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Research Article

Physico-Chemical Properties, Phytochemical Screening and Antioxidant Potential of Polar and Non-Polar Seed Extractions of Selected Medicinal Plants of *Solanaceae*

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Abstract: The objective of this study was to investigate the various bioactive components and assess the biological activity in the seeds of five medicinal plants (*Datura innoxia* Miller., *Solanum elaeagnifolium* Cav., *Solanum nigrum* L., *Solanum surattense* Burm., and *Withania coagulans* (Stocks) Dunal.) in *Solanaceae* family. In this regard, ash values, acid-insoluble ash and water-soluble ash values, extractive values and screening of various phytochemicals (alkaloid, carbohydrate, protein, flavonoid, tannins, saponins, and fixed oil) and quantitative estimation of carbohydrate and protein of seeds of selected plants have been calculated. The 2,2-diphenyl-1-picryl hydrazyl (DPPH) free radical scavenging assay was used to evaluate the antioxidant potential. Captivatingly, a phytochemical screening assay affirmed the presence of saponins, terpenoids, phenolic compounds, and flavonoids in the seeds of all examined plants. The highest amounts of DPPH radical-scavenging activity were found in the aqueous extract of *W. coagulans* (IC_{50} = 65.6 µg/ml). Consequently, current findings showed all selected plants of the family *Solanaceae* contain highly bioactive components, which could be used for the treatment of different diseases and as a potential resource for discovering new drugs.

Keywords: Medicinal Plants, Bioactive Components, *Solanaceae* Family, Phytochemical Analysis, Quantitative Estimation, Antioxidant Activity.

1. INTRODUCTION

Medicinal plants have been employed in healthcare systems from the beginning. Studies have been done all around the world to verify their efficacy, and some of the findings have spurred interest in the production of plant-based medications [1]. As per the World Health Organization, 80 percent of the world's population takes plant-based treatment as their primary form of health care and about 11% of the 252 drugs are exclusively derived from plants. In modern society regardless of the advancement of synthetic drugs plants are the fundamental source of new healthcare and pharmaceutical products [2]. The majority of research supports the effectiveness of medicinal herbs, which has sparked interest from all over the world. Medicinal plants offer therapeutic benefits and have fewer negative side effects than synthetic drugs [3].

Depending on their biological roles, the phytochemicals that plants generate might be classified as primary or secondary metabolites. Their primary elements include common sugars, proteins, amino acids, purines, and pyrimidines from nucleic acids, as well as other materials like chlorophyll. Secondary components encompass the remaining plant compounds, which include alkaloids, phenolics, glucosides, saponins, terpenes, lignans, flavonoids, and plant steroids [4]. Although secondary metabolites are produced in all part of the plant, including the bark, stems, leaves, roots, fruits, flowers, seeds, and so on, plants may not require them. These substances are thought to be therapeutic mediators and effective in treating a wide range of illnesses in both people and animals. exhibit a variety of significant pharmacological properties, including antibacterial, antiviral, antirheumatic, cholesterol synthesis inhibition, anticancer, and antiparasitic effects [5].

Plant species have strong antioxidant activity, which could be used to find effective treatments for tissue damage caused by free radicals. Various

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synthetic antioxidants, such as tertiary butyl hydroquinone (TBHQ), butylated hydroxyanisole (BHA), and butylated hydroxytoluene (BHT), have been incorporated into food products. However, these artificial antioxidants have been linked to liver problems [6]. With 84 genera and 3000 species, the *Solanaceae* family is extensively distributed in tropical and temperate regions of both hemispheres, but primarily in Western and Southern America. It is represented in Pakistan by 14 genera and 52 species. 27 species are native, 6 naturalized and others are cultivated and found rarely as escapes [7].

Various plants of the *Solanaceae* family have been traditionally used as medicine all over the world. It is because of the presence of various secondary metabolites. These are the most potent known anticholinergics, which means they suppress the neurological impulses provided by the endogenous neurotransmitter acetylcholine [8]. It has been reported that *Solanaceae* plants, including *Capsicum*, *Datura*, and *Solanum*, contain anti microbial peptides (AMP) and peptide-rich extracts from their seeds, leaves, fruits, and tubers. Notable antifungal, antimicrobial, or antiviral effects against human pathogenic microorganisms and phytopathogenic strains have been reported for these peptides. *Datura stramonium* L. seed extracts have been reported to have antibacterial activity against *Escherichia coli* and *Klebsiella pneumonia* [9]. It has been demonstrated that an extract from the leaves of *Solanum lycopersicum* shows potential antitumor properties against breast cancer cells by modifying the expression of genes linked to the development and spread of cancer. *Withania somnifera* leaf extract activates tumour suppressor proteins and has cytotoxic effects on human osteosarcoma, fibrosarcoma, and lung cancer epithelial cells [10].

The focus of this study was to investigate the seeds of five different plants from the *Solanaceae* family that were taken for physicochemically, phytochemically, and free radical scavenging ability analysis, i.e., *Datura innoxia* Miller., *Solanum elaeagnifolium* Cav., *Solanum nigrum* L., *Solanum surattense* Burm., and *Withania coagulans* (Stocks) Dunal. This research will aid in the verbalization of the phytochemical standard. As a result, basic knowledge regarding phytochemicals will be important in determining therapeutic effectiveness.

2. MATERIALS AND METHOD

2.1. Collection and Sample Preparation

An appropriate amount of seeds (150 g) of selected medicinal plants were collected from different areas of Karachi, Pakistan, sourced from their natural habitats (wild). With the seeds collection, plant specimens were also collected for identification purposes. Identified plants (Herbarium sheets) were deposited in S.I. Ali Herbarium, Center for Plant Conservation, University of Karachi. The seeds were dried at room temperature. Clean and well-dried seeds (100 g) of each plant species were blended into a fine powder using an electric blender.

2.2. Preparation of Crude Extracts

20 g powdered sample was soaked in 200 ml of different solvents, *viz.,* acetone (99.5%), chloroform (99.8%), methanol (99.8%) (BDH Laboratory Supplies), and water, separately, and placed for 48 hours for continuous shaking in an orbital shaker. After 48 hours of continuous shaking, the extracts were filtered with the help of Whatmann No. 1 paper and left for evaporation until all the solvent had evaporated. The dried material was collected, and the extractive value was calculated by weighing the sample with the help of an electrical weighing machine (Sartorious TE214S).

2.3. Physicochemical Analysis

Physicochemical analysis was carried out such as moisture content as per the standard method. At 105 ºC, 2 g crude powder of plant seeds was dried, weight was noted, drying loss was computed, and percentage was determined based on the initial sample [11]. Total ash content was calculated, 2 g of plant seed powder were ignited in a crucible at 500 °C until it turned white, signifying the absence of carbon. The ash was then rapidly cooled and weighed. The percentage of ash was calculated based on the initial sample weight. Additionally, the physical state, color, and ash values (water-soluble ash and acid-insoluble ash) were determined according to standard methods [12, 13].

2.4. Qualitative Phytochemical Analysis

For the phytochemical assessment, standard methods were employed. Alkaloids were identified

using Mayer's test, Wagner's test, and Hager's test [14, 15]. Carbohydrates were detected through Benedict's test and Fehling's reagent [16]. Proteins and amino acids were assessed using the Biuret test and Millon's test [17]. Phenolic compounds were identified by the Lead acetate test and Ferric chloride test [18]. Fixed oils were evaluated using the spot test, and saponins were detected through the Foam test. Glycosides were assessed using Borentrager's test [14].

2.5. Quantitative Estimation of Carbohydrate and Protein

The total carbohydrate of seeds was estimated using Anthrone reagent [19]. 4 ml of the anthrone reagent was mixed with 1 ml of the sample. After 10 minutes of incubation in a boiling water bath, the absorbance at 620 nm was measured with the spectrophotometer in comparison to a reagent blank. The results were reported as mg/g sample, and the estimation was carried out in triplicate.
The Bradford assay was used to determine the total protein content by mixing 3 ml of Bradford reagent with 0.1 ml of the sample. Following an incubation period of 10 minutes at room temperature, the absorbance at 595 nm was measured using spectrophotometer [20].

2.6. Determination of Antioxidant Activity

The approach utilized to test the antioxidant activity was based on an assessment of the extract's free radical scavenging activity using the method outlined by Brand-Williams *et al.* [21] with some modifications. The DPPH scavenging activity of different extracts of acetone, chloroform, methanol, and water were determined. As a blank and standard solvent, methanol and ascorbic acid were utilized, respectively. From each extract, stock solutions (2.5 mg/ml) were produced in methanol. These stock solutions were used to make working solutions (50, 100, 150, 200, and 250 µg/ml). 1 ml of DPPH solution (0.96 mM) was added to each working solution before incubating for 30 minutes at room temperature. 1 ml DPPH was added to 4 ml methanol to make the control. The absorbance of the samples and the control was measured using a UV/VIS spectrophotometer at the 517 nm range. The DPPH scavenging activities were calculated as a percentage of inhibition using the equation:

$$\%\, inhibition = \frac{A_0 - A_1}{A_0} \times 100$$

Where A_0 is the absorbance of control and A_1 is the absorbance of the sample. The IC_{50} is a measure of a compound's effectiveness in suppressing a particular function. The IC_{50} of the DPPH radical scavenging activity of seed extracts from selected plants was determined through linear regression analysis of concentrations and percent inhibition.

2.7. Statistical Analysis

All analyses were carried out in triplicate, with the results statistically analyzed and expressed as the mean (n = 3) and standard deviation (SD).

3. RESULTS AND DISCUSSION

3.1. Physicochemical Analysis

Physicochemical analysis of seed powder of selected medicinal plants is shown in Table 1. The loss on drying of powdered seeds was observed at 12% in *D. innoxia*, 2.5% in *W. coagulans,* 7% in *S. elaeagnifolium,* 3.9% in *S. nigrum, and* 14% *in S. surattense.* The total amount of ash was highest for *W. coagulans* seeds, i.e., 12.5%; whereas the lowest was observed in *S. elaeagnifolium,* i.e., 5%. The highest water-soluble ash was found 5.21% in *S. elaeagnifolium* and the lowest 3.91% in *S. surattense.* The acid-insoluble ash was highest

Table 1. Physicochemical properties of seeds of selected medicinal plants.

Parameter	*D. innoxia*	*W. coagulans*	*S. elaeagnifolium*	*S. nigrum*	*S. surattense*
The physical state of ash	Granulated	Granulated	Granulated	Fine powder	Fine Powder
Color of ash	Brown	Black	Grayish white	Light brown	Creamy white
% loss on drying	12	2.5	7	3.9	14
Ash content (%)	6	12.5	5	7.88	6.5
Water soluble ash	4.2%	4.37%	5.21%	4.73%	3.91%
Acid insoluble ash	2.5%	3.21	4.1	3.9	0.5415

for *S. elaeagnifolium,* i.e., 4.1% and the lowest for *S. surattense,* i.e., 0.54%. Extractive values of selected medicinal plants seeds of *Solanaceae* in different solvents are shown in Figure 1. The highest extractive value was observed in chloroform extract of *W. coagulans, S. elaeagnifolium, and S. surattense.* The highest extractive value of seeds of *S. nigrum* was found in methanol and lowest in acetone. The solubility of *D. innoxia* seed extract was highest in water and lowest in chloroform.

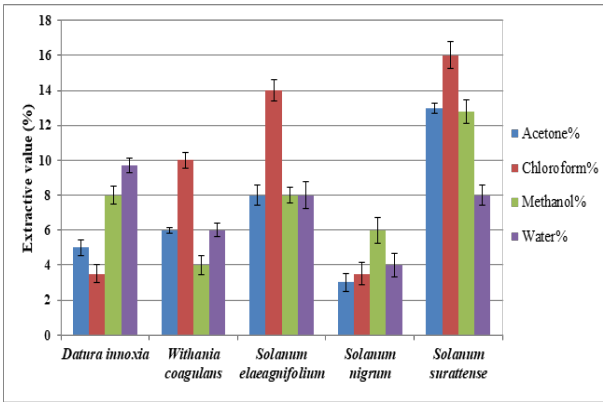

Fig. 1. Extractive values of seeds of selected medicinal plants in four different solvents.

3.2. Phytochemical Analysis

Historically members of the family *Solanaceae* were highly valued for their alkaloid content, which was utilized for poisoning and psychotropic effects; however, a study of the bioactive chemicals of this family is now becoming more important for medicinal applications [22]. The leaves of *Solanum torvum* have been reported to possess antimicrobial activity [23]. An ethanolic extract of *Datura stramonium* leaf showed a considerable anti-inflammatory effect in rats with carrageenan-induced paw edema [24]. Extracts of *S. nigrum* are analgesic and anti-inflammatory [25]. Ramadan *et al.* [26] reported that the seeds of *Datura innoxia* contain fatty acids and fat-soluble compounds. Pharmacological screening of the plant extracts

Table 2. Qualitative analysis of seeds of selected medicinal plants of family *Solanaceae* in four solvent extracts.

	D. innoxia				*W. coagulans*				*S. elaeagnifolium*				*S. nigrum*				*S. surattense*			
Solvent Extract	**Ac**	**Chl**	**Met**	**W**	**Ac**	**Ch**	**Met**	**W**	**Ac**	**Chl**	**Met**	**W**	**Ac**	**Chl**	**Met**	**W**	**Ac**	**Chl**	**Met**	**W**
Alkaloid																				
Wagner's reagent	+++	+++	+++	++	-	-	+	+	+++	+	+++	+++	-	-	+	+	-	-	++	++
Mayer's reagent	+++	+++	+++	++	+	+	+	+	+++	++	+++	+++	-	+	+	+	+	++	+++	+++
Carbohydrate																				
Benedict's test	++	++	+++	++	-	+	-	-	+++	+++	+++	+	+	-	+	+	++	+++	++	+
Fehling test	++	++	+++	++	+	-	+	+	++	++	++++	+++	-	+	+	+	++	+++	++	-
Protein																				
Biuret test	+	+	+	+	-	-	+	+	+	+	+	+	-	-	-	-	-	++	+++	+++
Millions test	+	-	++	++	++	++	+	+	++	++	++	++	++	++	++	++	++	++	++	++
Phenolic Compounds																				
Lead acetate	++	++	++	+++	+	+	+	+	++	-	++	+++	++	++	++	++	++	++	+++	+++
Ferric chloride test	++	++	++	++	+	+	+	+	++	-	+	++	++	++	++	++	+	++	+++	+++
Flavonoids	++	++	++	++	-	-	-	-	+++	+	++	++	+	+	+	+	-	+	+++	+++
Glycoside																				
Salkowsk's test	+	-	+++	+++	-	-	-	-	+	+	+	-	-	-	-	-	-	-	-	-
Terpenoids	++	+++	++	++	+	+	+	+	+++	+	++	+	+	++	++	++	++	+++	+++	+++
Fixed Oil																				
Spot test	+	++	++	+	-	-	-	-	++	++	++	-	-	-	-	-	+	++	++	++
Saponin																				
Foam test	+++	+	++	+++	-	-	-	+	-	-	-	-	-	-	+	+	-	-	+	+++

Key: Ac = Acetone, Chl = Chloroform, Met = Methanol, W = Water, + = present, - = absent, ++ = present in moderate amount, +++ = present in high amount.

provided insights into both their beneficial and toxic properties. Phytochemical investigation of selected medicinal plant seed extracts in various solvents revealed the presence of alkaloids, carbohydrates, proteins, phenolic compounds, flavonoids, terpenoids, fixed oil, and saponins (Table 2). These Phytoconstituents are considered remedies for a variety of ailments in both humans and animals. Alkaloids are present in all 5 selected plants, and have been attributed to medical use. Alkaloids are associated with cytotoxicity, analgesic, and antibacterial properties [27]. The present results show that most plants contain carbohydrates, except for *Withania coagulans*, which exhibited weak indications of carbohydrates in all solvent extracts of its seeds. Carbohydrates provide energy while also aiding digestion and nutritional absorption. Furthermore, some of the carbs in these plants have therapeutic properties. All five selected medicinal plants showed positive results for flavonoids. Flavonoids have strong anticancer activity and also help in managing diabetes and oxidative stress [28]. Flavonoids offer protection from hepatotoxins, tumours, viruses, and Alzheimer's disease. They possess antioxidant, antifungal, anti-carcinogenic, hepatoprotective, and cytotoxic properties [29]. The preliminary screening tests for phytochemicals can be helpful in exploring their bioactive values, which could lead to the discovery and development of new drugs [30].

3.3. Quantitative Estimation of Carbohydrate and Protein

The total carbohydrate content of various medicinal plants was assessed, and it was revealed that the maximum carbohydrate content was found in the aqueous extract of *Solanum elaeagnifolium,* i.e., 8.32 ± 0.11 mg/ml, and minimum carbohydrate content was obtained in the acetone extract of *Withania coagulans,* i.e., 0.68 ± 0.01 mg/ml (Figure 2). It was found that the protein was present in all of the plants studied in this investigation. The highest protein concentration was observed in the methanolic extract of seed of *Solanum surattense* 1447.68 ± 8.3 µg/µl and the lowest was present in the chloroform extract of *Datura innoxia* 57.43 ± 5.8 µg/µl (Figure 3).

3.4. Antioxidant Activity

The antioxidant capacity of extracts obtained from different solvents was investigated in this work utilizing DPPH scavenging activity tests. As shown in Figure 4 different extracts possessed varying free-radical scavenging activities. The IC_{50} value of antioxidants with strong scavenging ability should be low. Among the extracts analyzed antioxidant activity with different solvents varied IC_{50} value of 65.6 to 123.7 µg/ml after 30 min. The highest antioxidant value was observed by *W. coagulans* in the range of 65.62 µg/ml - 123.78

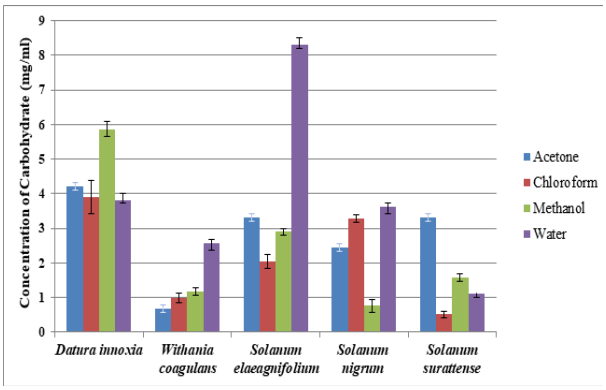

Fig. 2. Quantitative estimation of Carbohydrate in seed extract of selected plants

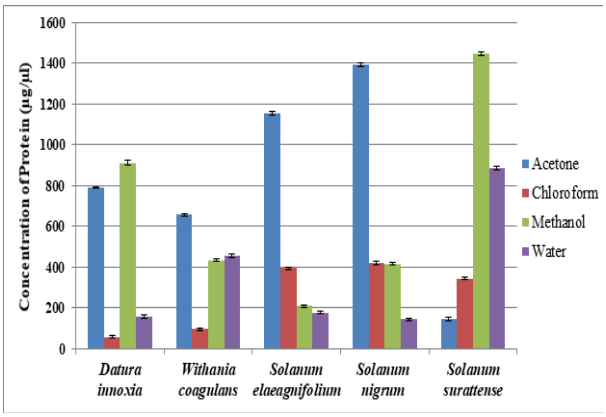

Fig. 3. Quantitative estimation of Protein in seed extract of selected plants.

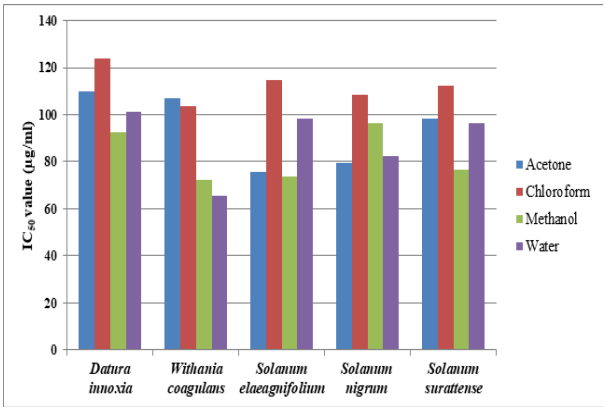

Fig. 4. IC_{50} values of DPPH free radical scavenging activity of seed extracts of selected *Solanaceae* species.

µg/ml, followed by *S. elaeagnifolium* in the range of 73.87 µg/ml -114.65 µg/ml, followed by *S. surattense* in the range 76.62 µg/ml - 112.5 µg/ml followed by *S. nigrum* in the range 79.5 µg/ml - 108.3 µg/ml, followed by *D. inoxia* 92.34 µg/ml - 123.78 µg/ml. Previous research has shown that changing the polarity of the solvent can be used to selectively concentrate antioxidant chemicals [31]. The presence of alkaloids and phenols in plant extract is often linked to its high activity, as these groups of phytochemicals include the bulk of active antioxidant substances. Secondary metabolites such as phenol and alkaloids play an important role in increasing antioxidant capacity [32]. Phenolic components are beneficial electron donors because their hydroxyl groups can contribute to the antioxidant process. Total phenol and antioxidant capacity have a strong positive correlation [33]. These findings could be utilized as markers for identifying and standardizing of the drug as an herbal treatment, as well as in the development of a monograph for the plant. Significant differences in free radical scavenging capabilities were observed in the current investigation for seed extracts of selected plants depending on the kind of extract utilized. The greatest scavenging capacity was demonstrate by aqueous and methanol extracts, followed by acetone and chloroform extracts.

4. CONCLUSIONS

The seeds of selected medicinal plants, due to their various bioactive substances, could potentially treat diseases such as rheumatism, diuresis, viral infections, cancer, malaria, fungal infections, and bacterial infections. Given their effects on human health, switching from synthetic to natural antioxidants could be advantageous. According to our findings, seeds of selected medicinal plants have the potential to be a source of valuable pharmaceuticals. Phytochemical screening of medicinal plants is important for both research institutes and pharmaceutical companies in the industrialized of the new drug.

5. CONFLICT OF INTEREST

The authors declare no conflict of interest.

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