

In vitro Effects of *Ganoderma lucidum* Extract on Breast Cancer Cells

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Abstract--- *Ganoderma lucidum*, an oriental fungus, has a long history of use for promoting health and longevity. The aim of this study was to determine the effects of *Ganoderma lucidum* extract on MCF7 line cell (breast cancer cell) viability in cell culture. In this laboratory experimental study, we used MTT assay to determine cell viability following administration of different doses of *Ganoderma lucidum* extract in cell culture. The data were statically analyzed using ANOVA. The results showed that the administration of 10 µg/ml and 100 µg/ml of *Ganoderma lucidum* extract resulted in decreased viability of MCF7 cells ($P < 0.05$), however, administration of 1 mg/ml *Ganoderma lucidum* extract resulted in increased viability of MCF7 cells ($P < 0.05$). Our findings indicate that appropriate doses of *Ganoderma lucidum* extract have antiproliferative effects on breast cancer cells.

Keywords--- *Ganoderma lucidum*, MCF7 Cell Line, Viability.

I. INTRODUCTION

GANODERMA lucidum (Reishi), a basidiomycetous fungus, is widely used in China and other Asian countries to treat various human diseases, such as hepatitis, hepatopathy, hypertension, nephritis, and cancers [1]-[3]. In addition, many bioactive components isolated from *Ganoderma lucidum* have been demonstrated to possess antioxidative, antihypertensive, and anticancer effects [4],[5]. For example, polysaccharides from *Ganoderma lucidum* exert anticancer effects against HL-60 and U937 leukemic cell lines [6]. Some of the triterpenes isolated from *Ganoderma lucidum* also exhibit cytotoxic activity against mouse sarcoma and mouse lung carcinoma cells in vitro [7].

One third of all newly diagnosed cancers among women are breast cancers [8]. MCF-7 is a breast cancer cell line isolated in 1970 from a 69-year-old Caucasian woman. MCF-7 is the acronym of Michigan Cancer Foundation-7, referring to the institute in Detroit [9]. Because breast cancer often progresses from the therapy-responsive phenotype to the highly invasive and metastatic phenotype, breast cancer is the second leading cause of cancer death in the U.S. female population [10]. A comprehensive review by the World Cancer Research Fund and the American Institute of Cancer Research clearly demonstrates the importance of nutrition in the prevention of cancer, which could also contribute to the

low incidence of breast cancers among Asian women [11]. Therefore, it was suggested that some nutritional products have chemopreventive and therapeutic effects against cancer. These anticancer effects also significantly increased the popularity of dietary supplements in cancer patients [12].

Despite considerable reports on inhibitory effects of *Ganoderma lucidum* on cancer cells, there is not considerable report on the effects of Iranian *Ganoderma lucidum* extract on cancer cells, particularly MCF7 line cell. The main aim of this study was to determine the effects of *Ganoderma lucidum* extract on breast cancer cells.

II. MATERIAL AND METHODS

A. Extract preparation

Ganoderma lucidum was prepared and different concentrations of extract (10 µg/ml, 100 µg/ml, 1mg/ml and 10mg/ml) were used in our study.

B. Protocol of Study

We used MTT assay in this work to determine the effects of ganoderma extract on vero cells viability in cell culture. Briefly, the procedure was carried out in the following steps:

DAY ONE: 100 µl of cells (15000 cells) was added into each well (96 well plate) and incubate at 37 with 5% CO_2 overnight.

DAY TWO: The media was removed and extract was added and incubated at 37 with 5% CO_2 overnight. For control 10% FBS was added to media.

DAY THREE: extract was removed from media. 20 µl of 5 mg/ml MTT was added to each well and incubated for 4 hours at 37°C. 150 µ isopropanol was added and covered with tinfoil and agitate cells on orbital shaker for 15 min. Absorbance was read at 570 nm with a reference filter of 630 nm and recorded.

C. Statistical Analysis

Statistical significance was evaluated by one-way analysis of variance (ANOVA) using SPSS 19. Significance was measured using Turkey's test. Differences with $P < 0.05$ were considered significant

III. RESULTS

Figure I represents viability of MCF7 cells in response to different doses of ganoderma extract.

Our results show that administration of 10 and 100 µg/ml, and also 10 mg/ml of ganoderma extract have anti-proliferative effects on MCF7 cell line, however, administration of 1mg/ml of ganoderma extract resulted in decreased MCF7 cell proliferation.

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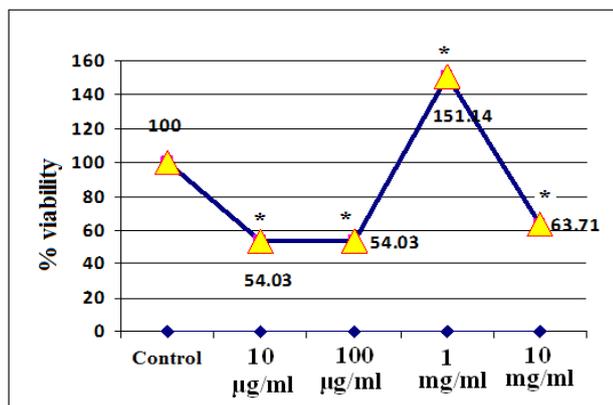


Fig. 1 Viability of MCF7 cells in response to different doses of *Ganoderma lucidum*. * represents significant difference compared with control group.

IV. DISCUSSION

In our study, we reported the dose dependent inhibitory or excitatory effect of *Ganoderma lucidum* extract on cell viability of MCF7 cell line. *Ganoderma lucidum* is a popular edible mushroom, the dried powder of which has been used in traditional Chinese medicine in many Asian countries. We have previously reported that spores or the fruiting body of *Ganoderma lucidum* downregulated the expression of uPA and uPAR, which resulted in the inhibition of cell migration of highly invasive human breast and prostate cancer cells [13]. Here we show that *Ganoderma lucidum* inhibits the growth of breast cancer cells, causes cell cycle arrest at G2/M phase, and induces apoptosis. Different biologically active compounds with possible anticancer activities were recently isolated from *Ganoderma lucidum*. For example polysaccharides from *Ganoderma lucidum* demonstrated activation of the immune response through the stimulation of production of interleukin-1 β (IL-1 β), IL-6, tumor necrosis factor- (TNF-), and interferon- γ (IFN- γ) by macrophages and T-lymphocytes [26]. Triterpenes were cytotoxic for hepatoma and lung carcinoma cells [14],[15], phenols demonstrated antioxidant properties [16], and lipids inhibited the growth of hepatoma and sarcoma in vivo [17]. Although the identification of biologically active components of *Ganoderma lucidum* is important for the mechanistic characterization of their specific activity, some of these components demonstrated cytotoxicity (14,15). In addition, there is some evidence that certain components in the natural herbal products can reduce the cytotoxicity of the whole product, and the interaction between different biologically active components is responsible for their in vivo effects [18]. In the present study, we show that *Ganoderma lucidum* inhibits the growth of breast cancer cells by the cell cycle arrest at G2/M phase. Since aberrantly active cell cycle regulatory proteins are responsible for the uncontrolled growth of cancer cells, these proteins are suitable therapeutic targets. However, other studies have shown that alcohol extracts of *Ganoderma lucidum* can arrest the cells at G1/G0 phase in cervical and breast cancer cells [19], [20]. Therefore, it is possible that specific cancer cell lines respond differently to *Ganoderma lucidum*. The inhibition of proliferation of breast cancer cells by *Ganoderma lucidum* might also be caused by the induction of apoptosis. Apoptosis is a physiological process by which cells are removed when

an agent damages their DNA [21], and the inhibition of apoptosis, rather than enhanced cell proliferation, is the critical factor that contributes to the development of cancer [22], [23]. Therefore, apoptosis can be considered an ideal way to remove cells [24], [25]. Briefly, our data demonstrate that *Ganoderma lucidum* inhibited the growth of human breast cancer cells. The biological effects of *Ganoderma lucidum* are mediated by the inhibition of multiple signaling pathways. Additional in vivo studies are necessary to establish *Ganoderma lucidum* as a potential agent for the prevention and/or treatment of breast cancer.

V. CONCLUSION

We have shown that appropriate dose of *Ganoderma lucidum* extract has anti-proliferative effects on MCF7 cancer line cells in cell culture, indicating the potential power of *Ganoderma lucidum* extract in treatment of breast cancer cells.

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