

## *In Vitro* Cytotoxic Activity of *Lagerstroemia speciosa* (L.) Pers. Leaf Extracts against MCF-7 Breast Cancer Cells

Ernie Halimatushadyah\*

Dyah Ayuwati Waluyo

Krismayadi

Binawan University, East Jakarta, DKI Jakarta, Indonesia

\*email: [ernie@binawan.ac.id](mailto:ernie@binawan.ac.id)

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### Abstract

Breast cancer is the most commonly diagnosed cancer in women, and chemotherapy is limited by toxicity and drug resistance. Bungur (*Lagerstroemia speciosa* (L.) Pers.) leaves contain flavonoids and other secondary metabolites that may have cytotoxic potential, but data comparing extracts prepared with different solvents against MCF-7 breast cancer cells remain limited. This study aimed to determine the secondary metabolite content of *L. speciosa* leaf extracts and to compare their *in vitro* cytotoxicity against MCF-7 cells. Leaf powder (*simplicia*) was macerated with methanol, 70% ethanol, or ethyl acetate. The extracts were screened qualitatively for secondary metabolites, and cytotoxicity was determined using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay. The extract yields were 10.05% (methanol), 12.63% (70% ethanol), and 2.56% (ethyl acetate). All three extracts contained alkaloids, saponins, tannins, phenolics, flavonoids, triterpenoids, steroids, and glycosides. The half-maximal inhibitory concentration ( $IC_{50}$ ) values were 98.46 ppm for the methanol extract, 163.67 ppm for the 70% ethanol extract, and 182.72 ppm for the ethyl acetate extract. The methanol extract had the lowest  $IC_{50}$  and therefore the strongest cytotoxic activity among the three extracts. Based on these results, *L. speciosa* leaf extracts showed *in vitro* cytotoxic activity against MCF-7 cells and warrant further investigation.

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## INTRODUCTION

Breast cancer (*carcinoma mammae*) is the most commonly diagnosed cancer and the leading cause of cancer death among women worldwide. Global estimates for 2022 showed that female breast cancer accounted for 11.6% of all new cancer cases and 6.9% of all cancer deaths (1). In Indonesia, GLOBOCAN 2024 estimated 93,236 new breast cancer cases and 38,012 breast-cancer deaths; among women, breast cancer accounted for 34.4% of new cancer diagnoses and ranked first by incidence (2). Breast cancer is treated with surgery, radiotherapy, endocrine therapy, targeted therapy, and chemotherapy (3). Chemotherapies, however, have toxic effects, and breast cancer cells that initially respond to first- and second-line drugs may eventually acquire resistance (4). These limitations have driven research on natural products as candidate anticancer agents, which have been reported to act on multiple targets and to cause fewer side effects (4,5).

Indonesia is a megabiodiversity country, and many of its plants are potential sources of medicinal compounds. Bungur (*Lagerstroemia speciosa* (L.) Pers., Lythraceae) is a flowering tree native to Southeast Asia and South Asia. Its phytoconstituents include glycosides, flavones, corosolic acid, ellagic acid, triterpenes, and tannins, which have been reported in different parts of the plant, including the leaves (6). Flavonoids and phenolic compounds are of particular interest because they have been associated with anticancer mechanisms. Flavonoids have been reported to modulate cell-

cycle progression at the transition from the G1 to the S phase, whereas saponins and triterpenoids have been reported to disrupt the permeability of mitochondrial membranes and thereby trigger cell death (7).

Studies on *L. speciosa* in relation to breast cancer have so far explored, for example, leaf extract-mediated zinc oxide nanoparticles tested against MCF-7 cells (8) and in silico analysis of phytosterols from the seed extract against breast cancer target proteins<sup>6</sup>. However, the effect of the extraction solvent on the yield, phytochemical profile, and cytotoxic activity of crude *L. speciosa* leaf extracts against MCF-7 cells remains poorly documented. The type of solvent affects the characteristics of a plant extract, because solvents of different polarity dissolve different groups of compounds (9).

The cytotoxicity of plant extracts is commonly screened *in vitro* using the MTT assay. In this assay, metabolically active cells reduce the yellow tetrazolium salt MTT to purple, water-insoluble formazan crystals. The absorbance of the dissolved formazan is then measured and used to estimate cell viability and the half-maximal inhibitory concentration (IC<sub>50</sub>) (10). MCF-7 cells, a hormone-dependent breast carcinoma cell line, were used as the target cells. This study aimed to determine the yield and the qualitative secondary metabolite content of methanol, 70% ethanol, and ethyl acetate extracts of *L. speciosa* leaves and to compare their cytotoxic activity against MCF-7 breast cancer cells, expressed as IC<sub>50</sub> values, using the MTT assay. The novelty of this study is the side-by-side comparison of three solvents of different polarity in the same MCF-7 model, which provides preliminary data to guide the selection of extracts for further investigation.

## METHODS

### Materials and equipment

Bungur (*Lagerstroemia speciosa* (L.) Pers.) leaves were identified at LIPI Cibinong. The dried leaf powder (simplicia) was used for extraction. The solvents were methanol, ethanol, and ethyl acetate (Merck). The reagents for phytochemical screening were 2N HCl (ROFA Laboratorium); Liebermann–Burchard, Mayer, Dragendorff, and Wagner reagents (Medical and Laboratory); and magnesium powder and 1% FeCl<sub>3</sub> (Merck). The MTT reagent and dimethyl sulfoxide (DMSO) were obtained from Sigma. MCF-7 cells (hormone-dependent breast carcinoma cells) were obtained from PSSP IPB, Bogor. The equipment comprised a rotary evaporator (Büchi), a spectrophotometer (Shimadzu), a microplate reader, and glassware (Pyrex).

### Extraction

Extraction was performed by maceration using three different solvents: methanol, 70% ethanol, and ethyl acetate. For each extraction, 1 kg (1,000 g) of dried *Lagerstroemia speciosa* leaf powder (simplicia) was macerated with 10 L of solvent (1:10, w/v) in a tightly closed container at room temperature for 24 hours. The plant material was then remacerated with fresh solvent at a 1:5 (w/v) ratio, corresponding to 5 L of solvent, for an additional 24 hours. The macerates obtained from both extraction cycles were combined and concentrated using a rotary evaporator at 50 °C until a thick extract was obtained. The methanol, 70% ethanol, and ethyl acetate extracts yielded 100.5 g, 126.3 g, and 25.6 g of thick extract.

### Identification of qualitative phytochemical content

The procedures below were used for the qualitative identification of alkaloids, saponins, tannins, phenolics, flavonoids, triterpenoids and steroids, and glycosides in each extract, using chemical detection reagents (11–13).

#### Alkaloids

A 0.5 g sample was mixed with 1 mL of 2 N hydrochloric acid and 9 mL of distilled water, heated in a water bath for 2 minutes, cooled, and filtered. The filtrate was divided into three test tubes, with 0.5 mL placed in each tube. Two drops of Mayer reagent were added to the first tube, two drops of Wagner reagent to the second tube, and two drops of

Dragendorff reagent to the third tube. The formation of a white to yellowish precipitate with Mayer reagent, a brown precipitate with Wagner reagent, and an orange to reddish-brown precipitate with Dragendorff reagent indicated a positive alkaloid reaction. A sample was considered positive for alkaloids when precipitation or turbidity was observed in at least two of the three tests.

### ***Saponins***

A 0.5 g sample was placed in a test tube with 10 mL of hot distilled water, cooled, and shaken vigorously for 10 seconds. The formation of a stable foam 1–10 cm high that persisted for at least 10 minutes indicated the presence of saponins. One drop of 2N hydrochloric acid was then added; foam that did not disappear confirmed the presence of saponins.

### ***Tannins***

A 1 g sample was boiled for 3 minutes in 100 mL of distilled water, cooled, and filtered. Two milliliters of the filtrate were mixed with 1–2 drops of 1% iron (III) chloride. A dark blue or blackish-green color indicated the presence of tannins.

### ***Phenolics***

Phenolic compounds were identified using the ferric chloride test. A portion of the sample solution was treated with 1% ferric chloride ( $\text{FeCl}_3$ ) solution. The development of a dark green, bluish-green, blue, or black coloration was considered a positive reaction for phenolic compounds.

### ***Flavonoids***

A 10 g sample was mixed with 10 mL of hot water, boiled for 5 minutes, and filtered while hot. Five milliliters of the filtrate were mixed with 0.1 g of magnesium powder, 1 mL of concentrated hydrochloric acid, and 2 mL of amyl alcohol, shaken, and left to separate. A red color in the amyl alcohol layer indicated the presence of flavonoids.

### ***Triterpenoids and steroids***

A 1 g sample was macerated in 20 mL of non-polar n-hexane for 2 hours and filtered. The filtrate was evaporated in an evaporating dish, and 3 drops of Liebermann–Burchard reagent were added to the residue. A green color indicated steroids, and a red or purple color indicated triterpenoids.

### ***Glycosides***

A 0.5 g sample was mixed with 2 mL of glacial acetic acid containing 1 drop of 1% iron (III) chloride ( $\text{FeCl}_3$ ) solution. Subsequently, 1 mL of concentrated sulfuric acid ( $\text{H}_2\text{SO}_4$ ) was carefully added along the wall of the test tube to form a separate layer. The formation of a purple to reddish-brown ring at the interface was considered a positive indication for glycosides.

### ***Cytotoxicity test (MTT assay)***

Cytotoxic activity against MCF-7 cells was determined using the MTT assay(14). Cells were seeded at  $1 \times 10^4$  cells per well in 100  $\mu\text{L}$  of complete culture medium in 96-well microplates and incubated overnight in a  $\text{CO}_2$  incubator at 37  $^\circ\text{C}$ . The medium was then removed, and 100  $\mu\text{L}$  of each extract concentration was added to the wells. The extracts were dissolved in DMSO, and the concentrations tested were 80 – 200 ppm. Each concentration was tested in three replication. Untreated MCF-7 cells were used as the negative control, whereas cells treated with the same final concentration of DMSO used to dissolve the extracts served as the vehicle control. Doxorubicin was used as the positive cytotoxic control. Blank wells containing complete culture medium and MTT reagent without cells were included to correct for background absorbance. The plates were incubated again for 24 hours, and the condition of the cells was documented for each treatment at the end of the incubation. The medium containing the treatment was then removed, and 100  $\mu\text{L}$  of MTT solution (0.5 mg/mL) was added to each well, including the control wells. The cells were incubated with MTT for 2 hours

at 37 °C in a CO<sub>2</sub> incubator. The MTT solution was then discarded, and 100 µL of isopropanol was added to each well to dissolve the purple formazan crystals. The plates were placed on a shaker to ensure complete solubilization of the formazan before absorbance measurement. Absorbance was measured with a microplate reader at 570 nm. Cell viability (%) was calculated as (absorbance of treated cells – absorbance of blank) / (absorbance of untreated cells – absorbance of blank) × 100%. The IC<sub>50</sub> value was determined by linear regression analysis of the relationship between extract concentration and cell viability using Microsoft Excel 2021. The linear regression equation ( $y = ax + b$ ) was generated, and the IC<sub>50</sub> value was calculated by substituting  $y = 50$  and solving for  $x$ , where  $x$  represents the extract concentration required to reduce cell viability to 50%.

## RESULTS

### Extraction yield

The percentage yields of the three extracts are shown in Table 1. The 70% ethanol extract gave the highest yield (12.63%), followed by the methanol extract (10.05%) and the ethyl acetate extract (2.56%).

Table 1. Percentage yield of *L. speciosa* leaf extracts obtained by maceration

| No | Extract               | Yield (%) |
|----|-----------------------|-----------|
| 1  | Methanol extract      | 10.05     |
| 2  | 70% ethanol extract   | 12.63     |
| 3  | Ethyl acetate extract | 2.56      |

### Qualitative phytochemical content

The methanol, 70% ethanol, and ethyl acetate extracts gave positive results for all secondary metabolite groups tested: alkaloids, saponins, tannins, phenolics, flavonoids, triterpenoids, steroids, and glycosides (Table 2).

Table 2. Qualitative secondary metabolite content of *L. speciosa* leaf extracts

| No | Secondary metabolite | Methanol | 70% ethanol | Ethyl acetate |
|----|----------------------|----------|-------------|---------------|
| 1  | Alkaloids            | +        | +           | +             |
| 2  | Saponins             | +        | +           | +             |
| 3  | Tannins              | +        | +           | +             |
| 4  | Phenolics            | +        | +           | +             |
| 5  | Flavonoids           | +        | +           | +             |
| 6  | Triterpenoids        | +        | +           | +             |
| 7  | Steroids             | +        | +           | +             |
| 8  | Glycosides           | +        | +           | +             |

+ = positive result.

### Cytotoxic activity against MCF-7 cells

The cytotoxic activity of the extracts against MCF-7 cells was evaluated using the MTT assay. The IC<sub>50</sub> values obtained for the three extracts are presented in Table 3. The methanol extract showed the lowest IC<sub>50</sub> value at 98.46 ppm, followed by the 70% ethanol extract at 163.67 ppm and the ethyl acetate extract at 182.72 ppm.

Table 3. IC<sub>50</sub> values of *L. speciosa* leaf extracts against MCF-7 cells

| No | Extract               | IC <sub>50</sub> (ppm) |
|----|-----------------------|------------------------|
| 1  | Methanol extract      | 98.46                  |
| 2  | 70% ethanol extract   | 163.67                 |
| 3  | Ethyl acetate extract | 182.72                 |

The morphology of MCF-7 cells after 24 hours of treatment with 100 ppm of each extract is shown in Figure 1. Fewer adherent cells were observed in wells treated with the methanol and 70% ethanol extracts than in the well treated with the ethyl acetate extract, where the cell layer appeared denser.

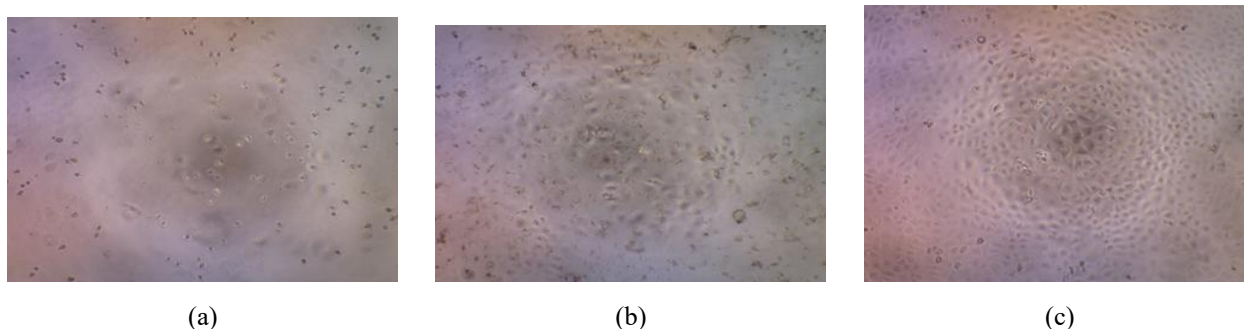


Figure 1. Morphology of MCF-7 cells after 24 hours of treatment with 100 ppm of (a) methanol extract, (b) 70% ethanol extract, and (c) ethyl acetate extract of *L. speciosa* leaves

## DISCUSSION

### Extraction yield

The extraction yield varied according to the solvent used. The 70% ethanol extract produced the highest yield (12.63%), whereas the ethyl acetate extract showed the lowest yield (2.56%). Solvents with different polarities dissolve different groups of compounds; therefore, solvent polarity can substantially influence the amount of material extracted from plant samples(9).

The comparatively high yield obtained with 70% ethanol may be related to the ability of the ethanol–water mixture to extract compounds across a relatively broad polarity range. In contrast, the substantially lower yield obtained with ethyl acetate suggests that a considerable proportion of extractable constituents in *L. speciosa* leaves may have greater affinity for more polar solvents.

The Indonesian Herbal Pharmacopoeia specifies a minimum extract yield of 9.2% for maceration using 70% ethanol (15). The 70% ethanol extract obtained in the present study had a yield of 12.63%, which exceeded this reference value. However, this comparison is applicable only to the 70% ethanol extract because the pharmacopoeial specification is linked to the extraction method and solvent described in the monograph. Therefore, the yields obtained using methanol and ethyl acetate should not be directly evaluated against the same standard.

### Qualitative phytochemical content

All three extracts showed positive qualitative reactions for alkaloids, saponins, tannins, phenolics, flavonoids, triterpenoids, steroids, and glycosides. These findings are generally consistent with previous studies reporting the presence of alkaloids, saponins, tannins, phenolics, steroids, glycosides, and triterpenoids in *L. speciosa* leaves (16), as well as flavonoids, glycosides, triterpenes, and tannins in this species (6).

The detection of similar classes of secondary metabolites in all three extracts indicates that methanol, 70% ethanol, and ethyl acetate were each capable of extracting representatives of these compound groups under the conditions used. Nevertheless, qualitative phytochemical screening only demonstrates the presence or absence of a compound class and does not provide information regarding its concentration.

Consequently, differences in biological activity among the extracts cannot be explained solely on the basis of these qualitative findings. Quantitative determination of relevant compound groups, particularly total phenolic content and total flavonoid content, would be required to evaluate whether differences in phytochemical concentration are associated with differences in cytotoxic activity.

### Cytotoxic activity against MCF-7 cells

The MTT assay is based on the reduction of the tetrazolium salt MTT to insoluble formazan by metabolically active cells; the amount of formazan formed, measured as absorbance, therefore reflects the number of metabolically active cells<sup>10</sup>. The IC<sub>50</sub> values of the three extracts against MCF-7 cells are presented in Table 3. A lower IC<sub>50</sub> means that a lower concentration is needed to inhibit 50% of the cells, so the methanol extract had the strongest cytotoxic activity of the three. The IC<sub>50</sub> values of the 70% ethanol and ethyl acetate extracts were about 1.7 and 1.9 times higher, respectively, than that of the methanol extract. All three extracts were classified as moderately cytotoxic because their IC<sub>50</sub> values were within the 21–200 µg/mL range (17).

The stronger activity of the methanol extract may be related to the phenolic and flavonoid compounds detected in all three extracts. However, because only qualitative screening was performed, the content of phenolics and flavonoids in each extract is unknown, and the difference in activity cannot be attributed to the level of any compound group. Quantitative analysis, such as total phenolic and total flavonoid content, is needed to test this relationship. Phenolic compounds in general have been reported to act on several cancer cell checkpoints and to induce apoptosis, autophagy, and cell-cycle arrest, as well as to inhibit angiogenesis and metastasis (18,19). At the molecular level, they have been reported to decrease Bcl-2 expression and mitochondrial membrane potential, suppress HIF-1 $\alpha$  expression, and increase apoptosis through downregulation of p-Akt (19). Flavonoids have been reported to exert antineoplastic effects by modulating reactive oxygen species (ROS) levels in tumor cells, inhibiting carcinogens, pro-inflammatory pathways, and angiogenesis, and by inducing apoptosis and inhibiting tumor proliferation and invasiveness (20). These mechanisms were not examined in the present study and are offered only as possible explanations for the observed cytotoxicity.

Several limitations should be considered. First, selectivity was not evaluated because normal cells were not tested; the extracts therefore cannot yet be described as having anticancer activity, only as being cytotoxic to MCF-7 cells *in vitro*. Second, MTT results depend on assay conditions such as cell density, MTT concentration, and incubation time, and should be interpreted with this in mind<sup>10</sup>. Further studies should include normal cell lines to calculate a selectivity index, a standard cytotoxic drug as a positive control, quantitative phytochemical analysis, and fractionation of the most active extract.

## CONCLUSION

The methanol, 70% ethanol, and ethyl acetate extracts of *Lagerstroemia speciosa* (L.) Pers. leaves contained the same groups of secondary metabolites qualitatively and showed *in vitro* cytotoxic activity against MCF-7 breast cancer cells, with IC<sub>50</sub> values of 98.46, 163.67, and 182.72 ppm, respectively. The methanol extract was the most active. These extracts demonstrated *in vitro* cytotoxic activity and warrant further investigation, including tests on normal cells to determine selectivity, quantitative phytochemical analysis, and fractionation of the most active extract.

## DECLARATIONS

**Author Contributions:** Ernie Halimatushadyah served as the principal researcher and was responsible for coordinating the overall research activities, supervising the study, and analyzing and interpreting the MTT cytotoxicity assay data. Dyah Ayuwati Waluyo and Krismayadi contributed to the extraction procedures and preparation of the plant extracts. Dyah Ayuwati Waluyo also contributed to the English translation and language preparation of the manuscript. All authors contributed to the preparation and revision of the manuscript, reviewed the final version, and approved the manuscript for publication.

**Conflicts of Interest:** The authors declare that they have no financial, personal, professional, or institutional conflicts of interest that could have influenced the conduct or reporting of this study.

**Declaration of Generative AI and AI-Assisted Technologies:** During the preparation of this manuscript, the authors used ChatGPT (OpenAI) solely to assist with English-language checking, grammatical correction, and improvement of sentence clarity and readability. The AI tool was not used to generate or analyze research data or to make scientific conclusions. All AI-assisted text was reviewed and revised by the authors, who take full responsibility for the final content of the manuscript.

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**Ethical Approval and Informed Consent:** Ethical approval and informed consent were not applicable to this study because the research did not involve human participants or experimental animals. The cytotoxicity study was conducted *in vitro* using the MCF-7 cell line.

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## REFERENCES

1. Bray F, Laversanne M, Sung H, Ferlay J, Siegel RL, Soerjomataram I, et al. Global cancer statistics 2022: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA: A Cancer Journal for Clinicians*. 2024;74(3): 229–263. <https://doi.org/10.3322/caac.21834>.
2. Sung H, Filho AM, Laversanne M, Ferlay J, Siegel RL, Soerjomataram I, et al. Global cancer statistics 2024: GLOBOCAN estimates of incidence and mortality worldwide for 34 cancers in 186 countries. *CA: A Cancer Journal for Clinicians*. 2026;76(4). <https://doi.org/10.3322/caac.70090>.

3. Debela DT, Muzazu SGY, Heraro KD, Ndalama MT, Mesele BW, Haile DC, et al. *New approaches and procedures for cancer treatment: Current perspectives*. SAGE Open Medicine. 2021. <https://doi.org/10.1177/20503121211034366>.
4. Yuan L, Cai Y, Zhang L, Liu S, Li P, Li X. *Promoting Apoptosis, a Promising Way to Treat Breast Cancer With Natural Products: A Comprehensive Review*. Frontiers in Pharmacology. 2022. <https://doi.org/10.3389/fphar.2021.801662>.
5. Naeem M, Iqbal MO, Khan H, Ahmed MM, Farooq M, Aadil MM, et al. *A Review of Twenty Years of Research on the Regulation of Signaling Pathways by Natural Products in Breast Cancer*. Molecules. 2022. <https://doi.org/10.3390/molecules27113412>.
6. Raju L, Lipin R, Eswaran R. Identification, ADMET evaluation and molecular docking analysis of Phytosterols from Banaba (*Lagerstroemia speciosa* (L.)Pers) seed extract against breast cancer. *In Silico Pharmacology*. 2021;9(1). <https://doi.org/10.1007/s40203-021-00104-y>.
7. Satari A, Ghasemi S, Habtemariam S, Asgharian S, Lorigooini Z. *Rutin: A Flavonoid as an Effective Sensitizer for Anticancer Therapy; Insights into Multifaceted Mechanisms and Applicability for Combination Therapy*. Evidence-based Complementary and Alternative Medicine. 2021. <https://doi.org/10.1155/2021/9913179>.
8. Dhorai N, Panneerselvam S, Rangaraj S, Thirumoorthy P, Jenni R, Venkatachalam R. Anti-cancer Potential of Biosynthesised of Zinc Oxide Nanoparticles against Human Breast Cancer Cells (MCF-7). *Nanochemistry Research*. 2025;10(3): 337–349. <https://doi.org/10.22036/ncr.2025.479525.1415>.
9. Julyantika P, Dewi N, Ganda Putra GP, Suhendra L. Effect of Solvent Type and Maceration Time on Characteristics and Stability of Lime Orange Extract (*Citrus amblycarpa*) as Natural Antioxidants in Foods. *Media Ilmiah Teknologi Pangan (Scientific Journal of Food Technology)*. 2022;9(1): 1–14.
10. Ghasemi M, Turnbull T, Sebastian S, Kempson I. The mtt assay: Utility, limitations, pitfalls, and interpretation in bulk and single-cell analysis. *International Journal of Molecular Sciences*. 2021;22(23). <https://doi.org/10.3390/ijms222312827>.
11. Reubun YTA, Kumala S, Setyahadi S, Simanjuntak P. Freezed Drying of Kelor Leaves Extract (*Moringa oleifera* Lam.). *Jurnal Sains dan Kesehatan*. 2021;3(4): 470–474. <https://doi.org/10.25026/jsk.v3i4.427>.
12. Nurmazela V, Ridwanto, Rani Z. Antioxidant Activity Test Of Barangan Banana Hump's Ethanol Extract (*Musa Paradisiaca* (L.)) With Dpph (1,1 Diphenyl-2-Picrylhydrazyl) Methods. *International Journal of Science*. 2022;3(5): 1478–1483. <http://ijstm.inarah.co.id>
13. Djamil R, Putri YE. Determination of quality parameters, toxicity test, antioxidant activity, and  $\alpha$ -glucosidase inhibitory activity of 70% ethanol extract bungur leaves (*Lagerstroemia speciosa* L. Pers.). *Asian Journal of Pharmaceutical and Clinical Research*. 2018;11(Special Issue 1): 217–221. <https://doi.org/10.22159/ajpcr.2018.v11s1.26611>.
14. Vajrabhaya L ongthong, Korsuwannawong S. Cytotoxicity evaluation of a Thai herb using tetrazolium (MTT) and sulforhodamine B (SRB) assays. *Journal of Analytical Science and Technology*. 2018;9(1). <https://doi.org/10.1186/s40543-018-0146-0>.
15. Kementerian Kesehatan Republik Indonesia. *Farmakope Herbal Indonesia Edisi II*. 2017.
16. Anil P, Manish S, Garvendra S. R, Vijay B, Tarachand K. In vitro antioxidant studies of *Lagerstroemia speciosa* leaves. *Pharmacognosy Journal*. 2010;2(10): 357–360. [https://doi.org/10.1016/s0975-3575\(10\)80109-9](https://doi.org/10.1016/s0975-3575(10)80109-9).

17. Permatasanti A, Hidayat W. *Potential of Indonesian Herbal as an Anti-Cancer Therapy: A Systemic Review of in vitro Studies*. Cancer Management and Research. 2023. p. 837–850. <https://doi.org/10.2147/CMAR.S414457>.
18. Mumtaz MZ, Kausar F, Hassan M, Javaid S, Malik A. Anticancer activities of phenolic compounds from *Moringa oleifera* leaves: in vitro and in silico mechanistic study. *Beni-Suef University Journal of Basic and Applied Sciences*. 2021;10(1). <https://doi.org/10.1186/s43088-021-00101-2>.
19. Bouyahya A, El Omari N, Bakrim S, El Hachlafi N, Blahbib A, Wilairatana P, et al. Advances in Dietary Phenolic Compounds to Improve Chemosensitivity of Anticancer Drugs. *Cancers*. 2022;14(4573): 1–20.
20. Gupta D, Guliani E. *Flavonoids: Molecular mechanism behind natural chemoprotective behavior-a mini review*. Biointerface Research in Applied Chemistry. 2022. p. 5983–5995. <https://doi.org/10.33263/BRIAC125.59835995>.