In vitro evaluation of the anti-cancer efficacy of Tecoma stans hydroethanolic leaf extract against a liver cancer cell line (HepG2).

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ABSTRACT

Introduction

Flavonoids, alkaloids, quinones, tannins, and traces of saponins and amino acids were found in Tecoma stans. The presence of phytochemicals has been recorded in almost all sections of the plant (leaves, root, flower, seed, fruit, bark) and hence the plant's usage in medical domains. Cancer, along with cardiovascular illnesses, is one of the main causes of death worldwide. The goal of this study is to see if tecoma stans hydro ethanolic leaf extract has anti-cancer properties against liver cancer.

Materials and methods

Human liver cancer cell line (HepG2) was brought from NCCS, Pune, India. Cell viability test and Gene expression analysis were carried out for Wnt and beta catenin mRNA Gene expression using MTT and PCR respectively. The results were analyzed using appropriate statistical tools.

Results

The Wnt and beta catenin gene expression is reduced on induction of 300 and 400 g (dosage) of hydroethanolic extract of *Tecoma stans* with significant difference in comparison with control . Thus Hydroethanolic extract of *Tecoma stans* showed significant anticancer property with the increase in dosage the anticancer activity increased contrastingly .

Conclusion

From this study the obtained results showed that *Tecoma stans* has anti cancer activity and can be used as an anti cancer drug in the medical field through the years of development of these drugs .

Key words ; *Tecoma stans* ; anti cancer ; hepatic cell line ; Wnt/beta catenin; Innovative technique.

INTRODUCTION

The plant *Tecoma stans* is a part of the family of Bignoniaceae. It is dicotyledonous in nature It has a wide range of medicinal and pharmacological applications. *Tecoma stans* is in diverse distribution in nature following the tropical and subtropical American region including Mexico to Argentina. It is also found in the Caribbean and the Bahamas. The species is characteristic of rocky slopes, often limestone outcrops , alluvial soils accompanied with drainage. It is common among the deforested and other disturbed sites and along roadways. Yellow elder has naturalized in much of tropical and subtropical Africa, Asia, the Pacific Islands and Australia (1). *Tecoma stans* is used in some parts of america as an anti anti diabetic drug. (2) .Even though *Tecoma stans* has been in medicinal use traditionally since ancient times its present implementation on smooth muscle cells remain very scarce (3) . The presence of phytochemicals has been recorded in almost all sections of the plant (leaves, root, flower, seed, fruit, bark) and hence the plant's usage in medical domains. (4) (5)

Cancer is one of the leading causes of death (6) . Normal cells have a property of contact inhibition that is during cell growth if the cell comes in contact with the cell membrane of another cell , it stops growing . Cancer cells lose the property of contact inhibition due to which they have uncontrolled cell growth and cell multiplication(4) . There are two types of cancer ; benign and malignant cancer . Benign cancer is location specific and only grown in that particular area of the body causing cancer . It can be cured by removing the tumor through surgery . The experience from our previous studies (7)

(8,9) (8)(10)(11)(12)(13)(11,13)(14)(15) (16) have led us to concentrate on the study.

Primary liver cancer, which starts in the liver, as well as malignant tumours from other regions of the body, can harm the liver. The majority of cancers are secondary or metastatic, meaning they began elsewhere in the body. Every year, cancer kills millions of people around the world. Surgery, radiation, or therapy like as chemo, hormone, and biological therapy can all be used to treat it.. Since ancient times, traditionally plants have been the source of medicines for the treatment of various diseases.(1) According to the WHO, a considerable portion of the world's population relies on plant-based medications and therapies to meet their main health-care needs.(17) Drugs derived from plants have fewer side effects compared to synthesised drugs , hence the scope for plant based medicines increases day by day . Studies at molecular levels were performed by our team of researches which insisted us to proceed this study (18–25),(26),(27),(28),(29,30),(31),(32),(33–37) The aim of this study is the evaluate the anti cancer activity of the hydroethanolic extract of leaf of *Tecoma stans* against cancer cells of liver (38)

MATERIALS AND METHODS

Cell line centre, Pune, India, provided the Human liver cancer cell line (HepG2). Tissues were cultured in RPMI media with 10% foetal bovine serum, 100 U/ml penicillin, and 100 g/ml streptomycin at 37 degrees Celsius and 5% CO2.The MTT test was used to measure cell growth. HepG2 tissues were sown in 96-well plates with 5x104/200l and grown overnight. Six duplicate wells were used in each treatment. All of the tissues were then grown for another 48 hours. The experiment was carried out three times. The MTT absorbance in negative control tissues was employed as a 0 percent cell inhibition measurement. The expression status of m RNA was analysed by Polymerase chain reaction for identifying the fold change of Wnt and beta catenin Gene expression m RNA expression over control samples. The samples were assessed using (ANOVA) and Duncan's multiple range test with p value at 0.05.

RESULT

Tecoma stans was found to reduce the abnormal proliferation of cells by reducing its cell viability at concentration (100-400 µg/ml. [Fig 1]. Effect of tecoma stans Wnt and Beta catenin, the cells were reduced in its proliferation and the reduction in in mRNA expression is observed. [Fig 2] [Fig 3]

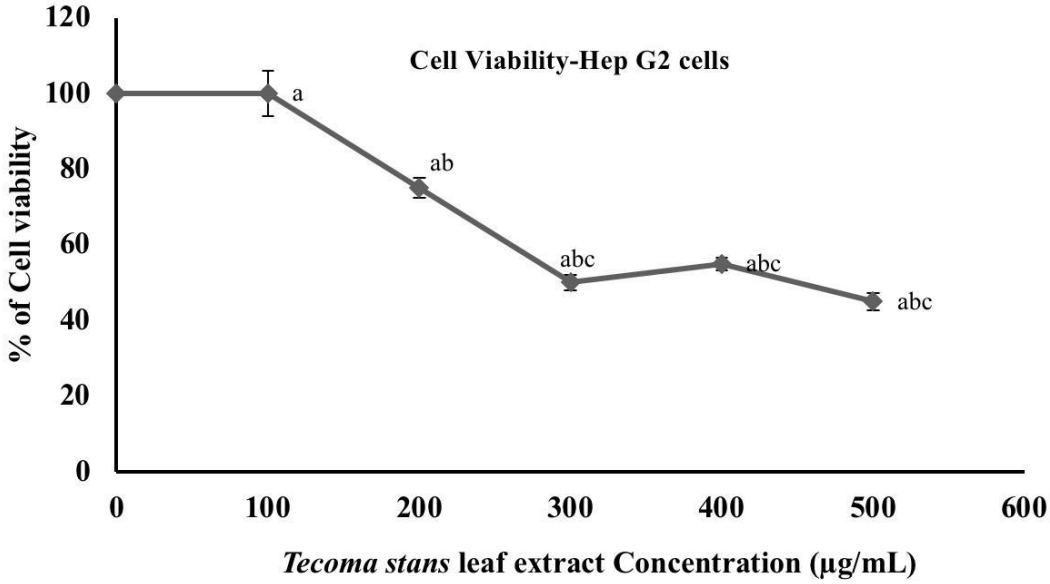

Figure 1: shows the *Tecoma stans* leaf extract on cell viability in HepG2 cells.

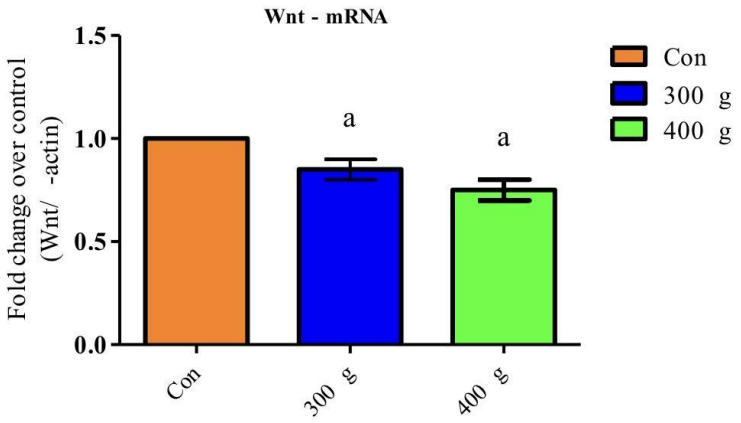

Figure:2 shows the effect of *Tecoma stans* leaf extract on Wnt mRNA expression in HepG2 cells.

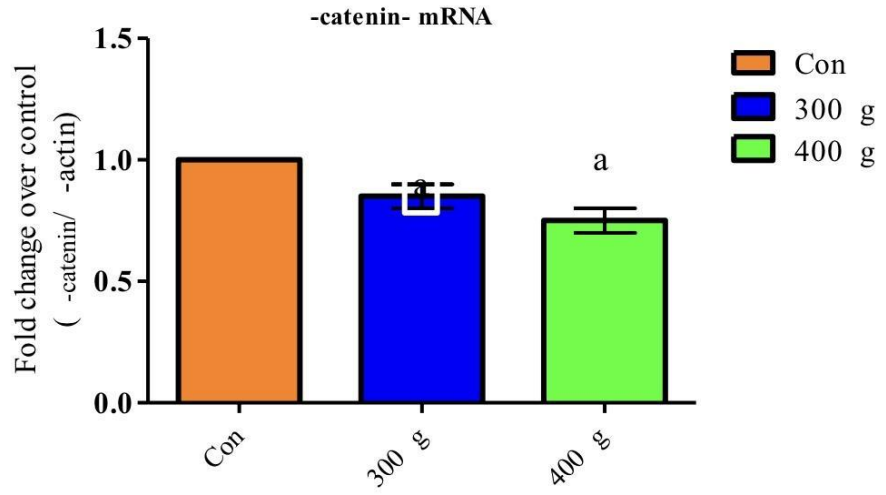

Figure :3 shows Effect of *Tecoma stans* leaf extract on B- Catenin mRNA expression in HepG2 cells

DISCUSSION

From this study it can be suggested that the Wnt and beta catenin gene expression is reduced on induction of 300 and 400 **µ**g/mL (dosage) of hydroethanolic extract of *Tecoma stans* with significant difference in comparison with control . Thus Hydroethanolic extract of *Tecoma stans* showed significant anticancer property with the increase in dosage the anticancer activity increased contrastingly .From this study the obtained results showed that *Tecoma stans* has anti cancer activity and can be used as an anti cancer drug in the medical field through the years of development of these drugs .

Wnt qualities encode emitted glycoproteins, which intercede intercellular flagging either over short or significant distances relying upon the tissues where they are being expressed. In creatures for the most part, Wnt flagging is engaged with a wide assortment of cell connections from early advancement through the grown-up stage. The current audit centers around the early formative jobs of Wnt qualities. The Wnt quality family, which has gone through broad quality duplications during metazoan development, can be partitioned into thirteen subfamilies, which fall into three classes varying in the sign transduction falls they trigger. Two of the flagging pathways are interrupted by Disheveled(39,40). In one, Disheveled represses GSK3, which brings about movement of β-catenin to the core. This pathway is associated with pivotal designing and detail of cell destiny. (41) This pathway intervenes planar cell extremity and concurrent augmentation developments. The third Wnt flagging pathway includes G-proteins and results in changes in intracellular Ca2+ levels, which assume parts in cell cycling and tissue partition.(41,42)

One of the critical components that direct cell growth, cell extremity, and cell destiny assurance during early stage improvement and tissue homeostasis is motioning by the Wnt group of secreted glycol lipoproteins.(43) Therefore, changes in the Wnt pathway are regularly connected to human birth deformities, malignant growth and different sicknesses. A basic and most contemplated Wnt pathway is authoritative Wnt flagging, what capacities by managing the measure of the transcriptional co-activator β-catenin that controls key formative quality articulation programs. This audit centers around our present comprehension of Wnt/β-catenin flagging, drawing primarily from hereditary,

formative and biochemical examinations in Drosophila, Xenopus, mice and people. For a more exhaustive and notable viewpoint we allude to prior audits and the Wnt landing page . The nematode Caenorhabditis elegans shows comparative yet additionally different Wnt/β-catenin pathways,(39) which are canvassed somewhere else and in the going with the survey . Wnt likewise initiates various non-sanctioned flagging pathways that are autonomous of β-catenin and have been as of late inspected.(44)

In tumour hosts, a decrease in viable cell count and an increase in non viable cell count toward normal suggest an anticancer effect.(45). Limitations of this study is that this is an in vitro study and future studies should be carried out invivo with large sample size to make the context evident..

CONCLUSION

From the obtained results and within limits of the study , it may be concluded that the 0hydroethanolic extract of leaf of *Tecoma stans* has a massive anti cancer potential against liver cancer by decreasing level of Wnt and beta catenin in the gene expression further studies have to be done in the future as hydroethanolic extract of leaf of *Tecoma stans* can be used as an effective anticancer drug.

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CONFLICT OF INTEREST

All the authors declare that there was no conflict of interest in the present study .

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