

Phytochemical Screening and Antioxidant Activity Evaluation of Selected Philippine Fruit Peels and Pulps

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Submission Date: 13-02-2024; Revision Date: 11-03-2024; Accepted Date: 12-04-2024.

ABSTRACT

Aim: This research aimed to explore the potential of underutilized plant waste products from Philippine fruits as sustainable sources of phytochemicals, assessing the viability of green extraction methods. It focused on the antioxidant activities of extracts from fruit peels and pulps, comparing these to the benchmark antioxidant, L-Ascorbic acid (Vitamin C). **Materials and Methods:** Peels and pulps from selected local fruits, including *Annona squamosa* (sugar apple; *atis*), *Musa acuminata* (banana; *lakatan*), *Sandoricum koetjape* (cotton fruit; *santol*), *Mangifera altissima* (mango; *paho*) and *Ananas comosus* (pineapple; *piña*), were utilized. Standard phytochemical screening methods were employed to identify the presence of secondary metabolites in aqueous extracts. Quantification of antioxidant activities was conducted against DPPH. **Results:** Antioxidant activities of *S. koetjape* (cotton fruit; *santol*) and *M. altissima* (mango; *paho*) fruit extracts demonstrated better or comparable efficacy to L-Ascorbic acid (Vitamin C). Evidence was established that water-based extraction of secondary metabolites, which are polar, is both feasible and environmentally sustainable. **Conclusion:** Extracts from Philippine fruit waste products are viable sources of phytochemicals with significant antioxidant activities. Furthermore, water-based, green extraction methods are beneficial for environmental sustainability and the promotion of green waste management.

Keywords: Antioxidant, DPPH Assay, Fruit Peel and Pulp, Phytochemicals.

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INTRODUCTION

Numerous fruit-derived household wastes, such as fruit peels, are rarely consumed yet usually contain more phenolics, among others, than the commonly edible tissues, which mean that many bioactive chemicals could be recovered from them. Utilizing fruit wastes as sources of bioactive chemicals may therefore have significant economic advantages and is increasingly attractive in terms of the future environmental sustainability.^[1-4]

The intricate chemical composition of plants is a testament to their potential health benefits, a notion that has been acknowledged through the varied biological roles of secondary metabolites. These compounds have been foundational to the medicinal practices of ancient civilizations due to their demonstrated variability in effects.^[5-7] As a defense mechanism, plants produce an array of phytochemicals, some of which possess antibacterial, antifungal and antiviral properties, thereby protecting the plant from various pathogens.^[8,9] Over a thousand of cataloged phytochemicals exists, deriving from diverse sources such as whole grains, fruits, vegetables, nuts and herbs. Among them, polyphenols, phytosterols, isoprenoids, carotenoids, saponins, dietary fibers and specific polysaccharides have been identified as key bioactive substances. Although certain phytochemicals share similarities with vitamins, they are not deemed essential for survival but are crucial for

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DOI: 10.5530/ajbls.2023.13.25

protecting cells against damage from environmental and metabolic byproducts. These phytochemicals play a significant role in neutralizing free radicals, thus safeguarding cellular integrity and reducing disease risk. The antioxidant capacity of many phytochemicals not only provides a shield against various diseases but also supports a balanced immune system and exhibits antibacterial, antidepressant and antihemorrhagic properties.^[10-15]

This study utilized the local Philippine fruits, which are widely available both as commercially cultivated produce and as natural sources of folkloric remedies, in which parts of the fruit trees have been used as home cures for several human diseases including viral, bacterial and fungal infection treatments. The aforementioned fruits' therapeutic benefits are supported by both legendary claims and scientific results from several studies, such as the study from the University of the Philippines where both the *atis* (sugar apple) and *santol* (lolly fruit) were considered as one of the top five Philippines fruits having the highest phenolic and flavonoid contents among the 30 different fruits investigated.^[16] In addition, fruit peels and pulps of *Mangifera indica* (mango) and *Ananas comosus* (pineapple) were also in the top list with their very high antioxidative capacities.^[17]

The purpose of this study was to identify phytochemical constituents in the peels and pulps of five distinct Philippine fruits and to evaluate their antioxidant properties. This could provide a scientific validation for their potential applications beyond being merely discarded as waste. The study was structured to achieve several key goals: firstly, to conduct an initial phytochemical screening using established protocols; secondly, to assess antioxidant capacities through the DPPH (2,2-diphenyl-1-picrylhydrazyl) assay; and thirdly, to compare the percentage of DPPH radical scavenging

ability of the peels and pulps from *Annona squamosa* (*atis*), *Musa acuminata* (*lakatan*), *Sandoricum koetjape* (*santol*), *Mangifera altissima* (*paho*) and *Ananas comosus* (*piña*) with that of the known antioxidant, L-ascorbic acid (Vitamin C).

MATERIALS AND METHODS

The native fruits, namely: *atis* (sugar apple; *Annona squamosa*), *lakatan* (banana; *Musa acuminata*), *paho* (mango; *Mangifera altissima*), *piña* (pineapple; *Ananas comosus*) and *santol* (cotton fruit; *Sandoricum koetjape*) were collected from the local area of Misamis Oriental, Philippines. The experimental instrumentations and analyses were conducted in the Department of Material Science laboratory of the University of Maryland, College Park, Maryland, USA. All chemicals used were high purity grade and supplied by Flinn and Fisher Scientific companies.

Preparation of Fruit Peel and Pulp Extract

About 300 g of peels and pulp from each fruit were collected, washed, chopped into smaller pieces, spread on a drying pan and subjected to drying. To accelerate the drying process, an oven equipped with a thermohygrometer was used and maintained at 55°C and 25% relative humidity. Representative portions of about 1 g for each sample was isolated in a petri dish, to monitor the loss of moisture and was placed in the oven together with the bulk sample. The constant-weight drying method was used to determine the drying duration. A constant weight indicated by a weight difference of less than 0.001 g between 3 hr weighing intervals of the one-gram portions. Then, the dried samples were vacuum-sealed and stored in a freezer until its extraction. Fruit peel and pulp extract was prepared by measuring 25 g of the dried sample in 100 mL of

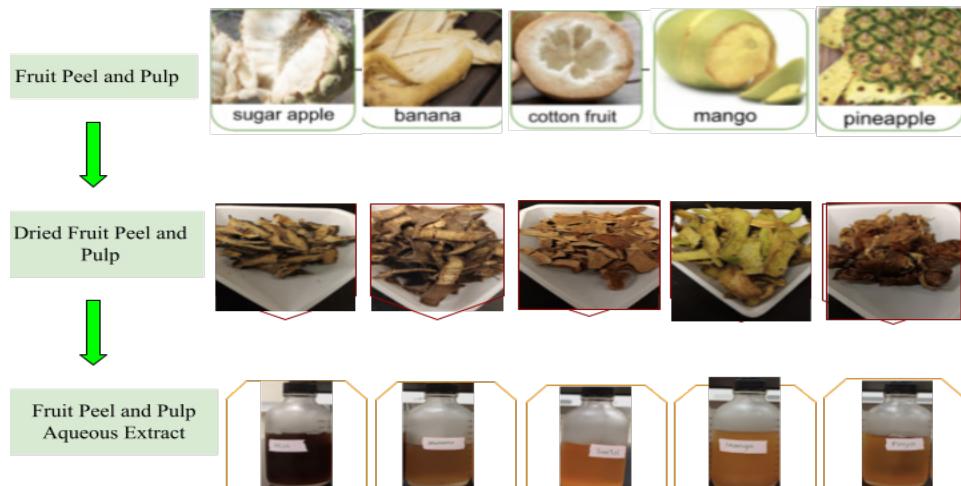


Figure 1: Dried fruit peels and pulps and their corresponding aqueous extracts.

deionized water and heating up the mixture to 55°C for about 50 min. The crude aqueous extracts were cooled to room temperature, filtered and stored at 4°C until its use. Figure 1 shows the fresh, dried and extracted states of the fruit peels and pulps.

Phytochemical Screening

Qualitative phytochemical analysis was carried out using different reagents, detailed in Table 1, to identify the secondary metabolites present in the obtained fruit peel and pulp aqueous extracts.^[18,19]

Table 1: Phytochemical screening methods and reagents used.

Phytochemicals	Reagents/Test	Indicators
Phenols	FeCl₃ Test 2 mL of 5% FeCl ₃ solution combined with 1 mL of extract.	A dark reaction mixture indicates phenolic presence.
	Wagner's Reagent Test Add 2 drops of iodine solution (2 g KI and 3 g iodine diluted to 100 mL) to 3 mL of extract.	Reddish-brown sediment suggests alkaloids.
Alkaloids	Alkaline Reagent Test 3 mL of extract mixed with 1 mL of 10% NaOH.	Intense yellow hue denotes flavonoids.
	Bontrager's Test 5 mL chloroform and 5 mL ammonia mixed with 0.2 mL extract.	Bright pink in aqueous phase signals anthraquinones.
Flavonoids	Salkowski's Test 5 mL extract, 2 mL chloroform, 3 mL sulfuric acid layered.	Reddish-brown interface layer shows terpenoids.
	FeCl₃ Test 1 mL water, 2 drops FeCl ₃ to 0.5 mL extract.	Blue-black colorization points to tannins.
Anthraquinones	Frothing Test 5 mL extract shaken vigorously, left to stand for 5 min.	Persistent honeycomb froth indicates saponins.
Terpenoids		
Tannins		
Saponins		

DPPH Assay: Antioxidant activity of fruit peel and pulp extracts

The capacity for quenching DPPH radicals was assessed to gauge the antioxidant strength of various fruit peel and pulp extracts. The propensity of plant-derived compounds to offer antioxidant protection has been well-documented in literature.^[10-12,17] Such antioxidants are crucial in safeguarding the body from

the detrimental impact of free radicals. In this context, the present work involved a scrutiny of the antioxidant potential inherent in the extracts of fruit peel and pulp by deploying the DPPH (1,1-diphenyl-2-picrylhydrazyl) radical neutralization technique. The DPPH radical, characterized by its deep violet hue, is stable due to the delocalization of the spare electron over the molecule, making it an ideal candidate for evaluating the hydrogen-donating capacity of antioxidant substances. The assay is lauded for its straightforwardness, rapidity and cost-effectiveness, being the preferred method for assessing the antioxidant capabilities of various plant-based and food entities.

For the assay, the colorimetric change induced by the reaction of antioxidants with the DPPH radical was utilized as an indicator of activity. This reaction typically leads to a color transition from violet to yellow, signifying the acceptance of a hydrogen atom by DPPH. To conduct the assay, a 0.2 mM solution of DPPH was formulated by dissolving 7.89 mg in 99.5% ethanol and the solution was then shielded from light for 2 hr. Subsequently, a volume of 2 mL of this solution was amalgamated with an equal amount of the fruit extract. The mixture was agitated to ensure thorough mixing and then left to stand at ambient temperature for 30 min. The interaction of the DPPH radical with an antioxidant, which donates a hydrogen atom, leads to a reduction in DPPH, observable through a UV-visible Spectrophotometer (Cary 300 Bio) at an absorbance wavelength of 524 nm. L-ascorbic acid, at a concentration of 0.5 mM, served as a comparative standard.

To quantify the Radical Scavenging Activity (RSA) exhibited by the fruit extracts, the following formula was employed:

$$\text{RSA (\%)} = (A_1 - A_2)/A_1 \times 100$$

Here, A_1 represents the absorbance of the control DPPH solution with ethanol, while A_2 the absorbance after the addition of the fruit extract.

RESULTS

Phytochemical Screening

Preliminary qualitative screening of the aqueous extracts from selected Philippine fruit peels and pulps identified the presence of secondary metabolites, including phenolic compounds, alkaloids, flavonoids, anthraquinones, terpenoids, tannins and saponins. The phytochemical presence was determined using various reagent tests, with results summarized in Table 2. Notably, the cotton fruit and mango exhibited a relatively

higher concentration of these secondary metabolites compared to the other tested fruits. In addition, using Salkowski reagent terpenoids were detected at a greater

intensity for all five-fruit peel and pulp extracts. On the other hand, using Bontrager's test, anthraquinones were not detected in banana and pineapple extracts.

Table 2: Qualitative phytochemical screening of the different fruit peel and pulp extracts.

Phytochemicals (Reagent Test)	<i>A. squamosa</i> (sugar apple; <i>atis</i>)	<i>M. acuminata</i> (banana; <i>lakatan</i>)	<i>S. koetjape</i> (cotton fruit; <i>santol</i>)	<i>M. altissima</i> (mango; <i>paho</i>)	<i>A. comosus</i> (pineapple; <i>piña</i>)
Phenols (Ferric chloride test)	+	+	+++	+++	+
Alkaloids (Wagner's reagent)	+	++	+++	++	++
Flavonoids (Alkaline reagent)	+	++	++	+++	++
Anthraquinones (Bontrager's test)	+	-	++	+++	-
Terpenoids (Salkowski test)	++	+++	+++	+++	++
Tannins (Ferric chloride test)	+	+	+++	+++	+
Saponins (Frothing test)	+	+	++	+	+++

Note: presence (+) and absence (-).

DPPH Assay: Antioxidant Activity

The antioxidant activity, measured through the DPPH radical scavenging assay, varied among the fruit extracts. The results, expressed as the percentage of radical scavenging activity, are detailed in Table 3 and visualized in Figure 2. The mango extract exhibited the highest DPPH radical scavenging activity with a percentage of 98.53%, followed by cotton fruit with 90.47%. These results indicate a higher antioxidant potential relative to the reference standard, L-ascorbic acid (Vitamin C), which showed an 80.02% activity. The banana and sugar apple extracts also demonstrated significant antioxidant activity, comparable to the reference standard. In contrast, the pineapple extract exhibited the lowest activity with 64.12%.

Table 3: DPPH radical scavenging activity of the different fruit peel and pulp extracts.

Fruit Peel and Pulp Extract	Radical Scavenging Activity (%)	Std Deviation
<i>L-Ascorbic Acid</i> (Vitamin C)	80.02	0.13
<i>A. squamosa</i> (sugar apple; <i>atis</i>)	78.86	0.45
<i>M. acuminata</i> (banana; <i>lakatan</i>)	81.59	0.64
<i>S. koetjape</i> (cotton fruit; <i>santol</i>)	90.47	0.45

<i>M. altissima</i> (mango; <i>paho</i>)	98.53	0.55
<i>A. comosus</i> (pineapple; <i>piña</i>)	64.12	0.80

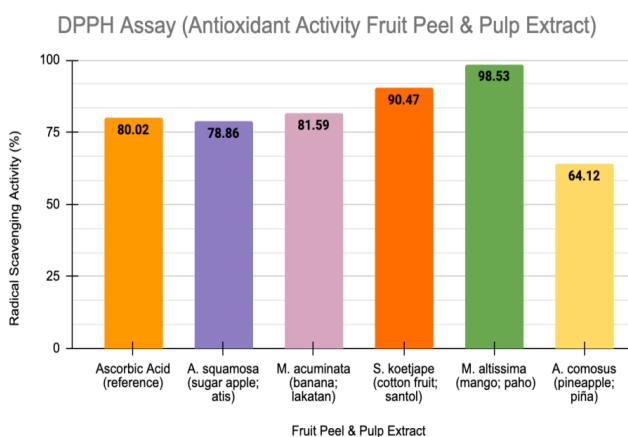


Figure 2: DPPH radical scavenging activity of the different fruit peel and pulp extracts in comparison with the Ascorbic acid reference.

The assay's colorimetric changes were visually represented in Figure 3, illustrating the degree of radical scavenging manifested by the color transition in the samples tested. When the free radical DPPH solution and antioxidants were combined, DPPH-H (2,2-diphenyl-1-hydrazine) solution was formed. The purple color of DPPH was changed into bright yellow when scavenged, which implies that the fruit peel and pulp extract are

excellent hydrogen sources (antioxidants) resulting in the reduction of DPPH to DPPH-H. The degree of discoloration indicates the scavenging potential of the antioxidant compounds present in fruit peel and pulp extracts in terms of hydrogen donating ability. Figure 4 shows the chemical reaction between the DPPH and antioxidant to produce DPPH-H.^[20]



Figure 3: Colorimetric changes associated with the radical scavenging activities of the different fruit peel and pulp extracts.

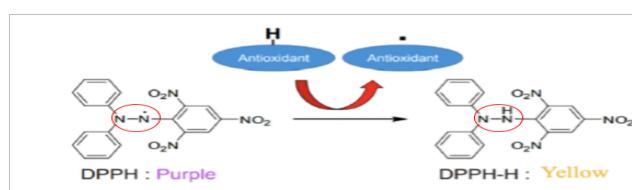


Figure 4: The redox reaction, DPPH + antioxidant → DPPH-H, accompanying the colorimetric change in the DPPH assay.

DISCUSSION

The qualitative phytochemical investigation revealed a varied presence of secondary metabolites among the selected Philippine fruit peel and pulp aqueous extracts. The higher intensity of secondary metabolites such as phenolic compounds, alkaloids, flavonoids, anthraquinones, terpenoids and tannins in cotton fruit and mango peels and pulps suggests a robust phytochemical profile that could correlate with their marked antioxidant activities. Terpenoids, known for their diverse biological activities, were identified across all five fruit extracts, indicating a potential for broad-spectrum biological effects.

The findings of this study are in congruence with the results of previous related studies where fruits peel and pulp exhibited remarkable amounts of phenols, flavonoids, alkaloids, tannins, terpenoids, anthraquinones and saponins.^[1-3,10,13,16-17] Additionally, our results were comparable to the results of other studies which

utilized other parts of the plants including leaves, stem, pods and roots.^[8-9,11-12,19] This underscores the potential of fruits peel and pulp as viable phytochemical source without adversely affecting the fruit trees while also repurposing such waste products. Whereas previous research has predominantly utilized organic solvents such as methanol for extraction, our study diverges by exclusively employing water as the extraction medium. This decision is grounded in water's polar nature, which effectively dissolves a broad range of secondary metabolites, particularly phenols and flavonoids, which are considered potent antioxidants. The use of water not only reduces the toxicity associated with the process but also promotes safer and more sustainable extraction practices.

The DPPH assay's results, exhibiting high radical scavenging activity for the *S. koetjape* (cotton fruit; *santol*) and *M. altissima* (mango; *paho*) peel and pulp extracts, underscore the potential of these fruit wastes as sources of natural antioxidants. The extracts' performance exceeded the radical scavenging activity of L-ascorbic acid, positioning them as compelling candidates for natural antioxidant sources. This finding supports previous studies that have highlighted the rich antioxidant potential of various fruit peels and pulps. Interestingly, the sugar apple and banana peel and pulp extracts also displayed significant antioxidant activities, relative to L-ascorbic acid. The lower antioxidant activity in the pineapple peel and pulp extract, as indicated by the persistence of the purple color in the DPPH assay, suggests a lesser concentration of effective radical scavenging compounds or a different mechanism of action that may require further investigation to fully understand.

The results of the phytochemical screening and DPPH assay are in harmony, suggesting a promising avenue for the use of these fruit waste extracts in therapeutic and nutraceutical applications. The potent antioxidant activities of the fruit peel and pulp extracts; *A. squamosa* (sugar apple; *atis*), *M. acuminata* (banana; *lakatan*), *S. koetjape* (cotton fruit; *santol*), *M. altissima* (mango; *paho*) and *A. comosus* (pineapple; *piña*), lend scientific credence to their traditional uses in treating various ailments. The distinct presence of phenolic components like flavonoids, terpenoids, alkaloids and other phenolic compounds may be responsible for the antioxidant activity seen in the selected fruits peel and pulp under study. The hydroxyls in their phenolic groups that are capable of donating hydrogen attributed to their antioxidant characteristics.^[10-11]

Moreover, by adhering to the principles of green chemistry, including waste prevention, atom economy

and the use of safer solvents and auxiliaries, this study exemplifies a commitment to more sustainable, safer laboratory practices.^[4] Despite the diversity in extraction methods and the parts of the plant used across studies, our findings underscore the comprehensive value of even the traditionally overlooked components—such as peels, pulp and pomace—as rich sources of beneficial bioactive compounds. This underlines the importance of adopting environmentally friendly practices not only in scientific research but also in broader applications, paving the way for future endeavors that aim to bridge the gap between traditional knowledge and modern scientific validation. By emphasizing innovative waste repurposing and safer extraction methods, this research highlights the pathway to harnessing phytochemicals from fruit waste. The study contributes to a deeper understanding of the bioactive potential of fruit waste products, reinforcing the benefits of their valorization in a circular economy and sustainable waste management practices.

CONCLUSION

This study unveiled the substantial phytochemical content and antioxidant capabilities within the peels and pulps of five selected Philippine fruits: *A. squamosa* (sugar apple; *atis*), *M. acuminata* (banana; *lakatan*), *S. koetjape* (cotton fruit; *santol*), *M. altissima* (mango; *paho*) and *A. comosus* (pineapple; *piña*). Through both phytochemical screening and DPPH radical scavenging assays, it was determined that *S. koetjape* (cotton fruit; *santol*) and *M. altissima* (mango; *paho*) possess remarkable bioactive compounds and pronounced antioxidant potential, underscoring the bioactive compound richness in these fruit residues.

The findings substantiate the traditional utilization of these fruits in various therapeutic practices, attributing their health benefits to the identified phytochemicals with known antimicrobial, anti-inflammatory, antioxidant and potential antitumor properties. These attributes, coupled with their role in neutralizing free radicals, pave the way for leveraging fruit waste as a resource for developing functional food derivatives and enhancing the value of herbal medicines. Moreover, the research highlights the dual benefits of employing fruit waste for antioxidant extraction: promoting environmental sustainability through waste reduction and offering economic advantages by tapping into underutilized resources. This aligns with the green chemistry principles, advocating for more sustainable practices in both food and pharmaceutical industries. In essence, this study not only contributes to the scientific

validation of the medicinal and nutritional value of Philippine fruits but also promotes a circular economy model, encouraging the repurposing of fruit waste into value-added products.

ACKNOWLEDGEMENT

We would like to acknowledge the financial support of NSF, REU/RET Site: Summer Research Experiences in Renewable and Sustainable Energy Technology (ReSET) and the Maryland Energy Innovation Institute (MEI²) and University of Maryland Department of Materials Science and Engineering To Dr. Isabel K. Lloyd and Dr. Mohamad I. Al-Sheikhly for the outstanding laboratory assistance, coordination and exceptional support for the completion of this study.

FUNDING

This work was supported in part by the NSF [grant number DMR2149982].

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

DPPH: 2,2-diphenyl-1-picrylhydrazyl; **DPPH-H:** 2,2-diphenyl-1-hydrazine; **RSA:** Radical Scavenging Activity; **UV:** Ultraviolet; **NaOH:** Sodium hydroxide; **KI:** Potassium iodide; **FeCl₃:** Ferric chloride

SUMMARY

This study explored the rich phytochemical and antioxidant potential of peels and pulps from selected Philippine fruits (*A. squamosa*, *M. acuminata*, *S. koetjape*, *M. altissima* and *A. comosus*), with *S. koetjape* and *M. altissima* demonstrating particularly pronounced bioactive properties. Through qualitative phytochemical screenings and quantitative DPPH assays, the research underscores the viability of utilizing fruit waste as a sustainable source for extracting valuable antioxidants, thereby aligning with green chemistry principles. The significant antioxidant activity observed suggests these extracts could be pivotal in developing new health-promoting products, potentially reducing the risk of chronic diseases associated with oxidative stress. Moreover, the study reinforces the importance of sustainable practices in the food and pharmaceutical industries, promoting the reuse of agricultural by-products to mitigate environmental impact. By

advocating for the repurposing of fruit waste into functional foods and nutraceuticals, this research contributes to a circular economy, offering a dual solution to environmental waste management and the valorization of underexploited resources. Ultimately, this study bridges the gap between traditional knowledge and contemporary scientific validation, underscoring the untapped potential of Philippine fruit wastes in promoting health, sustainability and economic development, thereby encouraging further exploration and utilization of these abundant natural resources.

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Cite this article: Pates MD, Walag AMP, Rosario RMD. Phytochemical Screening and Antioxidant Activity Evaluation of Selected Philippine Fruit Peels and Pulps. *Asian J Biol Life Sci.* 2024;13(1):197-203.