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Effect of Papaya Wastes on Quality Characteristics of Meat Burger

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Abstract

Large amounts of fruit byproducts are discarded daily by the food industry, representing a significant loss of nutrients. The present study compared between the bioactive compounds in papaya wastes, including peels and seeds. Furthermore, this study evaluated the effect of papaya (*Carica papaya*) peels powder (PP) addition on technological and microbial characteristics of beef burgers. Four formulas of beef burgers were prepared as follows: C (control beef burger), T1 (beef burger with 1% PP), T2 (beef burger with 2% PP), and T3 (beef burger with 3% PP). The results indicated that PP had greater contents of total phenolics (838 mg GAE/100g sample) and flavonoids (270.66 mg Quercetin/ 100g sample), as well as higher antioxidant activity than papaya seeds powder (SP). The incorporation of PP into beef burgers resulted in burgers with lower color L* and a* values, while b* values increased. Besides, a significant increase in crude fiber contents of beef burger samples was found with the addition of PP, as compared to control. Furthermore, the addition of PP enhanced physical quality (cooking loss, cooking yield and shrinkage) and thiobarbituric acid reactive substance (TBARS) values during storage. The obtained results suggest that PP can be utilized as a functional ingredient in meat products because of its high bioactive compounds content.

Keywords: Papaya wastes, Antioxidant activity, GC-MS, Antimicrobial, Beef burger.

1. Introduction

Substantial amounts of food wastes were dumped into the environment without processing. This is not only a threat to the environment, but also a wasted opportunity to reuse these byproducts and convert them into value-added materials for different industries. The reuse of these wastes will support the agricultural sector economically and will also be beneficial to the environment (**Laufenberg et al., 2003).** In this concept, several studies have explored the potential effects of using fruit byproducts on food technology **(Almeida et al., 2015).**

Recently, consumer awareness about health, encouraged the meat industry to produce healthier meat products to enhance the nutritional quality. Furthermore, synthetic additives were considered unhealthy in consumers opinions, while using plant-based natural additives in processed meat products considered as demand (**Martín-Mateos et al., 2022**).

Carica papaya belongs to the family *Caricaceae* and is widely cultivated in tropical regions (**Joymak et al., 2021**). *Carica papaya* is a nutraceutical plant, with a delicious fruit taste and healthy, besides the whole plant parts (roots, bark, peel, seeds, and pulp) have medicinal properties with a wide range of pharmacological activities and a rich source of powerful antioxidants **(Aravind et al., 2013**). While papayas world production (*Carica papaya L.*) in 2020 was 13894705 tones (**FAOSTAT, 2022**), papaya byproducts make up about 20-25% of the weight of the fruits, and consist of peels and seeds **(Pavithra et al., 2017).**

It was found that papaya peels are rich in fiber, protein, carbohydrates, ash, fat, and minerals (phosphorous and potassium), and it is also a source of antioxidants (**Jamal et al., 2017).** The large amount of fiber in papaya peels increased the opportunity to development new products (**Calvache et al., 2016**). Although papaya seeds have a very pungent taste, which makes them almost unpalatable, it has a strong medicinal value and antibacterial properties (**Aravind *et al.*,2013**). In this concept, several studies used papaya peels and seeds in various processed products (**Joymak et al., (2021); Azevedo and Campagnol, (2014**)).

However, there is a lack of information about influence of papaya wastes powder on quality properties of meat products during storage. Thus, this study was conducted to evaluate the effect of papaya PP addition on physicochemical, microbiological, and sensory properties of beef burgers during storage.

2. Materials and Methods

2.1. Materials

Boneless grounded beef meat was purchased from a local butcher shop at Assiut city, Egypt in the day before the experiment was done and stored in a refrigerator at 5±1°C overnight. The papaya fruits (*Carica papaya* L) were obtained from Agriculture Research Center, Giza, Egypt. Other ingredients of beef burger (spices, white and black pepper, onion powder, garlic powder and salt) were obtained from the local market, Assiut. Egypt. Furthermore, all chemicals used in this study were purchased from EL-Gomhouria for Trading Chemicals and Drugs Co., Assiut city, Egypt, while 2, 2-diphenyl-1-picrylhydrazyl (DPPH), Folin Ciocalteu's phenol reagent, and Gallic acid obtained from Sigma-Aldrich Chemie Gmbh Munich, Germany.

2.2. Methods

2.2.1. Preparation of papaya wastes powder

Ripe papaya fruits (*Carica papaya* L) were selected. Papaya fruits were washed and then manually peeled; the peels were cut using a stainless-steel knife and the seeds were removed. Peels and seeds were rinsed with water to remove any adhering mucilage, and then they were blanched at 100 ºC for 3-4 min. The peels and seeds were spread uniformly on a 40 x 28 cm stainless-steel trays and dried at 50 ˚C for 16-18 h using an electric air oven until constant weight. Then, the dried peels and seeds were ground using coffee mill and the powder (Fig.1) were kept separately in polyethylene bags and stored at -20˚C until further analysis.

2.2.2. Preparation of beef burger samples

Beef burgers were prepared according to **Shokry (2016)** using the ingredients listed in Table 1. A sample of beef burgers without addition of papaya PP was served as a control. The other three samples of beef burgers were prepared in the same manner with the addition of papaya PP at concentrations of 1, 2 and 3% as a partial replacement of meat. From the meat mixture, beef burgers weighing approximately fifty grams were shaped using a machine for making burger patties. Beef burgers were put in polyethylene bags and stored at -18 °C for 3 months. Fresh and stored samples were subjected to physical, chemical, and microbiological analysis.

2.2.3. Physical characteristics

2.2.3.1. Color measurement

Samples were analyzed at Cairo University Research Park (CURP)/ Faculty of Agriculture. Color was measured by chroma meter (Konica Minolta, model CR 410, Japan) calibrated with a white plate and light trap supplied by the manufacturer. Color was expressed using the CIE L*, a*, and b* color system (**CIE, 1976**). A total of three spectral readings were taken for each sample. Lightness (L*) (dark (0) to light (100)), the redness (a*) values (+) reddish to (-) greenish) and the yellowness (b*) values ((+) yellowish to (-) bluish) were estimated. The total color difference (ΔE) was calculated from the Hunter L*, a* and b* values according to equation: $\Delta E = (L^2+a^2+b^2)^{0.5}$. The chroma value indicates color intensity or saturation, and it is equal $(a^2+b^2)^{0.5}$. Hue angle was calculated as H° and is equal $\tan^{-1}$ (b*/a*).

2.2.3.2. Cooking loss

Cooking loss values were determined according to the method of **Piñero et al. (2008)** by using the following equation:

$$\text{Cooking loss (g/ 100g)} = \frac{Wr - Wc}{Wr} \times 100$$

Where: Wr: the weight of raw sausage (g); Wc: the weight of cooked sausage (g).

2.2.3.3. Cooking yield

Beef burger cooking yield values were measured by subtracting cooking loss from 100 according to **El-Nemr, (1979).**

2.2.3.4. Shrinkage

The percentage of shrinkage was calculated as described by **Serdaroğlu and Değırmencioğlu (2004)** by using the following equation:

$$\%\text{Shrinkage} = \frac{(\text{Raw thickness} - \text{Cooked thickness}) + (\text{Raw diameter} - \text{Cooked diameter})}{\text{Raw thickness} \text{ þ } \text{Raw diameter}} \times 100$$

2.2.3.5. pH measurement

pH values were measured with a standard combined electrode attached to a digital pH meter (Hanna Instruments, Padova, Italy) as described by **Vareltzis *et al.* (1997).**

2.2.4. Analytical methods

2.2.4.1. Proximate composition analysis

Moisture, protein, crude fat, crude fibers, and ash contents were determined according to the methods described in the **AOAC (2000).** Total carbohydrates were calculated by difference according to **Aly et al. (2021),** meanwhile caloric value was calculated according to **Livesy (1995).**

2.2.4.2. Determination of minerals content

ICAP6200 Inductively Coupled Plasma Emission Spectrometry (ICP-OES) was used in the determination of calcium, magnesium, iron, cupper, and zinc contents (**Isaac and Johnson, 1985**). Sodium and potassium contents were determined by a flame photometer corning 400 (**Chapman and Pratt, 1961**), whereas phosphorus content was determined by spectrophotometer (**Jackson, 1967**).

2.2.4.3. Determination of the total phenolics, flavonoids content, and antioxidant activity assay

- Determination of the total phenolics content: the total phenolics content was determined using Folin Ciocalteu reagent according to **Singleton and Rossi (1965).** Sample extracts were prepared using 80% methanol containing 1% hydrochloric acid. Extracts were mixed with diluted Folin Ciocalteu reagent (10-fold with distilled water) and allowed to stand at 22 °C for 5 min; after that, sodium bicarbonate (60 g/L) solution was added to the mixture. Absorbance was measured at 725 nm using a spectrophotometer (67 Series, Jenway, UK, USA).

- Determination of the total flavonoids content: The method described by **Jia et al. (1999)** was used to determine total flavonoids content**,** with slight modifications. The sample extract (prepared for total polyphenols content) was mixed with distilled water (2 mL) and subsequently with 5% NaNO2 (0.15 ml). After 6 min, 10% AlCl3 (0.15 ml) was added and allowed to stand further 6 min. after that, 4% NaOH (2 ml) was added to the mixture. Then, the final volume was made to be 5 mL by using distilled water. After 15 min the absorbance was determined by spectrophotometer (67 Series, Jenway, UK, USA) against a blank at 510 nm., and results were expressed as mg of quercetin equivalent (QE) /100 g of dry sample weight.
- Determination of antioxidant activity: The antioxidant activity was determined using the stable radical DPPH **Brand-Williams et al., (1995)**. Samples extracts were prepared by maceration in 70 % methanol for 24 h at the room temperature. The mixture was filtered, and the supernatant was used for antioxidant activity determination. A known volume of methanolic extract was dissolved in 70 % methanol then added to 335 µl DPPH (9.85 mg in 100 ml 70 % methanol). Vortex (2500 rpm) were used to shake the mixtures for 1 min and then placed in the dark at room temperature for 30 min. Absorbance at 517 nm was estimated with a spectrophotometer (JENWAY 6315). Control absorbance (without adding sample) was determined. The following formula was used to calculate DPPH radical inhibition percentage. I % = [(AB - AS)/AB] x 100

where I% is the DPPH radical inhibition %; AB is the control sample absorbance AS is the absorbance of tested sample at the end of reaction. Each assay was carried out in triplicate.

2.2.4.4. Identification of papaya wastes extracts components by Gas chromatography and mass spectrometry (GC-MS)

The GC-MS analysis of samples was performed at the Analytical Chemistry Department, Faculty of Science, Assiut University, to determine the volatile and semi-volatile chemicals compounds. Papaya PP and SP extracts were made with methanol. The GC-MS analysis was performed with a Thermo Scientific TM TRACE 1300 coupled to an ISQ-7000 and a Thermo Scientific TM TG-6MB 5 ms (30 mx0.250 mmx1.00 m) column. The temperature in the GC oven was kept at 100 °C for 15 min, then increased to 150 °C at a rate of 10 °C/min, and then to 200 °C at a rate of 5 °C/min. The temperature was raised to 250 °C at a rate of 5 °C per min, and then to 280 °C at a rate of 5 °C per min. Helium gas was used as the carrier, with a flow rate of 0.5 mL/min. The mass spectrometer was set to electron ionization mode, with a temperature of 320 °C for the ion source and 280 °C for the MS transfer line. The NIST 17 mass spectrum library (mainlib) was used to identify volatiles, and the results are represented as a percentage of the total GC area.

2.2.4.5. Determination of lipid oxidation:

Lipid oxidation in fresh beef burger samples and at the end of storage time was estimated through the determination of the thiobarbituric acid reactive substance (TBARS) according to the method of **Lemon (1975)** to evaluate efficiency of PP as a natural antioxidant.

2.2.5. **Microbiological analysis**

2.2.5.1. Antimicrobial activity

The antimicrobial activity of papaya PP and SP methanolic extracts was tested against six bacterial strains (*Bacillus cereus* AUMC No. B-52, *Escherichia coli* AUMC No. B-53, *Micrococcus luteus* AUMC No. B-112, *Pseudomonas aeruginosa* AUMC No. B-73, *Serratia marcescens* AUMC No. B-55, and *Staphylococcus aureus* AUMC No. B-54), and two fungal strains (*Fusarium oxysporum* AUMC No. 215 and *Geotrichum* *candidum* AUMC No. 226), which were obtained from Mycological Center, Faculty of Science, Assiut University, Egypt. Bacterial strains were individually inoculated in nutrient broth medium for 48 h to prepare inoculum, while fungi were grown for 7 days in Sabouraud's dextrose broth medium. Nutrient agar and Sabouraud's dextrose agar media were respectively used for bacteria and fungi in bioassay (**Johnson and Case, 2018**). After media solidification, sterile cork borer was used to cut 5 mm diameter cavities in the solidified agar (4 cavities per plate). Papaya wastes methanolic extracts were pipetted in the cavities (50 µL per cavity). Plates were incubated at 28°C for 48 h and diameter of inhibition zone around cavities were measured in mm (**Kwon-Chung and Bennett, 1992**). This was done on two replicates.

2.2.5.2. Microbiological counts:

Total bacteria count (TBC) was enumerated according to the method of **Suleiman et al. (2011)** on a nutrient agar medium after incubation at 37 ˚C for 48 hr. Mycological analysis was performed by dilution plate method (**Pitt and Hocking 2009**), and fungal count were enumerated on czapek's agar medium. Two replicates at each time point were analyzed for microbiological analyses.

2.2.6. Sensory evaluation

Sensory evaluation of samples was conducted using 10 semi-trained panelists who were asked to score color, texture, odor, and taste. Hedonic ranking test where 9 (extremely accepted) to 1 (extremely rejected) (**Gelman and Benjamin, 1989**)

2.2.7. Statistical analysis

Analysis of variance and significant differences among means were tested using SPSS software (version 16.0 for Windows, SPSS Inc., Chicago, IL). Analysis of Variance (ANOVA) was completed using Duncan's multiple comparison for mean difference testing **(Steel and Torrie, 1980).**

3. Results

3.1. Phytochemicals in papaya wastes

3.1.1. The content of total phenolics, flavonoids, and antioxidant activity

Data in Fig. 2 revealed that the total phenolic recorded 838 and 367.66 mg GAE/ 100 g sample in peels and SP, respectively. Whereas total flavonoids recorded 270.66 and 137 mg QE/ 100g in peels and seeds, respectively. Furthermore, Fig.2 reveals that the antioxidant activity of peels and seeds extracts were 63.2 and 60.37%, respectively.

3.1.2. Gas chromatography and mass spectrometry (GC-MS) analysis:

Gas chromatography and mass spectrometry analysis (GC-MS) of compounds in methanolic extracts of PP and SP was carried out, and the results are shown in Table 2 and Table 3. Thirty-two compounds were detected in PP while SP showed twenty four compounds. Several compounds found in both PP and SP methanolic extracts like Acetic acid,7- (4- hydroxy- 5- methoxy- 1,5-dimethylhexyl- (4,4,10,13,14-pentamethy l2,3,4,5,6,7,10,11,12,13,14,15,16,17tetradeca hydrocyclopenta [a] phenanthryl ester (recorded the highest percent (18.59%) in PP sample), 9-Octadecenoic acid (Z)-,3-[(1-oxohexadecyl)oxy]-2-[(1-oxooctadecyl)oxy]propyl ester, Octadecanoic acid,2-[(1-oxohexadecyl)oxy]-1-[[(1oxohexadecyl)oxy]methyl]ethyl ester, Hexadecanoic acid,1-[[[(2aminoethoxy)hydroxyphosphinyl]oxy]methyl]-1,2-ethanediyl ester and Eicosanoic acid,2-[(1oxohexadecyl)oxy]-1-[[(1-oxohexadecyl)oxy]methyl]ethyl ester; which all higher in papaya peels than papaya seeds.

Meanwhile, other compounds recorded higher values in SP than PP like Docosanoic acid, 1,2,3-propanetriyl ester, 5-Chloro-6beta-nitro-5alphacholestan-3-one, Hexadecanoic acid,1(hydroxymethyl)-1,2-ethanediylester, Octadecanoic acid,3-[(1-oxohexadecyl)oxy]-2-[(1-oxotetradecyl)oxy]propyl ester, Ethyl iso-allocholate, 5H-Cyclopropa[3,4]benz[1,2-e]azulen-5-one,3,9,9atris(acetyloxy)-3-[(acetyloxy)methyl]-2-chloro-1,1a,1b,2,3,4,4a,7a,7b,8,9,9a-dodecahydro-4a,7b-dihydroxy-1,16,8tetramethyl-1,αR1aα,1bα,2α,3α,4aα,7aα,7bα,8α,9α,9aα)]-, 7,8-Epoxylanostan-11-ol, 3-acetoxy, 3,5,9-Trioxa-5-phosphaheptacos-18-en-1-

aminium,4-hydroxy-N,N,N-trimethyl-10-oxo-7]-(1-oxo-9-octadecenyl)oxy-[,hydroxide, inner salt, 4-oxide, (R)- (recorded the highest percent (12.244%) in SP sample), and Oleic acid, eicosyl ester.

On the other hand, papaya PP contained different compounds, (which did not appeared in SP sample) like Acetic acid,6-morpholin-4-yl-9-oxobicyclo[3.3.1]non-3-yl este, 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl, 2-Furancarboxaldehyde5-(hydroxymethyl)-, Tetraacetyl-d-xylonic nitrile, 2-Myristynoyl pantetheine, α-D-Glucopyranoside, O-α-D-glucopyranosyl-(1.fwdarw.3)-α-D-fructofuranosyl, 9,10-Secocholesta-5,7,10-(19)triene-3, 24,25-triol, (3α,5Z,7E), and 3,5,9-Trioxa-4-phosphapentacosan- 1-aminium,4- hydroxy- N,N,N- trimethyl- 10-oxo-7]- (1-oxohexadecyl)oxy]-, hydroxide,inner salt, 4-oxide (which recorded the highest percent between these different compounds (3.857%)).

Furthermore, papaya SP showed new compounds when compared with PP sample, like 2,4,6-Decatrienoic acid,1a,2,5,5a,6,9,10,10a-octahydro-5,5a-dihydroxy-4 (hydroxymethyl)- 1,1,7,9-tetramethyl- 11- oxo- 1H 2,8 amethanocyclopenta [a] cyclopropa[e]cyclodecen-6 ylester,1]αR(1aα,2α,5α,5aα,6α,8aα,9α,10aα)-], 4-Piperidineacetic acid,1-acetyl-5-ethyl-2-[3-(2-hydroxyethyl(-1H-indol-2-yl]-α-methyl-, methyl ester (which recorded the highest percent between these different compounds (8.328%)), 9-Octadecenoic acid (Z),-tetradecyl ester, 3'H-Cycloprop(1,2)-5α-cholest-1-en-3-one, 1',1'-dicarboethoxy-1α,2α-dihydro. Moreover, it could be seen from the data in our study that *C. papaya* seed extract was dominated by esters.

3.2. Antimicrobial activity

The antimicrobial potentials of PP and SP methanolic extracts (50 µl) against eight pathogens (6 bacterial and 2 fungal strains) were assessed in terms of zone inhibition of microorganism's growth, and the results are illustrated in Table 4. PP recorded the same zones against all bacterial strains (8mm), while it had negative results against fungal strains under investigation. On the other hand, SP extract recorded the maximum zone formation against *Bacillus cereus, Pseudomonas aeruginosa and Serratia marcescens* (9mm), while SP extract recorded 10mm against fungal strains under investigation.

Indeed, the data in Fig.2 and Table 2 encouraged us to use papaya PP in our study, due to its higher content of phenolics and flavonoids, as well as antioxidant activity, when compared to SP.

3.3. Papaya peels analysis

3.3.1. Chemical composition of papaya PP

Proximate composition of PP is presented in Fig. 3. The fiber content of PP was 11.02%, while protein, ash, crude fat, and carbohydrates content were 12.47%, 10.98%, 6.5 and 59.025% (on dry weight basis), respectively. On the other hand, the caloric value was calculated in this study and recorded 344.48 kcal/100g.

3.3.2. Minerals composition

Fig. 4 shows the minerals composition of papaya PP, and the most abundant mineral was potassium (3183.57mg/ 100g), followed by phosphorus (732.34mg/ 100g). Moreover, the intake of 100 g papaya peel powder, will supply 58.58%, 67.74%, 24.90%, 58.87%, 51.47%, 546.36%, 27.48%, and 699.46% of P, K, Na, Mg, Ca, Fe, Zn, and Cu needs, respectively.

3.4. Burger manufacturing

3.4.1. Color values

Beef burger color was evaluated by CIE Lab color scale (L*, a* and b*). The L* value indicates the lightness, 0-100, representing (0) dark to (100) light. The statistical analysis in Table 5 illustrated that there were non-significant differences between sample T1 and control sample in the L* value of beef burger, while increase the level of PP addition was significantly affected L* values, and sample T3 recorded the lowest L* value. Furthermore, the a* value which gives the degree of the red-green color (higher positive a* value indicating more redness). In this study, the redness or a* values of beef burger significantly decreased from 9.05 for control sample to 5.69 for sample T3. On the other hand, the b* value indicates the degree of yellow-blue color (higher positive b* value indicating more yellowness). The b* values of beef burgers significantly influenced with the PP addition and ranged from 14.24 to 16.75. Moreover, Table 5 shows that the highest total color difference (ΔE) value was observed for control sample (C), while the same value insignificantly decreased after meat substation with 1% PP. The increase in the level of meat substitution with PP resulted in a significant negative affect on ΔE value. The statistical analysis in Table 5 indicated that the chroma values of beef burger were insignificantly ($p > 0.05$) affected with the PP addition. Meanwhile, Table 5 shows that Hue value increased significantly ($p < 0.05$) with the increase in substitution level of meat with PP.

3.4.2. Chemical composition

The proximate composition of beef burgers without and with the PP addition are presented in Table 6. Our data revealed that beef burgers formulated by replacing 1% meat with PP showed the highest significant ($p < 0.05$) increase in moisture content, when compared with control sample. This increase could be attributed to the enhancement in the water holding capacity of the beef burgers after PP addition, while the increase in PP addition resulted in a negative effect due to the increase in fat content (as PP contains 6.5% crude fat). On the other hand, Table 6 illustrates that protein content increased with the replacement of meat by 1% PP, while the increase in PP addition did not show a real effect on protein content. Regarding of fat content, Table 6 shows non-significant increase ($p >0.05$) in sample T1, while increasing the level of PP addition in beef burgers had significant effect on fat content, due to the lipid content of the PP flour. Furthermore, crude fiber content of manufactured beef burger significantly increased ($p< 0.05$) with the increase of substitution level of PP, and sample T3 recorded the highest value.

3.4.3. Sensory properties

Sensory evaluation of beef burgers without and with PP addition (at different levels) after cooking and their appearance before cooking is shown in Table 7 and Fig.5, respectively. Our data showed that beef burger samples which contained 1% PP recorded the best scores (non-significant as $p > 0.05$) in all studied parameters, when compared to control sample (except the color parameter, which recorded higher significant value). Although the scores of organoleptic properties reduced by increasing the level of PP, no significant differences were found between samples and control (except for taste parameter which varied significantly, and sample T3 recorded the lowest score).

3.5. Effect of storage on beef burgers

3.5.1. Physical properties

Cooking properties such as cooking yield, cooking loss and shrinkage are some of the most important factors for meat industry to predict the behavior of products during cooking. Partial replacing of meat with PP (at different levels) during processing burgers and its effect on cooking yield, cooking loss and shrinkage are shown in Table 8. Using PP during processing beef burgers increased cooking yield significantly ($p < 0.05$) and decreased the cooking loss for all samples. Whereas during storage the mean of groups values illustrated that cooking yield increased significantly ($p < 0.05$), while cooking loss decreased, when compared to zero time. On the other hand, Table 8 illustrates the effect of PP addition on beef burgers shrinkage. Control sample recorded the highest shrinkage percent, while using PP during processing burgers decreased shrinkage non-significantly ($p > 0.05$). Furthermore, after three months of storage shrinkage decreased significantly ($p< 0.05$) in all studied samples.

Table 8 shows that partial replacing of meat by PP caused significant decreases ($P < 0.05$) in pH values, when compared to the control sample, while T2 recorded the lowest pH value (5.973) at zero time. By the end of storage period, pH values increased significantly ($P <0.05$), when compared to zero time. Furthermore, mean square and p-values for cooking yield, cooking loss, shrinkage and pH showed a significant effect ($P<0.05$) of treatments (addition of PP with different levels), storage time and their interactions, except for the non-significant effect of treatment × storage time interaction on shrinkage value (Table 8.1).

3.5.2. Chemical changes

Fig. 6 shows the effect of PP addition on thiobarbituric acid reactive substances (TBARS) values of beef burger at zero time and after storage for three months. Non-significant differences ($p > 0.05$) were found between all samples under investigation (without and with PP addition) at zero time. Control sample recorded the highest value of TBARS (0.2964 mg/kg), while addition of PP caused non-significant decrease in TBARS values.

By the third month of storage, the control sample recorded the highest TBARS value (0.8398 mg/kg) between all studied samples. Furthermore, increase of the PP level in burger samples caused a significant ($p < 0.05$) decrease in TBARS values, and sample T3 recorded the lowest TBARS value (0.4082 mg/ kg).

3.5.3. Microbiological changes

Table 9 illustrates the effect of using PP in partial replacement of meat during processing beef burgers on TBC and fungal count. Mean of groups (related to treatments) in the same Table showed non-significant ($p > 0.05$) differences in TBC between treatments and the control sample.

Meanwhile values of groups mean (related to storage time) in Table 9 showed non-significant ($P > 0.05$) decrease in TBC as storage prolonged up to 3 months. This could be attributed to the decrease of water activity during freezing.

On the other hand, data in Table 9 revealed that the control sample recorded the highest fungal count, while using 1% of PP in partial replacement of meat during burgers processing decreased fungal count significantly ($p < 0.05$), and sample T1 recorded the lowest count. Regarding the effect of storage period, fungal count decreased by the end of storage period.

Furthermore, data in Table 10 regarding mean square and p-values for bacterial and fungal counts and illustrated non-significant effect of treatments and interaction of treatment × storage time, while storage time significantly affected on microbial count.

4. Discussion

In this work, we study the phenolic compounds, flavonoids, and antioxidant activity, as well as the antimicrobial activity of papaya wastes to evaluate its importunate as additives in processing meat products (beef burger).

The results in our study demonstrated that papaya peels had higher content of phytochemicals than papaya seeds. The phenolics content in this study were higher than those obtained by **Santos et al. (2014),** as they found phenolic content in two cultivars of papaya ranged between 5.53 and 5.75mg tannic acid/g in peels, and from 2.66 to 3.01 mg tannic acid/ g in seeds. Whereas **Martial-Didier et al. (2017)** reported that total phenolic content in papaya (*Carica papaya L. var solo 8*) peels recorded 65.48 mg (GAE)/100 g on dry weight; besides, **Maisarah et al. (2014)** found that total phenolic content in papaya seeds recorded lower values (30.32 mg GAE/100 g of dry weight), when compared to this study. Fig. 2 shows that papaya PP had higher significant values of total phenolics than papaya SP, as papaya peels protect the fruit and seeds from environmental factors (**Khan et al. 2022**). Likewise**, Santos et al. (2014)** observed that the papaya peels had higher contents of phenolic compounds than seeds. Different results were found by **Asghar et al. (2016),** as they revealed that *Carica* papaya methanolic extract of phenolics in seeds was higher than peels. Furthermore, lower flavonoids contents were found by **Gaye et al. (2019),** when compared with this study, as they found that total flavonoids content in peels 0.23-0.34 mg QE/g, while seeds contained 0.22-1.01 mg QE/g. Whereas **Ovando-Martinez et al. (2018)** found that flavonoids content in papaya skin recorded approximatrly1000 mg QE/100 g, which was higher than our findings. In this study we found lower values of antioxidant activity than those obtained by **Martial-Didier et al. (2017); Lydia et al., (2016)**. These differences may be attributed to the solvent type **(Jiménez-Moreno et al., (2019); Noriega-Rodríguez et al., (2020)).** Regarding GC-MS analysis of PP and SP (Tables 2 and 3), we found higher numbers of bioactive compounds than those detected by **Enearepuadoh and Victor (2021),** as they found 11 compounds in hexane extract of *Carica* papaya L. peels, and the main component of *Carica* papaya L. peels oil was the Oleic acid. Whereas **Abd El-Zaher (2014)** detected 15 compounds in Egyptian C. papaya seed extract, and the volatiles were dominated by esters. On the other hand, **Macleod and Pieris. (1983)** found 37 components in Sri Lanka variety. Moreover, **Farhan et al. (2021)** showed that methanolic extract of C. Papaya seeds analysis of GC-MS indicated presence of many components like phenolic compounds, antioxidants, saturated fatty acid, unsaturated fatty acid, terpenes, and others, which in agreement with this study. Therefore, the presence of all these bioactive compounds in the papaya wastes highlights the importance of the papaya wastes to human health.

The antimicrobial analysis in this study was conducted to compare between PP and SP, and the results showed that using methanolic extract of papaya seeds against pathogens microorganisms revealed excellent antimicrobial activity, which may be attributed to active compounds like Oleic acid **(Igbinaduwa et al., 2018).** Whereas **Asghar et al. (2016)** revealed that methanolic extract of papaya peels recorded 8.12 mm against *B. cereus*, 9.1 mm against *E. coli*, and 8.1 mm against *S. aureus*, which were close to our findings. Likewise, **Pathak et al. (2019)** referred to the antibacterial activity of papaya peels against some Gram-positive and Gram-negative microorganisms, while effectiveness depends on extraction solvent.

Indeed, the data in Fig.2 and Table 2 referred to the valuable findings of papaya PP, when compared to SP, which resulting in a beneficial effect on health, although these compounds can contribute to changes in taste and color of food products (**Sancho et al., 2010)**. Thus, papaya (*Carica papaya*) seeds are known to contain toxic substances, and the toxicity effect of pawpaw (papaya) SP had a positive correlation with exposure time from 24 to 96 h, for the sharp-tooth *catfish C. gariepinus* (**Ayotunde et al., 2010**). Furthermore, **Pathak et al. (2000)** referred to that ingestion of papaya seeds may affect the human male's fertility.

So, for all previous reasons, papaya PP was chosen to use in this study, and we determined the chemical composition of PP to understand its potential effect on the final product.

The chemical analysis of PP showed higher content as compared with those obtained by **Martial-Didier et al. (2017),** as they found that proteins, ash, fat, and carbohydrate content of papaya peels recorded 11.67, 5.98, 2.51 and 47.33%, respectively. Furthermore, **Santos et al. (2014)** determined the content of protein (15.03-18.18 g/ 100g) and ash (11.31-11.85g/ 100g) in papaya peels of two cultivars, which were higher than data obtained in this study. Potassium was the most abundant mineral in PP; other studies reported similar results in papaya peels (**Santos et al.2014, Martial-Didier et al., 2017**). Regarding minerals contribution in daily needs, different findings were reported by **Santos et al. (2014),** as they found the intake of 100 g papaya peels flour, will supply 78%, 44%, 81%, 34%, 22%, 47%, and 121% of phosphorus, potassium, magnesium, calcium, iron, zinc, and copper, respectively. These differences may be attributed to cultivars and RDI reference comparison (as the updated daily values were used in this study **(FDA, 2020**).

In this study, we used three levels of PP (1, 2, 1nd 3%) as a partial replacement of meat during processing beef burger, we estimated the effect of these addition on color, and we noticed that the addition of papaya PP to beef burger causes a change in lightness (L*) (Table 5), the decrease in L* values of beef burger samples with the increase of the level of PP addition could be due to the increase in the concentration of the carotenoid pigments in papaya peels (which absorb part of the visible spectrum) (**Velasco-Arango et al., 2020**). Another potential explanation for L* reduction in burger samples with PP in this study related to the more water retention in beef burger with PP (with high fiber contents), when compared to control sample; as the less water retention caused an increase in light reflectance and showed lighter appearance (**Baublits et al., (2006)**) in control sample. Whereas the increase in b* value with the increase of PP addition was expected because the high b* values reflected yellowness during ripening of papaya fruits due its content of β-carotene (**Ovando-Martinez et al., 2018**). Similarly with this study, **Zahid et al. (2020**) illustrated non-significant decrease in chroma value of meat patties samples formulated with clove extract, when compared with control. Meanwhile, the increase in substitution level of PP increased ˚Hue values, which indicate orange color (**Ovando-Martinez et al., 2018**).

On the other hand, a change in the nutritional composition of beef burger samples after addition PP was detected (Table 6). The data showed that moisture content increased with the first proportion (1%), due to the increase in moisture retention. The increase in free moisture content with its lubricating nature on processing caused an improvement in the product structure compactness and results in a juicy tissue protein structure **(Jiang, 2020).** Whereas increasing PP levels caused a negligible effect on protein content. Furthermore, fat content increased in beef burger with PP addition, due to the ability of fiber to absorb fat and interact with the protein of the ground meat matrix to prevent migration of fat from the product (**Alakali et al., 2010**). Our findings in fat content of beef burger samples were on the line with previous study was conducted by **Azevedo and Campagnol (2014).** On contrary, **Abd EL - Halim (2014)** found that fat content of camel burger decreased with addition papaya extract. Furthermore, PP contains fiber (Fig. 3), and by increasing the percentage of PP in the burger formulation, fiber content increase in the burgers.

Regarding sensory evaluation of beef burger, PP addition at low levels affect non-significantly (Table 7), as compared with control sample. Likewise, **Azevedo and Campagnol (2014)** found similar trend when they used papaya seeds in manufacturing beef burgers.

Furthermore, we study the effect of storage on beef burger samples, and we found that the cooking yield in beef burgers formulated with PP enhanced due to the ability of PP fibers to keep water; as cooking yield is correlated to the significant increase in moisture retention, which related to fibers content interactions with meat proteins and formation of a complex network (which could keep water from migration to the surface) (**Azevedo and Campagnol (2014); Anderson and Berry (2001)).** Similar results to our study were obtained by **Shokry (2016)** for the cooking yield in meat burger formulated with quinoa flour, who illustrated that the cooking yield increased with increasing level of quinoa flour incorporated in beef burger. Whereas **Azevedo and Campagnol (2014)** referred to the improvement of processing burger cooking yield with the increase in the level of papaya seeds flour, while they found lower cooking yield values than ours. Another study illustrated the role of high fiber content in enhancement of cooking yield, WHC, cooking loss and shrinkage (**Gad EL Rab et al., 2019**). Meanwhile **França et al. (2022**) found higher cooking loss values than our findings when they used shiitake byproducts in manufacturing low-salt beef burgers. Furthermore, **Zahid et al. (2020)** found non-significant ($p>0.05$) changes in the cooking loss in all patties during storage times. Our findings were on the line with previous study was carried out by **Azevedo and Campagnol (2014**), as they noticed a decrease in shrinkage% after papaya seeds addition during processing burgers.

In this study, the decrease in the pH values with partial replacing of meat by PP was observed. Likewise, **Mokhtar and Eldeep (2020**) found similar trend when they used mango peel extract during processing beef burgers, and the pH values of the fresh burgers ranged between 5.86 and 5.91. However, **Van der Wal et al. (1988)** revealed that the pH of raw beef was ranging from 5.5 to 6.0. Thus, the addition of papaya PP did not lead to denaturation of meat proteins and were satisfactory for the use in beef burgers. Whereas a slightly lower pH (5.6-5.68) was found by **França et al. (2022)** in raw burgers with shiitake byproducts. By the end of storage period, pH values increased. Likewise, **Ozer and Saricoban (2010)** referred to the significant impact of frozen storage on the increase of pH value in meat products. The increase in pH value during frozen storage, could be due to alkaline byproducts production by the microbial growth and protein liberation metabolites (**Amiri et al., 2019; Kalaikannan et al., 2007).** Similar trend in pH values during storage in meat products were reported in other studies **(Zahid et al., 2020; Abdelhakam et al., 2019).**

Non-significant decrease in TBARS values was associated with PP addition to beef burger. Similar trend was observed by **Abd EL- Halim (2014**), as he found that addition of papaya extracts decreased TBARS values, while control sample recorded lower TBARS value than our findings. The storage period in this study caused an increase in TBARS values in beef burger samples. likewise, **Mokhtar and Eldeep (2020)** referred to the increase of TBARS values by progress of storage, and control sample recorded the highest value (0.95 mg malondialdehyde/kg). The results of TBARS in our study indicated that PP (rich in phenolics and flavonoids) may works as a potential natural antioxidant could deal with free radicals and inhibit oxidative processes. As TBARS values measure oxidative rancidity development in meat products during storage (**Abdelhakam et al., 2019**).

Regarding TBC, non-significant difference at the beginning of storage time between beef burger containing PP and control. Similar findings were reported by **Mokhtar and Eldeep (2020).** Whereas **Abd EL - Halim (2014)** found higher content of TBC in fresh control camel burger, when compared with this study, and he noticed a decline in TBC content with papaya extract addition. Moreover, TBC and fungal count decline during frozen storage. Similar observations were reported by **Abdelhakam et al. (2019)** when they used red grapes pomace powder during processing beef hamburger.

These results showed different formulas of beef burgers containing PP, indicating the importance of the PP as a functional ingredient during processing beef burgers and its positive effects on quality characteristics of beef burgers. Although the enhancement in beef burger quality with the addition of PP, the bitter taste appeared and limited the use of high concentrations of PP. Further studies can be conduct in the future by using PP extracts during making different food products.

5. Conclusion:

As a waste papaya PP has promising healthy nutrients and contains higher contents of fiber, protein, and minerals. Indeed, the present study indicated that PP is a good source of phenolics and flavonoids, which could be used as an interesting alternative to synthetic antioxidants to prevent the oxidation in meat products. Whereas beef burger is one of the most popular foods between consumers, so using healthy, nutritious ingredient like PP during processing beef burger could be considerable. Besides PP addition improved cooking yield, decreased shrinkage without a significant change in sensory properties especially in sample T1. The addition of papaya peel flour in the beef burger formulation not only allows the improvement of the nutritional and technological quality of this widely consumed product but also can be effective to reduce the environmental impact of the improper disposal of industrial waste.

Therefore, the production and use of papaya PP could involve in food industries, supports environment sustainability, and suitable for consumers.

Conflicts of Interest/ Competing interest: The authors declare that there is no conflict of interest**.**

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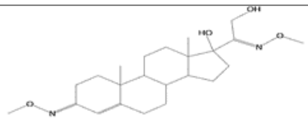
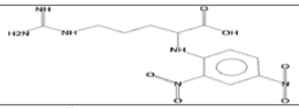
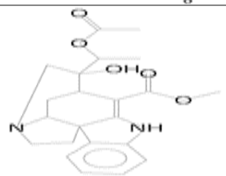
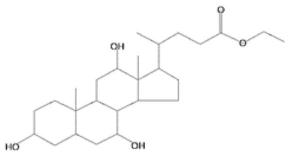
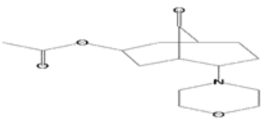
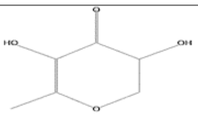
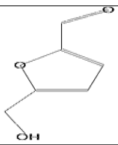
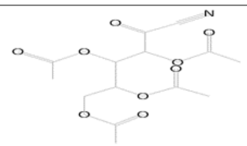
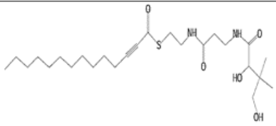
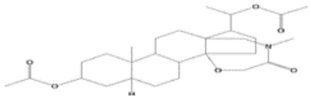
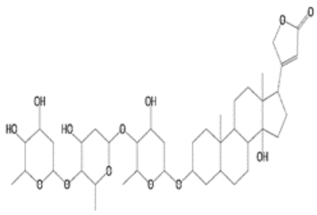
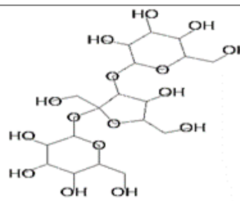
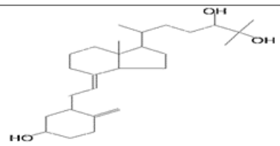
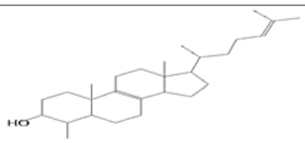
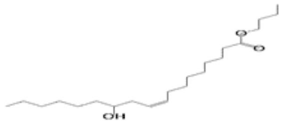
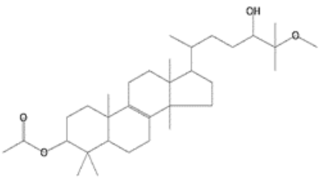
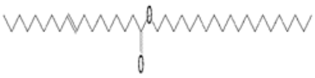
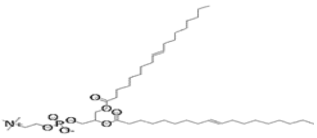
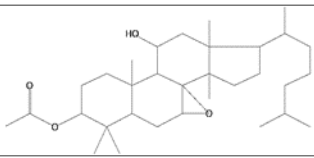
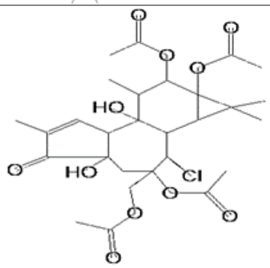
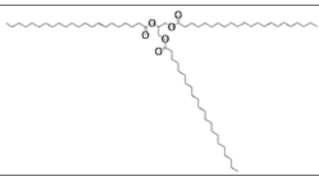
Table 1: Ingredients (%) of beef burger samples.

Samples Ingredients	Control	T1	T2	T3
Lean meat	71.0	70.0	69	68
Fat	10.0	10.0	10.0	10.0
Cold water	15.4	15.4	15.4	15.4
Salt	1.0	1.0	1.0	1.0
White pepper	0.2	0.2	0.2	0.2
Black pepper	0.2	0.2	0.2	0.2
Garlic powder	0.2	0.2	0.2	0.2
Onion powder	2.0	2.0	2.0	2.0
Papaya PP	0.0	1.0	2.0	3.0

Table 2: Gas chromatography (GC)-Mass Spectrometry (MS) analysis results of methanolic extract of papaya PP.

No.	Retention time (min)	Compound	Chemical structure	Molecular formula	Molecular weight	Area %	Biological activity
1	4.25	Pregn-4-ene-3,20-dione, 17,21-dihydroxy-, bis(O-methyloxime)		C23H36N2O4	404	0.453	Anti-inflammatory (Al-Gara'awi et al., 2019)
2	4.62	N-2,4-Dnp-L-arginine		C12H16N6O6	340	0.693	It has a therapeutic effect on tumours according to Kamstra et al., (2015)
3	5.51	Curan-17-oic acid, 19-(acetyloxy)-2,16-didehydro-20-hydroxy-, methyl ester, (19S)-		C22H26N2O5	398	0.420	Anti-yeast activity was reported for Curan-17-oic acid,2,16-didehydro-20-hydroxy-19 oxo, methyl ester (Hussein et al., 2016)
4	9.09	Ethyl iso-allocholate		C26H44O5	436	0.508	which has antieczematic (Brintha et al., 2017), antimicrobial and anti-inflammatory effects according to Hameed et al., (2015)
5	11.06	Acetic acid, 6-morpholin-4-yl-9-oxobicyclo [3.3.1] non-3-yl ester/ Clindamycin		C15H23NO4	281	1.977	-
6	11.6	Octadecane, 3-ethyl-5-(2-ethylbutyl)-		C26H54	366	0.430	Antioxidant, Antibacterial (Addai et al., 2022); anti-inflammatory effect (Al-Marzoqi et al., 2015)
7	13.27	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl		C6H8O4	144	2.326	Anti-diabetic and antioxidant activity (Hameed et al 2015)
8	15.7	2-Furancarboxaldehyde, 5-(hydroxymethyl)-		C6H6O3	126	1.489	Antimicrobial effect according to Premathilaka and Silva (2016)

9	16.26	Tetraacetyl-d-xylonic nitrile		$C_{14}H_{17}NO_9$	343	1.364	Antitumour and antioxidant (Hameed et al 2015)
10	17.42	2-Myristynoyl pantetheine		$C_{25}H_{44}N_2O_5S$	484	2.128	-
11	18	Cyclopropanebutanoic acid 2-[[2-[[2-[(2-pentylcyclopropyl) methyl] cyclopropyl] methyl] cyclopropyl] methyl]-, methyl ester		$C_{25}H_{42}O_2$	374	0.548	Anti-inflammatory, antioxidant, antimalarial, antituberculosis and antifungal (Hussein et al., 2015)
12	19.57	(5α) Pregnane-3,20α-diol, 14α,18α-[4-methyl-3-oxo-(1-oxa-4-azabutane-1,4-diyl)]-, diacetate		$C_{28}H_{43}NO_6$	489	0.576	Löfgren et al. (1992) referred to anaesthetic properties of (5α) Pregnane-3 alpha-ol-20 one, in human beings
13	20.4	Digitoxin		$C_{41}H_{64}O_{13}$	764	0.428	which has anticancer effect according to Elbaz et al., (2012), and has anesthetic, cardiotonic and diuretic effects (Brintha et al., 2017); antimicrobial, anticancer effects, anti–inflammatory, Treatment of heart disease (Al-Gara'awi et al., 2019)
14	22.54	α -D-Glucopyranoside, O-α -D-glucopyranosyl-(1. fwdarw.3)-α-D -fructofuranosyl		$C_{18}H_{32}O_{16}$	504	2.004	Sosa et al., 2016 reported that α glucopyranosyl -D-Glucopyranoside, O -(1. fwdarw.3)-ß-d-fruc-α-D- has anti-diabetic activity and antitumour. Besides Tayade et al. (2013) referred to its important as a preservative
15	26.96	9,10-Secocholesta-5,7,10(19)-triene-3, 24,25-triol, (3α,5Z,7E)-		$C_{27}H_{44}O_3$	416	0.966	which has anti-eczematic, anti-osteoporotic and inhibits prostaglandin-E2 9-reductase (Brintha et al., 2017); anti-inflammatory, and anticancer, antihepatoprotective (Al-Marzoqi et al., 2015); antieczematic (Brintha et al., 2017)
16	37.37	Cholesta-8,24-dien-3-ol, 4-methyl-, (3α,4α)-		$C_{28}H_{46}O$	398	0.462	Which has antimicrobial and diuretic effects according to Muthulakshmi et al. (2012)

17	37.6	n-Butyl ricinoleate		$C_{22}H_{42}O_3$	354	0.630	Kuppala et al. (2016) referred to antibacterial and anti-mycobacterial properties of ricinoleic acid-derived compounds
18	39.61	Acetic acid,7-(4-hydroxy-5-methoxy-1,5-dimethylhexyl)-4,4,10,13,14-pentamethyl-2,3,4,5,6,7,10,11,12,13,14,15,16,17-tetradecahydrocyclopenta[a]phenanthryl ester		$C_{33}H_{56}O_4$	516	18.59	-
19	43.04	Oleic acid, eicosyl ester		$C_{38}H_{74}O_2$	562	1.141	which has anti-inflammatory and anticancer effects and lowering cholesterol according to Kumar et al., (2018)), Anti-Candida activity (Sosa et al., 2016)
20	43.69	3,5,9-Trioxa-5-phosphaheptacos-18-en-1-aminium, 4-hydroxy-N, N, N-trimethyl-10-oxo-7-[(1-oxo-9-octadecenyl)oxy]-, hydroxide, inner salt, 4-oxide, (R)-		$C_{44}H_{84}NO_8P$	785	1.835	considered a phospholipid
21	46.12	7,8-Epoxylanostan-11-ol, 3-acetoxy-		$C_{32}H_{54}O_4$	502	1.165	which has antimicrobial, anti-inflammatory effect according to Hassan et al., (2014)
22	47	5H-Cyclopropa[3,4]benz[1,2-e]azulen-5-one,3,9,9a-tris(acetyloxy)-3-[(acetyloxy)methyl]-2-chloro-1,1a,1b,2,3,4,4a,7a,7b,8, 9,9a-dodecahydro-4a,7b-dihydroxy-1,1,6,8-tetramethyl-, [1aR-(1aα,1bα,2α,3α,4aα,7aα,7bα,8α,9α, 9aα)]-		$C_{28}H_{37}ClO_{11}$	584	1.305	-
23	48.81	Docosanoicacid, 1,2,3-propanetriyl ester		$C_{69}H_{134}O_6$	1058	3.235	which has emulsifying properties according to Daffodil et al., (2012))

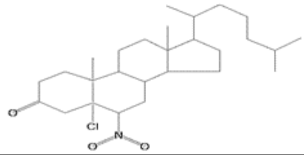
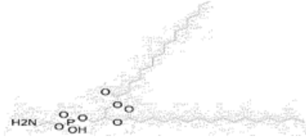
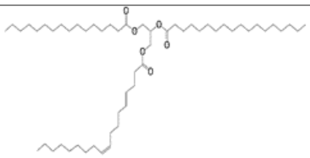
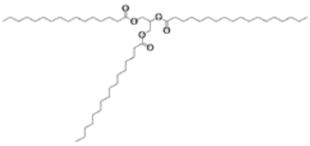
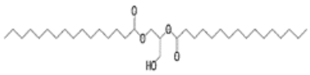
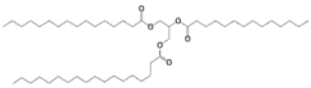
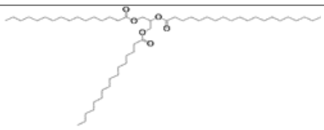
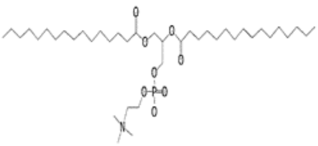
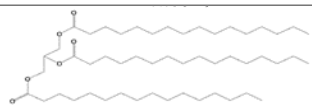
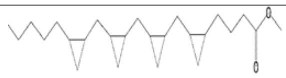
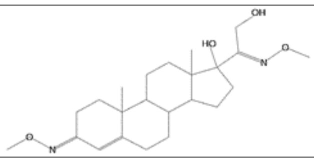
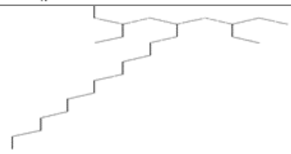
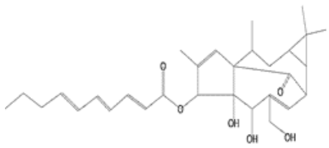
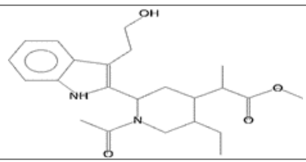
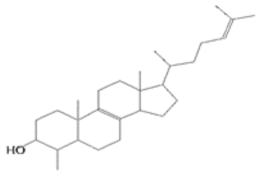
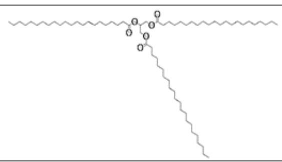
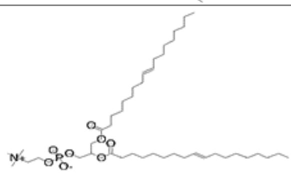
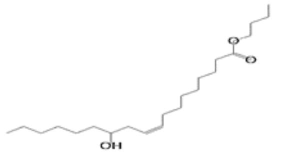
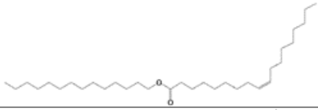
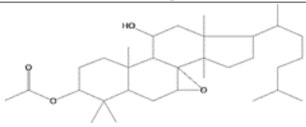
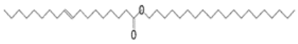
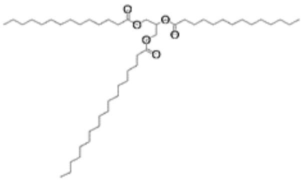
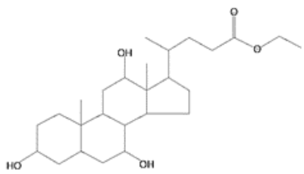
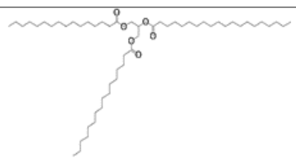
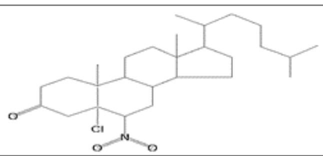
24	50.11	5-Chloro-6beta-nitro-5alpha-cholestan-3-one		C27H44ClNO3	465	5.225	steroids
25	51.23	Hexadecanoic acid,1-[[[(2-aminoethoxy) hydroxyphosphinyl] oxy] methyl]-1,2-ethanediyl ester		C37H74NO8P	691	4.991	-
26	51.79	9-Octadecenoic acid (Z)-,3-[(1-oxohexadecyl) oxy]-2-[(1-oxooctadecyl) oxy] propyl ester		C55H104O6	860	8.151	-
27	52.14	Octadecanoic acid,2-[(1-oxohexadecyl) oxy]-1-[[(1-oxohexadecyl) oxy] methyl] ethyl ester		C53H102O6	834	8.081	-
28	54.6	Hexadecanoic acid,1-(hydroxymethyl)-1,2-ethanediyl ester		C35H68O5	568	6.245	-
29	55.82	Octadecanoic acid,3-[(1-oxohexadecyl) oxy]-2-[(1-oxotetradecyl) oxy] propyl ester		C51H98O6	806	10.89	-
30	57.37	Eicosanoic acid,2-[(1-oxohexadecyl) oxy]-1-[[(1-oxohexadecyl) oxy] methyl] ethyl ester		C55H106O6	862	7.174	Mohammed et al. (2016) revealed the anti-inflammatory and anti-melasma properties of Eicosanoic acid, 2-(acetyloxy0-1-[(acetyloxy)methyl] ethyl ester
31	58.93	3,5,9-Trioxa-4-phosphapentacosan-1-aminium,4-hydroxy-N, N, N-trimethyl-10-oxo-7-[(1-oxohexadecyl) oxy]-, hydroxide, inner salt, 4-oxide		C40H80NO8P	733	3.857	-
32	61.02	Tripalmitin		C51H98O6	806	0.697	-

Table 3: Gas chromatography (GC)-Mass Spectrometry (MS) analysis results of methanolic extract of papaya SP.

No.	Retention time (min)	Compound	Chemical structure	Molecular formula	Molecular weight	Area%	Pharmacological action
1	4.25	Cyclopropanebutanoic acid 2-[[2-[[2-[(2-pentylcyclopropyl) methyl] cyclopropyl] methyl] cyclopropyl] methyl]-, methyl ester		C25H42O2	374	0.761	Anti-inflammatory, antioxidant, antimalarial, antituberculosis and antifungal (Hussein et al,. 2015)
2	4.61	Pregn-4-ene-3,20-dione, 17,21-dihydroxy-, bis(O-methyloxime)		C23H36N2O4	404	0.378	Anti-inflammatory (Al- Gara'awi et al,.2019)
3	11.6	Octadecane, 3-ethyl-5-(2-ethylbutyl)-		C26H54	366	0.635	Antioxidant and Antibacterial (Addai et al., 2022); antioxidant and anti-inflammatory effect (Al-Marzoqi et al., 2015)
4	21.15	2,4,6-Decatrienoic acid,1a,2,5,5a,6,9,10,10a-octahydro-5,5a-dihydroxy-4-(hydroxymethyl)-1,1,7,9-tetramethyl-11-oxo-1H-2,8a-methanocyclopenta[a]cyclopropa[e]cyclodecen-6-yl ester1,αR (1aα,2α,5α,5aα,6α,8aα,9α,10aα)-[		C30H40O6	496	0.834	Sosa et al., 2016 referred to the anticancer activity of 2,4,6-Decatrienoic acid, 1a,2,5,5a,6,9,10,10a-octahydro-5,5adihy
5	37.51	4-Piperidineacetic acid,1-acetyl-5-ethyl-2-[3-(2-hydroxyethyl-(1H-indol-2-yl]-α-methyl-, methyl ester		C23H32N2O4	400	8.328	-
6	38.35	Cholesta-8,24-dien-3-ol, 4-methyl, (3α,4α)-		C28H46O	398	0.513	Which has antimicrobial and diuretic effects according to Muthulakshmi et al. 2012
7	39.67	Docosanoic acid, 1,2,3-propanetriyl ester		C69H134O6	1058	11.299	which has emulsifying properties according to Daffodil et al., (2012)
8	40.90	3,5,9-Trioxa-5-phosphaheptacos-18-en-1-aminium,4-hydroxy-N, N, N-trimethyl-10-oxo-7]-(1-oxo-9-octadecenyl) oxy,-[hydroxide, inner salt, 4-oxide, (R)		C44H84NO8P	785	1.94	considered a phospholipid

9	42.24	n-Butyl ricinoleate		C22H42O3	354	0.865	Kuppala et al. (2016) referred to antibacterial and anti-mycobacterial properties of ricinoleic acid-derived compounds
10	43.04	9-Octadecenoic acid (Z)-, tetradecyl ester		C32H62O2	478	1.624	-
11	48.58	7,8-Epoxylanostan-11-ol, 3-acetoxy-		C32H54O4	502	1.734	which has antimicrobial, anti-inflammatory effect according to Hassan et al., (2014); Lee et al., (2013)
12	49.77	Oleic acid, eicosyl ester		C38H74O2	562	1.637	which has anti-inflammatory, anticancer effects and lowering cholesterol according to Kumar et al., (2018)), anti-Candida activity (Sosa et al., 2016)
13	51.44	Octadecanoic acid, 2,3-bis[(1-oxotetradecyl) oxy] propyl ester		C49H94O6	778	5.382	-
14	53.44	Ethyl iso-allocholate		C26H44O5	436	2.314	which has antieczematic (Brintha et al., 2017), antimicrobial and anti-inflammatory effects according to Hameed et al., (2015); Anticancer activity, (Al-Gara'awi et al., 2019)
15	55.10	Eicosanoic acid,2-[(1-oxohexadecyl) oxy]-1-[[(1-oxohe xadecyl) oxy] methyl] ethyl ester		C55H106O6	862	2.566	Mohammed et al. (2016) referred to the anti-inflammatory and anti-melasma properties of Eicosanoic acid, 2-(acetyloxy0-1-[(acetyloxy)methyl] ethyl ester
16	55.79	5-Chloro-6beta-nitro-5alpha-cholestan-3-one		C27H44ClN O3	465	10.177	steroids

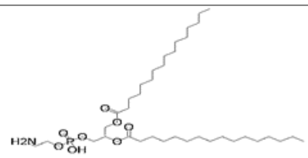
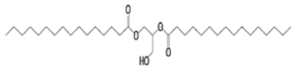
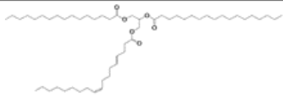
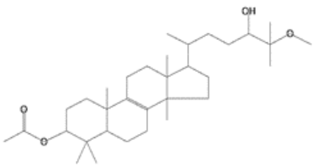
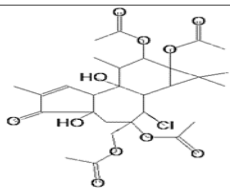
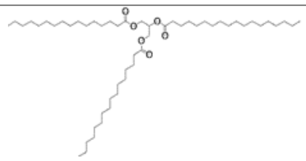
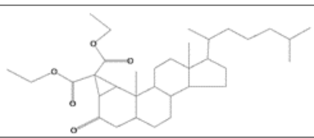
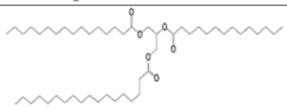
17	56.60	Hexadecanoic acid,1-[[[(2-aminoethoxy) hydroxyphosphinyl] oxy] methyl]-1,2-ethanediyl ester		C37H74NO8P	691	4.469	-
18	57.32	Hexadecanoic acid,1-(hydroxymethyl)-1,2-ethanediyl ester		C35H68O5	568	8.228	-
19	58.00	9-Octadecenoic acid (Z),-3-[(1-oxohexadecyl) oxy]-2-[(1-oxooctadecyl) oxy] propyl ester		C55H104O6	860	5.403	-
20	58.46	Acetic acid,7-(4-hydroxy-5-methoxy-1,5-dimethylhexyl)-4,4,10,13,14-pentamethyl-2,3,4,5,6,7,10,11,12,13,14,15,16,17-tetradecahydrocyclopenta[a]phenanthryl ester		C33H56O4	516	5.812	-
21	58.77	5H-Cyclopropa [3,4] benz[1,2-e] azulen-5-one,3,9,9a-tris (9,9a-dodecahydro-4a,7b-dihydroxy-1,1,6,8-tetramethyl-, [1aR-(1aα,1bα,2α,3α,4aα,7aα,7bα,8α,9α, 9aα)]-		C28H37ClO11	584	6.186	-
22	59.68	Octadecanoic acid,2-[(1-oxohexadecyl) oxy]-1-[[(1-oxohexadecyl) oxy] methyl] ethyl ester		C53H102O6	834	3.831	-
23	60.41	3'H-Cycloprop (1,2)-5α-cholest-1-en-3-one, 1',1'-dicarboethoxy-1α,2α-dihydro		C34H54O5	542	2.835	-
24	60.84	Octadecanoic acid,3-[(1-oxohexadecyl) oxy]-2-[(1-oxotetradecyl) oxy] propyl ester		C51H98O6	806	12.244	-

Table 4: Antimicrobial activity of papaya PP and papaya SP methanolic extracts.

Microorganisms	Inhibition zone (mm)			
	Papaya PP methanolic extract	Papaya SP methanolic extract	Methanol	Standard cont.
Bacillus cereus (+ve) AUMC No. B-52	8	9	-	20
Escherichia coli (-ve) AUMC No. B-53	8	8	-	23
Micrococcus luteus (+ve) AUMC No. B-112	8	8	-	22
Pseudomonas aeruginosa (-ve) AUMC No. B-73	8	9	-	18
Serratia marcescens (-ve) AUMC No. B-55	8	9	-	22
Staphylococcus aureus (+ve) AUMC No. B-54	8	8	-	20
Fusarium oxysporum AUMC No. 215	-	10	-	24
Geotrichum candidum AUMC No. 226	-	10	-	25

AUMC No: Assiut University Mycological Center.
Standard cont.: Chloramphenicol as antibacterial standard, clotrimazole as antifungal standard.

Table 5: Color estimation of beef burger samples without and with papaya PP.

Burger samples	L*	a*	b*	Total color index (Δ E)	Chroma	Hue
C	53.32 ± 0.67^{a}	9.05 ± 0.24^{a}	14.24 ± 0.66^{c}	55.93 ± 0.44^{a}	16.87 ± 0.68^{ab}	57.56 ± 0.53^{c}
T1	52.03 ± 1.46^{a}	7.16 ± 0.43^{b}	14.95 ± 0.43^{bc}	54.60 ± 1.41^{a}	16.57 ± 0.32^{b}	64.39 ± 1.78^{b}
T2	49.82 ± 1.16^{b}	6.23 ± 0.31^{c}	16.75 ± 0.62^{a}	52.93 ± 0.86^{b}	17.87 ± 0.68^{a}	69.61 ± 0.34^{a}
T3	49.65 ± 0.21^{b}	5.69 ± 0.16^{c}	15.55 ± 0.73^{b}	52.34 ± 0.05^{b}	16.56 ± 0.64^{b}	69.90 ± 1.36^{a}

C: control beef burger; T1: beef burger with 1% papaya PP as partial replacement of meat; T2: beef burger with 2% PP as partial replacement of meat; T3: beef burger with 3% PP as partial replacement of meat; $^{a-c}$ Means of triplicate ±SD (standard deviation) with different small letters in the same column differ significantly at $p<0.05$.

Table 6: Effect of partial replacement of meat with papaya PP on chemical composition (on dry weight basis %) in beef burgers.

Burger samples	Moisture	Protein	Fat	Crude fiber
C	56.82 ± 0.22^{b}	46.077 ± 0.28^{b}	28.84 ± 1.37^{c}	0.321 ± 0.00^{c}
T1	58.76 ± 0.01^{a}	47.07 ± 0.13^{a}	30.85 ± 0.43^{bc}	0.268 ± 0.00^{d}
T2	55.48 ± 0.01^{c}	46.24 ± 0.14^{b}	33.82 ± 0.48^{a}	0.785 ± 0.02^{b}
T3	55.39 ± 0.01^{c}	46.27 ± 0.14^{b}	31.76 ± 0.85^{ab}	0.849 ± 0.00^{a}

C: control beef burger; T1: beef burger with 1% papaya peels (PP) as partial replacement of meat; T2: beef burger with 2% PP as partial replacement of meat; T3: beef burger with 3% PP as partial replacement of meat.; a-d Means of duplicate ±SD (standard deviation) with different small letters in the same column differ significantly at $p<0.05$.

Table 7: Sensory properties of beef burger samples substituted with papaya PP (at different levels).

Sausage samples	Color (9 points)	Texture (9 points)	Odor (9 points)	Taste (9 points)
C	7.87±0.84^{bc}	8.63±0.74^{ab}	8.50±0.75^{ab}	8.75±0.88^{a}
T1	8.75±0.71^{a}	9.00±0.75^{a}	9.00±0.75^{a}	9.00±0.64^{a}
T2	8.13±0.64^{ab}	8.75±0.88^{ab}	8.13±0.84^{b}	8.00±0.54^{b}
T3	7.25±0.88^{c}	8.13±0.64^{b}	7.75±0.71^{b}	7.00±0.75^{c}

C: control beef burger; T1: beef burger with 1% papaya PP as partial replacement of meat; T2: beef burger with 2% PP as partial replacement of meat; T3: beef burger with 3% PP as partial replacement of meat.; $^{a\text{-}c}$ Means ±SD (standard deviation) with different small letters in the same column differ significantly at p<0.05

Table 8: Effect of different levels of papaya PP addition on cooking yield, cooking loss, shrinkage, and pH values of beef burgers at zero time and during storage.

Item	Beef burger samples	Storage time (months)			Mean of groups
		0	1	3	
Cooking yield	C	79.43±0.12	86.66±0.18	81.79±0.02	82.63^{c}
	T1	86.73±0.22	85.41±0.08	87.25±0.23	86.46^{b}
	T2	85.12±0.84	90.89±0.03	90.73±0.13	88.91^{a}
	T3	85.93±0.13	90.27±1.73	92.19±0.06	89.46^{a}
	Mean	84.30^{B}	88.31^{A}	87.99^{A}	
Cooking loss	C	20.57±0.12	13.34±0.18	18.21±0.02	17.37^{a}
	T1	13.27±0.22	14.58±0.08	12.75±0.23	13.54^{b}
	T2	14.87±0.84	9.11 ±0.03	9.27±0.13	11.08^{c}
	T3	14.07±0.13	9.73±1.73	7.8012±0.06	10.54^{c}
	Mean	15.69^{A}	11.69^{B}	12.01^{B}	
Shrinkage	C	10.60±4.39	15.58±8.25	5.67±2.15	10.62^{a}
	T1	7.85±0.02	12.96±4.82	7.17±3.81	9.59^{a}
	T2	5.52±1.11	6.84±1.79	1.37±0.06	4.98^{b}
	T3	10.23±4.77	7.26±00	5.21±1.05	7.94^{ab}
	Mean	8.55^{A}	10.97^{A}	4.95^{B}	
pH	C	6.41±0.03	6.43±0.01	6.25±0.02	6.37^{a}
	T1	6.04±0.11	6.16±0.01	6.16±0.02	6.13^{b}
	T2	5.97±0.03	6.06±0.01	6.03±0.04	6.03^{c}
	T3	6.01±0.02	6.03±0.02	6.06±0.01	6.03^{c}
	Mean	6.11^{C}	6.17^{A}	6.13^{B}	

C: control beef burger; T1: beef burger with 1% papaya PP as partial replacement of meat; T2: beef burger with 2% PP as partial replacement of meat; T3: beef burger with 3% PP as partial replacement of meat; Means of triplicate ±SD (standard deviation); $^{A\text{-}C}$ Different the capital letters in the same row means significantly difference (p<0.05) between storage periods; $^{a\text{-}c}$ Different the small letters in the same column means significantly difference (p<0.05) between treatments.

Table 8.1: Mean squares and P-values (in parentheses) for cooking yield, cooking loss, shrinkage, and pH values.

Factor	df	Mean Square (P-values)			
		Cooking yield	Cooking loss	Shrinkage	pH
Treatment[1]	3	58.148(0.000*)	58.148(0.000*)	55.48 (0.029)	0.253 (0.000*)
Storage time[2]	2	39.69(0.000*)	39.69(0.000*)	81.85 (0.013)	0.017 (0.000*)
treatment × storage time	6	10.502(0.000*)	10.502(0.000*)	12.48 (0.561)	0.014 (0.000*)

[1] Treatments: C: control beef burger; T1: beef burger with 1% papaya PP as partial replacement of meat; T2: beef burger with 2% PP as partial replacement of meat; T3: beef burger with 3% PP as partial replacement of meat.
[2] Storage time: 0,1 and 3 months; df: degrees of freedom; *Statistically significant at P<0.05

Table 9: Microbiological counts (log CFU/g) in beef burger samples without and with papaya PP addition at zero time and during storage for three months.

Item	Beef burger samples	Storage period (months)				Mean of groups
		0	1	2	3	
TBC	control	4.04±0.03	3.88±0.02	4.12±0.16	4.29±0.131	4.08^{a}
	T1	4.21±0.39	3.67±0.09	4.25±0.18	4.03±0.113	4.04^{a}
	T2	4.23±0.14	3.93±0.26	4.05±0.14	4.12±0.058	4.08^{a}
	T3	4.41±0.38	3.77±0.25	4.39±0.05	4.23±0.073	4.20^{a}
	Mean of groups	4.22^{A}	3.82^{B}	4.20^{A}	4.17^{A}	
Fungal count	control	3.88±0.02	2.991±0.30	3.57±0.12	2.83±0.75	3.32^{a}
	T1	3.03±0.25	2.699±0.00	3.23±0.13	2.60±0.43	2.89^{b}
	T2	3.63±0.40	2.94±0.34	3.38±0.15	2.81±0.47	3.19^{ab}
	T3	3.29±0.64	2.95±0.07	3.36±0.29	2.5±0.28	3.03^{ab}
	Mean of groups	3.46^{A}	2.89^{B}	3.38^{A}	2.68^{B}	

C: control beef burger; T1: beef burger with 1% papaya PP as partial replacement of meat; T2: beef burger with 2% PP as partial replacement of meat; T3: beef burger with 3% PP as partial replacement of meat; Means of duplicate ±SD (standard deviation);
$^{A-B}$ Different the capital letters in the same row means significantly difference (p<0.05) between storage periods; $^{a-b}$ Different the small letter in the same column means significantly difference (p<0.05) between treatments.

Table 10: Mean squares and P-values (in parentheses) for TBC and fungal count.

Factor	df	Mean Square (P-values)	
		TBC	Fungal count
Treatment[1]	3	0.038 (0.405)	0.285(0.123)
Storage time[2]	3	0.294 (0.002*)	1.134(0.001*)
treatment × storage time	9	0.035 (0.508)	0.043(0.949)

[1] Treatments: C: control beef burger; T1: beef burger with 1% papaya peels (PP) as partial replacement of meat; T2: beef burger with 2% PP as partial replacement of meat; T3: beef burger with 3% PP as partial replacement of meat.
[2] Storage time: 0,1,2 and 3 months; df: degrees of freedom; *Statistically significant at P<0.05.

Figures

Fig.1. Ripe papaya fruits, peels, and seeds before and after drying.

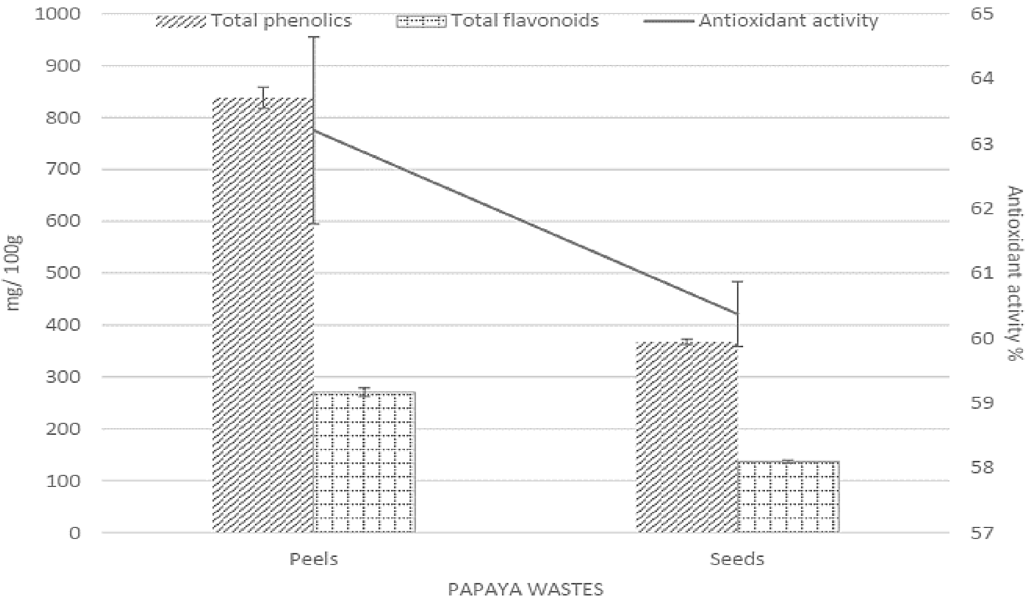


Fig.2: Total phenolics (mg GAE/100g sample on dry weight basis), Total flavonoids (mg Quercetin equivalent/ 100g sample on dry weight basis), and antioxidant activity% (by DPPH radical scavenging) values in papaya wastes. Means of triplicate ±SD (standard deviation).

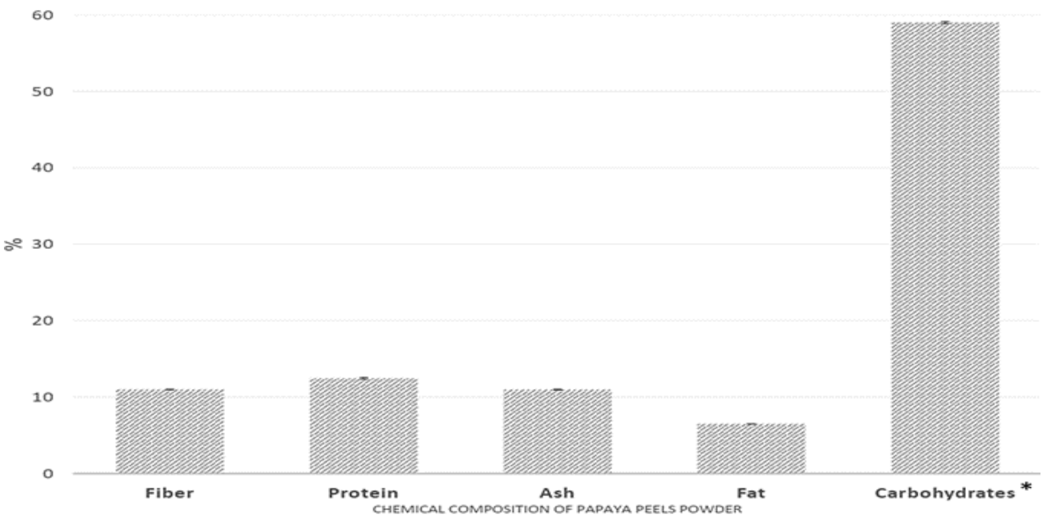


Fig.3. Proximate composition of papaya PP.
Means of duplicate ±SD (standard deviation) (g/ 100g on dry weight basis); *Carbohydrates calculated by differences.

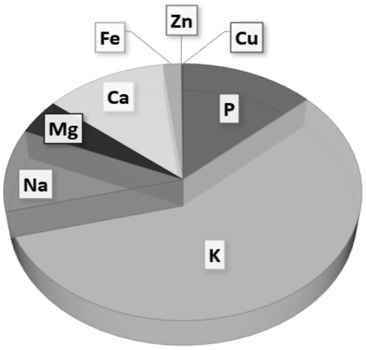
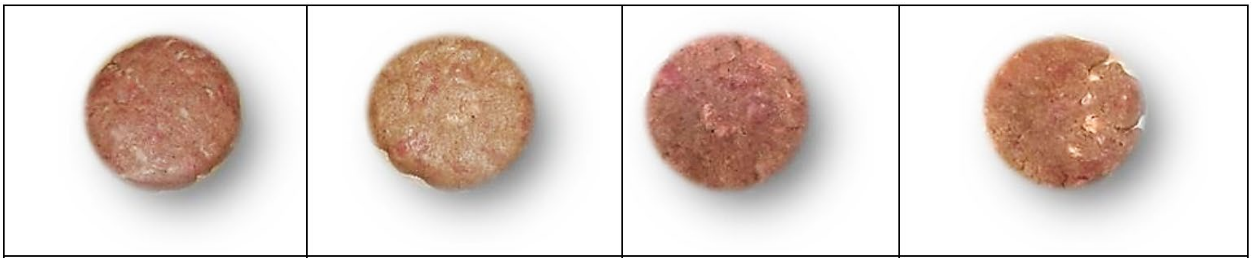


Fig. 4. Minerals content (mg/ 100 g) of papaya PP.



C	T1	T2	T3

Fig. 5: Appearance of burger samples without and with using papaya peels.
C: control beef burger; T1: beef burger with 1% papaya PP as partial replacement of meat; T2: beef burger with 2% PP as partial replacement of meat; T3: beef burger with 3% PP as partial replacement of meat.

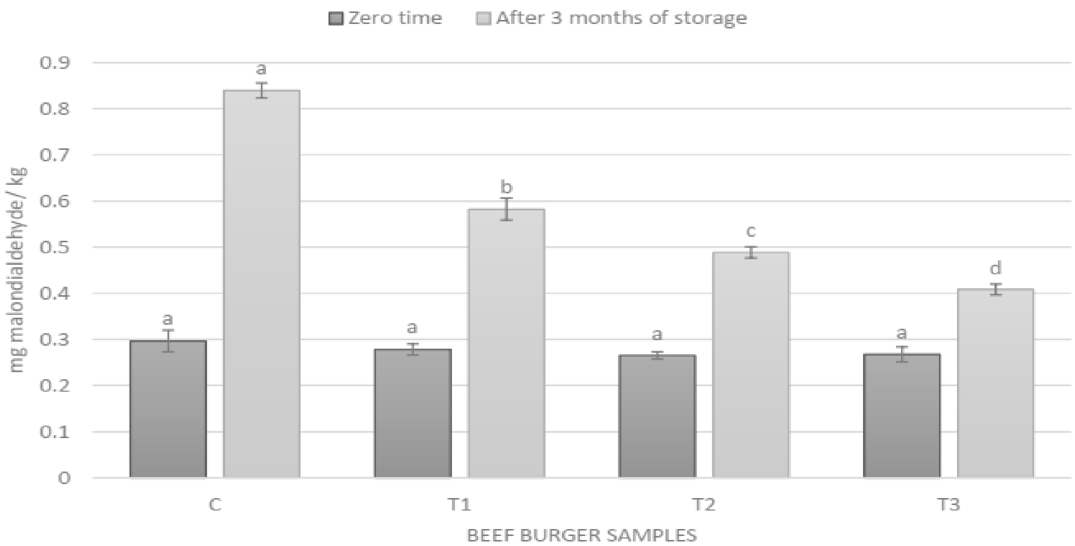


Fig.6: Effect of papaya PP addition on thiobarbituric acid reactive substances (TBARS) values of beef burger at zero time and after storage for three months.

C: control beef burger; T1: beef burger with 1% papaya PP as partial replacement of meat; T2: beef burger with 2% PP as partial replacement of meat; T3: beef burger with 3% PP as partial replacement of meat; [a-d] Means of triplicate ±SD (standard deviation) with different small letters in the same column differ significantly at $p<0.05$.