

## ONION PEEL AS A PROSPECTIVE ANTIOXIDANT SOURCE

**M. Selva Susi Priya** (Asst.Prof of Microbiology, Sardar Raja Arts & Science College  
Vadakkangulam)

### ABSTRACT

Onion peels, a common agricultural waste product, are rich in bioactive compounds, including antioxidants, that have potential applications in the food and pharmaceutical industries. This study investigated the antioxidant activity of extracts from Bhima Shubhra onion peels using various solvents—ethyl acetate, chloroform, ethanol, and methanol. The peels were subjected to extraction, and the resulting bioactive compounds, including saponins, phytosterols, total phenolics, and flavonoids, were quantified. Antioxidant activity was assessed using DPPH and ABTS assays. The results demonstrated that the type of solvent significantly influenced both the bioactive compound content and antioxidant potential. The ethanol extract exhibited the highest total saponin and phytosterol levels, while methanolic extracts showed the strongest antioxidant activity in both assays. Acetone extracts had the highest levels of total saponins and phytosterol. However, antioxidant activity was not directly correlated with the total phenolic or flavonoid content, indicating the importance of specific compounds, such as quercetin, in driving the antioxidant effects. This study highlights the potential of onion peel waste as a sustainable source of natural antioxidants and suggests that solvent selection plays a crucial role in optimizing the extraction of bioactive compounds. Further research is required to identify and characterize the specific compounds responsible for the antioxidant activity in onion peel extracts, with the aim of enhancing their utility in food and pharmaceutical applications.

### Introduction

About 107 million tons of onions (including shallots) emerged worldwide in 2021, putting them as the second most grown vegetable crop in the world [FAOSTAT,2013]. Environmental pressures are increased by the industrial processing of onions, which produces a lot of agricultural waste, principally dry peels, that is burned or dumped in landfills. Because of potential negative effects on the environment, there is continuous concern about the growing amounts of plant-residue agricultural waste that arise from the increase in crop food production worldwide [Maji et al., 2020]. Global environmental sustainability efforts may be negatively impacted and hampered by this created garbage [Ugwuoke et al., 2018]. As a result, it is now recognized that efforts should be taken to transform the agricultural crop waste produced during production and processing into products and resources that will benefit humanity. Research has been conducted on onion peel waste as a source of bioactive chemicals and dietary fibre for the food sector [Benitez et al.,2011], as well as a source of valuable biomolecules [Sagar et al, 2022], organic fertilizer [mallek et al 2007], and renewable energy source [Romano et al 2011,Segundo et al., 2022].

"Mining" the leftover biomass for chemicals generated from plants is one possible application for agricultural waste. Since plant tissues are known to contain a wide variety of physiologically active substances, they may be utilized as a source of raw materials to extract valuable phytochemicals [Kumar et al,2023,Mihajilov et al 2020]. Various agricultural residues have, indeed, come to light as possible sources for therapeutic phytochemicals

demonstrating a range of protective properties, including antioxidant and antibacterial capabilities [Kasapiduo et al 2015]. Today, agro-industrial wastes' bioactive derivatives can be found in a variety of commodities, that include functional dinner and cosmetics [Rodrigues et al 2017,Ngwairi et al, 2022,Oleszek et al, 20233].There is growing interest in using natural antioxidants obtained from herbs that are free of crop tissues in view of the possible health hazards that have been found to be connected to some synthetic antioxidants [Lourenco et al, 2019]. Given the great number of biologically active substances it contains, as well as its low cost and global availability, onion peel tissue is one of the greatest potential sources of natural inhibitors in this context [Bains et al, 2023].

In onion peel extracts, flavonoids, flavanols, phenolics, anthocyanins, tannins, ferulic acid, and vanillic acid were each found in numerous investigations [Kumar et al., 2022]. The flavonoid quercetin, which can be found in mono- and diglyco-sides or as a free aglycon, is the most often isolated antioxidant-containing molecule from onion peels [Stoica et al , 2013].Onion cultivar and variety have a significant impact on the chemical makeup of onion peels; red peels often exhibit more antioxidant activity than yellow and white peels [Albishi et al,2013]. This is consistent with data for other plant species, which also documented genotypic variation in the chemical makeup of atoms that are actively used in plant tissues [Gorelick et al, 2020, Shiponi et al 2021, Dangiger et al, 2021,Saloner et al 2022, Kumeret al 2022]. The quantities of Phyto molecules in the extracts can also be influenced by the solvents and extraction method used [Kumar et al 2022]. The amount of bioactive natural components in the extracted extracts can also be greatly impacted by mechanically preparing the peel before extraction [AbouZid et al 2008].Many studies has been conducted into the potential use of onion peel bio-waste as a source of pharmacological and antioxidant chemicals because of its potential for significant economic benefit [Hazama et al 2005, Prakash er al 2007]. Dried onion peel powder and ethanolic extract were found to increase the total antioxidant status of plasma in elderly rats while lowering the levels of 8-isoprostane in brain tissue and thiobarbituric acid reactive substances (TBARS) in liver tissue [park et al 2007]. However, adding pills containing 60% aqueous ethanol onion peel extract to the diet of young Korean women did not significantly alter erythrocyte antioxidative parameters or plasma antioxidative vitamins, but it did improve the lipid profile in plasma, suggesting that onion peel has a cardioprotective effect [Kim et al 2013]. Additionally, extracts from onion peels have antibacterial, anti-inflammatory, anti-obesity, antidiabetic, and anti-cancer properties [Stoica et al 2023]. Many parts of the world, traditional medicine and homemade cures have been known to use onion peel teas to treat a variety of illnesses, including gastrointestinal issues [Ayanniyi et al 2022]. The present study, therefore, focused on the antioxidant activity of onion peel extracts, with the goal to evaluate the potential of this bio-waste as a source of natural bioactive compound.

## **MATERIALS AND METHODS**

### **PLANT MATERIAL**

The experiments were conducted with peels of a white skinned onion commonly known as Bhima Shubhra (*Allium cepa* L.). Its can grow well in kharif seasons (June to October).These breed was grown mostly in Tamil Nadu.

### Onion Peel Extraction

The onion peels were extracted by three organic solvents that differ in polarity in the following order: ethyl acetate>chloroform>ethanol. For the extraction, dry onion peel was pulverized in a blender, and pure solvents were poured over the pulverized plant material (with a 5 mL:1 g ratio of solvent: plant material). The flasks containing the extracted samples were closed and kept in the dark for seven days at room temperature, and the extraction solvents were then removed using a rotary evaporator and the obtained crude extracts were kept at 4°C until analyses.

### Total Saponins and Phytosterol Content

For total Saponins content (TFC) the modified Flores et al. [14] protocol was followed to perform the extraction using chemical solvents. An Erlenmeyer flask containing 150 ml of 70% ethanol was filled with 50 g of seeds of each genotype of quinoa for 48 hours. After filtering the mixture, the process was carried performed again after waiting for twenty-four hours. After that, the extracts were incorporated and dried in a water bath set at 65°C. N-butanol was used to remove the residue after it had previously been dissolved in 10 ml of distilled water. In a rotary evaporator, the extracts were then dried out until a dry deposit was produced. (Flores et.al., 2013)

For Total phytosterol content the Cold Saponification and Liquid-Liquid Extraction method was used. This involves saponifying lipids from the plant material with an alcoholic solution (e.g., KO in ethanol) followed by liquid-liquid extraction using solvents like ethyl acetate or hexane to separate non-saponifiable compounds, including phytosterols (Dikshit et.al., 2020).

### Activity for DPPH Scavenging

The 2,2-diphenyl-1-picrylhydrazil (DPPH) radical scavenging method, which employs the stable DPPH radical as a reagent, was used to assess the antioxidant activity of the onion peel extracts and two chosen standard compounds (vitamin C and butylated hydroxyanisole, BHA) [Blois et al 1958]. 2700 µL of DPPH methanol solution (0.04 mg/mL) was mixed with 300 µL of the examined extracts, which were made by dissolving different quantities of the crude extract in methanol at different concentrations ranging from 0.01 to 0.15 mg/mL. The absorbance of the residual DPPH radical at 517 nm was measured after the reaction mixture was let to stand at room temperature in the dark for 30 minutes. For the samples, vitamin C, and BHA, all measurements were made three times, using methanol as a blank. The following formula was used to determine the DPPH-radical scavenging activity:

$$\text{Scavenging activity (\%)} = \frac{(A_0 - A_1)}{A_0} \times 100$$

where A<sub>0</sub> is the absorbance of the original DPPH solution and A<sub>1</sub> is the absorbance of the samples. The IC<sub>50</sub> value was the sample concentration that resulted in a 50% reduction in the initial DPPH concentration.

### Radical Scavenging Activity with ABTS

The modified Miller and Rice-Evans method [35] was used to perform the radical-scavenging activity of ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)). 5 mL of potassium persulfate (2.46 mM) was combined with 19.2 mg of ABTS to create the ABTS<sup>+</sup> (ABTS radical cation) solution. 1 mL of the ABTS<sup>+</sup> solution was diluted in 100–110 mL of water after being stored at room temperature for 12–16 hours in the dark. This

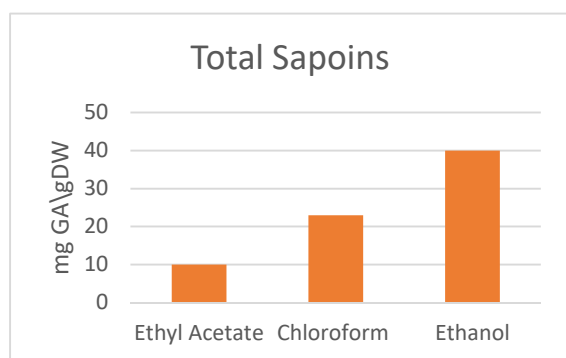
resulted in an absorbance of 0.7–0.02 at 734 nm. 75  $\mu$ L of the extract in methanolic solution (the extract concentration was 0.1 mg/mL) or BHA solution was combined with 3 mL of diluted ABTS $\cdot^+$  solution to create the reactive medium, which was then incubated for 30 minutes at 30 °C. Using water as a blank, the absorbance was measured at 734 nm. A Vitamin C calibration curve (0–2 mg/L) was used to determine the extracts' ABTS radical scavenging activity, which was then expressed as Vitamin C (VitC)/g of DW of the extract. Three duplicates of each measurement were made.

## RESULT

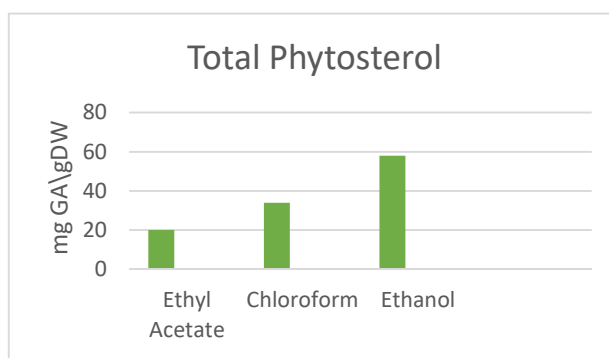
### Antioxidant Activity

Total sapoins and phytosterol contents in the extract were significantly affected by solvent used for extraction (Figure 1). Total sapoins content ranged from 10.5 to 40.3 mg GA/g DW. The ethanol extract had the highest content of sapoins compound, followed by chloroform and ethyl-acetate extract contain the lowest content of total sapoins compound. The Highest total phytosterol level was also identified in the ethanol extract (Figure 1), lower level was detected in ethyl-acetate. The order of total phytosterol levels in the extracts was ethanol > chloroform > ethyl-acetate extracts.

A

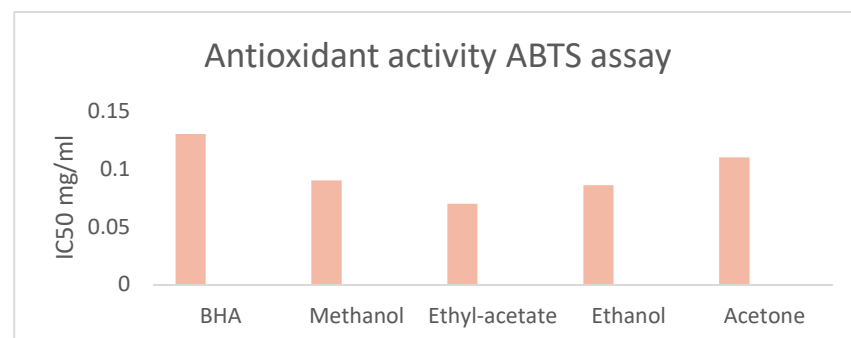


B

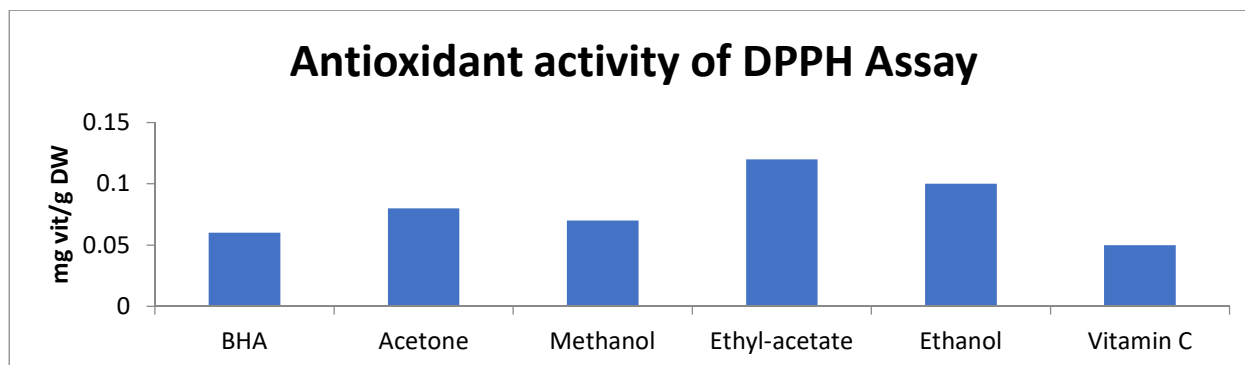


**Figure 1:** Total Sapoin (A) and phytosterol (B) contents in ethyl-acetate, chloroform extracts. Presented results are means  $\pm$  SD. Different letters above the means represent significant difference by Tukey HSD post hoc test ( $p < 0.05$ ). The antioxidant activity of the extracts was determined by colour reduction of the DPPH and ABTS solutions used for testing the radical scavenging potential. The acetone extract exhibited similar DPPH activity to the positive control, BHA indicating a high applicable potential. The lowest value was found for positive control vitamin C (0.08 mg/ml) followed by the acetone extract and positive control BHA (0.11 mg/ml and 0.13 mg/ml). The methanol, ethyl-acetate, ethanol had a higher DPPH activity with no statistical difference between extracts.

A



B



**Figure 2** : Antioxidant activity measured by the ABTS(A) and DPPH (B) methods in methanol , ethanol, ethyl-acetate and acetone extracts. The antioxidant activity was compared to that of butylated hydroxyanisole (BHA) and Vitamin C as standard .Presented results are means SD different letters above the means represent significant difference following Tukey HSD post hoc Test ( $p < 0.05$ )

The values obtained from the antioxidant activity of ABTS assay range from 0.95 to 2 mg vit C/g DW figure. The highest activity was noted on the acetone extract and lowest for the ethyl acetate.

## DISCUSSION

Waste from onion peels is a rich source of diverse chemicals with a range of biological activity that may find application in the food and pharmaceutical industries. Many factors influence the amount of phytochemicals in the waste. This is the type of onion used to make the trash [Beesk et al 2010]. The chemical and biological properties of waste from the onion variety Bhima Subhra , which is distinguished by the outer skins' distinctive dark bronze color and their abundance surrounding the bulb, were the main focus of this investigation. To ascertain which of the four solvent types—methanol, ethanol, acetone, and ethyl acetate—is most suited for producing extracts with strong antioxidant and antibacterial properties, chemicals were extracted from the onion waste. In the studied onion peel extracts from the Bhima Subhra variety, saponins were detected in the range of 10 to 40.58 mg GA/g DW, and total phytosterol content was in the range of 20 to 60.08 mg Qu/g DW. Other onion types were found to have equivalent amounts of flavonoids and total phenolics in their peel extracts [Benitez et al 2011, Burri et al 2017]. In line with the findings of Skerget et al. (2009), and she found higher quantities of total phenols in extracts made with pure acetone as opposed to ethanol extracts [Skerget et al, 2009], acetone extraction produced the highest levels of total phenols and flavonoids. However, methanol outperforms ethanol and acetone in the extraction of individual flavonoids and total phenols from a yellow onion variety, according to (Kim et al 2013). Ethyl-acetate was found to be an effective solvent for the extraction of polyphenols of intermediate polarity [Singh et al, 2009], while ethanol is a preferred solvent for the extraction of polyphenols, methanol is a good solvent for the extraction of low-molecular-weight polyphenols, and acetone is the best solvent for the

extraction of higher-molecular-weight flavonols [Do et al 2014]. These variations can be explained by the different chemical compositions of the various onion peels, which comprise a complex mixture of compounds that are selectively soluble in solvents of different polarities. As a result, it was discovered that the more polar fractions (methanol and ethanol) of an onion peel methanol extract had a larger total phenol concentration than the fractions with less polar solvents (ethyl acetate, chloroform, and n-hexane) [Nile et al 2017]. This is congruent with our results, as the use of ethyl acetate for extraction provided the lowest total phenol levels. In contrast, when comparing the extraction efficiency of a series of solvents (water, methanol, ethyl acetate, and n-butanol ethanol), Khalili et al. (2022) reported that ethyl acetate was the best solvent for the extraction of total phenols and flavonoids. In our study, only the content of total flavonoids was higher in the ethylacetate extract compared to the methanol and ethanol extracts (Figure 1). Two in vitro antioxidant tests, DPPH and ABTS, which are based on the single electron and/or hydrogen atom transfer (SET/HAT) mechanism, were used to assess the extracts' antioxidant activity [Prior et al 2005]. In the DPPH experiment, the IC<sub>50</sub> values of the other extracts did not differ statistically, but in the ABTS assay, the methanolic extract had the highest antioxidant activity in both tests, followed by the ethanol, acetone, and ethyl acetate extracts (Figure 2). The primary carrier of onion peel's antioxidant capacity is quercetin, which has been demonstrated to possess greater antioxidant power than its mono- and di-glycosides because of the presence of free OH groups on the ring [Celano et al 2021]. The studied methanolic extract had the highest levels of quercetin and quercetin hexoside. In contrast to the acetone and ethyl acetate extracts, the ethanol extract with the lowest quercetin quantity found in the ABTS test demonstrated a greater antioxidant ability. This suggests that additional substances may also enhance the extract's antioxidant potential. According to Ramos et al. (2006), quercetin's oxidative byproducts in yellow onion peel functioned as superior antioxidants. It can be noted that the antioxidant activity levels obtained for the extracts in this study by both antioxidant assays are not correlated with the total contents of phenols and flavonoids. Onion peel extracts' antioxidant properties have previously been demonstrated to be strongly correlated with their total phenol and flavonoid levels [Sagar et al. 2020], while there are some instances where the antioxidant. The primary determinant of extracts' antioxidant capacity is their individual component properties rather than their total phenol and flavonoid concentrations [Burri et al 2017]. In contrast to red onion extract, which included larger quantities of isorhamnetin, kaempferol, quercetin 4-monoglucoside, quercetin aglycone, and quercetin 3,4-diglucoside, Fredotović et al. showed that yellow onion peel extract inhibited DPPH radicals at a higher degree [Piechowiak et al 2022]. The chemicals found in onion skins that have the greatest impact on the antioxidant activity of onion peels include quercetin, vanillic acid, kaempferol, and p-hydroxybenzoic acid [Burri et al 2017].

## CONCLUSION

Onion peels, a waste product from onion processing, are a promising source of natural antioxidants with potential applications in the food and pharmaceutical industries. This study investigated the antioxidant potential of extracts from Bhima Subhra onion peels using various solvents. The findings demonstrate that the type of solvent used for extraction significantly affects the content of bioactive compounds and the antioxidant activity of the extracts. Acetone extract exhibited the highest levels of total saponins and phytosterol, while

the methanolic extract showed the strongest antioxidant activity in both DPPH and ABTS assays. This suggests that methanol may be a more effective solvent for extracting antioxidants from Bhima Subhra onion peels. The study also highlights that the antioxidant activity of extracts is not solely dependent on the total content of phytosterol and Saponins, but rather on the specific properties of the individual components present. Further research is needed to identify and characterize the specific compounds in onion peels that contribute most significantly to their antioxidant potential. Overall, this study confirms the potential of onion peels as a valuable source of natural antioxidants. By optimizing extraction methods and identifying the most potent bioactive compounds, onion peels can be utilized as a sustainable and cost-effective source of antioxidants for various applications.

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