Genotoxicity Analysis of Jojoba Oil on Oral Cancer Cell Line by DNA Fragmentation

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ABSTRACT

The aim of this study is to determine the genotoxicity of Jojoba oil on oral cancer cell line by DNA fragmentation. The objective of the study is to analyse the genotoxicity of jojoba oil on oral cancer cell line by DNA fragmentation. Genotoxicity is a word in genetics defined as a destructive effect of a compound on a cell’s genetic material (DNA, RNA) affecting its integrity. Genotoxins are mutagens; they can cause mutations. Genotoxins include both radiation and chemical genotoxins. Jojoba oil (Simmondsia chinensis) is the most important of the commercially available products of jojoba for organic farming and medicines. Jojoba is unique in that, unlike most other vegetable oils, it closely resembles sebum, a waxy substance produced by our skin glands, so it can act as a natural skin conditioner. It has nearly replaced animal fats in the manufacture of skin lotions and creams. The genotoxic substance invades the nucleus and causes damage to the nucleic acid. This changes caused can be viewed by DNA fragmentation. This study is to analyse the genotoxicity of jojoba oil on oral cancer cell line by DNA fragmentation. The genotoxicity of jojoba oil on oral cancer cell line was studied.

Keywords: Genotoxicity, jojoba oil, anti-cancer drug, oral cancer.

INTRODUCTION

Genotoxicity refers to the property of a chemical agent which can alter the genetic information of an organism that can cause mutations which may lead to cancer. All mutagens are genotoxic. The genotoxic ability is hindered or prevented by the DNA repair mechanism or apoptosis.

To understand the genotoxic activity of various chemical agents, the scientists conduct biological assays in the DNA which is exposed to various toxic substrates.

Genotoxicity and mutagenicity testing are an important part of the hazard assessment of chemicals for regulatory purposes. The mutagenicity of a compound cannot be assessed by a single assay system. For this reason, the experts have been attempting to suggest a strategy to better investigate the genotoxic potential of the jojoba oil products taking into consideration the needs of the cancer medicine industry.

To assess genotoxicity, different endpoints must be taken into considerations: beside point mutations induction, a compound can induce changes in chromosomal number (polyploidy or aneuploidy) or in chromosome structure (breaks, deletions, rearrangements). However, aneuploidy can arise as a result of both genotoxic and non-genotoxic events, since loss of chromosomes can be caused either by direct effects on the chromosome to produce an acentric fragment or by interference with the site of attachment of the chromosome on the spindle.

At the early testing stages, the genotoxicity assays for predicting potential heritable germ cell damage are the same as used for predicting carcinogenicity because the endpoints measured in genotoxicity tests are common precursors for both of these adverse health outcomes.

Jojoba oil is the liquid produced in the seed of the Simmondsia chinensis (Jojoba) plant, a shrub, and the oil makes up approximately 50% of the jojoba seed by weight and is native to southern Arizona, California and north-western Mexico.

Jojoba wax refers to the raw hydrocarbon extracted from the seeds and is comprised mostly of triglyceride esters, hydrolysis of this (with the removal of glycerol) leaves Jojoba oil, which is composed mostly of free fatty acids.

The oil has a higher cloud point and better cosmetic properties. Unrefined jojoba oil appears as a clear golden liquid at room temperature with a slightly nutty odour.

When refined it appears, colourless and odourless. Melting point is approximately 10°C.

Jojoba oil is used as a replacement for whale oil and its derivatives such as cetyl alcohol.

DNA fragmentation is the separation or breaking of DNA strands into pieces which can be done intentionally by laboratory personnel or by cells, or can occur spontaneously.

Spontaneous or accidental DNA fragmentation is fragmentation that gradually accumulates in a cell. Leukemic cells have been shown to generate several classes of DNA fragments after treatment with cytotoxic cancer chemotherapy agents.

However, it is unclear which of these fragmentation events are direct effects of DNA-damaging chemotherapy.

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agents and which fragmentation events are caused by downstream processes, such as apoptosis.

**MATERIALS AND METHODS**

Chemicals used was procured from Himedia. Cell line was purchased from ATCC.

**Maintenance of Cell Lines**

The oral cancer cell lines i.e., KB (ATCC CCL-17) were acquired from ATCC. Oral cancer cells were seeded in 24 well plate and kept in CO2 incubator. Cells were treated with the jojoba oil in three different concentrations (100 µl, 200 µl, 300µl) for 24 hrs. Treated cells were subjected to DNA fragmentation assay according to Alexei G.9

**Isolation of Genomic DNA**

1*106 cells were incubated with 100µl of cell lysis buffer at room temperature for one hour.

This was centrifuged for 15 min at 3000rpm at 4°C to sediment the cell debris. To the supernatant equal volume of phenol: chloroform: isoamylalcohol mixture was added to the supernatant and mixed well. This was centrifuged at5000 rpm for 15min. The supernatant was transferred to new tube.

The 3rd step was repeated once. To the final aqueous phase 40µl of 3.5M ammonium acetate was added, to this ice cold isopropanol was added to precipitate the DNA. This was incubated at -20°C for 1hour, followed by the centrifugation at10000 rpm for 15min. The pellet was retained and washed with 70% ethanol and stored in 20-50µl of TE buffer. The samples were analysed in 2% agarose gel stained with Ethidium bromide.

**Analysis of DNA Fragmentation by Agarose Gel Electrophoresis Method**

The extracted DNA is loaded to Agarose gel with the loading dye, DNA fragments was visualised under UV transilluminator.

**RESULTS**

![Figure 1: Gel showing the DNA Fragmentation in lanes 1, 2 and 3.](image)

Lane 1 – DNA from KB cells treated with 100µl sample
Lane 2 – DNA from KB cells treated with 200 µl sample
Lane 3 – DNA from KB cells treated with 300 µl sample

DNA fragmentation was observed with all the three concentrations of jojoba oil on oral cancer cell lines by agarose gel electrophoresis method. Apoptosis has been characterised biochemically by the activation of a nuclear endonuclease that cleaves the DNA into multimers of 180-200 base pairs and can be visualised as an ‘oligosomal ladder’ by standard agarose gel electrophoresis.10 This proves that jojoba oil shows genotoxicity on the oral cancer cells by degrading its DNA. Hence jojoba oil has the potential to be an anti-cancerous drug.

**DISCUSSION**

Genotoxicity is a word in genetics defined as a destructive effect on a cell’s genetic material (DNA, RNA) affecting its integrity. Genotoxins are mutagens; they can cause mutations. Genotoxins include both radiation and chemical genotoxins. A substance that has the property of genotoxicity is known as a genotoxin. All mutagens are genotoxic, but not all genotoxins are mutagens as they may not cause retained alterations in DNA sequence. A genotoxic agent is a chemical or another agent that damages cellular DNA, resulting in mutations or cancer. Toxic to the genome. Genotoxic substances are known to be potentially mutagenic or carcinogenic when inhaled, ingested or penetrate the skin. The drug used in chemotherapy to kill cancer cells depends on its ability to halt cell division. Usually, cancer drugs work by damaging the RNA or DNA that tells the cell how to copy itself in division. If the cancer cells are unable to divide, they die. Chemotherapeutic techniques have a range of side-effects that depend on the type of medications used. The most common medications affect mainly the fast-dividing cells of the body, such as blood cells and the cells lining the mouth, stomach, and intestines. Chemotherapy-related toxicities can occur acutely after administration, within hours or days, or chronically, from weeks to years.11 Thus the property of genotoxicity is explored in herbs as it is safe. Herbs possessing genotoxicity property will act only on the cancerous cells leaving the normal cells safe. Herbs with genotoxicity can be tested for its anti cancer property.

**CONCLUSION**

From the above experiment and research its evident that jojoba oil has all the vitality to treat oral cancer12 jojoba oil is the most commercially available product of *Simmondsia chinensis* and is easily available in the market. Though research is still going on various parts of the world to make use of this plant extract to treat cancer, oral-cancer in specific, there is less awareness among the masses.13 In future the phytochemical properties of Jojoba oil may be used to design anti-cancer drugs. Also the medicinal property of the various natural
herbs should be explored because than the other chemotherapeutic drugs, they don’t affect the normal and healthy cells and they don’t cause any side effects. 14

REFERENCES


