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ARAŞTIRMA MAKALESİ **RESEARCH PAPER**

The Anti-Proliferative Effects of *Ficus carica* Latex on Cancer and Normal Cells

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Abstract: The stems of unripe fig fruits or leaves contain a white liquid called fig latex. Fig. latex contains compounds that are effective against cancer cells. It is used for various purposes in folk medicine and has been proven to have superior pharmacological properties in modern studies. The industrial value of latex from a cosmetic perspective is increasing as well. In this research, the MTT test was used to see how fig latex from *Ficus carica* affected lung cancer (A549), breast cancer (MCF-7), triple-negative breast cancer (MDA-MB-231), and non-tumorogenic lung (MRC-5) cell lines. All cell lines were treated with various dilutions of fig latex ranging from 1/1500 to 1/12 for 48 hours. In cancer cells, it induced cell death in a concentration-dependent manner, and high doses caused a more serious cytotoxic effect. On the other hand, it showed less cytotoxicity in healthy cells than cancer cells. The IC50 values of Fig latex were found to be 1/26, 1/40, 1/45, and 1/7 for A549, MCF-7, MDA-MB-231, and MRC-5 cells, respectively. Furthermore, we acquired a high tumor specificity index for both breast and lung cancer cells, with a greater one for breast cancer cells compared to lung cancer cells. Based on our findings, fig latex showed selective cytotoxicity against cancer cells, making it a possible target for future cancer drug development. However, further research is necessary to fully understand the potential use of fig latex and its bioactive compounds in cancer treatment, particularly in terms of their functional properties.

Keywords: Breast cancer, fig latex, lung cancer, lung fibroblast, MTT.

Ficus carica Sütünün Kanser ve Normal Hücre Üzerindeki Anti-proliferatif Etkileri

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Öz: İncir sütü, olgunlaşmamış incir meyve veya yaprak saplarının içerisinde bulunan beyaz renkli bir sıvıdır. İncir sütü, halk hekimliğinde çeşitli amaçlar için kullanılmaktadır ve modern çalışmalarda üstün farmakolojik özelliklere sahip olduğu kanıtlanmıştır. Ayrıca incir sütünün kozmetik açıdan endüstriyel değeri gitgide artmaktadır. Mevcut çalışmada, *Ficus carica* (*F. carica*) türünden elde edilen incir sütünün akciğer kanseri (A549), meme kanseri (MCF-7), üçlü negatif meme kanseri (MDA-MB-231) ve tümörojenik olmayan akciğer (MRC-5) hücre hatları üzerindeki sitotoksik etkisi MTT analiziyle değerlendirildi. Tüm hücre hatları, incir sütünün 1/1500-1/12 arası değişen konsantrasyonlarıyla 48 saat boyunca muamele edildi. İncir sütü, kanserli hücrelerde konsantrasyona bağlı bir şekilde hücre ölüm oranını arttırdı ve yüksek dozlarında hücreler üzerinde daha ciddi bir sitotoksisik etkiye neden oldu. Sağlıklı hücrelerde ise kanser hücrelerine göre daha az toksik etki gösterdi. İncir sütünün IC_{50} değeri, A549 hücrelerinde 1/26, MCF-7 hücrelerinde 1/40, MDA-MB-231 hücrelerinde 1/45 ve MRC-5 hücrelerinde ise 1/7 olarak belirlendi. Meme kanseri ve akciğer kanseri hücrelerinde yüksek tümör spesifite değeri elde edildi. Bu değer meme kanseri hücreleri için daha yüksek bulundu. Bulgularımız, incir sütünün kanser hücrelerine karşı seçici sitotoksisiteye sahip olduğunu ve gelecekte kanserle ilgili ilaç araştırmalarındaki ilerlemelerde faydalı olabileceğini göstermektedir. İncir sütünün ve muhtevasındaki biyoaktif bileşiklerinin fonksiyonel özellikleri açısından kanser tedavilerinde potansiyel kullanımına dair bilimsel araştırmalara daha fazla ihtiyaç vardır.

Anahtar kelimeler: Akciğer kanseri, akciğer fibroblastı, incir sütü, meme kanseri, MTT.

INTRODUCTION

Fig. (*Ficus carica*, *F. carica*) trees are members of the Moraceae family. It originated in southwest Asia and is widely cultivated in the Mediterranean region. The world's leading fig producers are Turkey, Greece, Egypt, Morocco, Italy, Spain, Brazil, and Morocco (Soni et al., 2014). Bark, fruits, leaves, roots, and latex are valuable parts of the fig tree that are frequently used in folk medicine (Badgujar et al. 2014). In plants, latex is a milky, natural adhesive polymer extracted from different parts of the plant. In figs, the latex is milky white, which makes it commonly called "fig milk." Different metabolites found in fig latex include proteins, amino acids, fatty acids, minerals, vitamins, antioxidants, phenols, terpenoids, sterols, volatile components, and proteases (Castelli & Lopez 2022). In addition to the health benefits mentioned in alternative medicine, modern pharmacological studies have shown that fig latex can fight cancer, viruses, bacteria, parasites, high blood pressure, blood clotting, inflammation, and angiogenesis, and protect the liver (Upadhyay, 2011; Ghandehari & Fatemi, 2018; Almehmadi, 2023). These characteristics enable the use of fig latex as a therapeutic and preventative tool for various oxidative stress-related illnesses (Shiraishi et al., 2023). Furthermore, studies highlight the significant industrial potential of the bioactive components derived from fig latex (Shiraishi et al., 2023). As an alternative to animal enzymes, fig latex is utilized in the industry to produce cheese, natural rubber, and bioactive peptides and to soften meat (Rasool et al., 2023; Castelli & Lopez, 2022). According to Hegazy et al. (2023), the biological activities of fig latex are likely caused by flavonoids and terpenoids, which are types of secondary metabolites (Hegazy et al., 2023). These factors have led to a recent increase in the value of fig latex in the pharmaceutical and cosmetic industries (Teruel-Andreu, 2021). Therefore, the current research was conducted to assess the cytotoxic potential of fig latex derived from *F. carica* against several cell lines, including lung cancer (A549), breast cancer (MCF-7), triple-negative breast cancer (MDA-MB-231), and non-tumorigenic lung (MRC-5) cell lines.

MATERIAL AND METHOD

Chemicals and Cell Lines: The cell lines were obtained from the East Anatolia High Technology Research and Application Center (DAYTAM) (Atatürk University, Erzurum, Turkey). These cells are A549 (human lung carcinoma) (Cat. No. CCL-185™, ATCC®), MRC-5 (human lung fibroblast) (Cat. No. CCL-171™, ATCC®), MCF-7 (human breast cancer) (Cat. No. HTB-22, ATCC®), and MDA-MB-231 (triple negative breast cancer) (Cat. No. HTB-26, ATCC®). The analytically pure chemicals used in the study were all purchased from Sigma-Aldrich (St. Louis, Missouri, USA).

***Production of Fig Latex*:** The fig latex production process involved the harvesting of immature fruit stalks from *F. carica* trees in the Altınova region of Balıkesir, Turkey, during the summer months. The latex was collected drop-by-drop. We first placed the recovered fig latex in sun-protected glass bottles, then transferred it to 1 ml Eppendorf tubes and stored them at -80 °C until analysis. We identified the sample in the ethnobotany lab of the Faculty of Science, Atatürk University.

Cytotoxic Evaluation: The viability of the cells was assessed using the MTT (3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) assay, a versatile test commonly employed to monitor mitochondrial activity. First, high glucose Dulbecco's Modified Eagle Medium (DMEM) and Dulbecco's Modified Eagle Medium/Nutrient Mixture F-12 (DMEM/F12) media were supplemented with 10% fetal bovine serum (FBS), 100 IU/ml penicillin, and 100 µg/ml streptomycin. The final medium was used to cultivate cell lines A549, MCF-7, MDA-MB-231, and MRC-5. Every cell line was cultured in an incubator with 95% humidity, 5% CO_2, and a temperature of 37°C. One day before treatment, each cell line was uniformly seeded in 96-well plates at a density of 1×10^4 cells per well. The cells were then incubated in a CO_2 incubator. Fig latex was diluted with the complete medium at the following dilutions: 1/1500, 1/900, 1/900, 1/300, 1/180, 1/60, 1/36, and 1/12. We treated each cell line with varying concentrations of fig latex and then incubated it for 48 hours. 500 µM H_2O_2 served as the positive control. After 48 hours, each well received 20 µl (5 mg/ml) MTT containing 100 µl of complete media, and the cells were once again incubated in a CO_2 incubator at 37 °C for 4 hours. Following the incubation period, the supernatant was removed from the wells, and 150 µl of dimethyl sulfoxide (DMSO) was used to dissolve the formazan product, which appeared as blue-violet crystals. The absorbance of the formazan product secreted by live cells was quantified by an Epoch microplate reader (BioTek, USA) at a wavelength of 570 nm. The control group's absorbance was taken as 100%, and the percentage of viable cells was calculated by dividing the absorbance of the blank well by the absorbance of the control cell and multiplying the result by 100. Furthermore, the IC_{50} values were determined by fitting a curve with varying slopes, using the log of the latex concentration vs. the normalized response data. The software used for this analysis was GraphPad Prism version 6.00.

The tumor selectivity index of the fig latex was assessed by evaluating its cytotoxic effects on the lung fibroblast cell line (MRC-5). The specificity value of each cell was computed using the provided formula.

Tumor Specificity Index: IC_{50} value for MRC-5 cell / IC_{50} values for the specific cancer cells.

Statistical Analysis: The data are shown as mean ± S.E.M. We used one-way analysis of variance (ANOVA) and Tukey multiple comparisons in GraphPad Prism 6.0 (GraphPad, La Jolla, CA, USA) to identify variance differences. A significance level of $p<0.05$ was deemed acceptable for distinguishing the treatment groups from the control.

RESULTS

Cytotoxic effect of fig latex on cancer cells: Figure 1 indicates the effects of fig latex treatment on the A549 cell viability in the 1/1500-1/12 concentration range for 48 hours. Compared to the control group, we obtained 89.55% viability of A549 cells ($p < 0.05$) at the 1/1500 concentration of fig latex and 79.63%, 78.41%, 63.42%, and 61.05% ($p < 0.0001$) at the 1/900-1/60 concentrations of fig latex, respectively. On the other hand, high concentrations of fig latex (1/36 and 1/12) significantly decreased A549 cell viability to 55.18% and 46.80%, respectively. Moreover, H_2O_2 (500 µM) treatment on the same cell line for 48 hours decreased cell viability to 20.62%. The 50% inhibitory concentration (IC_{50}) value of fig latex was determined to be 1/26 for A549 cells.

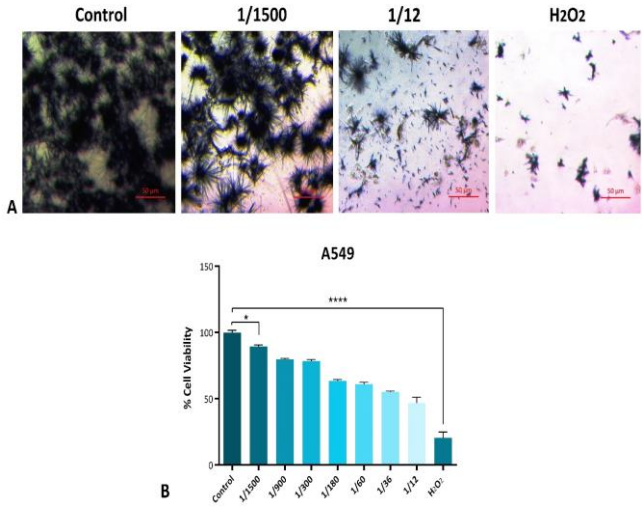

Figure 1. Effects of fig latex treatment at different concentrations on A549 cell viability. **A)** Representative MTT staining microscopy images of A549 cells exposed to fig latex at low and high doses. Irregular cell shape and low cell density in A549. **B)** Results by MTT assay are presented as percentages of cell viability calculated relative to the control group without any substance. Error bars correspond to the standard error of the mean of three repeated experiments. * $p < 0.05$; **** $p < 0.0001$ indicates significant differences between the control and other treated groups.

The cytotoxic effects of fig latex on MCF-7 cells are demonstrated in Figure 2. As can be seen from the figure, there is a significant decrease in the cell viability of breast cancer cells in a dose-dependent manner. For the 1/1500 and 1/900 concentrations, these values were found to be 92.25% and 90.48% ($p < 0.01$, $p < 0.001$), respectively. Furthermore, we acquired cell viability values of 83.65%, 77.97%, 68.13%, 41.56%, and 20.63% for the fig latex concentrations between 1/300 and 1/12 ($p < 0.0001$). For the H_2O_2 treatment, the viability of MCF-7 cells was determined to be 11.11%, which is lower than that of A549 cells. The IC_{50} value of fig latex on MCF-7 cells was calculated as 1/40. This value is lower than that of A549 cells, implying the profound cytotoxic effect of fig latex on MCF-7 cells.

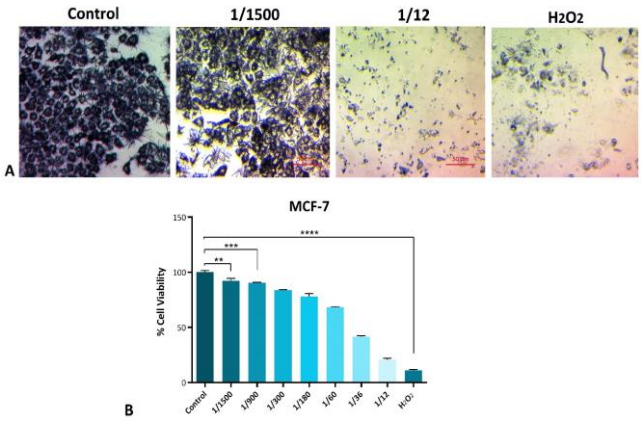

Figure 2. Effects of fig latex treatment at different concentrations on MCF-7 cell viability. **A)** Representative MTT staining microscopy images of MCF-7 cells exposed to fig latex at low and high doses. Irregular cell shape and low cell density in MCF-7 **B)** Results from the MTT assay are presented as percentages of cell viability calculated relative to the control group without any substance. Error bars correspond to the standard error of the mean of three repeated experiments. ** $p < 0.01$; *** $p < 0.001$; **** $p < 0.0001$ indicates significant differences between the control and other treated groups.

To determine the cytotoxic effect of this latex on breast cancer, we also performed an MTT viability assay on the MDA-MB-231 breast cancer cell line. The viability results with various concentrations of fig latex exposure are shown in Figure 3. The 1/1500-1/12 concentrations of fig latex had a significant effect on the viability of MDA-MB-231 cells ($p < 0.0001$). The cell viability values were 26.25% and 16.66% when exposed to high fig latex concentrations (1/36 and 1/12). Furthermore, treatment with H_2O_2 (500 µM) for the same duration decreased cell viability to 11.18% in this cell line, which is the same as that of MCF-7 cells. The IC_{50} value was found to be 1/45 for MDA-MB-231 cells, very close to the value of MCF-7 cells. Based on these findings, we can assume that fig latex increases the cell death rate in cancer cell lines in a concentration-dependent manner, and this effect is dramatic at higher doses.

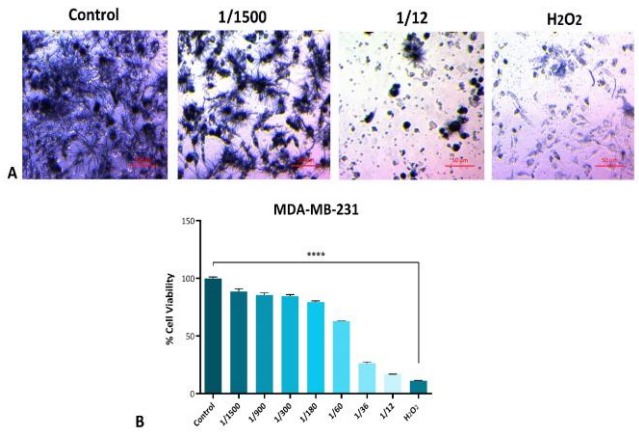

Figure 4. Effects of fig latex treatment at different concentrations on MRC-5 cell viability. **A)** Representative MTT staining microscopy images of MRC-5 cells exposed to fig latex at low and high doses. Irregular cell shape and low cell density MRC-5 **B)** Results from the MTT assay are presented as percentages of cell viability calculated relative to the control group without any substance. Error bars correspond to the standard error of the mean of three repeated experiments. ns: not significant; **** $p < 0.0001$ indicates significant differences between control and other treated groups.

DISCUSSION

Since the late 19th century, scientists have been trying to develop strategies to strengthen the immune system and balance its dysfunctions to eliminate cancer cells (World Health Organization, 2019). The National Cancer Institute (NCI) lists cancer treatment in eight categories: surgery, radiotherapy, chemotherapy, targeted therapy, immunotherapy, stem cell transplantation, and precision medicine (Hanahan Weinberg 2011). However, despite all scientific advances, patients receiving treatment continue to experience resistance to cancer chemotherapy and side effects, which remain the main problem. Therefore, there is a need to discover selective natural compounds with fewer side effects or agents with efficient pharmacological activities, low cost, and minimal disease resistance in cancer treatment (Muhammad et al., 2022). The fact that chemotherapy and radiotherapy cause drug resistance and unwanted toxic side effects. This situation leads to a preference for plant-derived natural compounds in cancer treatment (Garcia-Oliveira et al., 2021). Natural compounds are being developed as a popular therapeutic tool in cancer research due to their easy accessibility and low cost. The discovery and development of anticancer agents has moved from single-target drugs with high side effects to natural plant-based drugs with no or less toxicity (Tuli et al. 2019). The ability of plants to effectively offset complications in cancer and their versatile potential indicate that they are suitable resources for cancer prevention. Therefore, researchers are encouraged to study plants and their active ingredients in order to find therapeutic approaches that have few side effects and are inexpensive. (Muhammad et al., 2022). It is well known that natural products derived from plants are the main source of lead compounds with direct pharmaceutical applications in all disease classes (Li et al., 2014). One of the commonly used plants is fig which has strong therapeutic potential. In this respect, all forms of figs are potential pharmacological candidates as they are a natural source of polyphenols and bioactive metabolites. Fig latex has been scientifically proven to be a source of natural antioxidants (Lansky et al., 2008; Shahinuzzaman et al., 2020). Therefore, the present study was designed to evaluate the anticancer potential of fig latex from *F. carica* fruit stems.

Up to now, there have been many in vitro studies about the antiproliferative or anticancer activities of fig latex from leaves and fruit stems in different cancer types. For instance, a previous study compared the antiproliferative activity of latex from the leaves of *F. carica* and *F. salicifolia* species in MDA-MB-231 human breast cancer cells. That study indicated that *F. carica* species had a stronger ability to kill cells and control the growth of cancer cells in a dose-dependent manner (AlGhalban et al., 2021). Similarly, Abe (2020) also suggested that fig leaf extracts could suppress breast cancer cell growth and migration. Supporting these findings, the cytotoxic activity of fig leaf latex was also reported for human hepatocellular carcinoma (HepG2), breast cancer (MCF-7), colon cancer (HCT 116), and human skin fibroblast (CCD 45SK) cell lines. Among the studied cells, the greatest cytotoxic effect was observed in HepG2 cells (Yahiaoui et al., 2022). In addition to these studies, a recent study also reported that the latex inhibited the growth and migration of non-small cell lung cancer (NSCLC), and it also enhanced the death of these cells. Baohong et al. (2023) also observed that it suppressed tumor growth in A549 xenograft mice, causing no significant damage to normal mouse tissues such as the liver or kidney. Supporting these studies, our results revealed that fig latex profoundly inhibited lung and breast cancer cell lines with respect to the control ones. Thus, we can assume that the fig latex has anti-cancer potential against lung and breast cancer cells.

There have been many in vitro studies regarding the IC_{50} dose of fig latex for cytotoxic activity. In these studies, this dose has been reported to range from µg/mL to mg/mL. In several studies, this value was found to be lower than 5 µg/mL. For instance, in one study, the possible effects of fig latex from the fruit stem on HPV-associated cervical cancer cell lines (CaSki and HeLa) were investigated, and the IC_{50} values were determined to be 0.17 µg/mL and 0.24 µg/mL, respectively. Fig latex was shown to inhibit features associated with HPV-positive cervical cancer-transformed cells, such as rapid growth and invasion, and greatly reduced the expression of p16 and

HPV oncoproteins E6 and E7. Ghanbari et al. 2019). Similarly, another study revealed the IC_{50} values as 0.8 µg/mL for U251 (human glioma) cells, 0.25 µg/mL for SMMC-77721 (human hepatocellular carcinoma) cells, and 0.8 µg/mL for L02 normal liver cells. That study investigated how fig latex treated cells for 72 hours. On day 4, at 1 µg/mL, cell viability dropped below 20% in U251 cells, 0.5 µg/mL on day 2 in SMMC7721 cells, and 2 µg/mL on day 3 in L02 cells. In addition to these studies, the IC_{50} doses of fig latex were also reported to be higher than 10 µg/mL. In one of these studies, it was demonstrated that fruit and leaf latex reduced HeLa cell viability at concentrations as low as 2 µg/mL, with an IC_{50} value of approximately 17 µg/mL (Khodarahmi et al. 2011). Another study reported IC50 values of 0.25 mg/mL for U-138, T98G, and U-87 MG. (Tezcan et al., 2015). The same study demonstrated that these cytotoxic effects synergistically increased in combination with temozolomide, one of the anticancer drugs. On the other hand, a similar anticancer efficacy of fig fruit latex to doxorubicin was also demonstrated in MCF-7 cells (Wang et al., 2008). In that study, the IC_{50} value of this latex was found to be 25.30 µg/mL, close to the efficacy of doxorubicin (IC50: 24.50 µg/mL). Furthermore, the IC50 value was reported for HepG2 cells as 32.25 µg/mL, and for HCT116 cells as 38.75 µg/mL. The same study showed that *F. carica* fruit latex did not kill normal human melanocyte cells (HFB4) (Abdel-Aty et al., 2019).

There are several studies revealing that the IC_{50} dose of fig latex is between 0.1 -10 mg/mL. In a previous research, fig fruit latex was used at different concentrations (5–200 µg/mL) to treat HPV-positive cervical cancer (HeLa HPV18+ and CaSki HPV16+), HPV-negative cervical cancer (C33A), and normal human cervical keratinocyte (HCKT1) cell lines. In that study, the Sulforhodamine B (SRB) colorimetric viability assay revealed that the IC_{50} values of fig latex in HeLa, CaSki, and C33A at 72 hours were 106 µg/mL, 110 µg/mL, and 108 µg/mL, respectively. It was also reported that fig latex did not cause cytotoxicity in normal human cervical keratinocytes compared to cancer cells (Cakir et al., 2023). In another study, MTT assay results revealed apoptosis induction in HT-29 and HCT-116 cells after being treated with fig latex for 48 hours. In that study, this latex was reported to stop cell growth, with IC50 values of 182 µg/mL in HT-29 cells and 206 µg/mL in HCT-116 cells (Soltana et al., 2019). Additionally, the effect of fig tree latex on gastric cancer and peripheral blood mononuclear cells at doses between 0.125-5 mg/mL was evaluated after 72 hours of treatment. In the same study, the optimum concentration for inhibition of cell growth was determined to be 5 mg/mL (Hashemi et al. 2011). Furthermore, this value was found to be 10 mg/mL for esophageal cancer (KYSE-30) cells 72 hours after latex treatment (Hashemi et al., 2013). All these findings obtained from previous studies proved that fig latex carries potential anticancer activity even at very low high doses. In our studies, the lower IC_{50} values for cancer cells confirmed these previous findings.

Similar to in vitro studies, an in vivo study revealed that injecting fig latex directly into the tumor of rats with breast cancer reduced tumor size without any adverse reaction (Ghandehari et al. 2018). In another study, fig latex caused a significant reduction in vascular formation (Mostafaie et al. 2011). In addition, an *in vitro* study showed that fruit latex inhibits angiogenesis (Pawlus et al. 2008).

As well as the cytotoxic activities of fig latex, researchers evaluated the phototoxicity of fig latex from fruits and leaves on human melanoma cells (A375). According to the results of this study, it was revealed that fruit latex had the best antiradical activity with an IC_{50} value of 0.05 mg/mL, and leaf latex showed strong antiproliferative activity with an IC_{50} value of 1.5 µg/mL on A375 after UVA dose (1.08 J/cm^2) irradiation (Menichini et al., 2012).

Previous studies reported that the cytotoxic effects of fig latex arise from its phenolic compounds and Ficin, a cysteine proteinase, (Devaraj et.al., 2008; Rubnov et. al., 2001). An earlier study looked at how fig latex from fruits can help fight cancer in human oral cancer cells called FaDu. The study discovered that fig latex contains a substance known as cysteine protease fisin, which, depending on the dose, inhibits cell growth (Shin et al., 2017). In a different study, fig latex from fruit stems, leaves, and fresh branches significantly slowed down the growth of human prostate (PC3) and colon (HT-29) cancer cells. It was thought that this cytotoxic and apoptotic effect might not just be due to the ficin activity in the latex content, but also to other parts of the latex (Boyacıoğlu et al., 2021). Therefore, we can infer that these compounds in fig latex may contribute to our anticancer activity.

CONCLUSION

Our study contains important findings for developing drugs that respond favorably to all aspects of cancer treatment. Although there has been an increase in research focusing on fig latex and its bioactive compounds, more scientific evidence is needed to determine the possible anticancer properties of these products. Future research should focus on *in vitro* and *in vivo* studies to elucidate the pharmacological mechanism of fig latex and its bioactive compounds.

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REFERENCES

Abdel-Aty, A.M., Hamed, M.B., Salama, W.H., Ali, M.M., Fahmy, A.S. & Mohamed, S.A. (2019). *Ficus carica*, *Ficus sycomorus* and *Euphorbia tirucalli* latex extracts: Phytochemical screening, antioxidant and cytotoxic properties. *Biocatalysis and Agricultural Biotechnology*, ***20***, 101199.

Abe, T. (2020). Fig (*Ficus carica* L.) leaf tea suppresses allergy by acceleration disassembly of IgE-receptor complexes. *Bioscience, Biotechnology, and Biochemistry*, ***84***(5), 1013-1022.

AlGhalban, F.M., Khan, A.A. & Khattak, M.N.K. (2021). Comparative anticancer activities of *Ficus carica* and *Ficus salicifolia* latex in MDA-MB-231 cells. *Saudi Journal of Biological Sciences*, ***28***(6), 3225-3234.

Almehmadi, A. (2023). Biological Activities of *Ficus carica* L. Fig Leaves and Latex. *Journal of Food and Dairy Sciences*, ***14***(7), 173-179.

Badgujar, S.B., Patel, V.V., Bandivdekar, A.H. & Mahajan, R.T. (2014). Traditional uses, phytochemistry and pharmacology of *Ficus carica*: A review. *Pharmaceutical Biology*, ***52***(11), 1487-1503.

Baohong, L., Zhongyuan, L., Ying, T., Beibei, Y., Wenting, N., Yiming, Y., Qinghua, C. & Qingjun, Z. (2023). Latex derived from *Ficus carica* L. inhibited the growth of NSCLC by regulating the caspase/gasdermin/AKT signaling pathway. *Food & Function*, ***14***(4), 2239-2248.

Boyacıoğlu, O., Kara, B., Tecimen, S., Kılıç, M., Delibaş, M., Erdoğan, R., Özdemir, E., Bahadır, A. & Örenay-Boyacıoğlu, S. (2021). Antiproliferative effect of *Ficus carica* latex on cancer cell lines is not solely linked to peroxidase-like activity of ficin. *European Journal of Integrative Medicine*, ***45***, 101348.

Cakir, M.O., Bilge, U., Naughton, D. & Ashrafi, G.H. (2023). *Ficus carica* Latex Modulates Immunity-Linked Gene Expression in Human Papillomavirus Positive Cervical Cancer Cell Lines: Evidence from RNA Seq Transcriptome Analysis. *International Journal of Molecular Sciences*, ***24***(17), 13646.

Castelli, M.V. & Lopez, S.N. (2022). Chemistry, Biological activities, and uses of *Ficus carica* latex, In: *Gums, Resins and Latexes of Plant Origin: Chemistry, Biological Activities and Uses*, 801-822p, Cham: Springer International Publishing.

Devaraj, K.B., Kumar, P.R. & Prakash, V. (2008). Purification, characterization, and solvent-induced thermal stabilization of ficin from Ficus carica. *J. Agric. Food Chem.*, ***56,*** 11417-23.

Garcia-Oliveira, P., Otero, P., Pereira, A. G., Chamorro, F., Carpena, M., Echave, J., Fraga-Corral, M., Simal-Gandara, J. & Prieto, M.A. (2021). Status and challenges of plant-anticancer compounds in cancer treatment. *Pharmaceuticals*, ***14***(2), 157.

Ghanbari, A., Le Gresley, A., Naughton, D., Kuhnert, N., Sirbu, D. & Ashrafi, G.H. (2019). Biological activities of *Ficus carica* latex for potential therapeutics in Human Papillomavirus (HPV) related cervical cancers. *Scientific Reports*, ***9***, 1013.

Ghandehari, F. & Fatemi, M. (2018). The effect of *Ficus carica* latex on 7, 12-dimethylbenz (a) anthracene-induced breast cancer in rats. *Avicenna Journal of Phytomedicine*, ***8***, 286-295.

Hanahan, D. & Weinberg, R.A. (2011). Hallmarks of cancer: the next generation. *Cell*, ***144***(5), 646-674.

Hashemi, S.A. & Abediankenari, S. (2013). Suppressive effect of fig (*Ficus carica*) latex on esophageal cancer cell proliferation. *Acta Facultatis Medicae Naissensis*, ***30***(2), 93-96.

Hashemi, S.A., Abediankenari, S., Ghasemi, M., Azadbakht, M., Yousefzadeh, Y. & Dehpour A.A. (2011). The effect of fig tree latex (*Ficus carica*) on stomach cancer line. *Iranian Red Crescent Medical Journal*, ***13***(4), 272-275.

Hegazy, M.M., Mekky, R.H., Afifi, W.M., Mostafa, A.E. & Abbass, H.S. (2023). Composition and Biological Activities of *Ficus carica* Latex, In: Ramadan, M.F. (Ed) *Fig (Ficus Carica): Production, Processing, and Properties*, 597-641p, Cham, Springer International Publishing.

Khodarahmi, G.A., Ghasemi, N., Hassanzadeh, F. & Safaie, M. (2011). Cytotoxic effects of different extracts and latex of *Ficus carica* L. on HeLa cell line. *Iranian Journal of Pharmaceutical Research*, ***10***(2), 273-277.

Koch, A., Tamez, P., Pezzuto, J. & Soejarto, D. (2005). Evaluation of plants used for antimalarial treatment by the Maasai of Kenya, *J. Ethnopharmacol*. ***101***(1-3) 95-99.

Lansky, E.P., Paavilainen, H.M., Pawlus, A.D. & Newman, R.A. (2008). *Ficus* spp.(fig): Ethnobotany and potential as anticancer and anti-inflammatory agents. *Journal of Ethnopharmacology*, ***119***(2), 195-213.

Li, A.N., Li, S., Zhang, Y.J., Xu, X.R., Chen, Y.M. & Li, H.B. (2014). Resources and biological activities of natural polyphenols. *Nutrients*, ***6***(12), 6020-6047.

Menichini, G., Alfano, C., Provenzano, E., Marrelli, M., Statti, G.A., Somma, F., Menichini, F. & Conforti, F. (2012). Fig latex (*Ficus carica* L. cultivar dottato) in combination with UV irradiation decreases the viability of A375 melanoma cells in vitro. *Anti-Cancer Agents in Medicinal Chemistry*, ***12***(8), 959-965.

Mostafaie, A., Mansouri, K., Norouznezhad, A.H. & Mohammadi, M.H.R. (2011). Anti-angiogenic activity of *Ficus carica* latex extract on human umbilical vein endothelial cells. *Cell Journal*, ***12***(4), 525-528.

Muhammad, N., Usmani, D., Tarique, M., Naz, H., Ashraf, M., Raliya, R., Tabrez, S., Zughaibi, T.A., Alsaieedi, Hakeem, I.J. & Suhail, M. (2022). The role of natural products and their multitargeted approach to treat solid cancer. *Cells*, ***11***(14), 2209.

Pawlus, A.D., Cartwright, C.A., Vijjeswarapu, M., Liu, Z., Woltering, E. & Newman, R.A. (2008). Anti-angiogenic activity from the fruit latex of *Ficus carica* (Fig). *Planta Medica*, ***74***, PA97.

Rasool, I.F.U., Aziz, A., Khalid, W., Koraqi, H., Siddiqui, S.A., Al-Farga, A., Lai, W.F. & Ali, A. (2023). Industrial Application and Health Prospective of Fig (*Ficus carica* L.) By-Products. *Molecules*, ***28***(3), 960.

Rubnov, S., Kashman, Y., Rabinowitz, R., Schlesinger, M. & Mechoulam, R., (2001). Suppressors of cancer cell proliferation from fig (*Ficus carica*) resin: isolation and structure elucidation. *J. Nat. Prod*., ***64***, 993-996.

Shahinuzzaman, M., Yaakob, Z., Anuar, F.H., Akhtar, P., Kadir, N.H.A., Mahmut Hasan, A.K., Sobayel, K., Nour, M., Sindi, H., Amin, N., Sopian, K. & Akhtaruzzaman, Md. (2020). In vitro antioxidant activity of *Ficus carica* L. latex from 18 different cultivars. *Scientific Reports*, ***10***(1), 10852.

Shin, B.S., Lee, S.A., Moon, S.M., Han, S.H., Hwang, E.J., Kim, S.G., Kim, D.K., Kim, J.S., Park, B.R. & Kim, C.S. (2017). Latex of *Ficus carica* L. induces apoptosis through caspase and bcl-2 family in FaDu human hypopharynx squamous carcinoma cells. *International Journal of Oral Biology*, ***42***(4), 183-190.

Shiraishi, C.S.H., Zbiss, Y., Roriz, C.L., Dias, M.I., Prieto, M.A., Calhelha, R.C., Alves, M.J., Heleno, S.A., da Cunha Mendes, V., Carocho, M., Abreu, R.M.V. & Barros, L. (2023). Fig Leaves (*Ficus carica* L.): Source of Bioactive Ingredients for Industrial Valorization. *Processes*, ***11***(4), 1179.

Soltana, H., Pinon, A., Limami, Y., Zaid, Y., Khalki, L., Zaid, N., Salah, D., Yerzhan Sabitaliyevich, U., Simon, A., Liagre, B. & Hammami, M. (2019). Antitumoral activity of *Ficus carica* L. on colorectal cancer cell lines. *Cellular and Molecular Biology*, ***65***(6), 6-11.

Soni, N., Mehta, S., Satpathy, G. & Gupta, R.K. (2014). Estimation of nutritional, phytochemical, antioxidant and antibacterial activity of dried figsg (*Ficus carica*). *Journal of Pharmacognosy and Phytochemistry*, ***3***(2), 158-165.

Teruel-Andreu, C., Andreu-Coll, L., López-Lluch, D., Sendra, E., Hernández, F. & Cano-Lamadrid, M. (2021). *Ficus carica* fruits, by-products and based products as potential sources of bioactive compounds: A review. *Agronomy*, ***11***(9), 1834.

Tezcan, G., Tunca, B., Bekar, A., Yalcin, M., Sahin, S., Budak, F., Cecener, G., Egeli, U., Demir, C., Guvenc, G., Yilmaz, G., Erkan, L.G., Malyer, H., Taskapilioglu, M.O., Evrensel, T. & Bilir, A. (2015). *Ficus carica* latex prevents invasion through induction of let-7d expression in GBM cell lines. *Cellular and Molecular Neurobiology*, ***35***, 175-187.

Tuli, H.S., Tuorkey, M.J., Thakral, F., Sak, K., Kumar, M., Sharma, A.K., Sharma, U., Jain, A., Aggarwal, V. & Bishayee, A. (2019). Molecular mechanisms of action of genistein in cancer: Recent advances. *Frontiers in Pharmacology*, ***10***, 1336.

Upadhyay, R.K. (2011). Plant latex: A natural source of pharmaceuticals and pesticides. *International Journal of Green Pharmacy (IJGP)*, ***5***(3), 169-180.

Wang, J., Wang, X.J., Jiang, S., Lin, P., Zhang, J., Lu, Y.R., Wang, Q., Xiong, Z.J., Wu, Y.Y., Ren, J.J. & Yang, H.L. (2008) Cytotoxicity of fig fruit latex against human cancer cells. *Foodand Chemical Toxicology*, ***46***(3), 1025-1033.

World Health Organization. (2019). International agency for research on cancer. World Health Organization: Geneva, Switzerland.

Yahiaoui, S., Kati, D.E., Ali, L.M., El Cheikh, K., Morére, A., Menut, C., Bachir-bey, M. & Bettache, N. (2022). Assessment of antioxidant, antiproliferative, anti-inflammatory, and enzyme inhibition activities and UPLC-MS phenolic determination of *Ficus carica* latex. *Industrial Crops and Products*, ***178,*** 114629.