

Antioxidant Properties of *Pyrus communis* and *Pyrus pyrifolia* Peel Extracts

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Abstract

With the rise of global health awareness by embracing a healthy lifestyle and natural product consumption, the search for natural antioxidant sources has invited more research performed especially in fruits as a whole product or in its components, especially the ones that are usually discarded/not the mainly consumed component. *Pyrus communis* and *Pyrus pyrifolia* are two of the most popular species of pear fruits consumed in the world, and while there exist several studies about the antioxidant potential of its flesh, specific studies about the antioxidant properties of their peels are still lacking. To achieve an understanding of antioxidant properties of peel extract of *P. communis* and *P. pyrifolia*, several tests have been performed in controlled laboratory conditions to gauge phenolic and flavonoid contents through three different extraction measurements. This research has shown that peel extracts of both *P. communis* and *P. pyrifolia* contain natural antioxidants that are beneficial for health, with *P. communis* extracted with methanol having a higher concentration of antioxidant contents compared to *P. pyrifolia*.

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INTRODUCTION

In food research, phenolic compounds with antioxidant activity holds an important position. This is related to the fact that phenolic compounds play a beneficial effect on preventing and/or treating various diseases, such as cancer, diabetes, cardiovascular and neurodegenerative diseases, among others (Cory *et al.*, 2018; Panche *et al.*, 2016; Pandey & Rizvi, 2009). Phenolic is an important bioactive and possess strong antioxidant activity, and fruits and vegetables usually are the primary sources of Phenolic compound (Tungmunnithum *et al.*, 2018). It is widely believed that the ingestion of fresh fruits and vegetables is related to the reduction of cardiovascular and cancer diseases (Aghajanpour *et al.*, 2017).

Pear (*Pyrus* spp) have been known to contain a number of phytochemical compounds with important bioactive properties like phenolics and flavonoids, which are essential for their health benefits (Öztürk *et al.*, 2015). Those health benefits of polyphenol consumption derive from their antioxidant and anti-inflammatory properties (Borges *et al.*, 2010). Other pro-health properties that are attributed to pears are related to the content of triterpenoids, due to their antioxidative, anti-inflammatory, and anticancer properties (Reiland & Slavin, 2015).

During recent years, some researchers have been focused on analyses and comparison of the nutritional components contained in the edible part of pear fruit such as total sugars, vitamins, minerals, and so on (Slavin

& Lloyd, 2012; Kahle *et al.*, 2005; Tanriöven & Ekşi, 2005). A part from some common reported compounds such as arbutin, chlorogenic acid, catechin, quercetin, kaempferol, various hydroxycinnamoylmalic acids, and their ethyl esters, hydroxycinnamoyl malates, procyanidins, and triterpenes compound has also been found in the peel of pear (Lee *et al.*, 2011a; Lee *et al.*, 2011b; Ma *et al.*, 2012).

Pears is one of the fruits imported into Indonesia which is often found in supermarkets or traditional markets. Pears which are widely circulating in Indonesia are *Pyrus communis* or green pears/European pears and *Pyrus pyrifolia* or yellow pears/Asian pears (Novera *et al.*, 2015). Pears are usually consumed along with the peels, although consuming the fruit while discarding the peels are also common, because the peels are often viewed as non-beneficial (Sagar *et al.*, 2018). With this fact, understanding the potentials and bioactive compounds of pear peel as a side product of the fruit is very important (Li *et al.*, 2014).

Based on our study in this matter, we found that the information on the composition and constituents of pear peel are limited. Therefore, we propose that it is important for food products research to have a systematic study on pear peel, especially on the phenolic and flavonoid compounds. The aim of this research is to identify the total phenolic and antioxidant contents, and performing qualitative determination of phytochemical contents for the two pear varieties.

MATERIALS AND METHODS

Tools and material

The main material used in the study are *P. communis* and *P. pyrifolia*. Both samples of pears were obtained from Caringin's Traditional Market, Bandung, West java that imported from Australia and China, respectively. Other materials used were methanol, ethyl acetate, *n*-hexane,

ethanol, ascorbic acid, gallic acid, quercetin, Folin-Ciocalteu's phenol reagent (Sigma), aquadest, chloroform, toluene, acetone, 25% ammonia 2 N, 10% hydrochloric acid, Dragendorff reagent, Mayer reagent, Mg powder, amyl alcohol, sodium hydroxide, gelatin, Fe (III) chloride 1%, and anhydrous acetic acid. The tools used are sifters, cups, oven, desiccators, maceration chamber, erlenmeyer flasks, vacuum rotary evaporator, analytical scales, and micropipette. Spectrophotometric measurements were performed on ultraviolet (UV)-1600 spectrophotometer (Shimadzu).

Sample preparation

Both pear peel samples that have been collected and cleaned are dried by drying the cabinet at 40-50°C until smooth and dry.

Extractions

Pear peels were extracted by maceration in 200 ml of *n*-hexane, ethyl acetate and methanol at room temperature for 24 hours. Residues and extracts are separated by filtering using filter paper. The residue obtained was re-extracted twice with a fresh portion of the extraction solvent. Extracts obtained from the three extraction processes were combined and excess solvents were concentrated using a vacuum rotary evaporator at 50°C. Semisolid extracts obtained quantitatively were transferred to extraction solvents and stored at 10°C until used for further experiments.

Phytochemical screening

Phytochemical compounds were carried out for all extracts with the following methods:

Steroids and terpenoids

As much as 2 ml of extract was added to 2 ml of acetic anhydride and the concentrated H₂SO₄. The formation of blue with green rings indicates the presence of terpenoids and the brown rings indicate the presence of steroids (Ayoola *et al.*, 2008).

Alkaloids

Extracts were dissolved individually in dilute hydrochloric acid and filtered. Filtrates were treated with Dragendroff's reagent (solution of Potassium Bismuth Iodide). The Formation of red precipitate indicates the presence of alkaloids (Altemimi *et al.*, 2017).

Saponins

As much as 0.5 g of extract was shaken with 2 ml of water. If foam produced persists for ten minutes it indicates the presence of saponins (Altemimi *et al.*, 2017).

Tannins

To the extract, 1% gelatin solution containing sodium chloride was added. The Formation of white precipitate indicates the presence of tannins (Altemimi *et al.*, 2017).

Phenolics

Extracts were treated with 3-4 drops of ferric chloride solution. The Formation of bluish-black color indicates the presence of phenols (Altemimi *et al.*, 2017).

Flavonoids

Extracts were treated with a few drops of sodium hydroxide solution. Formation of intense yellow colour, which becomes colourless on the addition of dilute acid, indicates the presence of flavonoids (Altemimi *et al.*, 2017).

Determination of total phenolic contents

Total polyphenol was determined by Folin-Ciocalteu method (Aryal *et al.*, 2019). A dilute extract of each plant extract (0.5 ml of 1 : 10 g/ml) or gallic acid (standard phenolic compound) was mixed with Folin-Ciocalteu reagent (5 ml, 1 : 10 diluted with distilled water) and aqueous Na₂CO₃ (4 ml, 1 M). The mixtures were allowed to stand for 15 minutes and the total phenols were determined by colorimetry at 765 nm. The standard curve was prepared using 0, 50, 100, 150, 200, and 250 mg/L solutions of gallic acid in methanol : water (50 : 50, v/v). Total phenol values are expressed in terms of gallic

acid equivalent (mg/g of dry mass), which is a common reference compound.

Determination of total flavonoid contents

Aluminum chloride colorimetric method was used for flavonoids determination (da Silva *et al.*, 2015). Each plant extracts (0.5 ml of 1 : 10 g/ml) in methanol were separately mixed with 1.5 ml of methanol; 0.1 ml of 10% aluminum chloride; 0.1 ml of 1 M potassium acetate; and 2.8 ml of distilled water. It remained at room temperature for 30 minutes. The absorbance of the reaction mixture was measured at 415 nm with UV-Vis spectrophotometer. The calibration curve was prepared by preparing quercetin solutions at concentrations 12.5 to 100 g/ml in methanol.

Data analysis

Results are given as the mean of three independent determinations ± standard deviation using SPSS version 19.0. The data were statistically analyzed by ANOVA and *t*-tests. The level of statistical significance was set at $p < 0.05$.

RESULTS AND DISCUSSION

Phytochemical screening

Phytochemical screening of the two varieties of peel pears shows that both have the same content, namely phenolic, flavonoids, tannins, saponins, and alkaloids as shown in **Table I**. All of these phytochemicals are important elements of herbal medicine and are directly related to various health-promoting activities such as anticancer, antifungal, anti-inflammatory, antihyperglycemic, immunomodulatory, and wound healing properties (Forni *et al.*, 2019). However, flavonoid content in *P. communis* is greater than *P. pyriformis*.

Flavonoids have been found to possess antitumoral, antiallergic, and anti-inflammatory activities (Panche *et al.*, 2016). Phenolic content is the greatest content found in

both *P. communis* and *P. pyrifolia*. In particular, many phenolic compounds have been identified in pear fruits such as arbutin, chlorogenic acid, hydroxycinnamoyl malates, catechins, and procyanidins (Sun *et al.*, 2019).

Table I. Qualitative phytochemical screening in *P. communis* and *P. pyrifolia* peels

Samples	Phenolics	Flavonoids	Tannins	Saponin	Alkaloids	Steroids and terpenoids
<i>P. communis</i>	++	++	+	+	+	-
<i>P. pyrifolia</i>	++	+	+	+	+	-

Note: (++) highly presence; (+) presence; (-) absence

Total extraction yields of pear peels

In *P. communis* peels, total extraction yields from *n*-hexane, ethyl acetate, and methanol were 11.35%; 17.95%; and 30.85%, respectively as shown in **Table II**. In *P. pyrifolia* peels, total extraction yields were 13.50%; 18.75%; and 37.50%, respectively as presented in **Table III**. In both *P. communis* and *P. pyrifolia* peels, the highest yields were observed in methanol extract and higher in the *P. pyrifolia* peel extracts. The extractable compounds from various plant materials are strongly dependant on the nature of extractable solvents. Methanol is one of the polar solvents used for active component extraction which can attract many compounds such as anthocyanins, terpenoids, saponins, tannins, xanthoxylines, quassinoids, lactones, flavones, phenones, and polyphenols (Mujeeb *et al.*, 2014).

Table II. Total extraction yields of *P. communis* peels

Extract solvent	Total extraction yields (%)
<i>n</i> -hexane	11.35
Etil acetate	17.95
Methanol	30.85

Table III. Total extraction yields of *P. pyrifolia* peels

Extract solvent	Total extraction yields (%)
<i>n</i> -hexane	13.50
Etil acetate	18.75
Methanol	37.50

Total phenolic contents

The antioxidant activity of plant phenolics is due to the reactivity of phenol moieties (hydroxyl group on the aromatic ring) which have the ability to scavenge free radicals via hydrogen donation or electron donation or electron donation. Total phenolics were determined by Folin-Ciocalteu reagent method. In this method, phenols from a blue-colored phosphomolybdic-phosphotungstic complex in the presence of an alkaline solution (Saeed *et al.*, 2012).

The total phenolic contents in *P. communis* peels were 7.20 ± 0.03 mg/g; 33.35 ± 0.57 mg/g; and 68.86 ± 0.78 mg from *n*-hexane, ethyl acetate, and methanol extracts, respectively. Statistically significant the lowest yields were observed in *n*-hexane extracts as compared to ethyl acetate and methanol extract while the latter two have the same significantly different too. In the case of *P. pyrifolia* peels, the contents from *n*-hexane, ethyl acetate, and methanol extracts were 8.45 ± 0.11 mg/g; 31.22 ± 0.68 mg/g; and 63.50 ± 0.41 mg/g, respectively. Statistical significance was similar to *P. communis* peels as presented in **Figure 1**. Methanol extract have the highest content which were higher in the *P. communis* peels (68.8 mg GAE/g) than in the *P. pyrifolia* peels (63.5 mg GAE/g). This result is in agreement with Singh & Rajini (2004) which reported that the maximum antioxidant yield was obtained with methanol compared to acetone and water.

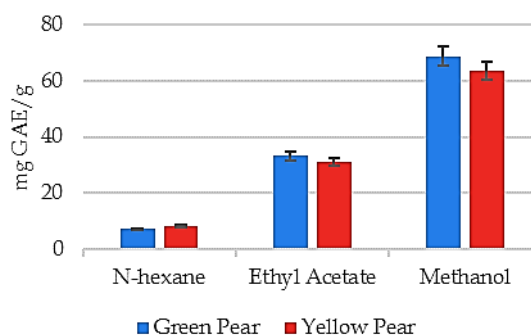


Figure 1. Total phenolic contents of *P. communis* and *P. pyrifolia* peels by solvents

Total flavonoid contents

The total flavonoid contents in *P. communis* peels from *n*-hexane, ethyl acetate, and methanol were 11.72 ± 0.72 mg/g; 49.75 ± 1.72 mg/g; and 62.84 ± 0.77 mg/g, respectively. In the case of *P. pyrifolia* peels, the observed contents were 10.91 ± 0.14 mg/g; 40.69 ± 0.84 mg/g; and 50.9 ± 1.45 mg/g, respectively. Analytic results show that *n*-hexane, ethyl acetate, and methanol extracts on *P. communis* peels were significantly different from *P. pyrifolia* peels. From the result, the highest content was observed in methanol extracts there are 62.84 mg QE/g in *P. communis* peel and 50.9 mg QE/g in *P. pyrifolia* peel as presented in **Figure 2**. Methanol is an effective solvent for polyphenols, and then it is commonly used in the laboratory and in industrial extraction process (Zhang *et al.*, 2018).

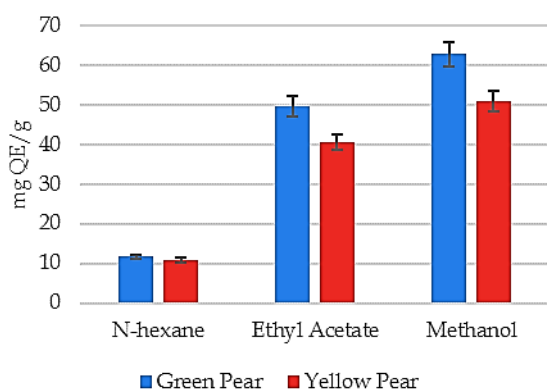


Figure 2. Total flavonoid contents of *P. communis* and *P. pyrifolia* peels by solvents

It can be seen that the total contents of polyphenol and flavonoid have the same trends. Total phenolic content and total flavonoid contents for both the varieties of pear peels showed that methanol extracts are the highest values and *P. communis* peels contained more phenolic and flavonoid contents than *P. pyrifolia* peels.

CONCLUSION

From this study, it is concluded that two varieties of pear peel show the presence of phytochemicals, there are

phenolics, flavonoids, tannins, alkaloids, and saponins. Methanol extracts is the highest amount of total phenol and total flavonoids contents both *P. communis* and *P. pyrifolia* peels which *P. communis* peels exhibited more amounts of phenolic and flavonoids than *P. pyrifolia* peels. The presence of these bioactive compounds in pears varieties establishes themselves as the sources of natural therapeutic agents that can act as potent free radical scavengers.

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