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Physicochemical and Antioxidant Potential of Garlic: Heat Processing Effects

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Abstract—*Heat processing method of fruits and vegetables has been developed over the centuries to make the final product more attractive in flavour, appearance, taste and consistency. Besides consumer preferences, the selected heat processing methods for food preparation is an important factor affecting not only the nutritional composition, but also the intake of bioactive compounds under normal dietary conditions. In this study garlic was evaluated for their, physicochemical, and antioxidant potential. The antioxidant content was significantly affected on application of different processing methods and processing time. Roasted sample exhibited higher % DPPH and total phenolic content as compared to the other heat processing methods. One of the roasting methods, microwave processing retained maximum ascorbic acid content, percent acidity and pH value. Hence roasting and microwave treatment may be recommended over other heat processing methods for better retention of bioactive compounds.*

Keywords*: Antioxidant potential, Garlic, Heat treatment, Physicochemical potential.*

1. INTRODUCTION

Antioxidant compounds in food are observed to possess health-protecting effects. Primary sources of natural antioxidants are whole grains, fruits and vegetables. Garlic (*Allium sativum*) has been used in world cuisines as well as in herbal medicine for thousands of years and has been claimed to prevent several diseases including cancer. Garlic (*Allium sativum*) is widely used in many parts of the world as vegetables as well as folk and modern medicine. In recent years garlic has received considerable attention in research for its pharmacological potential, which include lipid-lowering effects, anticancer activity, antioxidant activity, anti atherosclerotic activity, antimicrobial activity [1,2,3]. The cardiovascular effects of garlic are among the best investigated of all medicinal plants species [4]. Garlic contains minerals: calcium, copper, iron, manganese, phosphorus, potassium, selenium and Vitamins, A, B and C. Garlic contains various bioactive compounds including Allicin and its derivatives [5]. Garlic has also been proposed as one of the richest sources of total phenolic compounds among the generally consumed vegetables, and highly appreciated regarding its contribution of phenolic compounds in human diet.

Despite several therapeutic potential, raw garlic could not be consumed in large quantity due to its unpleasant taste and odour. Therefore, it is cooked as spice to make the final product more attractive in flavour, appearance, taste and consistency. Heat treatment, one of the most widely used post harvest processing method, is supposed to cause various physicochemical changes, including changes in flavor, colour and nutrient content. Heat treatment also leads to non-enzymatic browning reactions such as the Millard reaction, caramelization and chemical oxidation of phenols [6]. This study therefore has been planned to explore the effect of various heat processing methods as boiling, roasting, frying, steaming and microwaving on physico-chemical attributes and, consequently bioactive potential of garlic.

2. MATERIALS AND METHODS

2.1 Materials

Garlic was procured from the local market, cleaned removed extraneous materials and debris; edible bulbs were collected for further analysis. All standard chemicals were obtained from Sigma Chemicals (USA). Other chemicals, reagents, solvents used in this study were of analytical, extra pure grade.

3. METHODS

3.1 Heat processing methods

Five heat processing methods viz., boiling, steaming, frying, roasting and microwaving were selected for the study. These methods were proceeded for 5, 10, and 15 minutes respectively. All these experiments were performed in triplicate, each using 100 g of garlic sample.

3.2 Boiling

About 200 ml of distilled water was poured into a 500 ml beaker and put on the hot plate (Bezij 15090012008, India). As the water began to boil, the chopped garlic sample (100g) was added and boiled for 5, 10, 15 minutes respectively. After that sample was drained off and cooled [7].

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3.3 Roasting

100 g of crushed garlic sample was roasted in tray drier (Science tech. India) at 190º C for 5, 10, and 15 min. After roasting, the roasted garlic sample was collected and cooled [8].

3.4 Frying

100 ml of soybean oil was placed in a frying pan (30 cm in diameter) and heated on a hot plate for 1 min. 100 g of garlic sample was then placed in the pan, and the heating was reduced to medium and fried for 5, 10, and 15 min. Next, the fried garlic was drained off and cooled [9].

3.5 Steaming

100 g of crushed garlic sample was placed on a tray in a stainless steel steam cooker, which was covered with a lid, and steamed over 95ºC, for 5, 10, and 15 min under atmospheric pressure. After steaming, the garlic was drained for 1 min using a wire mesh strainer and then cooled [7].

3.6 Microwaving

Garlic (100g) was added to 200 ml of distilled water in a 500 ml beaker and then cooked in a domestic microwave oven (Whirlpool 021128620) for 5, 10, 15 min. The beaker was covered with watch glass to prevent water loss. The sample was drained off and cooled [10].

4. PHYSICOCHEMICAL ANALYSIS OF FRESH AND PROCESSED GARLIC

4.1 Determination of Moisture content

The moisture content of garlic was determined using standard method of AOAC [11]. Dishes used for the moisture determination were first dried at 105°C for 1 hour in drying oven. It was then transferred to the desiccators, cooled for 30 minutes, and weighed. The prepared samples were mixed thoroughly and about 5 g of the samples were transferred to the dried and weighed dishes. The dishes and their contents were placed in the drying oven and dried for 3 hrs at 105°C, and then the dishes and their contents were cooled in desiccators to room temperature and reweighed.

4.2 Determination of pH

10g garlic paste was homogenized with 50ml-distilled water in a mixer grinder. The ground sample was filtered and the pH was determined by dipping the combined glass electrode of a digital pH meter (Khera model, Indian make) into the filtrate.

4.3 Determination of percent acidity

Acidity of garlic was determined using the method of Ranganna [12]. To prepare the sample, 10 gm sample was boiled in 100 ml of distilled water for one hour, replacing the water lost by evaporation. It was then cooled, filtered and transferred to a volumetric flask and made up to 100 ml with distilled water. 10 ml of the aliquot was pipette out and titrated with 0.1N NaOH using few drops of phenolphthalein as indicator. The titre value was noted and percent acidity was calculated using the following equation:

$$\% \text{ Acidity} = \frac{\text{Titre x Normality of NaOH x Volume made up x Eq.wt. of Citric Acid x 100}}{\text{Volume of Sample taken for Estimation x wt. or volume of sample taken x 1000}} \times 100$$

4.4 Determination of color

Color determination was carried out using the Hunter *L*, *a*, *b* system with a colorimeter (Minolta CR-300). Both fresh and processed samples were analyzed. The assessments were carried out at room temperature (25ºC). *L** values indicate white to dark (lightness, black = 0, white = 100), *a** values green to red (redness > 0, greenness < 0), and *b** values blue to yellow (yellowness, b* > 0, blue < 0). The individual differences in L*,a*and b*values of each heat treatment with respect to the color of the fresh sample was evaluated using ΔE calculation [13]. The total color difference (Δ*E*) was calculated using the following equation:

$$\Delta E = (\Delta L^2 + \Delta a^2 + \Delta b^2)^{1/2}$$

5. ANALYSIS OF ANTIOXIDANT POTENTIAL OF FRESH AND PROCESSED GARLIC

5.1 Total polyphenol content

The total phenolic content of extract was determined using the Folin-Ciocalteu reagent according to modified method described by Singleton *et al.* [14]. Standard solution or extract (0.2 ml) was mixed with 2 ml of 2% Na_2CO_3 solution and 0.1 ml of 50% Folin-Ciocalteu reagent. After 30 min, the absorbance was read at 750 nm, and TPC was calculated from a calibration curve that was obtained using gallic acid as the standard. The results were expressed as milligrams of gallic acid equivalents per 100 g fresh weight. Extract was analyzed in triplicates.

5.2 DPPH radical scavenging activity

The free radical scavenging activity of the extract was measured using DPPH (1, 1- diphenyl 2-picryl hydrazyl) method of Shimada *et al.* [15] with slight modification. 10 mg of crushed garlic sample was mixed with 10 ml acidified methanol and heated at 40°C in water bath for 30 min. 100 µl of sample extract thus prepared was kept in a test tube and diluted with 2.9 ml of pure methanol. Sample was mixed with 150 µl of DPPH solution, incubated for 15 min. in dark and absorbance was measured in UV visible spectrophotometer at 515 nm. The % radical scavenging activity was calculated using following formula:

Control Absorbance-Sample Absorbance/Control Absorbance × 100

5.3 Determination of Ascorbic acid

Ascorbic acid content was estimated by standard A.O.A.C [11] method. 2g sample was crushed in Meta phosphoric acid solution and volume was made up to 50 ml in volumetric flask.15 g Meta phosphoric acid added in 40 ml acetic acid containing 200 ml distilled water. Standard ascorbic acid solution of 1mg/ml was made.

6. STATISTICAL ANALYSIS

The results were reported as mean ± standard deviation (SD) values. The significant differences among the means were determined with one way analysis of variance (ANOVA) using IBM SPSS statistics version 20 at a significance level of 0.05.

7. RESULTS

7.1 Effect of processing methods on moisture content

The moisture content of fresh and processed garlic was presented in Fig. 1. The moisture content of garlic was significantly ($p<0.05$) affected by boiling, steaming, frying, microwaving and roasting. The moisture content of fresh garlic was 65.76 mg/100g. It varied from 41.5 to 83.72 mg/100g (dwb).

Moisture content of fresh and processed garlic

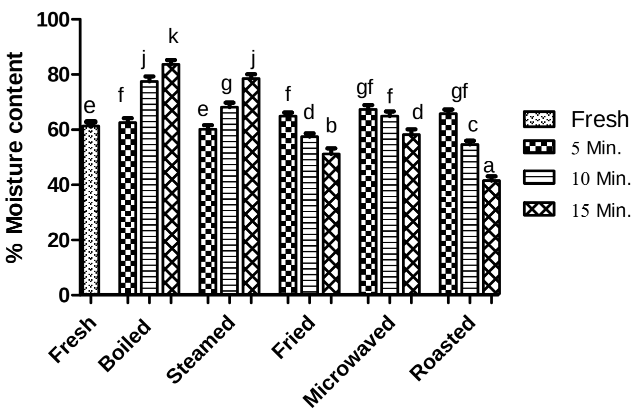

Fig. 1: Effect of processing methods and processing time on the moisture content of garlic. Mean values on the bars with different letters are significantly different at p<0.05.

7.2 Effect of processing methods on pH value

The garlic sample was treated with boiling, steaming, frying, roasting and microwave processing, and the pH value of the sample was examined. Changes in pH at various temperatures are shown in Fig. 2. The pH of fresh garlic was 6.54, whereas after heat treatments boiling, steaming, frying, microwave and roasting, the pH value of the garlic sample was 5.26, 5.13, 4.89, 4.99 and 4.49 respectively.

pH Value of fresh and processed garlic

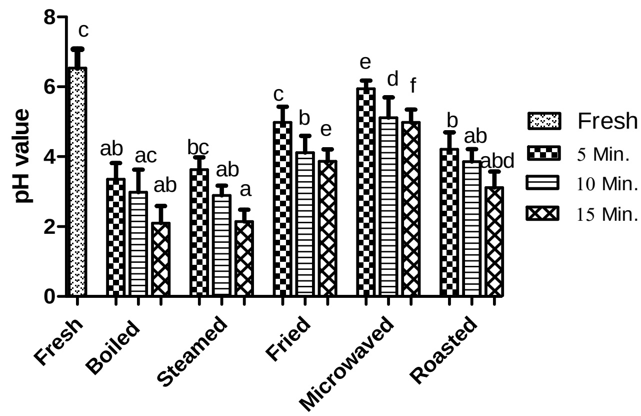

Fig. 2: Effect of processing methods and processing time on the pH value of garlic. Mean values on the bars with different letters are significantly different at p<0.05.

7.3 Effect of processing methods on percent acidity

In investigating the effects of heat treatment on the percent acidity of garlic, it was revealed that the percent acidity decreased as the processing time increased. In fresh garlic, percent acidity was 1.94 %. For boiling, steaming, frying, roasting, and microwaving, the percent acidity decreased by 0.12～0.37, 0.31～0.38, 1.21～1.32, 1.45～1.62 and 1.75～1.86 % respectively Fig. 3.

% Acidity of fresh and processed garlic

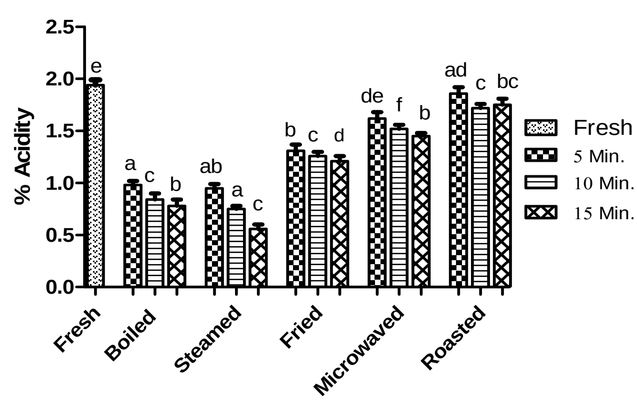

Fig. 3: Effect of processing methods and processing time on the percent acidity of garlic. Mean values on the bars with different letters are significantly different at p<0.05.

7.4 Effect of processing methods on color value

The Hunter color L*, a* and b* values of fresh garlic were 62.84, -1.97 and 15.75, respectively. Effect of the processing methods and time on the Hunter color value is shown in Table 1. Lower L* value in processed garlic reveals that after various type of processing the brightness of sample was less as compared to fresh samples. With the increase in the processing time the L* value decreased further. a* value of samples increased after processing which indicates that the sample redness increased after processing. b* value increased after roasting, frying and microwave treatment.

Table 1: Colour value of the fresh and processed garlic during different processing method and processing time.

Heat Treatments		L*	a*	b*	ΔE
Fresh		62.84 ±1.61^{de}	-1.97± 0.53^{bc}	15.75±0.35^{c}	-
Boiling	5 min	60.35±1.46^{d}	-2.37±0.09^{d}	11.05±1.24^{b}	5.33^{a}
	10 min	58.21±1.28^{cd}	-2.29±0.06^{a}	10.25±1.41^{b}	7.19^{b}
	15 min	57.05±1.83^{bcd}	-2.20±0.13^{a}	9.64±0.10^{ab}	8.42^{cd}
Steaming	5 min	54.42±1.34^{ab}	-1.58±0.02^{ad}	10.35±0.04^{b}	10.01^{efg}
	10 min	48.41±1.42^{a}	-1.98±0.52^{ab}	9.35±0.05^{ab}	15.78^{i}
	15 min	43.41±1.54^{abc}	-2.86±0.68^{a}	8.35±0.05^{d}	20.81^{hi}
Frying	5 min	50.14±1.46^{abc}	2.62±0.06^{cd}	20.08±0.34^{a}	14.18^{j}
	10min	49.56±1.35^{ab}	3.45±0.06^{def}	19.25±1.41^{d}	14.76^{hi}
	15 min	48.2±0.36^{de}	4.47±0.47^{fg}	18.82±0.89^{d}	16.28^{i}
Microwaving	5 min	61.29±0.35^{e}	1.15±0.89^{cd}	24.88±0.45^{a}	9.77^{fe}
	10 min	59.28±0.29^{cd}	1.26±0.69^{de}	22.24±1.25^{efg}	7.83^{cd}
	15 min	58.29±0.43^{d}	1.35±0.58^{defg}	20.45±1.36^{d}	7.33^{c}
Roasting	5 min	60.29±0.38^{b}	1.78±0.05^{cde}	26.46±1.42^{g}	11.63^{g}
	10 min	59.19±0.25^{cd}	1.88±0.05^{efg}	24.65±1.34^{f}	10.37^{fg}
	15 min	57.66±0.85^{cd}	1.95±0.72^{g}	22.5±0.12^{e}	9.36^{def}

Values are expressed as mean ± standard deviation of triplicate experiment. Mean values in a column with different letters are significantly different at $p< 0.05$.

7.5 Effect of processing methods on total phenolic content

The Total Phenolic Content in fresh and processed garlic, expressed as milligrams of gallic acid equivalent per 100 g of fresh weight, is presented in Fig. 4. The TPC in fresh garlic was 78.45 mg/100 g. For boiling, steaming, frying, microwaving, and roasting, TPC decreased by 34.18～52.87, 59.12～62.59, 69.84～72.43, 71.25～75.25 and 74.39～76.67 mg/100 g respectively.

Total Phenolic Content of fresh and processed garlic

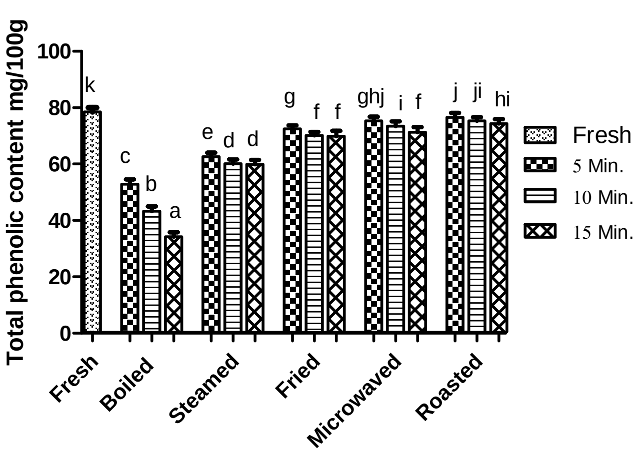

Fig. 4: Effect of processing methods and processing time on the Total Phenolic Content of garlic. Mean values on the bars with different letters are significantly different at p<0.05.

7.6 Effect of processing methods on DPPH radical scavenging activity

The DPPH radical scavenging activity of fresh and processed garlic, expressed as milligrams of AA equivalent per 100 g of fresh weight, is presented in Fig. 5. The DPPH radical scavenging activity of garlic extract was significantly reduced ($p<0.05$) after processing.

DPPH Radical Scavenging Activity of fresh and processed garlic

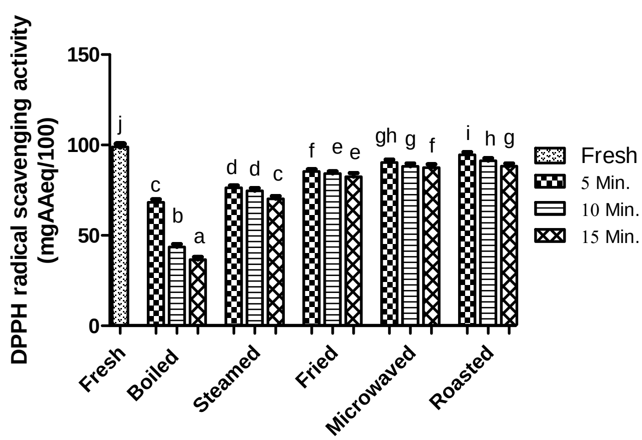

Fig. 5: Effect of processing methods and processing time on the DPPH radical scavenging activity of garlic. Mean values on the bars with different letters are significantly different at p<0.05.

The DPPH radical scavenging activity of fresh garlic was 98.92 mg AA eq/100 g and after processing, decreased by 36.55～68.29, 70.15～76.30, 82.45～85.35, 87.48～90.36 and 88.28～94.56 mg AA eq/100 g for boiling, steaming, frying, microwaving and roasting respectively.

7.7 Effect of processing methods on ascorbic acid content

The Ascorbic Acid levels of fresh and processed garlic are presented in Fig.6. The Ascorbic acid content of fresh garlic was 27.53 mg/100 g. All processing methods lead to a significant loss ($p<0.05$) in the amount of Ascorbic acid as compared to fresh garlic. As processing time increased, the Ascorbic acid content of boiled, steamed, fried, microwave and roasted garlic decreased, with a range of 10.40～12.26, 15.68～18.51, 19.48～22.64, 21.46～23.48 and 17.54～20.24 mg/100 g, respectively. For boiling, steaming, frying, microwaving, and roasting, the mean Ascorbic acid content lost after for 15 min were 55.4%, 32.7%, 26.4, 17.7%, and 14.7% respectively. The result of the effect of five different heat-processing methods indicated that the highest reduction was noted after boiling, followed by steaming, roasting, frying and microwaving.

Ascorbic acid content of fresh and processed garlic

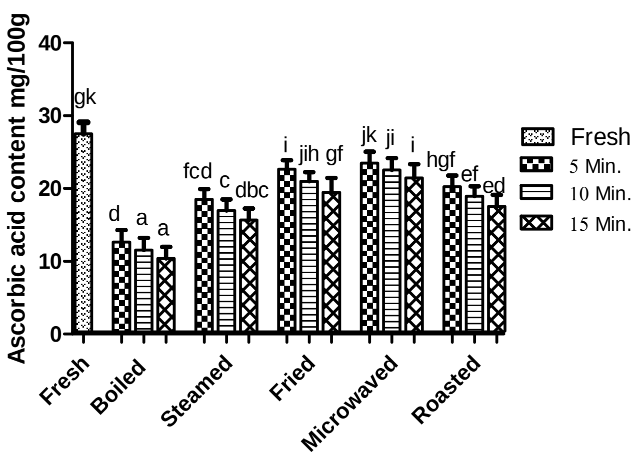

Fig. 6: Effect of processing methods and processing time on the ascorbic acid content of garlic. Mean values on the bars with different letters are significantly different at p<0.05.

8. DISCUSSION

Changes eminent in the moisture content of the garlic is due to the application of various heat treatments. During the dry heat treatment process (roasting, frying and microwaving), lower moisture contents were at higher temperatures [16] whereas during moist heat treatment process i.e. boiling and steaming moisture content were increased continuously over time. Boiling has higher moisture content due to the absorption of water during processing.

It was found that the pH value of five minutes microwave treated sample decreased by lesser amounts than that of other heat-treated samples. A significantly decrease ($p<0.05$) in pH value was observed with the progression of heat treatment process. Thereafter, the pH decreased gradually with increased temperature. Similar results were also observed in the red ginseng manufacturing process, and the decrease in pH progressed as the heating temperature increased [17].The pH decrease in the heated garlic sample was, in part associated with the production of browning materials during heat treatment. The formation of carboxylic acids (which are produced by the oxidation of aldehyde group in aldohexose, acidic compounds and decrease in basic amino acid by combing with sugar) has been reported responsible for the decrease in pH, in browning reaction and during heat treatment.

All the five heat processing methods i.e. boiling, steaming frying, microwaving and roasting significantly ($p<0.05$) affected the percent acidity in garlic compared with fresh garlic. The highest reduction was observed after boiling (80.9～93.8%), followed by steaming (80.8～84.0%), frying (31.9～37.6%), microwaving, (5.6～9.7%) and roasting (16.4～25.2%), whereas maximum retention was observed in microwaving (5.6～9.7%). Similarly the mean acidity of boiled vegetables was found to be decreased with increased boiling time [18].

The change in color calculated from color parameters 'L' 'a' 'b' of fresh and processed samples in the range of 0.10 to 1.35 reveals, an imperceptible color difference.

Dry heat processing methods i.e. frying, microwaving and roasting did not significantly ($p<0.05$) affect the total phenolic content in processed garlic when compared to fresh garlic, whereas boiling and steaming significantly decreased ($p<0.05$) total phenolic content. The highest reduction was observed after boiling (32.60～56.4%), followed by steaming (20.2～24.6%), frying (7.6～10.9%), microwaving, (9.1～4.0%) and roasting (5.1 ～2.3%). Similar kind of result was obtained by Chuah *et al.* [19] and found that boiling significantly reduced the total phenolic content in colored peppers, while fry and microwave cooking had no impact on total phenolic content. Zhang [10] and Ismail *et al.* [20] reported that cooking vegetables significantly decreases, total phenolic content. Reduced phenolic compounds in boiled or steamed foods have been attributed to the dissolution of phenolic compounds into the cooking water [21]. The loss of phenolic compounds also depend on the processing time and food size [22]. In contrast, other studies found that cooking increased Total phenolic contents in some vegetables due to the disruption of cell walls, which liberated soluble phenolic compounds from insoluble ester bonds [23,24].

The DPPH radical scavenging activity was reduced by 30.9～63.0% of its initial capacities after boiling, followed by steaming (22.8～29.0%), frying (13.7～16.6%), microwaving (8.6～11.5%) and roasting (4.4～10.7%).

The results show that moist-heat processing methods i.e. boiling and steaming result in high losses of ascorbic acid, while dry heat processing methods i.e. frying, roasting and microwave treated sample revealed only small losses. Various studies have shown that cooking reduces Ascorbic acid content in fruits and vegetables; therefore, as a commonly perceived idea, Ascorbic acid is destroyed during heat processing because it is unstable at high temperature. Chuah *et al.* [25] reported that the Ascorbic acid levels in peppers decreased during cooking procedures such as boiling, microwave cooking, and stir-frying. Significant reductions were documented for boiling particularly due to the diffusion of Ascorbic acid into cooking water. Somsub *et al.,* [26]**,** and Leskova *et al.* [27] reported higher Ascorbic acid retention values in foods processed by stir frying, roasting and microwave cooking than in those processed by boiling or blanching. Various studies report that cooking reduces Ascorbic acid content in food, including sweet chestnuts, potatoes, tropical leafy vegetables, selected Thai vegetables, broccoli and sweet peppers [28,29]. The amount of cooking related loss of Ascorbic acid depends on several factors, including cooking method, heating temperature, cooking time, enzymatic oxidation during preparation, and surface area exposed to water and oxygen.

9. CONCLUSION

It has been observed that change in physiochemical and antioxidant potential depends on type of heat processing methods used. Antioxidant potential was significantly affected on the application of different processing methods and time. Roasted samples exhibited higher total phenolic content and radical scavenging activity as compared to the other heat processing treatments. Ascorbic acid content, percent acidity and pH value retained better in microwave processed sample. Therefore, mild roasting and microwave treatment may be considered better and healthy processing treatment over others.

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