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Protective effects of lemongrass (*Cymbopogon citratus* STAPF) extract mediated mitochondrial fission and glucose uptake inhibition in SW1417

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Abstract

This study investigated the anticarcinogenic effect of lemongrass extract on human SW1417 colon cancer cells. SW1417 cells were cultured and allocated into five groups to apply the treatment with the lemongrass extract using 5 separate fluctuated concentrations: 0, 50, 100, 150 and 200 µg/mL for 24 h. The mRNA expressions of the oxidative pressure genes (*HO-1* and *NQO1*) were measured using the RT-PCR technique. Additionally, mitochondrial morphology was evaluated using Biotium (100 nM MitoView™ 405-Blue) within a free culture. The antitumor activity value (IC_{50}) in SW1417 cells was done at 150 µg/mL. Besides, the changes of mitochondrial morphology in the treated SW1417 cells at all the concentrations of lemongrass extract were markedly observed as mitochondrial fission, which increased with increasing the concentrations and led to an apoptotic effect. For more validation, mRNA levels of the oxidative pressure genes *HO-1* and *NQO1* were confirmed the obtained data, *HO-1* and *NQO1* genes expressions recorded significant ($P < 0.05$) increase which associated with increasing the concentrations of lemongrass extract standardized to β-actin housekeeping gene and contrasted to untreated cells (0 µg/mL). In conclusion, our findings indicate that lemongrass extract provided an anticarcinogenic action against the cell proliferation of human colon cancer cells.

Keywords: lemongrass (*Cymbopogon citratus* STAPF) extract; human colon cancer cells (SW1417); anticancer; apoptosis; mitochondrial fission.

Practical Application: The extract of lemongrass acts as an anti-carcinogenic agent against the cell proliferation of SW1417.

1 Introduction

Lemongrass (Cymbopogon citratus STAPF) leaves have been widely consumed as infusions in Brazilian folk medicine to treat ailments through the anti-spasmodic, analgesic, antiinflammatory, antipyretic, diuretic and sedative properties of this species (Blanco et al., 2009; Negrelle & Gomes, 2007). Essential oil of lemongrass is of immense commercial value as a food preservative, flavoring agent and ingredient in fragrances and cosmetics (Ganjewala & Luthra, 2010). In addition, various in vitro and in vivo pharmacological activities of lemongrass essential oil (LGEO) have been described, including anxiolytic and anticonvulsant activities (Blanco et al., 2009; Silva et al., 2010) and antibacterial, antifungal and antiprotozoal properties (Duarte et al., 2007; Irkin & Korukluoglu, 2009; Oliveira et al., 2009; Santoro et al., 2007; Silva et al., 2008). Previous studies have shown antimutagenic and antioxidant activities of lemongrass extracts, or their specific compounds (i.e. citral, b-myrcene and geraniol) in different in vitro and in vivo systems (Aboagye et al., 2021; Cheel et al., 2005; Faheem et al., 2022; Mitić-Ćulafić et al., 2009; Pereira et al., 2009; Rabbani et al., 2006; Tapia et al., 2007). Moreover, geraniol has been found to reduce the proliferative activity of Caco-2 human colon and MCF-7 human breast cancer cells lines (Agnihotri et al., 2022; Zhi et al., 2021). In addition, lemongrass ethanolic extract given orally to male Fischer 344 rats inhibited both colonic aberrant crypt foci (ACF) and hepatic glutathione S-transferase placental form (GST-P) positive foci development induced by carcinogens azoxymethane and diethylnitrosamine, respectively (Nomier et al., 2021; Okada et al., 2021; Zhi et al., 2021).

Cancer chemoprevention is defined as the prevention, inhibition, or reversion of cancer by the administration of natural or synthetic agents (Flora & Ferguson, 2005; George et al., 2021). Chemopreventive agents may inhibit cancer development either by limiting exposure to carcinogens (e.g. carcinogenformation inhibitors and blocking agents) or by decreasing tumor promotion/progression stages (e.g. suppressing agents) (George et al., 2021). Many compounds of medicinal or dietary plants have been identified as potential chemopreventive agents capable of inhibiting DNA damage, and even retarding or reversing the carcinogenesis process in both in vitro and in vivo bioassays (Aggarwal & Shishodia, 2006; Flora & Ferguson, 2005; Malik et al., 2022; Patra et al., 2021). Furthermore, there are various epidemiological studies that associate dietary intakes of fruit, cereal, vegetables, and teas with a lower risk of several human cancers (Khan et al., 2008; Patra et al., 2021). Therefore, the impressive findings of basic research and clinical trials are stimulating the search for potential cancer chemoprevention agents. This study aimed to validate the impact of lemongrass (*Cymbopogon citratus* STAPF) essential oil mediated mitochondrial fission contributed to induced apoptosis in human colon cancer cells.

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2 Materials and methods

2.1 Lemongrass essential oil extraction

Lemongrass leaves (*Cymbopogon citratus* STAPF) were collected from the local medicinal plants (Biology Department Farm, College of Science, Princess Nourah bint Abdulrahman University) and deposited with a voucher specimen (496). The essential oil was extracted from fresh lemongrass leaves through 3 h of boiling hydrodistillation using a Clevenger apparatus and as described in (Bidinotto et al., 2011). The extract was stored at 4 °C in a dark receptacle until the moment of use as.

2.2 Cell culture

Human SW1417 colon cancer cell line was bought from the American Type Culture Collection (ATCC, Manassas, VA, USA) and kept in DMEM/Ham's F-12 (1:1 v/v) medium enhanced with 100 mL/L FBS, 1.5 g/L sodium bicarbonate, 400 µg/mL hydrocortisone, 10 mL/L penicillin and streptomycin (0.1 mg/mL).

2.3 Cell treatment

From the humidified hatchery, cells seeding was additionally done at 1×10^6 cells/well or 1×10^5 cells/well in 96 well tissue culture plates separately. The extract from (Cymbopogon citratus STAPF) lemongrass was put on to a culture media and the cells were then treated with 5 separate fluctuated concentrations 0, 50, 100, 150 and 200 µg/mL for 24 h.

2.4 Cytotoxicity assay

The extract from lemongrass (0.10 mL) were dissolved in 9.90 mL of DMSO to get a working concentration of 1 mg/mL. The active concentration was prepared freshly and filtered through 0.45 µ filter before each assay. In brief, 10 mL of extract was prepared in a concentration of 1 mg/mL. For each sample, 500 µL were poured in ten Eppendorf tubes. The samples were syringe-filtered using 0.45 µM filter to remove contaminants. 500 µL of the sample's working concentration was further added to the first Eppendorf tube and mixed well. Then, 500 µL of this volume was transferred from first to last tube by serial dilution to obtain the desired concentration of the lemongrass extract. As a result, the volume remains constant, but there was a gradual change in concentration. The cytotoxicity assessment was performed using MTT assay.

For this assay, SW1417 cells were plated in 96-well culture plates (1 x 10^4 cells/well). The cells were exposed to five concentrations of 0, 50, 100, 150 and 200 µg/mL of lemongrass extract for 24 hours. The measurements were performed in triplicate. The colors developed in the plates were read at 550 nm by using DMSO as a blank. The percentage of cell viability was expressed using the following formula (Equation 1):

$$\%\ Cell\ viability = mean\ absorbance\ of\ treated\ cells / mean\ absorbance\ of\ control\ cells \times 100 \quad (1)$$

2.5 Evaluating mitochondrial morphology

Non-controlled living SW1417 cells have been cultivated for 15 minutes in the same medium that was preliminary warmed and incorporated Biotium (specifically 100 nM MitoView™ 405-Blue) within a free culture. After two time-washing with PBC, nuclei were stained by Hochest 33342-blue stain for 10 min. Afterwards, the cells have been investigated using the inverted phase-contrast microscopy (developed by Carl Zeiss Microscopy, Germany) with 40X magnitude to view the mitochondrial morphology and following the methods described in previous report (Alkhateeb et al., 2021). All chemical agents used in the experiment were retrieved from Sigma (USA).

2.6 Measurement of glucose uptake

2-[N-(7-nitrobenz-2-oxa-1,3-diazol-4-yl)amino]-2-deoxy-D-glucose (2-NBDG) was used to treat SW1417 cells for 30 min, after lemongrass extract-treatment for 24 h. SW1417 colon cancer cells glucose uptake was assayed using flow cytometry (Becton-Dickinson, San Jose, CA).

2.7 Determination of (ROS)-Reactive Oxygen Species production

Subcellular ROS was examined fluorometrically by estimating the of a non-fluorescent test 2,7-dichloro-fluorescein diacetate (DCF-DA) oxidation to a fluorescent metabolite dichlorofluorescein (DCF) via mitochondrial ROS just as depicted beforehand with slight adjustments (Elbekai et al., 2004). Gathered cells were suspended in 500 mL of PBS and mixed in with 10 mM (last centralization) of dichloro-dihydro-fluorescein diacetate (DCFH-DA) for 20 min at 37 °C. The cells suspension was deposited at 1200 rpm for 5 min. Therefore, the cells were washed thrice with 500 mL of Phosphate-Buffered Saline (PBS)/pellet to evacuate excess DCFH-DA. The ROS level was tested by flow cytometry (Becton-Dickinson, San Jose, CA).

2.8 RNA extraction and cDNA synthesis

All out RNA was separated utilizing Invitrogen-TRI-zol reagent as indicated by the maker's guidelines and evaluated by estimating the absorbency at 260 nm. The quality of RNA was controlled by estimating 260/280 proportions. From that point, the synthesizing of the cDNA-strand was produced utilizing the High-Amplitude cDNA turn around interpretation pack (Applied Biosystems) as indicated by the maker's directions (Zordoky et al., 2008).

2.9 Measurement of mRNA expressions by Real-Time Polymerase Chain Reactions (RT-PCR)

The primers were utilized in the present examination (Table 1) were bought from (Invitrogen, USA). Measure controls were consolidated in separated wells but onto a similar plate, to be more specific. All the samples and controls were run in triplicates on an ABI 7500 Fast Real-time PCR. The quantitative

Table 1. The sequence of primers.

Gene	Forward primer	Reverse primer
HO-1	ATGGCCTCCCTGTACCACATC	TGTTGCGCTCAATCTCCTCCT
NQO1	CGCAGACCTTGTGATATTCCAG	CGTTTCTTCCATCCTTCCAGG
β-actin	GCACCACACCTTCTACAATG	TGCTTGCTGATCCACATCTG

RT-PCR data was breaking down by a near edge (Ct) strategy, and the overlap acceptances of treated examples were contrasted and the untreated examples. Relative quality expression (i.e., ΔΔCT) strategy as earlier outlined was used to analyse the data on the RT-PCR (Livak & Schmittgen, 2001). β-actin was utilized as an interior reference gene to standardize the declaration of the selected genes.

2.10 Statistical analysis

Analytical examinations were performed by use of SigmaStat programming adaptation 3.5 (Systat Software, San Jose, CA, USA). Quantitative outcomes were presented as mean standard deviations. Esteems of p being lower than 0.05 were deemed statistically imperative.

3 Results

3.1 Effect of lemongrass leaves extract on SW1417 cell proliferation

To determine the ability of lemongrass leaves extract to inhibit growth and proliferation of SW1417 colon cancer cells were treated with steadily increasing concentrations of lemongrass leaves extract (0, 50, 100, 150 and 200 µg/mL) for 24 h, after which cell reasonability and expansion were determined using MTT assay. Figure 1 exhibits that endurance of SW1417 cells were altogether diminished after incubation with lemongrass leaves separate in a focus subordinate way when contrasted with untreated SW1417 cells (Figure 1), proposing that lemongrass leaves extract is tumor cell selective. The determined IC_{50} for lemongrass leaves extract is around 150 µg/mL.

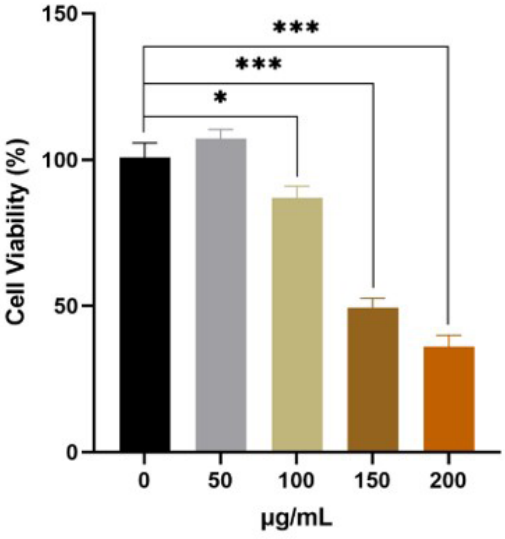

Figure 1. Cytotoxicity assessment by MTT assay on SW1417 colon cancer cells exposed to various concentrations of lemongrass leaves extract for 24 h. Stars indicate statistically significant differences of cytotoxicity and cell viability assessment between the concentrations (0, 50, 100, 150 and 200 µg/mL). Values are put as percentages of the control (mean ± SEM, n = 5) ***P < 0.001, **P < 0.01, *P < 0.05 in comparison to the control (0 µg/mL).

3.2 Impact of lemongrass extract on mitochondrial morphology

During the study, the mitochondrial morphology (MM) has been investigated utilizing the Biotium stain in relation to living SW1417 cells covering both treated and non-treated cells. The special effect was obtained by applying lemongrass extract at five separate doses, namely (0, 50, 100, 150 and 200 µg/mL) throughout 24-hour processing. Non-treated SW1417 cells manifested standard structure with non-affected mitochondrial conditions; however, upon adding lemongrass extract, MM was visibly modified demonstrating an elevation in the fragmentation effects as well as punctiform colonial morphology. This happened supposedly due to recorded mitochondrial damage that caused an elevation in the consistencies of lemongrass extract in the processed SW1417 cells. Technically, the top mitochondrial damage was noticed at doses 100, 150 and 200 µg/mL (Figure 2).

3.3 Glucose uptake inhibition

During glucose metabolism, ATP production and cell proliferation, both are significant in cell growth. Nevertheless,

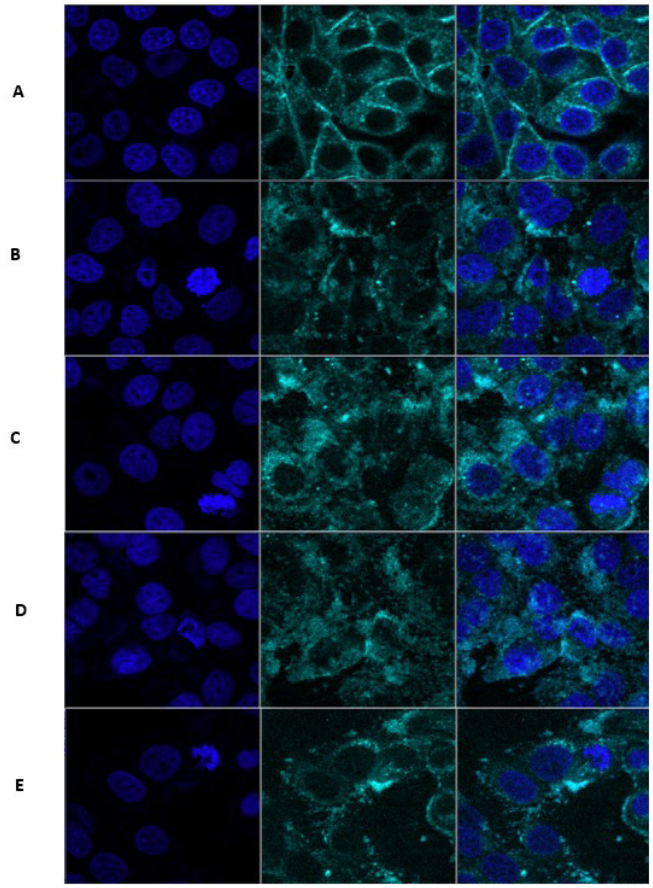

Figure 2. The effect of lemongrass extract on SW1417 cells showing abnormal shape of nuclei by (Hochest 33342-blue stain) and mitochondrial fusion by (100 nM MitoView™ 405- Blue stain). A) represents untreated SW1417 cells with normal structure and undamaged mitochondrial status. In contrast, other SW1417 cells were treated by lemongrass extract had mitochondrial changes in shape with fragmented patterns and punctiform morphology when SW1417 cells treated at four different concentrations (50, 100, 150 and 200 µg/mL) as seen in B, C, D, and E respectively.

if glucose absorption was inhibited there is a subsequent of cell growth suppression. We discovered that the uptake of glucose (2-NBDG) uptake was affected by lemongrass extract. The glucose take-up restraint was eased by a dose-subordinate way in SW1417 cells with lemongrass extract for 24 hours as shown in (Figure 3).

3.4 Effect of lemongrass extract on the expression of oxidative stress genes and ROS production in SW1417 cells

To examine if lemongrass extract interceded oxidative stress, we determined the capacity of lemongrass extract to balance the declaration of oxidative pressure genes in SW1417 human colon cancer cells. Consequently, SW1417 cells were treated with same convergence of lemongrass extract for 24, from that point ROS creation and *NQO1* and *HO1* mRNA levels were estimated by DCF and RT-PCR measure, respectively. Our outcomes demonstrated that lemongrass extract essentially expanded the ROS creation at all concentrations with a most extreme acceptance of 3, 6, 9 and 10-overlays accomplished by 100, 150 and 200 µg/mL of lemongrass extract, respectively (Figure 4). While it fundamentally initiated *HO-1* mRNA levels in SW1417 cells in a fixation subordinate way (Figure 5a) nonetheless; actuated *HO-1* mRNA levels just at the most noteworthy concentrations tried (150 and 200 µg/mL) for 24 h. Interestingly, increasing the concentrations of lemongrass extract was associated with decrease the mRNA levels of *NQO1* except the first concentration of lemongrass extract (50 µg/mL) which recorded significant ($P < 0.05$) increase in the gene expression of *NQO1* (Figure 5b).

4 Discussion

The prominent cultivation of lemongrass (Cymbopogon spp.) relies on the pharmacological incentives of its essential oil. Lemongrass essential oil (LEO) carries a significant amount of numerous bioactive compounds, such as citral (mixture of geranial and neral), isoneral, isogeranial, geraniol, geranyl acetate, citronellal, citronellol, germacrene-D, and elemol, in addition to other bioactive compounds. These components confer various pharmacological actions to LEO, including antifungal, antibacterial, antiviral, anticancer, and antioxidant properties (Mukarram et al., 2022; Pan et al., 2022).

The outcomes from the phytochemical subjective examination of lemongrass extract demonstrated that most of the credited bioactivty, as cancer prevention agent has been ascribed to its compounds (e.g. citral and b-myrcene) have shown antioxidant and antigenotoxic/antimutagenic activities against different mutagens (Bidinotto et al., 2011; Rao et al., 2009; Rabbani et al., 2006).

The findings showed that the treatment with lemongrass extract reduced the cell viability of SW1417 cells were gradually with increase the concentrations of lemongrass extract, especially after the dose 100 µg/mL of this extract in comparison to the control (0 µg/mL), the decrease in the number of living cells is due to glucose uptake inhibition that affects ATP production and cell proliferation, both are significant in cell growth. Nevertheless, if glucose absorption was inhibited there is a subsequent of cell growth suppression, apoptotic genes and oxidative stress activation, and intracellular ROS accumulation (Alkhateeb et al., 2021; Manosroi et al., 2006). Moreover, significantly high ROS levels in mitochondria can result in free radicals' attacks on membrane phospholipids that go before mitochondrial film depolarization. Mitochondrial depolarization, viewed as an irreversible advance in apoptosis (Manosroi et al., 2006). The improvement of ROS creation prompted expanded apoptosis occasions (Figure 2). Provided that mitochondrial morphology influences imbalances in energy and is ceaselessly changed via fission and fusion events, tight coordination

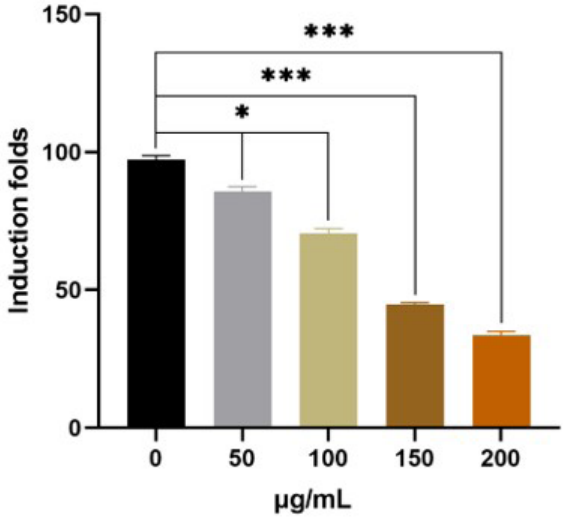

Figure 3. Restraint of glucose take-up was measured by flow cytometry on SW1417 colon cancer cells that were treated by four concentrations of lemongrass leaves extract for 24 h. Stars indicate statistically significant differences of the values based on the concentrations (0, 50, 100, 150 and 200 µg/mL). Values were recorded as mean ± SEM, (n = 5) ***P < 0.001, **P < 0.01, *P < 0.05 contrasted with control (0 µg/mL).

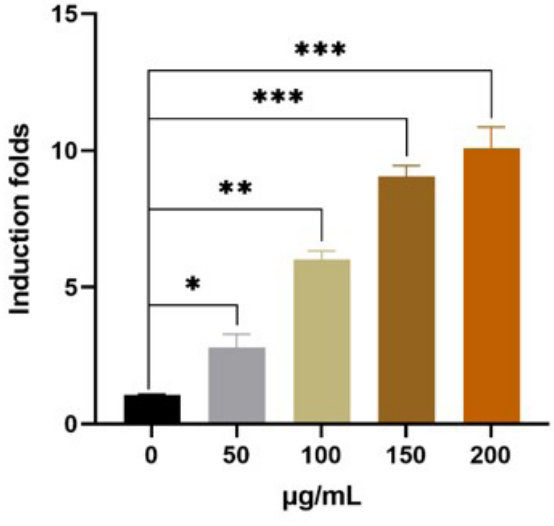

Figure 4. ROS creation in SW1417 cells was treated for 24 h with different groupings of lemongrass leaves extract (0, 50, 150, and 250 µg/mL) for 24 h. DCF arrangement was estimated fluorometrically utilizing excitation/outflow frequencies of 484/535 nm. Values were introduced as means ± SEM, (n = 10). ***P < 0.001, **P < 0.01, *P < 0.05 contrasted with control (0 µg/mL).

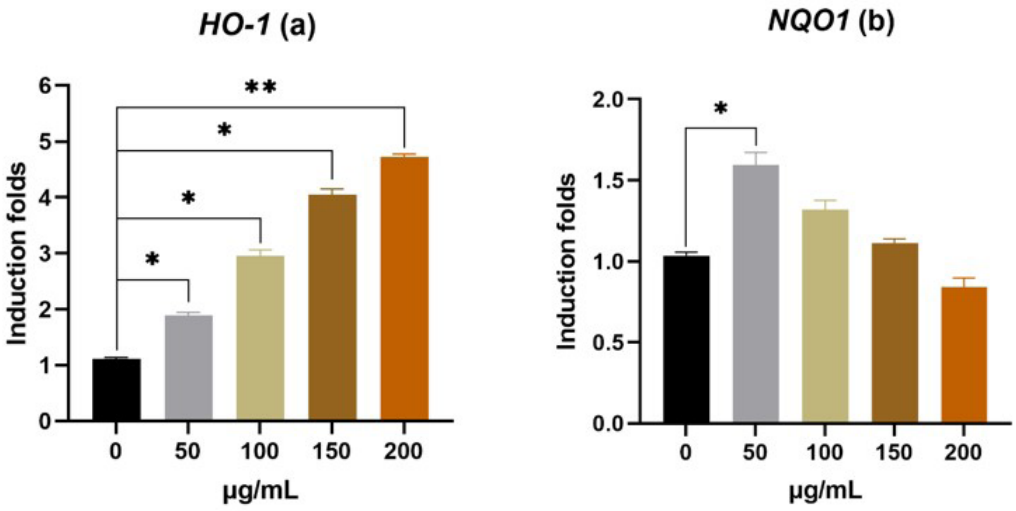

Figure 5. Effect of lemongrass leaves extract on oxidative pressure genes *HO-1* (a) and *NQO1* (b) mRNA levels in SW1417 cells treated for 24 h with different concentrations of lemongrass leaves extract (0, 50, 100, 150, and 200 µg/mL). From there on, the mRNA levels of *HO-1* and *NQO1* were measured utilizing RT-PCR and standardized to β-actin housekeeping gene. Data were recorded as means ± SEM (n = 5) of three free investigations. ***P < 0.001, **P < 0.01, *P < 0.05 contrasted to untreated cells (0 µg/mL).

betwixt inter-organelle interactions and mitochondrial dynamics is vital. Mitochondrial splitting outcomes in a disabled insulin-subordinate glucose take-up (Hsu et al., 2015). Apoptosis is a firmly controlled procedure heavily influenced by a few flagging pathways, for example, mitochondrial pathways and caspases (Bonora et al., 2021). Apoptosis induction with ROS generation by malignant growth chemoprotective agents, for example, doxorubicin (Elbekai et al., 2004), incites disease cell passing as well as purposes DNA harm and genomic insecurity (George et al., 2021; Elbekai et al., 2004; Zordoky et al., 2008). Nevertheless, a large portion of these malignant growth chemoprotective treatments are cytotoxic and their utilization is related to toxicities. Thus, the generation of new chemopreventive specialists ready to repress cell expansion and actuate apoptosis in malignant growth cells however with less or no reactions is significant and foreseen. Along these lines, to display the in vivo circumstance, human colon malignancy SW1417 cell lines were utilized in the present examination to anticipate human reactions to lemongrass extract by researching the limit of this extract to hinder SW1417 cells development and expansion and investigate the job of apoptosis in lemongrass extract extricate—interceded impact. These data are consistent with a recent study that applied the lemongrass extract to $APC^{min/+}$ transgenic mice and led to the reduction of intestinal tumors using oral administration lemongrass extract (Ruvinov et al., 2019), this study showed that the lemongrass extract was well tolerated and effective at inhibiting colon cancer xenograft growth in mice. The effect of lemongrass leaves extract on oxidative pressure gene *HO-1* mRNA level in SW1417 cells were treated for 24 h with different concentrations (0, 50, 100, 150, and 200 µg/mL), showed significant increase with increasing the concentrations which consistent with the data reported in (Alkhateeb et al., 2021).

Furthermore, *NQO1* mRNA levels in SW1417 cells treated for 24h with different concentrations of lemongrass leaves extract (0, 50, 100, 150, and 200 µg/ mL) and recorded significant increase at only the first concentration of lemongrass extract (50 µg/mL) followed by nonsignificant and gradually decrease in the gene expression at (100, 150 and 200 µg/mL) with increasing the concentrations, these data indicated that the higher concentrations of lemongrass extract returned the levels of *NQO1* gene expression to the normal lower levels. Yang et al. (2022) reported that the analysis of *NQO1* mRNA gene expression indicated 50-fold higher levels in untreated liver tumors and in the tissue surrounding the tumors of patients with hepatocarcinoma than in normal individuals (Yang et al., 2022). Also, previous studies presented that *NQO1* gene expression is elevated in some human cancers such as breast, colon, and lung and colorectal cancer before applying the treatment (Licznerska et al., 2021; Mizumoto et al., 2019; Preethi et al., 2022; Yadav et al., 2018; Yang et al., 2022).

5 Conclusion

The findings of the present study indicate that lemongrass extract showed a potential anticarcinogenic activity (i.e. suppressing effect) in vitro using human colon cancer cell line (SW1417) by inducing mitochondrial fission, ROS production, and apoptosis in addition to glucose uptake inhibition. However, these results require further testing to identify whether or not these findings will be happened in healthy cell lines and further investigations for its potential contributions as a cancer treatment for other types of cancer, prevention, and prevention of relapse.

Ethical approval

Not applicable.

Conflict of interest

The author declares that there is no conflict of interest.

Availability of data and material

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

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Author contributions

MAA: Conceptualization, Methodology, Software, Formal analysis, Writing - Original Draft, Review & Editing.

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References

Aboagye, G., Tuah, B., Bansah, E., Tettey, C., & Hunkpe, G. (2021). Comparative evaluation of antioxidant properties of lemongrass and other tea brands. *Scientific African*, 11, e00718. http://dx.doi.org/10.1016/j.sciaf.2021.e00718.

Aggarwal, B. B., & Shishodia, S. (2006). Molecular targets of dietary agents for prevention and therapy of cancer. *Biochemical Pharmacology*, 71(10), 1397-1421. http://dx.doi.org/10.1016/j.bcp.2006.02.009. PMid:16563357.

Agnihotri, S., Dobhal, P., Ashfaqullah, S., Chauhan, H. K., & Tamta, S. (2022). Review of the botany, traditional uses, pharmacology, threats and conservation of Zanthoxylum armatum (Rutaceae). *South African Journal of Botany*, 150, 920-927. http://dx.doi.org/10.1016/j.sajb.2022.08.038.

Alkhateeb, M. A., Al-Otaibi, W. R., AlGabbani, Q., Alsakran, A. A., Alnafjan, A. A., Alotaibi, A. M., & Al-Qahtani, W. S. (2021). Low-temperature extracts of purple blossoms of basil (*Ocimum basilicum L.*) intervened mitochondrial translocation contributes prompted apoptosis in human breast cancer cells. *Biological Research*, 54(1), 2. http://dx.doi.org/10.1186/s40659-020-00324-0. PMid:33407904.

Bidinotto, L. T., Costa, C. A., Salvadori, D. M., Costa, M., Rodrigues, M. A., & Barbisan, L. F. (2011). Protective effects of lemongrass (Cymbopogon citratus STAPF) essential oil on DNA damage and carcinogenesis in female Balb/C mice. *Journal of Applied Toxicology*, 31(6), 536-544. http://dx.doi.org/10.1002/jat.1593. PMid:21089157.

Blanco, M. M., Costa, C. A. R. A., Freire, A. O., Santos, J. G. Jr., & Costa, M. (2009). Neurobehavioral effect of essential oil of Cymbopogon citratus in mice. *Phytomedicine*, 16(2-3), 265-270. http://dx.doi.org/10.1016/j.phymed.2007.04.007. PMid:17561386.

Bonora, M., Missiroli, S., Perrone, M., Fiorica, F., Pinton, P., & Giorgi, C. (2021). Mitochondrial control of genomic instability in cancer. *Cancers*, 13(8), 1914. http://dx.doi.org/10.3390/cancers13081914. PMid:33921106.

Cheel, J., Theoduloz, C., Rodríguez, J., & Schmeda-Hirschmann, G. (2005). Free radical scavengers and antioxidants from lemongrass (Cymbopogon citratus (DC.) Stapf.). *Journal of Agricultural and Food Chemistry*, 53(7), 2511-2517. http://dx.doi.org/10.1021/jf0479766. PMid:15796587.

Duarte, M. C. T., Leme, E. E., Delarmelina, C., Soares, A. A., Figueira, G. M., & Sartoratto, A. (2007). Activity of essential oils from Brazilian medicinal plants on Escherichia coli. *Journal of Ethnopharmacology*, 111(2), 197-201. http://dx.doi.org/10.1016/j.jep.2006.11.034. PMid:17210236.

Elbekai, R. H., Korashy, H. M., Wills, K., Gharavi, N., & El-Kadi, A. O. (2004). Benzo [a] Pyrene, 3-Methylcholanthrene and ß-Naphthoflavone induce oxidative stress in Hepatoma Hepa 1c1c7 cells by an AHR-dependent pathway. *Free Radical Research*, 38(11), 1191-1200. http://dx.doi.org/10.1080/10715760400017319. PMid:15621696.

Faheem, F., Liu, Z. W., Rabail, R., Haq, I. U., Gul, M., Bryła, M., Roszko, M., Kieliszek, M., Din, A., & Aadil, R. M. (2022). Uncovering the industrial potentials of lemongrass essential oil as a food preservative: a review. *Antioxidants*, 11(4), 720. http://dx.doi.org/10.3390/antiox11040720. PMid:35453405.

Flora, S., & Ferguson, L. R. (2005). Overview of mechanisms of cancer chemopreventive agents. *Mutation Research. Fundamental and Molecular Mechanisms of Mutagenesis*, 591(1-2), 8-15. http://dx.doi.org/10.1016/j.mrfmmm.2005.02.029. PMid:16107270.

Ganjewala, D., & Luthra, R. (2010). Essential oil biosynthesis and regulation in the genus Cymbopogon. *Natural Product Communications*, 5(1), 163-172. http://dx.doi.org/10.1177/1934578X1000500137. PMid:20184044.

George, B. P., Chandran, R., & Abrahamse, H. (2021). Role of phytochemicals in cancer chemoprevention: insights. *Antioxidants*, 10(9), 1455. http://dx.doi.org/10.3390/antiox10091455. PMid:34573087.

Hsu, W. H., Lee, B. H., & Pan, T. M. (2015). Leptin-induced mitochondrial fusion mediates hepatic lipid accumulation. *International Journal of Obesity*, 39(12), 1750-1756. http://dx.doi.org/10.1038/ijo.2015.120. PMid:26119995.

Irkin, R., & Korukluoglu, M. (2009). Effectiveness of Cymbopogon citratus L. essential oil to inhibit the growth of some filamentous fungi and yeasts. *Journal of Medicinal Food*, 12(1), 193-197. http://dx.doi.org/10.1089/jmf.2008.0108. PMid:19298215.

Khan, N., Afaq, F., & Mukhtar, H. (2008). Cancer chemoprevention through dietary antioxidants: progress and promise. *Antioxidants & Redox Signalling*, 10(3), 475-510. http://dx.doi.org/10.1089/ars.2007.1740. PMid:18154485.

Licznerska, B., Szaefer, H., & Krajka-Kuźniak, V. (2021). R-sulforaphane modulates the expression profile of AhR, ERα, Nrf2, NQO1, and GSTP in human breast cell lines. *Molecular and Cellular Biochemistry*, 476(2), 525-533. http://dx.doi.org/10.1007/s11010-020-03913-5. PMid:33064289.

Livak, K. J., & Schmittgen, T. D. (2001). Analysis of relative gene expression data using real-time quantitative PCR and the 2− ΔΔCT method. *Methods*, 25(4), 402-408. http://dx.doi.org/10.1006/meth.2001.1262. PMid:11846609.

Malik, S., Kaur, K., Prasad, S., Jha, N. K., & Kumar, V. (2022). A perspective review on medicinal plant resources for their antimutagenic potentials. *Environmental Science and Pollution Research International*, 29(41), 62014-62029. http://dx.doi.org/10.1007/s11356-021-16057-w. PMid:34431051.

Manosroi, J., Dhumtanom, P., & Manosroi, A. (2006). Anti-proliferative activity of essential oil extracted from Thai medicinal plants on KB and P388 cell lines. *Cancer Letters*, 235(1), 114-120. http://dx.doi.org/10.1016/j.canlet.2005.04.021. PMid:15979235.

Mitić-Ćulafić, D., Žegura, B., Nikolić, B., Vuković-Gačić, B., Knežević-Vukčević, J., & Filipič, M. (2009). Protective effect of linalool, myrcene and eucalyptol against t-butyl hydroperoxide induced genotoxicity in bacteria and cultured human cells. *Food and*

Chemical Toxicology, 47(1), 260-266. http://dx.doi.org/10.1016/j.fct.2008.11.015. PMid:19049815.

Mizumoto, A., Ohashi, S., Kamada, M., Saito, T., Nakai, Y., Baba, K., Hirohashi, K., Mitani, Y., Kikuchi, O., Matsubara, J., Yamada, A., Takahashi, T., Lee, H., Okuno, Y., Kanai, M., & Muto, M. (2019). Combination treatment with highly bioavailable curcumin and NQO1 inhibitor exhibits potent antitumor effects on esophageal squamous cell carcinoma. *Journal of Gastroenterology*, 54(8), 687-698. http://dx.doi.org/10.1007/s00535-019-01549-x. PMid:30737573.

Mukarram, M., Choudhary, S., Khan, M. A., Poltronieri, P., Khan, M., Ali, J., Kurjak, D., & Shahid, M. (2022). Lemongrass essential oil components with antimicrobial and anticancer activities. *Antioxidants*, 11(1), 20. http://dx.doi.org/10.3390/antiox11010020. PMid:35052524.

Negrelle, R. R. B., & Gomes, E. C. (2007). Cymbopogon citratus (DC.) Stapf: chemical composition and biological activities. *Revista Brasileira de Plantas Medicinais*, 9(1), 80-92. Retrieved from https://api.semanticscholar.org/CorpusID:56066104

Nomier, Y., Asaad, G. F., Alshahrani, S., Safhi, S., Medrba, L., Alharthi, N., Rehman, Z., Alhazmi, H., & Sanobar, S. (2021). Antidepressant and anxiolytic profiles of Cymbopogon Flexuosus ethanolic extract in chronic unpredictable mild stress induced in rats. *Biomedical & Pharmacology Journal*, 14(1), 175-185. http://dx.doi.org/10.13005/bpj/2112.

Okada, F., Izutsu, R., Goto, K., & Osaki, M. (2021). Inflammation-related carcinogenesis: lessons from animal models to clinical aspects. *Cancers*, 13(4), 921. http://dx.doi.org/10.3390/cancers13040921. PMid:33671768.

Oliveira, V. C., Moura, D. M., Lopes, J. A., Andrade, P. P., Silva, N. H., & Figueiredo, R. C. (2009). Effects of essential oils from Cymbopogon citratus (DC) Stapf., Lippia sidoides Cham., and Ocimum gratissimum L. on growth and ultrastructure of Leishmania chagasi promastigotes. *Parasitology Research*, 104(5), 1053-1059. http://dx.doi.org/10.1007/s00436-008-1288-6. PMid:19085009.

Pan, D., Machado, L., Bica, C. G., Machado, A. K., Steffani, J. A., & Cadoná, F. C. (2022). In vitro evaluation of antioxidant and anticancer activity of lemongrass (Cymbopogon citratus (DC) Stapf). *Nutrition and Cancer*, 74(4), 1474-1488. http://dx.doi.org/10.1080/01635581.2021.1952456. PMid:34282694.

Patra, S., Pradhan, B., Nayak, R., Behera, C., Das, S., Patra, S. K., Efferth, T., Jena, M., & Bhutia, S. K. (2021). Dietary polyphenols in chemoprevention and synergistic effect in cancer: clinical evidences and molecular mechanisms of action. *Phytomedicine*, 90, 153554. http://dx.doi.org/10.1016/j.phymed.2021.153554. PMid:34371479.

Pereira, R. P., Fachinetto, R., Prestes, A. S., Puntel, R. L., Silva, G. N. S., Heinzmann, B. M., Boschetti, T. K., Athayde, M. L., Bürger, M. E., Morel, A. F., Morsch, V. M., & Rocha, J. B. T. (2009). Antioxidant effects of different extracts from Melissa officinalis, Matricaria recutita and Cymbopogon citratus. *Neurochemical Research*, 34(5), 973-983. http://dx.doi.org/10.1007/s11064-008-9861-z. PMid:18853256.

Preethi, S., Arthiga, K., Patil, A. B., Spandana, A., & Jain, V. (2022). Review on NAD (P) H dehydrogenase quinone 1 (NQO1) pathway. *Molecular Biology Reports*, 49(9), 8907-8924. http://dx.doi.org/10.1007/s11033-022-07369-2. PMid:35347544.

Rabbani, S. I., Devi, K., Khanam, S., & Zahra, N. (2006). Citral, a component of lemongrass oil inhibits the clastogenic effect of nickel chloride in mouse micronucleus test system. *Pakistan Journal of Pharmaceutical Sciences*, 19(2), 108-113. PMid:16751120.

Rao, B. S. S., Shanbhoge, R., Rao, B. N., Adiga, S. K., Upadhya, D., Aithal, B. K., & Kumar, M. R. S. (2009). Preventive efficacy of hydroalcoholic extract of Cymbopogon citratus against radiation-induced DNA damage on V79 cells and free radical scavenging ability against radicals generated in vitro. *Human and Experimental Toxicology*, 28(4), 195-202. http://dx.doi.org/10.1177/0960327109104822. PMid:19734270.

Ruvinov, I., Nguyen, C., Scaria, B., Vegh, C., Zaitoon, O., Baskaran, K., Mehaidli, A., Nunes, M., & Pandey, S. (2019). Lemongrass extract possesses potent anticancer activity against human colon cancers, inhibits tumorigenesis, enhances efficacy of FOLFOX, and reduces its adverse effects. *Integrative Cancer Therapies*, 18, 1534735419889150. http://dx.doi.org/10.1177/1534735419889150. PMid:31845598.

Santoro, G. F., Cardoso, M. G., Guimarães, L. G. L., Freire, J. M., & Soares, M. J. (2007). Anti-proliferative effect of the essential oil of Cymbopogon citratus (DC) Stapf (lemongrass) on intracellular amastigotes, bloodstream trypomastigotes and culture epimastigotes of Trypanosoma cruzi (Protozoa: Kinetoplastida). *Parasitology*, 134(Pt 11), 1649-1656. http://dx.doi.org/10.1017/S0031182007002958. PMid:17686189.

Silva, C. B., Guterres, S. S., Weisheimer, V., & Schapoval, E. E. S. (2008). Antifungal activity of the lemongrass oil and citral against Candida spp. *The Brazilian Journal of Infectious Diseases*, 12(1), 63-66. http://dx.doi.org/10.1590/S1413-86702008000100014. PMid:18553017.

Silva, M. R., Ximenes, R. M., Costa, J. G. M., Leal, L. K. A. M., Lopes, A. A., & Viana, G. S. B. (2010). Comparative anticonvulsant activities of the essential oils (EOs) from Cymbopogon winterianus Jowitt and Cymbopogon citratus (DC) Stapf. in mice. *Naunyn-Schmiedeberg's Archives of Pharmacology*, 381(5), 415-426. http://dx.doi.org/10.1007/s00210-010-0494-9. PMid:20237771.

Tapia, A., Cheel, J., Theoduloz, C., Rodríguez, J., Schmeda-Hirschmann, G., Gerth, A., Wilken, D., Jordan, M., Jiménez-González, E., Gomez-Kosky, R., & Mendoza, E. Q. (2007). Free radical scavengers from Cymbopogon citratus (DC.) stapf plants cultivated in bioreactors by the temporary immersion (TIS) principle. *Zeitschrift für Naturforschung C*, 62(5-6), 447-457. http://dx.doi.org/10.1515/znc-2007-5-620. PMid:17708453.

Yadav, U., Kumar, P., & Rai, V. (2018). NQO1 gene C609T polymorphism (dbSNP: rs1800566) and digestive tract cancer risk: a meta-analysis. *Nutrition and Cancer*, 70(4), 557-568. http://dx.doi.org/10.1080/01635581.2018.1460674. PMid:29652514.

Yang, X., Duan, J., & Wu, L. (2022). Research advances in NQO1-responsive prodrugs and nanocarriers for cancer treatment. *Future Medicinal Chemistry*, 14(5), 363-383. http://dx.doi.org/10.4155/fmc-2021-0289. PMid:35102756.

Zhi, H., Cui, J., Yang, H., Zhang, Y., & Zhu, M. (2021). Research progress of geraniol in tumor therapy. *Proceedings of Anticancer Research*, 5(1), 26-30. http://dx.doi.org/10.26689/par.v5i1.1882.

Zordoky, B. N., Aboutabl, M. E., & El-Kadi, A. O. (2008). Modulation of cytochrome P450 gene expression and arachidonic acid metabolism during isoproterenol-induced cardiac hypertrophy in rats. *Drug Metabolism and Disposition: the Biological Fate of Chemicals*, 36(11), 2277-2286. http://dx.doi.org/10.1124/dmd.108.023077. PMid:18725507.