بعض الفاكهة والخضر النامية في جمهورية مصر العربية

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إن تواجد بقايا نشاط الانزيمات الداخلية المرتبطة بجودة الخضر والفاكهة سواء في المنتجات الخام أو المصنعة قد يسبب فقد في الجودة أثناء التصنيع والتخزين. ويختلف محتوى وخصائص هذه الانزيمات بدرجة كبيرة تبعاً لنوع و صنف المصدر وطبيعة البيئة التي نما فيها. استهدفت هذه الدراسة تقدير الظروف المثلى من درجة الأس الهيدروجيني ودرجة الحرارة وكذلك النشاطات الأولية الكلية لثلاثة من الأنزيمات المؤكسدة المتعلقة بالجودة وهي البيروكسيداز، البولي فينول أوكسيداز والليبوأوكسيجيناز في ثلاثة أنواع من الفاكهة هي المانجو صنف Zebda، الموز صنف Enana والزيتون صنف Picual وثلاثة أنواع من الخضروات هي الفاصوليا الخضراء صنف Little Marvel الطماطم صنف Marmand والخيار صنف Ria والتي تزرع في جمهورية مصر العربية. أوضحت النتائج وجود نشاط للأنزيمات الثلاثة في الفاكهة والخضر تحت الدراسة. اختلفت قيم الأس الهيدروجيني ودرجة الحرارة المثلى لأعلى نشاط لهذه الأنزيمات باختلاف نوع الفاكهة والخضر. حيث كان أعلى نشاط نوعي للبيروكسيداز وجد في مستخلص الفاصوليا الخضراء (0.207 وحدة لكل مجم بروتين) وأقل نشاط في مستخلص قشر الموز (0.87 وحدة لكل مجم بروتين). أحتوى مستخلص الفاصوليا الخضراء على أقل نشاط لأنزيم البولي فينول أوكسيداز في حين أن أعلى نشاط سجل في قشر الموز. وبالنسبة لنشاط أنزيم الليبوأوكسيجيناز فقد أحتوى مستخلص المانجو على أعلى نشاط نوعي (0.249 وحدة لكل مجم بروتين) عنه في كل من مستخلصي الخيار والطماطم (0.228 و 0.211 وحدة لكل مجم بروتين على الترتيب)، كما أن النشاط النوعي لهذا الأنزيم كان أقل ما يمكن في لب الموز (0.068 لكل مجم بروتين).