



EXTRACTION AND EVALUATION OF BIOACTIVE COMPOUNDS FROM SOME FRUIT AND VEGETABLE PEELS

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ABSTRACT

Different research reports have revealed that food industry by-products can be considered as good sources of potentially valuable bioactive compounds. This study was performed to evaluate the antioxidant capacity of the peel extracts of pomegranate (PoP), mango (MaP), prickly-pear (PrP), Cantaloupe (CaP) and pea (PeP) using different solvent polarity (methanol, ethanol, ethylacetate and water) to particular attention to their content of total phenolics and flavonoids. Among all tested extracts, ethanolic extracts of PoP, MaP and PrP and aqueous extract of CaP and PeP were shown to exhibited a significant ($p \leq 0.05$) highest extraction yield, total phenolic and flavonoid contents and antioxidant capacity (measured by DPPH, ABTS and FRAP methods). The antioxidant capacity of different peel extracts was showed the following descending order, MaP > PoP > PrP > CaP ≈ PeP. Also there was a strong positive correlation between the DPPH, ABTS, FRAP values and total phenolic as well as, flavonoid contents of different peels extracts. Seventeen phenolic compounds were identified and quantified by HPLC-UV analysis in different peel extracts. Galic acid was identified as the major compound in MaP and PoP extracts, while it was rutin in the case PrP, CaP and PeP extracts. From the results obtained, fruit and vegetable peels can be considered as good sources of valuable bioactive compounds.

INTRODUCTION

Considerable amount of solid wastes in the form of peels and seeds are generated by the fruit

and vegetable industries, these wastes if not disposed correctly are seen to cause serious environmental problems and economic losses. Recently, it was reported that 39% of food waste is produced by the food manufacturing industries in developed countries. There are several reports highlighting the integral exploitation of bioactive compounds from these wastes and their potential application as antioxidant, antimicrobial, flavoring, colorant, and texturized agents. Peels are the major by-products obtained during the processing of various fruits and these were shown to be a good source of polyphenols, carotenoids and other bioactive compounds which possess various beneficial effects on human health (Mirabella et al 2014 and Roy and Lingampeta 2014).

One of the principal causes of food quality deterioration is lipid peroxidation. Lipid peroxidation results in formation of reactive oxygen species and free radicals; which are purportedly associated with carcinogenesis, mutagenesis, inflammation, DNA changes, aging and cardiovascular diseases. To prevent the lipid peroxidation in fats and oils, synthetic antioxidants (BHA and BHT) have been used as food additives for over 50 years (Shahidi et al 1997).

Nowadays great interest is awarded to natural antioxidants due to problems related to safety and toxicity of synthetic antioxidant. Many natural antioxidants have already been isolated from different plants such as oil seeds, cereals, vegetables, leaves, roots, herbs and spices (Chaouch et al 2015).

Pomegranate peels (*Punica granatum L.*) are considered non-edible parts or by products (50% of fruit weight) obtained during juice processing. It is characterized by the significant presence of polyphenols such as ellagitannins, ellagic acid, gallic acid and flavonoids associated with biological

properties such as antioxidant and antimicrobial agents (Akhtar, 2015; Faria & Calahau, 2010; Glazer et al 2012; Marchi et al 2015; Negi et al 2003)

Major by-product of mango (*Mangifera indica* L.) processing are peels and seeds, amounted of 35 and 60% of the total fruit weight respectively. The peels of mango has a significant potential benefit due to its powerful antioxidant properties and high content of phenolic compounds. The major phenolic compounds of ripe and unripe mango peels are gallic acid, syringic acid, gentsyl-protocatechic, mangiferin, ellagic acid and quercetin (Tunchaiyaphum et al 2013).

Peels of Prickly pear represent a large proportion of the whole fruit (from 40 to 50 %) and constitute a source of bioactive compounds, notably phenolics, flavonoids and betalains (Arrizan et al 2006 and Kuti, 2004). Chougui et al (2015) found that hydro-ethanolic extract of prickly pear peels (*Opuntia-ficus-indica*) was rich in total phenolics (1512.58 mg GAE/100gDM). HPLC analyses allowed the identification of sixteen compounds belonging to hydroxyl benzoic acid, hydroxycinnamic acids and flavonoids.

Hadrich et al (2014) indicated that the peel of pea (*Pisum sativum*) represented a source of active compounds essentially phenols and flavonoids with antioxidant activity. The ability of the peel pea extracts, and BHT as a positive control, to reduce Fe^{3+} to Fe^{2+} was determined. The reducing capacity of peel of pea extracts (ethylacetate, methanol and water) differed significantly in their activity.

Extraction is an important step in the isolation and later in the identification and quantification of phenolic compounds (Cacace and Mazza, 2003). Since the phenolic compounds of different plants differ structurally, it is very difficult to develop a standardized extraction method that would simultaneously extract all inherent phenolic compounds (Nacz and Shahidi, 2006).

Optimization of extraction conditions to yield the maximum level of food grade antioxidant phenolics involves, the type of solvent, or solvent ratio, the particle size of the plant material, the solvent-solid ratio, extraction temperature and time (Akhtar et al 2015).

Methanol, ethanol, or propanol and their mixtures in water as well as the acetone, ethyl acetate and dimethyl from a mixture are so far the most commonly used solvents in the extraction of phenolic compounds from the plant materials (Escribano-Bailon & Santos-Buelga 2003 and Nacz & Shahidi, 2006).

The objective of this study was carried out to determine the content of total phenolics, total flavonoids and antioxidant capacity extracted from peels of pomegranate, mango, prickly pear cactus, by pea, cantaloupe and using five different solvents.

In addition, the phenolic acids profile of these peel extracts were identified using HPLC technique to get insight into the compounds responsible for the antioxidant activity of these by-product.

MATERIALS AND METHODS

Chemicals

Gallic acid, 2,2-diphenyl-1-picryl-hydrazyl (DPPH), 2,2-azino-bis(3-ethyl-benzothiazoline-6-sulfonic acid) (ABTS), 2,4,6-tripyridyl-s-triazine (PTZ), Folin-Ciocalteu reagent solvents for HPLC analysis and the individual phenolic standards were purchased from Sigma-Aldrich, Inc. (Louis, USA). All other chemicals, solvents and reagents used were of analytical grade and the experiment was carried out in triplicate.

Plant materials and sample preparation

Pomegranate (*Punica granatum* L.), mango (*Mangifera indica* L.), prickly pear cactus (*Opuntia ficus-indica*), pea (*Pisum sativum*), cantaloupe (*Cucumis melo*) were obtained from local market, in Cairo, Egypt during 2014/2015

The peels of different materials were manually removed, cut into pieces and dried in an air oven at 55 ± 2 °C for 8 h. The dried sample was grounded using coffee grinder, passed through a 40-mesh sieve and stored in polyethylene bags at -18°C until analysis.

Preparation of plant peel extracts

Bioactive compounds of plant peel samples were extracted using different solvents including (methanol, ethanol, ethyl acetate & their combinations (in 1:1:1) and water). Twenty gram of each peel powder sample were stirred with 100 ml of each solvent for 4 hr at room temperature. The extracts were filtrated through Whatman No 42 filter paper to remove peel particles and dried at 40°C under vacuum. The yield was calculated as grams of extract obtained after drying. Each of the dried peel extract sample was stored in closed vial at 4°C (Roy et al 2014).

Determination of total phenolic content

Total phenolics of different peel extracts were determined according to the method of **Jayaprakasha and Lingam Peta (2001)** using Folin-Ciocalteu reagent. Portions 400 µl of diluted extracts were taken in test tubes; 1.0 ml of Folin-Ciocalteu reagent (diluted 10-fold with distilled water) and 0.8 ml of 7.5% sodium carbonate were added. The tubes were mixed and allowed to stand for 30 min in dark at room temperature before the absorbance was measured at 760nm using Labomed. Inc. spectrophotometer (Spectro UV – VIS Double Beam / UVD-3500) against blank, which contained 400 µl of ethanol in place of sample. The amount of phenolics present in the sample was determined from a standard curve prepared with gallic acid in 95% ethanol. The total phenolic content was expressed as gallic acid equivalents in mg/g dried extract.

Determination of total flavonoid content

Total flavonoids were estimated by using spectrophotometric assay as described by **Kim et al (2003)** with simple modifications. Each extract (50µl) was mixed with water (500µl) and 200µl sodium nitrate (150g/l). After 6min of agitation, 100µl of aluminum chloride (100g/l) was added. Then the mixture was indicated for 5 min., before 1ml of 40 g/l NaOH solution was added. The mixture was diluted to final volume of 2.5 ml with distilled water and mixed thoroughly. The absorbance was measured at 510nm. Catechine was used to make the calibration curve. Total flavonoid content was expressed as mg catechine equivalent (CE) / g dried extract.

Determination of antioxidant activity of different peel extracts

DPPH radical scavenging activity

The capacity of peels extracts to scavenging DPPH was measured based on the method of **Hwang and Do Thi (2014)**. This method depends on the reduction of purple DPPH radicals to yellow colored DPPH and the remaining DPPH radicals that show maximum absorption at 517 nm was measured. Briefly 100 µl of peel extract was allowed to react 3.95 ml of DPPH solution (0.2 mM DPPH in methanol). The decrease in absorbance was determined at 517nm using Labomed. Inc. spectrophotometer, against blank of pure methanol

after 60 min of incubation in a dark condition. Trolox was used to make the calibration curve. The antioxidant activity was expressed as mg trolox equivalent / g extract.

ABTS cation radical-scavenging assay

The stock solutions of ABTS reagent was prepared according to **Hwang and Do Thi (2014)** by reacting equal quantities of a 7mM aqueous solution of ABTS with 2.45 mM potassium persulfate for 16 h at room temperature (25 °C) in the dark. The working solution was then prepared by diluting 1ml ABTS solution with 60 ml of ethanol : water (50:50,v/v) to obtain an absorbance of 1.0 ± 0.02 units at 734 nm. Extracts ((50µl) were allowed to react with 4.95 ml of the ABTS solution for 1h in a dark condition. Then the absorbance was taken at 734nm using the spectrophotometer. Results was expressed as mg Trolox equivalents (TE)/g sample.

Ferric-reducing antioxidant power assay (FRAP)

The FRAP assay was carried out according to the method of **Hwang and Do Thi (2014)**. FRAP reagent was prepared with 25 ml acetate buffer (300 mM, pH 3.6), 2.5 ml 10 mM TPTZ solution in 40 mM HCl, and 2.5 ml of 20mM ferric chloride hexahydrate. FRAP reagent was prepared freshly. The mixture was warmed at 37 °C before use. 50µl of peel extract samples were allowed to react with 3.95 ml FRAP solution for 30 min in a dark condition. Readings of the color product (ferrous tripyridyltriazine complex) were then taken at 593 nm. The standard curve was prepared using trolox. Results was expressed as mg Trolox equivalents (TE)/g sample. Additional dilution was needed if the FRAP value measured was over the linear range of standard.

Analysis of phenolic compounds by HPLC

Phenolic compounds were analysed by HPLC using Agilent Technologies 1100 series liquid chromatograph equipped with an auto sampler and diode-array detector. The analytical column was a Eclipse XDB-C18 (150x 4.6 µm, 5 µm) with a C18 guard column (Agilent, USA). The mobile phase consisted of acetonitrile (solvent A) and 2% acetic acid in water (v/v) (solvent B). The flow rate was kept at 0.8 ml/min for total run time of 70 min and

the gradient program was as follows :100%B to 85%B in 30 min ,85%Bto 50%B in 20 min , 50%B to 0% B in 5 min .Extract were prepared as mentioned previously and were evaporated at 40°C using a rotary evaporator. The injection volume was 50µl and peaks were monitored simultaneously at 280 and 320 nm for the benzoic acid and cinnamic acid derivatives, respectively. All samples were filtered through a. 45 Acrodisc syringe filter (Gelman Laboratory, MI) before injection . Peaks were identified and compared with those standars. (Yahia and Mondragon, 2011).

Statistical analysis

All samples were analyzed in triplicates and the results were expressed as means \pm standard deviation (SD). Analysis was assessed using the Statical Analysis System Software System for windows (SAS, 2008). One way analysis of variance (ANOVA) was performed to evaluate the significant differences between sample means, with significant level being considered at $p < 0.05$. Means comparisons were assessed by Duncan 's test.

RESULTS AND DISCUSSION

Extraction of bioactive compounds from pomegranate, mango, prickly pear, cantaloupe and pea

peels using different solvents was comparative with antioxidant activity of the extracts as follows

Extraction yield

Table (1) represents the yield values obtained from fruit and vegetable peels extracted with solvents of a different polarity. Depending on the extraction solvent, yield % significantly ($p < 0.05$) differed among peel extracts .The yield % of different solvents varied from 34.86 to 50.37%, 24.28-40.57%, 24.22-33.86%,12.42-20.21% and 11.01-25.05% from the extracts of (PoP), (MaP), (PrP), (CaP) and (PeP) respectively. Among all tested extracts, it appears that ethanolic extract showed the highest yield from PoP, MaP and PrP, while water extract exhibited the most yield from CaP and PeP.

This result is in a contrast with those obtained by **Ismail et al (2010)**, **Li et al (2006)** and **Marchi et al (2015)** for cantaloupe skin, and **Shiban et al (2012)** for pomegranate peels. Whereas, it differed from **Hardich et al (2014)** who proved that methanol is the most suitable solvent for extraction of bioactive compounds from peel of pea. Moreover, ethanol and water are safer and therefore more suitable for the food industry than the other organic solvents (**Huh et al 2004**).

Table 1. Yield (%w/w) of different solvent extracts from fruit and vegetable peels

Peels Extracts	Pomegranate (PoP)	Mango (MaP)	Prickly pear (PrP)	Cantaloupe (CaP)	Pea (PeP)
Methanol(M)	47.95 ^{AB} ±2.0	34.52 ^B ±1.5	30.74 ^{AB} ±2.0	13.24 ^B ±0.45	15.22 ^B ±0.0
Ethanol(E)	50.37 ^A ±1.45	40.57 ^A ±0.0	33.86 ^A ±0.0	17.51 ^A ±1.12	18.54 ^B ±0.24
Ethyl acetate (EA)	45.61 ^B ±2.5	28.16 ^C ±1.84	29.31 ^B ±1.48	12.42 ^B ±2.0	11.01 ^C ±1.11
M:E:EA (ratio1:1:1)	47.62 ^{AB} ±0.0	34.28 ^B ±0.04	31.94 ^A ±1.11	16.25 ^{AB} ±1.5	16.27 ^B ±1.56
Water (W)	34.86 ^C ±1.55	24.28 ^C ±0.85	24.22 ^B ±1.18	20.21 ^A ±0.12	25.05 ^A ±0.45

Values (mean±S.D.) with different letters within the same column are significantly different ($p \leq 0.05$)

Total phenolic and flavonoid contents

Several studies have revealed that the phenolic content in the plants are associated with their antioxidant activities, probably due to their redox properties, which allow them to act as reducing agent, hydrogen donors and singlet oxygen quenchers (Chang et al 2001). Flavonoids are the most common and widely distributed group of phenolic compounds, which usually are very effective antioxidants (Yanish lieva-Maslarova, 2001).

It can be noticed from Tables (2) and (3) that total phenolic (TP) and flavonoid (TF) contents of different peel extracts were significantly ($p \leq 0.05$) differed depending on the peel source and solvent

type. Based on the solvent system, ethanolic extracts of PoP, MaP and PrP and water extract of CaP and PeP exhibited the highest levels of TP and TF contents. However, among the different tested peel samples ethanolic extracts of PoP and MaP recorded the highest TP (93.77 and 181.65 mg gallic acid /g) and TF (17.57 and 14.71 mg catechine/g) respectively. These results were accordance with the results of extraction yield. Similar results were obtained by Negi et al (2003) and Shibani et al (2012) for pomegranate peels, Tunchaiyaphum et al (2013) for mango peels, Abdel-Hameed et al (2014) for prickly pear peels, Ismail et al (2010) for cantaloupe skin and Hardish et al (2014) for peel of pea.

Table 2. Total phenolic (TP) content (mg gallic acid/g) of different extracts from some fruit and vegetable peels

Peels Extracts	Pomegranate (PoP)	Mango (MaP)	Prickly pear (PrP)	Cantaloupe (CaP)	Pea (PeP)
Methanol(M)	81.35 ^F ±0.03	134.99 ^B ±3.09	30.15 ^{HIJ} ±0.98	30.83 ^{HIJ} ±0.10	24.33 ^{KIJ} ±0.31
Ethanol(E)	93.77 ^D ±2.06	181.65 ^A ±2.6	61.62 ^G ±1.6	15.00 ^K ±0.15	27.02 ^{HIJ} ±0.1
Ethyl acetate(EA)	87.13 ^E ±5.6	123.75 ^C ±1.03	24.69 ^{IKJ} ±0.36	32.12 ^{HI} ±0.00	20.38 ^{KJ} ±0.41
M:E:EA (ratio1:1:1)	76.71 ^F ±1.5	124.70 ^C ±2.06	33.23 ^{HI} ±0.15	25.52 ^{HIJK} ±0.36	26.19 ^{HIJ} ±0.20
Water (W)	52.47 ^H ±4.11	35.51 ^{H±} 0.05	32.63 ^{HI} ±1.03	33.64 ^{HI} ±1.03	48.45 ^G ±0.46

Values (mean±S.D.) with different letters within the same column and row are significantly different ($p \leq 0.05$)

Table 3. Total flavonoid (TP) content (mg catechine/g) of different extracts from some fruit and vegetable peels

Peels Extracts	Pomegranate (PoP)	Mango (MaP)	Prickly pear (PrP)	Cantaloupe (CaP)	Pea (PeP)
Methanol(M)	17.24 ^A ±0.03	10.39 ^C ±0.16	0.82 ^G ±0.12	1.70 ^G ±0.03	6.78 ^D ±0.18
Ethanol(E)	17.57 ^A ±2.06	14.71 ^B ±0.00	0.99 ^G ±0.03	2.27 ^G ±0.06	6.58 ^D ±0.13
Ethyl acetate(EA)	13.75 ^B ±5.6	13.56 ^B ±1.43	0.96 ^G ±0.03	1.66 ^G ±0.00	6.94 ^D ±0.13
M:E:EA (ratio1:1:1)	13.41 ^B ±1.5	13.74 ^B ±0.23	1.25 ^{ED} ±0.04	2.38 ^{GF} ±1.02	6.17 ^G ±0.07
Water (W)	2.07 ^G ±4.11	4.32 ^{EF} ±0.48	0.57 ^G ±0.04	9.82 ^C ±0.00	7.08 ^D ±0.16

Values (mean±S.D.) with different letters within the same column and row are significantly different ($p \leq 0.05$)

Antioxidant activity

The antioxidant capacity of different fruit and vegetable peels extracts measured by the use of ABTS, FRAP and DPPH assays are presented in **Table (4)**. Overall comparison of different solvents showed ethanol, methanol, ethylacetate and their mixture extracts of PoP and MaP exhibited significant ($p<0.05$) stronger activity(DPPH and ABTS values), while water extract of the same peels showed lower activity. Similar results were obtained with FRAP assay for PoP and MaP extracts. Prickly pear (PrP) extracted with ethanol and mixture of methanol, ethanol and ethylacetate had significantly ($p<0.05$) higher antioxidant activity than those extracted with water. On the contrary, water extract of CaP and PeP significantly ($p<0.05$) exhibited higher antioxidant activity compared to other tested solvents (**Table 4**).

Among all tested extracts ethanolic extracts exhibited the strongest antioxidant capacity measured by ABTS,FRAP and DPPH assays, being 243.05,357.9 and 257.38 mg torloex /g PoP, 315.5, 319.5 and 343.02 mg torolex /g MaP, and 199.8, 222.08 and 209.70 mg torolex /g PrP respectively. On the other hand, aqueous extracts of CaP and PeP recorded higher antioxidant activity measured by ABTS,FRAP and DPPH being 164.9,211.4 and 182.5,215.3 mg torolex /g PeP and 195.2 and 200.4 mg torelex /g CaP respectively. In conclusion, the antioxidant capacity of different peels extracts was showed the following depending order; mango> pomegranate>prickly pear (ethanolic extract) > cantaloupe≈ pea (water extract).

Previously, **Li et al (2006)** found extraction of pomegranate peel with a mixture of ethanol and acetone had a higher antioxidant capacity than pulp extract. Also, **Negi et al (2003)**, found that acetone, ethyl acetate and methanol peel extracts of pomegranate exhibited a higher antioxidant capacity, but the water extract was the lowest. **Abdel-Hameed et al (2014)** showed that the juices of peels and pulps of red cactus exhibited a higher reducing power activity (149.49 and 123.23 mg ascorbic acid equivalent/100ml juice respectively. Our results are in contrast with the reports of **Lee et al (2007)** and **Hadrich et al (2014)** showed that water is the most suitable solvent for extraction of phenolic compounds from the peel of pea (*pisum sativum*). **Jahural et al (2015)** reported that mango peel extracts were proven to be a good source of polyphenols, anthocyanins and carotenoids and

thus, they may be used in nutraceutical and functional foods as a source of antioxidant agents.

Table 4. Antioxidant activity of different solvent extracts obtained from some fruit and vegetable peels

Solvents	Antioxidant assay		
	ABTS mg Trolox/g	FRAP mg Trolox/g	DPPH mg Trolox/g
Pomegranate peel			
Methanol(M)	229.9 ^A ±29.57	314.5 ^B ±3.4	237.70 ^A ±14.1
Ethanol(E)	243.05 ^A ±9.20	357.9 ^A ±13.4	257.38 ^A ±6.08
Ethyl acetate(EA)	231.64 ^A ±21.9	344.5 ^A ±4.1	245.30 ^A ±11.12
M:E:EA (ratio1:1:1)	239.8 ^A ±15.8	335.7 ^{AB} ±1.06	228.3 ^A ±18.1
Water (W)	162.9 ^B ±19.34	202.6 ^C ±4.11	128.9 ^B ±1.2
Mango peel			
Methanol(M)	282.2 ^B ±2.0	389.7 ^A ±2.5	324.8 ^B ±1.2
Ethanol(E)	315.5 ^A ±17.4	391.5 ^A ±4.3	343.02 ^A ±2.4
Ethyl acetate(EA)	306.8 ^{AB} ±4.7	383.2 ^{AB} ±12.4	325.8 ^B ±1.8
M:E:EA (ratio1:1:1)	302.3 ^{AB} ±5.3	367.02 ^B ±3.5	321.7 ^B ±3.0
Water (W)	129.19 ^C ±6.1	156.9 ^C ±3.22	226.6 ^C ±0.6
Prickly – pear peel			
Methanol(M)	146.8 ^C ±5.5	136.22 ^C ±13.4	208.5 ^A ±7.2
Ethanol(E)	199.8 ^A ±1.5	222.08 ^A ±17.5	209.7 ^A ±2.4
Ethyl acetate(EA)	146.6 ^C ±8.1	129.15 ^D ±9.06	178.7 ^B ±0.6
M:E:EA (ratio1:1:1)	165.34 ^B ±10.02	178.15 ^B ±7.9	101.09 ^D ±9.02
Water (W)	116.34 ^D ±12.5	139.9 ^C ±5.6	138.9 ^C ±0.00
Cantaloupe peel			
Methanol(M)	147.7 ^B ±13.2	151.8 ^B ±11.7	148.4 ^C ±2.4
Ethanol(E)	135.6 ^B ±9.5	148.4 ^B ±12.6	175.18 ^{AB} ±7.8
Ethyl acetate(EA)	117.5 ^C ±11.04	127.4 ^B ±6.2	156.8 ^B ±14.4
M:E:EA (ratio1:1:1)	140.9 ^B ±2.5	194.8 ^A ±13.8	172.9 ^{AB} ±1.8
Water (W)	182.5 ^A ±15.2	215.3 ^A ±11.6	200.4 ^A ±1.8
Pea peel			
Methanol(M)	124.8 ^{AB} ±6.2	158.6 ^B ±1.9	155.9 ^C ±1.2
Ethanol(E)	83.7 ^B ±11.8	100.3 ^C ±10.02	109.8 ^D ±0.00
Ethyl acetate(EA)	110.8 ^{AB} ±14.9	131.7 ^{BC} ±11.9	165.4 ^B ±3.6
M:E:EA (ratio1:1:1)	88.11 ^B ±1.4	165.3 ^B ±9.7	110.9 ^D ±0.00
Water (W)	164.9 ^A ±12.02	211.4 ^A ±8.4	195.2 ^A ±0.6

Values (mean±S.D.) with different letters within the same column for each peel waste extract are significantly different ($p\leq0.05$)

Correlation between antioxidant capacity and total phenolic and flavonoid contents

The correlation between total phenolic, flavonoid contents and antioxidant capacity of different peel extracts were calculated and the results are given in **Table (5)**.

Table 5. Correlation coefficient between antioxidant activity total phenolic and flavonoids contents of different peel extracts

Trait	Flavonoid	phenol	ABTS	FRAP	DPPH
Flavonoid	1.00	0.73	0.71	0.82	0.61
phenol	0.73	1.00	0.85	0.89	0.82
ABTS	0.71	0.85	1.00	0.80	0.78
FRAP	0.82	0.89	0.80	1.00	0.75
DPPH	0.61	0.82	0.78	0.75	1.00

A strong linear relationships could be observed between the total phenolic content and antioxidant capacity of different peels extracts ($R^2 = 0.82, 0.89$ and 0.85 for DPPH, FRAP and ABTS values, respectively). Also, there was a positive linear correlation between the total flavonoids and antioxidant capacity of the peel extracts ($R^2 = 0.61, 0.82$ and 0.71 for DPPH, FRAP and ABTS values, respectively). The correlations between the results of antioxidant activity and the total phenolic and flavonoid contents indicated that polyphenol compounds largely contributed to the antioxidant capacities of the selected fruit and vegetable peels. The positive linear relationships between the total phenolic contents and antioxidant capacity were in accordance with the result of other researches (**Chen et al 2014; Fu et al 2011 and Song et al 2010**).

Comparisons were also evaluated between the results obtained by the DPPH, FRAP and ABTS assays. A high correlation was observed between FRAP and ABTS values ($R^2 = 0.80$), ABTS and DPPH values ($R^2 = 0.78$) and FRAP and DPPH values ($R^2 = 0.746$), which suggested that the com-

pounds capable of reducing oxidants also could scavenge free radicals in these peels extracts. Similar results were obtained by **Chen et al (2014)**. Also, there was a high correlation between total phenolic and flavonoids contents of different peels extracts ($R^2 = 0.73$).

Identification and quantification of phenolic compounds of different peel extracts

The ethanolic extracts of PoP, MaP and PrP and aqueous extracts of CaP and PeP were subjected to HPLC analysis so as to investigate the types of phenolic compounds responsible for the antioxidant activity of these peel extracts.

In this study, seventeen phenolic compounds were identified and quantified in the peel samples extracts by comparing the HPLC chromatograms of them with the HPLC chromatograms of standard compounds based on the retention at the same conditions (**Table 6**).

As seen from **Table (6)**, rutin, coumarin and ferulic acid were identified in all tested peel extracts, whereas, gentisic, chlorogenic acid, syringic acid, vanillic acid and chrysin were identified in only one sample of different peel extracts.

Gallic acid was identified as the major compounds in MaP (95.04%) and PoP (65.73%) extracts, while, it was rutin in the case of PrP, CaP and PeP extracts. Also, it is clearly noticed that MaP extract exhibited the highest amount of gallic acid (11739.35 $\mu\text{g/g}$) followed by PoP extract (7818.80 $\mu\text{g/g}$) and CaP extract (123.33 $\mu\text{g/g}$) On the other hand, CaP extract contained the most amount of rutin (2007.17 $\mu\text{g/g}$) followed by PrP and PeP extracts (1263.04 and 309.23 $\mu\text{g/g}$ respectively).

HPLC analysis showed that the content of phenolic compounds of the peel wastes extracts have the following order, mango>pomegranate> cantaloupe>prickly-pear and pea. This finding is supported by the fact that the ethanolic extract of MaP and PoP which has shown the best antioxidant activity in the present study has more content of phenolic compounds (mainly gallic acid) as seen in **Table (6)**.

Table 6. Quantity of phenolic compounds ($\mu\text{g/g}$) identified by HPLC analysis of different peel extracts

Compounds	Rt (min)	Peel extracts of				
		pomegranate	Mango	Prickly -pear	cantaloupe	Pea
Gallic acid	5.5	7818.801	11739.354	0.000	123.331	0.00
protocatechuic acid	9.5	220.687	238.541	48.367	0.000	35.196
gentisic acid	16.5	0.000	0.000	27.666	0.000	0.000
catechine	17.9	682.248	119.104	0.000	0.000	141.218
chlorogenic acid	20.0	0.000	26.805	0.000	0.000	0.000
caffeic acid	20.6	11.916	0.000	0.000	731.056	4.272
syngic acid	22.1	142.697	0.000	0.000	0.000	0.000
vanillic acid	23.7	0.000	0.000	0.000	137.080	0.000
Rutin	35.8	205.284	58.240	1263.043	2007.169	309.230
coumarin	36.7	293.773	24.696	49.316	75.664	50.917
ferulic acid	31.5	792.468	50.941	11.724	2.435	1.484
sinapic acid	33.1	501.825	85.588	13.360	5.154	0.000
rosmarinic acid	40.0	651.772	0.000	55.305	19.178	6.160
cinnamic acid	42.5	0.000	9.096	13.736	18.935	0.865
Quercetin	43.2	274.705	0.000	23.440	0.000	5.404
Kaempferol	46.1	42.464	0.000	0.000	0.000	3.356
chrysin	51.5	245.451	0.000	0.000	0.000	0.000
Total		11884.08	12352.36	1505.96	3120.01	558.10

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