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Preliminary Phytochemical Screening and Estimation of Total Phenolic Content of Aqueous Cladode Extract of *Opuntia dillenii*

Thana Lakshme PS, R Gayathri*, V Vishnu Priya

Department of Biochemistry, Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University, Chennai, India

ABSTRACT

Opuntia dillenii commonly known as prickly pear belonging to the family Cactaceae. Opuntia dillenii is ideally suited to water-scarce dry zones of the world as an alternate source of food and fodder. Generally phytochemicals in plants include alkaloids, flavonoids and etc. It also includes phenol content which is a good antioxidant. The aim of the study is to analyse the phytochemicals and estimate total phenol content of Opuntia dillenii. Opuntia dillenii was purchased from a nursery in Chennai. Aqueous cladode extract of Opuntia dillenii was prepared. Phlobatannins, carbohydrates, flavonoids, alkaloids and terpenoids were qualitatively analysed. Total phenol was quantitatively estimated with Gallic acid as standard. From the Phytochemical analysis it was evident that the leaf extract shows significant presence of phytochemicals such as phlobatannins, flavonoids and terpenoids. Carbohydrates are in trace amounts. Total phenolic content was found to be 12.6 mg GAE/gm. Thus it was evident that Opuntia dillenii was significantly rich in phenolic compounds and this a good source of antioxidant. This study indicates the valuable presence of micro nutrients like phytochemicals and phenols. Phenols are the most important group of natural compounds considered to be of high scientific and therapeutic interest as it controls the accumulation of free radicals. From this study it was evident that the plant Opuntia dillenii is rich in phytoconstituents and further studies are required to isolate the chemical constituents and explore its biological value.

Key words: Water scarce dry zones; phytochemicals; phenols; antioxidants; free radicals; oxidative stress

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Corresponding author: R Gayathri

e-mail✉: gayathri.sdc@saveetha.com

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INTRODUCTION

Term phytochemicals implies that these are chemicals of plant source produced through primary and secondary metabolism [1]. Phenols and acids are part of phytochemicals. Their foremost vital role is plant growth and has a defense against competitors, pathogens and predators [2]. Phytochemicals can be classified into carotenoids and polyphenols which include flavonoids, phlobatannins, terpenoids, etc. [3]. Compounds which are recognised as essential nutrients are comprised as phytochemicals and essential for normal physiological functions [4]. Certain phytochemicals are phytotoxins which are toxic to humans and some include aristolochic acid which is carcinogenic at low doses [5]. Some phytochemicals are polyphenols, flavonoids which are prooxidants and act as antioxidants that shield us from free radicals [6]. The presence of these phytochemicals and their estimation is done with the extract of any part of the plant or the whole plant. For this, plants are chosen based on their origin mostly. For example, plants that exist in desert and arid regions have complex metabolic activities for their survival which leads to effective secondary metabolites. Thus in our study we are going to deal with *Opuntia dillenii* which also own its own traditional purposes.

Opuntia dillenii is a large sized species of cactus which is endemic to subtropical and tropical coastal areas. It belongs to sub family Opuntioideae which fit into the family Cactaceae and order Caryophyllales. The common names include prickly pear, Ethem Indian fig. This plant was included in the sub kingdom Tracheophytes, Super-division Embryophyta, division Eudicots. *Opuntia dillenii* (Ker Grawl) is the variety of species stricta and genus Opuntia Mill [7]. It has

its origin in South eastern USA. Nowadays plants grown in desert and semi desert areas are of more focus due to their diverse pharmacological activities such as anti- oxidant, anti-inflammatory activities, etc. The fruits have a rich amount of valuable constituents like ascorbic acid, total phenol and every element that makes the plant a significant complementary therapy against several pathological conditions, disorders and cancer also [8].

Previous studies involving phytochemical analysis carried out in Opuntia elatior mill resulted in the high presence of colour pigment betacyanin and sugar content burr low acidity of fruit. This pigment betacyanin possesses antioxidant potential and the phenolic content of fruit supports the nutritional needs [9]. But the extent and amount of phenolic content was not estimated . In cactus pear, phytochemical activity and phenol content was estimated for the whole plant and it has been concluded that pulp is rich in phenol and peels have antioxidant property but there was no discussion about leaf extract [10]. Total phenolic content and presence of phytochemicals have significant relationship with anti-oxidant and anti-cancer properties [11]. In Opuntia ficus, phytochemicals, nutritionals and anti-oxidant properties were examined. It indicated the nutritional and pharmaceutical potential in all parts of the plant that had linear correlation with antioxidant and total phenolics [12].

In recent years, there has been a global trend towards the use of natural sources as antioxidants, involved in drug therapy and also as functional foods [13]. In such a prevailing situation, the potent knowledge about the phytochemicals and phenolic content would pave the way for establishing *Opuntia dillenii* as a better source for therapeutics. Usage of aqueous leaf extract of the plant for the study distinguishes the present study from the prior studies. The aim of the current study is to evaluate and phytochemical screening and estimate the total phenolic content of aqueous leaf extract of *Opuntia dillenii*.

MATERIALS AND METHODS

Collection of plant material

The plant *Opuntia dillenii* herb was purchased from a nursery in Ambattur. The leaf of the plant was used to prepare the aqueous extract preparation.

Preparation of plant extract

Cladode of the plant was carefully removed. Washed and cut into pieces. 50 grams of the cladode was weighed and crushed with 100ml of sterile water. Then the mixture was boiled at 50-60˚C for 20 minutes. Then the resulting solution was filtered through a whatman filter paper. The resulting filtrate was centrifuged for 15 minutes for 3000rpm. The resultant supernatant was collected, dried and stored in an airtight container at 4˚C for further use.

Phytochemical analysis

The phytochemical analysis was done qualitatively which involved the analysis of Phlobatannins, carbohydrates, flavonoids, alkaloids and terpenoids using specific reagents.

Test for phlobatannins: 1 ml of sample and 2 ml of 5% concentrated hydrochloric acid was added and boiled for 5 minutes. Red colour deposition is observed indicating phlobatannins.

Test for carbohydrates

Fehling's test: 1 ml of sample and 2 ml of fehling's solution A and B was added and boiled for 3 minutes in a boiling water bath. Reddish brown precipitate was observed.

Benedict's solution: 1 ml of sample and 2 ml of Benedict's solution was added and boiled for 3 minutes in a boiling water bath. Reddish green or brown colour is formed.

Test for flavonoids: 1 ml of sample was taken and 1 ml of 5% liquid ammonia solution was added and mixed. Presence of flavonoids yields a yellow colour.

Test for alkaloids: 2 ml of concentrated hydrochloric acid and 6 drops of hexane was added continuously to 1 ml of sample. 3 ml of picric acid was added. Creaming pale yellow colour is observed indicating alkaloid.

Test for terpenoids: To 1 ml of sample, 5 ml of chloroform was added and mixed well and 1 ml of concentrated sulphuric acid was added. Red colour change is observed.

Test for Tannins: To 5ml of the sample, a few drops of 0.1% Ferric chloride was added to 5ml of the extract. The presence of a blue black color or brownish green indicates the presence of Tannins.

Test for Diterpenes: 4-5 drops of copper acetate solution was added to the extract with mild stirring. The presence of diterpenes is indicated by the formation of emerald green colour.

Total phenolic content

Phenolic content is measured via Folin-Ciocalteu assay, an electron transfer based assay and gives us the reducing capacity which is expressed as the total phenolic content using Gallic acid as standard. 500 µL of various concentrations -depending on solubility or fractions of extracts in water was mixed with 2.5 mL of Folin-Ciocalteu reagent (0.2 N). After a few min 2 mL of Na2CO3 solution (75 g/L) was added, after 120 min standing in dark, the optical density was determined at 760 nm. The total phenolic contents were calculated and expressed as gallic acid equivalents (GAE), on the basis of the calibration curve of gallic acid.

C (GAE)=c x V/M

where:

C=Concentration determined from standard curve (mg/ml)

V=Volume used during the assay (ml)

M=Mass of the assay extract used (g) to give GAE in mg/g extract

RESULTS AND DISCUSSION

From the results, it can be inferred that cladode of *Opuntia dillenii* is rich in alkaloids and has a significant amount of phenols. The total phenolic estimation was found to be 12.6 mg GAE/gm (Table 1). The results showed that cladode extract of the plant is rich in phenols, invariably it includes the antioxidant property due to its redox mechanism [14]. Thus a plant with antioxidant property plays a vital role in free radical scavenging activity. Antioxidants have the potential to suppress cancer and to reduce the risk of cancer development by scavenging ROS [15]. This free radical scavenging mechanism makes the plant potent to fight against disorders. Also, the fruit of Opuntia constitutes a rich amount of phytochemicals and phenols [16].

Table 1: Estimation of total phenol content.

S.no	Plant extract	Concentration of mg GAE/gm
1	Aqueous cladode extract of *Opuntia dillenii*	12.6

Table 2: Preliminary phytochemical screening (Qualitative screening).

S.no	Phytochemicals	Result
1	Phlobatannins	+ +
2	Carbohydrates	+
3	Flavonoids	+ +
4	Alkaloids	+ + +
5	Terpenoids	+ +
6	Tannins	++
7	Diterpenes	+

This involves the therapeutic approach of phenol content of *Opuntia dillenii*. The phytochemical analysis makes the plant more closer to medicinal needs as it gives the sources of medicine such as alkaloids, flavonoids, etc. In our study, various phytochemicals are present of which alkaloids are predominant (Table 2).

Also, moderate amounts of flavonoids, phlobatannins and terpenoids and lesser amounts of carbohydrates were present. This extent or quantity of phytochemicals or phenols will differ according to species even if it is of the same genus [17]. This makes the researchers search for a better plant with high curative value when there is an outcome of disorders or diseases. Then ultimately the selection would be a species with high biological activity, less harm and been used traditionally [18,19]. Generally, plants are medicinal plants due to the presence of rich and diverse phytochemicals such as alkaloids, flavonoids and terpenoids, etc., For example, alkaloids are used in the treatment of cancer [20]. Flavonoids prevent the incidence of many diseases and play a better role in DNA repair and most specifically it is cardioprotective in action [21] in green foods are always a source of attraction in research. The secondary metabolites which are present in plants usually play a protective role in them. As plants are immobile, these phytochemicals are vital in shielding them. They are a part of all ancient medicine and moreover, have little or no side effects. Oxidative stress is found to have a major role in disorders such as diabetes, cardiovascular diseases, and other lifestyle disorders. Antioxidants play an important role in curing all these disorders. Synthetic medicine can never give a cure but only can control the symptoms. Thus, it is important to explore the rich phytochemicals and phenolic compounds of all the herbs which were a part of ancient medicine. One such important herb is Opuntia and from the study it was proved to possess a good

content of phenol and other phytoconstituents and can serves as an important source for pharmaceutical drug preparations. Thus, natural products are still important sources to discover new anticancer drugs and other therapeutics [22].

CONCLUSION

The study result suggests that the phytochemicals (Alkaloids, flavonoids, terpenoids) present in the aqueous extract of *Opuntia dillenii* can be a potential source for chemotherapeutic compounds used in cancer therapy. The phytochemicals and the rich phenol content assessed in this study validates its application as a folkloric medicine in curing diabetes. Further extraction with various solvents and methods needs to be considered to explore all the phytochemicals as solvents are selective in extracting different compounds. The high total phenol content of the extract suggests good antioxidant activity and thus the biomedical applications of all the phyto ingredients need to be explored. Higher the content of bioactive substances, more significant will be its biotherapeutic role.

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CONFLICT OF INTEREST

None declared.

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