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# Comparative study on anti-oxidant potential and αglucosidase inhibitory activity of crude extract of *Allium cepa* dry peel over *Allium cepa* ø

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**Abstract**
**Background:** *Allium cepa* (onion) is not only a widely consumed vegetable but also a plant with well recognized medicinal value. It contains flavonoids, phenolic compounds, quercetin, and sulfur compounds that help in reducing oxidative stress, regulating blood sugar, and promoting overall health. Although the onion bulb is commonly used in traditional and homoeopathic preparations, the dry outer peel usually discarded as waste has been found to contain even higher concentrations of these beneficial compounds.
**Aim and objective:** The present study was undertaken to evaluate whether the ethanolic extract of onion dry peel demonstrates stronger antioxidant and antidiabetic activities compared to the standard homoeopathic preparation, *Allium cepa* Ø (mother tincture).
**Methodology:** Dry onion peels were collected, processed, and subjected to ethanolic extraction. The resulting extract, along with a commercially available *Allium cepa* Ø (mother tincture), was analyzed at Biogenix Laboratory, Trivandrum. Antioxidant potential was assessed using DPPH and ABTS radical scavenging assays, while antidiabetic activity was evaluated through α-amylase and α-glucosidase inhibition studies.
**Results:** The ethanolic extract of onion peel exhibited consistently stronger activity than the mother tincture across all assays. In antioxidant tests, the peel extract achieved $IC_{50}$ values of 19.40 µl/mL (DPPH) and 209.10 µl/mL (ABTS), compared with 140.70 µl/mL and 1058.57 µl/mL for the tincture. Similarly, in enzyme inhibition assays, the peel extract recorded lower $IC_{50}$ values of 215.40 µl/mL (α-amylase) and 217.09 µl/mL (α-glucosidase), in contrast to 847.60 µl/mL and 700.00 µl/mL for the tincture.
**Conclusion:** These findings demonstrate that onion dry peel possesses markedly higher antioxidant and antidiabetic activity than the commonly used bulb based mother tincture. As onion peel is generally discarded as agricultural waste, this work highlights its potential as a low-cost, sustainable source of bioactive compounds with promising applications in medicine, nutraceuticals, and functional food industries.

**Keywords:** *Allium cepa* (onion), onion peel extract, antioxidant activity, antidiabetic activity, DPPH assay, ABTS assay, α-amylase inhibition, α-glucosidase inhibition, quercetin, agricultural waste utilization

## Introduction
*Allium cepa* is a multipurpose food plant and Medicinal herb, which belongs to the botanical family of Amaryllidaceae [1]. This homeopathic drug is suggested to be effective in treating disorders including asthma, inflammatory disorders, dysentery, wounds, scars, keloids, and pain [2].
*Allium cepa* contains saponins, aglycones, quercetin, cepaenes, flavonoids, organosulfur, and phenolic compounds, showing various pharmacological properties and therapeutic effects [4]. Hence it possessed many pharmacological effects including anti-oxidant, anti-microbial effects, anti-cancer effects, hypolipidemic, hypotensive, detoxification, anti-inflammatory, and analgesic [5]. It also contains a high concentration of folic acid, vitamin B6, magnesium, calcium, potassium, and phosphorus as well as vitamins and minerals [3].
Onions are particularly notable for their rich content of quercetin, a flavonoid that enhances their value both as a traditional remedy and as a modern functional food. This powerful antioxidant is found in higher concentrations near the bulb's outer layers, especially close to the skin, where it may contribute to the prevention of cardiovascular diseases and certain

cancers. Because the outer layers contain more quercetin, minimal peeling is recommended to preserve its health-promoting benefits [6].

Diabetes mellitus is a metabolic disorder, which is characterized by an abnormal postprandial increase of blood glucose level. α-Glucosidase secreted from intestinal chorionic epithelium is responsible for the degradation of carbohydrates. α-Glucosidase inhibitors slow down the process of digestion and absorption of carbohydrates by competitively blocking the activity of glucosidase [7].

According to the World Health Organization (2006), diabetes affected at least 171 million people globally, and its prevalence is rising at an alarming rate. Estimates from the American Diabetes Association (2005) predict that by 2030, this figure could double. The condition is widespread in both developed and developing nations with the steepest growth in cases anticipated in Asia and Africa. In the United States alone, around 23.6 million individuals approximately 7.8% of the population were living with diabetes in 2007. The National Diabetes Information Clearinghouse reported that managing the disease costs the country about $132 billion annually. In India, projections suggest an increase from 15 million cases in 1995 to 57 million by 2025, positioning it as the nation with the largest diabetic population in the world (Boyle et al., 2001) [8].

Antioxidants are believed to play a very important role in the body defense system against the various reactive oxidant species that are generated during several physiological and pathological processes [9]. Excess in the generation of free radicals leads to cellular stress that can damage the DNA, proteins, and other cellular structure and function [10].

The rise of multidrug-resistant bacteria has intensified the search for alternative therapeutic agents. While synthetic antibiotics remain widely produced and used, their declining effectiveness and potential health risks highlight the urgent need for safer options. In contrast, medicinal plants offer a promising solution. Their bioactive compounds such as phenolics, alkaloids, saponins, and terpenoids occur naturally as complex mixtures within the plant cell, making it harder for pathogens to develop resistance. Over the past three decades, these compounds have demonstrated potent antibacterial effects through mechanisms including membrane disruption, protein binding, metabolic interference, anti-quorum sensing, and biofilm inhibition. Harnessing this potential will require advanced omics technologies and network pharmacology to identify optimal combinations, either among plant molecules themselves or in synergy with existing antibiotics [11].

According to the Homeopathic Pharmacopoeia of India, the British Homeopathic pharmacopoeia, and the Homeopathic pharmacopoeia of United States. *Allium cepa* Mother tincture is prepared from it's Red matured bulb. But studies have shown that the outermost dry peels of onion also have several biochemical properties such as Anti-oxidant, Anti-inflammatory, and Analgesic, Hypolipidemic, Hypotensive, Detoxification [12]. So, it's important to study whether there is any difference in action between the extract of *Allium cepa* made from the outer dry peel alone and the one commercially purchased. Highlighting their Anti-oxidant potential and α-glucosidase. So, this study is taken up to fulfill this gap in Homoeopathic phytochemical research.

## Materials and Methods

### Preparation of ethanolic extract of dried onion peel

This experimental in vitro study was conducted over a period of 2-3 weeks. The crude extract of *Allium cepa* dry peel and the commercially available *Allium cepa* Ø were designated as Sample A and Sample B, respectively.

For preparation of Sample A, fresh onions were thoroughly washed, and the outer dry peel was carefully separated and collected. A total of 15 g of the dried peel was mixed with 60 ml of 90% ethanol and kept undisturbed at 25 °C for 14 days to allow proper extraction. After the extraction period, the mixture was filtered first through muslin cloth and then through Whatman No. 1 filter paper to remove solid residues. The resulting filtrate was stored in a sterile dark glass bottle under refrigeration until further use.

### α-amylase inhibition assay

**Principle**

α-Amylase is an important digestive enzyme that breaks down starch into simple sugars. By inhibiting its activity, the rate of carbohydrate digestion and subsequent rise in blood glucose levels can be reduced, which is beneficial in the management of diabetes [13].

**Procedure**

Different concentrations of the test samples (250-1000 µL/mL) were prepared in 25 mM phosphate buffer (pH 6.9). Each reaction mixture contained 25 µL of porcine pancreatic α-amylase (0.5 mg/mL) and was pre-incubated at 25 °C for 10 minutes. Following this, 25 µL of 0.5% starch solution was added as a substrate, and the mixture was further incubated for 10 minutes. The reaction was stopped by adding 50 µL of 96 mM 3,5-dinitrosalicylic acid (DNSA) reagent. The tubes were placed in a boiling water bath for 5 minutes, cooled to room temperature, and the absorbance was recorded at 540 nm using a microplate reader (Erba, Lisascan). A control was maintained without test samples, and the percentage inhibition was calculated by comparing the absorbance of the test with that of the control [13].

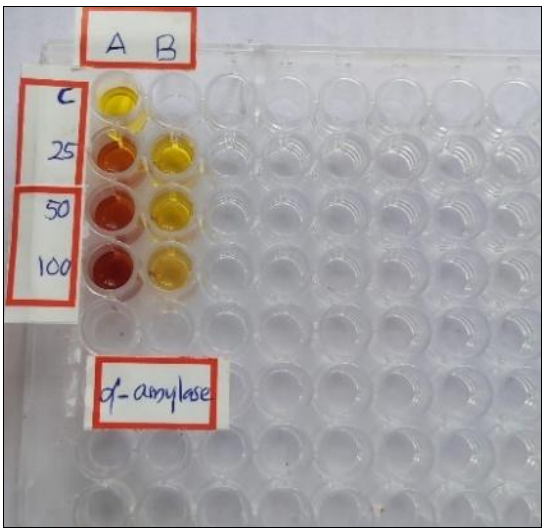



**Fig 1:** Alpha amylase inhibitory assay of Sample A and Sample B

### α-glucosidase inhibition assay

**Principle**

α-Glucosidase is another digestive enzyme that helps convert disaccharides into glucose. Inhibiting this enzyme slows glucose release into the bloodstream and helps reduce postprandial hyperglycemia. This makes α-glucosidase inhibition a key strategy in controlling type 2 diabetes [14].

**Procedure**

Varying concentrations of the test samples (250-1000 µL/mL) were prepared in 0.1 M phosphate buffer (pH 7.2) to a final volume of 1000 µL. Each reaction mixture contained 25 µL of α-glucosidase enzyme (Sigma-Aldrich, 1 U/mL) and was pre-incubated at 25 °C for 10 minutes. After this, 1 mL of 37 mM sucrose solution was added as a substrate, and the mixture was incubated at 37 °C for 30 minutes. The reaction was terminated by placing the tubes in a boiling water bath for 2 minutes. To each tube, 250 µL of glucose reagent was then added, incubated for 10 minutes, and the absorbance was measured at 510 nm using a microplate reader (Erba, Lisascan). A control was maintained with buffer and enzyme only, and the percentage inhibition was determined by comparing the absorbance values of test samples against the control [14].

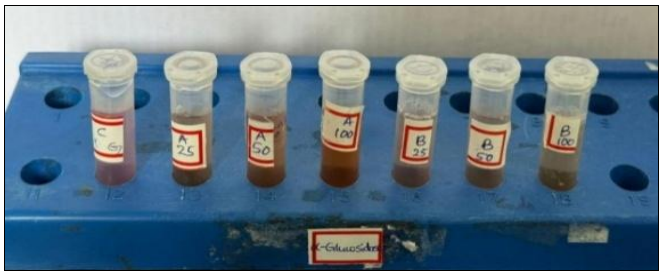


**Fig 2:** Alpha glucosidase inhibitory assay of Sample A and Sample B

**DPPH radical scavenging Assay**

**Principle**

The DPPH (1,1-diphenyl-2-picrylhydrazyl) assay is a widely used method to evaluate the free radical scavenging ability of plant extracts. DPPH is a stable free radical with a deep violet color that turns yellow when neutralized by an antioxidant. The degree of discoloration reflects the radical scavenging activity of the sample [15].

**Procedure**

Different concentrations of the test samples (12.5-200 µL/mL) were prepared in methanol. To each sample, 1.48 mL of 0.1 mM DPPH solution was added, and the mixture was incubated in the dark at room temperature for 20 minutes. A control was maintained with methanol and DPPH solution only. After incubation, the absorbance was measured at 517 nm using a UV-Visible spectrophotometer (Shimadzu UV-1900i). The percentage inhibition of DPPH radicals was calculated by comparing the absorbance of test samples with the control. Ascorbic acid was used as a standard antioxidant reference [15].

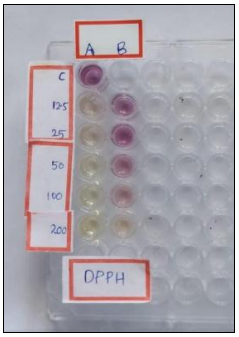



**Fig 3:** DPPH radical scavenging assay of Sample A and Sample B

**ABTS assay**

**Principle**

The ABTS assay measures the ability of antioxidants to quench the $ABTS^+$ radical cation, a blue-green chromogen formed by the reaction of ABTS (2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)) with potassium persulfate. Antioxidants reduce the $ABTS^+$ radical, leading to decolorization, which can be quantified spectrophotometrically [16].

**Procedure**

The $ABTS^+$ radical was generated by mixing 20 mM ABTS solution with 17 mM potassium persulfate solution and allowing it to stand in the dark at room temperature for 12-16 hours. For the assay, different concentrations of the test samples (125-2000 µL/mL) were prepared. To each sample, 0.16 mL of the prepared $ABTS^+$ solution was added and the volume was adjusted to 1.36 mL with distilled water. A control was maintained without the test sample. After 20 minutes of incubation at room temperature, absorbance was measured at 734 nm using a UV-Visible spectrophotometer (Shimadzu UV-1900i). The percentage inhibition of ABTS radicals was calculated by comparing the absorbance values of the test samples with the control. Ascorbic acid was used as a standard reference [16].

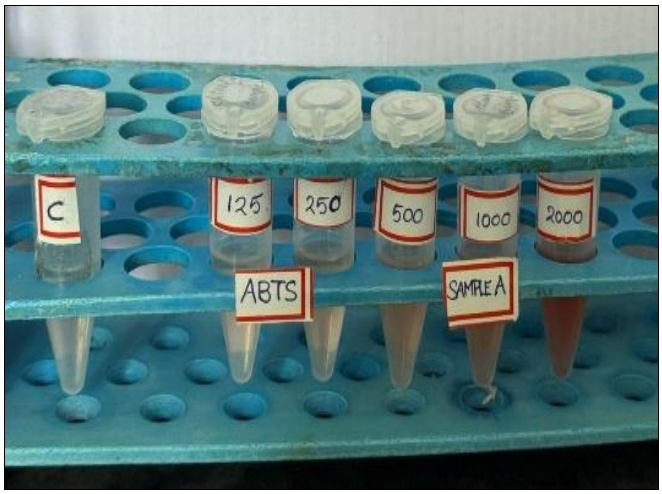


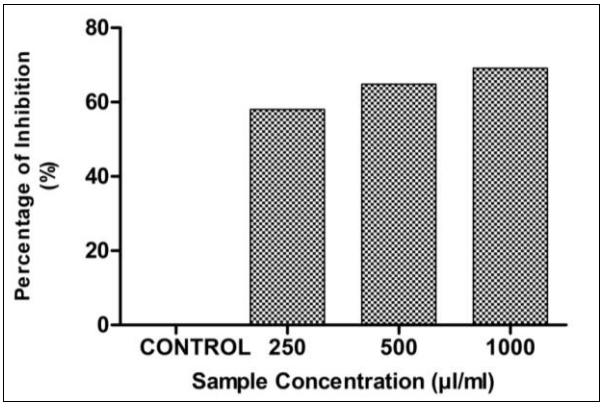

**Fig 4:** ABTS assay of Sample A and Sample B

**Results**

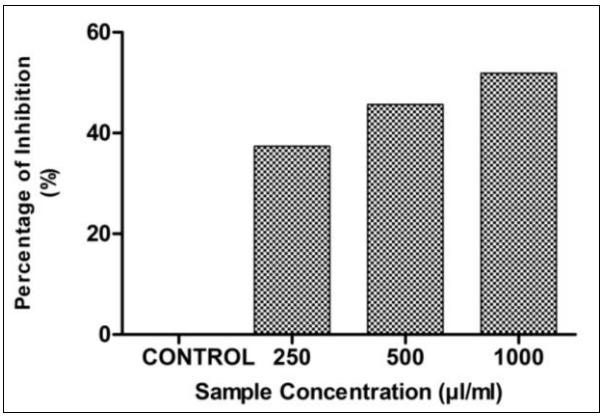
**α-amylase inhibition activity**

The ethanolic extract of dried onion peel (*Allium cepa* L., Sample A) demonstrated strong inhibitory activity against α-amylase, an enzyme responsible for starch digestion. A clear concentration-dependent increase in inhibition was observed for both the peel extract and the mother tincture (*Allium cepa* Ø, Sample B), though Sample A consistently showed higher activity.

At 250 µL/mL, Sample A inhibited 58.03% of enzyme activity, compared with 37.38% for Sample B. At 500 µL/mL, inhibition increased to 64.83% for Sample A and 45.67% for Sample B. At the highest concentration tested (1000 µL/mL), Sample A reached 69.09% inhibition, while Sample B achieved 51.89%. The $IC_{50}$ value of Sample A (215.40 µL/mL) was substantially lower than that of Sample B (847.60 µL/mL), confirming that the onion peel extract is a more effective inhibitor of α-amylase activity.




**Graph 1:** Graphical representation depicting the estimation of α-amylase in A. Along Y axis Percentage inhibition, Along X axis varied concentration of A




**Graph 2:** Graphical representation depicting the estimation of α-amylase in B. Along Y axis Percentage inhibition, Along X axis varied concentration of B

**Table 1:** Analysis of percentage inhibition at different concentrations of Sample A in α-amylase assay

| Sample concentration (µl/mL) | OD at 540nm | Percentage inhibition |
|---|---|---|
| **Sample code: A (*Allium cepa* peel extract)** | | |
| Control | 0.9748 | 0.000 |
| 250 | 0.4091 | 58.032 |
| 500 | 0.3428 | 64.834 |
| 1000 | 0.3013 | 69.091 |

**Table 2:** Analysis of percentage inhibition at different concentrations of Sample B in α-amylase assay

| Sample concentration (µl/mL) | OD at 540nm | Percentage inhibition |
|---|---|---|
| **Sample code: B (*Allium cepa* medicine)** | | |
| Control | 0.9748 | 0.000 |
| 250 | 0.6104 | 37.382 |
| 500 | 0.5296 | 45.671 |
| 1000 | 0.4689 | 51.898 |

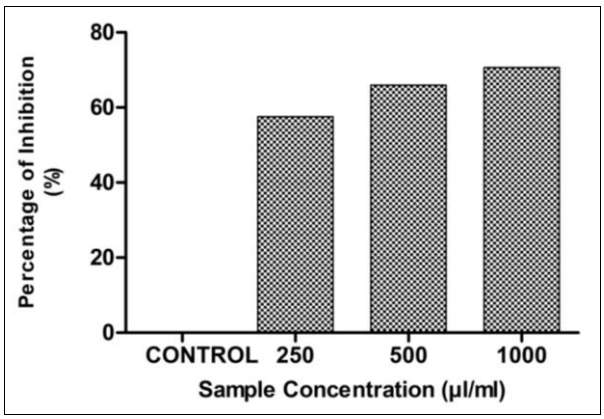
$IC_{50}$ Value-A: 215.398 µl/mL (Calculated using ED50 PLUS V 1.0 Software)

$IC_{50}$ Value-B: 847.599 µl/mL (Calculated using ED50 PLUS V 1.0 Software)

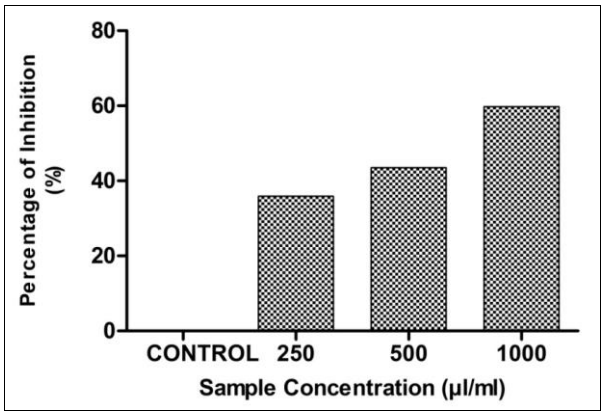
### α-glucosidase inhibition activity

The onion peel extract (Sample A) also showed significant inhibition of α-glucosidase, the enzyme that breaks down disaccharides into glucose. This activity directly supports its role in controlling postprandial blood sugar levels.

At 250 µL/mL, Sample A inhibited 57.58% of enzyme activity, compared to 35.90% for Sample B. At 500 µL/mL, inhibition rose to 65.98% for Sample A and 43.47% for Sample B. At 1000 µL/mL, Sample A achieved 70.65% inhibition, while Sample B reached 59.79%. The $IC_{50}$ value of Sample A (217.09 µL/mL) was considerably lower than that of Sample B (700.00 µL/mL), further demonstrating the superior potency of the onion peel extract.




**Graph 3:** Graphical representation depicting the estimation of α-glucosidase inhibitory assay. Along Y axis Percentage inhibition, Along X axis varied concentration of A




**Graph 4:** Graphical representation depicting the in vitro α-glucosidase inhibitory assay. Along Y axis, Percentage of inhibition (%) and along X axis, concentration of B

**Table 4:** Analysis of percentage inhibition at different concentrations of Sample B in alpha glucosidase assay

| Concentration (µl/ml) | Absorbance | Percentage of inhibition |
|---|---|---|
| Control | 0.8456 | 0.000 |
| **Sample code: A (*Allium cepa* peel extract)** | | |
| 250 | 0.3587 | 57.580 |
| 500 | 0.2877 | 65.977 |
| 1000 | 0.2482 | 70.648 |

**Table 3:** Analysis of percentage inhibition at different concentrations of Sample A in alpha glucosidase assay

| Concentration (µl/ml) | Absorbance | Percentage of inhibition |
|---|---|---|
| Control | 0.8456 | 0.000 |
| **Sample code: B (*Allium cepa* medicine)** | | |
| 250 | 0.542 | 35.904 |
| 500 | 0.478 | 43.472 |
| 1000 | 0.34 | 59.792 |

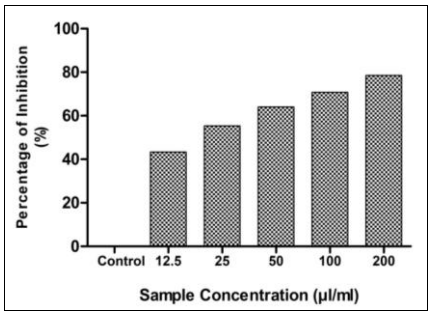
$IC_{50}$ Value -A-217.089µl/mL (Calculated using ED50 PLUS V1.0 Software)

$IC_{50}$ Value -B-700µl/mL (Calculated using ED50 PLUS V1.0 Software)

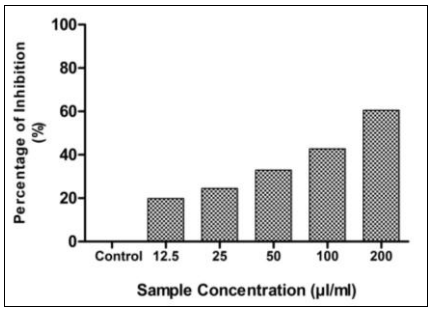
**DPPH radical scavenging activity**

The antioxidant activity of the samples was evaluated using the DPPH radical scavenging assay. Both samples exhibited concentration-dependent radical scavenging activity, with Sample A showing much stronger inhibition across all concentrations.

At 12.5 µL/mL, Sample A recorded 38.61% inhibition, whereas Sample B showed only 25.34%. At 50 µL/mL, Sample A reached 57.49% inhibition compared to 42.58% for Sample B. At the highest concentration (200 µL/mL), Sample A achieved 66.30% inhibition, slightly higher than Sample B (65.51%). The $IC_{50}$ value for Sample A was 19.40 µL/mL, compared to 140.70 µL/mL for Sample B, confirming the stronger antioxidant capacity of the onion peel extract.




**Graph 5:** Graphical representation of DPPH Radical scavenging assay in A




**Graph 6:** Graphical representation of DPPH radical scavenging assay in B

**Table 5:** Analysis of percentage inhibition at different concentrations of Sample A in DPPH radical scavenging assay

| Concentrations (µl/mL) | Absorbance | Percentage of inhibition |
|---|---|---|
| Control | 0.415 | 0.000 |
| 12.5 | 0.235 | 43.346 |
| 25 | 0.185 | 55.400 |
| 50 | 0.149 | 64.079 |
| 100 | 0.121 | 70.829 |
| 200 | 0.089 | 78.544 |

**Table 6:** Analysis of percentage inhibition at different concentrations of Sample B in DPPH radical scavenging assay

| Concentrations (µl/mL) | Absorbance | Percentage of inhibition |
|---|---|---|
| Control | 0.415 | 0.000 |
| 12.5 | 0.332 | 19.913 |
| 25 | 0.313 | 24.566 |
| 50 | 0.278 | 32.980 |
| 100 | 0.237 | 42.768 |
| 200 | 0.164 | 60.535 |

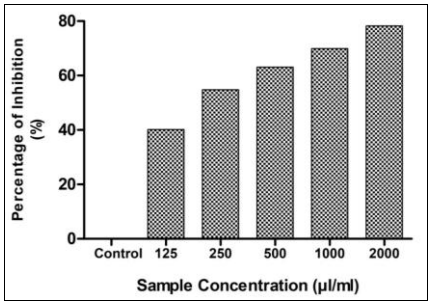
$IC_{50}$ Value-A: 19.4 µl/mL (Calculated using ED 50 PLUS V1.0 Software)

$IC_{50}$ Value-B: 140.7 µl/mL (Calculated using ED 50 PLUS V1.0 Software)

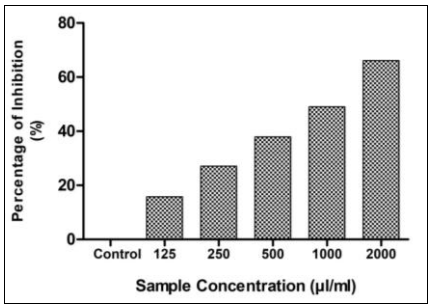
**ABTS radical scavenging activity**

The ABTS assay further validated the antioxidant potential of onion peel extract. Sample A consistently demonstrated greater activity compared to Sample B.

At 125 µL/mL, inhibition was 33.33% for Sample A and 20.29% for Sample B. At 500 µL/mL, Sample A showed 53.67% inhibition, whereas Sample B reached 37.68%. At the highest concentration tested (2000 µL/mL), Sample A achieved 68.72% inhibition, while Sample B recorded only 49.86%. The $IC_{50}$ value of Sample A was 209.10 µL/mL, significantly lower than that of Sample B (1058.57 µL/mL), indicating that onion peel extract is almost five times more potent in scavenging ABTS radicals.




**Graph 7:** Graphical representation depicting ABTS assay. Along X axis concentration of A, along Y axis percentage of inhibition




**Graph 8:** Graphical representation depicting ABTS Assay. Along X axis concentration of B, Along Y axis percentage of inhibition

**Table 7:** Analysis of percentage inhibition at different concentrations of Sample A in ABTS assay

| Concentration(µl/mL) | Absorbance | Percentage of inhibition |
|---|---|---|
| Control | 0.41 | 0.000 |
| **Sample: A (*Allium cepa* peel extract)** | | |
| 125 | 0.2453 | 40.171 |
| 250 | 0.1854 | 54.780 |
| 500 | 0.1513 | 63.098 |
| 1000 | 0.1234 | 69.902 |
| 2000 | 0.0893 | 78.220 |

**Table 8:** Analysis of percentage inhibition at different concentrations of Sample B in ABTS assay

| Concentration(µl/mL) | Absorbance | Percentage of inhibition |
|---|---|---|
| Control | 0.41 | 0.000 |
| **Sample: B (*Allium cepa* medicine)** | | |
| 125 | 0.3454 | 15.756 |
| 250 | 0.2989 | 27.098 |
| 500 | 0.2547 | 37.878 |
| 1000 | 0.2091 | 49.000 |
| 2000 | 0.1391 | 66.073 |

$IC_{50}$ Value- Sample A: 209.101 µl/mL (Calculated using ED50 PLUS V1.0 Software)

$IC_{50}$ Value- Sample B: 1058.57 µl/mL (Calculated using ED50 PLUS V1.0 Software)

## Discussion

This study compared the antioxidant capacity and α-glucosidase/α-amylase inhibitory effects of *Allium cepa* dry peel extract (Sample A) with those of a commercially prepared *Allium cepa* Ø (Sample B). In all assays performed, the *Allium cepa* dry peel extract consistently showed stronger activity than the *Allium cepa* Ø, indicating higher levels of bioactive compounds in the peel.

### Antioxidant activity

In both the DPPH and ABTS assays, Sample A displayed greater free radical scavenging activity and lower $IC_{50}$ values than Sample B. These results point to the *Allium cepa* peel's rich content of flavonoids especially quercetin and other phenolics that can effectively neutralize Reactive Oxygen Species (ROS). *Allium cepa* peels are known to contain much higher concentrations of quercetin than the edible bulb, which likely explains the observed potency. By reducing oxidative stress, such antioxidants can help protect against the development of diabetes, cardiovascular problems, and certain cancers.

### Antidiabetic activity

The α-glucosidase and α-amylase inhibition tests revealed that Sample A also outperformed Sample B in slowing carbohydrate breakdown. Lower $IC_{50}$ values for the *Allium cepa* dry peel extract suggest a stronger ability to limit starch and sugar digestion, which can help moderate post-meal spikes in blood glucose. This effect is likely linked to the phenolics, saponins, and organosulfur compounds in *Allium cepa*, which can bind to and block the active sites of these digestive enzymes.

### Sustainability and practical value

An important outcome of this research is the demonstration that *Allium cepa* peel normally thrown away as waste has greater antioxidant and antidiabetic properties than the commonly used bulb-based tincture. This positions *Allium cepa* peel as a sustainable, inexpensive source of health-promoting compounds that could be developed into herbal products, nutraceuticals, or functional food ingredients. Making use of such plant waste not only adds economic value but also supports environmentally responsible healthcare approaches.

## Conclusion

The study found that *Allium cepa* (onion) peel extract was much more effective than the tincture in controlling enzymes linked to diabetes. It showed lower $IC_{50}$ values for α-amylase (215.40 vs. 847.60 µl/mL) and α-glucosidase (217.09 vs. 700.00 µl/mL). The peel extract also had stronger antioxidant effects, with $IC_{50}$ values of 19.40 µl/mL (DPPH) and 209.10 µl/mL (ABTS), compared to 140.70 µl/mL and 1058.57 µl/mL for the tincture. These results suggest that onion peels usually discarded as waste are actually a valuable, low cost source of natural compounds that could support diabetes management and antioxidant therapy. Further research is needed to confirm their safety and effectiveness in real world use.

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