In Vitro Cytotoxicity of Gallic Acid Derivatives (Alkyl gallates) Against Breast MCF-7 Cancer Cells

Ade Arsianti[1,2(✉)], Maya Dorothea[3], Naura Syafira[3], Ananda Tony[3], and Anton Bahtiar[4]

[1] Department of Medical Chemistry, Faculty of Medicine, University of Indonesia, Depok, Indonesia
arsi_ade2002@yahoo.com
[2] Faculty of Medicine, Drug Development Research Cluster (DDRC), Indonesia Medical Education and Research Institute (IMERI), University of Indonesia, Salemba Raya 6 Street, Jakarta 10430, Indonesia
[3] Faculty of Medicine, University of Indonesia, Depok, Indonesia
[4] Department of Pharmacology, Faculty of Pharmacy, University of Indonesia, Kampus UI, Depok, Indonesia

Abstract. Breast cancer is the most common cancer in women. Based on WHO data in 2011, it is estimated that more than 508,000 women worldwide died of breast cancer. In Indonesia, the prevalence of breast cancer is 26.5% of all cancer cases, in which become the second frequent cancer. To date, the treatments for breast cancer are surgery, radiation therapy, chemoradiation, or combination therapy. However, the high rate of complications and side effects caused led to the need for the development of other therapies. Gallic acid is known to have potential anticancer effects. Structure modification on gallic acid by esterification of carboxyl group into alkyl gallate derivatives would change the hydrophobicity, as well as change its solubility and cytotoxicity against cancer cells. In this research, ten gallic acid derivatives (alkyl gallates) have been synthesized from the esterification of gallic acid and corresponding alkyl alcohol. Purification of the derivatives were conducted by flash column chromatography on silica gel by using the mixture of chloroform and methanol as mobile phase. Cytotoxic activities of gallic acid and its synthesized alkyl gallates against breast MCF-7 cancer cells were evaluated by MTS (3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium) assay. The data were analyzed by linear regression method to generate IC_{50} value. Among alkyl gallate derivatives, isoamyl gallate, heptyl gallate and octyl gallate showed a moderate cytotoxicity on breast MCF-7 cancer cells with IC_{50} values of 58.11; 25.94 and 42.34 μg/mL, respectively. Thus, isoamyl gallate, heptyl gallate and octyl gallate should be further developed as potent and promising agents for the treatment and therapy of breast cancer.

Keywords: Gallic acid derivative · Alkyl gallate · Cytotoxicity · MCF-7 cell

© The Author(s) 2023
A. Lelono et al. (Eds.): ICOLIB 2021, ABSR 27, pp. 266–276, 2023.
https://doi.org/10.2991/978-94-6463-062-6_26

1 Introduction

The Breast cancer is the most common cancer in women and is the second most common type of cancer in the world. Based on GLOBOCAN (2012), the incidence of breast cancer worldwide reaches 11.9%, while in Indonesia, breast cancer is the most common cancer with a prevalence of 26.5% [1]. The survival rate of breast cancer is low. American Cancer Society states that the 5-year survival rate of patients with stage IV of breast cancer is only 22% [2]. The high mortality rate and the low survival rate of breast cancer are caused by inadequate breast cancer treatment. According to guideline from ESO (European School of Oncology) and ESMO (European Society of Medical Oncology), breast cancer treatment includes surgery, chemotherapy, and radiation. Determination of the treatment modality applied to breast cancer case is based on the stage in which breast cancer is detected. In cases of early-stage breast cancer, the recommended treatment is surgery. The treatment of choice for advanced breast cancer is chemotherapy and radiation [3]. However, chemotherapy and radiation are not selective to kill the cancer cells, it can also affect the normal cells. Therefore, research to find the more effective anti-breast cancer agents are needed.

Gallic acid or trihydroxybenzoic acid is a bioactive compound which is found in many plants and fruits. The previous studies revealed that gallic acid showed antifungal, antibacterial and anti-cancer effects. According to Inoue et al. [4], three adjacent phenolic hydroxyl groups in gallic acid are essential to the cytotoxicity. Meanwhile, carboxyl group in gallic acid is likely to be necessary for the selectivity of gallic acid to cancer cells. It seems that mitochondrial membrane potential is a common target of these compounds to cause apoptotic process in the cancer cells [4]. Wang et al. reported that gallic acid also has an anti-cancer effect against human breast cancer MCF-7 cells [5]. Study by Inoue et al. [4] and Serrano et al. [7] have revealed that gallic acid prompted apoptosis in cancer cells as an accompaniment of oxidative stresses from mitochondrial dysfunction, reactive oxygen species (ROS), and an incline in intracellular Ca^{2+} level. However, its hydrophilicity restricts gallic acid from passing the target cell membrane [6, 7]. To increase the potential anticancer activity and hydrophobicity of gallic acid, further study in gallic acid derivatives is needed. Modifying structure of gallic acid by esterification its carboxyl group into alkyl gallate derivatives would change the hydrophobicity, as well as change its solubility and cytotoxicity against cancer cells [8]. Locatelli et al. [8] concluded that the effectiveness of gallic acid derivatives (alkyl gallates) against some cancer cells was caused by the differences in their hydrophobicity which was influenced by the length of the carbon chain of the alkyl group in alkyl gallates. The longer and more branched carbon chain of the alkyl group in alkyl gallate, the stronger its hydrophobicity, so it will be easier to penetrate cell walls and provide therapeutic effects in cells. However, the longer carbon chain, the greater molecular weight of an alkyl gallate, causing it more difficult to diffuse through the cell membrane [8]. This fact indicates that it is very important to search the alkyl gallate with a certain carbon chain length of alkyl group that can optimally penetrate into the cell wall and inhibit the growth of cancer cells effectively. In this study, cytotoxicity of a series of synthetic alkyl gallates with linear and branch carbon chain of alkyl groups would be tested against MCF-7 breast cancer cells using the MTS assay. We hypothesized that alkyl gallates

with linear or branch alkyl substituent show greater cytotoxicity against MCF-7 cells compared to original compound, gallic acid.

2 Materials and Methods

Flowchart of experimental design (conceptual framework) is displayed in Fig. 1, as follows.

2.1 Gallic Acid and Alkyl gallates

Gallic acid was purchased from Sigma-Aldrich and was analyzed for its purity by Nuclear Magnetic Resonance (NMR). From the results of the analysis, the purchased gallic acid has a purity of 99%. Ten alkyl gallate derivatives of gallic acid, namely methyl gallate, ethyl gallate, propyl gallate, butyl gallate, isobutyl gallate, t-butyl gallate, amyl gallate, isoamyl gallate, heptyl gallate, and octyl gallate, have been synthesized previously through esterification reaction between gallic acid and the corresponding alkyl alcohol. The synthesized alkyl gallates were purified by flash column chromatography on silica gel (gradient elution method), with the mixture of chloroform and methanol as mobile phase. The purity of the synthesized alkyl gallates have been confirmed by thin layer chromatography (TLC) with UV lamp at 254 nm and 366 nm as a visualized spot, which showed there was only one spot and no other impurity spots on the TLC plate. Confirmation of the purity and structure elucidation of the synthesized alkyl gallates were determined by ^{1}H (proton) and ^{13}C (carbon) Nuclear Magnetic Resonance (NMR), which showed the purity of the synthesized alkyl gallate ranging from 96–98%.

Gallic acid and its derivatives of alkyl gallates were dissolved in DMSO and then diluted using a medium containing DMEM, 10% of fetal bovine serum, 1% of penicillin

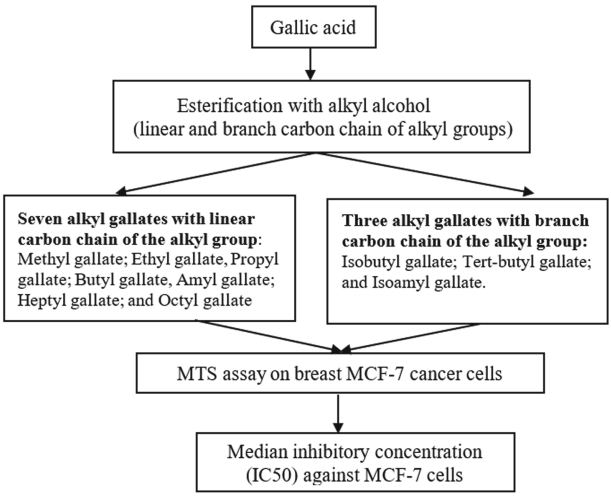

Fig. 1. Conceptual framework of research.

streptomycin and 1% of Fungizon to reach a concentration of 51.2 μg/mL. This solution was then taken partially to be diluted to obtain the variety concentration of 0.4; 0.8; 1.6; 3.2; 6.4; 12.8 and 25.6 μg/mL, which will be used for experiment.

2.2 Medium Preparation and Cell Culture

MCF-7 breast cancer cells (Fig. 2) obtained from Department of Pathological Anatomy, Faculty of Medicine, University of Indonesia. MCF-7 cells were cultured by DMEM medium with 10% Fetal Bovine Serum supplementation. The medium is made by addition of DMEM into distilled water, the solution is stirred while bicarbonate is added to reach pH of 7.2 to 7.4. Subsequently, the solution is diluted by distilled water until it reaches the desired concentration. Fetal Bovine Serum (FBS), PenStrep and Fungizon were then added into the solution until their concentration of 10%, 1% and 1% in the solution, respectively. The solution was filtered with minisart syringe filter. The filtrate was transferred to a sterile container by syringe.

The culture was performed using sterile technique in a laminar flow hood, incubated in an incubator with 5% of CO2 at the temperature of 37 °C. The MCF-7 cancer cells were counted using the Neubauer counting chamber. A total of 10 μL of the cells was mixed with 10 μL of 0.1% trypan blue dye. The mixture was homogenized and then put into the Neubauer counting chamber and viewed under a light microscope. The cells were counted using the principle that cells attached to the right and top lines would be counted, while those attached to the left and bottom lines were not counted. The counted cells will be cultured for experiments.

Cell culture was performed by placing the cells in a TC flask filled with the medium. Cell culture was carried out in a sterile condition on biosafety cabinet to prevent contamination. Then, the cells in TC flask were put into an incubator at 37 °C with 5% of CO_2, the cells growth was observed every day. If the cells have grown to fill the TC flask, the cells will be harvested for storage or replanting, or used as experimental material.

Harvesting cells was conducted by removing the medium and washing the cells with PBS, then carefully removing the PBS. Add 1 ml of trypsin to release the attached cell in the TC flask, then the cells were transferred to a 15 mL of tube containing the stock

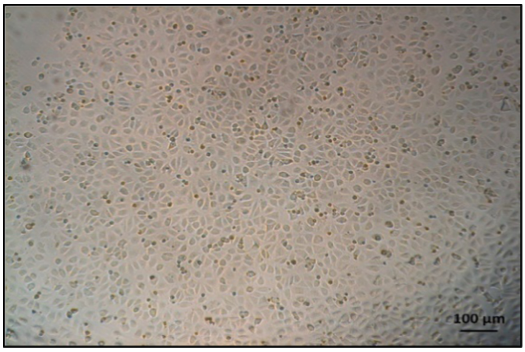

Fig. 2. MCF-7 breast cancer cells before treatment.

medium. The solution was centrifuged and the supernatant was discarded, and cells were counted using a Neubauer counting chamber.

2.3 MTS Assay

The cytotoxicity evaluation of gallic acid and its derivatives of alkyl gallates against MCF-7 breast cancer cells were carried out by MTS assay. MTS assay gives convenient step by adding reagent directly to the cell culture without additional intermittent step needed in the MTT assay. Besides that, the purple formazan product in MTS assay is soluble, whereas the formazan product in MTT assay is not soluble [9]. The cultured MCF-7 breast cancer cells were put into a 96-well microwell of 1×10^4 cells/well. The cells were left overnight to allow its attachment to the well. The medium will then be replaced with the testes compound, gallic acid and alkyl gallates, with the variety concentration of 51.2; 25.6; 12.8; 6, 4; 3.2; 1.6; 0.8 and 0.4 μg/mL. The mixture was incubated for 48 h. Then, 20 μL of MTS solution which had been mixed with PMS in a ratio of 20:1 was added to each well and then incubated for 2 h. After incubation, the absorbance of the solution was measured on a microplate reader with a wavelength of 490 nm.

2.4 Data Analysis

In this work, the data were analyzed by linear regression method, which has advantages such as easy to be implemented and interpreted, as well as it can be applied well for linearly separable data.

The data obtained in the form of absorbance (Abs.) in ELISA reader with a wavelength of 490 nm. From the data obtained, the percentage of viability (% viability) is calculated using the formula:

$$\%\ \text{viability} = \frac{\text{Abs. of sample - abs. of blank}}{\text{Abs. of control - Abs. of blank}} \times 100\% \quad (1)$$

The linear regression equation, $y = ax + b$, was obtained by plotting of log concentration of the sample in x axis versus percentage of viability in y axis. R-squared (R^2) is used for data analysis. In this research, we used high standard of R-squared with value of 0.95 or above, which shows high level of correlation and statistically significant.

3 Result and Discussion

In this research, cytotoxicity evaluation of gallic acid, doxorubicin (positive control) and alkyl gallates were determined by MTS assay. MTS assay is an assay technique using MTS tetrazolium reagent which is used to detect cell viability. MTS tetrazolium can be reduced by viable cells to form purple formazan product which can be dissolved in the cell culture medium [9]. In this work, the MTS assay of heptyl gallate against MCF-7 cells represented the cytotoxicity results for all alkyl gallate derivatives. The purple formazan was formed after MCF-7 cells treated by MTS and heptyl gallate in concentration of 51.2 μg/mL (Fig. 3a) and 1.5 μg/mL (Fig. 3b). As shown in Fig. 2,

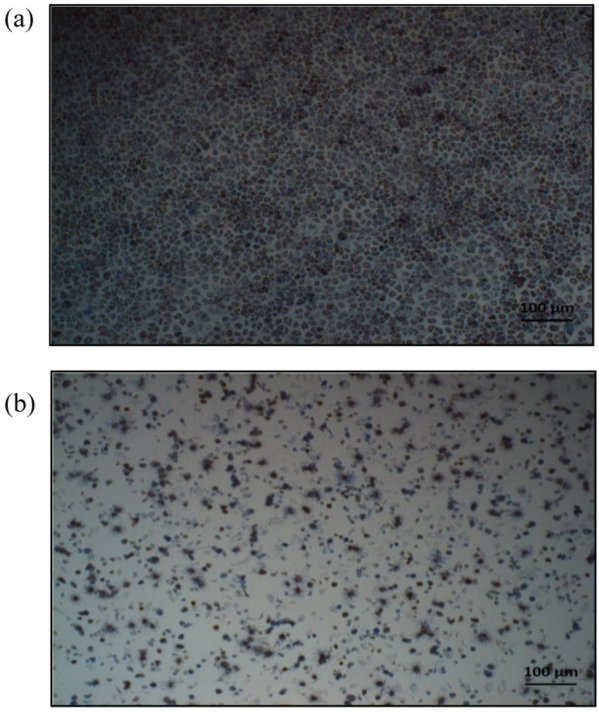

Fig. 3. MCF-7 cells after treated by MTS and 1.5 µg/mL of heptyl gallate (a), and after treated by MTS and 51.2 µg/mL of heptyl gallate (b).

the greater purple formazan was formed when MCF-7 cells treated by heptyl gallate in concentration of 1.5 µg/mL, whilst the treatment of MCF-7 cells with heptyl gallate in concentration of 51.2 µg/mL resulted in decreasing the formation of purple formazan. This result indicating that heptyl gallate inhibiting the growth of breast MCF-7 cancer cells in dose-dependent manner. In which, the higher concentration of heptyl gallate, the stronger its inhibition on MCF-7 cells, thus the lower MCF-7 cell viability, as shown by the lower absorbance.

Linear regression of log concentration in x-axis versus percentage of cell inhibition in y-axis for the best three cytotoxic compounds on MCF-7 cells, namely isoamyl gallate, heptyl gallate and octyl gallate, are displayed in Figs. 4, 5 and 6, respectively.

Cytotoxic activity of gallic acid and alkyl gallate derivatives expressed in IC_{50} value, which is defined as the concentration is required to inhibit 50% of cancer cells growth. The smaller IC_{50} value, the greater cytotoxic activity. Substituting y with 50 in linear regression equation of $y = 45.071x + 28.894$ ($R^2 = 0.9807$) for isoamyl gallate in Fig. 4 to give log of IC_{50}, thus anti log or inverse log of IC_{50} will give the IC_{50} value of isoamyl gallate (58.11 µg/mL). We used the same way to calculate the IC_{50} value for heptyl gallate (Fig. 5), octyl gallate (Fig. 6), and other derivatives of alkyl gallate.

Cytotoxicities (IC_{50} values) of gallic acid, doxorubicin (positive control) and ten alkyl gallates against breast MCF-7 cancer cells are summarized in Table 1.

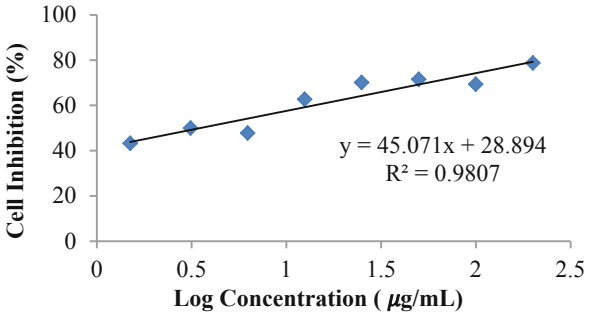

Fig. 4. Linear graph of isoamyl gallate on MCF-7 cells.

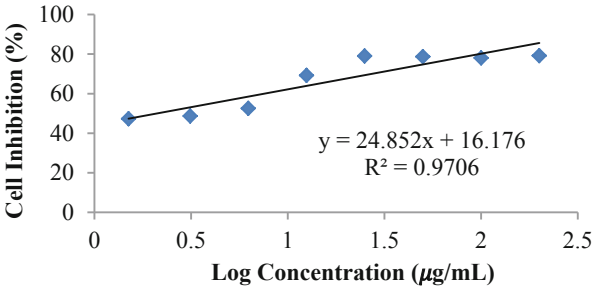

Fig. 5. Linear graph of heptyl gallate on MCF-7 cells.

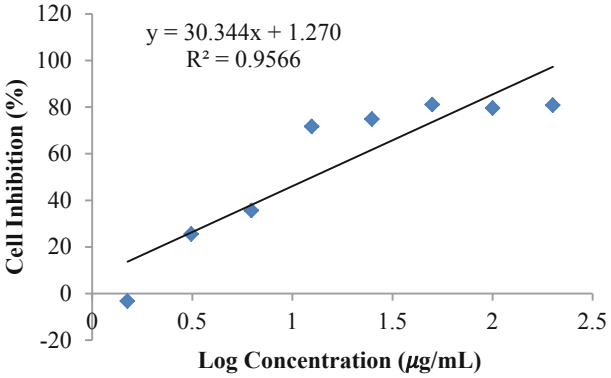

Fig. 6. Linear graph of octyl gallate on MCF-7 cells.

Based on classification provided by Atjanasuppat et al., an extract or compound has a very active cytotoxicity against cancer cells if the IC_{50} value < 20 μg/mL; moderate cytotoxicity (IC_{50}:20–100 μg/mL), weak cytotoxicity (IC_{50}:100–1000 μg/mL, and no cytotoxic activity ($IC_{50} > 1000$ μg/mL) [10].

As shown in Table 1, doxorubicin as a positive control (IC_{50}: 6.58 μg/mL) showed the most active cytotoxicity against breast MCF-7 cancer cells. In contrast to doxorubicin,

Table 1. Cytotoxicity (IC_{50} value) of gallic acid, doxorubicin and alkyl gallates against breast MCF-7 cells

Compound	IC_{50} (μg/mL)
Gallic acid	**166.90**
Methyl gallate	113.25
Ethyl gallate	130.12
Propyl gallate	>1000
Butyl gallate	>1000
Isobutyl gallate	227.83
Tert-butyl gallate	151.20
Amyl gallate	>1000
Isoamyl gallate	**58.11**
Heptyl gallate	**25.94**
Octyl gallate	**42.34**
Doxorubicin	6.58

propyl gallate, butyl gallate and amyl gallate with IC_{50} value over than 1000 μg/mL were assigned no cytotoxicity against breast MCF-7 cancer cells.

Methyl gallate (IC_{50}: 113.25 μg/mL) and ethyl gallate (IC_{50}: 130.12 μg/mL), Tert-butyl gallate (IC_{50}: 151.20 μg/mL), isoamyl gallate (IC_{50}: 58.11 μg/mL), heptyl gallate (IC_{50}: 25.94 μg/mL) and octyl gallate (IC_{50}: 42.34 μg/mL) have higher cytotoxicity against breast MCF-7 cells compared to gallic acid (IC_{50}: 166.90 μg/mL). Isobutyl gallate and tert-butyl gallate show a greater cytotoxicity than butyl gallate, as well as isoamyl gallate has a stronger cytotoxicity against breast MCF-7. This is suggesting that branched carbon chain of alkyl group in isobutyl gallate, tert-butyl gallate and isoamyl gallate more important for cytotoxicity against MCF-7 cells compared to linear carbon chain of alkyl group in butyl gallate and amyl gallate. Previous research by Frey et al. [11] gave similar results with this study, in which, isobutyl gallate that has branched-chain substituent demonstrated a stronger cytotoxicity compared to butyl gallate which has a linear chain substituen against sarcoma 786A, adenocarcinoma TA3, and multiresistant variant TA3-MTX-R cell lines. Correspondingly, cytotoxicity of isoamyl and isopropyl gallate against sarcoma 786A, adenocarcinoma TA3 and TA3-MTX-R were higher than the cytotoxicity of amyl gallate and propyl gallate [11]. Another study by Mannhold et al. [12] revealed that the addition of branched carbon chain to the derivative of gallic acid will change the solubility. Chain branching allows the derivative compound to be more lipophilic owing to the decreasing in polarity, thus making it easier to penetrate the lipid bilayer [12].

Among the series of alkyl gallates, Isoamyl gallate, heptyl gallate and octyl gallate demonstrated a strong cytotoxicity against MCF-7 cells. The strongest cytotoxicity on breast MCF-7 cancer cells has been shown by heptyl gallate with IC_{50} value of 25.94 μg/mL. This result indicating that modifying of carboxyl group in gallic acid into

six linear carbon chain of heptyl group in heptyl gallate has been successfully improved its cytotoxicity against breast MCF-7 cancer cells. The apparently more cytotoxic activity of heptyl gallate seemed to be related with alkyl chain lengths. The longer alkyl chain is formed, the greater partition coefficient value, signifying higher lipophilicity. Hence, the length of the lipophilic alkyl side chain may apply a powerful impact on the membrane affinity of these compounds, a hypothesis that is in accordance with other studies on alkyl gallates [13, 14]. The lipophilic property increases the permeability for cell membranes so that it can easily diffuse through transmembrane and perform localization in cell line [8]. Locatelli et al. proved that less lipophilic substances are less efficient in reaching the interior of the cells or sending cell-death signals [15]. As a result, heptyl gallate has higher cytotoxic effect than other alkyl gallate with shorter side chains, such as methyl gallate, ethyl gallate, propyl gallate, butyl gallate, and amyl gallate. Compared to heptyl gallate, modifying of carboxyl group in gallic acid into eight linear carbon chain of octyl group in octyl gallate resulted in decreasing in its cytotoxicity against MCF-7 cells. This fact suggesting that the existence of six linear carbon chain in heptyl gallate contributed to the optimalization of its cytotoxicity against MCF-7 cells. Thus, heptyl gallat shows the most ability to inhibit the growth of MCF-7 cancer cells effectively and optimally compared to others alkyl gallate derivatives.

As a conclusion, we have evaluated cytotoxic activity of ten alkyl gallate derivatives of gallic acid against breast MCF-7 cancer cells. Among them, heptyl gallate showed the greatest cytotoxicity against MCF-7 cells, thus heptyl gallate should be further developed as potent and promising anti-breast cancer agent.

Acknowledgments. Authors thank to Ministry of Research and Technology-Badan Riset dan Inovasi Nasional (Kemenristek-BRIN) for PDUPT research grant fiscal year 2021 with the contract number: NKB 092/UN2.RST/HKP.05.00/2021, and for PUTI Saintekes research grant fiscal year 2020 from University of Indonesia with contract number: NKB-4767 and 4794/UN2.RST/HKP.05.00/2020.

Authors' Contributions. AA gives the contribution to synthesize alkyl gallate and write the paper manuscript. MD, NS, AT and AB contribute to evaluate the cytotoxicities of alkyl gallates and analyze the data. All authors read and approved the final manuscript.

References

1. J. Ferlay, I. Soerjomataram, M. Ervik, R. DikshitS. Eser, C. Mathers, et al. GLOBOCAN 2012 v1.0, Cancer Incidence and Mortality Worldwide: IARC CancerBase No. 11 [Internet]. Lyon, France: International Agency for Research on Cancer, 2013 [cited 2015 Jan 8]. Available from: http://globocan.iarc.fr
2. American Cancer Society. Breast Cancer Survival Rates by Stage [Internet]. 2014 [cited 2015 Jan 8]. Available from: http://www.cancer.org/cancer/breastcancer/detailedguide/breast-cancer-survival-by-stage
3. F. Cardoso, A. Costa, L. Norton, E. Senksus, M. Aapro, F. Andre, et al. ESO-ESMO 2nd international consensus guidelines of advanced breast cancer (ABC2). J Ann Oncol. 2014, Vol. 25(10), pp.1871–1888 DOI: https://doi.org/10.1093/annonc/mdu385.

4. M. Inoue, R. Suzuki,N. Sakaguchi, Z. Li, T. Takeda, Y. Ogihara, B.Y. Jiang, Y. Chen. Selective induction of cell death in cancer cells by gallic acid. Biological and pharmaceutical bulletin. 1995, Vol. 18(11), pp.1526–1530. DOI: https://doi.org/10.1248/bpb.18.1526.
5. K. Wang, X. Zhu, K. Zhang, L. Zhu, F. Zhou. Investigation of gallic acid induced anticancer effect in human breast carcinoma MCF-7 cells. J Biochem Mol Toxicol. 2014, Vol. 28(9), pp. 387–393. DOI: https://doi.org/10.1002/jbt.21575
6. M. Inoue, N.S akaguchi, K. Isuzugwa, H. Tani, Y. Ogiwara. Role of reactive oxygen species in gallic acid-induced apoptosis. Biol. Pharm. Bull. 2000, Vol. 23(10), pp.1153–1157. DOI: https://doi.org/10.1248/bpb.23.1153.
7. A. Serrano, C. Palacios, G. Roy, C. Cespon, M.L. Villar, M. Nocito, P. Gonzales-Porque. Derivatives of gallic acid induce apoptosis in tumoral cell lines and inhibit lymphocyte proliferation. Arch. Biochem. Biophys. 1998, Vol. 350(1), pp. 49–54. DOI: https://doi.org/10.1006/abbi.1997.0474.
8. C. Locatelli, F.B. Filippin-Monteiro, T.B. Creczynski-Pasa. Alkyl esters of gallic acid as anticancer agents: A review. Eur J Med Chem. 2013, Vol. 60(1), pp. 233–239.
9. T.L. Riss, R.A. Moravec, A.L. Niles, et al. Cell Viability Assays. 2013 [Updated 2016]. In: G. S. Sittapalam, N.P. Coussens, H. Nelson, et al., editors. Assay Guidance Manual [Internet]. Bethesda (MD): Eli Lilly & Company and the National Center for Advancing Translational Sciences; 2004. Available from: http://www.ncbi.nlm.nih.gov/books/NBK144065/
10. K. Atjanasuppat, W. Wongkham, P. MeepowpanP. Kittakoop, P. Sobhon, A. Bartlett, et al. In vitro screening for anthelmintic and antitumour activity of ethnomedicinal plants from Thailand. J. Ethnopharmacol. 2009, Vol. 123, pp. 475–482. DOI: https://doi.org/10.1016/j.jep.2009.03.010
11. C. Frey, M. Pavani, G. Cordano, S. Munoz, E. Rivera, J. Medina, et al. Comparative cytotoxicity of alkyl gallates on mouse tumor cell lines and isolated rat hepatocytes. Comp Biochem Phys A. 2007, Vol. 146, pp. 520–527. DOI: https://doi.org/10.1016/j.cbpa.2006.03.007
12. R. Mannhold, H. Kubinyi, H. Timmerman, editors. In: Todeschini R, Consonni V. Handbook of molecular descriptors methods and principal in medisinal chemistry. 11th ed. German: Wiley-VCH; 2008.
13. P. Tammela, L. Laitinen, A. Galkin, T. Wennberg, R. Heczko, H. Vuorela, J.P. Slotte, P. Vuorela. Permeability characteristics and membrane affinity of flavonoids and alkyl gallates in Caco-2 cells and in phospholipid vesicles. Archives of biochemistry and biophysics. 2004, Vol. 425(2), pp.193–199. DOI: https://doi.org/10.1016/j.abb.2004.03.023
14. Q. Feng, T. Kumagai, Y. Nakamura, K. Uchida, T. Osawa. Correlation of antimutagenic activity and suppression of CYP1A with the lipophilicity of alkyl gallates and other phenolic compounds. Mutation Research/Genetic Toxicology and Environmental Mutagenesis. 2003, Vol. 537(1), pp.101–108. DOI: https://doi.org/10.1016/S1383-5718(03)00057-3
15. C. Locatelli, R. Rosso, M.C. Santos-Silva, C.A. de Souza, M.A. Licinio, P. Leal, et al. Ester derivatives of gallic acid with potential toxicity toward L1210 leukemia cells. Bioorg Med Chem. 2008, Vol. 16(7), pp. 3791–3799. DOI: https://doi.org/10.1016/j.bmc.2008.01.049

Open Access This chapter is licensed under the terms of the Creative Commons Attribution-NonCommercial 4.0 International License (http://creativecommons.org/licenses/by-nc/4.0/), which permits any noncommercial use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons license and indicate if changes were made.

The images or other third party material in this chapter are included in the chapter's Creative Commons license, unless indicated otherwise in a credit line to the material. If material is not included in the chapter's Creative Commons license and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder.