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## EVALUATION OF PHYTOCHEMICAL AND ANTIMITOTIC POTENTIAL OF *LAGENARIA SICERARIA* FRUIT USING ONION ROOT MODEL

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Antimitotic assay,  
Onion root model

### ABSTRACT

Cancer is one of the leading causes of mortality worldwide. Many of the cucurbitaceae plants traditionally reported to possess antitumor activity. The present study was carried out to evaluate the anticancer activity for different extracts of *Lagenaria siceraria* Fruit. The present research work had carried out on laboratory level assay to avoid the use of different animal models. Preliminary phytochemical tests of successive extraction of *Lagenaria siceraria* fruit powder had performed to find out the different chemical moieties as per standard procedures. Preliminary anticancer screening by exposure of different extracts on Onion Root model was carried out to find out the lead extract which showed the promising cell growth inhibitory activity. The antimitotic assay by onion root method was selected because this is easy to done and give fastest promising results. Onion was selected for the antimitotic assay which shows the root growth inhibition that compared with standard antimitotic drug (Colchicine 100PPM and 100 PPM). n-butanol extract of *Lagenaria siceraria* fruit powder shows most promising anticancer activity than other extracts tested and control used. Further isolation of active moiety from n-butanol extract for anticancer activity by chromatographic techniques is almost completed.

### Abbreviations, Unit etc

NH -	n-Hexane
CHL-	Chloroform
DM-	Dichloromethane
EA-	Ethyl Acetate
NB-	n-Butanol
METH-	Methanol
WAT-	Water
PPM-	Parts Per Millions
P-	Prophase
M-	Metaphase
A-	Anaphase
T-	Telophase

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**INTRODUCTION:** Cancer is one of the most fatal diseases in human population and one of the most frequent causes of death worldwide. To prevent the cancer, synthetic and natural sources are used in alone or combination.

Today due to resistance of different allopathic medicines natural source is preferred mainly to block the development of cancer in human. Plant shows different chemical moiety including flavonoids<sup>1-3</sup>,

terpenoids<sup>1-3</sup> and steroids<sup>1-3</sup> which have the Pharmacological properties like antiulcer<sup>4</sup>, antihyperlipidemic<sup>5-6</sup>, antioxidant<sup>7-8</sup>, cytotoxic as well.

*Lagenaria siceraria* Standley, commonly known as bottle-gourd, belongs to the Cucurbitaceae family. The plant is widely available throughout India. It is a climbing or trailing herb, with bottle- or dumb-bell shaped fruits. Both its aerial parts and fruits are commonly consumed as a vegetable.

Traditionally, it is used as medicine in India, China, European countries, Brazil, Hawaiian island, etc. for its cardiogenic, general tonic and diuretic properties. *Lagenaria siceraria* Standley fruit has different biological activities, as traditional medicinal plants, such as antihyperlipidemic, antidiabetic, antiulcer and prominently antioxidant activity.

So, the present communication deals with successive extraction of *Lagenaria siceraria* Standley fruit for anticancer activity. This activity was screened by different laboratory based models. The Antimitotic assay was selected because this is easy to done and give fastest promising results.

**MATERIALS AND METHODS:** The ripened fruits of the plant *Lagenaria siceraria* Standley were collected from the local area of Vaduj, District Satara, Maharashtra, India. The plant was identified by botanist, Dr. S. R. Yadav Shivaji University Kolhapur, Maharashtra. Voucher specimen (No. 1 Sarang Sunil Mahamuni) was deposited in Dept. of Pharmacognosy, Bharati Vidyapeeth College of Pharmacy, Near Chitranagari, Kolhapur- 416013, Maharashtra.

**Reagents and chemicals:** n-Hexane, Chloroform, Dichloromethane, Ethyl acetate, n-Butanol, Methanol, Distilled water<sup>10</sup> preliminary phytochemical reagents, Colchicine, 0.1 N HCL and acetocarmine stain.

**Equipments and apparatus:** Soxhlet apparatus (1kg cap.), Mettler analytical balance, Rotamentle (J-SIL), Rotary film evaporator (Evator). All experiment performed in year 2011-12 at Bharati Vidyapeeth College of Pharmacy, Near Chitranagari, Kolhapur-416013, Maharashtra.

**Preparation of Extracts:** Dry powder (250g) was used for carrying out successive Soxhlet extraction with 2 liter each of n-Hexane, Chloroform, Dichloromethane, Ethyl acetate, n-Butanol, Methanol and Chloroform-water (IP) for 72h at room temperature by maceration. Each time after completion of cycle the extract was removed and powder was dried at 45°C for 24h. All the extracts were filtered and filtrates were evaporated using Rotary film evaporator and dried in vacuum drier. Extractive values are mention in **table 1**.

**Phytochemical Screening:** All the extracts obtained were subjected for phytochemical screening using standard procedure<sup>11</sup>. The dried extracts (few mg) were dissolved in sufficient amount of respective solvents and tested for various constituents. The results of the tests are mentioned in **table 2**.

**Antimitotic Assay:** The experiment was performed in three groups as per the standard procedures<sup>12-14</sup>; Test group (7 in No. of Extracts), Control group (7 in No. of Respective Solvents), Standard (colchicine) and each had two concentrations 100 PPM and 1000 PPM. All extracts dissolved in respective solvents and made desired dilution, 0.5 ml sample was added in respective labeled dram vials and solvents were evaporated. 12ml of tap water was added to each vial. Onion bulbs were placed on each vial. All vials were placed in dark condition.

The growth of roots was observed (**Fig. 1**) and sequentially added tap water if it gets absorbed by onion itself every day. Root length was measured after 2-3 days and also observed the mitotic phases were counted in 40x objective of Motic Electron microscope and measured mitotic index of each group. The experiment was repeated in triplicate. Results are mentioned in **table 3**.

The mitotic index was calculated using the following formula:

$$\text{Mitotic index} = \frac{P + M + A + T}{\text{Total cells}}$$

**RESULTS AND DISCUSSION:** The present study thus explores the potent antimitotic activity which may be either because of a direct cytotoxic effect of the extract on normal phytocells or restriction of cell division in normal cell cycle.

Fruit shows different chemical moieties mostly steroids, triterpenes, alkaloids and glycosides. For Antimitotic assay, n-Butanol extract shows 100% and 61.53% activity as compared to the given antimitotic drug (Colchicine 100PPM and 100 PPM) respectively.

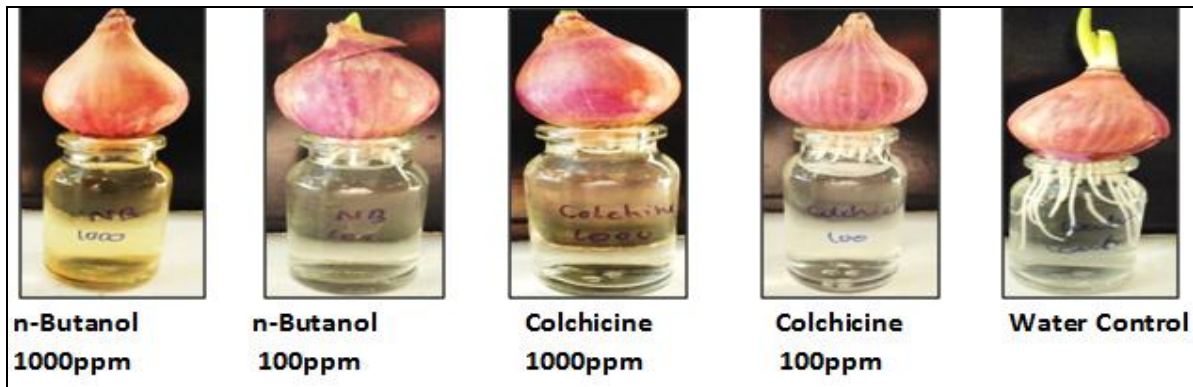


FIGURE 1: ONION ROOT MODEL

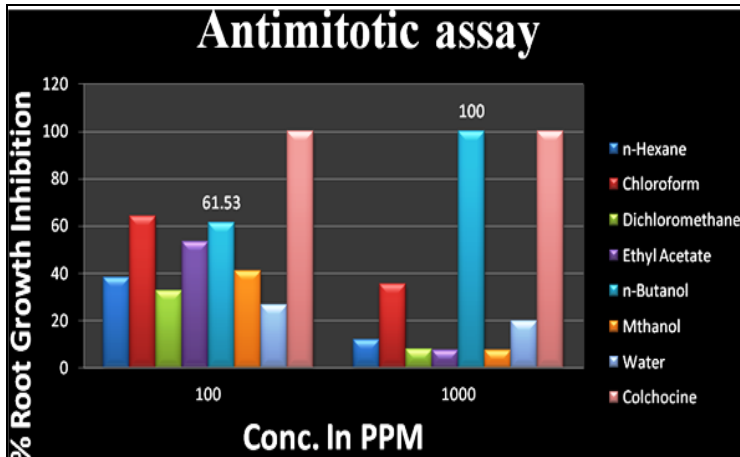
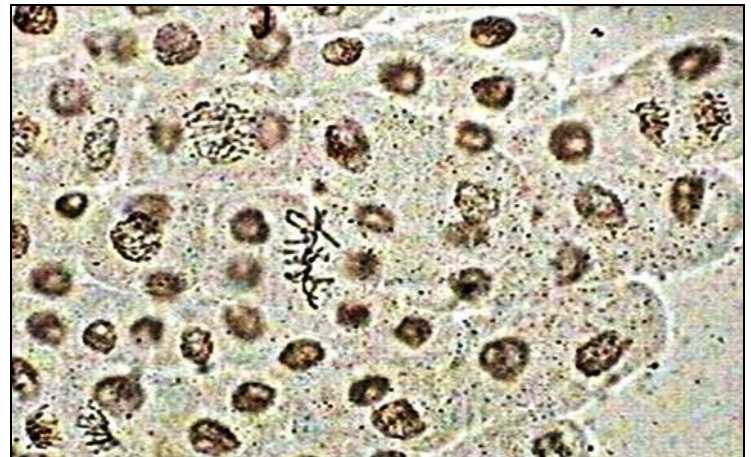
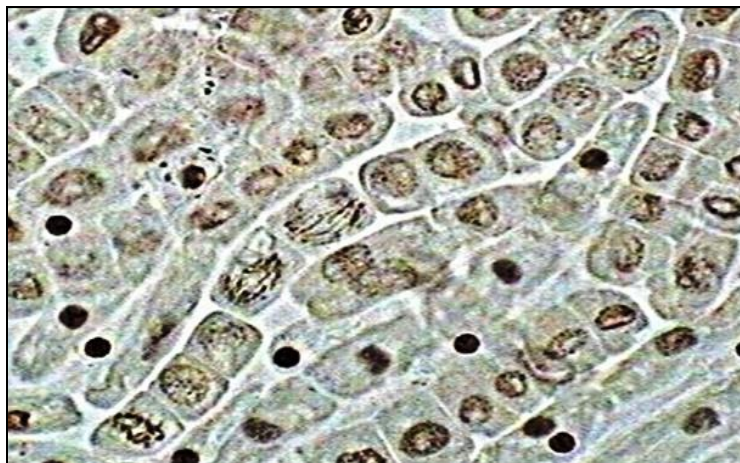


FIGURE 2: GRAPHICAL PRESENTATION OF ANTIMITOTIC ASSAY



B) N-BUTANOL 100ppm



A) WATER CONTROL



C) COLCHICINE 100ppm

FIGURE 3: OBSERVED MITOTIC PHASES

TABLE 1: PERCENTAGE YIELD OF *LAGENARIA SICERARIA* FRUIT EXTRACTS

Solvent	Color	Consistency	% Yield
n-Hexane	Yellow	Sticky	0.21
Chloroform	Green	Non sticky	3.35
Dichloromethane	Brown	Sticky	0.21
Ethyl Acetate	Brown	Sticky	15.85
n-Butanol	Brown	Sticky	13.72
Methanol	Brown	Sticky	2.31
Aqueous (Water: Chloroform)	Black Brown	Non sticky	25.80

TABLE 2: PRELIMINARY PHYTOCHEMICAL SCREENING OF DIFFERENT EXTRACTS *LAGENARIA SICERARIA* FRUIT

Constituents	Phytochemical Tests	Extracts (Fractions)						
		NH	CHL	DM	EA	NB	METH	WAT
Carbohydrates	Molisch's Test	-	+	+	-	+	+	-
Reducing sugar	Fehling's Test	-	+	-	-	-	-	+
Monosaccharide	Barfoed's	-	-	-	-	-	-	+
Pentose sugar	Bial's orcinol	-	-	-	-	-	-	-
Hexose (fructose)	Selvinoff's	-	-	-	-	+	+	+
Non reducing Sugar	Tannic acid	-	-	-	-	-	-	-
Proteins	Ninhydrin	-	-	-	-	-	-	-
Steroids and	Liebermann	+	+	+	+	+	+	+
Triterpenoids	Burchard Test	+	+	+	+	+	+	+
Anthraquinones	Borntrager's	-	-	-	+	-	-	+
Flavones	Shinoda	-	-	-	-	-	-	+
Alkaloids	Dragendroffs Test	-	-	-	-	-	-	+
Tannins	Ferric chloride Test	-	-	-	+	-	-	+

TABLE 3: RESULT OF EVALUATION OF ANTIMITOTIC POTENTIAL OF *LAGENARIA SICERARIA* FRUIT

Sr. No.	Drug/ Extracts	Conc. (in ppm)	Root Length (in cm)	% Root (Growth Inhibition)	Mitotic index
1	n-Hexane	100	4.2	38.09	22.36
		1000	3.3	12.00	24.61
	Control	-	4.1	-	24.28
2	Chloroform	100	4.7	64.04	30.76
		1000	4.5	08.88	35.36
	Control	-	3.3	-	22.50
3	Dichloromethane	100	4.9	32.65	32.50
		1000	5	08.0	32.85
	Control	-	5.3	-	28.57
4	Ethyl acetate	100	3	53.33	21.25
		1000	2.2	07.5	32.50
	Control	-	1.9	-	27.14
5	n-Butanol	100	2.6	61.53	20.00
		1000	0.4	100	19.56
	Control	-	0.2	-	36.66
6	Methanol	100	3.9	41.02	38.57
		1000	5.4	07.40	30.70
	Control	-	4.0	-	28.57
7	Water	100	6.0	26.66	43.75
		1000	2.0	20.00	40.56
	Control	-	4.5	-	25.00
8	Colchicine	100	1.6	100	0
		1000	0.4	100	0
	Control	-	4.5	-	25.00

**CONCLUSION:** n-Butanol extract of *Lagenaria siceraria* Fruit powder showed the promising Antimitotic activity so it was selected as a lead extract. Further isolation of active moiety from n-butanol extract for anticancer activity by chromatographic techniques is almost completed.

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## REFERENCES:

1. Gangwal A., Par mar S. K., Sheaths N. R: Triterpenoids, Flavonoids and Sterols from *Lagenaria Siceraria* Fruits. Scholars Research Library. 2010; 2 (1) 307-317.
2. Meenal S. Kubde, S. S. Khadabadi, I. A. Farooqui, S. L. Deore: *Lagenaria Siceraria*: Phytochemistry, Pharmacognosy And Pharmacological Studies Report And Opinion 2010; 2(3), 91-98.
3. Rakesh P. Prajapati. Manish Kalariya, Sachin K. Paramar, Navin R.Sheth: Phytochemical and Pharmacological review of *Lagenaria Siceraria*.2010; IP: 117.241.248.98.
4. Vivek Shrivastav, Dr.Ch.V.Rao, Anil Pasndy, Vivek yadav: Antiulcer activity of methanolic fruit extract of *Lagenaria Siceraria* (mol.) Standl. International journal of Pharmaceutical Research and Development (IJPRD) 2011; Vol 3(7): (187-192).
5. Pankaj Nainwal, Karan Dhamija, Sacchidanandmani Tripathi: Study of Antihyperlipidemic Effect on the Juice of the Fresh Fruits of *Lagenaria Siceraria*. International Journal of Pharmacy and Pharmaceutical Sciences 2011; Vol 3, Issue 1. ISSN- 0975-1491. 88-90.
6. Prerona Saha, Upal K. Mazumder, Pallab K. Haldar, Sriparna Kundu Sen, Sagar Naskar: Antihyperglycemic Activity Of *Lagenaria Siceraria* Aerial Parts On Streptozotocin Induced Diabetes in Rats. Original Research Article Accepted: May 9, 2011. 49-60.
7. S. L. Deore, S.S. Khadabadi, Q. R. Patel, S. P. Deshmukh, M. S. Jaju, N. R. Junghare, T.P. Wane and R.G. Jain: In Vitro Antioxidant Activity and Quantitative Estimation of Phenolic Content of *Lagenaria siceraria* Rasayan Journal.Vol.2, No.1 (2009), 129-132.
8. P. Erasto and Z.H. Mbwambo: Antioxidant Activity and HPTLC Profile of *Lagenaria sSiceraria* Fruits. Tanzania Journal of Health Research, April 2009 Vol. 11, No. 2. 79-83.
9. P. Saha, U. K. Mazumder, P. K. Haldar , A. Islam And R. B. Suresh Kumaz: Evaluation of Acute and Subchronic Toxicity of *Lagenaria siceraria* Aerial Parts. International Journal of Pharmaceutical Sciences and Research, 2011; Vol. 2(6): 748-753
10. Harborne, J. B. Phytochemical Methods. 1<sup>th</sup>ed Chapman and Hall, London Springer publication 1998; 10-23.
11. Khandelwal KR. Preliminary Phytochemical screening. Practical Pharmacognosy. 6<sup>th</sup> ed. Pune, India: Nirali Prakashan 2006; 149-539.
12. R. Sehgal, S. Roy, and V.L. Kumar: Evaluation of cytotoxic potential of latex of *Calotropis procera* and Podophyllotoxin in *Allum cepa* root model. BIOCELL 2006; 30(1): 9-13.
13. S. Kavitha Bagya, P.V. Rajashree and Kishor Gnana Sam: Preliminary Anticancer screening and Standardization of some Indigenous Medicinal Plant using cell-biology and Molecular Biotechnology Based models. Research journal of medicinal plant 20011; 5(6); 728-737.

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