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RESEARCH ARTICLE

Antioxidant activity and phytochemical profiling of peaches, Arabica coffee, and their combination

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ABSTRACT

Antioxidant compounds scavenge unstable free radical reactions and are abundant in certain plants. Peaches (*Prunus persica* L.) are rich in phytochemicals such as carotenoids, anthocyanins, phenolic acids, and flavonoids, while Arabica coffee (*Coffea arabica*) contains alkaloids, flavonoids, tannins, steroids, and terpenoids. This study evaluated the antioxidant activity and phytochemical profile of peaches, Arabica coffee, and their combination using ethanol, ethyl acetate, and water as solvents. Extraction was performed via maceration with 96% ethanol and fractionation using water and ethyl acetate. Antioxidant activity was assessed using the DPPH (1,1-Diphenyl-2-picrylhydrazyl) assay, and phytochemical tests evaluated secondary metabolites such as alkaloids, flavonoids, phenolics, steroids/terpenoids, tannins, and saponins. Results indicated that the ethyl acetate extract of peaches showed moderate antioxidant activity (IC_{50} = 141.91 ppm), while the water extract of Arabica coffee exhibited robust activity (IC_{50} = 22.30 ppm). Notably, combining water extracts from both sources demonstrated enhanced antioxidant capacity (IC_{50} = 37.85 ppm), improving peach extract activity by over threefold. These findings underscore the potential synergistic effects of combining these natural sources for health-promoting antioxidants.

1. Introduction

Therapeutic plants and fruits have been widely utilized for centuries as traditional medicines and remain valuable in contemporary health practices. These natural sources are rich in bioactive compounds, including alkaloids, phenols, terpenoids, and flavonoids, which confer various pharmacological, ecological, and even toxic effects on plants, humans, and animals (Mariyah, 2020). Such compounds contribute to their medicinal potential and play a significant role in promoting health and preventing diseases.

Antioxidant compounds are crucial for neutralizing unstable free radicals by donating electrons to stabilize them and prevent harmful chain reactions. These compounds are increasingly valued in both the food and health industries. In food, antioxidants serve as preservatives, enhancing shelf life, while in health, they reduce the risk of chronic diseases such as cancer and coronary heart disease (Purwanto *et al.*, 2017).

Antioxidants can be categorized into natural and synthetic types based on their source. Plant-derived antioxidants are favored for their efficacy and safety, as synthetic antioxidants carry potential side effects and toxicity risks. Various parts of plants—fruits, leaves, flowers, and stems—serve as rich sources of antioxidants, including phenol derivatives, tocopherols, coumarins, ascorbic acid, dihydroflavones, and hydroxycinnamic compounds (Hamzah, 2016).

Fruits of the Prunus genus, such as peaches

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(*Prunus persica* L.), are known for their phytochemical richness. They contain carotenoids, anthocyanins, phenolic acids, and flavonoids, which have demonstrated potential in preventing degenerative diseases and cancer. Peaches are also attributed with aphrodisiac, antipyretic, and circulatory benefits, and their seeds and oil are used in traditional medicine in various ways (Aziz & Rahman, 2013).

Arabica coffee (*Coffea arabica*) is one of the most popular beverages worldwide. It also has significant health benefits. Its bioactive compounds, such as alkaloids, flavonoids, tannins, saponins, steroids, and terpenoids, exhibit diverse biological activities, including antioxidant, antidiabetic, antihypertensive, antiviral, and antibacterial properties (Ajhar & Meilani, 2020).

Given the recognized health benefits of peaches and Arabica coffee, their combined use may offer synergistic antioxidant effects. This study aims to investigate the antioxidant activity and phytochemical profile of *Prunus persica* and *Coffea arabica* individually and in combination, using the DPPH (2,2-diphenyl-1-picrylhydrazyl) assay. Understanding their interaction and potential enhancement of antioxidant capacity is critical for advancing their application in medical science and health-related fields.

2. Materials and Methods

2.1. Sample Preparation, extraction, and fractionation of peaches and arabica coffee

Fresh peaches were sorted, washed, cut into small cubes, and blended to create a homogenized mixture. Arabica coffee beans were roasted until brown and then pulverized using a blender. Prepared samples of peaches and arabica coffee were macerated in 96% ethanol within a large Erlenmeyer flask and left to macerate for six hours with periodic stirring using a shaker. The macerate was filtered, and the filtrate was concentrated using a vacuum rotary evaporator to obtain a viscous ethanol extract. Peach extract fractionation was performed using a 10 g paste of the thick ethanol extract, which was mixed with 600 mL distilled water and 300 mL ethanol and then placed in a separatory funnel with 700 mL ethyl acetate. Arabica coffee fractionation was performed using a 10 g extract combined with 200 mL distilled water and 200 mL ethanol, followed by 600 mL ethyl acetate in a separatory funnel. Solutions were shaken to separate into an organic solvent layer (bottom) and an aqueous layer (top). The bottom layer was collected, and this process was repeated three times.

2.2. Antioxidant Activity Test

Antioxidant activity was measured using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay. A volume of 0.2 mL of the sample solution was pipetted into a vial. Subsequently, 3.8 mL of a 50 µM DPPH solution was added, and the mixture was homogenized. The solution was incubated in the dark for 30 min. Absorbance was measured using a UV-Vis spectrophotometer at a wavelength range of 400–600 nm (Cahyani, 2015). The antioxidant activity of the samples was determined from the level of inhibition of DPPH radical uptake by calculating the percentage of inhibition of DPPH uptake (Asra *et al.*, 2019). IC_{50} values were calculated using Equation 1, where abs Blank is absorbance 50 µM DPPH, and abs sample is the absorbance of Sample

$$\% \text{ inhibition} = \frac{Abs\ Blank - Abs\ Sample}{Abs\ Blank} \times 100\%$$

Antioxidant activity was classified based on the IC_{50} values as follows: strong when $IC_{50} < 50$ ppm, vital when $50 < IC_{50} \leq 100$ ppm, medium when $100 < IC_{50} \leq 150$ ppm, Weak when $150 < IC_{50} \leq 200$ ppm, and fragile: $IC_{50} > 200$ ppm (Mangkasa *et al.*, 2018; Molyneux, 2004; Sasmita *et al.*, 2014; Tristantini *et al.*, 2016).

2.3. Phytochemical Screening

Phytochemical screening was conducted using standard methods described by Trease and Evans (1996) and Lamara *et al.* (2018). A 2.5 g plant extract sample was mixed with 25 mL of deionized water in a marked container. Alkaloid was identified by adding 1 mL of Dragendorff's reagent to the extract, followed by heating and shaking. The absence of foam upon adding 2 N HCl indicated a positive result.

Saponins were screened by adding 2 mL extract, and 2 mL deionized water was shaken vigorously. Persistent foam for 30 minutes indicated the presence of saponins. Tannins and Phenols were identified by adding 13% of ferric chloride (FeCl3) to 1 mL of the extract, with color changes (e.g., green, blue, red) indicating positive results (Dabbou, 2017; Mariyah, 2020). Flavonoids were identified by adding 2 mL of 2 N NaOH to the extract, producing a characteristic yellow hue for flavonoids. A yellow color indicated the presence of coumarins upon mixing 1 mL extract with 1 mL of 10% NaOH solution. In addition, to detect the presence of flavonoids, approximately 1 mL of the sample is mixed with 0.5 g of magnesium powder. Then, ten drops of concentrated HCl are carefully added to the mixture. A positive reaction is indicated by developing an orange, pink, or red color in the solution, confirming the presence of flavonoids (Bintang, 2017). Phlorotannins were screened by mixing 1 mL sample, chloroform, acetic anhydride, and concentrated H_2SO_2 produced a red-orange or purple color for terpenoids or steroids (Purwanto *et al.*, 2017). Cardiac Glycosides (Steroids and Triterpenoids) were screened by adding

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A solution of 100% acetic acid with ferric chloride and concentrated H_2SO_2 produced layered color changes (e.g., chocolate, green, purple), confirming their presence (Hasnaenil & Aminah, 2019).

2.4. Data Analysis

Data from phytochemical Tests were analyzed descriptively. IC_{50} values from the antioxidant activity test were calculated and analyzed quantitatively. Statistical methods, including means, standard deviations, and ANOVA, were used to compare antioxidant activities among peach extract, Arabica coffee extract, and their combination.

3. Results

3.1 Extraction and Fractionation

This study involved the extraction of peaches (*Prunus persica* L.) and arabica coffee (*Coffea arabica*) and assessing their antioxidant activity using the DPPH method. The antioxidant activity was evaluated for each extract: peach, arabica coffee, and a combination. Each sample underwent three replicates to ensure reliability. The fruit was identified at the Bogoriense Herbarium, LIPI Biology Research Center in Bogor, West Java, confirming it as *Prunus persica* L. The fractionation yields (Table 1) indicate that the water fraction of Arabica coffee exhibited the highest yield (88%), while the ethyl acetate fraction of peaches had the lowest yield (6%).

Table 1. Yield of fractionation of peaches, arabica coffee, and a combination of both

Fraction	Mass (g)	Yield (%)
Peach ethyl acetate	1,2	6
Peach juice	10	50
Arabica coffee ethyl acetate	1,2	12
Arabica coffee water	8,8	88
Combined ethyl acetate	12,2	61
Combined water	7,8	39

3.2. Antioxidant Activity of Peach Fruit Extract, Arabica Coffee, and a Combination of Both

Table 2 shows that the ethyl acetate extract of peaches demonstrated the best antioxidant activity (IC_{50} = 141.91 ± 5.74 ppm, medium category). This aligns with earlier findings (Dhingra *et al.*, 2014). showing that peaches' ethyl acetate fraction had superior antioxidant properties compared to other fractions. Arabica coffee exhibited potent antioxidant activity across all fractions, with the water fraction yielding the best IC_{50} value (22.30 ± 1.02 ppm). This agrees with previous studies conducted in Jember (Hamzah, 2016; Setiadi, 2016). The combined extracts of peaches and Arabica coffee showed enhanced antioxidant activity, particularly in the water fraction (IC_{50} = 37.85 ± 1.91 ppm, powerful category). Combining the two extracts improved the antioxidant potential of peaches, increasing their IC_{50} value by 75.3% or approximately 3.5 times. One-way ANOVA revealed that fractionation significantly affects the antioxidant activity of peach extracts ($p < 0.001$). In contrast, fractionation did not considerably impact the antioxidant activity of arabica coffee ($p = 0.143$), indicating similar activity levels across fractions. However, the combination of peach and coffee extracts showed significant differences due to fractionation ($p < 0.001$), demonstrating enhanced antioxidant strength in the combined extracts.

3.3. Phytochemical Test Results

The phytochemical analysis in Table 3 shows that the ethanol extract of peaches contains flavonoids, while the ethyl acetate extract contains phenolics, flavonoids,

Table 2. Antioxidant activity of fractionation of peaches, arabica coffee, and a combination of both

Fraction	IC_{50} (ppm)			
	Peaches	Arabica Coffee	Combination Peacheas and Arabica Coffee	Vitamin C
Ethanol	298.01±7.46	25.05±2.17	55.06±3.28	3.91±0.19
Ethyl acetate	141.91±5.74	22.80±1.18	38.06±2.65	
Water	454.60±40.62	22.30±1.02	37.85±1.91	

Table 3. The phytochemical analysis of fractionation of peaches, arabica coffee, and a combination of both

No.	Metabolites	Test method	Peaches			Arabica Coffee		
			Ethanol	Ethyl acetate	Water	Ethanol	Ethyl acetate	Water
1	Phenolic	FeCl3	-	+	+	+	+	+
2	Flavonoid	HCL concentrated +Mg	+	+	+	+	+	+
		H_2SO_4 2N	+	+	+	+	+	+
		NaOH 10%	+	+	+	+	+	+
3	Steroid/Triterpenoid	Lieberman Burchard	-	-	-	-	-	-
4	Saponin	HCL+ H_2O	-	-	-	-	-	-
5	Tanin	FeCl3 1%	-	+	-	+	+	+

and tannins. The water extract also contains phenolics and flavonoids.

4. Discussion

The research emphasizes the phytochemical abundance and antioxidant capabilities of natural extracts from peaches (*Prunus persica* L.) and Arabica coffee (*Coffea arabica*) separately and in conjunction. The results validate the existence of bioactive substances, including phenolics, flavonoids, and tannins, which substantially enhance their antioxidative effects.

The antioxidant activity of peach extracts, especially in the ethyl acetate fraction (IC_{50} = 141.91 ppm), was determined to be moderate. This is consistent with earlier research by Dhingra *et al.* (2014), which shows that ethyl acetate fractions of peaches possess enhanced antioxidant activities relative to other solvents. Phenolics and flavonoids in peach extracts significantly contribute to this activity due to their capacity to neutralize free radicals and avert oxidative damage. These chemicals are crucial in alleviating oxidative stress, a substantial contributor to the onset of chronic diseases like cancer and cardiovascular conditions (Ishak, 2018).

Arabica coffee extracts exhibited robust antioxidant activity in all fractions, with the aqueous fraction showing the optimal IC_{50} value of 22.30 ppm. These findings validate earlier research (Hamzah, 2016; Setiadi, 2016) highlighting the enhanced antioxidant capacity of coffee extracts derived from polar solvents. The elevated polyphenols and flavonoids in Arabica coffee beans highlight their effectiveness in neutralizing free radicals. Polyphenols serve as micronutrients recognized for diminishing oxidative stress, safeguarding cellular integrity, and promoting general health (Ajhar & Meilani, 2020; Mangiwa & Maryuni, 2019).

The amalgamation of peach and Arabica coffee extracts exhibited improved antioxidant efficacy, particularly in the aqueous fraction (IC_{50} = 37.85 ppm). This synergistic impact underscores the potential to enhance their effectiveness by integrating natural antioxidants. The combination's enhanced antioxidant capacity, which augmented peaches' activity by roughly 3.5 times, indicates that the interaction among the phytochemical components in these extracts amplifies their free radical scavenging efficacy. This outcome is significant for developing functional foods or nutraceuticals with elevated antioxidant properties.

Phenolics, flavonoids, and tannins were consistently detected in all extracts, enhancing their antioxidant properties. Phenolic chemicals, distinguished by specific color alterations throughout the assessment, are acknowledged for their radical-scavenging capabilities and capacity to impede lipid peroxidation. Flavonoids, identified via colorimetric assays, demonstrate significant antioxidant and anti-inflammatory properties, underscoring their medicinal promise. Due to their astringent characteristics, tannins enhance the antioxidant activity of the extracts by forming complexes with free radicals.

Arabica coffee extracts included supplementary bioactive components such as triterpenoids, which are recognized for their anti-inflammatory and anticancer characteristics. The lack of steroids and saponins in these extracts indicates that their antioxidative properties are primarily attributed to polyphenolic substances (Ajhar & Meilani, 2020).

5. Conclusions

The study confirms that peaches (*Prunus persica* L.) and Arabica coffee (*Coffea arabica*) possess significant antioxidant activities, varying degrees depending on the solvent and preparation method. The ethyl acetate fraction of peach extract demonstrated moderate antioxidant potential (IC_{50} = 141.91 ppm), while the water fraction of Arabica coffee exhibited extreme activity (IC_{50} = 22.30 ppm). Significantly, the combination of peach and coffee extracts synergistically enhanced antioxidant activity, with the water fraction of the combination achieving an IC_{50} of 37.85 ppm, placing it in the robust category. Phytochemical analysis revealed the presence of phenolics, flavonoids, and tannins in all extracts, which are critical to their antioxidant efficacy. Additionally, Arabica coffee extracts contained triterpenoids, further enhancing their bioactive profile. These findings highlight the potential of combining natural extracts to develop antioxidant-rich formulations for therapeutic and nutraceutical applications. Future research should explore the mechanical interactions between these phytochemicals and their potential health benefits in clinical settings.

Conflict of interest

There is no conflict of interest in this research.

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