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PHYTOCHEMICAL ANALYSIS OF SOME SELECTED INDIAN MEDICINAL PLANTS

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ABSTRACT

Phytochemicals are bioactive compounds obtained from the plants and are widely applied in the traditional herbal medicine. These herbal medicines are used by the local people to cure the various diseases which include the major diseases such as Diabetes Mellitus, Cancer, HIV etc. The objective of the present study was to screen such a phytochemicals as well as the mineral content in selected medicinal plant extracts. Four different plants were taken for analysis viz. *Trigonella foenum-graecum*, *Syzygium cumini*, *Terminalia Chebula* and *Salvadora persica*. It was found that flavonoid is present abundantly in all species. Saponins and tannins are also present in almost all species studied. Finally among the four plant species analyzed for the mineral, we found the highest mineral content i.e. 19% in *Salvadora persica* plant. It was concluded that the plants studied were rich in phytochemicals with significant pharmacological and medicinal applications.

KEYWORDS: **Phytochemicals, Diabetes Mellitus, Flavonoid, Tannins, Saponins.**

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INTRODUCTION

Phytochemicals generally originated from the plant source are nothing but the bioactive compounds also known as secondary metabolites. There are two types of metabolites produced in plants viz. Primary metabolites and Secondary metabolites. Primary metabolites are important for the plants regular metabolism such as growth and development. Secondary metabolites produced by plants may have little need for them. These are synthesize in almost all parts of the plant like bark, leaves, stem, root, flower, fruits, seeds, etc. During past several years, phytochemicals have been used worldwide as the traditional herbal medicine. Because of this pharmaceutical industries as well as researchers put a greater emphasis on the phytochemical studies. Also these phytochemicals present in the different plant parts are used up by the local peoples for healing of certain disorders[1]. These are also widely used in the field of agriculture. Secondary metabolites are economically important in the production of drugs, flavor and fragrances, dye and pigments, pesticides and food additives. Many of the drugs that are derived from the secondary metabolites are simple synthetic modifications or copies of these naturally obtained substances[2].

Following are some of the commercial importance of phytochemicals

1. Limited quantities of nicotine, pyrethrins, and rotenone, are used as pesticides[3].
2. Tannins are generally acts as an astringent[4].
3. Quinones e.g. hypericin can be used as antimicrobial agent[5].
4. Some of the secondary metabolites are used as a pharmacological tool to study various biochemical processes. For example, diterpene esters derived from the latices of various species of Euphorbia is considered as a potent irritants and co-carcinogens, hence important in studies of chemical carcinogenesis[3,6].

Now a day, evolving commercial importance of secondary metabolites has acquired a great interest in analysis as well as production of these natural products and is extensively investigated as a source of medicinal agents[7]. Hence it seemed to be a significant to evaluate the phytochemicals from different plant extracts. In the present study, different plant parts such as seeds and leaves of four different plant species were qualitatively screened for phytochemicals using standard tests. These four medicinally important plant species are *Trigonella foenum-graecum* (Fenugreek), *Syzygium cumini* (Jambul), *Terminalia Chebula* (Hirada) and *Salvadora persica* (Miswak). *Trigonella foenum-graecum* is an annual plant belongs to the family of Fabaceae. It is grown worldwide as a semi-arid crop, and seeds are a common ingredient in an Indian diet. In India, especially it is widely used as a condiment. It also used medicinally to enhance lactation in the nursing mothers. Apart from this, fenugreek also used to cure indigestion and baldness, and possess hypoglycemic and antihyperlipidemic activities[8]. *Syzygium cumini* is an evergreen tropical tree in the flowering plant belongs to family Myrtaceae. Fruits and seeds extracts of *Syzygium cumini* are found to possess an antidiabetic, anti-inflammatory, hepatoprotective, anti-hyperlipidemic, diuretic and antibacterial properties contributed by its secondary metabolites such as saponins, tannins and flavonoids[9]. *Terminalia chebula* is a moderate tree belongs to the family Combretaceae. It is used in traditional medicines and has a common name Black myrobalan, Ink tree or Chebulic myrobalan. It is widely used in the preparation of drugs for various infectious diseases like chronic ulcers, leucorrhoea, pyorrhoea and fungal infections of the skin etc. It is also helpful to prevent aging. It found to impart longevity, increase immunity and body resistance against many diseases[10]. *Salvadora persica* is a natural toothbrush, popularly known as ‘chewing stick’ or ‘miswak’ and belongs to family Salvadoraeceae. It is one of the most popular medicinal plants used to cure the various diseases such as rheumatism, leprosy,

gonorrhoea, ulcers, scurvy, tumors and dental diseases[11].

MATERIALS AND METHODS

Plant Material

The required plant parts were collected from the region of Ahmednagar district, Maharashtra, India and authenticated by the , Department of Botany, Padmashri Vikhe Patil College, Pravaranagar (Loni), Tal.Rahata, District Ahmednagar, (MS), India.

Extraction of plant material

The extraction of plant material was done by Hot water extraction method. The plant material was allowed to dry naturally i.e. under shade drying. After completion of drying process, material was ground in a grinder and the powder was kept in an appropriately labeled plastic bottle. 5gm of ground material was weighed using an electronic weighing balance, dissolved in a 25 ml of sterile water and then boiled at 50-60°C for 30 minutes on water bath. The extract was filtered through Whatman No.1 filter paper and centrifuged the filtrate at 2500 rpm for 15 minutes. Resulting extract was stored in sterile bottles at 4-8°C for further analysis[12].

Phytochemical analysis

Preliminary qualitative screening for phytochemicals, of all these plant species was carried out with the following methods.

Test for Coumarins

2 ml of extract was treated with 3 ml of 10% NaOH. Observed the formation of yellow color indicating the presence of coumarins[12].

Test for Anthocyanins

2 ml of extract was treated with 2 ml of 2N hydrochloric acid and ammonia was added to it. Observed the appearance of pink-red color turning blue-violet. This indicates the presence of anthocyanins[12].

Test for Leucoanthocyanins

5 ml of extract was allowed to react with 5 ml of isoamyl alcohol. Appearance of upper layer red in color indicates the presence of leucoanthocyanins[12].

Test for Fatty acids

0.5 ml of extract was added to 5 ml of ether and allowed it to evaporate on filter paper. Then the filter paper was dried and observed the appearance of transparency on filter paper, the indication of the presence of fatty acids[12].

Test for Steroids

(Libermann Burchard Test)

1 ml of extract was dissolved in 10 ml of chloroform. To this mixture equal volume of concentrated sulfuric acid was added by sides of the test tube. The upper layer becomes red while lower layer of sulfuric acid turns yellow in color with green fluorescence indicating the presence of steroids[12].

Test for Saponins (Foam test)

2 ml of extract was taken in a test tube and 6 ml of distilled water was added to it. The mixture was then shaken vigorously. The persistence of foam was observed that indicates the presence of saponins[12].

Test for Terpenoids (Salkowski test)

2 ml of extract was treated with 2 ml of acetic anhydride. Few drops of concentrated sulfuric acid was then added to this solution and observed the formation of blue, green rings that indicates the presence of terpenoids[12].

Test for Quinones

1 ml of extract was added to the 2 ml of dilute NaOH. Formation of blue green or red coloration confirms the presence of quinones[13].

Test for Tannins (Braymer's test)

2 ml of extract was allowed to react with 10% alcoholic ferric chloride solution. Formation of blue or greenish color of the solution was observed. This was the indication of the presence of the tannins[13].

Test for Phlobatannins *(Precipitate test)*
About 2 ml of extract was added to 2 ml of 1% aqueous hydrochloric acid and the mixture was boiled. Deposition of a red precipitate confirmed the presence of phlobatannins[13].

Test for Phenolic Compounds *(Ferric chloride test)*
Few drops of the extract were treated with 5% aqueous ferric chloride. Formation of deep blue or black color indicates the presences of phenolic compounds[14].

Test for Flavonoids*(Alkaline reagent test)*
2 ml of extract was treated with few drops of 1N sodium hydroxide solution and observed the formation of intense yellow color. This yellow color becomes colorless on addition of dilute hydrochloric acid, indicating the presence of flavonoids[14].

Test for Alkaloids *(Mayer's Test)*
2 ml of extract was treated with 2 drops of Mayer's reagent. Presence of white creamy precipitate indicates the positive test[14].

Determination of Ash content
2 g of each plant sample was taken and weighed accurately in a clean silica dish. The dish was first heated over a low burner flame. After that the dish is transferred to a muffle furnace maintained at 300^0C-450^0C for 3-5 hours. The ash residue obtained was then cooled in desiccator and weighed. The percentage of total ash content was calculated by the formula as follows[15]:

Total Ash Percent of plant sample (%) = [Weight of dry ash residue (g) ÷ Weight of plant sample (g)] x 100

RESULTS AND DISCUSSION

Four plants *Trigonella foenum-graecum, Syzygium cumini, Terminalia Chebula* and *Salvadora persica* were screened for their phytochemical constituent and percent ash content. It was found that all the plants have considerable proportion of important phytochemicals that are easily detected by qualitative tests. In our analysis it was cleared that the *Trigonella foenum-graecum* is rich in alkaloids, flavonoids, saponins, tannins, coumarins, leucoanthocyanins etc. , *Syzygium cumini* is rich in phenolics, flavonoids, saponins, tannins etc., *Terminalia chebula* is rich in favonoids, saponins, tannins, fatty acids, quinones etc., while *Salvadora persica* has slight alkaloid content and rich in flavonoids, coumarins, leucoanthocyanins, steroids and quinones (Table 2). The important thing is that all plant samples contain one common and abundant secondary metabolite, flavonoid. From the literature survey it was found that flavonoids have wide range of biological properties such as anti-inflammatory, antibacterial, antiviral, anti-allergic, cytotoxic antitumor propetrties. It is used in the treatment of neurodegenerative diseases and has vasodilatory action. It is also reported that flavonoids involved in inhibition of lipid-peroxidation, platelet aggregation, capillary permeability and fragility, cyclo-oxygenase and lipoxygenase enzyme activities etc. Flavonoids are also known to inhibit variety of enzymes like hydrolases, hyalouronidases, alkaline phosphatases, arylsulphatases, cAMP phosphodiesterases, lipase, α-glucosidase and kinases. According to the literature, it also has been found that quercetin, plays an important role in diabetes. It leads to the regeneration of pancreatic islets and increases insulin release. It also stimulates Ca^{2+} uptake from isolated islet cells which is helpful in non-insulin dependent diabetes[16].Alkaloids and phenolic compounds along with hypoglycemic, antidiabetic properties [17-20] also exhibit anti-inflammatory, antimicrobial and antioxidant effects[21]. Moreover, saponins exhibit various biological activities like, it gives a permeability to the cell membrane, helpful in lowering the serum cholesterol levels, it possess abortifacient properties, it has immunomodulatory property, it has cytotoxic effects on malignant tumor cells and is involved in synergistic enhancement of the toxicity of

immunotoxins[22]. Saponins also show antidiabetic property[23]. Coumarins, a major class of flavonoids, possesses pharmacological properties like antidiabetic, antioxidant, hepato-protective , anticoagulant, antimicrobial, anti-inflammatory, analgesic, antioxidant, anticancer, antiviral, antimalarial activities etc.[24].Tannins are reported to have a cardio-protective, anti-inflammatory, anti-carcinogenic and antimutagenic properties. Tannins are also involved in treatment of non-insulin dependent diabetes mellitus by enhancing the glucose uptake and inhibiting adipogenesis[25].The most striking feature about quinones is its pharmacological properties that makes it different from other secondary metabolites. It inhibits HIV 1 reverse transcriptase and shows antitumor and immunomodulatory activities[26]. It also has antimicrobial, anticancer, antiviral and antibacterial properties[27].The main purpose to calculate the total ash content is to measure the total amount of minerals present in that plant samples. Among the four plants tested for total ash content, *Salvadora persica* shows highest percentage of the ash i.e. it content 19% (Table 3) of ash indicating high amount of minerals in it. Minerals are used as coenzymes and cofactors in the biochemical processes[28]. Therefore it is necessary to evaluate the mineral values in the extract.

Table 1
Ethnobotanical information of selected medicinal plant species for phytochemical analysis in Ahmednagar district of Maharashtra.

S.No.	Plant species	Local name	Part used
1	*Syzygium cumini*	Jambul	Seed
2	*Terminalia chebula*	Hirada	Seed
3	*Trigonella foenum-graecum*	Fenugreek	Seed
4	*Salvadora persica*	Miswak	leave

Table 2
Preliminary phytochemical analysis of screened medicinal plant species

Sr. No.	Test	*Syzygium cumini*	*Terminalia chebula*	*Trigonella foenum-graecum*	*Salvadora Persica*
1	Alkaloids	-	-	+	+
2	Phenolic Compounds	+	-	-	-
3	Flavonoids	+	+	+	+
4	Saponins	+	+	+	-
5	Tannins	+	+	+	-
6	Phlobatannins	-	-	-	-
7	Coumarins	-		+	+
8	Anthocyanins	-	-	-	-
9	Leucoanthocyanins	-	-	+	+
10	Terpenoids	-	-	-	-
11	Steroids	-	-	-	+
12	Fatty acids	-	+	-	-
13	Quinones	-	+	-	+

Table 3
Analytical data for ash content

Sr. No.	Plant sample	Ash content %
1	*Syzygium cumini*	3.5
2	*Terminalia chebula*	5.5
3	*Trigonella foenum- graecum*	4.0
4	*Salvadora persica*	19

CONCLUSION

From the overall scenario, it is concluded that as the plants studied, found to rich in phytochemicals, are full of pharmacological and medicinal significance. Out of all secondary metabolites flavonoid is found to be abundant in the everywhere almost all plant species studied. Further study is required to find their potentials in the mentioned biological properties such as antidiabetic, anti-tumor, etc.

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