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Invitro Determination of Flavonoids and Phenolic Compounds from Corm of Amorphophallus Campanulatus (Roxb.)

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Abstract:-

The flavonoids and phenols are very significant phytoconstituents having ability to play important role in control and prevention of tissues damage by activated by oxygen species.

Amorphophallus campanulatus (Roxb.) was analysed for flavonidal compounds (FC) by usining absorbance of different concentration (20 to 100 µg/ml) in term of rutin and total phenolic compounds (TPC) was calculated in form of catechol equivalent. TLC study methanolic and petroleum ether corm extracts were conducted for quantification of flavonoids in mg per gram of Rutin equivalent determined from test extracts. The results indicated that, methanol exctracts of A. campanulatus (Roxb.) contain 88 mg/g of Rutin equivalent while, minimum amount of flavonoids was found in petroleum ether extract.

Similarly, to calculate quantification of phenolic compounds from test extracts by using the absorbance of different concentration 0.5 to 2.5 μ g/ml of Catechol as phenolic compound obtain from corm extracts of methanolic was 2.0 mg/g of Catechol equivalent. While, petroleum ether extract found to be 1.0 mg/ml.

Introduction:-

Today attention has been focused on such species which have reactive oxygens and free radicals are which play vital role in the genesis of varied diseases which are imflammation, cataract, liver cirrliosis, reperfusion injury, cancer and others (Halliwel et.al., 1994).

The herbal drugs have such phytoconstituents are the gaining importance in the prevention and treatment of various organ toxicities due to xenobiotic challenges or environmental challenges.

A. campanulatus (RoxB.) from family Araceae a tuberous, stout indigenous herb commonly known as elephant foot yam, suran, grown as vegetable is widely available (Ghani A.; 1998) and is reported to contain flavonoids (Shilpi J.A., et.al., 2005). In Ayurvedic System of Medicine, tubers of A. campanulatus (RoxB.) has been indicated in treating various above mentioned patho-physiological conditions due to Rective oxygens species (ROS) (Narayana Das Prajapati, et.al., 2004; Sivaraj V.V., 1994; Kirtikar K.R. et.al., 1987; Nadkarni K. M. et.al., 2000; Guha Bakshi D.N. et.al., 1999; Pullilah T., 2000). Tubers are reported in management of haemmroids (Vastrad C.S., et.al) to have antiprotease activity (Pratibha S., et.al., 1995), antimicrobial activity (Alam Khan. et.al., 2008) and analgesic activity of its methanolic extract (Shilpi J. A. et.al., 2005).

This exhaustive literature survey revealed that the tubers (corm) are not yet screened for its quantitative evaluation of Flavonoids and Total Phenolic contents of the extracts of the corm. Hence in the present study an attempt is made to standardize the corm of *A. campanulatus* (*RoxB.*) in terms of its Flavonoidal content and Total Phenolic content.

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Materials and method:-

Tests for flavonoids

The flavonoids are structurally derived from the parent substance called flavone. The flavonoids, which occur in free form or bound to sugars, are called as glycosides. For this reason, when analyzing flavonoids, it was usually better to examine the flavonoids in hydrolyzed plant extracts.

Preparation of test solution:

Small amount of extract with equal volume of 2M hydrochloric acid was added and heated the test tube for 30-40 min at 100^oC, allowed to cool, filtered and extracted with ethyl acetate. The ethyl acetate extract was concentrated to dryness, followed the test for flavonoids to ethyl acetate fraction by dissolving the residues with ethyl acetate.

Shinoda test:

Test solution with few fragments of magnesium ribbon and concentrated. hydrochloric acid showed pink to magenta red colour.

Determination of flavonoids from corms extracts of A. companulatus (Roxb.)

Rutin was used as standard flavonoid. Different concentrations (20 to 100 μ g/ml) of rutin were analyzed at 510 nm and a calibration curve was plotted as absorbance versus concentration. 10 μ g/ml of each test substance (corm extracts of *A. companulatus (Roxb.)*) were analyzed using the similar procedure and quantity of flavonoids in mg per gram of rutin equivalent was determined for each extract.

Procedure:

- Known volume of samples was pitted out in series of test tubes and volume was made up to 0.5 ml with distilled water.
- Sodium nitrite (5%; 0.03 ml) was added to each tube and incubated for 5 minutes at room temperature.
- Aluminum chloride solution (10%; 0.6 ml) was added and incubated for 5 minutes at room temperature.
- Sodium hydroxide solution (1 M; 0.2 ml) was added and total volume was made up to 1 ml with distilled water.
- Absorbance was measured at 510 nm against a reagent blank.
- Standard curve using different concentrations of rutin was prepared.
- From the standard curve, concentration of flavonoids in the test samples was determined and expressed as µg of rutin equivalent.

Determination of Phenolic compounds from corm extracts of A. companulatus (Roxb).

Catechol was used as standard phenolic compound. Different concentrations (0.5 to 2.5 μ g/ml) of catechol were analyzed at 650 nm and a calibration curve was plotted as absorbance versus concentration. 1 μ g/ml of each test substance (corm extracts of *A. companulatus (Roxb.)*) was analyzed by using the similar procedure and quantity of phenolic compounds in mg per gram of catechol equivalent was determined for each extract.

Procedure:

• Aliquot of each sample was pipette out in series of test tubes and volume was made up to 3 ml with distilled water.

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- Folin-Ciocalteu Reagent (0.5 ml) was added to each tube and incubated for 3 minutes at room temperature.
- Sodium carbonate (20%; 2 ml) solution was added, mixed thoroughly and the tubes were incubated for 1 minute in boiling water bath.
- Absorbance was measured at 650 nm against a reagent blank.
- Standard curve using different concentrations of standard phenolic a ctechol was prepared.
- From the standard curve, concentration of phenols in the test samples was determined and expressed as µg of catechol equivalent.

Results and Discussion:-

Determination of flavonoids

In order to investigate quantity of flavonoids by using absorbance of different concentrations (20 to 100 μ g/ml) of Rutin and the results are summarized in the table- 1. A calibration curve shows linear correlation at measured concentrations. Table- 2 represents the quantity of flavonoids in mg per gram of Rutin equivalent determined for test extracts. The results indicate that methanol extracts of *A. campanulatus (Roxb.)* contain maximum amounts of flavonoids as 88 mg/g of Rutin equivalents respectively while minimum amount of flavonoids in presence of petroleum ether extract.

Determination of phenolic compounds

Determination of quantum of phenolic compounds by using the absorbance of different concentrations (0.5 to 2.5 µg/ml) of Catechol and the results are given in the table- 3. A calibration curve shows linear correlation at measured concentrations. Table- 4 represents the quantity of phenolic compounds in mg per gram of Catechol equivalent determined from each extract. The results indicate that, methanol extract of *A. campanulatus* (*Roxb.*) contain maximum amounts of phenolic compounds as 2.0 mg/g of catechol equivalents respectively. While, other tested extract was found to be minimum.

References:-

- 1. Alam Khan, Moizor Rahman, Islam M.S., Indian J Pharmacol, 2008, 40(1), 41.
- 2. Ghani A., Medicinal plants of Bangladesh, Dhaka, Bangladesh, Asiatic Society of Bangladesh, 1998, 77.
- 3. Guha Bakshi D. N., Sensarma P., Pal D. C., A Lexicon off Medicinal Plants of India, Naya Prakash, Calcutta, India, 1999, 127.
- 4. Halliwell H., Lancet, 1994,344, 721.
- 5. Kirtikar K. R. and Basu B. D., Indian Medicinal Plants, Interantional Book Distributors, Dehradun, 1987, IV, 2609.
- 6. Nadkarni K. M. and Nadkarni A. K., Indian Material Medica, Popular Prakashan Pvt. Ltd., Mumbai, 2000, 94.
- 7. Narayana Das Prajapati, Purohit, S.S., Arun K. Sharm, Tarun Kumar, A Hand Book of Medicinal Plants A complete source book, Agrobios (India), Jodhpur, 2004, 41.
- 8. Pratibha S., Nambison B., Leelamma S., Plant Food Alum Nutr., 1995, 40(1), 247.
- 9. Pullaiah T., Medicinal Plants of India, Regency publication, New Delhi, I, 2000, 49.
- 10. Shilpi J.A., Ray R. K., Sarder S. J., Vddin S. J., Fitotheropia, 2005, 76 (3-4), 367.
- 11. Shivraj V. V., Indira Balachandran, Ayurvedic Drugs and Their plant sources, Oxford and IBH publishing Co. Pvt. Ltd., New Delhi, Bombay and Calcutta, 1994, 457.
- 12. The Wealth of India Raw Material, CSIR, New Delhi, 2003, 1: A, 233.
- 13. Vastrad C.S., Pakkanavar R.V., Antiseptic, 99(9), 343.

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Figures and Tables

Table 1: Absorbance table for different concentrations of rutin

Sl. No.	Concentration of Rutin (in µg)	Optical Density (at 510nm)
1	Blank	0
2	20	0.19
3	40	0.35
4	60	0.56
5	80	0.74
6	100	0.91

Graph 1: Calibration Curve for Rutin

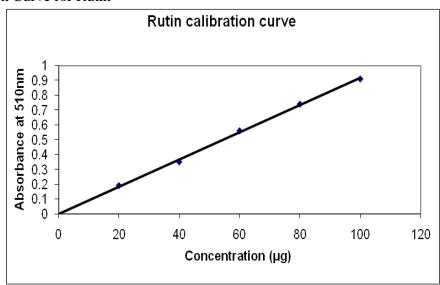


Table 2: Quantity of Flavonoids (Rutin equivalents) found in each extract

Sl. No.	Test Sample	mg/g of Rutin equivalents
1.	Methanolic extract of A. campanulatus (Roxb.)	88 mg/g
2.	Petroleum ether extract of A. campanulatus (Roxb.)	16 mg/g

 Table 3: Absorbance table for different concentrations of Catechol

Sl. No.	Concentration of Catechol (in µg)	Optical density (at 650nm)
1	Blank	0
2	0.5	0.175
3	1	0.325
4	1.5	0.48
5	2	0.64
6	2.5	0.81

Graph 2: Calibration curve for Catechol

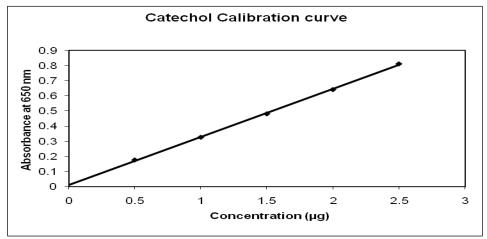
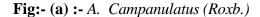


Table 4: Quantity of phenolic compounds (Catechol equivalents) found in each extracts

Sl. No.	Test sample	mg/g of Catechol equivalents
1.	Methanolic extract of A. Campanulatus (Roxb.)	2.0 mg/g
2.	Petroleum ether extract of A. campanulatus (Roxb.)	1.0 mg/g







(b) :- Corm of A. Campanulatus (Roxb.)