Tropical Journal of Pharmaceutical Research January 2024; 23 (1): 77-83
ISSN: 1596-5996 (print); 1596-9827 (electronic)

Available online at http://www.tjpr.org
http://dx.doi.org/10.4314/tjpr.v23i1.10

Original Research Article

Cytotoxicity of luteolin, a flavonoid compound isolated from *Anthemis palestina*

Yahia Z Tabaza[1]*, Zuh-Kyung Seong[1], Young-Mi Kim[2], Walhan Alshaer[3], Talal A Aburjai[1]

[1]Department of Pharmaceutical Sciences, School of Pharmacy, The University of Jordan, Amman 11942, Jordan, [2]College of Pharmacy and Research Institute of Pharmaceutical Sciences, Seoul National University, 1, Gwanak-ro, Gwanak-gu, Seoul 08826, Republic of Korea, [3]Cell Therapy Center, The University of Jordan, Amman 11942, Jordan

For correspondence: **Email: y.tabaza@ju.edu.jo; **Tel:** +962-6-5355000; **Fax:** +962-6-5300250*

Sent for review: 2 May 2023 *Revised accepted: 3 January 2024*

Abstract

Purpose: *To determine the active principle responsible for the cytotoxic effect of Anthemis palestina (Reut. ex Kotschy) Reut. ex Boiss. (Asteraceae).*
Methods: *A bioassay-guided fractionation was used to isolate the active principle, luteolin, which structure was elucidated using ^{1}H and ^{13}C NMR. The cytotoxic effects of luteolin and doxorubicin (positive control) in the breast cancer cell lines MDA-MB-231 and MCF-7, and in normal fibroblasts, were determined using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide (MTT) assay.*
Results: *Luteolin was isolated from the ethyl acetate extract of the aerial parts of Anthemis palestina. With an IC_{50} value of 14.91 ± 5.77 µM for MDA-MB-231 cells, luteolin was less active than doxorubicin. However, with respect to MCF-7 cells, there was no significant difference in the cytotoxicity values of luteolin and doxorubicin, with IC_{50} value of 29.28 ± 11.85 µM for luteolin. However, with an IC_{50} value of 51.39 ± 18.51 µM against fibroblasts, luteolin was significantly safer than doxorubicin.*
Conclusion: *Luteolin might be responsible for the cytotoxicity of Anthemis palestina. The high level of luteolin cytotoxicity indicates the potential benefits of Anthemis palestina, not only in terms of its taste, but also for its likely positive therapeutic impact on cancer, especially breast cancer.*

Keywords: *Cancer, Breast cancer, Cytotoxicity, Anthemis palestina, Luteolin*

Tropical Journal of Pharmaceutical Research is indexed by Science Citation Index (SciSearch), Scopus, Web of Science, Chemical Abstracts, Embase, Index Copernicus, EBSCO, African Index Medicus, JournalSeek, Journal Citation Reports/Science Edition, Directory of Open Access Journals (DOAJ), African Journal Online, Bioline International, Open-J-Gate and Pharmacy Abstracts

INTRODUCTION

Cancer and cardiovascular diseases are considered the leading causes of premature death globally, and it has been projected that cancer will be the major cause of premature death by the end of this century [1]. Therefore, it is obvious that cancer is a problem that should be tackled with utmost seriousness. As at 2020, breast cancer was the most frequently diagnosed type of cancer worldwide, followed by lung, liver, stomach and colon cancers [1]. In Jordan, mortality from cancer ranks second to that of coronary artery diseases [2]. This could be attributed to many risk factors, e.g., unhealthy lifestyles such as poor diet and lack of physical activity; exposure to various carcinogens, and last but not least, the extensive use of tobacco

products among Jordanians [2]. Consistent with the global trend, the most frequently occurring types of cancer in Jordan are breast, colorectal and lung cancers [3].

Natural products and their derivatives are major resources in the drug discovery process, especially for anticancer and cytotoxic agents. This may be attributed to several features of natural products, e.g., structural diversity, potential to target many receptors, presence of wide ranges of pharmacophores, and vastness of stereochemistry. Moreover, natural products have potential for delivery through many cellular transporter systems, and they conform with Lipinski's rule of five which enhance their oral absorption from the gastrointestinal tract. In fact, with specific reference to cancer, the percent of new anticancer drugs that are either natural products or synthetic analogues of natural products, increased from 53.3 % between 1946 and 1980, to 64.9 % from 1981 to 2019 [4]. Therefore, in this study, the search for cytotoxic agents that are effective against the most widely diagnosed cancer type, i.e., breast cancer, was focused on natural products.

Anthemis palestina (Reut. ex Kotschy) Reut. ex Boiss., also known as Palestine Chamomile, belongs to the Asteraceae, or the so-called Compositae family. From March to June, this annual plant grows wide in Jordan, most commonly in fields, waste grounds, and mountainous regions in Jordan Valley, Irbid, Ajloun, Jarash, Salt, and Amman. Generally, plants that belong to the genus *Anthemis* are rich in flavonoids, terpenes, and volatile oils. Thus, these plants usually exhibit anti-inflammatory, hepatoprotective, antioxidant, and anti-cancer properties [5].

The natives of the West Bank and Jordan employ *Anthemis palestina* for treating spasms, bacterial infections, inflammation and oxidative stress [6,7]. Analysis of hydro-distilled essential oil obtained from *A. palestina* flowers revealed that it contained mainly terpenes, and results from Brine Shrimp Lethality test showed that it had cytotoxic properties [8]. In addition, a study of another oil derived from hydro-distillation of *A. palestina* flowers revealed that it exerted antioxidant, antibacterial, and antifungal properties [6]. In addition, it showed cytotoxic properties against human cervix adenocarcinoma (HeLa), human Burkitt lymphoma B (BJAB), and human colon adenocarcinoma (Caco-2) cell lines [6]. Moreover, the methanolic extract of *Anthemis palestina* demonstrated inhibitory activities against xanthine oxidase, hormone-sensitive lipase, and α-amylase, in addition to antioxidant activities [9-11]. Furthermore, the endophyte *Chaetomium subaffine* obtained from *Anthemis palestina* yielded four new compounds, i.e., chaetomisides A – D which showed cytotoxic activities against both ZR-75 mammary cancer and A549 pulmonary cancer cells [12]. However, no phytochemical investigation has been performed to identify the compound(s) that might be responsible for the important biological activities of *A. palestina*, especially the cytotoxic effects. Therefore, the present work was carried out to study *A. palestina* from a phytochemical point of view, and to isolate the bioactive constituent that might be responsible for its cytotoxic activity.

EXPERIMENTAL

Materials

Methanol (HPLC grade), acetonitrile, and hexane were purchased from LabChem (Zelienople, Pennsylvania, USA); HPLC grade ethyl acetate and dichloromethane were purchased from Honeywell (France), while luteolin standard was obtained from Sigma-Aldrich (USA).

Plant material

Anthemis palestina was collected in May 2019 from Alaluk, Zarqa Governorate, Jordan. The plant material was identified by a plant taxonomist, Prof. Dawud Al-Eisawi, Department of Biological Sciences, School of Science, The University of Jordan. A voucher specimen (201905-01-ANP) was deposited at the Department of Pharmaceutical Sciences, The University of Jordan. Additionally, the name of the plant was checked at www.theplantlist.org which was accessed in May 2022.

Extract preparation and fractionation

The dried and chopped aerial parts and roots of *Anthemis palestina* (300 g) were cleaned, mixed and extracted twice with 2 liters of ethyl acetate, with the aid of 2-h sonication, after which the extract was left at room temperature for 3 days. Thereafter, the extract solution was filtered and evaporated using a rotary evaporator, resulting finally in a yield of 29 g of dried extract. After that, the extract was fractionated using a gravity column (120 × 70 cm, 5 cm ID, n-silica gel). Elution was done with stepwise gradient of n-hexane in ethyl acetate (i.e., 0, 10, 20, 30, 50, 70 and 100 % ethyl acetate, each at a volume of 1.5 L, yielding 10 sub-fractions. Using TLC, the eighth fraction (1.8 g) was seen to constitute the main metabolite. Therefore, it was fractionated via flash chromatography (using Silica SL 50 µm

irregular 20-g cartridge), with a stepwise gradient of dichloromethane and methanol, i.e., from 100 % dichloromethane to 80 % dichloromethane and 20 % methanol over 30 min. This yielded three sub-fractions. Thereafter, 23 mg (0.08 % yield) of a pure compound, later identified as luteolin, was obtained from the second fraction using a 1.5 × 40 cm Sephadex® LH-20 column chromatography and elution with 100 % methanol. The column chromatography procedures were performed over NP-silica (Fluka Silica Gel 60 F_{254}, Switzerland) and Sephadex LH-20 (25 – 100 LM, Sigma-Aldrich, Steinheim, Sweden). Büchi Pure C-810 system and EcoFlex Silica SL 50 µm irregular 20 g cartridge (Büchi, Switzerland) were used for flash chromatography. Evaporation was carried out with a Heidolph Laborota 4000 rotatory evaporator (Germany).

NMR

A 5-mg specimen was solubilized in 0.60 mL of DMSO- d_6 acquired from Cell Bio (Korea) and placed in an NMR tube sourced from Norell (USA). The NMR spectra were recorded on a Varian UNITY 500 FT-NMR spectrometer (Varian, Inc., Palo Alto, CA), with tetramethyl silane as an internal standard. The resultant spectra were analyzed using MestReNova 14.2.1, a software developed by Mestrelab Research (Spain).

Thin layer chromatography (TLC)

Thin layer chromatography (TLC) on pre-coated Merck Kiesel-gel 60 F_{254} 0.25 mm plates was employed to pool fractions and to compare the compound obtained (luteolin) with a standard reference, using capillary tubes purchased from Hirschmann (Germany). After the TLC plates were sprayed with 10 % sulphuric acid and heated, visualization of the spots was done under ultra violet (UV) at λ of 254 nm. The solvents used are indicated in the section on *Materials*. The fractions were all dissolved in acetone and applied as spots on activated TLC plates prior to development of the chromatogram in a mobile phase which rose to a distance of 5 to 6 cm. Subsequently, the spots were visualized under UV light at a wavelength of 254 nm. Thereafter, the TLC plates were sprayed with a reagent containing 10 % sulfuric acid, and subsequently heated to 200 degrees Celsius.

Culture of cell lines

Parent mammary carcinoma cells MCF-7 and MDA-MB-231, and healthy fibroblasts (human normal dermal fibroblasts) were sourced from ATCC. Culturing of MCF-7 cell line was done in RPMI) 1640 medium (EuroClone, Italy), whereas the MDA-MB-231 cells were grown in Minimum Essential Medium (MEM). The fibroblasts were grown in DMEM. All the mentioned media contained glutamine (0.002 M), 10 % (v/v) heat-inactivated FBS, and 1 % penicillin-streptomycin. All the cell lines were incubated as adherent single layers at 37°C in a 5 % CO_2 tissue culture incubator (Memmert, Schwabach, Germany).

Cell viability assay

The anti-proliferative effects of the fractions and the isolated compound against the mentioned cell lines were determined using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide (MTT) assay, described as per Ghasemi *et al* [13]. The MDA-MB-231, MCF-7 and fibroblast cells were plated in 96-well plates (TPP, Switzerland), with each well having approximately 8×10^3 cells. Samples of each cell line were either left untreated as negative control, exposed to doxorubicin as a positive control, or exposed to varying concentrations of the test samples. The test samples consisted of the crude extract and the fractions obtained after the chromatographic separation of the extract (62.2 µg/mL each), including the isolated compound, luteolin. Luteolin was used at concentrations of 0.9, 1.7, 3.4, 6.8, 13.7, 27.3, 54.7, 109.4, 218.8, 437.5, 875.0, and 1750.0 µM. Untreated cell lines served as controls. Following a 72-h incubation at 37 °C in a 5 % CO_2 atmosphere, the culture medium in each well was replaced with 100 µL of fresh medium containing MTT (Promega, USA). After a 3-h incubation at 37 °C, the MTT medium was discarded, and the resultant formazan crystals were dissolved in dimethyl sulphoxide (DMSO). The optical density of the solution in each well was read in a Glomax microplate reader at 560 nm. All bioassays were repeated thrice. Then, the half-maximal inhibitory concentration (IC_{50}) of luteolin was calculated using GraphPad Prism 7 (USA).

Statistical analysis

Student's *t*-test was conducted using Minitab 21 (Minitab LLC, USA), and the data are presented as mean ± SD. Statistical significance was assumed for *p*-values less than or equal to 0.05.

RESULTS

Structural identification and analysis of luteolin

The isolated compound obtained was a yellowish powder. Its identification was carried out with

both ^{1}H and ^{13}C NMR. Thereafter, the NMR data were compared with published data, and the isolated compound was identified as luteolin (Figure 1 and Table 1).

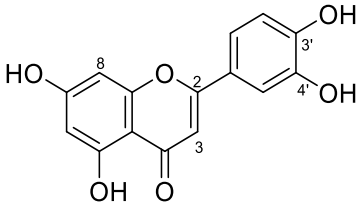

Figure 1: Luteolin structure

The identity of luteolin was further confirmed by direct comparison and co-elution with a standard reference sample using TLC in a solvent system of n-hexane: ethyl acetate (1:4 v: v).

In vitro cytotoxic effect of *Anthemis palestina*

The MTT assay was implemented to investigate the cytotoxic effects of *Anthemis palestina* extract and fractions, in addition to the isolated compound luteolin. The results of this assay are shown in Figures 2. Fraction 8 exhibited cytotoxic effects against both breast cancer cell lines, resulting in a total viability of less than 0.2 for each cell line. Due to its aforementioned cytotoxic effect and the fact that it contained the major metabolite, as shown in TLC, this fraction was subjected to further fractionation. In addition, it is worthy of note that fractions 1, 3, and 4 also exhibited cytotoxic activity against the tested cell lines. However, these fractions were put aside due to their high chlorophyll contents.

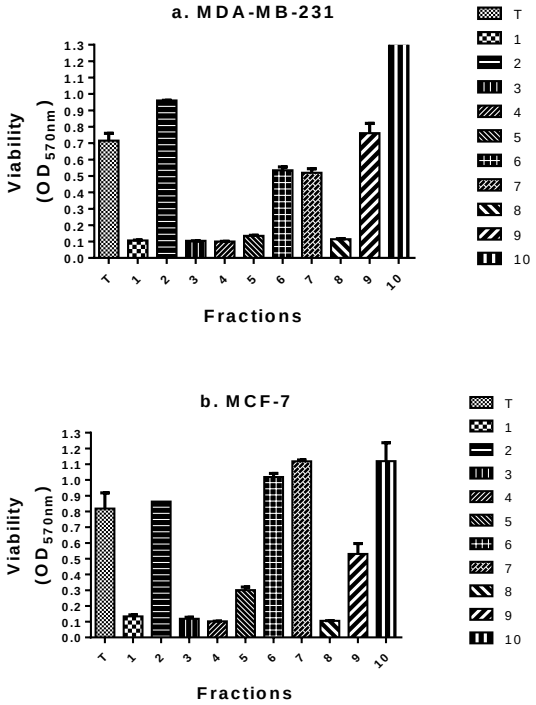

Figure 2: *In vitro* cytotoxic activity of crude extract (T) of *Anthemis palestina* and fractions (1 – 10) against the 2 cell lines (a & b)

Table 1: ^{1}H and ^{13}C NMR data for luteolin in relation to literature

Position	**Compound 1 in DMSO**		**Luteolin in $CDCl_3$ [14]**	***^{13}C NMR (100 MHz)***
	^{1}H NMR (500 MHz)	***^{13}C NMR (126 MHz)***	***^{1}H NMR (400 MHz,)***	
2		163.9		163.9
3	6.70 (1H, s)	103.7	6.66 (1H, s)	103.7
4		181.7		181.7
5		157.3		157.3
6	6.44 (1H, d, *J* = 1.8 Hz)	98.8	6.43 (1H, d, *J* = 2.0 Hz)	98.8
7		164.1		164.1
8	6.18 (1H, d, *J* = 1.8 Hz).)	93.8	6.17 (1H, d, *J* = 2.0 Hz).)	93.8
9		161.4		161.5
10		102.9		102.9
1’		118.9		119.0
2’	7.42 (1H, d, *J* = 2.3 Hz)	113.4	7.41 (1H, d, *J* = 2.2 Hz)	113.4
3’		149.7		149.7
4’		145.7		145.7
5’	6.88 (1H, d, *J* = 8.2 Hz)	116.0	6.80 (1H, d, *J* = 8.2 Hz)	116.0
6’	7.41 (1H, dd, *J* = 10.8, 2.3 Hz)	121.5	7.38 (1H, dd, *J* = 8.2, 2.2 Hz)	121.5
5-OH	12.97 (1H, s)		12.97 (1H, s)	
7-OH	10.84 (1H, s)		10.82 (1H, s)	
3’-OH	9.41 (1H, s)		9.41 (1H, s)	
4’-OH	9.931H, s)		9.91 (1H, s)	

Table 2: IC_{50} values of doxorubicin and luteolin against the various cell lines

Cell line	IC_{50} of doxorubicin (µM)	IC_{50} of luteolin (µM)
MDA-MB-231	0.28±0.09*	14.91±5.77*
MCF-7	0.14±0.03	29.28±11.85
Fibroblasts	1.80±0.90*	51.39±18.51*

Note: Values are presented as mean ± SD. *Significant difference at $p \leq 0.05$

In vitro cytotoxic effect of luteolin

The IC_{50} values for the positive control, doxorubicin, and the pure compound, luteolin, were calculated based on the percent of the remaining cells in three different experiments. The IC_{50} values are shown in Table 2 and in the dilution curves in Figure 3.

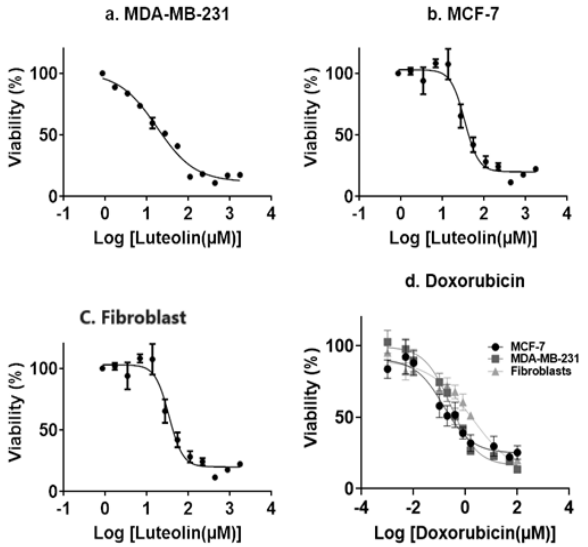

Figure 3: Dilution curves for luteolin (a - c) and doxorubicin (d) (in molar concentrations) used for the determination of IC_{50} values against the cell lines

DISCUSSION

In this phytochemical study of *Anthemis palestina*, a bioassay-guided fractionation approach was implemented through targeting various breast cancer cells. This led to the isolation of luteolin which was identified using ^{1}H and ^{13}C NMR, in addition to TLC. Thus, luteolin is the first pure compound to be isolated and identified from this plant. The cytotoxic effect of luteolin was established by comparison with doxorubicin which served as positive control. When the IC_{50} data of luteolin and doxorubicin were compared, it was shown that although luteolin exhibited good cytotoxic effect against MDA-MB-231 cells (with IC_{50} value of 14.91 ± 5.77 µM), it was still significantly less effective than doxorubicin. However, the LD_{50} value of luteolin against the other breast cancer cell line (MCF-7) was 29.28 ± 11.85 µM, indicating comparable effectiveness with doxorubicin. However, what sets luteolin apart from doxorubicin is its safety, as revealed in the IC_{50} values of both compounds against the normal human dermal fibroblasts (with IC_{50} values of 1.80 ± 0.90 µM and 51.39 ± 18.51 µM for doxorubicin and luteolin, respectively). This indicates the possibility of further investigations on luteolin as a safe anti-cancer agent.

Luteolin is a polyphenolic flavonoid compound. It is commonly found in various edible plants, including fruits, vegetables, and medicinal herbs [15]. Flavonoids are usually present in the vacuoles of plant cells, and their importance for the host organisms is well-known as they serve as antioxidants, anti-inflammatory agents, detoxicants, UV-filters, signaling molecules, and defense molecules [16]. In addition, flavonoids are of great benefit to humans when consumed in the diet, due to their antioxidant, anti-microbial and even estrogenic regulatory properties [16]. Moreover, flavonoids exert anticancer activity in humans via modification of the activities of ROS-scavenging enzymes and their role in regulation of cellular metabolism, e.g. cell cycle blockage, apoptosis induction, autophagy, and anti-proliferative effects [15].

In China, plants rich in the flavonoid luteolin have been used traditionally for cancer treatment [15,17]. It has been suggested that the biological activities of luteolin are functionally interrelated. For example, there is a link between its anticancer and anti-inflammatory activities [15]. Furthermore, the anti-cancer property of luteolin is linked to stimulation of apoptosis, as well as suppression of cell proliferation, metastasis and angiogenesis [15]. Moreover, luteolin inhibits several cell survival routes, e.g., PI3K/Akt, Nf-kB), and X-linked inhibitor of apoptosis protein (XIAP), leading to sensitization of cancer cells to induced cytotoxicity [15]. Hence, it has been proposed that luteolin could function as an anticancer agent for different cancer types and also as a cancer preventive agent [15].

The *in vivo* and *in vitro* cytotoxic effects of luteolin have already been reported in different cancers, including cancers of the breast, colon, pancreas, prostate, mouth, lung, kidney, cervix, placenta, ovary, skin, liver, gastric mucosa, esophagus, and bladder [17]. In MDA-MB-231 human breast cancer cells, luteolin regulated miRNAs and produced inhibition of cell cycle, tube formation, and the expression of Notch signaling-related proteins, in addition to apoptosis and cell cycle phase arrest [12, 17].

On the other hand, for MCF-7 cell lines, luteolin exhibited high cytotoxicity by decreasing ROS levels, increasing the anti-apoptotic protein B cell lymphoma, inhibiting IGF-1-stimulated MCF-7 cell proliferation, blocking the cell cycle, triggering apoptosis, regulating the estrogen signaling pathway, and suppressing the p-EGFR, p-Akt, p-STAT3 and p-Erk1/2 pathways [17]. These findings may explain the cytotoxic activity of luteolin against both MDA-MB-231 and MCF-7 cells as demonstrated in this work.

The consumption of biologically active molecules from the diet does indeed have a beneficial effect against several diseases, including cancer. One of these bioactive molecules is luteolin which has been shown to play therapeutic and preventive roles against various cancer types, including breast cancer [17]. Hence, luteolin may be used as a complementary therapy for cancer, especially as part of a diet of edible plants that contain this flavonoid compound. It is worthy of note that luteolin has also been isolated from other species of *Anthemis*, namely, *Anthemis chia*, *Anthemis cretica* and *Anthemis monantha* [18].

CONCLUSION

This study reports the isolation of a natural product luteolin from *Anthemis palestina.* The isolated compound, i.e., luteolin, exhibits significant cytotoxic effect on MDA-MB-231 and MCF-7 mammary carcinoma cells, pinpointing the benefits of *Anthemis palestina* not only for its dietary use but also for its positive impact on health, particularly, breast cancer.

DECLARATIONS

Acknowledgements

The authors would like to appreciate the support of the Deanship of Scientific Research at The University of Jordan (7/2018-2019, 96/2018-2019).

Funding

This work was generously funded by the Deanship of Scientific Research at The University of Jordan (7/2018-2019, 96/2018-2019).

Ethical approval

None provided.

Availability of data and materials

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

Conflict of Interest

No conflict of interest associated with this work.

Contribution of Authors

The authors declare that this work was done by the authors named in this article and all liabilities pertaining to claims relating to the content of this article will be borne by them. Yahia Z Tabaza conceptualized the work, obtained the funds, performed the extraction and the statistical analysis, and took part in writing and revising the manuscript. Zuh-Kyung Seong carried out the lab work, analyzed the results, and took part in writing and revising the manuscript. Young-Mi Kim performed the NMR analysis. Walhan Alshaer conducted the bioassays. Talal A Aburjai supervised the work and revised the manuscript. All authors read and approved the final manuscript.

REFERENCES

1. *Cao W, Chen H-D, Yu Y-W, Li N, Chen W-Q. Changing profiles of cancer burden worldwide and in China: a secondary analysis of the global cancer statistics 2020. Chin Med J (Engl) 2021; 134(07): 783–791.*
2. *Abdel-Razeq H, Attiga F, Mansour A. Cancer care in Jordan. Hematol Oncol Stem Cell Ther 2015; 8(2): 64–70.*
3. *Globocan. Jordan fact sheets 2020.*
4. *Newman DJ, Cragg GM. Natural products as sources of new drugs over the nearly four decades from 01/1981 to 09/2019. J Nat Prod 2020; 83(3): 770–803.*
5. *Boukhary R, Aboul-ElA M, El-Lakany A. Review on chemical constituents and biological activities of genus Anthemis. Pharmacogn J 2019; 11(5): 1155–1166.*

6. Bardaweel SK, Tawaha KA, Hudaib MM. Antioxidant, antimicrobial and antiproliferative activities of Anthemis palestina essential oil. BMC Complement Altern Med 2014; 14: 1–8.
7. Jaradat NA, Ayesh OI, Anderson C. Ethnopharmacological survey about medicinal plants utilized by herbalists and traditional practitioner healers for treatments of diarrhea in the West Bank/Palestine. J Ethnopharmacol 2016; 182: 57–66.
8. Tawaha KA, Alali FQ, Hudaib MM. Chemical composition and general cytotoxicity evaluation of essential oil from the flowers of Anthemis palestina Reut. ex Boiss. Growing in Jordan. J Essent Oil-Bearing Plants 2015; 18(5): 1070–1077.
9. Hudaib MM, Tawaha KA, Mohammad MK, Assaf AM, Issa AY, Alali FQ, Aburjai TA, Bustanji YK. Xanthine oxidase inhibitory activity of the methanolic extracts of selected Jordanian medicinal plants. Pharmacogn Mag 2011; 7(28): 320–324.
10. Bustanji Y, Issa A, Moulay A, Hudaib M, Tawaha K, Mohammad M, Hamed S, Masri I, Alali F. Hormone-sensitive lipase inhibition by selected medicinal plants. J Med Plants Res 2011; 5(18): 4405–4410.
11. Tawaha K, Alali FQ, Gharaibeh M, Mohammad M, El-Elimat T. Antioxidant activity and total phenolic content of selected Jordanian plant species. Food Chem 2007; 104(4): 1372–1378.
12. Tabaza Y, Edrada-Ebel R, Watson D, Young L. Implementing metabolomics tools in the search for new antiproliferative agents from the plant-associated endophytes. University of Strathclyde; 2018.
13. Ghasemi M, Turnbull T, Sebastian S, Kempson I. The MTT assay: utility, limitations, pitfalls, and interpretation in bulk and single-cell analysis. Int J Mol Sci 2021; 22(23): 12827.
14. Li G, Zhou J, Sun M, Cen J, Xu J. Role of luteolin extracted from Clerodendrum cyrtophyllum Turcz leaves in protecting HepG2 cells from TBHP-induced oxidative stress and its cytotoxicity, genotoxicity. J Funct Foods 2020; 74: 104196.
15. Lin Y, Shi R, Wang X, Shen H-M. Luteolin, a flavonoid with potential for cancer prevention and therapy. Curr Cancer Drug Targets 2008; 8(7): 634–646.
16. Birt DF, Hendrich S, Wang W. Dietary agents in cancer prevention: flavonoids and isoflavonoids. Pharmacol Ther 2001; 90(2–3): 157–177.
17. Imran M, Rauf A, Abu-Izneid T, Nadeem M, Shariati MA, Khan IA, Imran A, Orhan IE, Rizwan M, Atif M, et al. Luteolin, a flavonoid, as an anticancer agent: A review. Biomed Pharmacother 2019; 112: 108612.
18. López-Lázaro M. Distribution and biological activities of the flavonoid luteolin. Mini Rev Med Chem 2009; 9(1): 31–59.