

## EVALUATION OF CHEMOPREVENTIVE ACTIVITY OF SOME SPICES THROUGH CELL VIABILITY ASSAY AGAINST SELECTED CELL LINES

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### ABSTRACT

Spices have been traditionally used for prevention and cure of many diseases. Hence, this study was aimed to investigate the effects of aqueous and alcoholic extract of four different spices (asafoetida, ginger, cinnamon and cardamom) extracts on HeLa cell lines, human breast adenocarcinoma cell line (MCF) and HEP-G2 cancer cell lines through cell viability assay. Aqueous and alcohol extract (20 mg/ml) of asafoetida showed better Chemopreventive activity. Cell viability was 14% for aqueous extract whereas 9% for alcohol extract on HeLa cell line; 18% Cell viability for aqueous extract whereas 14% for alcohol

extract on MCF-7 cell line; 14% Cell viability for aqueous extract whereas 9% for alcohol extract on HEP-G2 cell line. Therefore, these spices might be used for natural healing of the tumor.

**KEYWORDS:** Chemopreventive activity, HeLa cell lines, human breast adenocarcinoma cell line (MCF), HEP-G2, Spices.

### INTRODUCTION

Spices are defined by the US Food and Drug Administration as, “aromatic vegetable substances, in the whole, broken, or ground form, whose significant function in food is seasoning rather than nutrition.”<sup>[1]</sup>

Although, spices have been well known for their medicinal, preservative and antioxidant properties, they have been currently used with primary purpose of enhancing the flavor of foods rather than extending shelf-life. Spices and their derivatives have been used in food housekeeping for thousands of years, bringing about differentiated flavor and aroma. The

inhibitory effect of various spices has been found against food borne microorganisms.<sup>[2]</sup>

Spices have been used for thousands of years to enhance the flavour, colour and aroma of food. In addition to boosting flavour, herbs and spices are also known for their preservative<sup>[3]</sup> and medicinal value, which forms one of the oldest sciences<sup>[4]</sup> A large number of plants are used to combat different diseases<sup>[5,6]</sup> and possess antimicrobial activity.<sup>[7,8,9,10,11,12,13,14]</sup>

The future of the natural habitat of medicinal plants is being threatened by ever increasing anthropogenic activities. Increased commercialization has resulted in over-harvesting of medicinally useful plants, which has diminished their number so they are now in danger of extinction. To overcome this alarming problem, the discovery of novel active compounds is the need of the day. Previously, all drugs and medicinal agents were derived from natural substances, especially from higher plants.

Considering the above discussion, chemopreventive activity of spice extracts i.e. asafoetida, ginger, cinnamon and cardamom were evaluated against HeLa cell lines, MCF and HEP-G2 cancer cell lines.

## **MATERIALS AND METHODS**

### **Spice samples and Extract Preparation**

Asafoetida, ginger, cinnamon and cardamom were bought from the local market in Kanpur. Extracts were prepared by the method of Clarkson and Bibby, 1969.<sup>[15]</sup> Both water and alcohol extracts of spices were used. Water extracts were made by extracting 5 gm of ground spice in 100 ml distilled water in a Soxhlet extraction apparatus for four hours at 100° C. To prepare alcohol extracts 5 gm of ground spice was added to 100 ml of absolute alcohol and agitated at room temperature for eight hours in a wrist-action shaker. Thereafter, the mixture was allowed to stand for 12 hours, the alcohol evaporated without heat, and the residue was mixed with 100 ml of distilled water at 80° C.

### **Cell Lines used**

Human epithelial carcinoma (HELA) Cell Line, Human breast adenocarcinoma cell line (MCF) and HEP-G2 cancer cell lines were used for determination of anti-cancer effects of selected spices.

### Maintenance of Cell Culture

The cells at a density of  $1 \times 10^5$  from each cell line were transferred to MEM media and media was replaced after every 2 days until the outgrowth had spread to cover at least 50% of the growth surface. Further, the cells were sub cultured by enzymatic method using trypsin and maintained at MEM medium.

### Cell Viability Assay

Results of cell viability assay of spice extracts at different concentrations on selected cell lines are shown in Figures 1-3.

Aqueous and alcohol extract (20 mg/ml) of asafoetida showed better chemopreventive activity. Cell viability was 14% for aqueous extract whereas 9% for alcohol extract on HeLa cell line; 18% Cell viability for aqueous extract whereas 14% for alcohol extract on MCF-7 cell line; 14% Cell viability for aqueous extract whereas 9% for alcohol extract on HEP-G2 cell line.

On HeLa cell lines, among the selected spices, asafoetida was found to be most effective. The aqueous and alcohol extracts of asafoetida at 20 mg/ml concentrations have shown 14% and 9% cell viability respectively. The aqueous and alcohol extracts of cinnamon at 20 mg/ml concentrations have shown minimum Chemopreventive action against the HeLa cell lines with 41% and 18% cell viability respectively (Figure 1).

On MCF-7 cell lines, among the selected spices, asafoetida was found to be most effective. The aqueous and alcohol extracts of asafoetida at 20 mg/ml concentrations have shown 18% and 14% cell viability respectively. The aqueous and alcohol extracts of cinnamon at 20 mg/ml concentrations have shown minimum Chemopreventive action against the MCF-7 cell lines with 35% and 24% cell viability respectively (Figure 2).

On HEP-G2 cell lines, among the selected spices, asafoetida was found to be most effective. The aqueous and alcohol extracts of asafoetida at 20 mg/ml concentrations have shown 14% and 9% cell viability respectively. The aqueous and alcohol extracts of cinnamon at 20 mg/ml concentrations have shown minimum chemopreventive action against the HEP-G2 cell lines with 36% and 27% respectively (Figure 3).

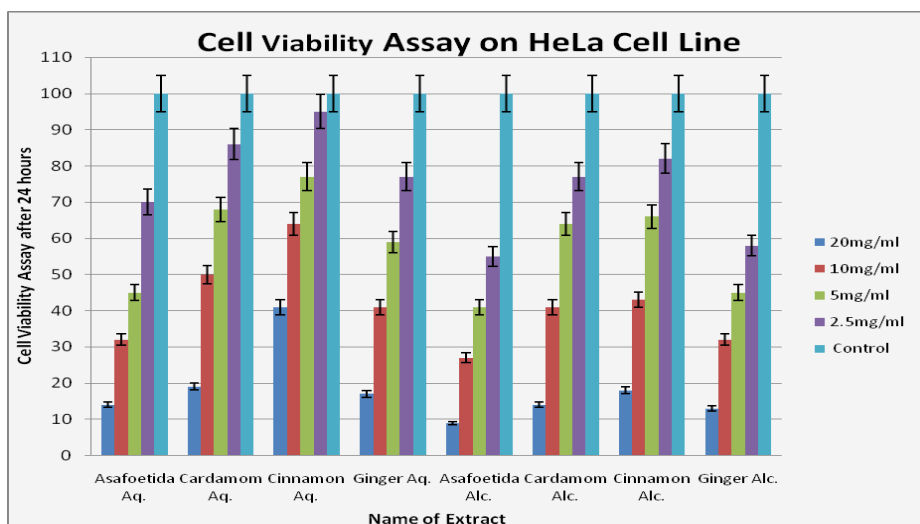
Chemopreventive potential of selected spices was observed to be in decreasing order of Asafoetida, Ginger, Cardamom and then Cinnamon.

Alcohol extracts of the spices has shown better chemopreventive activity for all the selected cell lines.

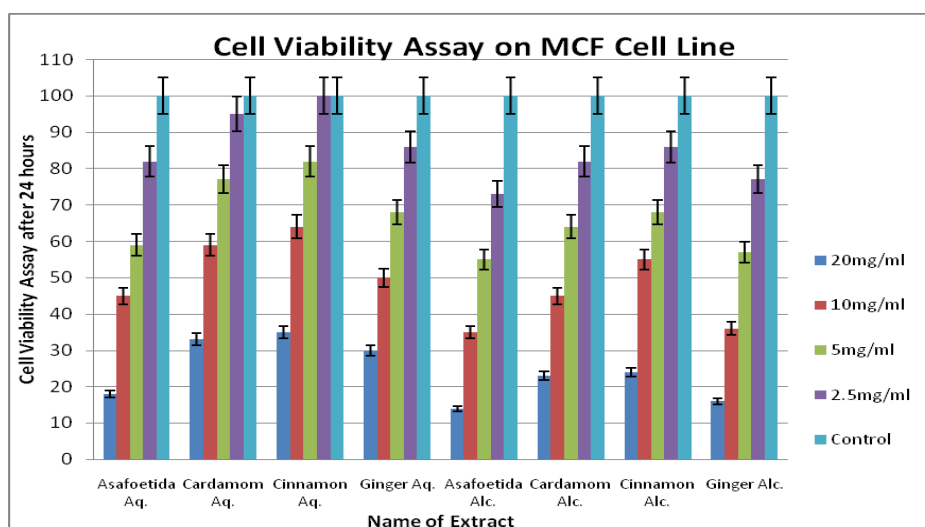
Similar results were reported by Lee and Surh (1998) that the ginger extract has been found to exert inhibitory effects on the viability of human HL-60 (promyelocytic leukemia) cells.

The ginger extract inhibited proliferation of cancer cell lines using the cell viability assay for chemopreventive activity (Sharma *et al.*, 2009; Abdullah *et al.*, 2010).

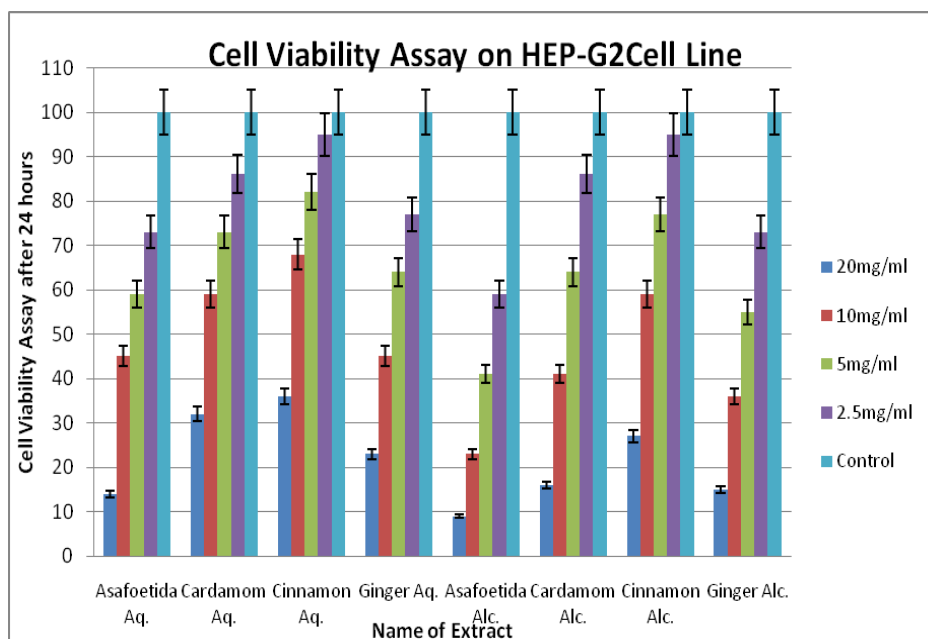
The chemopreventive activity of cinnamon using cell viability assay was reported earlier by Kwon *et al.* (2010).



**Figure 1:** Cell viability assays of spice extracts on HeLa cell line.



**Figure 2.** Cell viability assay (expressed in percentage) of spice extracts on MCF-7 cell line.



**Figure 3: Cell viability assay (expressed in percentage) of spice extracts on HEP-G2 cell line.**

## CONCLUSION

Herbs and spices are well known for their preservative and medicinal value, which can be investigated in discovering novel chemopreventive compounds. This study was aimed to investigate the effects of aqueous and alcoholic extract of four different spice (asafoetida, ginger, cinnamon and cardamom) extracts on HeLa, MCF and HEP-G2 cancer cell lines through cell viability assay. Spice extracts were used for Cell Viability assay on various selected cancerous cell lines that exhibited significant inhibitory effect. Aqueous and alcohol extract (20 mg/ml) of asafoetida showed better Chemopreventive activity,. Chemopreventive potential of selected spices was observed to be in decreasing order of Asafoetida, Ginger, Cardamom and then Cinnamon.

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