European Journal of Molecular & Clinical Medicine
ISSN 2515-8260 Volume 08, Issue 02, 2021

IN-VITRO CYTOTOXICITY ACTIVITY OF SOLANUM XANTHOCARPUM AGAINST MCF7, HELA, A549 AND CACO2 CELL LINES BY MTT ASSAY

G. Sreedhar , T Bhaskara Rao*

Dept of Chemistry, KLEF (Deemed To Be University), Vaddeswaram, Guntur-5222502

Abstract

The present investigation aims to evaluate anticancer activity of methonolic extract of Solanum xanthocarpum fruits. The extract is investigated for its inhibitory effect on MCF7, HeLa, A549, CaCo2 cell lines. Percentage viabilities of cell lines are assessed by adopting the MTT method. The extract has significant cytotoxicity on MCF 7, HeLa, A549, CaCo2 cell lines in the concentration range between 10 to 100 µg/ml. as per MTT assay. IC50 values of Solanum xanthocarpum on MCF7, HeLa, A549, CaCo2 cell lines are 27.99, 75.55, 54.66 and 156.36 respectively. From the performed assay, methanolic extract has more cytotoxic effect on MCF 7 and least activity on CaCo2 cell line. Thus the extract of Solanum xanthocarpum fruits has identified to have anticancer activity

Keywords: Cytotoxicity, Solanum xanthocarpum, methanol extract, clines, MTT

Introduction

Cells viability and proliferation rates are good indicators of cells health. Cell health and metabolism are affected by physical and chemical agent's .Viability of Cells and proliferation rates reflect health of the cells. Cell health and metabolism are affected by physical and chemical agents these reagents hinder the growth of cancer cells by various mechanisms viz., obliteration of cell-membranes, retarding the protein synthesis by binding over to receptors. Further, inhibition of the growth of cancer cells may also be caused due to elongation of oligodeoxynucleotide and prevention of enzymatic reactions

The reagents employed for this purpose must be bio-compactable and do not because side reactions. Many of the synthetic drugs have ill-effects and cussing other disorders. In this context, the chemicals derived from the plant materials are interesting researcher in the recent

past. Plants constitute one of the major sources of drugs in modern as well as traditional medicine throughout the world[2]. The bioactive substances in plants are mainly produced as secondary metabolites. **Methods have been developed** to cure various diseases like gonorrhea, rheumatism, cough, asthma, catarrhal fever and sore throat by using extracts from the bio-materials. In fact our research group has investigated Cytogenetic properties of some mangroves belong to Krishna-Godavari estuary[3].

Solanum xanthocarpum (SX) plant is medicinal herb [11]and is known for its medicinal values. The plant belongs to the Solanaceae family. It is commonly known as Indian night shade or Yellow berried nightshade. Alcoholic or aqueous extracts of Solanum xanthocarpum (SX) plant have shown hypotensive effect, antiviral activity (against Ranikhet disease virus) and against sarcoma-180 in mice[5]. In the present study, the cells viability and proliferation in the alcoholic extract of Solanum xanthocarpum furt has been investigated[12-19].

MATERIALS AND METHODS

Plant Collection

Fruits of the '*Solanum xanthocarpum'* were collected from the Kuragallu rural area of district Guntur, Andhra Pradesh, India. The voucher -specimen was preserved in the chemistry lab of K L university.

Preparation of Extract

The collected fruits of the *Solanum xanthocarpum* were cleaned and dried under shade for a period of one month. The dried fruits were ground to fine powder. Powders of the fruits were successively extracted with Methyl alcohol (60- 80 C) using a Sock let extractor. Extractions of

the solvent were collected, filtered and vacuum dried in a rotary evaporator at 40 ± 5^{O}C. The dried product obtained is analyzed for it Cytotoxic activity.

Cytotoxicity bioassays by MTT method

The various Cell lines tested by MTT methods were presented in Table1.The chemical used in this investigation were of Analytical Grade quality. DMEM (Dulbecco's modified Eagle's medium), MTT [3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyl, tetrazolium bromide, EDTA Phosphate Buffered Saline (PBS) and trypsin, were procured from Sigma Chemicals. Fetal Bovine Serum (FBS) was purchased from Gibco. 25 cm ^{2} and 75 cm^{2} flask and 96 well plated were purchased from Eppendorf India.

Table 1: Different Cell lines tested MTT method

S. No	Compound Name	Cell Line tested
1	Solanumxanthocarpum (SX) extract	MCF7
		HeLa
		A549
		CaCo2

The Cancer cell lines were purchased from NCCS, Pane. Cells were maintained in MEM supplemented with 10% FBS and antibiotics penicillin/streptomycin (0.5 mL^{-1}), in an atmosphere of 5% CO $_{2}$ /95% air at 37^{0} C.

Preparation of Testing Extract

The required quantity of the extra was correctly weighed and dissolved in DMSO so as to obtain the concentration of 1mg/ml. This solution was subsequently diluted to a series of concentrations ranging from 10 to 100 µg/ ml.

MTT ASSAY

This method is based on colorimetric estimation. In this method, the reduction of yellow 3-(4, 5-dimethylthiazol- 2-yl)-2, 5-diphenyl tetrazolium bromide (MTT) caused by 'mitochondrial *succinate dehydrogenase' present in live cells, was used as a basis.* In live cells, the presence of enzyme causes the reduction while in dead cell (or their compounds), the reduction is not

affected. Thus, the reduction is based on number of live cells and not on dead cells. In live cells the MTT penetrates into 'mitochondria' and get reduced to form insoluble deep purple coloured formazan crystals. On treating the crystals with DMSO, the cell were dissolved in the solvent and thereby releasing the Formosan. The Formosan is assayed spectrophotmetriacllly as max at 570 nm.

The IC50 values obtained for the extract wires pet to different cell lines were presented in Table 2.Cell viability was evaluated by the MTT Assay with three independent experiments with six concentrations of compounds in triplicates.

Cells were trypsinized and performed the tryphan blue assay to know viable cells in cell suspension. Cells were counted by haemocytometer and seeded at density of 5.0 X 10^{3} cells well in 100 µl media in 96 well plate culture medium and incubated overnight at 37 ^{0} C. After incubation, take off the old media and add fresh media 100 µl with different concentrations of test compound in represented wells in 96 plates. After 48 hrs Discarded the drug solution and added the fresh medic with MTT solution (0.5 mg / $mL^{-1)}$ was added to each and the plates were incubated at 37 ^{0} C for 3 hrs. At the end of incubation time, precipitates are formed as a result of the reduction of the MTT salt to chromophore formazan crystals by the cells with metabolically active mitochondria. The optical density of solubilized crystals in DMSO was measured at 570 nm on a microplate reader. The percentage growth inhibition was calculated using the following formula.

$$\% \text{ Inhibition} = \frac{100\ (\text{Control} - \text{Treatment})}{\text{Control}}$$

The Ic50 value was determined by using linear regression equation i.e. Y =Mx+C. Here,

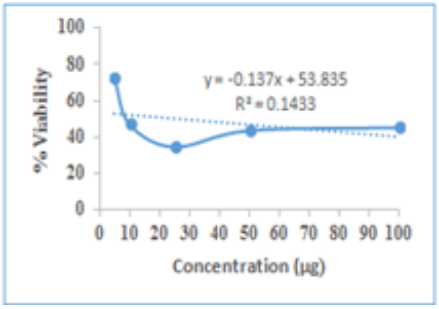
Y = 50, M and C values were derived from the viability graph.

Results and Conclusion

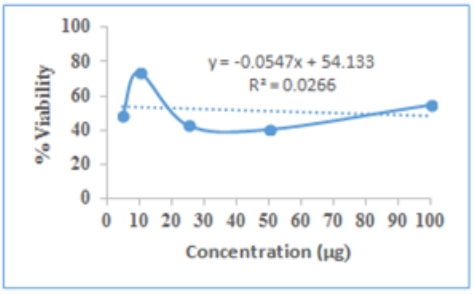
The IC_{50} values are tabulated in table2. The present study describes *in vitro* anti cancer activity of Solanum xanthocarpum alcoholic extract. From the result of the cytotoxicity evolution of the extract, it displayed a potent activity against MCF7 and A549 cell lines with the IC50 values 27.99 and 54.66.The cytotoxic effects also plotted in graph concentration versus viability percentage.

Table 2:Cytotoxicity by MTT assay of Solanum Xanthocarpum Methanolic extract

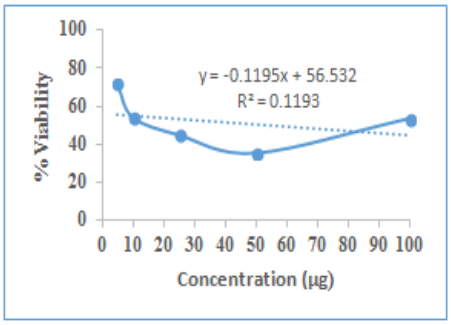
S.NO	Sample Name	Cell Line	**IC_{50} (µg)**
1	SX methanolic extract	MCF 7	27.99
2	SX methanolic extract	HeLa	75.55
3	SX methanolic extract	A549	54.66
4	SX methanolic extract	CaCo2	156.37



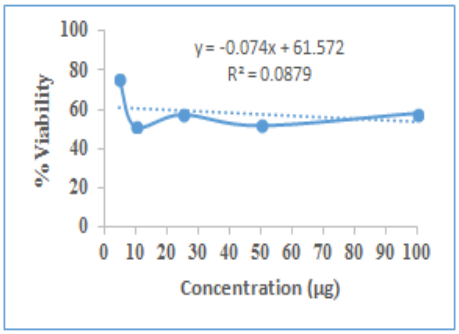
MCF7Cells Cell Lines



Hela Cells Cell Lines



A549 Cell Lines



Caco2 Cell Lines

Figure 1: Results of cell viability assay of different concentrations of Solanum xanthocarpum extracts on MCF7, HeLa, A549 AND CaCo2 are shown in the graphs.

REFERENCE

[1] Ishiyama M, Tominaga H, Shiga M, Sasamoto K, Okhura Y, Ueno KA. Violet.Biological & Pharmaceutical Bulletin.19, 1518, 1996.

[2] Sachin Parmar, Amit Gangwal, Navin Sheth, Der Pharmacia Lettre, 2, 373, 2010.

[3] T.Bhaskara Rao,B. Venkateswara Rao and K Ramanjaneyulu, International Journal of Pharma and Bio Sciences 5, 396, 2014.

[4] AK, Al-Asady, Majeed G H, and Al-Waely W A. "Study of rhamnolipids cytotoxicity, inhibitory effect on some microorganisms and applying in food products."International Journal of Bio-Technology and Research (IJBTR) 6.3 (2016):1-8.

[5] A Venkanna, B. Siva, B. Poornima, P.R. Rao Vadaparthi, K. Rajendra Prasad, K. Ashok Reddy, G. Bhanu Prakash Reddy, K. Suresh Babu, Fitoterapia 95,102,2014.

[6] Khizar Abbas1, Uzma Niaz, Talib Hussain, M. Asif Saeed, Zeeshan Javaid *, Arfat Idrees and Shahid Rasool Acta Poloniae Pharmaceutica Ñ Drug Research, 7,415, 2014.

[7] Borkataky, Munmi, and K. A. U. S. H. A. L. Sood. "Antibacterial, Antioxidant and Cytotoxic Activities of Cinnamomum Tamala Nees. Leaves."International Journal of Medicine and Pharmaceutical Sciences: 55-62.

[8] R. Udayakumar, K. Velmurugan And Raghu Ram Krishna, Vol: XXVI (2) October, November, December – 2004 3DJHV.

[9] Raikar, Shanthanu M., Vishal U. Kalebar, And Shakeel Ahmed Adhoni. "Screening Of Pharmacological And Cytotoxic Activities Of Fresh Water Lake Isolated Microalgae Chlorella Vulgaris As-13 And Chlorella Pyrenoidosa As-6." International Journal of Bio-Technology and Research (IJBTR) 8.4, Aug 2018, 1-8.

[10] Sanjay patel, Nirav Gheewaka, Ashok Suthar, Anand,Acta Poloniae Pharmaceutica ñ Drug Research, 71,415,2014.

[11] Suri, R.K., Chaudhari, D.C. and Jaffer, R. Commercially important medicinal plants from forests. J.Eco. Bot. Phytochemistry 3, 129, 1992.

[12] Chopra., R.N., Nayar, S.L. and Chopra, I.C. Glossary of Indian Medicinal plants National Institue of Science Communication, New Delhi (1996).

[13] Ol'ga, D., Ludmila P. Ovchinnikova, And Leonid I. Goryunov. "Antioxidant Activity And Cytotoxicity Of Polyfluorinated 1, 4-Naphthoquinones With Alkylating Functions In The Quinone Moiety."Therapy 4:5. International Journal of Medicine and Pharmaceutical Sciences (Ijmps) 5.1, Feb 2015, 11-20.

[14] Ashok Purohit. Contraceptive Efficacy of Solanum xanthocarpum berry in male rats. Ancient science of life, 182, 264, 1992.

[15] Ambasta, S.P. The Useful Plants of India. Publications and information Directorate, CSIR, New Delhi, 10, 1992.

[16] Udayakumar, R. and Hazeena Begum, V. Antimicrobial studies of some selected medicinal plants, ancient science of life, XXI, 230, 2002.

[17] *Malhotra, Sonam, Neetu Jha, and Krutika Desai. "A superficial synthesis of selenium nanospheres using wet chemical approach."Int J Nanotechnol Appl3 (2014): 7-14.*

[18] *Arora, D.S. and Bhardwaj, S.K, Antibacterial activity of some medicinal plants, Geo Bios., 24, 127, 1997.*

[19] *Binutu, O.A.Fitoterapia, LXIX, 1871998.*

[20] *Barnabas, C.G.G. and Nagarajan, S. Fitoterapia, 59, 508, 1988.*

[21] *Miski, M., Ulupeler, A., Johanson, C. and Mabry, T.J.J. Nat. Prod., 46, 874, 1988.*

[22] *Bader, G., Binder, K., Hiller, K. and Zieger Bohme, H.Pharmazie, 42, 140, 1987.*

[23] *Sachin Parmar*, Amit Gangwal, Navin Sheth, Der Pharmacia Lettre, 2, 373, 2010.*

[24] *Suresh G, Poornima B, Babu KS, Yadav PA, Rao MSA, Siva B, Fitoterpia 86,100,2013.*