



Genotoxicity Analysis of *Coriandrum Sativum* on Oral Cancer Cell Line by DNA Fragmentation

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ABSTRACT

The aim of the study is to determine the Genotoxicity analysis of *Coriandrum sativum* on oral cancer cell lines by DNA fragmentation method. Coriander grows wild over a wide area of Western Asia and southern Europe, prompting the comment, "It is hard to define exactly where this plant is wild and where it only recently established itself. Oral cancer, also known as mouth cancer, is a type of head and neck cancer and is any cancerous tissue growth located in the oral cavity. Oral or mouth cancer most commonly involves the tongue. It may also occur on the floor of the mouth, cheek lining, gingiva (gums), lips, or palate (roof of the mouth). The *Coriandrum sativum* extract showed effective anti-cancer activity against oral cancer cell lines.

Keywords: Computer aided drug design, Drug discovery process, Homology modeling, Lead optimization.

INTRODUCTION

Coriandrum Sativum is a commonly used herb for most of the time culinary and food seasoning purpose. *Coriandrum sativum* is commonly known as Dhania in south Asia and cilantro and Parsley in European countries. It has been used in food for adding up a fresh aroma to the food recipes.¹ The fresh leaves are an ingredient in many South Asian foods (such as chutneys and salads); in Chinese and Thai dishes; in Mexican cooking, particularly in salsa and guacamole and as a garnish; and in salads in Russia and other CIS countries.

Chopped coriander leaves are a garnish on Indian dishes such as dal. As heat diminishes their flavour, coriander leaves are often used raw or added to the dish immediately before serving. In Indian and Central Asian recipes, coriander leaves are used in large amounts and cooked until the flavour diminishes.² Roasted coriander seeds, called dhana dal, are eaten as a snack. They are the main ingredient of the two south Indian dishes: sambhar and rasam.³

One preliminary study showed coriander essential oil to inhibit Gram-positive and Gram-negative bacteria, including *Staphylococcus aureus*, *Enterococcus faecalis*, *Pseudomonas aeruginosa*, and *Escherichia coli*.⁴

Although seeds generally have lower content of vitamins, they do provide significant amounts of dietary fiber, calcium, selenium, iron, magnesium and manganese.⁵ Having a deeper, more intense flavour than the leaves, coriander roots are used in a variety of Asian cuisines, especially in Thai dishes such as soups or curry pastes.⁶

Oral cancer, also known as mouth cancer, a type of head and neck cancer and is any cancerous tissue growth

located in the oral cavity.⁷ There are several types of oral cancers, but around 90% are squamous cell carcinomas, originating in the tissues that line the mouth and lips. Oral or mouth cancer most commonly involves the tongue. It may also occur on the floor of the mouth, cheek lining, gingiva (gums), lips, or palate (roof of the mouth). Most oral cancers look very similar under the microscope and are called squamous cell carcinoma, but less commonly other types of oral cancer occur, such as Kaposi's sarcoma.⁸ In 2013 oral cancer resulted in 135,000 deaths up from 84,000 deaths in 1990. Five – year survival rates in the United States are 63%.⁹ Early stage symptoms can include persistent red or white patches, a non-healing ulcer, progressive swelling or enlargement, unusual surface changes, sudden tooth mobility without apparent cause, unusual oral bleeding or epitaixis and prolonged hoarseness.¹⁰

Late stage symptoms can include an indurated area, paresthesia or dysesthesia of the tongue or lips, airway obstruction, chronic serous otitis media, otalgia, trismus, dysphagia, cervical lymphadenopathy, persistent pain or referred pain and altered vision.¹¹ When glossectomy is performed for smaller tumors (< 4 cm), the adequacy of resection (margin status) is best assessed from the respected specimen itself.

The status of the margin (positive/tumor cut through versus negative/clear margin) obtained from the glossectomy specimen appears to be of prognostic value, while the status of the margin sampled from the post-glossectomy defect is not.

The method of margin sampling appears to correlate with local recurrence: preference for tumor bed/defect margins may be associated with worse local control.^{12,13}



MATERIALS AND METHODS

The chemicals used in this assay are procured from himedia.

Preparation of Plant Extract

The plants were washed, wiped and cut into pieces. They were homogenised with 100 ml of ethyl acetate kept overnight and then filtered.

Maintenance of Cell Line

The vial containing KB cell lines acquired from ATCC, was removed from liquid nitrogen freezer and the vial was thawed for 2 minutes by mild agitation in a 37°C water bath. Then it was centrifuged for 10 minutes at 150 to 200g, room temperature. Supernatant was disposed and cells were cleaned with Eagle's minimum essential medium to remove residual DMSO. The cell pellet was re-suspended in 3ml of DMEM with 10% FBS. It was then incubated in a CO incubator (eg: phenol red) in a medium which changed color as an indicator. The culture was then kept in a growth medium with 10% fetal bovine serum, until cell lines were re-established. The cell lines was incubated with varying concentration of wheat grass extract and assessed for its genotoxicity.

Isolation of DNA

1*10 to the power of 6 cells were incubated with 100µl of cell lysis buffer at room temperature for one hour. This was centrifuged for 15 min at 3000rpm at 4c sediment the cell debris. To the supernatant equal volume of phenol: chloroform: isoamyl alcohol mixture was added and mixed well. This was centrifuged at 5000rpm for 15 min. The supernatant was transferred to new tube.

And the centrifugation was repeated again. To the final aqueous phase 40 µl of 3.5M ammonium acetate was added, to this ice cold isopropanol was added to precipitate the DNA. This was incubated at -20degree C for 1 hour, followed by the centrifugation at 10000 rpm for 15min. The pellet was retained and washed with 70% ethanol and stored in 20-50 µl of TE buffer. The samples were analyzed in agarosegel stained with Ethidium bromide.

Genotoxicity by Agarose Gel Electrophoresis

The agarose gel has to be prepared with 1X TAE buffer and stained with 2µl of ethidium bromide. The % of agarose depends upon the molecule to be separated. DNA Samples isolated were loaded with loading dye (2 µl of loading dye is used). Electrophoresis of DNA fragments at 50 volts. Visualisation of DNA fragments in the UV trans-illuminator was done.

RESULTS AND DISCUSSION

DNA fragmentation was observed in different concentration of *Coriandrum sativum* on oral cancer cell line by agarose gel electrophoresis. The gel picture depicts the DNA fragmentation of the KB cells incubated in the *Coriandrum sativum* extract, thus showing the

genotoxicity effect of *Coriandrum sativum* extract. As the concentration of the *Coriandrum sativum* extract increases, the DNA fragmentation also increased.

In diseases such as cancer, induction of apoptosis has been a new target for mechanism-based drug discovery.

Cancer cells evolve to avoid apoptosis-inducing signaling pathway in order to survive.

Apoptosis has been characterised biochemically by the activation of nuclear endonuclease that cleaves DNA into multimers of 180-200 base pairs and can be visualised as an oligosome ladder by standard agarose gel electrophoresis.¹⁴

Thus, induction of apoptosis in cancer cells can be a promising treatment method in cancer therapy. Natural-derived products, extracts or isolated active compounds, had drawn growing attention as agent in cancer therapy due to their ability to modulate apoptosis.¹⁵ Apoptosis involves characteristic morphological and biochemical events ultimately leading to cell demise.¹⁶

CONCLUSION

The herb exhibits anti-cancer activity. From the present study, it was concluded that the extract of *Coriandrum sativum* acts against oral cancer cells.

Thus, the anti-cancer activity of *Coriandrum sativum* may be useful in the treatment of patients with Oral carcinoma.

Further research should be initiated to put light on the anti-cancerous property of the drug.

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