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Essential Oils as an Alternative to Inhibit the Growth of Lung Cancer A549 Cell Lines

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ABSTRACT

Lung cancer is the leading cause of cancer-related deaths worldwide and ranked in the top 10 of newly reported cases. Due to the fact that cancer manifests with the most common symptoms, such as cough, it is always discovered late. Some essential oils are viable medicinal sources due to their extensive history of usage in traditional medicine. This study aims to explore the efficacy of five possible essential oils, namely Clary sage, Frankincense, Marjoram, Myrrh, and Thyme, as a halal alternative therapy for lung cancer against the lung cancer A549 cell line. The evaluation of the cytotoxicity for the essential oils was done by MTT-assay test. According to these studies, Frankincense essential oils showed the most potential in suppressing the growth of cancer cells among the essential oils examined. It had the highest inhibition of cell growth, with 93.9% cell proliferation, compared to the others.

Keywords: Lung cancer; Halal pharmaceutical; Essential oil; Clary sage; Frankincense; Marjoram; Myrrh; Thyme; A549

1 Introduction

Lung cancer is the leading cause of cancer-related fatalities worldwide, following prostate cancer (for males) and breast cancer (for females). 82% of lung cancers are classed as non-small cell lung cancer (NSCLC), whereas 14% are classified as small cell lung cancer (SCLC) [1]. This category is further subdivided into adenocarcinoma, squamous cell carcinoma, and large cell carcinoma [2]. Overall, the relative 5-year survival rate for lung cancer is 22% (18% for males and 25% for women), 26% for NSCLC, and 7% for SCLC. Only 24% of lung cancers are detected at a localised stage, with a 60% 5-year survival rate. Symptoms typically do not present until cancer has reached an advanced stage, including chronic cough, blood-streaked sputum, chest pain, a raspy voice, increased shortness of breath, and recurrent pneumonia or bronchitis are among the symptoms [1].

Recently, there has been much focus on using essential oils (EOs) in pharmacological and therapeutic applications after it was found that they had chemopreventive potential. The evidence for this comes from the EO's capacity to cause apoptosis in various tumour cell lines.



Several EOs have impressive antibacterial activity against both facultative and obligate anaerobic bacteria. The antioxidant and anticancer properties of EOs isolated from traditional medicinal herbs were also found to be due to these ingredients [3]. The antibacterial and antiviral activities of these EOs, as well as their anticancer, antibiotic, anti-inflammatory, antioxidant, and antiviral abilities, have been extensively documented in a wide range of scientific literature [4]. Clary sage (*Salvia sclarea*) EOs show cancer cell growth inhibitory and antioxidant activities [4]. Frankincense (*Boswellia sacra*) EOs induced apoptosis to lung cancer A549 cell line [5]. Marjoram (*Origanum majorana*) EOs suppressed the proliferation of human HT-29 colorectal cancer cells and their ability to form new colonies [6]. Myrrh (*Commiphora myrrha*) EOs shows cytotoxicity against breast cancer [7]. Thyme (*Thymus vulgaris*) EOs induced a dose-dependent inhibition of cell proliferation [8]. This research aims to study the potential essential oils as a halal alternative therapeutic source for lung cancer.

2 Materials and Methods

2.1 Essential oils

Clary sage (*Salvia sclarea*), Frankincense (*Boswellia sacra*), Marjoram (*Origanum majorana*), Myrrh (*Commiphora myrrha*), and Thyme (*Thymus vulgaris*) EOs were obtained from Young Living, US.

2.2 Cancer cell cultivation

The lung cancer A549 cell line was cultivated in RPMI 1640 medium (Gibco) supplemented with 10% fetal bovine serum (FBS) (Gibco) at 37 °C in a humidified 5% CO₂ atmosphere. For subculturing, cells were harvested after accutase/EDTA treatment at 37°C. Cells were used when monolayer confluence had reached 75% [9].

2.3 Cell proliferation assay

All formulations were evaluated by MTT cytotoxic assay [5] using lung cancer A549 cells. The cells were seeded in 96-well plates at 1×10^4 cells/well density and incubated for 24 hrs. Then, the cells were treated with 4 mg/ml of EOs (Clary sage, Frankincense, Marjoram, Myrrh, and Thyme) for about 48 hrs. On the other hand, control cells were treated with DMSO, which was incorporated into the medium. After that, the cells were incubated with 1 mg/ml of MTT reagent at 37°C for 4 hrs, and then, it was discarded. The formed formazan crystals were dissolved using 100 ml of DMSO, followed by incubation and shaking. Finally, colourimetric analysis using a multiplate reader measured absorbance at 570 nm. The cell proliferation (%) was calculated and compared with the control.

3 Results & Discussions

3.1 Assessment of the cytotoxicity

The cytotoxic effect of five different types of EOs, including Clary sage, Frankincense, Marjoram, Myrrh, and Thyme, was investigated in the lung cancer A549 cell line. The

cytotoxic effect in the present study was measured and represented in one way: cell proliferation percentage (Table 1 and Figure 1). A discussion and a justification of the outcomes of this evaluation was presented with the presentations of those results.

Table 1: Cell proliferation (%) values for each essential oil (EO) towards lung cancer cell A549 cell line

Essential oils	Cell proliferation (%)
Clary sage (<i>Salvia sclarea</i>)	88.7
Frankincense (<i>Boswellia sacra</i>)	93.9
Marjoram (<i>Origanum majorana</i>)	86.6
Myrrh (<i>Commiphora myrrha</i>)	84.2
Thyme (<i>Thymus vulgaris</i>)	93.5

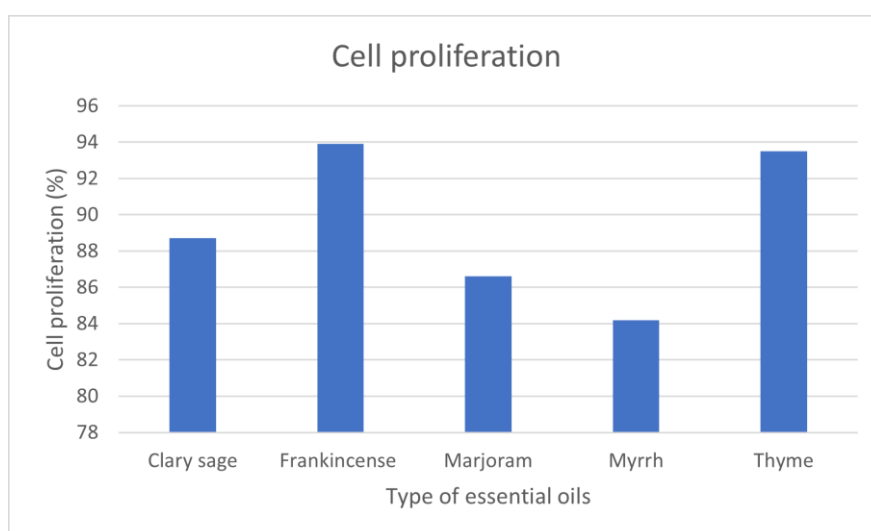


Figure 1: The graph for cell proliferation (%) of all five types of essential oils (EOs) against lung cancer A549 cell line.

3.2 Cytotoxicity of essential oils

As shown in Table 1 and Figure 1, Frankincense and Thyme EOs showed the highest percentage of cell proliferation, which was 93.9% for Frankincense and 93.5% for Thyme. It was shown that both EOs managed to inhibit a high number of cells, indicating that they have very good cytotoxicity. The other three EOs exhibited strong inhibitory effects. Clary sage EO inhibited approximately 88.7% of the enzyme, Marjoram EO inhibited approximately 86.6%, and Myrrh EO inhibited approximately 84.5%. After treatment with EOs, the cells were inhibited with a positive result. The fact that the proportion of inhibited cells was greater than 80% demonstrates that they had a good cytotoxic impact.

4 Conclusions

The cytotoxic effect of five different types of essential oils, including Clary sage, Frankincense, Marjoram, Myrrh, and Thyme, was investigated in the lung cancer A549 cell line. The cytotoxic effect seen in this study's present in vitro model, five different kinds of essential oils, including Clary sage, Frankincense, Marjoram, Myrrh, and Thyme, were tested against the lung cancer A549 cell line to see if any of them had an effect. Consequently, Frankincense

essential oils had the highest level of cell growth inhibition, with 93.9% cell proliferation. Products derived from plants are quickly becoming recognized as one of the most promising halal product choices. Essential oils created from plants have sparked the curiosity of numerous experts; they have been demonstrated to treat various ailments since antiquity. According to this research, using essential oils as a treatment for several types of cancer, notably lung cancer, was successful. Essential oils are not only able to serve as a halal substitute for pharmaceutical items, but they are also able to assist in the complementation of chemical products that could have potentially negative impacts on consumers. It is necessary to conduct additional research, including clinical trials, to enhance the treatment's efficacy.

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