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EFFECT OF EXTRACT MILK THISTLE AND EUPHORBIA MACULATA ON THE BREAST CANCER CELL LINE MCF-7

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ABSTRACT

Cancer has abnormal cell growth in the body that can lead to death. For thousands of years, plants have been used to cure many diseases. Given the prevalence of breast cancer in Iran, the ascending tilt of this malignancy over the past two decades, high mortality rates and on the other hand, with regard to oral intake, cheap and easy access by the general public to the extract of the milk thistle the effectiveness of this herbal product was investigated on MCF-7 breast cancer cells. Also, in recent decades, scientists have used natural compounds to treat cancer, including raw vegetable extract or a combination of different phytochemicals. Study on cytotoxic activity of raw ethanol extracts from the plant Euphorbia maculate strain Euphorbiacea on the MCF-7 cell line. One of the most valuable biological activities in these species is anti-cancer. In summary, the results of the research confirm the effectiveness of these plants against breast cancer and given the nature of the product, its low cost and public access, entering this herbal supplement in the diet may be effective in treating breast cancer. Based on the results of the extract of E. maculata L. Has a poor inhibitory effect on MCF-7 cells.

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Introduction

Breast cancer is the most common neoplastic malignancy of women in the world And it is a major cause of death from cancer in women. (1-2) Although the annual outbreak of this malignancy is increasing in the world, however, its prevalence has been reported differently in different countries.(3) Although the highest prevalence of this malignancy is for developed countries, but research shows that the incidence of breast cancer incidence is higher in developing countries the average life expectancy of these patients is lower (4). Studies have shown that the prevalence of breast cancer in Iran is lower than in developed countries, However, this malignancy is still the most common cancer in Iranian women available information indicates the prevalence of this malignancy in Iran over the past two decades (5). One of the treatment options for this malignancy can be surgery, chemotherapy, and radiotherapy. However, mortality rates in these patients are high, which itself indicates the ineffectiveness of these therapeutic approaches. One of the main causes of the incidence of cancers is the influence of environmental factors on the development of mutations and genetic changes responsible for the onset of malignancies (6). It has been shown that some food products have an effective role in preventing cancer due to their antioxidant properties (7). These products include silymarin; a polyphenolic flavonoid extracted from the milk thistle and about 90% of it is a combination of silibinin (8). Silibinin is an antioxidant compound which prevents the liver from acute and chronic damage. It is also noted that this compound is effective in lowering blood cholesterol levels and enhancing the immune system (9). Studies conducted over the past years have shown that silibinin has anti-cancer effects against lung cancer cells (11-10), glioblastoma (12), skin (13), bladder (14), liver (15), prostate (17-16), and clone (18-19). Today, the extract of milk thistle is widely used as a very effective dietary supplement in the United States and European countries. Given the prevalence of breast cancer in Iran, the ascending tilt of this malignancy over the past two decades, high mortality rates and on the other hand, with regard to oral intake, cheap and easy public access to this herbal product, we review the effectiveness of Silibinin in breast cancer cells. Cells (MCF-7) Human Cell Line Breast Cancer, the cell line is very suitable for in vitro studies of breast cancer and for the first time in 1970, 69-year-old female breast cancer patients were isolated.

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milk thistle

Although much progress has been made in clinical and basic research and has led to a high rate of cure for many diseases, but cancer remains the second leading cause of death after cardiac abnormalities in developed and developing countries. Due to the problem of microbial resistance to chemical drugs and their unwanted side effects, over the past decade, the use of natural medicines has become of global importance. Among the human diseases that have been tried with natural medicines, cancer, which seems to be the most important genetic disease. Plants are one of the main sources of biologically active substances that over the past decade have become increasingly important for the development of natural remedies for cancer treatment (20). The use of medicinal plants is as old as human life, since diseases have been born with the advent of mankind, and several thousand years of documents in the history of medicine and medicine contain the valuable experiences and knowledge of medicinal plants. For the last few decades, what was used as a medicine was obtained from natural sources, mainly from plants. On the one hand, with the advent of science on the one hand, and economic issues, on the other hand, the use of medicinal plants was reduced in the past, and chemical drugs in many cases replaced the plants. The experience of recent decades has shown that chemical drugs with many efficiencies have many harmful and adverse effects. Today, it has been proven that there is less pure matter that has no harmful effects. For this reason, in recent decades, the return to the use of medicinal plants has been a matter of great interest and universities, research centers, factories and the World Health Organization Widespread use of medicinal plants has been proposed and the role of medicinal plants in the 21st century is considered crucial. Some of the medicines and especially the anticancer effects.Frafion species have been studied in the world, but about the impact of E. Maculata L. There is no research on available breast cancer cells. This plant is from the category of Angiosperms, Malpighiales, Euphorbiaceae family, Euphorbia genus and E. maculata L. species. This is a one-year-old native to North America. In Iran, this species was collected by Naseh et al. (2006) from Mazandaran province, the city of Noshahr, and was first reported in Iran and Iran in the Flora Iranica region. Different species of Euphorbia, including plant species, have a long history of traditional drugs. Today, many Euphorbia species are also used as drugs in many drug delivery systems (21). Therefore, due to the compounds present in available sources, the purpose of this research is to review the effects of cytotoxicity of E. maculata L. ethanolic extract on MCF-7 cancer cells.

Maculata

Euphorbiaceae is one of the great species of two-leaved nematodes with 300 genera and about 5000 species. These herbaceous plants are often laced with shrubs, with simple leaves, alternate. The chemical properties of Euphorbia species are very interesting and diverse among the flowering plants families. The main secondary metabolites found in Euphorbia species can

be classified as lipids and derivatives, terpenes, aramytic compounds, amines, alkaloids, and amino acids. There are many studies that measure these secondary metabolites in these species. For example, it has been reported that E. hiberna seeds contain 33% oil and fatty acids such as stearic, myristic, palmitic, linoleic and oleic acids. E. lathyris and E. chrcias were also reported to contain 30 and 27% fatty acids, respectively. Simple and also very complex alkaloids have been discovered in the family Euphorbiaceae. Alkaloids are nitrogen compounds found in plants. They form salts with plant acids, and when they are combined with sugars, they form glycoalkalohydes. Two alkaline Yorasil and Uridine, separated from E. altotibetic (22). Flavonoids have been reported in members of the Euphorbiaceae family. In addition, terpenes are a group of molecules that are made up of isoprene units. There are reports that there are a wide variety of Diterepen in Euphorbiaceae. The plants of the Euphorbiaceae family are rich in terpenes. Triucllol and euphorbol are often found in the Latex members of the genus Euphorbia (23). Several steroidal estrogens from E.chamaesyce were also extracted. A new steroid called geniculatoside F was obtained from E. geniculata Linn (24). There are reports of various sugars in these plants. From E. pulcherrima, glucose and arabinose were extracted and sucrose and glucose from E. roots. Ebracteolata and E. paralias were also reported, and the E. caudicifolia seed contains free sugars (23). The anticancer role of E. maculata on the MCF-7 cell line is the use of medicinal plants for human life, since diseases have been born with the advent of mankind, and several thousands of years of history in medicine and medicine with experiences and the valuable information of medicinal plants. One of the human diseases that have been tried to treat natural medicines is cancer, which seems to be the most important genetic disease. Plants are one of the main sources of biologically active substances that, over the past decade, have become increasingly important for the development of natural remedies for cancer treatment (25). There are various reports about the chemical composition of the E. maculata L. species. For example, Matsunaga et al. (1988) extracted the tripletpine and cytosterol extract by examining E. maculata L. extract (26). Amakura et al. (1997) also used tannins in E. maculata L., a medicine used to treat injuries in Mexico and China, as well as anti-diarrhea in China, and tested a new dimeric hydrolyzed tannin compound, Eumaculin A was extracted (27). Further studies on the polyphenolic components in the leaf extract of this plant confirmed the presence of euphorbin A and its analogues, B euphorbin and D (28). In the case of the effects of the drug, and in particular the anticancer, some Frafion species have been studied in the world, But about the impact of E. Maculata L. There is no research on available breast cancer cells. This plant is from the category of Angiosperms, Malpighiales, Euphorbiaceae family, Euphorbia genus and E. maculata L. species. This is a one-year-old native to North America. Therefore, due to the compounds present in the available sources, the aim of this study was to investigate the effects of cellular toxicity of E. maculata L. ethanolic extract on MCF-7 cancer cells.

Discussion and conclusion

Breast cancer is the most common female malignancy in the world and the prevalence of this malignancy in most countries in the world, including Iran, is increasing. Among the causes associated with an ascending incidence of cancer, environmental factors such as air pollution, stress, life patterns and diet of individuals can be mentioned. It has been shown that the use of antioxidant foods plays an important role in preventing and reducing the risk of cancers (29-30). On the other hand, despite the use of therapeutic approaches such as surgery, chemotherapy, and radiotherapy, mortality rates are still high in patients with cancer, which itself indicates the inefficiency of these therapeutic approaches. In addition, the destructive effects of chemotherapy and radiation therapy on divided normal cells are among other disadvantages associated with these therapeutic processes. (31) Therefore, considering the above points, the tendency to pay attention to the use of natural products and food supplements has increased the anti-cancer effect in recent years. Among the nutritional supplements to be considered in this regard, we can mention the extract of milk thistle, which has recently been studied on the anticancer effect of this herbal product. Recently, Momeni and colleagues have shown that silibinin, which is the most important part of the extracts of milk thistle, The invasive properties of glioblastoma cells are reduced by inhibiting cathepsin B (32). In a study by Zhang, it was found that silibinin inhibits the growth of gastric cancer cells of SGC-7901 by reducing the expression of p34cdc2 protein. (33) In addition, Kunatz also showed that silibinin enhances apoptosis induced by TRAIL in intestinal adenocarcinoma (34). Other studies also suggest an antitumor effect of silibinin against lung, bladder, prostate, and clone cancer cells (35-36) In line with the results of other studies, the results of the study also confirm the efficacy of this herbal drug against breast cancer cells. The results of this study showed that the treatment of MCF-7 cancer cells by silibinin is dependent Dose and time reduce the proliferation of these cells. Apoptosis is a highly regulated process that plays a very important role in maintaining hemostasis in multicellular organisms (37). It has been shown that apoptosis is controlled by many intracellular and extracellular factors; among the intracellular factors, the balance between Bcl-2 (apoptotic inhibitor) and Bax (pro-apoptotic equivalence (Bcl-2 as the most important determinant of fate) The cell is introduced in response to an extracellular stimulus (38). The Bax protein acts as a key protein in apoptosis induced by various factors in the internal pathway of apoptosis. This protein, through interaction with mitochondrial membrane proteins, increases the mitochondrial membrane penetration and releases cytochrome c from mitochondria and activates caspases and ultimately apoptosis. On the other hand, Bcl-2 has an anti-apoptotic effect in response to various stimuli of apoptosis by preventing the release of cytochrome c from mitochondria. For this reason, in order to investigate the effect of silibinin on cell death in MCF-7 cells, protein expression and mRNA expression of Bax and Bcl-2 genes were measured after cell treatment. The results show that Silibinin significantly increases the expression of the expression of the Bax mRNA expression, While it did not affect the amount of Bcl-2 gene expression. Also, as expected, the expression level of Bax parietal increased significantly with the result of mRNA expression, but did not change in the amount of Bcl-2 protein. In summary, the results of the research confirmed the pro-apoptotic properties of silybin and the effectiveness of this herbal supplement against the MCF-7 breast cancer cell line. Natural, low cost and public access to this product are among the benefits. According to the results, it seems that the use of this herbal supplement in the diet may be effective in reducing the incidence of breast cancer. The results of this study showed that E. maculata L. has a weak anticarcinogenic effect on the breast cancer cell line. Different studies have shown that many species of this genus have anticancer effects. This anticancer therapeutic effect of Euphorbia species can be attributed to the presence of a large number of secondary metabolites in these species, because E. maculata L. is rich in valuable drug compounds, including secondary metabolites of triplepenoids, B-propenoids, flavonoids, phenolic compounds, and tannins. The therapeutic effects of these

compounds have been reported in various studies. Matsunaga et al. (1988) extracted the tripletpine and cytosterol extract by examining E. maculata L. extract (3).

Table. Effect of Different Concentrations of E. maculata L. on Optical Absorption and Inhibition of Growth of MCF-7 Cells

Extract concentration E. maculata L.) mg/ml(	***Absorption rate***	***Growth inhibition (%)***	***IC50 (mg/ml)***	***P-value***
0/156	0/039* ± 0/011	33/54		0/012
0/312	0/047* ± 0/016	32/02		0/021
0/625	0/052* ± 0/026	23/30		0/012
1/25	0/079 ± 0/017	16/45		0/170
2/5	0/112 ± 0/050	10/91	2/41	0/494
5	0/157 ± 0/041	12/60		0/273
7/5	0/115 ± 0/012	4/826		0/153
10	0/243 ± 0/065	0/7064		0/714
Control	0/095 ± 0/003	-		-
Blood	0/251 ± 0/099	-42/62		0/076
DMSO	0/042* ± 0/017	22/08		0/017

*** The number of stars had a significant difference with the control group ($p \leq 0.05$).

Flavonoids have been reported in members of the Euphorbiaceae family. Some investigators have reported on the mechanisms of their antioxidant effect on these compounds, which Euphorbia species can act as a cleanser of active oxygen species due to the presence of phenolic compounds and flavonoids, and ultimately can Have anticancer effects. Because flavonoids and phenolic compounds are one of the most important groups of secondary metabolites with antioxidant properties and kiln-like properties. For example, ethanolic extracts of E. acanthothamnos and E. macroclada exhibited higher antioxidant activity in contrast to the DPPH test (4). Jyothi et al. (2008) Aqueous extracts of Linn parts. E. antiquorum for its hepatic and anti-oxidant activity and confirmed that the extract exhibited a high antioxidant activity and reported in its mechanism of action that it was reduced by hydroxylic acid and activity Radical anion superoxide was applied. It has also been suggested that flavonoids not only increase the permeability of the capillaries, they may also prevent certain inflammatory processes. The usual mechanisms for anti-inflammatory drugs are to prevent the pathways of arachidonic acid metabolism. However, these mechanisms can also be activated by luteolin glycosides. In addition, other mechanisms such as inhibiting the release of histamine and cleansing of free radicals have also been reported for this compound (5). Patil et al. (2011) suggested that, unlike the EL4 cell line, Euphorbia hirta extract suggested that flavonoids extracted from this species may have a protective role in cancer through their effects on signaling in the proliferation and angiogenesis of the cell (6). Yang et al. (2008) described the phenolic acid composition of Gallic acid in E. helioscopia for the first time as an anti tumor composition in this species (7).

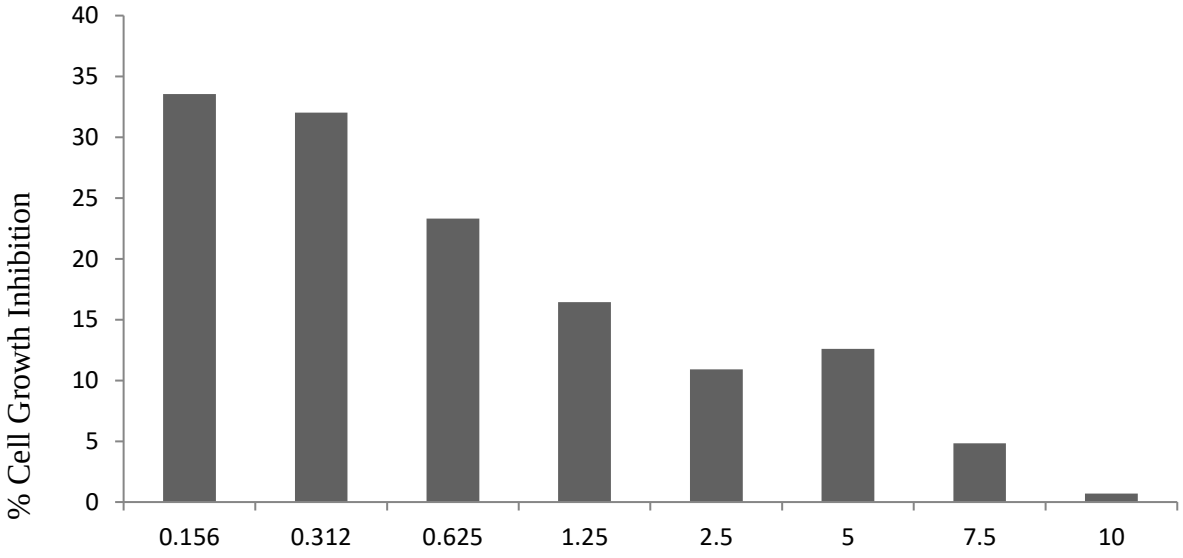

Chart. Cytotoxicity factor of E. maculata L. extract on MCF-7 cancer cell line

* Stars with star sign had a significant difference with the control group ($p \leq 0.05$).

According to the results of this experiment, E. maculata L. extract has marginal inhibitory effect on MCF-7 cells. Of course, since this is the first study in this area, more research is needed, and further studies are needed to isolate the plant's components, as well as review it on other cancerous lines, including the cervix and prostate in the future be done. Various studies have shown that many species of Euphorbia have anticancer effects. The results of this study showed that E. maculata L. had a weak anticarcinogenic effect on the breast cancer cell line. This anticancer therapeutic effect of Euphorbia species can be attributed to the presence of many secondary metabolites in these species, because E. maculata L. is rich in valuable pharmaceutical compounds, including secondary metabolites of triplepenoids, B-propenoids, flavonoids, phenolic compounds, and tannins. The therapeutic effects of these compounds have been reported in various studies. Some researchers, by studying the mechanisms of the antioxidant effect of flavonoids, reported that Euphorbia species, acting as phenolic compounds and flavonoids, can act as a cleanser for active oxygen species, and can ultimately Have anticancer effects. Because flavonoids and phenolic compounds are one of the most important groups of secondary metabolites with antioxidant properties and kiln-like properties. Jyothi et al (2008) Aqueous Extract airports parts of Linn. E. antiquorum for its hepatic and anti-oxidant activity and confirmed that the extract exhibited a high antioxidant activity and reported in its mechanism of action that it was reduced by hydroxylic acid and activity Radical anion superoxide was applied. It has also been suggested that flavonoids not only increase the permeability of the capillaries, they may also prevent certain inflammatory processes. The usual mechanisms for anti-inflammatory drugs are to prevent the pathways of arachidonic acid metabolism. However, these mechanisms can also be activated by luteolin glycosides. In addition, other mechanisms such as inhibiting the release of histamine and cleansing of free radicals have also been reported for this compound (8). In contrast to the EL4 cell line, Patil et al. (2011) suggested that flavonoids extracted from this species may have a protective role in cancer through their effects on signaling in the proliferation and angiogenesis of the cell (9). Yang et al. (2008) described the phenolic acid composition of gallic acid in E. helioscopia for the first time as an anti tumor composition in this species (10). According to the results of this experiment, E. maculata L. extract has marginal inhibitory effect on MCF-7 cells. Of course, since this is the first study in this area, more research is needed, and further studies are needed to isolate the plant's components, as well as review it on other cancerous lines, including the cervix and prostate in the future be done.

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