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PHYTOCHEMICAL SCREENING FOR ACTIVE COMPOUNDS IN *PEDALIUM MUREX* L.

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Abstract

Pedalium murex (Pedaliceae) has been traditionally used for the treatment of puerperal diseases, digestive tonics, ulcers, fevers, wounds other ailments and general debility. The present investigation was intented to evaluate the preliminary phytochemical characters of this plant. Phytochemical studies facilitate new discovery for the synthesis of more potent drugs.

Keywords: Pedalium murex, leaves, fruits, solvents and active compounds

Introduction

For past few decades compounds from natural sources have been gaining importance because of the vast chemical diversity that they offer. This had led to phenomenal increase in the demand for herbal medicines in the last two decades and or need has been felt for ensuring the quality, safety and efficacy of herbal drugs. The medicinal value of these plants lies in some chemical substances that produce a definite physiological action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids and phenolic components (Hill, 1952). Ayuerveda, an ancient system of the Indian medicine, cites several plants that are useful in the treatment of urogenital disorders. Prostane, a herbal formulation consist of Tribulus terrestris, Areca catechu, Pedalium murex, Caesalpinia bonducella, and Asparagus racemosus. T. terrestris and Pedalium murex have been reported to be good diuretics (Prasad and Sastry, 1987).

Pedalium murex is an important medicinal plant that contains several alkaloids like pedalitin, Diosmetin, Dinatin, Pedalin dinatin-7-glucuronide (Subramanian and Nair, 1972). The leaf decoction is used to control white discharge due to excessive body heat. Root decoction is used as an antibilious agent, while the juice of the fruit is used as an emmenagogue and to promote lochial discharge (Satyavathi et al., 1987). The decoction of the seeds and glycosides obtained from it showed mild diuretic activity and the alcoholic extract of the fruits reduced blood pressure in dog and rat (Haravey, 1996). It is reported that many Indian medicinal plants show beneficial effects against renal injury (Ali et al., 2006). A good example is a succulent herb, Pedalium murex Linn, commonly called Gokhru a member of family Pedaliaceae. It is commonly found in Deccan and in some parts of Ceylon and Gujarat and in the coastal areas of southern India (Nadkarani, 1982). It is about 15 to 40 cm in height, having four angle spiny brownish colour fruits (1-2 cm). The fruits are rich in flavonoids, sapogenin (diosgenin-0.06%) and soluble proteins (20.14mg/ym), (Mukherjee, 2002). An infusion extract prepared using cold water from the leaves, stems and fruits of *P.murex* is demulcent, diuretic and also found to be useful in the treatment of disorders of urinary systems such as gonorrhea, dysuria, incontinence of urine, etc. (Chopra *et al.*, 1999; Shukla *et al.*, 2004).

The knowledge of the chemical constituents of plants would further be valuable in discovering the actual value of folkloric remedies (Farnsworth, 1996). Research on medicinal plants has been increased and screened for their antimicrobial activities in number of studies. Hence, an attempt was made to evaluate the phytochemical properties of *Pedalium murex* leaves and fruits.

Material and Methods

Pedalium murex L. leaves and fruits were collected from Gopalapuram, Theni Dist, Tamilnadu, India and authenticated by Dr.V.Balasubramanian, Associate Professor, PG and Research Department of Botany, Kongunadu Arts and Science College, Coimbatore, Tamilnadu, India. The voucher specimen of *Pedalium murex* (78/Jan/2009) was deposited in the Research Dept of Botany, Kongunadu Arts and Science College, Coimbatore, Tamilnadu, India.

The fresh leaves and fruits were shade dried and ground using mechanical motor at the PG and Research Department of Botany, Kongunadu Arts and Science College, Coimbatore. The powdered material (50gm) was transferred into a Soxhlet apparatus containing 200ml of respective solvents (Petroleum ether, Chloroform, Acetone and Methanol). The extract was concentrated to dryness under vacuum dessicator.

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The extracts obtained was stored and later used for phytochemical screenings.

Phytochemical screenings

The leaves and fruit extracts of *Pedalium murex* was analysed for the presence of flavonoids, alkaloids, glycosides, steroids, phenols, saponins, terpenoid, cardiac glycosides and tannins according to standard methods (Harborne, 1973; Sofowora, 1982; Trease and Evans, 1989).

Steroids (Salkowski's test)

About 100mg of *Pedalium murex* dried extract was dissolved in 2ml of chloroform. Sulphuric acid was carefully added to form a lower layer. A reddish brown colour at the interface was an indicative of the presence of steroidal ring.

Cardiac glycosides (Keller killiani's test)

About 100mg of extract was dissolved in 1ml of glacial acetic acid containing one drop of ferric chloride solution. This was then underlayer with 1ml of concentrated sulphuric acid. A brown ring obtained at the interface indicated the presence of a de oxy sugar characteristic of cardenolides.

Saponins

A drop of sodium bicarbonate was added in a test tube containing about 50ml of an aqueous extract of samples. The mixture was shaken vigorously and kept for 3minuts. A honey comb like froth was formed and it showed the presence of saponins.

Resins

To 2ml of chloroform or ethanolic extract, 5 to 10ml of acetic anhydrite was added and dissolved by gentle heating. After cooling, 0.5ml of H_2SO_4 was added. Bright purple colour was produced. It indicated the presence of resins.

Phenols (Ferric Chloride Test)

To 1ml of alcoholic solution of sample, 2ml of distilled water followed by a few drops of 10% aqueous ferric chloride solution were added. Formation of blue or green colour indicated the presence of phenols.

Tannins (Lead acetate test)

In a test tube containing about 5ml of an aqueous extract, a few drops of 1% solution of lead acetate was added. Formation of a yellow or red precipitate indicated the presence of tannins.

FeCl₃ test

A 2ml filtrate [200mg of plant material in 10ml distilled water, filtered], and 2ml of FeCl3 were mixed. A blue or black precipitate indicated the presence of tannins.

Terpenoid

2ml of chloroform and 1ml of conc. H2SO4 was added to 1mg of extract and observed for reddish brown colour that indicated the presence of terpenoid.

Glycosides

A small amount of alcoholic extract of samples was dissolved in 1ml water and then aqueous sodium hydroxide was added. Formation of a yellow colour indicated the presence of glycosides.

Flavonoids

In a test tube containing 0.5ml of alcoholic extract of the samples, 5 to 10 drops of diluted HCl and small amount of Zn or Mg were added and the solution was boiled for few minutes. Appearance of reddish pink or dirty brown colour indicated the presence of flavonoids.

Alkaloids (Mayer's test)

1.36gm of Mercuric chloride and 5gm of KI were dissolved in 60ml and 10 ml of distilled water respectively. These two solvents were mixed and diluted to 100ml using distilled water. To 1ml of acidic aqueous solution of samples few drops of reagent was added. Formation of white or pale precipitate showed the presence of alkaloids.

Results and Discussion

This study on *Pedalium murex* leaves extract revealed the presence of glycosides, flavonoids, phenols, steroids, saponins, terpenoid and tannins. Presence of alkaloid, resins and cardiac glycosides was not noticed [Table 1]. Fruit extract of this plant contained flavonoids, alkaloids, glycosides, steroids, phenols, saponins, terpenoid, tannins and cardiac glycosides. There was no resins [Table 2].

Phytochemical constituents		Petroleum ether	Chloroform	Acetone	Methanol
Flavonoids		-	-	-	+++
Alkaloids		-	-	-	+
Glycosides		-	++	+++	+++
Steroids		-	-	-	+++
Phenols		-	-	++	+++
Terpenoid			-	-	+++
Saponins		-	+	+	+++
Resins		-	-	-	-
Tannins FeCl :	a test	-	-	++	+++
Lead	acetate test	-	-	-	+++
Cardiac glycosides		-	-	-	-

Table 1: Phytochemical screenings of Pedalium murex leaf extracts

- =absent, + = present, ++ = moderately present, +++ =Appreciable amount

Phytochemical constituents		Petroleum ether	Chloroform	Methanol
Flavonoids		-	-	+++
Alkaloids		-	++	+
Glycosides		-	++	+++
Steroids		++	+	+++
Phenols		-	+	++
Terpenoid		+	++	+++
Saponins		+	-	+++
Resins		-	-	-
Tannins	FeCl 3 test			+++
	Lead acetate test	++		++
Cardiac glycosides		+++	++	

Table 2: Phytochemical screenings of Pedalium murex fruit extracts

- =absent, + = present, ++ = moderately present, +++ =Appreciable amount

These findings are in agreement with the studies of Kelmanson *et al.* (2000) and Nair *et al.* (2005). Preliminary phytochemical analysis of *Pedalium murex* methanol extract showed the presence of reducing sugars, phenolic compounds, saponins, xanthoprotein, alkaloids, triterpenoids, tannins and flavonoids. Srinivasa Rao *et al.* (1999), Ananthakrishnan (2002) were used ethanol as a solvent for the extraction of different secondary metabolities of these plants. Since the polarity of ethanol is higher, most of the secondary metabolities of *Pedalium murex* dissolved in ethanol.

Cardiac glycosides on the other hand are known to work by inhibiting the Na^{+/}K⁺ pump. This causes an increase in the level of sodium ions in the calcium ion. This inhibition increases the amount of Ca²⁺ ions available for concentration of the heart muscle which improves cardiac output and reduces distention of heart; thus are used in the treatment of congestive heart failure and cardiac arrhythmia (Schneider and Wolfling, 2004)

Flavonoids have been referred to as nature's biological response modifiers because of strong experimental evidence of their inherent ability to modify the body's reaction to allergen, virus and carcinogens. They show anti-allergic, anti-inflammatory, anti-microbial and anti-cancer activity [http://en.wikipedia.org/ wiki / flavonoids].

Flavonoids and tannins are phenolic compounds that are a major group of compounds act as primary antioxidants or free radical scavengers (Polterait, 1997).

Tannins are known to possess general antimicrobial and antioxidant activities (Rievere *et al.*, 2009). Recent reports show that tannins may have potential value as cytotoxic and antineoplastic agents (Aguinaldo *et al.*, 2005). Other compounds like saponins also have antifungal properties (Aboada and Efuwape, 2001; Mohanta *et al.*, 2007).

Saponins is a mild detergent used in intracellular histochemistry staining to allow antibody access to intracellular proteins. In medicine, it is used in hyper cholestrolaemia, hyperglycemia, antioxidant, anticancer, anti inflammatory and weight loss, etc. It is also known to have anti-fungal properties (De-Lucca *et al.*, 2005). Saponins have been implicated as a bioactive antibacterial agents of plants (Mandal *et al.*, 2005: Manjunatha, 2006).

Plant steroids are known to be important for their cardiotonic activities, possess insecticidal and antimicrobial properties. They are also used in nutrition, herbal medicine and cosmetics (Callow, 1936).

Plant derived natural products such as flavonoids, terpenoids and steroids etc have received considerable attention in recent years due to their diverse pharmacological properties including antioxidant and antitumor activity (De Feudis *et al.*, 2003; Takeoka and Dao, 2003).

Phenolic phytochemicals have antioxidative, antidiabetic, anticarcinogenic, antimicrobial, antiallergic, antimutagenic and anti-inflammtory (Arts and Hollman, 2005; Scalbert *et al.*, 2005).

Conclusion

Pedalium murex is a valuable source of medicinally useful compounds that have been used traditionally for various aliments. Leaf and fruit extracts of this plant showed good source for the bioactive compounds. Thus plant studied here can be a potential source for useful drugs, if it is involved in further research.

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