

CHEMOPROFILE INVESTIGATION OF SELECTED PLANTS USED IN DIABETES

Kalyani Thakare, Dr. Sahadeo P. Rothe*

P.G. Department of Botany, Shri Shivaji College of Arts, Commerce and Science, Akola

Address for correspondence: sprothe@rediffmail.com; Contact - +919822239825

ABSTRACT

Diabetes mellitus (DM) is a chronic, metabolic disease characterized by hyper-glycemia. High blood glucose levels may result in microvascular complications such as retinopathy, nephropathy, and neuropathy. More than 1000 plant species i.e. traditional herbal medicines are used against diabetes as folk medicine. The knowledge of system of treating Diabetes mellitus, as the history reveals, existed with the Indians since prehistoric age. Present study deals to find out the phytoconstituents of 5 medicinal plants viz. Punica granatum L., Acacia nilotica (L.) Del., Bacopa monnieri (L.) Wettst., Ficus racemosa L. and Butea monosperma (Lamk.) Taub. which are frequently used in hyperglycaemic conditions.

Keywords: Hyper-glycemia, traditional herbal medicines, phytoconstituents, etc.

INTRODUCTION:

Diabetes mellitus (DM) is a chronic, metabolic disease of the endocrine system characterized by hyper-glycemia. High blood glucose level may be a result of several factors such as insufficient insulin production of pancreas, or resistance of the body to the action of insulin. Uncontrolled blood glucose levels may result in microvascular complications such as retinopathy, nephropathy, and neuropathy. It may also lead to vascular complications such as coronary artery disease resulting in strokes and myocardial infarctions (Ghosh and Collier, 2012). Thus, it is aimed to lower elevated blood glucose levels to prevent morbidity and mortality resulting from diabetes. It is estimated that there are 300 million people suffering from diabetes in the world. This number keeps increasing as a result of insulin resistance triggered by unhealthy eating habits and sedentary lifestyles. In modern medicine, insulin and/or oral agents such as sulfonylureas, biguanides, meglitinides, alpha-glycosidase inhibitors, thiazolidinediones and incretinmimetics are used for the treatment of diabetes. On the other hand, traditional herbal medicines are used all over the world by diabetic patients. It is estimated that more than 1000 plant species are used against diabetes as folk medicine (Marles and Farnsworth, 1995). Research on herbal medicines is encouraged to come up with alternatives for treatment of diabetes since oral antidiabetic agents have side effects in the long run (Samad *et al.*, 2009). Plants are considered to be the basis for deriving natural or semi-synthetic constituents that can be used against diabetes. The antidiabetic activity of some herbal medicines is attributed to the presence of compounds such as flavonoids, terpenoids, coumarins, and phenolic compounds, among others (Jarald *et al.*, 2008; Rao *et al.*, 2010). Investigations must be carried out to identify the chemical constituent(s) responsible for the antidiabetic activity of the medicinal plants, and to elucidate their mechanism of action. However, little knowledge exists on the medicinal use of some plants within the Cypriot culture (Georgiades, 1987, 1992; Della *et al.*, 2006; Lardos, 2006; Ozan, 2011).

Also, with increasing incidence of diabetes mellitus in rural population throughout the world and due to adverse effects of synthetic medicine, there is a clear need for development of indigenous, inexpensive botanical sources for anti-diabetic crude or purified drugs (Venkatesh *et al.*, 2003). As per ancient literature, more than 800 plants are reported to have anti-diabetic properties (Eddouks and Maghrani, 2004). Ethnopharmacological surveys indicate that more than 1200 plants are used in traditional medicine for their alleged hypoglycemic activity (Kesari

et al., 2007). Medicinal plants, since ancient times, have been used in virtually all cultures as a source of medicine. A study of ancient literature indicates that diabetes was fairly well known and well-conceived as an entity in ancient India. The knowledge of system of treating Diabetes mellitus, as the history reveals, existed with the Indians since prehistoric age. Its earliest reference (1000 BC in the Ayurvedic literature) is found in mythological form where it is said to have originated by eating Havisha, (Latha and Pari, 2003); a special food, which was offered at the times of yagna organized by Dakshaprajapati. Ayurvedic antidiabetic herbs improve digestive power, increase one of the Rasas (gastric secretions); being Laghu, get easily digested in the body; and being Ruksha, decrease output of overall body fluids e.g. urine, sweat etc. Food items, which are 'madhumehaghna' (antidote), are an important underlying principle of therapy for the prameha (diabetes) patient. Food items which correct the metabolic imbalance by their action e.g. foods exhibiting 'rasa', 'katu', 'laghu', 'medaghna', properties are old cereals, roasted cereals, barley, jawar, ragi, mung dal, horsegram, tur dal, drumstick, leaves, bittergourd, jamun, amla, fig, raw papaya, milk, meat of animals that live in dry region, etc. The indigenous diet may not be useful in lowering the blood sugar to the same extent as insulin and other hypoglycaemic agents do. But it has some other influences, which may be useful for the management of the disease and its complications (Subbulakshmi and Naik, 2001). Present study deals to find out the phytoconstituents of 5 medicinal plants which are frequently used in hyperglycaemic conditions.

MATERIALS AND METHODS

Collection of plant material

The plants selected for the study are *Punica granatum* L., *Acacia nilotica* (L.) Del., *Bacopa monnieri* (L.) Wettst., *Ficus racemosa* L. and *Butea monosperma* (Lamk.) Taub. were collected during month of November - December, 2016 and February, 2017 from various forest localities of Akola district (MS). The plant material and specimens was identified by using standard floras like Cooke 1907, Dhore 2005, Naik 1989. The voucher specimens were preserved in the institute herbarium library.

The collected plant material (Leaves, Flowers and Fruits) were washed with tap water and then distilled water. Then the material is shade dried for 4-5 days and grinded well to obtain homogenous fine grade powder. The 5gm powdered material soaked in each 50 ml of distilled water and alcohol for 1 hour. The solvent was filtered and the preliminary tests were carried out.

Preliminary phytochemistry:-

The preliminary phytochemical studies are done for detection of various constituents i.e. alkaloids, glycosides, carbohydrates, etc. present in plant extract, which is responsible for the pharmacological activity. Chemical tests were carried out on the successive extracts separately using standard procedures to identify the constituents as described by (Harborne, 1973; Sofowora, 2000).

Test for Alkaloid: 2 ml of plant extract in separate test tube and warmed with 2% sulphuric acid for 2 min. and it was filtered in separate test tube and few drops of Dragendorff's reagent was added and observed for the presence of orange red precipitate for the presence of alkaloid.

Test for glycosides: 2 ml of plant extract in separate test tube with 2ml of glacial acetic acid containing of drops of ferric chloride solution and observe for brown ring formation at the interface, confirms the presence of cardiac glycosides.

Test for Terpenoids: Take about 2 ml extract in separate test tube and add 2 ml chloroform and 3 ml of sulphuric acid in it. A reddish brown coloration at interface confirms the presence of terpenoids.

Test for Reducing sugar: 1ml extract in 4 ml distilled water taken in test tube and shake well, filter the extract and add few drops of Fehling solution A and B and boil for 2 min. Orange red precipitate confirms the presence of reducing sugar.

Test for steroids: To 2 ml of the plant extract add 2ml of acetic anhydride and add 2 ml of ethanol with 2ml of sulphuric acid. Violet to blue or green colour indicates the presence of steroids.

Test for saponins: 2 ml of sample was added in 10 ml of distilled water and shaken well. Froth formation confirms the presence of saponins.

Test for Tannins and Phenolics: 2 ml of plant extract was added in 2 ml distilled water and heated in water bath, then filtered and 5% Ferric chloride was added. Dark green-black colour indicates the presence of tannins.

Test for flavonoids: To 2ml of plant extract add 10% NaOH, yellow colour appears which faint on addition of concentrated HCl, which confirms the presence of flavonoids.

Test for carbohydrates: A small portion of filtrate was treated with Molish reagent and sulphuric acid, formation of a violet ring indicates the presence of carbohydrates.

Test for proteins and amino acids: Add few drops of Millon's Reagent to 2 ml of plant extract. White colored precipitate confirms the presence of proteins.

Test for coumarins: 1 gm of powder drug kept with water in a test tube, covered with paper, soaked in NaOH is diluted and boiled. Yellow fluorescence indicates the presence of coumarins after examination under UV lamp.

Test for Anthocyanin and Betacyanin: 2 ml extract was added to 1ml of 2N Sodium hydroxide and heated for 5 minutes at 100°C. Formation of bluish green colour indicates the presence anthocyanin and yellow colour indicates betacyanin.

Test for Anthraquinone: To 2 ml of extract, add 5 ml of dil. H₂SO₄. The solution was then boiled and filtered. Add equal volume of benzene to the filtrate. The solution was shaken well and the organic layer was separated. Equal volume of dilute Ammonia solution added to the organic layer. The Ammonia layer turn pink showing the presence of Anthraquinone.

OBSERVATION AND RESULT:

Preliminary phytochemistry was done in order to find out the chemical composition in the shade dried plant materials. The results observed are enumerated in the table.

Table No. 1: Qualitative Phytochemical tests for the selected medicinal plant parts.

Sr.No.	Tests	<i>P. granatum</i> L. Fruit		<i>A. nilotica</i> (L.) Del. Fruit		<i>B. monnieri</i> (L.) Wettst. Leaf		<i>F. racemosa</i> L. Fruit		<i>B. monosperma</i> (Lamk.) Taub. Flower	
		EtOH	D.W	EtOH	D.W	EtOH	D.W	EtOH	D.W	EtOH	D.W
1	Sterols	-	-	-	-	-	-	-	-	+	-
2	Terpenoids	-	-	-	-	-	-	-	-	+	+
3	Saponins	-	-	+	+	-	+	-	-	-	+
4	Glycosides	+	+	-	-	-	-	-	-	-	-
5	Alkaloids	-	-	-	-	-	-	-	-	-	-
6	Carbohydrates	+	-	-	+	+	-	-	-	-	-
7	Tannin & Phenolics	+	+	+	+	-	-	-	-	+	+
8	Flavonoids	-	-	-	+	-	-	-	-	+	+
9	Protein & amino acids	-	-	-	-	+	-	-	+	+	-
10	Coumarins	-	-	-	-	-	-	-	-	-	-
11	Anthocyanin	-	-	-	-	-	-	-	-	+	-
12	Betacyanin	-	-	-	-	-	-	-	-	-	-
13	Anthraquinine	-	-	-	-	-	-	-	-	-	-

*EtOH = Ethanol; D.W= Distilled water

DISCUSSION AND CONCLUSION

Phytochemical analysis

For the phytochemical analysis, extract of the above mentioned plant parts were prepared in two different solvents viz. Ethanol and Distilled water.

Preliminary Phytochemistry

The preliminary phytochemical studies were carried out in the solvents viz. Ethanol and Distilled water.

In ethanol solvent of fruit of ***Punica granatum L.***, when the extract were studied the test were positive for glycosides, carbohydrates, tannins and phenols whereas, negative for sterols, terpenoids, saponins, alkaloids, flavonoids, amino acids and proteins, coumarins, anthocyanin, betacyanin and anthraquinines.

In aqueous solvent of fruit of ***Punica granatum L.***, when the extract were studied the test were positive for saponins, carbohydrates, whereas, negative for glycosides, terpenoids, sterols, alkaloids, tannins and phenolics, flavonoids, amino acids and proteins, coumarins, anthocyanin, betacyanin and anthraquinines.

In ethanol solvent of fruit of ***Acacia nilotica(L.) Del.***, when the extract were studied the test were positive for saponins, tannins and phenols whereas, negative for sterols, terpenoids, glycosides, alkaloids, carbohydrates, flavonoids, amino acids and proteins, coumarins, anthocyanin, betacyanin and anthraquinines.

In aqueous solvent of fruit of ***Acacia nilotica(L.) Del.***, when the extract were studied the test were positive for saponins, carbohydrates, tannins and phenols and flavonoids whereas, negative for sterols, terpenoids, glycosides, alkaloids, amino acids and proteins, coumarins, anthocyanin, betacyanin and anthraquinines.

In ethanol solvent of leaves of ***Bacopa monnieri (L.)Wettst.***, when the extract were studied the test were positive for carbohydrates, amino acids and proteins whereas, negative for sterols, terpenoids, saponins, glycosides, alkaloids, tannins and phenols, flavonoids, coumarins, anthocyanin, betacyanin and anthraquinines.

In aqueous solvent of leaves of ***Bacopa monnieri (L.)Wettst.***, when the extract were studied the test were positive for saponins only whereas, negative for sterols, terpenoids, glycosides, alkaloids, carbohydrates, tannins and phenols, flavonoids, amino acids and proteins, coumarins, anthocyanin, betacyanin and anthraquinines.

In ethanol solvent of fruit of ***Ficus racemosa L.***, when the extract were studied the test were positive for none whereas, negative for sterols, terpenoids, saponins, glycosides, carbohydrates, tannins and phenols, alkaloids, flavonoids, amino acids and proteins, coumarins, anthocyanin, betacyanin and anthraquinines.

In aqueous solvent of fruit of ***Ficus racemosa L.***, when the extract were studied the test were positive for amino acids and proteins only whereas, negative for sterols, saponins, terpenoids, glycosides, carbohydrates, tannins and phenols, alkaloids, flavonoids, , coumarins, anthocyanin, betacyanin and anthraquinines.

In ethanol solvent of flower of ***Butea monosperma (Lamk.)Taub.***, when the extract were studied the test were positive for sterols, terpenoids, tannins and phenols, flavonoids, amino acids and proteins and anthocyanins, whereas, negative for saponins, glycosides, alkaloids, carbohydrates, coumarins, betacyanin and anthraquinines.

In aqueous solvent of flower of ***Butea monosperma (Lamk.)Taub.***, when the extract were studied the test were positive for terpenoids, saponins, flavonoids, tannins and phenols whereas, negative for sterols, glycosides, alkaloids, carbohydrates, amino acids and proteins, coumarins, anthocyanin, betacyanin and anthraquinines.

REFERENCES:

- Cooke, Theodore (1907). "The Flora of the Presidency of Bombay". Vol. I & Vol. II, Botanical Survey of India; Calcutta.
- Dhore M. A., 2005, Flora of Amravati District. Publ. S. G. B. Univ. Amravati.
- Della A, Paraskeva-Hadjichambi D, Hadjichambis A (2006). An Ethnobotanical Survey of Wild Edible Plants of Paphos and Larnaca Countryside of Cyprus. *J. Ethnobiol. Ethnomed.* 2:34.
- Eddouks M, Maghrani M. Phlorizin-like effect of *Fraxinus excelsior* in normal and diabetic rats. *J Ethnopharmacol*2004;9:149-54.
- Georgiades CC (1987). Flowers of Cyprus, Plants of Medicine. Nicosia, Cyprus. 1:103.
- Georgiades CC (1992). Flowers of Cyprus, Plants of Medicine. Nicosia, Cyprus. 2:99.
- Ghosh S, Collier A (2012). Churchill's Pocketbook of Diabetes. 2nd edition. Elsevier Health Sci.
- Harbone, J.B.: Phytochemical Methods- A Guide to Modern Techniques of Plant Analysis, Chapman and Hall London, 1998.
- Jarald E, Joshi SB, Jain DC (2008). Diabetes and herbal medicines. *Iranian J. Pharmacol. Ther.* 7(1):97-106.
- Kesari AN, Kesari S, Santosh KS, Rajesh KG, Geeta W. Studies on the glycemic and lipidemic effect of *Murrayakoenigi* in experimental animals. *J Ethnopharmacol*2007; 112(2): 305-11.
- Lardos A (2006). The botanical *materiamedica* of the *Iatrosophikon* -A collection of prescriptions from a monastery in Cyprus. *J.Ethnopharmacol.* 104:387-406.
- Latha M, Pari L. Antihyperglycaemic effect of *Cassia auriculata* in experimental diabetes and its effects on key metabolic enzymes involved in carbohydrate metabolism. *ClinExpPharmacolPhysiol*2003;30(1-2):38-43.

- Marles R, Farnsworth N (1995). Antidiabetic plants and their active constituents. *Phytomedicine* 2:137-189.
- Naik V.N 1989, *Flora of Marathwada* Vol. I & Vol. II AmrutPrakashan, railway station road, Aurangabad.
- Ozan Ç (2011). KuzeyKıbrıs'takullanılantibbitkiler. [An Investigation on the Medicinal Plants of Northern Cyprus.]. Master of Science thesis, Near East University Institute of Health Sciences Department of Pharmaceutical Botany/ Pharmacognosy, Nicosia. Turkish Republic of Northern Cyprus.
- Rao M, Sreenivasulu M, Chengaiah B, Reddy K, Chetty C (2010). Herbal medicines for diabetes mellitus: A review. *Int. J. Pharm. Tech. Res.* 2(3):1883-1892.
- Samad A, Shams MS, Ullah Z, Wais M, Nazish I, Sultana Y, Aqil M (2009) Status of herbal medicines in the treatment of diabetes: a review. *Curr. Diabetes Rev.* 5(2):102-111.
- Sofowora, E.E., 2000. Phytochemical screening of Nigerian medicinal plants lloydia J. *Integrative Med.*, 41: 234-246
- Subbulakshmi G, Naik M. Indigenous foods in the treatment of diabetes mellitus. *Bombay Hospital J.* 2001;43(4):548-61.
- Venkatesh S, Reddy GD, Reddy BM, Ramesh M, Apparao AVN. Antihyperglycemic activity of *Carullumaasttenuate*. *Fitoterapia.* 2003;74:274-7.

