Author! Ensure that your manuscripts should be completely grammatical error free . Do not form complex sentences unless it is needed.

KAEMPFEROL EXERTS A DIFFERENTIAL EFFECT ON K… CELLS AND NORMAL HUMAN BUCCAL C…

Title of the manuscript should be 15 words

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Affiliation of the authors

ABSTRACT

Abstract should be not more than 250 words and should have some lines on introduction, your objective, some important results, discussion and conclusion

Kaempferol is a natural plant derived flavonoid that has been show… against several cancer cells, but the differential effect of the compour… cells is largely unknown. In the present study, we attempted to id… kaempferol in KB oral cancer cell line and its effect on cultured norma… assays showed that kaempferol significantly reduced the viability of KB cells(64% , p<0.01), whereas in normal buccal cells, the viability was comparatively higher (86% , p<0.01%). Fluorescent staining of kaempferol treated KB cells showed changes in nuclear morphology typical to apoptotic cells . Our results showed that kaempferol significantly induces apoptotic cell death in oral carcinoma cells leading to G_2/M cell cycle arrest. Collectively, our findings indicate the potency of kaempferol as a future candidate for the development of oral cancer chemotherapeutic agent, which can induce death in cancer cells while protecting the non-canceroous cells.

KEYWORDS: **Kaempferol, buccal cells, oral carcinoma**

Keywords should be 5-6 words.

INTRODUCTION

Cancer is a major health problem in many parts of the world. The American Cancer Society recently reported that cancer is the second leading cause of death in the United States and in the next few years, it may surpass heart diseases as the leading cause of death.[1] Oral cancer is the sixth most frequent type of cancer among the various types of cancers, in which, malignant cells form in the lips, oral cavity or oropharynx.[2] Most oral cancers start in squamous cells, the thin, flat cells that line the lips, oral cavity, and oropharynx. Cancer that forms in squamous cells is called squamous cell carcinoma. Tobacco and alcohol, chronic inflammation, UV radiation, human papilloma virus or *Candida* infections, immunosuppression, genetic predisposition and diet are associated with tumour development.[3] Surgery, radiation and chemotherapy are the various strategies successfully employed for the management of cancer and yet they are not without limitations.[4] Several shortcomings, such as severe side effects of radiation and chemotherapy have lead to the rise in interest in Complementary and Alternative Medicine (CAM). The search for natural compounds, that show evidence of low toxicity towards normal cells while effectively killing cancer cells, has emerged as an important area of cancer research in recent times.[5] Flavonoids are a large group of polyphenolic compounds that occur ubiquitously in plants as secondary metabolites. Around 9,000 types of flavonoids are classified in human food and are found in fruits and vegetables.[6] A plethora of epidemiological, preclinical and clinical studies have established the beneficial effects of these compounds.[7] Kaempferol is an abundant flavonoid found in a wide variety of food components such as, tea (*Camellia sinensis*), strawberries (*Fragaria ananassa*), green chillies (*Capsicum frutescens*), carrots (*Daucus carota*), pumpkins (*Cucurbita pepo*), brinjal (*Solanum melongena*), broccoli (*Brassica oleracea*), grape fruit (*Citrus paradisi*), apples (*Malus domestica*), beans (*Phaseolus vulgaris*) and onions (*Allium cepa*).[8-10] Epidemiological studies have shown an inverse relationship between the high intake of kaempferol-rich diet and the incidence of several types of cancer, namely, colon,[6] ovarian,[7] pancreatic,[8] gastric[9] and lung cancer.[11] Kaempferol has been reported to exert anti proliferative effect *in vitro* on a number of human cancer cell lines including human breast carcinoma (MCF-7) cells, bladder (EJ), colon (HT-29, HCT116), human hepatoma (HepG2 and Hep3B), human stomach carcinoma (SGC-7901) cells, human cervical carcinoma (HeLa and SiHa) cells and human lung carcinoma (A549) cells.[12-15] Kaempferol has been documented to alter several important components of cellular signal transduction pathways associated with apoptosis, angiogenesis, inflammation and metastasis. Significantly, while kaempferol has been reported to inhibit the growth and induce apoptotic cell death of cancer cells, it has also been shown to maintain normal cell viability and exert protection to normal cells.[10] In comparison to standard chemotherapeutic drugs, kaempferol is known to be less toxic to normal cells.[16-17] The anti-cancer properties of kaempferol have been attributed to its ability to induce apoptosis in cancer cells and inhibit cancer cell proliferation.[10] Apoptosis or

Programmed Cell Death is a natural process which plays a vital role in the maintenance of physiological balance between cellular proliferation and death. Disruption o is and resistance to therapy. Hence, targeting apoptosis in c c approach.[18] Cultured human cells have long been employed f compounds of interest on several toxicological and pharmacologica cancer. Several studies have documented the apoptosis-inducing ability of kaempferol in s of cancer cell lines, such as ovarian cancer cells, HCT116 human colon cancer cells, A549 lung cancer cells, human osteosarcoma U-2 OS cells and glioblastoma cells.[20-24] So far, the comparative effect of kaempferol in cancers and non-cancerous cells has not been reported. Hence, the present study was at evaluating the anticancer activity of kaempferol on an oral cancer cell line (KB) in comparison with its effect on normal buccal cells.

Objective or aim of your study should be present.

MATERIALS AND METHODS

Kaempferol was purchased from Sigma Aldrich n.

For explaining your methods keep the tense as past tense (preferable) throughout the manuscript

Cell lines and culture conditions

KB oral cancer cell line of human origin was obtained from National Centre for Cell Science, Pune, India. The cells were cultured in DMEM (Gibco), supplemented with 10% FBS (Gibco). Penicillin and streptomycin (Gibco) were also added to the medium to 1X final concentration from a 100X stock and maintained in CO_2 incubator with 5% CO_2 and 95% humidity. The cells at confluency were harvested using trypsin – EDTA (Gibco) and 10^5 cells were seeded into sterile 6-well and 96 well plates. In each well of the 6-well plates, a sterile cover slip was placed before the cells were seeded and were then incubated overnight in a CO_2 incubator with 5% CO_2 and 95% humidity atmosphere for adherence. The cells were then exposed to the test compounds and / or etoposide at a final concentration of 200 µM for 24 hours. After treatment, the cover slips from the 6-well plates were removed and placed on a glass slide and sealed with vaseline. These slides were used for various staining techniques, whereas in 96-well plates, the medium was removed, replaced with fresh medium and used for viability assays. Buccal cells isolated from healthy individuals were cultured following the method of Michalczyk *et al.*[25] Healthy individuals in the age group of 20-30 years, and not on any medication, were included in the study as voluntary donors of buccal cells. The study was scrutinized and approved by the Institutional Human Ethics Committee (Approval No. AUW/IHEC-14-15/XPD-08). After rinsing the mouth with 5 ml of Listerine mouthwash solution for 2-3 minutes, followed by rinsing with sterile saline (5 ml × 2), the buccal cavity was scrapped using sterile soft wood cell scrapers. The mouth was rinsed with 15 ml of sterile saline, which was collected in a dish. The scraper was washed with 10 ml of sterile saline into the same dish. The cells were spun at 3000g for 5 minutes and the pelleted cells were resuspended in 1ml sterile PBS. The cells were incubated for 30 minutes in 1X Penstrep (Penicillin-Streptomycin, from a 100X stock (MP Biomedicals, USA) with 12µg/ml flucanazole (Cipla) to get rid of any bacteria and fungi that may be contaminating the oral cavity. The cells were then washed twice with PBS and resuspended in DMEM supplemented with 2% FBS, 50U/ml penicillin and 50µg/ml streptomycin solution with 1.2µg/ml flucanazole in the culture flask. The buccal cells were incubated overnight at 37ºC in a CO_2 incubator (Napco, UK). Following this, the medium was replaced with one containing 5% FBS, antibiotics and flucanazole, as done earlier. The cells were incubated overnight at 37ºC in a CO_2 incubator (Napco, UK). The cells (10^5 cells / ml) were seeded in a 96-well plate and the suspension culture was incubated at 37ºC for 24 hours. At the end of the incubation period, the cells were treated with etoposide, in the presence and the absence of kaempferol at a final concentration of 200µM for 24 hours at 37ºC. The treated cells were harvested by rapid pipetting. The collected cells were centrifuged at 4000 rpm for 3 minutes, washed twice with PBS and finally suspended in PBS. The cell suspension was then used for the viability assays and staining analysis.

Optimization of Dose

In order to determine the optimal dose of kaempferol, cell viability assays namely, MTT and SRB, were carried out with 25 µM, 50 µM, 100 µM and 150 µM concentrations of kaempferol.

Cell viability assays

MTT[26] assay and SRB[27] assay were employed to assess the viability of KB cells exposed to the flavonoid kaempferol, both in the presence and absence of etoposide, a standard chemotherapeutic drug. To compare the effects of kaempferol on cancerous and non-cancerous cells, normal human buccal cells were also studied in a similar manner.

MTT dye reduction assay

The MTT [3-(4,5-dimethyl-thiazol-2-yl)-2,5-diphenyltetrazolium bromide] reduction assay works on the principle that mitochondrial succinate tetrazolium reductase system converts yellow coloured MTT into its purple formazon derivative.[26] The amount of dye produced is proportional to the number of live cells. After exposure of the cells to kaempferol and/or the standard drug, etoposide for 24 hours, the medium was removed and the cells were treated with 50µl of MTT and incubated for 3 hours at 37ºC with mild shaking. At the end of the incubation period, 200µl of PBS was added to all the samples and the liquid was carefully aspirated. Acid-propanol (200 µl) was added to the wells and left overnight in the dark. The absorbance was read at 650nm in a microtitre plate reader (Bio-rad, USA). The optical density of the oxidant-induced cells was

fixed as 100% viability and the per cent viability of the cells in the other treatment groups were calculated relative to this.

SRB assay

Sulphorhodamine B (SRB) is a pink coloured aminoxanthane dye with two sulphonic groups. In TCA fixed cells, SRB binds to basic amino acids in the proteins, under midly acidic conditions, to provide a sensitive index of cellular protein content, which is directly proportional to cell viability.[27] After treatment of the cells, the medium was completely removed from each well and washed with 200µl PBS to remove any trace of medium and serum. Ice cold 40% TCA (350µl) was layered on top of the cells and incubated at 4ºC for one hour, after which the cells were washed 5 times with cold PBS (200 µl). SRB stain (350 µl) was added to each well and left in contact with the cells for 30 minutes at room temperature. The cells were then washed 4 times with 350 µl of 1% acetic acid to remove any unbound dye. Then, 350µl of 10 mM Tris (pH 10.5) was added to solubilize the protein-bound dye and was shaken gently for 20 minutes on a gyratory shaker. The tris layer in each well was transferred to a new 96-well plate and the absorbance was read in a microtitre plate reader (Bio-rad, USA) at 496 nm. The optical density of the oxidant-induced cells was fixed as 100% viability and the percent viability of the cells in the other treatment groups were calculated relative to this.

Acridine orange and ethidium bromide staining

Acridine orange and ethidium bromide staining (AO / EtBr) staining technique was used to differentiate between quiescent and actively proliferating cells. In this technique, apoptotic cells, which have a larger fraction of DNA in the denatured form, display an intense red fluorescence and a reduced green emission when compared to non-apoptotic cells.[28] To the treated cells in cover slips, 10 µl of AO / EtBr was added and spread by mounting a cover slip over it. The stained slides were incubated at room temperature for 5 minutes. The apoptotic cells with condensed chromatin and fragmented nuclei were identified by their red fluorescence and the normal cells were visualized by their green fluorescence, which was counted using an upright fluorescence microscope (Nikon, Japan) using B2A filter at 400X magnification.

Propidium iodide staining

The protocol elaborated by Sarker *et al.* [29] was followed to differentiate between apoptotic and normal cells wherein propidium iodide binds to DNA in cells undergoing death and enables the observation of nuclear changes in apoptotic cells. After exposure to the oxidant/compound, the KB cells were permeabilised with acetone:methanol (1:1) mixture at -20ºC for 30 minutes in the dark. Apoptotic cells were scored using the green filter of a flourescence microscope (Nikon, Japan) at 400X.

Cell cycle analysis

The distribution of kaempferol treated KB cells in various phases of the cell cycle was analyzed by flow cytometry using PI stain. KB cells were exposed to 50µM kaempferol for 24 hours. After treatment, the cells were pelleted, washed with phosphate-buffered saline (PBS) and fixed in 70% ethanol at -20ºC. The fixed cells were then resuspended in PI solution (50µg/ml) containing sodium citrate (10mg/10ml), RNase (400µg/10ml) and tween 20 (3µl/10ml). Stained cells were analyzed in FACSverse flow cytometer (BD Biosciences, USA). The FACSuite software program was used to determine the percentage of cells in different phases of the cell cycle.

Statistical analysis

The results are presented as mean ± SD of triplicates. The various parameters analyzed were subjected to statistical analysis using SigmaStat (Version 3.1). Statistical significance was determined by one-way ANOVA, with $p<0.05$ being considered significant.

RESULTS

Optimization of Dose

KB cells were exposed to various doses of kaempferol (25 µM, 50 µM, 100 µM and 150 µM) for 24 hours and the viability of the cells was tested by MTT and SRB assays (Graphs 1A & 1B). It was found that, with an increase in the concentration of kaempferol, there was a marked decrease in the viability of the treated cells. The IC_{50} value for kaempferol was found to be 50 µM in KB cells. Hence further studies were carried out with this concentration.

Effect of kaempferol on the viability of KB oral carcinoma and normal buccal cells

In this study, the cytotoxicity of kaempferol on KB oral cancer cells and its comparative effect on normal human buccal cells was assessed using MTT and SRB assays. As shown in Graphs 2A & 2B, the viability of KB cells decreased significantly on treatment with kaempferol which further reduced in the presence of etoposide, whereas in normal buccal cells the extent of cell death was comparatively low (Graphs 3A & 3B).

Observation of nuclear changes in apoptosis following kaempferol treatment

KB cells and normal buccal cells treated with kaempferol were stained using various dyes that stain apoptotic nuclei such as propidium iodide, acridine orange and ethidium bromide. The nuclear changes in apoptosis were followed in the presence and the absence of kaempferol (50µM) and/or the standard chemotherapeutic agent (etoposide). The results revealed that the number of apoptotic events increased in KB cells treated with etoposide and the proportion of dying cells increased in the presence of the selected compound, kaempferol, when compared to the group treated with etoposide alone (Graphs 4A & 4B; Figure 1). As shown in Graphs 5A & 5B and Figure 2, the number of apoptotic events observed was lower in normal buccal cells than that noted in oral cancer cells (KB).

Induction of cell cycle arrest by kaempferol in KB cells

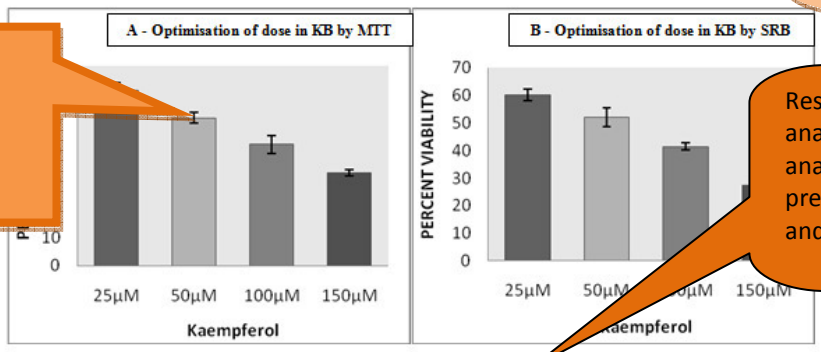
To investigate the ability of kaempferol to bring about cell cycle arrest in specific phases, KB cells were treated with 50 µM kaempferol for 24 hours, stained with PI and analyzed using flow cytometry. The distribution of cells in various phases of the cell cycle, namely, G_0-G_1, S and G_2/M, was significantly different between the control and kaempferol treated groups (Graphs 6 & 7). Kaempferol treatment resulted in a significant increase in the percentage of cells in G_2/M phase of the cell cycle.

All graphs, figures and table should have caption and foot notes

Graph 1

Optimisation of dose of kaempferol in KB oral carcinoma cel

Standard Error Bar (SEM) would be preferable for bar charts

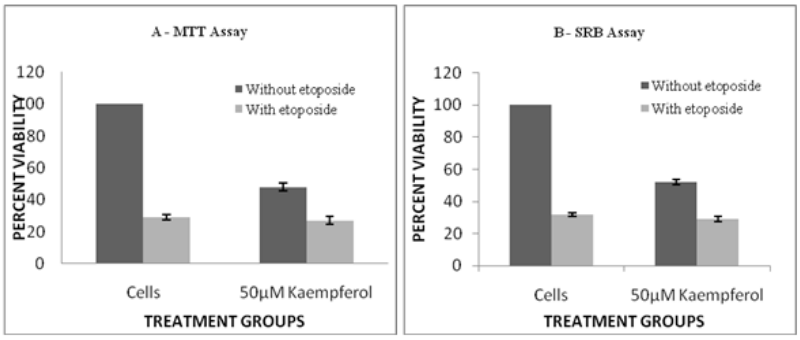


Results should preferentially analysed by suitable statistical analysis .Its SD or SEM should be present in foot notes with p value and if significant or not.

The values are Mean ± S.D. of triplicates(P<0.001)

Graph 2

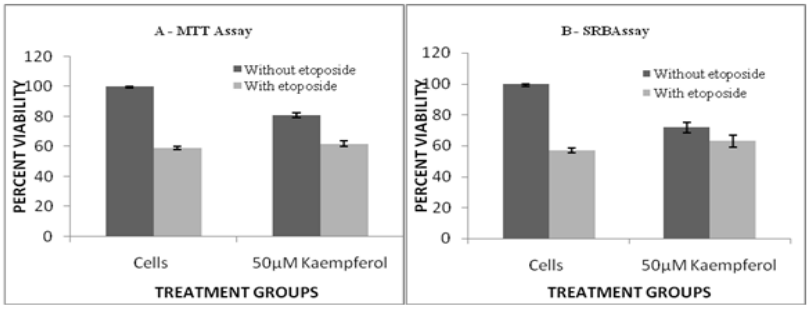
Effect of kaempferol on the viability of KB oral carcinoma and normal buccal cells



The values are Mean ± S.D. of triplicates(P<0.001)

Graph 3

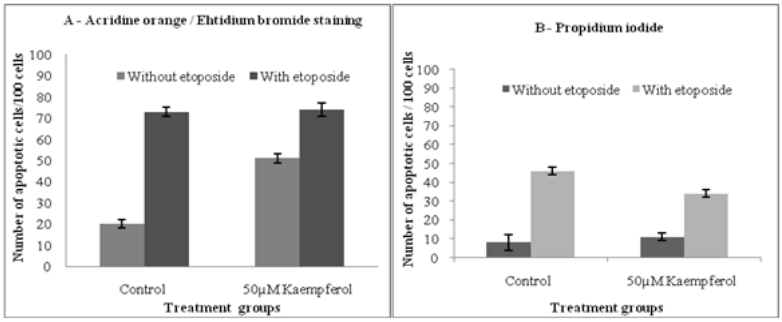
Effect of kaempferol on the viability of normal buccal cells



The values are Mean ± S.D. of triplicates(P<0.001)

Graph 4

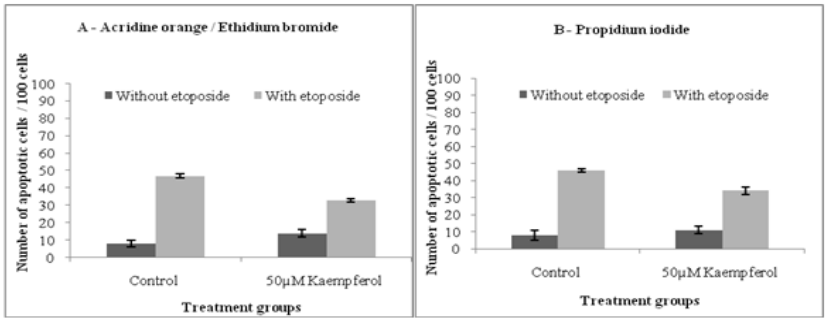
Effect of kaempferol on nuclear changes in KB oral carcinoma cells



The values are Mean ± S.D. of triplicates(P<0.001)

Graph 5

Effect of kaempferol on nuclear changes in cultured buccal cells



The values are Mean ± S.D. of triplicates(P<0.001)

Figure 1

Effect of Kaempferol on Nuclear Changes in KB Oral Carcinom orange / Ethidium bromide staining Propidium iodi

Photographs/figures must be clear and sharp and should be of higher resolution (300 dpi or higher) and in JPEG,GIF,TIFF or Provide

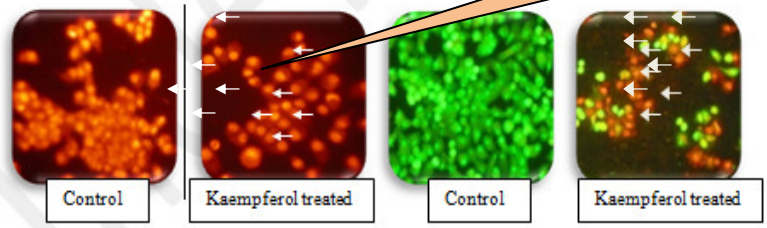


Figure 2

Effect of Kaempferol on Nuclear Changes in Cultured Buccal Cells Acridine orange / Ethidium bromide staining Propidium iodide staining

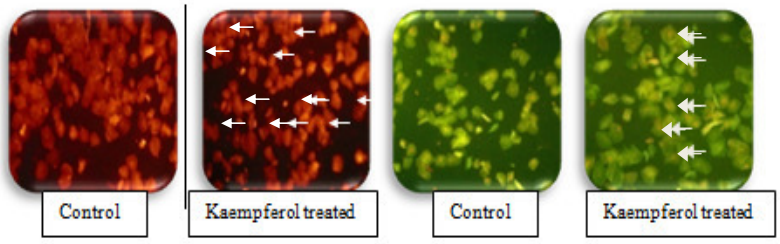


Table 1
The relative quantitation of hemolymph and supernatant of

Provide relevant ±SD values for your observation.

Sample	Concentrations µg/ml				
	10 µg/ml	20 µg/ml	30 µg/ml	40 µg/	50µ µg/ml
Control	0.00±0	0.00±0	0.00±0	0.00±0	0.00±0
Hemolymph	63.68±2.3 ^{a***}	76.20±2.9 ^{a***}	84.73±3.5 ^{a***}	97.09±0.12 ^{a***}	97.30±0.04 ^{a***}
Supernatant	47.37±1.9 $^{a*** b***}$	49.7± 1.3 $^{a*** b***}$	56.00±3.2 $^{a*** b***}$	71.40± 3.9 $^{a*** b***}$	74.8± 2.6 $^{a*** b***}$

The values are expressed in Mean ± S.D; significant* $p \leq 0.05$; highly significant *** $p \leq 0.01$; not significant^{ns} $p \geq 0.05$; a – Control vs Hemolymph and Supernat - Hemolymph vs Supernatant.

In the foot notes of the table we could not find "P- Value" and "n value" for your observations. You need to give explanation for that below the table.

Groups	Treatment	Mean ulcer index± SD	Mean volume of Gastric secretion ± SD	Total acidity (mEq/l/100gm)
A	Normal saline	0.1308 ± 0.04	2.633 ± 0.62	79.53 ± 3.34
B	Ranitidine	0.0234± 0.004*	1.316 ± 0.360 *	44.25 ± 4.67 *
C	*Tinospora cordifolia* (400mg/kg)	0.0348 ± 0.005*	1.483 ± 0.541	47.41 ± 3.87 *

Values are mean ± SD ; (n = 6)
** P< 0.01 when compared with control*

'P' value and 'n' value

In Discussion, results should be discussed towards achievement of your objective with citing references/literatures. Its better to have atleast few recent references to support that your work is of recent research

DISCUSSION

Kaempferol, a phytoestrogen has been reported to inflammatory, anti-oxidant and anti-cancer activities.[30-32] M to quantify cell viability and /or cytotoxicity.[19] In a study by and green tea extract on tumor cell lines, namely, MDA-MB-30 by the use of colorimetric method based on tetrazolium salt. Similarly, MTT assay was conducted to determine the growth inhibitory effect of ethanolic extracts of Papua's ant nest (*Myrmecodia tuberosa*) on KB oral carcinoma cells, which showed that, increasing concentrations of the extract resulted in decrease in the number of viable KB cells. Our results are also in accordance with report by Su *et al.*[34] which revealed that the flavonoid quercetin treatment significantly suppressed the proliferation of the bladder cancer cell line UMUC3 with IC_{50} 42.29 µM. Yanqiu *et al.*[35] showed the synergistic inhibition of the growth of Human myelogenous leukemia K562 cells and the adriamycin resistant variant K562/A cells by the flavonoids, quercetin and kaempferol, in a dose-dependent manner. Som *et al.*[36] analyzed the effects of quercetin on HL-60 leukemic cells by MTT approach and revealed that quercetin led to a dose-dependent inhibition on cell proliferation.Azevedo *et al.*[37] demonstrated the antiproliferative activity of kaempferol on MCF-7 breast cancer cells by SRB assay. Similarly, delphinidin, cyanidin, two of the most abundant anthocyanins found in bilberry, and trolox were shown to inhibit MCF7 breast cancer cell proliferation as determined by SRB assay.[38] In another study by Martı´nez-Pe´rez *et al.*,[39] the flavonoid oncamex brought about a significant reduction in cell density of MCF-7, MDA-MB-231, BT-549 and HBL-100 breast cancer cell lines. Studies on the effect of kaempferol on human-derived hepatoma cell lines HepG2 and Hep3B as well as on HCT-116 colon cancer cells using SRB assay revealed a significant reduction in cell viability.[40] Our results, thus, lend further support to the idea that kaempferol possesses potent antiproliferative effect on several cancer cell lines.In addition to this observation, our results showed that kaempferol exhibited protective effect in normal buccal cells and cytotoxicity in KB oral carcinoma cells. Faedmalek *et al.*[38] reported that nano-silver treatment showed stronger inhibitory effect in HepG2 liver cancer cells compared to mice primary liver cells. Kiruthika and Padma[41] showed that the aqueous, methanol and chloroform extracts of *Zea mays* leaves induced apoptosis in Hep2 cells, while exerting protective effect in non-cancerous primary chick embryo fibroblasts. Eriodictyol, a naturally occurring flavanone was studied for its antiproliferative activity against human hepatocellular carcinoma cells (Hep-G2) and normal liver hepatocyte cell line (AML12) and was shown to lead to a significant and concentration dependent growth inhibition of liver cancer cells with IC50 value of 37.6 µM while displaying very less cytotoxicity against AML12 normal liver hepatocyte cells.[42] Dual acridine orange/ethidium bromide (AO / EtBr) fluorescent staining is used to identify apoptosis-associated changes in cells and to discriminate between different stages of apoptosis. Viable cells appear light green, early apoptotic cells display bright green fluorescence and condensed chromatin while late apoptotic cells exhibit orange fluorescence and nonviable cells show red colored fluorescence. Studies on the cytotoxic effects of nimbolide, a limonoid present in leaves and flowers of the neem tree (*Azadirachta indica*) on human breast cancer cells MCF-7 and MDA-MB-231 by AO / EtBr dual staining showed significant morphological changes characteristic of apoptotic cells, such as cell

shrinkage and reduced cell density.[43] IMF-8, a dimethylamino substituted iminoflavone was demonstrated to possess comparable anticancer activity with standard drug doxorubicin when assessed by the AO / EtBr staining and the percentage of apoptotic cells after treatment was shown to be significantly higher compared to control in HCT- 116 cells.[44] Lv *et al*.[5] examined the apoptotic events in MDA-MB-231 and MCF-7 human breast cancer cells treated with curcumin and reported that AO / EtBr staining revealed intact nuclei in control cells as opposed to apoptotic nuclei and chromosomal condensation in treated cells. In line with these reports, the results of the present study showed that the exposure of KB oral carcinoma cells to kaempferol resulted in a significant raise in the number of apoptotic cells when compared to control cells.Nuclear fragmentation is a characteristic feature of cells undergoing apoptotic death. Propidium iodide is a fluorescent DNA binding dye which is used to examine the morphological changes in nuclei apoptotic cells. Mohankumar *et al*.[45] determined the cytotoxicity of an ortho-hydroxy substituted analog of curcumin (BDMC-A) in MCF 7 breast cancer cells and revealed that PI was taken up by treated cells and fragmented nuclei were observed in higher numbers than untreated cells. Jambunathan *et al*.[46] reported that the administration of methanol extract of leaves and the rhizomes of *C. amada* induced apoptosis in the breast cancer cell lines MCF-7 and MDA MB 231 and demonstrated differential effect in non-cancerous breast cells HBL-100 as observed by propidium iodide staining. Kiruthika *et al*.[47] quantified the nuclear changes in Hep2 cells caused by etoposide and its modulation in the presence of *Zea mays* leaf extracts using propidium iodide staining and the results showed that *Zea mays* leaf extract exhibted anticancer property and enhanced the chemotherapeutic action of etoposide in cancer cells.To examine whether kaempferol regulates cell cycle progression in KB oral carcinoma cells, flow cytometric analyses was carried out. Observation of the distribution of cells in the various phases of cell cycle indicated a G_2/M cell cycle arrest. Our results are in agreement with previous reports that demonstrated induction of cell cycle arrest at G_2/M phase by kaempferol in SK-HEP-1 human hepatic cancer cells.[16] In another study by Cho and Park[48] HT-29 human colon cancer cells were found to be arrested in G1 phase within 6 h and in G_2/M phase at 12 h of exposure to kaempferol. Morover, kaempferol has been shown to induce arrest of renal cell carcinoma (RCC) cells mainly at G_2/M stage of cell cycle after treatment for 24 hours.[49] Jaramillo-Carmona *et al*.[50] reported that kaempferol augmented the chemo preventive efficacy of quercetin through the arrest of HCT-116 human colon cancer cells in the G_2/M cell cycle phase.

CONCLUSION

Considered together, all these observations further strengthen our conclusion that kaempferol possesses potential antiproliferative activity against KB oral carcinoma cells and has a differential effect on normal buccal cells. Futher, our results show that kaempferol significantly induces apoptotic cell death in oral carcinoma cells leading to G_2/M cell cycle arrest. Collectively, our findings indicate the potency of kaempferol as a future candidate for the development of oral cancer chemotherapeutic agent, which can induce death in cancer cells while protecting the non-cancerous cells.

ACKNOWLEDGEMENTS

This study was supported by research grant from Department of Science and Technology, Government of India under the WOS-A scheme.

CONFLICT OF INTEREST

Conflict of interest declared none.

AUTHORS CONTRIBUTION STATEMENT

……https://goo.gl/j49MB1

Include author contribution statement before References section to know each authors contribution to the paper. You can visit or click this link (copy and paste the link in the browser) (https://goo.gl/j49MB1) to see some of the examples.

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