

Review of water-based extraction techniques affecting the antioxidant activity of pomegranate peel

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Article history:

Received: 16 April 2022

Received in revised form: 29 August 2022

Accepted: 31 December 2023

Available Online: 3 January 2025

Keywords:

Agriculture waste,
Bioactive,
Extraction,
Water extract,
Fruit peel

DOI:

[https://doi.org/10.26656/fr.2017.9\(1\).206](https://doi.org/10.26656/fr.2017.9(1).206)

Abstract

The demand in the agricultural sector is on the rise, which is indirectly linked to the accumulation of agricultural wastes via post-harvest operation, transportation, wholesale, retail, storage, processing and packaging, which occurs in the food supply chain has been one of the major challenges faced globally. Agricultural wastes contain diverse types of bioactive compounds, which can be further utilised for the enhancement of food values. In the present review, several water-based extractions were discussed including maceration, digestion, infusion, decoction, percolation, Soxhlet, hydrodistillation, steam-distillation, ultrasound-assisted and microwave-assisted extraction. The comparisons were based on the polyphenol content and antioxidant activities focussing on pomegranate peel (POMEPE). This review discussed various polyphenol compounds found in POMEPE (ellagic acid and punicalagin) that contributed to antioxidant activities. It was found that the acceptable limit of radical scavenging activities of POMEPE was 65% in water-based extractions, respectively. POMEPE revealed that water-based maceration, digestion, infusion, decoction, ultrasound-assisted and microwave-assisted extraction showed acceptable antioxidant activities. Therefore, water-based extractions portrayed great potential to be assimilated into the market based on their antioxidant potential and their non-toxic and environment-friendly characteristics. This review is important to food industries and stakeholders to further explore the possibilities of utilising fruit peels as functional food products.

1. Introduction

In recent years, there has been ascending demand in the agriculture industry as people have widely realised the importance of fruit and vegetable consumption. This realisation is a joint effort between each country's governmental policy, the World Health Organisation (WHO), the Food and Agriculture Organisation (FAO) and also the mission of the United Nations. Furthermore, fruits and vegetables rising in demand, which will eventually increase the production rate; and export and import value as well, consequently improving the economic level. Eventually, it also leads to the agriculture sector's growth. Department of Statistics Malaysia (DOSM) enclosed that the agriculture sector has contributed RM 101.5 billion which covered 7.1% of the Gross Domestic Product (GDP) in 2019 (DOSM, 2019).

However, along with the agriculture sector's growth, there are also consequences in agriculture waste accumulation. In 2019, FAO highlighted that 14% of the world's food was lost in terms of quality and quantity at the stage of post-harvest until it reached the retail level. Meanwhile, FAO (2019) also revealed that Central and Southern Asia had the highest food loss index with 20.7%. The lowest food loss index was recorded in Australia and New Zealand with 5.8% (FAO, 2019). Meanwhile, approximately 15% of municipal solid waste is contributed by crop waste (Agamuthu, 2009). This revealed that agro-waste loss had a huge proportion in waste generation. It was revealed that the global pomegranate market value in 2018 was USD 8.2 billion. Hence, by the year 2026, the global pomegranate market value will be USD 23.14 billion at a Compound Annual Growth Rate (CAGR) of 14.0% (Conidi *et al.*, 2020).

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It has been estimated that 998 million tonnes of agricultural waste are generated annually (Obi *et al.*, 2016). The spike in waste accumulation was up to 100.75% in the year 2018 (38,142 tonnes) as compared to the year 2005 (19,000 tonnes) (Muzamir, 2020). Meanwhile, 80% of the waste was contributed by unused parts of fruits and vegetables during food preparation, whereas the other proportion (20%) was contributed by leftover food generated by the consumer (Zulkipli and Dawum, 2016). Furthermore, Panzella *et al.* (2020) highlighted that the annual global food waste generation is approximately 1.3 billion tons which accumulated at primary (farm) and secondary (production plant) process levels including the food supply chain. Meanwhile, focusing on Central and Southern Asia, the agriculture waste mostly yielded from agricultural food and crop loss throughout transportation (25%), wholesale (15%) and retail; compared to on-farm post-harvest operation (7.7%), storage (5.91%) and processing and packaging (0.25%) level. This issue becomes a steppingstone for researchers to identify the potentials associated with agriculture wastes, particularly on crop wastes or agro-wastes such as peel, seed, leaf and stalk in order to reduce the severity of waste accumulation. Pomegranate peel (POMEPE) is the waste that are commonly found in the processing plant, hypermarket (pre-cut fruit) and household levels. A wide range of studies showed the potential of POMEPE to be utilised in food, beverages, poultry supplement, human supplement, industrial usage and medical purposes (Naguib and Tantawy, 2019; Vella *et al.*, 2019; Yap *et al.*, 2021). Excellent sources of diverse polyphenols made up of phenolic acid, flavonoids, tannins and stilbenes contributed to the significant impact of POMEPE to be utilised as functional ingredients (Ali *et al.*, 2014; Dabesor *et al.*, 2017; Naguib and Tantawy, 2019).

Although extensive research had been carried out on agriculture waste potential, there were limited reviews or statistical trends of each of the fruits' waste generated published. Instead, the proportion of fruits became an indicator towards its contribution to agriculture waste accumulation as illustrated in Table 1. It was revealed that peel's waste played a role in the waste accumulation

of pomegranate (Table 1). The rising demand for high fibre and minerals drinks has broadened the fruit juice market and eventually contributed to the rise of agricultural waste generated as well (Gunwantrao *et al.*, 2016). Despite its huge impact on the country's financial and economy, agriculture waste also has the potential to develop unpleasant odour, soil pollution, serious environmental pollution and harbourage of pests or insects. Hence, stern action needs to be taken towards agriculture recycling. The utilisation of these wastes remains a high challenge to food industries, scientists and government to create zero waste technology for the agricultural sector.

Thus, polyphenol content and antioxidant potential were reviewed by comparing varieties of water-based extraction on the selected fruit peels such as POMEPE extracts. The extraction includes water-based maceration extraction (WME), water-based digestion extraction (WDiE), water-based infusion extraction (WIE), water-based decoction extraction (WDE), water-based percolation extraction (WPE), water-based Soxhlet extraction (WSE), water-based hydrodistillation extraction (WHDE), water-based steam-distillation extraction (WSDE), water-based ultrasound-assisted extraction (WUAE) and water-based microwave-assisted extraction (WMAE).

2. Extraction techniques

Water-based conventional extraction technique is the recovery of bioactive compounds from plant matrices using conventional solvents (water) based on mass-transfer or diffusion phenomena such as WME, WDiE, WIE, WDE, WPE, WSE, WHDE and WSDE (Hussain *et al.*, 2019; Rocchetti *et al.*, 2019). In contrast, Rocchetti *et al.* (2019) indicated that water-based non-conventional extraction techniques involved certain energy inputs or processing aids in enhancing the extraction efficiency and selectivity while offering superior extraction efficiency in terms of yield, cost, selectivity and extraction time using water as the solution or based such as WUAE and WMAE. Nevertheless, based on the studies previously conducted, it was found that the

Table 1. Fruit waste proportion.

Type of fruit	Peel (%)	Other waste (%)	Reference(s)
Pomegranate	50% ^f	N.D.	Mansour <i>et al.</i> (2013) ^f ; Malviya <i>et al.</i> (2014) ^g ; Kharchoufi <i>et al.</i> (2018) ^h Sharayei <i>et al.</i> (2019) ⁱ Kaderides <i>et al.</i> (2019) ^j Balli <i>et al.</i> (2020) ^l
	60% ^g		
	40-50% ^h		
	26-30% ⁱ		
	40% ^j		
	40-50% ^k		
	41.6- 68% ^l		

Different superscripts indicated different sources of journal.

N.D. = No Data

compounds extracted from aqueous-based extraction also have its own potential. Although it does not as high as organic solvent, but it is safer and environment friendly. On the other hand, there were tremendous studies highlighted that aqueous solution has undoubted potential and some possess only small differences or no significant difference with other organic solvents in term of antioxidant activity.

Recently, Hüseyinbaş *et al.* (2020) highlighted that aqueous solution is the greenest solvent for being non-corrosive, non-toxic, naturally abundant, environmentally stable and low-cost solvent. The factors influencing the aqueous solution's capacity to influence solvent efficiency depend on the extraction's duration and temperature. Table 2 illustrates variations in duration and temperature which affect the polyphenol content in the crude extract. The study conducted among POMEF revealed that extraction's duration and temperature played huge roles in determining aqueous-based extraction's quality. However, based on the real world, aqueous-based extraction is more crucial and highly utilised in food or animal feeding including dairy cow feed as highlighted by Abarghuei *et al.* (2013).

Maceration is conducted by immersing the sample into the solvent and keeping it at ambient temperature (15-25°C) for at least 24 hrs in a covered or sealed container as affirmed by Saravanabavan *et al.* (2020). However, it is a time-consuming extraction technique as it commonly takes up to three to seven days duration to complete the extraction process (Zengin *et al.*, 2019; Saravanabavan *et al.*, 2020). The extracted sample was filtrated and undergoes lyophilisation. Digestion extraction is known as gentle extraction as described by Zengin *et al.* (2019). This method is identical to maceration extraction, but the temperature involved is slightly different (30-60°C). Digestion of the extraction utilised medium or gentle heat in the extraction process and commonly takes several hours ranging from one hour to 24 hrs. Infusion extraction methods involving macerating the sample in hot aqueous solution (61-99°C) for short period of time (at least 20 mins). However, there were also studies that used infusion extraction for several hours ranging from 20 mins to 24 hrs (Saravanabavan *et al.*, 2020). Percolation; the solvent will be poured into the percolator in downward displacement and the solvent passes through the beds of the sample to retrieve compound of interest. As for Soxhlet extraction, the extraction begins with the vapourisation of the mixture which the collected in the porous thimble, once the condensed sample is full in the porous thimble area, it will be sent back into the flask through the siphon. This cycle continued until the extraction was completed. Low boiling point solvent is

commonly used for this method in order to reduce the time of the extraction as boiling temperature is required and to preserve the bioactive compounds of the sample (Ferrentino *et al.*, 2018). Hydrodistillation or steam distillation extraction technique is low cost and requires simple instrumentation (Sharayei *et al.*, 2019). Besides, the oil retrieved is free from organic solvents. However, the high temperature (100°C) used in hydrodistillation extraction has its consequences. Besides the long extraction time taken, high energy consumption and products' thermal degradation are the significant drawbacks of hydrodistillation extraction as highlighted by Dangkulwanich and Charaslertrangsi (2020).

Sharayei *et al.* (2019) highlighted that ultrasound-assisted extraction (UAE) has shorter operation time, reduced solvent consumption, low temperature usage, easy operation and energy saver compared to conventional extraction. It was mentioned that UAE required equipment that has the capability to transmit sonic waves in the solvent to extract the compounds from the sample. Afterwards, the sample left for a time ranging from minutes to hours to cool down before filtration and lyophilisation. Table 2 shows that POMEF had great phenolic yield in aqueous-based UAE (WUAE). In simpler words, Boggia *et al.* (2016) stated that UAE used ultrasound energy to disrupt the plant's cell wall and consequently, intracellular liquids started to escape from the cell and dissolved in the solvent. Despite the expensive equipment and required highly skilled operators, UAE only required low temperatures to extract secondary metabolites from plant samples. Microwave-assisted extraction (MAE) has been identified as a promising extraction technique with short time, low solvent consumption and high yield of the interest compounds (Kaderides *et al.*, 2019). The association of microwave energy or irradiation transmitted through the sample has initiated molecular interactions with the electromagnetic field. Boggia *et al.* (2016) reported that the mass and heat gradient initiated the synergistic combination that accelerated the extraction process and at the same time yield high interest compounds. There are several variables in MAE that influence the extraction yield. This includes microwave output power, extraction time and liquid-solid ratio. Xueling *et al.* (2011) indicated that microwave output power has great influence towards phenolic yield. For instance, the curvilinear increase of phenolic yield was identified until it reached 80% of output power when the maximum power supplied was 750 W. On the other hand, Table 2 shows several variables of MAE that influence the phenolic content of the plant sample. There were numerous studies which revealed the potential of aqueous-based MAE (WMAE) in POMEF. Furthermore, it was asserted by Alias and

Table 2. Aqueous-based extraction yields by the variation of variables.

Extraction	Variable (s)	Sample	Phenolic content	Reference(s)
WME	25°C, 1 hr	POMEPE	23 mg GAE/g DW to 25 mg GAE/g DW	Jalal <i>et al.</i> (2018)
	100°C, 1 hr	POMEPE	α -punicalagins and β -punicalagins: 13 mg/g to 105 mg/g	Balli <i>et al.</i> (2020)
	55°C, 4 hrs	POMEPE	274.08 mg GAE/g DW	Kharchoufi <i>et al.</i> (2018)
	75°C, 4 hrs	POMEPE	286.58 mg GAE/g DW	Kharchoufi <i>et al.</i> (2018)
	25°C, 12 hrs	POMEPE	242.05±7.99 mg GAE/g DW	Kennas and Amellal-Chibane (2019)
	25°C, 24 hrs	POMEPE	1.9 mg QE/100 g DW	Karthikeyan and Vidya (2019)
	3 mins	POMEPE	542.5 mg GAE /g DW	Rongai <i>et al.</i> (2017)
WUAE	20 kHz 400 W	POMEPE	10.7 - 76.8 mg GAE/g DW	Sharayei <i>et al.</i> (2019)
	200 W; 26 kHz	POMEPE	100 - 200 mg GAE/g DW	Turrini <i>et al.</i> (2020)
	400 W	POMEPE	α -punicalagin = 562.26±47.14 mg/L β -punicalagin = 1,251.13±22.21 mg/L	Tamborlin <i>et al.</i> (2020)
	30 kHz; 10 mins	POMEPE	15 - 20 mg GAE/g DW	Hüseyinbaş <i>et al.</i> (2020)
	59 kHz; 60 mins; 200 W	POMEPE	180.5 mg GAE/ g DW	Aboelsoued <i>et al.</i> (2019)
WMAE	470 W; 10 mins	POMEPE	12.38±0.32 mg GAE/100 g DW	Vladić <i>et al.</i> (2020)
	800 W	POMEPE	13.34±0.20 mg GAE/100 g DW	Vladić <i>et al.</i> (2020)
	2000 W; 90 s	POMEPE	66.56±0.37 mg GAE/g DW	Skenderidis <i>et al.</i> (2020)
	4000 W; 90 s	POMEPE	95.52±0.54 mg GAE/g DW	Skenderidis <i>et al.</i> (2020)
	6000 W; 90 s	POMEPE	95.52±0.64 mg GAE/g DW	Skenderidis <i>et al.</i> (2020)
	470 W	POMEPE	17.11±0.26 mg GAE/100 g DW	Vladić <i>et al.</i> (2020)

POMEPE: Pomegranate peel, WME: Water-based maceration extraction, WDiE: Water-based digestion extraction, WIE: Water-based infusion extraction, WDE: Water-based decoction extraction, WPE: Water-based percolation extraction, WSE: Water-based Soxhlet extraction, WHDE: Water-based hydrodistillation extraction, WSDE: Water-based steam-distillation extraction, WUAE: Water-based ultrasound-assisted extraction, WMAE: Water-based microwave-assisted extraction (WMAE), Values expressed as mean±standard deviation.

Abbas (2017) that microwave power had a linear relationship with phenolic compounds ($R^2 = 0.97$). Vladić *et al.* (2020) highlighted that MAE was proven to be an efficient extraction method as it required short extraction time, low solvent consumption, indirect heat source, rapid energy transfer, selective heating and suitable for thermolabile compounds. Although MAE is a non-conventional extraction that has great potential in extracting bioactive compounds, but there are several drawback of MAE methods as postulated by Torres *et al.* (2017) and Panzella *et al.* (2020). First, excessive power applied may accidentally degraded the bioactive compounds extracted from the sample. Second, the equipment used in MAE is expensive. Lastly, it was highlighted that appropriate temperature, duration and frequency is required in MAE.

3. Polyphenol compounds in pomegranate peel

Polyphenol compounds are phenolic acids, flavonoids, stilbenes, phenolic alcohols and lignans. Furthermore, Panzella *et al.* (2020) highlighted POMEPE was rich in punicalagin and punicalin. Kharchoufi *et al.* (2018) emphasised that at the early maturity stage of

pomegranate, the fruit contains abundant polyphenols and keeps on rising. However, once it reached the peak in the maturation stage, the polyphenol concentration started to decrease over time. Besides, Walid *et al.* (2012) revealed that POMEPE's phytochemical compounds were significantly higher than pomegranate seed, leaves and flower in both water-based extraction and methanolic extraction with $p < 0.05$. Moreover, it was highlighted that POMEPE phenolic content was 10-fold higher compared to the pulp extract (Kennas and Chibane, 2019). Meanwhile, Balamurugan *et al.* (2020) reported that POMEPE contains thrice concentrations of polyphenols compared to pomegranate pulp. Furthermore, POMEPE is a waste commonly found in several levels of processing or consumption level. Therefore, POMEPE has huge potential to be utilised based on its characteristics and abundance in the global agriculture waste.

The polyphenol compounds and concentration of POMEPE was tabulated in Table 3. When compared to other solvents (hexane, ethyl acetate and acetone), it showed that WME had a higher TPC value. As highlighted by Hadrach *et al.* (2014), the TPC of WME

of POMEF were 175.10 ± 2.34 mg GAE/g DW and 6.6 ± 0.76 mg CE/g DW, respectively. From the study, when maceration extraction using hexane as the solvent, the polyphenol compounds were unable to be retrieved as analysed by TPC assay from POMEF sample (Hadrich *et al.*, 2014).

Next, WPE was uncommon in agriculture waste study. Usually, this method is used in the sample involved with brewing techniques such as coffee or tea. WSE, WHDE and WSDE are rarely opted in antioxidant studies based on a few exhaustion processes and high temperatures involved in a long period of time that may have contributed to the interest compounds' degradation.

Further, the growing stages, type of cultivars, ecological conditions, cultivated region and extraction conditions were among the factors pointed out by Jalal *et al.* (2018) and Jalili *et al.* (2020) which initiate these variations. This is due to the adaptation and defence responses towards the cultivated environment and condition (Laxa *et al.*, 2019).

4. Antioxidant potential in pomegranate peel

An antioxidant is a compound which delays or inhibits lipid oxidation of another biomolecule oxidation that prevents or repairs the damaged cell due to the oxidation process. The antioxidant potential in POMEF was tabulated in Table 4.

In the DPPH (IC₅₀) assay of POMEF, there was no significant difference ($p > 0.05$) between WME (163.50 ± 10.42 µg/ml) and methanolic maceration extraction (ME) (155.16 ± 13.24 µg/ml) were recorded (Walid *et al.*, 2012). Next, it was revealed that WDiE and methanolic digestion extraction (DiE) had no significant difference ($p > 0.05$) (Kharchoufi *et al.*, 2018). Next, 25%, 50% and 100% methanolic ultrasonic-assisted extraction (UAE) had lower DPPH values compared to WUAE (66.6%) with 51.84%, 55.06% and 63.95% radical scavenging activity, respectively (Kumar and Ali, 2015). WME had a higher DPPH value compared to ethanolic ME with $32.73 \pm 2.67\%$ and $20.75 \pm 3.26\%$, respectively (El-Nekeety *et al.*, 2017). Meanwhile, methanolic ME showed no significant difference ($p > 0.05$) compared to WME in the ABTS assay (Jalal *et al.*, 2018). Then, WMAE's EC₅₀ DPPH radical scavenging activity was 15.07 mg/mL which was significantly lower compared to ethanolic MAE (21.68 mg/mL) (Alias and Abbas, 2017). As highlighted by Jalal *et al.* (2018), ABTS radical cation was mostly reactive towards thiols, ascorbic acid and phenolics.

Chen *et al.* (2014) postulated that alkaloids seemed to have a stronger correlation with antioxidant activity compared to phenols. Furthermore, Walid *et al.* (2012)

emphasised that instead of solely depending on the polyphenol's concentration, the structure and interactions formed between the compounds with antioxidant potential also played significant roles in radical scavenging activity. Within WDiE of POMEF, α -punicalagin, β -punicalagin were the most concentrated compounds extracted from the sample and followed by HHDP-hexoside, ellagic acid-*O*-hexoside, punicalagin derivatives, pedunculagin II, ellagic acid-*O*-pentoside and ellagic acid (Kharchoufi *et al.*, 2018). These compounds contributed to antioxidant activity in WDiE of POMEF. El-Hamamsy and El-Khamissi (2020) revealed that WIE had better antioxidant activity compared to WME through DPPH assay (IC₅₀). The main difference between WME and WIE was temperature, where it was conducted at temperatures of 25°C and 80°C, respectively. This is because ambient temperature solvent has a higher dielectric constant compared to non-ambient temperature solvents, which reduces the aqueous solution's extraction power and mass kinetic transfer (Vladić *et al.*, 2020). In WDE, the comparison with methanol was conducted by Wang *et al.* (2011), which revealed that WDE DPPH antioxidant activities of POMEF were 53.74%. Therefore, based on the strong correlation between total polyphenols concentration and DPPH radical scavenging activities ($R^2 = 0.98$), it consequently revealed higher antioxidant activity in WDE of POMEF (Negi *et al.*, 2003; Wang *et al.*, 2011). It was indicated that at extraction temperature, extraction time, microwave power and the ratio of solid to liquid of 80°C, 50 mins, 2000 W and 40% (w/v); and 60°C, 50 mins, 6000 W and 10% (w/v) had the lowest antioxidant activity (71.43 ± 1.41 L and 71.43 ± 0.27 L, respectively) (Skenderidis *et al.*, 2020). Meanwhile, the highest antioxidant activity was identified in 60°C, 10 mins, 6000 W and 25% (w/v) with WMAE of POMEF which had 17.61 ± 0.59 L in extract equivalent volume for achieving the reference IC₅₀ value of DPPH (Skenderidis *et al.*, 2020). Next, Boggia *et al.* (2016) revealed that the WUAE (89 ± 0.74 mg GAE/100 g DW) had a higher TPC value compared to the WMAE (59 ± 0.91 mg GAE/100 g DW) of POMEF. It was revealed that WUAE had $41.0 \pm 0.92\%$ of radical scavenging activities, while, WMAE had $36.0 \pm 0.96\%$ of DPPH radical scavenging activities with $p < 0.05$. The explanation to the reported data was the rapid warming and direct heat interaction from MAE have the probability to degrade the polyphenols content in the sample as mentioned by Boggia *et al.* (2016). Therefore, across several aqueous-based extractions, decoction, UAE and MAE showed the most acceptable data to be used in POMEF antioxidant study.

Table 3. Polyphenol compounds of pomegranate peel.

Type of Sample Extract	Extraction Technique	Polyphenol Content	Polyphenol Analysis	Reference(s)
POMEPE	WME	Alkaloids, flavonoids, saponins, tannins alkaloids, steroids, terpenoids, anthocyanins, hydrolysable tannins, condensed tannins, protocatechuic acid, syringic acid, ellagic acid, punicalagin, flavan-3-ols	TPC 1. 85.60±4.8 mg GAE/g DW 2. 175.10±2.34 mg GAE/g DW 3. 175.10±2.34 mg GAE/g DW 4. 5.7 mg QE/ 100 g 5. 242.05±7.99 mg GAE/g 6. 0.30±0.501 mg GAE/ mg DW Other polyphenol content: 1. 6.6±0.76 mg CE/g DW 2. 6.6±0.76 mg CE/g DW 3. 11.5±0.54 µg QE/mg 4. 1.9 mg catechol/100 g	Hadrich <i>et al.</i> (2014), Jalili <i>et al.</i> (2020), Kennas and Chibane (2019), Walid <i>et al.</i> (2012)
	WDiE	Tannic acid, punicalagins derivatives, pedunculagin II, ellagic acid-O-hexoside, HHDP-hexoside, ellagic acid-O-pentoside	TPC 1. 400 - 435 mg TAE/g DW 2. 420.06±0.05 mg/g 3. 274.08 mg GAE/g DW 4. 274.08 mg GAE/g DW	Khan <i>et al.</i> (2017), Kharchoufi <i>et al.</i> (2018)
	WIE	Punicalagin, phenolics, flavonoids, alkaloids, saponins, tannins, steroids, terpenoids, protocatechuic acid, syringic acid, ellagic acid, iso-ferulic acid	TPC 1. 286.58 mg GAE/g DW 2. 286.58 mg GAE/g DW 3. 143.4±5.2 mg GAE/g DW 4. 286.5 mg GAE/g DW 5. 166.83 mg GAE/100 g DW Other polyphenol content: 1. 58.85±0.4 mg Rutin/g	El-Hamamsy and El-Khamissi (2020), Kharchoufi <i>et al.</i> (2018), Kushwaha <i>et al.</i> (2015)
	WDE	Ellagitannins gallic acid, ellagic acid, polymeric tannins	TPC 1. 20.27 - 28.95 mg GAE/g DW 2. 24.89 mg GAE/g DW 3. 150 - 300 mg GAE/g DW Other polyphenol content: 1. 43.19±2.24 g TCE/100 g DW	Balli <i>et al.</i> (2020), Canuti <i>et al.</i> (2020), Vladić <i>et al.</i> (2020), Wang <i>et al.</i> (2011)
	WHDE / WSHE	Camphene, 5-methyl-furfural, 1,8-cineole, butylbenzene, 2-methylbenzofuran, camphor, hexylbenzene, α-cadinol, dibutyl phthalate, palmitic acid, oleic acid	N. D.	Hadrich <i>et al.</i> (2014)
	WSE	Gallic acid	TPC 1. 40.8±0.2 - 59.8±0.6 mg GAE/g DW 2. 28.78 mg GAE/g DW	Negi <i>et al.</i> (2003), Nuamsetti <i>et al.</i> (2012)
	WUAE	Anthocyanin, tannin, ellagitannins, ellagic acid, gallic acid, catechin, chlorogenic acid, caffeic acid, catechin, quercetin, caffein, caffeic acid, syringic acid, naringenin cinnamic acid, punicalagin, ellagic acid-hex	TPC 1. 100 - 200 mg GAE/g DW 2. 542.5 mg GAE/g DW 3. 10.7 - 76.8 mg GAE/g DW 4. 15.00 - 20.00 mg GAE/g DW Other polyphenol content: 1. 102.8 mg RE/g DW	Boggia <i>et al.</i> (2016), Carrera <i>et al.</i> (2012), Hüseyinbaş <i>et al.</i> (2020), Kumar and Ali (2015), Rongai <i>et al.</i> (2017)

POMEPE: Pomegranate peel, WME: Water-based maceration extraction, WDiE: Water-based digestion extraction, WIE: Water-based infusion extraction, WDE: Water-based decoction extraction, WPE: Water-based percolation extraction, WSE: Water-based Soxhlet extraction, WHDE: Water-based hydrodistillation extraction, WSDE: Water-based steam-distillation extraction, WUAE: Water-based ultrasound-assisted extraction, WMAE: Water-based microwave-assisted extraction (WMAE), N.D.: No Data.

Table 4. Antioxidant potential of pomegranate peel.

Type of Sample Extract	Extraction Technique	Radical Scavenging Activity	Reference(s)
POMEPE	WME	<ul style="list-style-type: none"> DPPH (EC₅₀) assay = 11.48±2.29 µg/mL ABTS assay = 3.80±0.31 TEAC mmol/100 g DW Reducing power (EC₅₀) assay = 163.50±10.42 µg/mL IC₅₀ analysis = 31.2±5.4 µg/mL IC₅₀ β-carotene bleaching = 65.0±2.90 µg/mL IC₅₀ FRAP assay = 750.32±5.4 µg/mL Condensed tannins had a positive correlation with DPPH radical scavenging activity (R² = 0.94) 	Hadrich <i>et al.</i> (2014), Kennas and Chibane (2019), Walid <i>et al.</i> (2012)
	WDiE	<ul style="list-style-type: none"> DPPH value = 3497.02 mmol Trolox/g DPPH assay = 70-80% Correlation (R² = 0.992) relationship with antioxidant activities DPPH assay (IC₅₀) = 23.5±2.7 µg/mL 	El-Hamamsy and El-Khamissi (2020), Kharchoufi <i>et al.</i> (2018)
	WIE	<ul style="list-style-type: none"> DPPH assay = 63.63±2.15% DPPH (EC₅₀) assay = 183.71±8.71 µg/mL DPPH assay (IC₅₀) assay = 23.5±2.7 µg/mL 	El-Hamamsy and El-Khamissi (2020), Kennas and Chibane (2019), Kushwaha <i>et al.</i> (2015)
	WDE	<ul style="list-style-type: none"> DPPH antioxidant activities = 53.74% TPC and DPPH radical scavenging activities (R² = 0.98) 	Negi <i>et al.</i> (2003) Wang <i>et al.</i> (2011)
	WSE	<ul style="list-style-type: none"> DPPH activities = 956±12.7 µmol AAE/g DW Reducing activity = 2.056 Super anion scavenging activity = 2.012 IC₅₀ DPPH assay = 10.2 µg/mL to 13.1 µg/mL Antioxidant activity = 15% 	Kumar and Ali (2015), Mishra <i>et al.</i> (2017), Negi <i>et al.</i> (2003), Nuamsetti <i>et al.</i> (2012)
	WUAE	<ul style="list-style-type: none"> DPPH radical scavenging activity = 66.06% FRAP assay = 763.73 mg FSE/g DW DPPH assay = 15.8% to 88.8% Antioxidant activity = 66.6% DPPH assay = 41±0.92% 	Boggia <i>et al.</i> (2016), Kumar and Ali (2015), Sharayei <i>et al.</i> (2019)
	WMAE	<ul style="list-style-type: none"> IC₅₀ DPPH assay: 14.53 µg/mL DPPH radical scavenging activities = 90% Antioxidant activity = 17.61±0.59 L DPPH assay = 36.0±0.96% 	Boggia <i>et al.</i> (2016), Skenderidis <i>et al.</i> (2020), Xueling <i>et al.</i> (2011)

POMEPE: Pomegranate peel, WME: Water-based maceration extraction, WDiE: Water-based digestion extraction, WIE: Water-based infusion extraction, WDE: Water-based decoction extraction, WUAE: Water-based ultrasound-assisted extraction, WMAE: Water-based microwave-assisted extraction.

5. Comparison between water-based extract

Each extraction factor or variable influenced the quantity and quality of the sample extract. The comparison was tabulated in Table 5. There were findings regarding the effectiveness of WIE compared to WME and WDiE, and WDE compared to WUAE found. This indicated that the higher temperature used in the extraction helps in reducing solvent surface tension and viscosity which facilitated solvent penetration and solubilisation of the sample.

Besides, the high-temperature solvent has a low dielectric constant, which increases the water's extraction power and mass kinetic transfer (Vladić *et al.*, 2020). As explained by Turrini *et al.* (2020) POMEPE extracted by WDE (100°C) and WUAE (65°C) were

influenced by the temperature. Although sonic wave was used in WUAE, the temperature played a vital role in extracting the bioactive compounds. Next, WMAE showed better potential compared to WSE (Kumar and Ali, 2015; Alias and Abbas, 2017). This is due to WSE undergoing several exhaustions for the formation of vapour in the Soxhlet apparatus resulting in compound degradation.

Although the phenolic yield had a positive linear relationship with extraction time and a curvilinear increase with microwave output power, several studies have highlighted that excessive exposure to microwave power towards the plant cell may consequently deteriorate the polyphenol compounds and affect the TPC yield (Xueling *et al.*, 2011; Zengin *et al.*, 2019;

Table 5. Comparison between extraction techniques.

Sample	Technique(s)	Description	Reference(s)
POMEPE	WIE > WME	TPC WIE = 143.4±5.2 mg GAE/g DW WME = 126.0±1 mg GAE/g DW DPPH radical scavenging activity (IC ₅₀) WIE = 23.5±2.7 µg/mL WME = 27.6±2.9 µg/mL	El-Hamamsy and El-Khamissi (2020)
	WIE > WDiE	TPC WIE = 286.58 mg GAE/g DW WDiE = 274.08 mg GAE/g DW p>0.05	Kharchoufi et al. (2018)
	WIE > WDiE	TPC WIE = 11.15% WDiE = 8.26%	Wang et al. (2011)
	WIE > WDiE	TPC WIE = 1.4 g GAE/100 g WDiE = 1.0 g GAE/100 g	Gaye et al. (2019)
	WDE > WUAE	TPC WDE = 150 mg GAE/g DW WUAE = 100 mg GAE/g DW p<0.05	Turrini et al. (2020)
	WMAE > WSE	TPC WMAE = 207.72 mg GAE/g DW WSE = 28.78 mg GAE/g DW p<0.05	Alias and Abbas (2017)
	WMAE > WSE	TPC WUAE = 2.45 mg GAE/g WSE = 1.82 mg GAE/g DPPH activity WUAE = 66.06% WSE = 15%	Kumar and Ali (2015)
	WUAE > WMAE	TPC WMAE = 59.0±0.91 mg GAE/100 g DW WUAE = 89.3±0.74 mg GAE/100 g DW p<0.05 DPPH activity WUAE = 41.0±0.92% WMAE = 36.0±0.96% p<0.05	Boggia et al. (2016)

POMEPE: Pomegranate peel, WME: Water-based maceration extraction, WDiE: Water-based digestion extraction, WIE: Water-based infusion extraction, WSE: Water-based Soxhlet extraction, WUAE: Water-based ultrasound-assisted extraction, WMAE: Water-based microwave-assisted extraction.

Skenderidis et al., 2020). As reported by Alias and Abbas (2017), the phenolic content at 120°C was 99.42 mg GAE/g DW, while at 30°C (206.46 mg GAE/g DW), the phenolic compounds identified were much higher. Réblová (2012) highlighted that the antioxidant activity started to decline at higher temperatures with a linear relationship with $p<0.01$ ($R^2 = -0.816$). Besides high temperature influencing the extraction yield's quality, rapid warming and direct heat interaction from MAE also contributed to the degradation of the polyphenols content in the sample (Boggia et al., 2016).

6. Conclusion

Though the water-based extraction technique rarely chosen compared to other solvents-based extraction

techniques such as ethanol, methanol and acetone, water extracts showed medium to high antioxidant activities. Although there was evidence that water-based extraction had slightly lower antioxidant activities, there were tremendous studies which showed that proved otherwise. The radical scavenging activities in water-based extraction of the selected fruit peels ranged from 30 to 70%. Therefore, it was revealed that the non-toxic, non-corrosive and green solvent which are the unique property of water has great potential in the extraction of substances with antioxidant activities from fruit peels such as pomegranate.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgements

The authors are grateful to the Bestari Grant Scheme: 600-IRMI/DANA 5/3/BESTARI (125/2018) from Universiti Teknologi MARA.

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