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Research article

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Optimizing Bioactive Substances Extraction Procedures from Guava, Olive and Potato Processing Wastes and Evaluating their Antioxidant Capacity

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Abstract

Food wastes valorization has been employed dramatically at different fields due to their fine and functional components. This study is aiming at optimization of bioactive substances extraction conditions from guava, olive and potato processing wastes. After collecting, the solvent extraction technique was applied using different solvents and drying methods. Then, the bioactive substances, antioxidant capacity and antimicrobial activity were determined. Subsequently, methanol had peaked solvents either olive or potato wastes of extractable bioactive substances. Conversely, acetone was the better for both guava wastes. Hence, they also exhibited the highest scavenging activity against $DPPH^{\bullet}$ and $ABTS^{\bullet+}$ free radicals. Total phenolic compounds were interrelated with antioxidant activity than other bioactive substances. Both olive wastes and guava pomace displayed greater antioxidant and antimicrobial activities than other wastes. Also, the food wastes dried using oven-drying was recommended. Formerly, it could be useful as antioxidant and antimicrobial agents in food and drug industries.

Keywords

Food processing wastes, Extraction conditions, Antioxidant activity, Bioactive substances, Antimicrobial activity

Abbreviation

AOA: Antioxidant activity; GSE: Guava seeds extract; GPE: Guava pomace extract; OLE: Olive leaves extract; OPE: Olive pomace extracts; PPRE: Potato peels russet extract; PPH: Potato peels hermus extract, TPC: Total phenolic compounds; TF: Total flavonoids

Introduction

Food processing wastes cause solicitous problems world widely. Due to its equal 39% of total food losses [1]. Unquestionably, they occasioned from different fruits and vegetables after them converting to processed forms. However, they deemed as attractive sources of bioactive substances featured with valuable human health benefits [2, 3]. Guava (*Psidium guajava* L.) wastes have upper bioactive substances than twelve tropical fruits wastes, for instance [4]. Also, they are ranged within the limits 30% [5]. Another example, olive (*Olea europaea* L.) processing wastes are respectable sources for bioactive substances [6]. Especially they are remained with huge quantities after olive oil processing up to 70% of olive waste [7] and 10% of olive leaves [8]. However, Egypt is ranked globally as the first olive in quantity per hectare to be 97.88 Hg Ha^{-1} [9]. Tangibly, potato (*Solanum tuberosum* L.) is the fourth largest crop grown all over the world and

the foremost foods at more than 100 countries [10]. Whilst, potato peels (3-5%) are the major wastes of potato processing industries [11]. It is provided an excellent source for bioactive substances [12].

Generally, polar organic solvents are the most effective in bioactive substances solubilizing from plant tissues [13]. However, there are fluctuations at previous work about optimizing conditions for bioactive substances extraction. Acetone was mentioned before as a preferring solvent [14, 15]. Whereas, methanol was recommended for bioactive substances extraction [16]. At the same time, ethanol was suggested by some author for the same purposes [17], for instance. Therefore, the present study has been undertaken with the objective of comparison between four solvents (acetone, ethanol, methanol and water) in term of bioactive substances extraction. Also, the effect of the drying methods on yield obtained, total phenolic compounds (TPC), total flavonoids (TF) and flavonols will be assessed. Moreover, their antioxidant activity (AOA) by DPPH$^{\bullet}$ and ABTS$^{\bullet+}$ assays as well as antimicrobial activity will be evaluated. Furthermore, the correlation between these components individually and their AOA using Pearson correlation and regression factor will be realized.

Materials and Methods

Food processing wastes

a. Guava seeds and pomaces were obtained from Cairo for Agricultural Processing Co., industrial zone, El-Obour City, Egypt.

b. Olive leaves and pomaces Kronakii variety were obtained from Cairo for Oil Industry, industrial zone, 6th October City, Egypt.

c. Potato peels Hermus and Russet varieties were obtained from Egypt Foods Co., industrial zone, Quesna City, Egypt.

Food processing wastes preparation

Each waste was divided into two portions after removing the unsymmetrical parties. Then it was dried by both oven-dryers (Tit Axon S.R.L via Canova, Italy) at 40-50 °C gradually for 12 h and solar-dryers (locally made) at ~38-40 °C for 72 h till the constant weight. The dried wastes were milled (Severin, type 3871, Germany). The powder was passed through a 60 mesh sieve to obtain homogenous powder then kept at -18 ± 1 °C until use after packaging in dark glass jars.

Bioactive substances extraction

Experimentation, the effect of acetone, ethanol, methanol as (80%) and water on bioactive substances extraction from each waste was inspected. Guava seeds and olive pomace were defatted by *n*-hexane as (1:5, w/v) for 1 h before extraction. Each dried waste was mixed with solvent as (1:20, w/v) individually in dark bottles. The bottles were agitated (MLM Zentrifugenbau.TS21, Germany) for 24 h. The mixture was filtered through filter paper Whatman No.1. The filtrates were collected, then solvents were removed by rotary evaporator (Vacuum evaporator NE-1-Rikakikai Co., LTD, Japan) at 40 °C. They were lyophilized (CHRIST, ALPHA 1-4D plus, Germany) then kept at -18±1 °C until further uses according to Lafka et al. [18] with some modification.

Determination of total phenolic compounds

In brief, 200 µL of each extract was mixed with 1 mL of 10-fold diluted Folin–Ciocalteu reagent (Fluka Co., France). After 5 min the reaction stopping by 1 mL of 7.5 g 100mL^{-1} Na_2CO_3 and 1.5 mL distilled water was added also. The mixture was incubated in the dark for 60 min then the absorbance by (CE599- Automatic Scanning Spectrophotometer, GECIL, England) at 760 nm was measured according to Abaza et al. [19]. The TPC was expressed as milligrams of standard curve from gallic acid (Serva, fine Biochemical, New york) equivalents (mg of GAE 100 g^{-1}dw) using the following equation based on the calibration curve:

$$Y = 0.0201\ x + 0.0538 \qquad (R^2 = 0.99) \ldots\ldots\ldots\ldots\ldots\ldots (1)$$

Where: Y is the concentration and x is the absorbance.

Determination of total flavonoids

A 0.5 mL aliquot of 2 g 100mL^{-1} $AlCl_3$ ethanolic solution was added to 0.5 mL of extracts and mixed well. Then, they were kept for 1 h at room temperature and the absorbance at 420 nm was measured. As for flavones, A 5.0 g 100mL^{-1} sodium acetate solution were added then mixed well and kept for 2.5 h at room temperature. The absorbance at 440 nm was taken according to Mohdaly et al. [20]. The final concentration was expressed as quercetin (Sigma Aldrich, Germany) equivalents (mg QEg^{-1}dw) using the following equation based on the calibration curve:

$$Y = 0.037x+0.1363 \qquad (R^2 = 0.98) \ldots\ldots\ldots\ldots\ldots\ldots (2)$$

Where: Y is the concentration and x is the absorbance.

DPPH$^{\bullet}$ radical scavenging activity

A 0.1 mL from each extract and BHT 100 mgkg^{-1} were added to 3.9 mL DPPH$^{\bullet}$ (Sigma Aldrich, Germany) methanolic solution. Formerly, the absorbance at 517 nm was measured after the solution allowing to stand in the dark for 60 min according to Lafka et al. [18]. The final results were expressed as micromoles of Trolox equivalents per gram of dry weight (μmol TE g^{-1}dw) and as % inhibition using the following equation based on the calibration curve:

$$AOA\ (\%) = [(Ac_{517}-As_{517})/Ac_{517}] \times 100 \qquad \ldots\ldots\ldots\ldots\ldots (3)$$

Where: Ac_{517} is the absorbance of the blank and As_{517} is the absorbance of the extracts or BHT.

ABTS$^{\bullet+}$ radical cation scavenging activity

ABTS$^{\bullet+}$ radicals was produced by reacting ABTS$^{\bullet+}$ (Osaka, Japan) stock solution with 2.45 mmol L^{-1} potassium per-sulfate (final concentration) and allowing the mixture to stand in the dark at room temperature for 12-16 h before using. The ABTS$^{\bullet+}$ radicals' solution was diluted with ethanol to an absorbance of 0.70 ± 0.02 at 734 nm and equilibrated at 30 °C as well as measured at 734 nm according to Lu et al. [21]. The final results were expressed as (μmol TE g^{-1}dw) and

% inhibition using the following equation:

$$AOA\ (\%) = [(Ac_{734}-As_{734})/Ac_{734}] \times 100 \quad \ldots\ldots\ldots\ldots (4)$$

Where: Ac_{734} is control absorbance and As_{734} is extracts or BHT absorbance.

Evaluation of antimicrobial activity against some food pathogenic and spoilage strains

Determination of antibacterial activity

The screening of the antibacterial activity of tested extracts was performed using agar disc diffusion assay as described by Kotzekidou et al. [22]. The bacterial strains (*Bacillus cereus, Escherichia coli O157, Listeria monocytogenes, Salmonella typhi, S. typhimurium, Staphylococcus aureus* and *Yersinia enterocolitis*) from Institut für Gärungsgewerbe, Berlin, Germany were propagated. A loop full from each strain was added into Mueller Hinton Broth (Himedia, India) then incubated at 37 °C for 12 h. Appropriate volume from each culture was mixed with sterilized Mueller Hinton Agar to set an inoculums as ~10^{-6} cell mL^{-1}, then poured in sterilized Petri dishes. Consequently, the extracts were sterilized by 0.45 µm filters (Minisart®, Germany). Sterile filter paper discs 6 mm were immersed into sterilized extracts for 5 s then put immediately onto the solid cultures surface'. The plates were incubated at 37 °C for 24-48 h and the inhibition zones around discs were measured.

Determination of antifungal activity

The effect of wastes extracts on mycelia yield of selected fungi were investigated at (1, 2.5 and 5%) concentrations according to Tripathi et al. [23]. Potato dextrose broth (Himedia, India) was prepared in 50 mL Erlenmeyer flasks and inoculated with 10^5 spore mL^{-1} of (*Aspergillus niger, Alternaria alternata, Penicillium chrysogenum* and *Rhizopus stolonifer*). An equal amount of distilled water was added in the corresponding control (0% extracts). The flasks were incubated at 28 ± 1 °C with 120 rpm shaking. After 5 days, flasks containing mycelia were filtered and washed through filter paper Whatman No.1. The mycelia were allowed to dry at 60 °C for 6 h then at 40 °C overnight. The growth inhibition percentage was calculated as:

$$\text{Growth inhibition } \% = [(DW_{utf}-DW_{etf})/DW_{utf}] \times 100 \ldots (5)$$

Where: DW_{utf}: dry weight of untreated fungal strain and DW_{etf}: dry weight of treated fungal strain with extract.

Statistical analysis

The statistical analysis was carried out using SPSS program (ver. 19) with multi-function utility regarding to the experimental design under significance level of 0.05 for the whole results and multiple comparisons were carried out applying LSD according to Steel et al. [24]. Moreover, Pearson's correlation analysis was calculated and obtained correlation results were compared to critical values of Pearson's *r* table under levels of significance with two-tailed test.

Results and Discussions

Effect of extraction conditions on yield obtained and total phenolic compounds of guava, olive and potato wastes extracts

Regularly, both olive wastes had peaked in their content of yield obtained and TPC. It was followed by guava, while the potato extracts had bottomed (Table 1). For that reason, olive leaves extract (OLE) had pointed, conversely, either potato peels russet extract (PPRE) or potato peels hermus extract (PPHE) had bottomed. Likewise, it was preceded by guava seeds extract (GSE), guava pomace extract (GPE) and olive pomace extract (OPE), respectively. Indeed, oven-drying is

Table 1: Yield obtained and total phenolic contents of oven- and solar-dried guava, olive and potato wastes extracts using different solvents (Mean±SD), *n*=3.

Extracts	Item	Solvent / drying methods								Mean ±SD
		Acetone		Ethanol		Methanol		Water		
		OD	SD	OD	SD	OD	SD	OD	SD	
Guava pomace	YO*	120.45±1.03	103.17±2.76	98.79±1.19	90.42±0.97	107.33±0.89	98.37±1.39	39.95±0.25	29.67±1.51	86.01±31.43^{d}
	TPC**	134.83±1.38	99.98±2.06	109.91±1.39	70.59±2.17	124.16±1.30	80.59±1.24	5.64±0.33	3.51±0.32	78.65±48.06^{c}
Guava seeds	YO*	69.41±0.13	46.55±1.02	45.40±13.4	36.98±1.38	59.26±1.0	40.35±1.21	29.88±0.72	18.41±0.54	43.28±15.83^{c}
	TPC**	82.56±0.93	66.54±0.80	67.20±1.69	55.35±0.77	76.60±1.80	61.44±1.24	5.18±1.42	1.57±0.21	52.05±29.83^{b}
Olive leaves	YO*	261.43±0.88	239.32±0.78	280.33±0.92	251.25±1.10	299.43±3.19	273.15±2.30	89.38±0.81	60.93±1.88	219.40±87.14^{f}
	TPC**	276.04±4.56	115.52±4.98	349.70±17.91	303.11±3.24	387.08±6.68	313.21±2.05	99.99±3.75	78.31±0.97	240.37±117.49^{e}
Olive pomace	YO*	167.94±1.69	151.19±1.0	188.04±1.39	163.05±2.10	200.75±3.03	181.32±0.64	60.11±1.55	48.19±1.86	145.07±55.67^{e}
	TPC**	81.04±10.64	67.84±1.92	104.85±9.17	75.48±1.76	371.93±10.04	198.62±10.02	66.25±3.74	28.70±3.73	124.33±106.80^{d}
Potato peel Hermus	YO*	29.98±0.55	21.39±0.99	35.78±1.51	23.25±0.99	41.06±1.62	30.23±0.86	13.50±3.23	8.50±0.80	25.46±10.57^{a}
	TPC**	4.81±0.04	0.49±0.07	5.13±0.14	0.70±0.02	9.48±0.49	0.94±0.05	0.94±0.48	0.19±0.04	2.83±3.18^{a}
Potato peel Russet	YO*	34.57±2.01	23.33±1.23	40.19±0.91	30.51±1.09	46.43±1.40	38.07±1.07	18.37±1.31	13.26±1.13	30.59±10.99^{b}
	TPC**	6.73±0.44	0.90±0.07	7.60±0.37	1.24±0.14	12.00±0.58	2.50±0.35	0.92±0.04	0.35±0.04	4.03±4.07^{a}

*Yield obtained that measured as g Kg^{-1}dw: see materials and methods section,
** Total phenolics compounds that measured as mg GAE 100 g^{-1}dw, OD: Oven-drying, SD: Solar drying,
$^{a, b, c, \ldots}$: Means with the same letter in the same column are not significantly different (p>0.05).

the heightened manner for yield obtained and TPC isolation than solar-drying as mentioned before for other tissues [25]. A significant difference (p<0.05) among solvents was observed, and this matter is varied according to the solvent polarity [13]. Expressively, methanol had peaked (117.97 g kg^{-1}dw). Whilst, the water was the bottommost solvent in yield obtained and TPC being 35.84 and 24.29 mg GAE 100 g^{-1}dw, respectively. Methanol was resulted the maximum bioactive substances either olive or potato wastes. Whilst, the highest quantities from these components both guava wastes were attained using acetone as illustrated in Table 1 and Figure 1A. These results are agreement with Mohamed, Mohdaly and Jimenez et al. [14, 20, 26]. However, no results established about extraction optimization of these components from guava pomace.

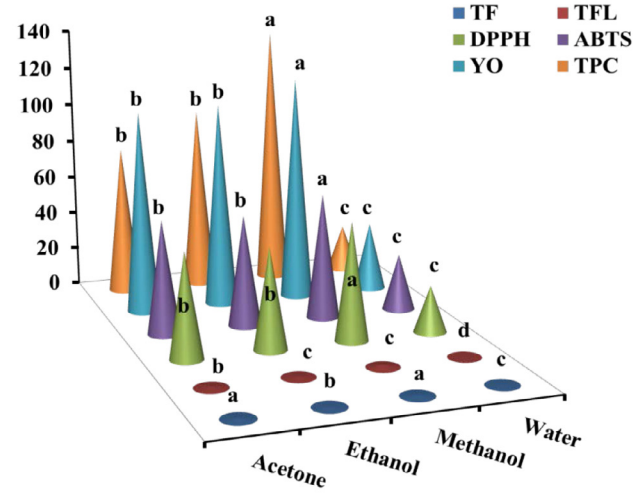

Figure 1A: Efficiency of different solvents in bioactive substances extraction from guava, potato and olive waste extract. (Mean±SD), *n=3*.

Determination of total flavonoids and flavonols compounds of guava, olive and potato processing wastes

Statistically, the significant differences (p<0.05) between both drying methods in order of TF and flavonols contents was initiated. Equally, a significant difference (p<0.05) in TF and flavonols substances was observed clearly among wastes (Table 2). Amazingly, the greater such components were taken out using acetone rather than other solvents both guava wastes. On opposite finding, olive and potato wastes methanolic extracts exhibited the highest TF and flavonols than other solvents as portrayed in Figure 1A. In the same table, water was the poorest solvent in TF and flavonols extraction except the waste kind. The OPE had pointed, whilst PPHE had bottomed from their contents of TF and flavonols. These results are in agreement with previous studies of Brahmi and Mohdaly et al. [27, 28], but are lower than published results by Abaza et al. [19]. However, no results found about such components either GPE or OPE as well extraction conditions.

Determination of antioxidant activity of guava, olive and potato processing wastes

DPPH· radical scavenging activity

In the present work, the AOA of tested extracts was evaluated *in vitro* compared with BHT. Table 3 symbolized that the AOA differed significantly among solvents. Exclusively, the methanolic extracts were the highest AOA. It was monitored by acetonic and ethanolic extracts. The efficiency of AOA using different solvents was varying. For example, guava wastes acetonic extracts were the highest AOA, whereas, olive and potato wastes methanolic were the uppermost AOA compared with other solvents. Regardless drying methods or solvents, olive wastes extracts had peaked, whereas potato wastes had bottomed. Undoubtedly, oven-drying had lower influences against AOA than solar-drying a rounding 59.83 and 39.21%, respectively (Figure 1B). To emphasize that, GPE exhibited greater AOA than GSE. Consequently, OLE was higher AOA than OPE. Similarly, PPRE was higher AOA than PPHE. Kui-Hua et al. [29] reported that a yellow skinned potato variety had lower AOA than the red skinned potato. The obtained data were evident that the BHT recorded AOA higher than GSE, PPHE and PPRE extracts and lower than OLE, OPE and GPE extracts in radical inhibition. Thus, OLE, OPE and GPE can be used a good natural AOA in food and/or stuffs. These results are agreement with the results published by Lu and Brahmi et al. [21, 27].

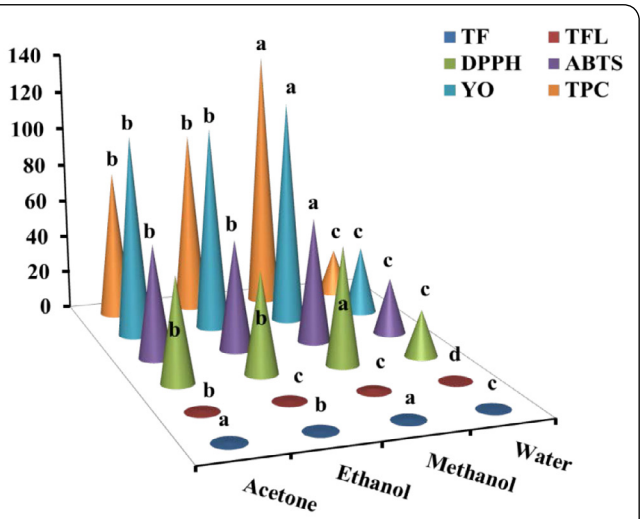

Figure 1B: Changes of bioactive substances between oven and solar-dried guava, olive and potato waste extract.(Mean±SD), *n=3*.

$ABTS^{\bullet+}$ radical cation activity

$ABTS^{\bullet+}$ was the second method used for AOA evaluation as tabulated in Table 4. The AOA had totally and significantly (p<0.05) differed among extracts regarding solvents. Expectation the drying methods, methanol and ethanol solvent exited the highest AOA, except both guava wastes which had peaked using acetone. Conversely, the water extracts exhibited lower AOA than all wastes both drying methods. Olive wastes extracts recorded the highest extracts in AOA. Conversely, potato wastes listed the lowest extracts regardless solvents or drying methods. Also, the maximum AOA was noticed with oven-dried OLE methanolic extract's reaching 99.91%, while the bottommost AOA was attained with solar-dried PPHE water extract's to be 5.74%. The obtained data manifested that the BHT scored higher AOA than GSE, PPHE and PPRE. Whilst it was recorded lower AOA than GPE, OPE and

Table 2: Total flavonoids and total flavonols compounds of oven- and solar-dried guava, olive and potato wastes extracts extracted using different solvents (Mean±SD), *n*=3.

Extracts	Item	Solvent/ drying methods								Mean ±SD
		Acetone		Ethanol		Methanol		Water		
		OD	SD	OD	SD	OD	SD	OD	SD	
Guava pomace	TF*	0.33±0.0	0.21±0.0	0.20±0.0	0.08±0.0	0.28±0.04	0.15±0.01	0.10±0.01	--	0.16±0.10^{c}
	TFL**	0.21±0.01	0.19±0.01	0.11±0.0	0.08±0.0	0.17±0.0	0.14±0.0	0.04±0.01	0.01±0.01	0.11±0.06^{c}
Guava seeds	TF*	0.38±0.02	0.26±0.02	0.21±0.02	0.09±0.02	0.34±0.02	0.22±0.02	0.18±0.01	0.07±0.01	0.21±0.10^{d}
	TFL**	0.25±0.04	0.21±0.02	0.13±0.01	0.10±0.01	0.20±0.01	0.17±0.01	0.03±0.0	--	0.13±0.08^{d}
Olive leaves	TF*	1.56±0.05	1.11±0.05	1.22±0.02	0.76±0.01	1.80±0.01	1.34±0.02	0.73±0.03	0.30±0.02	1.10±0.46^{e}
	TFL**	2.68±0.02	1.57±0.02	2.54±0.01	1.41±0.01	4.27±0.02	2.88±0.02	0.45±0.02	0.36±0.01	2.01±1.26^{e}
Olive pomace	TF*	1.79±0.02	1.35±0.02	1.24±0.07	0.85±0.02	1.88±0.06	1.44±0.06	0.89±0.03	0.47±0.02	1.23±0.46^{f}
	TFL**	2.95±0.02	1.85±0.01	2.79±0.02	1.68±0.02	4.30±0.02	3.20±0.02	0.41±0.01	0.30±0.01	2.18±1.32^{f}
Potato peel Hermus	TF*	0.08±0.01	--	0.18±0.01	0.07±0.01	0.22±0.01	0.10±0.01	--	--	0.08±0.08^{a}
	TFL**	0.04±0.0	--	0.02±0.0	--	0.12±0.0	0.09±0.01	--	--	0.03±0.04^{a}
Potato peel Russet	TF*	0.18±0.01	0.06±0.01	0.26±0.01	0.14±0.01	0.29±0.01	0.17±0.01	--	--	0.13±0.10^{b}
	TFL**	0.05±0.01	0.02±0.01	0.04±0.0	0.01±0.0	0.15±0.01	0.11±0.01	0.01±0.01	--	0.04±0.05^{b}

* Total flavonoids that measured as mg QEg^{-1}dw: see materials and methods section,

** Total flavonols that measured as mg QEg^{-1}dw, OD: Oven-drying, SD: Solar drying,

a, b, c,....: Means with the same letter in the same column are not significantly different (p>0.05).

Table 3: Effect of different solvents and drying methods on antioxidant activity of guava, olive and potato wastes extracts (Mean±SD), *n*=3.

Extracts	DPPH▪	Solvent/ drying methods								Mean ±SD
		Acetone		Ethanol		Methanol		Water		
		OD	SD	OD	SD	OD	SD	OD	SD	
Guava pomace	IP*	90.04±1.19	53.65±1.27	70.95±0.72	37.36±0.72	79.04±0.61	47.13±1.16	12.43±1.19	8.51±1.11	49.88±28.44^{d}
	TE**	16.24±0.26	8.11±0.27	12.08±0.16	4.64±0.15	13.84±0.13	6.72±0.25	--	--	7.70±5.81^{d}
Guava seeds	IP*	72.74±1.26	49.26±0.98	62.75±0.93	19.45±1.35	69.83±0.46	40.75±0.84	10.47±2.08	1.65±0.97	40.86±26.37^{c}
	TE**	12.47±0.28	7.34±0.21	10.29±0.20	0.84±0.29	11.83±0.10	5.49±0.18	--	--	6.03±5.04^{c}
Olive leaves	IP*	91.58±0.92	68.72±0.84	97.26±0.57	77.75±1.68	99.36±0.56	86.48±1.60	67.14±0.63	58.94±1.11	80.90±14.39^{f}
	TE**	16.05±0.45	11.78±0.38	17.93±0.13	13.65±0.37	18.39±0.12	15.56±0.35	11.32±0.14	9.52±0.24	14.27±3.09^{f}
Olive pomace	IP*	90.80±1.07	64.58±1.22	95.58±1.15	70.39±1.15	98.8±0.72	83.4±1.29	60.79±0.83	52.39±0.83	77.09±16.64^{e}
	TE**	16.73±0.41	11.49±0.50	17.94±0.26	12.29±0.26	18.66±0.16	15.21±0.29	10.14±0.19	8.25±0.19	13.84±3.67^{e}
Potato peel Hermus	IP*	14.87±1.07	7.21±0.96	20.57±0.72	11.61±1.15	30.34±1.16	19.31±1.60	5.43±0.67	2.02±0.83	13.92±8.94^{a}
	TE**	0.57±0.08	--	1.08±0.16	--	3.20±0.25	0.86±0.37	--	--	0.714±1.05^{a}
Potato peel Russet	IP*	28.00±1.45	19.73±1.04	36.8±1.15	26.45±0.97	48.14±1.37	32.47±0.72	10.41±0.83	2.02±0.83	25.50±14.05^{b}
	TE**	2.98±0.14	0.98±0.04	4.61±0.25	2.52±0.23	7.07±0.30	3.92±0.17	--	--	2.75±2.34^{b}
BHT▪▪	IP*	77.41±0.01								
	TE**	12.77±0.02								

▪ Antioxidant activity was determined by DPPH• method and calculated by two ways,

* Inhibitions percentage as (%), ** Trolox equivalent µmol TE g^{-1}dw calculated by certain equation,

▪▪ Butylated hydroxytoluene (100 ppm), OD: Oven-drying, SD: Solar drying, -- Not detected,

a, b, c,....: Means with the same letter in the same column are not significantly different (p>0.05).

OLE around 82.49%. These results are in agreement with the results published by Brahmi et al. [27]. However, until now there are no previous studies to estimate the AOA for guava wastes using ABTS•+ method.

Pearson correlation coefficient between bioactive substances of guava, olive and potato wastes extracts and their antioxidant activity

Obviously, the correlation between yield obtained and TPC was higher than its correlation with TF or flavonols

Table 4: Antioxidant activity of oven- and solar-dried guava, olive and potato wastes extracts measured using $ABTS^{\bullet+}$ method (Mean±SD), *n*=3.

Extracts	ABTS▪	Solvent/ drying methods								Mean ±SD
		Acetone		Ethanol		Methanol		Water		
		OD	SD	OD	SD	OD	SD	OD	SD	
Guava pomace	IP*	95.43±0.62	58.07±1.29	73.90±0.79	43.15±0.98	84.08±0.29	51.56±0.83	16.24±0.28	14.2±1.05	54.57±28.36[d]
	TE**	17.42±0.14	9.27±0.28	12.72±0.18	6.01±0.22	14.94±0.06	7.85±0.18	0.14±0.06	--	8.54±6.13[d]
Guava seeds	IP*	74.36±0.49	56.81±0.95	70.37±0.95	29.22±0.41	72.86±0.14	44.86±0.87	17.01±1.96	6.78±0.72	46.53±25.28[c]
	TE**	12.82±0.11	8.99±0.21	11.95±0.21	2.97±0.09	12.49±0.03	6.39±0.19	0.31±0.43	--	6.98±5.14[c]
Olive leaves	IP*	96.97±0.34	71.42±1.49	98.46±0.44	83.86±0.49	99.91±0.08	88.33±1.16	71.42±1.49	65.26±1.16	84.45±13.12[f]
	TE**	17.75±0.08	12.18±0.33	18.08±0.10	14.89±0.11	18.39±0.02	15.87±0.26	12.18±0.33	10.84±0.25	15.02±2.86[f]
Olive pomace	IP*	93.44±0.88	71.05±0.77	98.37±0.36	74.13±0.82	99.09±0.21	87.47±0.93	66.53±1.16	61.2±0.75	81.41±14.33[e]
	TE**	16.98±0.19	12.10±0.17	18.06±0.08	12.77±0.18	18.22±0.05	15.68±0.20	11.11±0.25	9.95±0.16	14.35±3.12[e]
Potato peel Hermus	IP*	20.44±1.02	12.30±1.70	29.49±0.82	17.28±0.82	35.37±1.02	22.03±0.68	8.23±1.02	5.74±0.68	18.86±9.78[a]
	TE**	1.06±0.22	--	3.03±0.18	0.36±0.18	4.31±0.22	1.40±0.15	--	--	1.27±1.54[a]
Potato peel Russet	IP*	34.06±0.75	23.88±1.38	43.19±1.63	32.11±0.75	57.53±2.18	39.35±1.03	15.24±0.89	6.38±0.55	31.46±15.51[b]
	TE**	4.03±0.16	1.81±0.3	6.02±0.36	3.60±0.17	9.15±0.47	5.31±0.23	0.04±0.07	--	3.74±2.99[b]
BHT▪▪	IP*	82.49±0.14								
	TE**	13.81±0.01								

▪ Antioxidant activity was determined by $ABTS^{\bullet+}$ method and calculated by two ways,

* Inhibitions percentage as (%), ** Trolox equivalent μmol TE g^{-1}dw calculated by certain equation,

▪▪ Butylated hydroxytoluene (100 ppm), OD: Oven-drying, SD: Solar drying, -- Not detected,

[a, b, c,...]: Means with the same letter in the same column are not significantly different (p>0.05).

as presented in Table 5. This may be due to the difference of chemical composition between phenolic and flavonoids extractability [30]. Also the correlation between yield obtained and AOA against $DPPH^{\bullet}$ or $ABTS^{\bullet+}$ had pointed compared with (TPC, TF, flavonols) and AOA. Due to the yield obtained included TPC, TF and flavonols but collectively. Surely, the highest correlation was observed between $DPPH^{\bullet}$ and $ABTS^{\bullet+}$ due to the mechanism are similar as mentioned before [31]. From the previous data, TPC was the major components in yield obtained. Thus, the regression between TPC and AOA towards $DPPH^{\bullet}$ and $ABTS^{\bullet+}$ assay was presented in Figure 2. A high content of TPC led to high AOA in each extract. For example, AOA was posted the highest incidence to be 84.45% when TPC rating 240.37 mg GAE 100 g^{-1}dw. In contrast the ratio of their decreased to the lowest levels when the TPC was decreased.

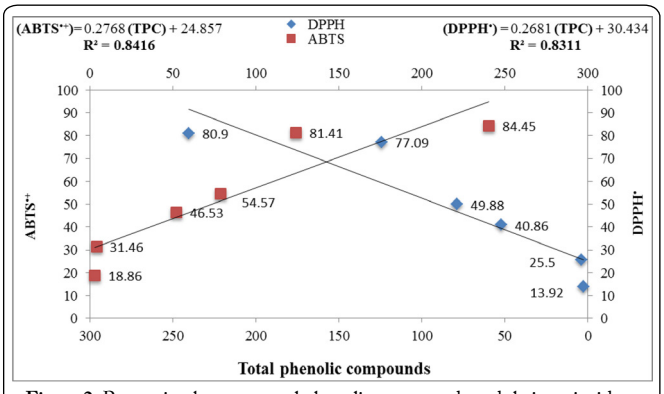

Figure 2: Regression between total phenolic compounds and their antioxidant activity of selected waste extracts.(Mean±SD), *n=3*.

Table 5: Pearson's correlation coefficients of bioactive substances and their antioxidant activity in guava, olive and potato extracts.

	Bioactive substances					
	YO (g kg^{-1} dw)	TPC (mg g^{-1} dw)	TF (mg QE g^{-1} dw)	TFL (mg QE g^{-1} dw)	$DPPH^{\bullet}$ (μmol TE g^{-1} dw)	$ABTS^{\bullet+}$ (μmol TE g^{-1} dw)
YO	1	0.92***	0.87***	0.89***	0.82***	0.81***
TPC		1	0.80***	0.84***	0.78***	0.76***
TF			1	0.95***	0.76***	0.75***
TFL				1	0.70***	0.68**
$DPPH^{\bullet}$					1	0.99***
$ABTS^{\bullet+}$						1

Asterisks (**, ***) represent a significant difference at (P<0.01, P<0.001), respectively.

Evaluation of the antimicrobial activities for guava, olive and potato wastes extracts

Antibacterial activity

According the aforementioned results, the best extracts for each waste was preferred. Then, its antibacterial activity *in vitro* against seven bacterial strains was studied; data were summarized in Table 6. Noticeably, OLE exhibited the highest antibacterial activity in different levels. It was followed by OPE and GPE. In contract, PPRE and GSE showed the lowest impacts against the same pathogenic strains. OLE was greater significantly (p<0.05) than OPE. Whilst, no significant differences (p>0.05) were evident between OPE and GPE.

Table 6: Effect of guava, olive and potato wastes extracts on the growth inhibition of some food pathogenic bacterial strains (Mean±SD), *n*=3.

Extracts▪	Strains / Inhibition zone (mm)						
	B. cereus	*E. coli* O157	*L. monocytogenes*	*S. typhi*	*S. Typhmurium*	*Staph. aureus*	*Yersinia enterocolitis.*
GPE	10.00 ± 0.0^{cA}	9.50 ± 0.71^{bB}	9.00 ± 0.0^{aB}	10.50 ± 0.71^{dB}	9.50 ± 0.71^{bB}	10.00 ± 0.0^{cB}	10.00 ± 0.0^{cC}
GSE	11.00 ± 0.0^{dB}	7.50 ± 0.71^{aA}	8.50 ± 0.71^{bA}	12.00 ± 0.0^{eC}	7.50 ± 0.71^{aA}	10.00 ± 0.0^{cB}	12.00 ± 0.0^{eD}
OLE	11.00 ± 0.0^{aB}	15.50 ± 0.71^{dE}	17.00 ± 5.66^{eE}	20.50 ± 0.71^{gE}	14.00 ± 1.41^{cD}	13.00 ± 0.0^{bC}	20.00 ± 0.0^{fF}
OPE	12.00 ± 0.0^{bC}	11.00 ± 0.0^{aD}	25.00 ± 0.0^{gF}	16.00 ± 0.0^{eD}	13.00 ± 1.41^{cC}	20.50 ± 0.71^{fD}	15.00 ± 0.0^{dE}
PPHE	12.50 ± 0.71^{cD}	10.00 ± 0.0^{bC}	13.00 ± 0.0^{dD}	10.00 ± 0.0^{bA}	--	13.00 ± 0.0^{dC}	7.50 ± 0.71^{aA}
PPRE	10.00 ± 0.0^{cA}	10.00 ± 0.0^{cC}	10.00 ± 0.0^{cC}	10.50 ± 0.71^{dB}	--	8.50 ± 0.71^{bA}	8.00 ± 0.0^{aB}

▪ Oven-dried wastes extracts and antibiotic disc, --Not detected.

$^{a,b,c,...}$: Means with the same letter in the same row are not significant different (p>0.05),

$^{A,B,C,...}$: Means with the same letter in the same column are not significant different (p>0.05).

Generally, the highest inhibition against *E. coli* O157, *S. typhi, S. typhimurium* and *Yersinia enterocolitis* was achieved by OLE. Specifically, OPE displayed the highest activity against *L. monocytogenes* approximately 25.00 mm. PPHE and PPRE didn't effect on *S. typhimurium*. Nevertheless, PPHE showed the uppermost effect against *B. cereus* to be 12.50 mm. the phenolic compounds could have an inhibiting effect on microbial growth according to their constitutions and concentrations [32, 33]. However, few studies were reported the potential antibacterial of GPE, GSE and OEL [17, 34]. Unfortunately, no available data about antibacterial of both potatoes peel and OPE were established.

Antifungal activity

Normally, OPE and OLE was upper inhibition than other extracts. It was trailed by GPE and GSE as rendered in Figure 3. Regardless the fungi strains species, the mean value of OPE and OLE inhibition at 5% was around (22.20 and 18.87%), respectively. Alternatively, the same concentration of PPHE had bottomed against fungal growth to be 3.72%. A significant difference (p<0.05) was found between each extract (2.5 and 5%) once. GPE affected the tested fungal strains higher than GSE in different concentrations. GSE and GPE had peaked (5%) against *A. alternata* about (13.10% and 15.05% inhibition), respectively. OLE (5%) was subdued significantly (p<0.05) fungal strains growth than OPE with the equal concentration. To the best of our knowledge, there is no publication was found about this issue.

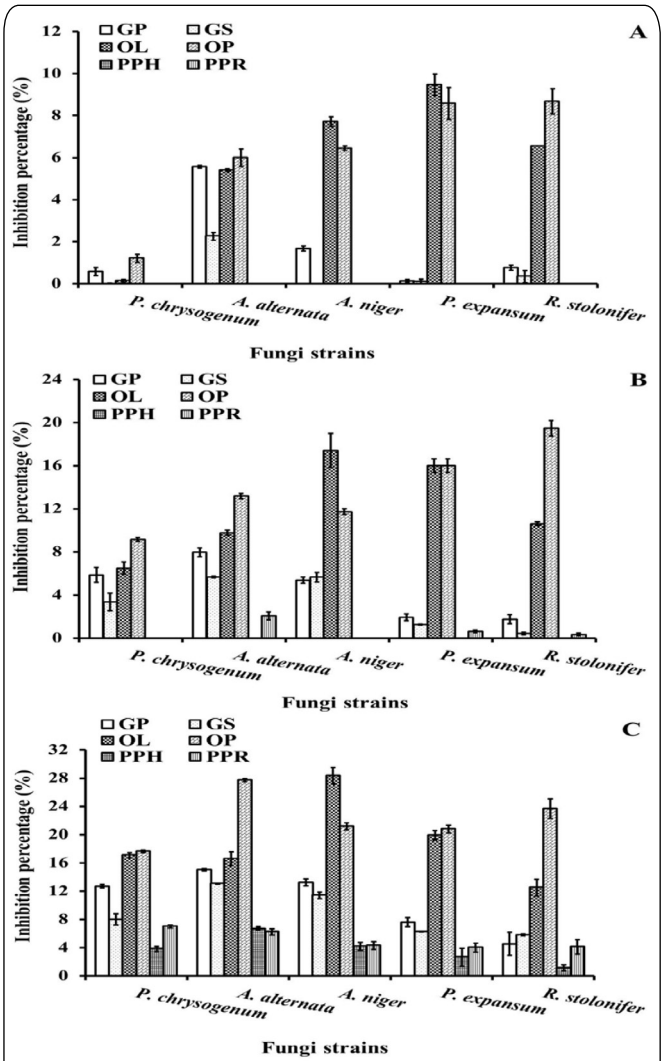

Figure 3: Evaluation of antifungal activity for guava, olive and potato wastes extracts against some spoilage fungal strains at different concentrations 1% (A), 2.5% (B) and 5% (C). (Mean±SD), *n=3*.

Conclusion

A successful and innovative comparison between four familiar solvents and tow drying methods were carried out. Methanol was the best solvents in olive and potato processing wastes according to their content of yield obtained, TPC, TF, flavonols and AOA. Conversely, acetone was the best solvents for both guava wastes, while water recorded lower bioactive substances and AOA than all solvents among extracts. Also,

oven-drying led to obtain a high content of these components compared to solar-drying. Noteworthy, OLE, OPE and GPE may be used as antioxidants agent rather than BHT when they extracts were taken using the recommended conditions.

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