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Phytochemical Screening of *Pistacia chinensis* var. *integerrima*

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Abstract: Phytochemical screening is an important step which leads to the isolation of new and novel compounds. *Pistacia chinenis* var. *integerrima* different parts such as leaves, bark, roots and galls have been selected for phytochemical screening to identify the different classes of secondary metabolites. The galls extract is common practice in folk medicine which revealed the presence of alkaloids, terpenoids, flavonoids and tannins. The bark showed the presence of terpenoids, flavonoids whiles the leaves and roots extracts showed the presence of terpenoids and tannins.

Key word: Phytochemical screening · *Pistacia chinenis* var. *integerrima* · Alkaloids · *Terpenoids* · flavonoids · Tannins

INTRODUCTION

Medicinal plants are used by 80% of the world population for their basic health needs. The relationship between human, plants and drugs derived from plants describe the history of mankind. Plants are the important source of natural drugs. The plants are assumed to contain compounds which have potential to be used in modern medicine for the treatment of diseases which are not curable. Pakistan is rich in countless floras, especially the Khyber Pakhtunkhwa province. The country has more than 6000 species of wild plants, out of them about 400-600 are considered to be medicinally important. During the past decade, traditional systems of medicine have become a topic of global importance. Current estimates suggested that in many developing countries, a large population relies heavily on traditional practitioner and medicinal plants to meet the primary health care needs, although modern medicine may be available in these countries. Herbal medicines have often maintained popularity for historic and cultural reasons. Currently, many people in the developed countries have begun to turn to alternative or complementary therapies, including medicine herbs.

Pistacia chinensis var. *integerrima* (Anacardiaceae) has known as kakra shingi. It's an 18 m moderate sized deciduous tree found in the Himalayas from Indus to Kumaon [1]. The insect of Pemphigus species formed hard horn shaped, rugose, hollow galls like excrescences are on the leaves and petioles of the plant [2]. Dried crushed galls have a very sharp and to some extent bitter in taste and terebinthine odour. The galls are aromatic, astringent, expectorant and has high valued in Ayurvedic medicine as a remedy for asthma, phthisis and other disorders for the respiratory tract, dysentery, chronic bronchitis, hiccough, vomiting of children, skin diseases, psoriasis, fever, snake bite, scorpion sting and also to increase hunger and remove bed humors [3]. In Pakistan, the galls of *Pistacia chinensis* var. *integerrima* are used for the treatment of hepatitis and liver disorders. It has been reported to have depressant [4], analgesic, anti-inflammatory activities [5-8] and hyperuricemic effect [9]. Monoterpenes [5, 6, 10], triterpenoids [5, 6, 11-13], sterol [14, 15], dihydromalvalic acid [16] and flavonoids [15, 17] have been reported from the different parts of others species of *Pistacia*.

METERIALS AND METHODS

Plant Material: *Pistacia chinensis* var. *integerrima* parts such as bark, leaves, roots and galls were collected from Razagram area of district Dir, Khyber Pukhtun Khawa province of Pakistan in the month of February, 2010. The plant materials were identified by plant Taxonomist, Department of Botany, University of Peshawar, Pakistan where the voucher specimen was deposited.

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Extraction: The Plant materials were dried at room temperature for 10 days. The dried plant material (galls, leaves, barks and roots) of *Pistacia chinensis* var. *integerrima* were crushed to make fine powder. The powdered materials were soaked in methanol for 5 days and subjected to extraction until exhaustion of plant materials. The extracts were then concentrated under reduced pressure using rotary evaporator at temperature below 60 °C. Then the crude of methanolic extract was suspended in water and successively partitioned with n-hexane, chloroform, ethyl acetate.

Phytochemical Screening: Chemical tests were carried out on the n-hexane, chloroform, ethyl acetate and methanol extracts of different parts (galls, leaves, barks and roots) of *Pistacia integerrima* using standard procedures to identify the constituents as described by Sofowora [18], Trease [19] and Evans and Harborne [20].

Alkaloids: About 0.2g of each extract was wormed with 2% H_2SO_4 for two minutes. It was filtered and a few drops of Dragendroffs reagent were added. Orange red precipitate indicated the presence of alkaloids.

Tannins: A small quantity of each extract was mixed with water and heated on water bath and filtered. A few drops of ferric chloride were added the filtrate. A dark green solution indicates the presence of tannins.

Anthraquinones: About 0.5 g of each extract was boiled with 10 % HCl for few minutes in water bath. It was filtered and allowed to cool. Equal volume of $CHCl_3$ was added to the filtrate. Few drops of 10% ammonia was added to the mixture and heated. Formation of rose-pink color indicates the presence of anthraquinones.

Glycosides: The extracts were hydrolyzed with HCl and neutralized with NaOH solution. A few drops of Fehling solution A and B were added. Red precipitate indicates the presence of glycosides.

Reducing Sugars: The extracts were shaken with distilled water and filtered. The filtrate was boiled with few drops of Fehling solution A and B for few minutes. An orange red precipitate indicates the presence of reducing sugars.

Saponins: About 0.2 g of the extract was shaken with 5ml of distilled water and then heated to boil. Frothing (appearance of creamy miss of small bubbles) shows the presence of saponin.

Flavonoids: Extract of about 0.2 g was dissolved in diluted NaOH and Hcl was added. A yellow solution that turns colorless indicates the presence of flavonoids.

Phlobatanins: The extract (0.5 g) was dissolved in distilled water and filtered. The filtrate was boiled with 2% HCl solution. Red precipitate shows the presence of phlobatanins.

Steroids: 2 ml of acetic anhydride was added to 0.5 g of the extract of each with 2 ml of H_2S0_4. The colour changed from violet to blue or green in some samples indicate presence of steroids.

Terpenoids Test: 0.2 g of the each extract was mixed with 2 ml of chloroform and concentrated H_2SO_4 (3ml) was carefully added form a layer. A reddish brown coloration of the interface was formed to indicate positive results for the presence of terpenoids.

RESULTS

The weight percentage yield of the crude extracts of n-hexane, chloroform, ethyl acetate and methanol of *Pistacia chinensis* var. *integerrima* galls, leaves, barks and roots are given in Table 1.

The phytochemical screening of n-hexane, chloroform, ethyl acetate and crude methanol extracts of *Pistacia integerrima* galls, leaves, barks and roots are shown is in Tables 2, 3, 4 and 5, respectively.

Table 1: Extractive values of different parts of *Pistacia integerrima.*

Parts	Methanol	Hexane	Chloroform	Ethyl acetate
Galls	3%	7.6%	10.7%	34.4%
Leaves	2%	12.2%	24.6%	35%
Bark	1.8%	10%	12.83%	41.66%
Roots	4%	9.5%	11.7%	20%

Table 2: Phytochemical screening of n-hexane, chloroform, ethyl acetate and methanol crude extracts of *Pistacia integerrima* galls.

Chemical components	n-Hexane extract	Chloroform extract	Ethyl acetate extract	Methanol extract
Alkaloids	–	–	+	+
Steroids	–	–	–	
Terpenoids	–	+	+	+
Flavonoids	+	+	+	+
Anthraquinones	–	–	–	–
Tannins	–	+	+	+
Phlobatanins	–	–	–	–
Saponins	–	–	–	+
Glycoside	–	–	–	–
Reducing sugars	–	+	+	+

Table 3: Phytochemical screening of hexane, chloroform, ethyl acetate and methanol crude extracts of *Pistacia integerrima* Leaves.

Chemical components	n-Hexane extract	Chloroform extract	Ethyl acetate extract	Methanolextract
Alkaloids	–	–	–	–
Steroids	–	–	–	–
Terpenoids	-	+	+	+
Flavonoids	–	–	–	–
Anthraquinones	–	–	–	–
Tannins	–	–	+	+
Phlobatanins	–	–	–	–
Saponins	–	–	–	–
Glycoside	–	–	–	–
Reducing sugars	–	–	–	–

Table 4: Phytochemical screening of hexane, chloroform, ethyl acetate and methanol crude extracts of *Pastacia integerrima* barks.

Chemical components	n-Hexane extract	Chloroform extract	Ethyl acetate extract	Methanolextract
Alkaloids	–	–	–	–
Steroids	–	–	–	–
Terpenoids	–	+	+	+
Flavonoids	–	+	+	+
Anthraquinones	–	–	–	–
Tannins	–	–	–	–
Phlobatanins	–	–	–	–
Saponins	–	–	–	–
Glycoside	–	–	–	–
Reducing sugars	–	+	–	+

Table 5: Phytochemical screening of n-hexane, chloroform, ethyl acetate and methanol crude extracts of *Pastacia integerrima* roots.

Chemical components	n-Hexane extract	Chloroform extract	Ethyl acetate extract	Methanolextract
Alkaloids	–	–	–	–
Steroids	–	–	–	–
Terpenoids	–	+	+	+
Flavonoids	–	–	–	–
Anthraquinones	–	–	–	–
Tannins	–	–	+	+
Phlobatanins	–	–	–	–
Saponins	–	–	–	–
Glycoside	–	–	–	–
Reducing sugars	–	+	–	+

DISCUSSION

Phytochemical screening of the plant material revealed the presence of the major secondary metabolites; alkaloids, terpenoids, tannins and flavonoids. Quantitative estimations of bioactive constituents are summarized in Tables 2-5.

The crude methanolic extract of *Pistacia integerrima* contains all the polar and non polar components present in the galls. Secondary metabolite constituents detected in the ethyl acetate and methanolic crude fraction of plant is alkaloids which have not been reported. Alkaloids have numerous functions and among them fore most are their analgesics, anti inflammatory, antispasmodic and bactericidal effects [21]. Apart of alkaloids the medicinal properties such as analgesic and anti inflammatory activates of the galls extract may also be attributed to the presence of triterpenes, tannins and flavonoids. The specific activities that may be attributed to the presence of alkloids need further investigation. The use of galls in the liver disorder and the presence of flavonoids and tannins attribute its use in folk medicine.

Small amount of extract was obtained from barks and leaves as compared to roots and galls. The crude methanolic, ethyl acetate and chloroform extracts obtained from leaves showed the presence of terpenoids and tannins, while bark the extract showed the presence of terpenoids and flavonoids. The presence of such components in bark may have good anti inflammatory and analgesic activities but no work have been reported so far. Similarly the roots extracts showed the presence of flavonoids only. Medicinal plants is also rich in tannins and contribute property of astringency i.e., fasten the healing of wounds and inflamed mucous membranes [21]. The presence of tannins in the galls, leaves and roots fractions were indicated the medicinal importance of this plant that associated with reported activities [3-9, 21].

The present screening study proved useful tool for the comparative studies of the amount of bioactive principles present in different parts of the plant with its other species and populations belonging to different regions with different climatic conditions. These data can also help us to choose the superior race of this valuable herb with greater quantity of medically and therapeutically important phytochemically.

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