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Research Article

Extraction, Separation Isolations and Phytochemical Investigation of the Insulin Plant '*Costuspictus*'

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ABSTRACT

Costuspictus (spiral ginger) commonly known as 'Insulin plant' was introduced from Mexico to India (kerala) very recently. The literature survey reveals that the fresh raw leaves should be consumed for anti-diabetic. Despite the preliminary studies the detailed phytochemical investigation has not been reported so far. Therefore, the rhizome extracts of costus pictus in different organic solvents (acetone, ethyl alcohol, chloroform, n-hexane and water). The five extract yielded by cold maceration procedure with the solvent Acetone, n-Hexane, Ethyl alcohol, Chloroform and Water. The maximum extract yield was obtain in ethyl alcohol cold extract which was preferably used for further to phytochemical studies, even though there was negligible difference in the presence of chemical constituents in the extract of samples. The ethanol extract of rhizome has shown maximum number concentration of secondary metabolites. Further phytochemical testing, The phytochemical Investigation reports revels that Insulin Plant '*Costuspictus*' The preliminary phytochemical tests indicated the presence of steroids, alkaloids, glycoids, flavonoids, saponin, tannins, in the extracts of rhizome of C. Pictus. The test which gave a positive results are mentioned in above phytochemical tests. The further work will be carried out for investigation of rhizome ethanol extract by thin layer chromatographic and Column chromatographic analysis, and spectroscopic analysis like GC-MS, ^{1}HNMR. Based on the above results it is evident that the leaves of C. pictus have ant diabetic effect and must be developing remarkable interest and potential in the new researcher for future studies on diabetes mellitus.

KEYWORDS

Costuspictus, Insulin plant.

1. INTRODUCTION

Diabetes mellitus is a known metabolic disorder of varied etiology characterized by chronic hyperglycemia (4) due to relative deficiency of insulin or its resistance. Diabetes is associated with disturbances of carbohydrate, fat and protein metabolism [5]. Since oral hypoglycemic agents cause side effects, there is a growing interest in herbal remedies [11] for the treatment of diabetes mellitus. Many plant preparations are used in folk medicine to manage diabetes [10] mellitus. New oral hypoglycemic compounds from medicinal plants may provide a useful source for development of pharmaceutical entities or as a dietary adjunct to existing therapies. Herbal drugs are considered to be less toxic and more free from side-effects compared to synthetic drugs.

Insulin plant (costus pictus) a folk medicine used for the treatment of diabetes mellitus is subjected to phytochemical analysis. This plant belongs to Costaceae family, which has been separated from Zingiberaceae on the basis of the presence of spirally arranged leaves and rhizomes being free from aromatic essential oils. The plant commonly known as Spiral ginger, which is originated in Mexico and is found to have antidiabetic properties. The plant preparations are used in folk medicine to manage diabetes mellitus [8, 9]. Herbal drugs [11] are also considered to be less toxic and freer from side effects compared to synthetic drugs. The ethnobotanical information reports that about 800 plants possess antidiabetic [13] potential. Plants have always been an exemplary source of drugs and many of the currently available drugs were derived directly or indirectly from them with this background, the present study was undertaken to examine the antidiabetic activity [6,7] of costus pictus plants from the freshly taken rhizomes. Stems are also intake as food by boiling or removing its outer skin. Decoctions of stem are used in fever and dysentery young stem juice is internally for eye and ear infection. More than 100 species of genus are distributed in the tropics all over the world. It is perennial, upright, spreading plant reaching about two feet tall, with the tallest stems falling over and lying on the ground. Leaves are simple; alternate, entire, evergreen, 4-8 inches in length with parallel venation. The large, smooth, dark green leaves of this tropical evergreen have light purple undersides and are spirally arranged around stems, forming attractive, arching lumps arising from underground rootstocks. With this background, the present study was undertaken to examine the anti-diabetic activity of the fresh leaves of C. pictus in streptozotocin-induced diabetic rats. C. pictus D. Don commonly known as ‘spiral ginger’ ‘step ladder’ or ‘insulin plant’ is a member of Zingiberacea family and is a newly introduced plant in India; originated probably in Mexico. In India it is grown in gardens especially in the state of Kerala where the fresh raw leaves are eaten by diabetic people. It is used as a munching supplementary food for the treatment of diabetes.

2. MATERIALS AND METHODS

2.1 Preparation of samples

The portion of fresh costus pictus plant (rhizome) was collected from local area punawale, district-Pune and wash with water for 2 to 3 times. Then separately harvested, chopped and dried at room temperature for 10 days. Around 100grams of rhizomes powder were collected for further analysis. The rhizome extracts of costus pictus in different organic solvents (acetone, ethyl alcohol, chloroform, n-hexane and water)

Figure-1. Dry powder of Costus pictus rhizomes.

2.2. Extraction

The rhizome extract of costus pictus is carried out by two methods.

2.2.1. Cold maceration

This is conventional extraction technique in which solvent recovery is not possible and it is time consuming process but important is that there is no decomposition of the chemical constituents, as it was observed in the hot continuous extraction process and hence for this extraction experiment we choose cold maceration technique to avoid decomposition of any compound.

2.2.2. Hot continuous extraction

In this technique the desired compounds are continuously extracted, this extract soxhlet apparatus is used. This method is best technique for extraction as it reduces time for extraction but large number of compounds are decomposed and hence not used in present study.

In the cold maceration extraction methods we have been used 100gms of air dried powder material of rhizome was separately macerated with different organic solvent (ethyl acetate, ethanol, water, acetone etc.).Successively for 48 hrs with occasional stirred and it was then filtered. The filtrate was evaporated to dryness using a rotary evaporator at 400^0C under reduced pressure to avoid decomposition of the chemical constituents.

3. RESULTS AND DISCUSSION

3.1. Cold maceration extraction

The filtrate obtained by was evaporated to dryness using a rotary evaporator at 400C under reduced pressure to avoid decomposition of the chemical constituents. The residue of different solvent were weighed in grams and is shown in Table 1.

Table 1. Solvent used for extraction and residue weight in grams.

Sr.No.	Solvent used for extraction	Weight of Residue in g rams
1	The weight of residue obtained by Solvent ethyl acetate	0.30
2	The weight of residue obtained by ethanol	2.13

3	The weight of residue obtained by acetone	0.76
4	weight of residue obtained by Chloroform	0.46
5	The weight of residue obtained by water	0.10

The crude ethanol extract was later subjected to photochemical tests and further analysis.

Figure 2. Crude extract of ethanol

3.2. Phytochemical test

The includes Detection of Alkaloids, Detection of Saponins, Detection of Tannins and Detection of flavonoids were carried out by standard methods rhizome extract of costus pictus in different organic solvents (acetone, ethyl alcohol, chloroform, hexane and water). Shows positive phytochemical test. The crude ethanol extract was yielded in bulk residue and was subjected to photochemical tests and further analysis and investigation study shows that this ethanol extract shows good positive phytochemical tests.

3.2.1 Detection of Alkaloids

1. Mayer's test -To a few ml of extract, a drop of Mayer's reagent was added by the side of the test tube. A white or creamy precipitate observed, which indicates the test as positive.

Figure 3. Mayer's test

2. Wagner's test -To a few ml of extract, few drops of Wagner's reagent were added by the side of the test tube. A reddish brown precipitate confirms that the Wagner's test is positive.

Figure 4. Wagner's test

3. Hager's test-To a few ml of extract, 1 or 2 ml of Hager's reagent was added. A prominent yellow precipitate indicates the Hager's test as positive.

Figure 5. Hager's test

3.2.3. Detection of Saponins

1. Foam test –The extract was dissolved in 20 ml. of distilled water. The suspension was shaken in a graduated cylinder for 15 minutes. A two cm. layer of foam indicates the presence of Saponins formed at the as a foam.

Figure 6. Foam test

3.2.4. Detection of Tannins

Ferric chloride test -The extract was dissolved in 5 ml of distilled water. To this few drops of 5% Ferric chloride were added. A dark green color indicates the presence of tannins.

Figure 7. Ferric chloride test

3.2.5 Detection of flavonoids

Magnesium and hydrochloric acid reduction test

The extract was dissolved in 5 ml of alcohol and few fragments of magnesium ribbon and concentrated hydrochloric acid drop wise were added. It can develop pink Crimson color, which clearly indicate the presence of flavonoids

Figure 8. Detection of flavonoids

Table 2. Preliminary phytochemical tests of ethanol extract of rhizomes costus pictus obtained by cold maceration.

Extraction solvent	**Plant material**	**Phytochemicals**									
		Steroid	Saponin	Glycosides	Phenols	Alkaloids	Quinones	Coumarins	Furanoids	Flavonoids	Tannins

t											
Ethanol	Rhizome	+	++	++	-	++	-	-	-	++	-

4. CONCLUSION

The extraction and photochemical tests result are observed as indicated below;

4.1. Extraction of plant material of rhizomes costus pictus

The five extract yielded by cold maceration procedure .The solvent use to was Acetone, n-Hexane, Ethyl alcohol, Chloroform and Water. The weight of residue obtained by Solvent ethyl acetate was 0.30 Gms, The weight of residue obtained by ethanol was found to be 2.13 Gms, The weight of residue obtained by acetone was 0.76 Gms, while weight of residue obtained by Chloroform 0.46 Gms and the weight of residue obtained by water 0.10 Gms. The maximum extract yield was obtained in ethyl alcohol cold extract which was preferably used for phytochemical analysis.

4.2. Phytochemical tests

Preliminary phytochemical tests of ethanol extract of rhizomes costus pictus obtained by cold maceration. The preliminary phytochemical tests indicated the presence of steroids, alkaloids, glycosides, flavonoids, saponin, tannins, in the extracts of rhizome of C. Pictus. The test which gave positive results are mentioned in above phytochemical tests. The further work will be carried out for investigation of rhizome ethanol extract by thin layer chromatographic and Column chromatographic analysis, and spectroscopic analysis (12) like GC-MS, 1HNMR9 (5). Based on the above results it is evident that the leaves of C. pictus have ant diabetic effect and must be develop remarkable interest and potential in the new researcher for future studies on diabetes mellitus

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