ejpmr, 2022,9(8), 145-148

SJIF Impact Factor 6.222

EUROPEAN JOURNAL OF PHARMACEUTICAL AND MEDICAL RESEARCH

www.ejpmr.com

Review Article

ISSN 2394-3211

EJPMR

EVALUATION OF ANTI – CANCER ACTIVITY OF METHANOLIC AND AQUEOUS EXTRACTS OF *ABUTILON INDICUM* LINN BY IN- VITRO METHOD

[1]Tarique Hussain, *[2]Ravi Kumar Vishwakarma, [3]Asha Rani, [4]Randhir Kumar Gupta and [5]Deepak Kumar

[1]Department of Pharmacology, Glocal University, New Delhi Yamunotri Marg, State Highway-57, Mirzapur Pole, Dist-Saharanpur, UP-247121, India.

[2]Department of Pharmacology,Mangalayatan University, NCR 33rd Milestone, Aligarh-Mathura Highway, Beswan, Aligarh, UP-202145, India.

[3]Government Pharmacy Institute, Bariatu, Ranchi, Jharkhand-834001, India.

[4,5]Ranchi College Pharmacy, Kutetoli, Chandaghasi, Ranchi, Jharkhand-834003, India.

***Corresponding Author: Ravi Kumar Vishwakarma**

Department of Pharmacology,Mangalayatan University, NCR 33rd Milestone, Aligarh-Mathura Highway, Beswan, Aligarh, UP-202145, India.

Article Received on 30/05/2022 **Article Revised on 20/06/2022** **Article Accepted on 10/07/2022**

ABSTRACT

Abutilon indicum belonging to family Malvaceae is distributed throughout number of tropical and subtropical areas and subtropical zones and has been used for various disorders in traditional and folk medicine. Medicinal herbs play an important role in primary health care system among rural population since synthetic anti-cancer remedies are beyond the reach of common man because of the cost factor. The herbal medicines have a vital role in the prevention and treatment of cancer which execute their therapeutic effect by inhibiting cancer activating enzymes and hormones, stimulating DNA repair mechanism. Methodology: Extract were tested for the presence of active principle agents such as Triterpenoids,Steroids,Glycosides,Saponins, Alkaloid, Flavonoids, Tannins, Proteins, Free Amino Acid, Carbohydrate and lignin. Results: The result of the study shows Methanolic and Aqueous Extract possessed an Anti-neoplastic property comparing these two extracts Methanolic extract is more potent than Aqueous extract.) Were evaluated against six different cell lines (Lung A549, NCI-H23, Colon -205, SW- 620, Liver- HEP- 2, Ovary –OVCAR—5, Breast-mcf- 7, Prostate – DU – 145). Comparing Methanolic and Aqueous Methanolic extract exhibited 95% of the Anti-neoplastic potential against the Human Cancer cell origins.

KEYWORDS: *Abutilon indicum*, Anti-Cancer, Flavonoid's, Malvaceae.

INTRODUCTION

India has a rich Culture of Medicinal herbs and spices, which includes about more than 2000 species and has a vast geographical area with high potential abilities for Ayurvedic, Unani, Siddha traditional medicines but only few have been studied chemically and Pharmacologically for their potential medicinal value. Herbal molecules are safe and could overcome the resistance produced by the pathogens as they exist in a combined form or in a pooled form of more than one molecule in the protoplasm of the plant cell. *Abutilon indicum* is a small shrub in the Malvaceae family, native to tropic and subtropical regions and sometimes cultivated as an ornamental. Plant remedies and their preparation as therapeutic treatment, Natural products chemistry has brought forth compound lead to advanced cancer treatment. Cancer disease kills yearly about 3500 per millions populations around the worldwide. Naturally occurring chemical constituents from medicinal plants are known to be chemically balanced, effective, and least injurious with none reducedside effects as compared to Synthetic compound. This plant is often used as a medicinal plant and is considered invasive on certain tropical islands. It is known as "Atibala" in Hindi and found in the outer Himalayan tracts from Jammu to Bhutan up to an altitude of 1500m and extending through the whole of northern and central India.

PHYTOCHEMISTRY: *Abutilon indicum* has been explored phytochemically by various researchers and found to possess number of chemical constituents.

WHOLE PLANT: The whole plant contains mucilaginous substances and asparagines. saponins, flavonoids, alkaloids, hexoses, n-alkane mixtures (C22-34), alkanol as main classes of compounds. Some important constituents reported in the plant are β-sitosterol, vanillic acid, p-coumaric acid, caffeic acid, fumaric acid, Abutilon A,(R)-N-(1'-methoxycarbonyl-2'phenylethyl)-4-hydroxybenzamide,hydroxybenzoic, galacturonic, p Dglycosyloxybenzoic and amino acids.The plant *Abutilon indicum* contains of essential oil which mainly consists of α-pinene -caryophyllene, caryophyllene oxide, endesmol, farnesol, borenol, geraniol, geranyl acetate, elemene and α-cineole.[4]

METHODOLOGY

Processing of Plant Material: The leaf extracts of the plant *Abutilon indicum* under study were obtained using the Soxhlet Extraction Method. A Soxhlet Extractor was used for this purpose. A Soxhlet extractor is a piece of laboratory apparatus invented in 1879 by Franz von Soxhlet. The Soxhlet extraction of leaves were carried out by taking 50 grams of whole plant leaves of *Abutilon indicum*, washed and dried were taken and placed in a thimble made up from thick filter paper, which was loaded into the main chamber of the Soxhlet extractor. The Soxhlet extractor was placed onto a round bottom flask containing the extraction solvent i.e. distilled water. The Soxhlet extractor was then equipped with a condenser. The solvent was heated at 90^{O} C to reflux. As the solvent vapour travelled up a distillation arm into a condenser, the condensed vapours dripped back down into the chamber housing and the material solidified. The chamber containing the solid material slowly filled up with warm solvent. When the Soxhlet chamber was almost full, the chamber was automatically emptied by a siphon side arm, with the solvent running back down to the distillation flask. This cycle was allowed to repeat 18 times. After extraction the solvent was removed by means of a rotary evaporator. The extracted compound was collected. The non-soluble portion of the extracted solid in the thimble was discarded. The aqueous extract was then used for further investigation. Similarly, methanolic extract was also obtained by using Soxhlet extraction method.

PHYTOCHEMICAL SCREENING OF AQUEOUS AND METHANOLIC EXTRACTS OF *ABUTILON INDICUM*

Phytochemical Analysis

Extracts were tested for the presence of active principle such as Triterpenoids, Steroids, Glycosides, Saponins, Alkaloid, Flavonoids, Tannins, Proteins, Free Amino Acid, Carbohydrate and lignin. Following standard procedures were used.

S.No	Test For	Test	Result
1	Carbohydrates	Benedict's Test	+
2	Tannins	Gelatine Test	+
3	Alkaloids	Hager's Test	+
4	Saponins	Foam Test	+
5	Flavonoids	Ferric chloride Test	+
		Alkaline reagent Test	+
		Lead acetate solution Test	+
6	Proteins	Biuret Test	+
7	Glycosides	Keller Killani Test	+
8	Free Amino Acids	Ninhydrin Test	+
9	Steroid and Terpenoids	Liebermann Burchard Test	+

PREPARATION

- Firstly Dilute the Dye Wash Solution by adding 1-part 10X Dye Wash Solution to 9 parts distilled water. You require ~0.8ml per well to sufficiently wash the wells.
- In a plastic container or clean amber glass, add 100ml 1X Dye Wash Solution. Remove 1ml 1X Dye Wash Solution and add to the SRB Dye vial, pipette up and down to resuspend and transfer the entire contents to the 100ml 1X Dye Wash Solution. Stir to mix. The SRB Dye Solution can be stored at room temperature protected from light. Crystals may form during storage, remove by filtering with a syringe filter prior to use in the assay.

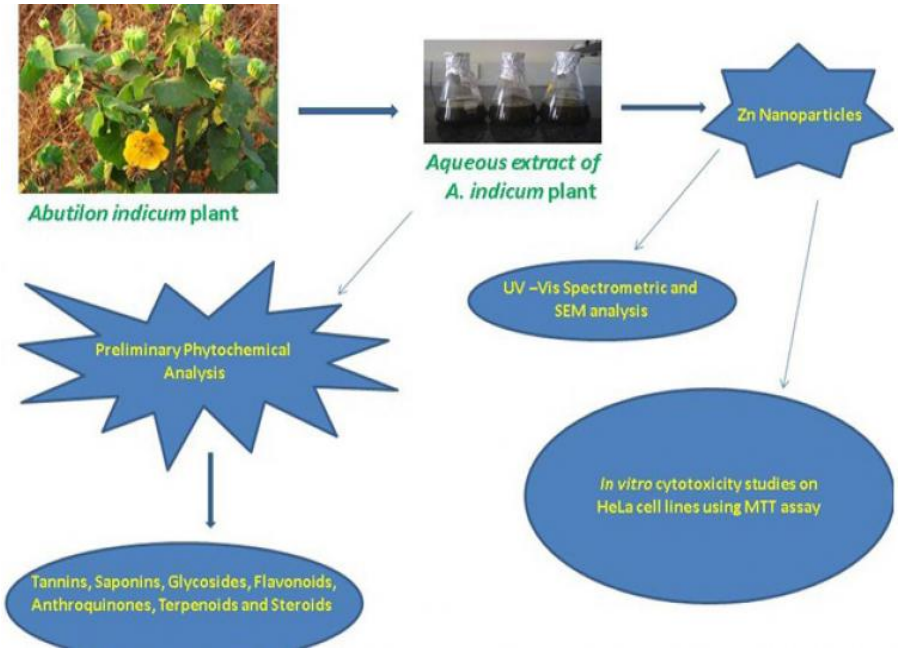
SRB ASSAY PROCEDURE

The monolayer cell culture was trypsinized and the cell count was adjusted to 0.5-1.0 x 105 cells/ml using medium containing 10% new born sheep serum. To each well of the 96 well microtiter plate, 0.1ml of the diluted cell suspension (approximately 10,000 cells) was added. After 24 hours, when a partial monolayer was formed, the supernatant was flicked off, washed once and 100 µl of different test compound concentrations were added to the cells in microtiter plates. The plates were then incubated at 37^{o}C for 72 hours in 5% CO_2 incubator, microscopic examination was carried out, and observations recorded every 24 hours. After 72 hours, 25 µl of 50% trichloro acetic acid was added to the wells gently such that it forms a thin layer over the test compounds to form overall concentration 10%. The plates were incubated at 4^{o}C for one hour. The plates were flicked and washed five times with tap water to remove traces of medium, sample and serum, and were then air-dried. The air-dried plates were stained with 100µl SRB and kept for 30 minutes at room temperature. The unbound dye was removed by rapidly washing four times with 1% acetic acid. The plates were then air dried.

100 µl of 10mM Tris base was then added to the wells to solubilize the dye. The plates were shaken vigorously for 5 minutes. The absorbance was measured using microplate reader at a wavelength of 540 nm. The percentage growth inhibition was calculated using following formula; the percentage growth inhibition was calculated using following formula.

%cell inhibition= 100-{(Ac-Ab)/(At-Ab)} x100

Where, At= Absorbance value of test compound, Ab = Absorbance value of blank, Ac=Absorbance value of control.



RESULT AND DISCUSSION

The result of the study shows Methanolic and Aqueous Extract possessed Anti-Cancer properties comparing these to extract Methanolic extract is more potent than Aqueous extract.The result of the study shows Methanolic and Aqueous Extract possessed Anti-cancer properties comparing these to extract Methanolic extract is more potent than aqueous extract.) were evaluated against six different cell lines (Lung A549,NCI-H23, Colon -205,SW- 620, Liver- HEP- 2, Ovary –OVCAR—5, Breast-mcf- 7, Prostate – DU – 145). Comparing Methanolic and Aqueous extracts the Methanolic extract exhibited 95% of the Anti-Cancer potential against the Human Cancer cell origins. The growth inhibition of 70% was considered active while testing extracts, but in testing of active ingredients at different molar concentrations, the growth inhibition of 50% or above was the criteria of activity. These findings therefore justify the use of *Abutilon Indicum* medicinal plants in the treatment of several ailments in india.

Table 1: Preliminary Phytochemical Screening of Aqueous and Methanolic Extract of Whole Plant of *Abutilon Indicum*.

Test	Drug coarse powder	Aqueous	Methanol
Tannins	+	-	+
Terpenoids	-	-	-
Saponins	+	+	+
Volatile oil	+	+	+
Alkaloids	+	+	+
Flavonoid’s	+	+	+
Glycosides	+	+	+
Mucilage’s	+	-	+
Sterols	+	-	-
Carbohydrates	+	+	+
Proteins	+	+	+

The preliminary phytochemical screening of *Abution indicum* whole plant extracts showed presence of sterols, Carbohydrates, Proteins, Flavonoid's, Glycosides, Mucilage's, Tannins, Saponins of Volatile oil respectively in different extracts.

Table 2: Anti Cancer Activity of *Abutilon Indicum* In Different Cell Lines.

Extract	Concentration	Colon		Breast		Lung		Liver HEP2	Prostate DU 145	Ovary-OVCAR5
		colo-205	Sw 620	mcf-7	2R-75-1	Ncl-H23	A-549			
Growth Inhibition										
Methanol	100	20	45	88	88	72	0	72	80	0
Aqueous	100	0	0	64	0	63	0	0	0	0
Positive controls	Concentration (molar)	-	-	-	-	-	-	-	-	-
Paclitaxel	1×10^{-6}M	-	-	70	-	71	60	-	66	62
5-fluro – uracil	2×10^{-5}M	-	-	-	60	-	-	-	-	-
Mitomycin	1×10^{-6}M	-	-	-	-	-	-	73	-	-
Adriamycin	1×10^{-6}M	82	83	-	-	-	-	-	-	-

CONCLUSION

Natural products discovered from medicinal plants have played an important role in the treatment of Cancer or Tumour. The present study points to the potential anti-cancer activity of Aqueous and Methanol extract of *Abutilon Indicum.* Further studies to characterize the active principles and elucidate the mechanism of the action of Aqueous a Methanol extract are in Progress. Hence the plant extract may have clinical and therapeutic proposition in the most life threaten disease like cancer and further studies are required to investigate the plant sample as antineoplastic agent. Therefore, it is anticipated that plant can provide potential bioactive compounds for the development of new 'leads' to combat cancer diseases.

REFERENECES

1. Bagi M.K. Kalyani G.A., Dennis T.J. Kumar AK, Kakrani HK.A Preliminary Pharmacology Screening of *Abutilon Indicum.*
2. Kirtiar K.R. Basu B.D., Indian Medicinal Plants, Edn 2, Vol. I, Dehradun – 1994, 314–317.
3. Pacific Island Ecosystems at Risk Retrieved, 2008 – 06 – 18.
4. Riss TL, Moraves, R.A., Moravee Assay Drug Dev. Technol, Feb, 2004; 2(1): 51–52.
5. Breast Cancer Treatment, NCL 23 May 2014, Retrieved, 29, June 2014. 21.
6. Breast Cancer NCL, Retrieved 29, June 2014.
7. Ruddon, Raymond (2007), Cancer Biology, Oxford University Press, 223.
8. J. Raamachandran, "Herbs of Siddha Medicines – The First 3D Book on Herbs, P4.
9. M.J. Thun J.O. Delancey M.M. Center, A Journal and E.M. Ward, "The Global burden of Cancer, Priorities for Prevention, "Carcinogenesis, 2009; 31(1): 100–110.
10. R. Parideens, L. Biganzoli. P. Burning, Journal of Clinical Oncology, 2000; 18(4): 724.
11. Shipi Arora, Devendra Pratap Singh, Ankit Saini, Ashish Kumar Pharmacognostic investigation on roots and leaves extract of *abutilon Linn* Int. J. Res. Dev Pharm. Life Sci., 2013; 2(5): 567-573.
12. M.S. Mohite, P.A. Shelar, V.N Raje, S.J. Babar, R.K. Sapkal Review on pharmacological properties of *Abutilon indicum* Asian J.Pharmaceut. Res., 2012; 24): 156-160.
13. Sidharth Tiwari, Saumya Mishra, Dharm Raj Misra, Richa Upadhyay Identification of new bioactive compound from fruit of Abutilon indicum through GCMS analysis Biol. Forum Int. J., 2016; 8(1): 548-554.
14. Pandey DK, Tripathy RD, Tripathy NN, Dixit SN Antifungal activity of some seed extracts with special reference to that of Pimpinella diversifolia DC. Indian journal of crude drug research, 1983; 21(4): 177-182.
15. Markham KR, Ternai B, Stanley R, Geiger H, Mabry TJ. ^{13}C NMR studies of flavonoids. III. Naturally occurring flavonoid glycosides and their acylated derivatives. Tetrahedron, 1978; 34: 1389-1397.