GSC Biological and Pharmaceutical Sciences
eISSN: 2581-3250 CODEN (USA): GBPSC2
Cross Ref DOI: 10.30574/gscbps
Journal homepage: https://gsconlinepress.com/journals/gscbps/

(RESEARCH ARTICLE)

Check for updates

Metabolite profiling of Minnieroot (*Ruellia tuberosa* L.) leaves using gas chromatography-mass spectrometry (GC-MS)

Juswardi Juswardi [1,*], Nur Salamah [2], Nina Tanzerina [1], Kamila Alawiyah [1], Syafrina Lamin [1] and Mustafa Kamal [1]

[1] *Department of Biology, Faculty of Mathematics and Natural Sciences, University of Sriwijaya, Indonesia.*
[2] *Program of Biology, Faculty of Mathematic and Natural Sciences, University of Sriwijaya, Indonesia.*

GSC Biological and Pharmaceutical Sciences, 2025, 30(02), 234-241

Publication history: Received on 17 December 2024; revised on 13 February 2025; accepted on 16 February 2025

Article DOI: https://doi.org/10.30574/gscbps.2025.30.2.0031

Abstract

The Minnieroot plant (*Ruellia tuberosa* L.) is widely used in traditional medicine to treat various ailments due to its pharmacological properties due to it containing various metabolites. Further research is necessary to analyze the metabolomic profiling of the ethanol extract of Minnieroot leaves using GC-MS. This research will help identify metabolite compounds with therapeutic value and their pharmacological activities. The metabolite profile of the Minnieroot leaf reveals 17 active compounds derived from three biosynthesis pathways: the pentose phosphate pathway, the shikimic acid, and the mevalonate. The proportion based on the classification consists of 9 classes with the most furan class. Among these, nine dominant compounds have been identified, including *5-Hydroxymethylfurfural*; *2,6-Octadien-1-ol, 3,7-dimethyl-, acetate*; and *Acetic acid*. These compounds exhibit a variety of biological activities, which have the potential for developing treatments based on these metabolites.

Keywords: *Ruellia tuberosa* L.; Metabolite Profiling; Bioactivity*; 5-Hydroxymethylfurfural*

1. Introduction

The Minnieroot plant (*Reullia tuberosa* L.) belongs to the Acanthaceae family and is known as Kencana Ungu in Indonesia. It is widely utilized in traditional medicine [1], [2]. This plant is used to treat various ailments due to its pharmacological properties, including antioxidant, antimicrobial, anticancer, anti-inflammatory, antifungal [3], antidiabetic, antipyretic, antihypertensive, analgesic, and antidotal effects [4]. Additionally, Minnieroot is a component of herbal beverages in Taiwan and serves as a thirst quencher [5], [2].

The diverse pharmacological activities of Minnieroot are attributed to the presence of secondary metabolites, such as flavonoids, alkaloids, saponins, and steroids [6]. Noted [7] that the leaf extract of Minnieroot contains several secondary metabolites with antidiabetic properties, functioning as amylase enzyme inhibitors, as well as exhibiting antifungal and antioxidant activities. Additionally, [8] identified compounds in Minnieroot leaf extract, including *hexadecanamide*; *9-octadecenamide*; *(Z)-octadecenamide*; and 1,*2-benzenedicarboxylic acid*. Furthermore, according to [9], the essential oils present in Minnieroot leaves including *e-phytol, tributyl acetyl citrate*; and *heptacosane* contribute to its therapeutic effects.

To further elucidate the metabolite compounds and their bioactive functions, a metabolomics approach was employed, utilizing metabolite profiling. Metabolite profiles are instrumental in identifying and characterizing the composition of plant metabolites. Research conducted by [10] revealed four active compounds in the ethanol extract of Minnieroot leaves: *9-octadecenamide*; *hexadecanamid; octadecenamide*; and *1,2-benzenedicarboxylic acid*, identified using Gas

* Corresponding author: Juswardi Juswardi

Chromatography Mass Spectrometry (GC-MS). According to [11], GC-MS is a widely used method for analyzing non-target metabolite compounds.

Given its composition and beneficial pharmacological activity, Minnieroot is considered an important medicinal plant. Therefore, further research is necessary to analyze the metabolomic profiling of the ethanol extract of Minnieroot leaves using GC-MS. This research will help identify metabolite compounds with therapeutic value and their pharmacological activities, contributing to the development of herbal medicines.

2. Materials and Methods

2.1. Plant Materials

Minnieroot leaf samples were collected in Jambu Ilir Village, Tanjung Lubuk District, Ogan Komering Ilir Regency, South Sumatra Province, Indonesia. The research site is located at coordinates 3°28'18.5" South Latitude and 104°46'02.8" East Longitude, in a lowland area.

2.2. Maceration and Extraction

Mature leaves from the 3rd to 5th positions on the shoot were selected from the Minnieroot plant. The leaf samples were dried in a shaded area, protecting them from direct sunlight. After drying, the leaves were ground into a powder, producing a material known as simplicia. Subsequently, 200 gs of simplicia was macerated with 1000 ml of 70% ethanol for three days. The mixture was then filtered through filter paper, and the resulting filtrate was evaporated using an evaporator to produce a thick extract.

2.3. Metabolite Content Analysis Using GC-MS

The extract from the Minnieroot leaves was analyzed using Gas Chromatography-Mass Spectrometry (GC-MS). A 0.1 mM sample was prepared by adding 2 ml of methanol (MeOH) and 2 ml of chloroform. This mixture was sonicated for 10 minutes and then centrifuged for 5 minutes at 10,000 rpm. The supernatant was then injected into the GC-MS system following the Agilent 7890A gas chromatography protocol.

2.4. Data Analysis

The GC-MS data were analyzed to identify the chemical components, their structures, retention times, and relative areas. The detected metabolites were further examined for their biosynthetic pathways, properties, and bioactive characteristics using resources such as PubChem, KEGG, ChEBI, PlantCyc, and SpectraBase.

3. Results and Discussion

3.1. Metabolite Profile of Minnieroot Leaves (*Ruellia tuberosa* L.)

Analysis of the metabolite profile in the ethanol extract of Minnieroot leaves using GC-MS obtained results in a chromatogram (Figure 1.). Then continued identification of the metabolite profile compounds with the conformity of the MS spectrum with the database to determine the type of compound.

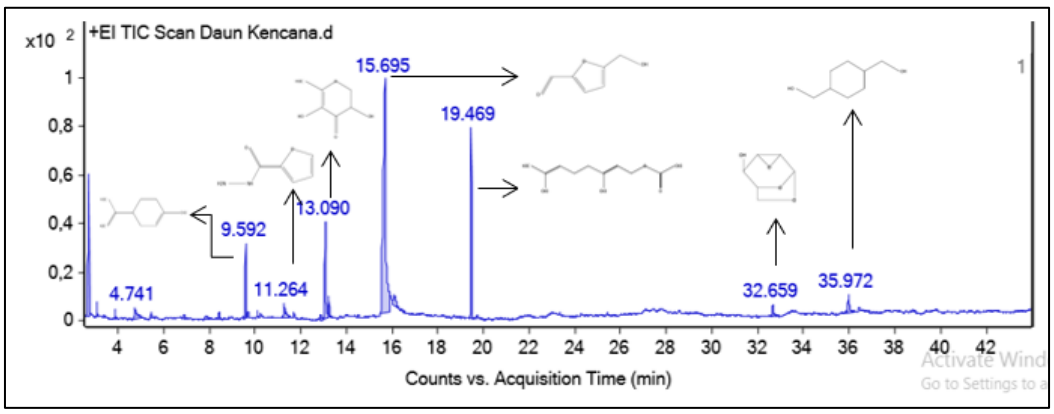

Figure 1 Fingerprint of metabolite compounds of ethanol extract of Minnieroot (*Ruellia tuberosa* L.) leaves with GC-MS

The fingerprint of metabolite compounds is based on the chromatogram in Figure 1. Seventeen detected compounds and nine main compounds were obtained. Each compound at different retention times has varying compound abundance. According to [12], GC chromatography can detect compounds with the lowest concentration, allowing for the identification of secondary metabolites in plants with varying chromatograms and mass spectra. The dominant compounds in Minnieroot leaves include: 5-*Hydroxymethylfurfural* (48.96%); *2,6-Octadien-1-ol, 3,7-dimethyl-, acetate* (11.89%); *Acetic acid* (11.58%); *4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl* (8.37%); *D-Limonene* (4.57%); *2-Furancarboxylic acid, hydrazide* (2.04%); *1,4-Cyclohexanedimethanol* (1.96%); *1-Octanol, 2-nitro* (1.28%); *2,3-Anhydro-d-mannosan* (1.02%).

The dominant compound with higher abundance is *5-Hydroxymethylfurfural* (*5HMF*) with a total abundance of 48%. This compound can be produced in plants in response to biotic or abiotic stress. The abundance of *5-HMF* compounds in plant metabolites can vary depending on the type of plant, growth conditions, and environmental factors. According to [13], the phytochemical content of a plant is influenced by several factors such as genes, light, temperature, humidity, pH, differences in altitude that can produce different compounds, and the type of solvent used.

3.2. Identification and Bioactivity of Metabolite Compounds of Minnieroot (*Ruellia tuberosa* L.) Leaves

The identification of metabolite compounds detected from Minnieroot leaves can be seen in Table 1, which explains the class of metabolite compounds detected in Minnieroot leaves, abundance, and bioactivity based on several references.

Table 1 Identification of Compounds, Classification, and Bioactivity of Metabolites of Minnieroot (*Ruellia tuberosa* L.) Leaves

Compound Names	Area (%)	Class	Bioactivity	References
Acetic acid	11.58	Alkanoic acid	Antibacterial	[14]
Acetic anhydride	0.7	Alkanoic acid	Antimicrobial	[15]
2-Propenoic acid, ethenyl ester	0.5	Cinnamic acid	Antibacterial, Anti-inflammatory	[16]
Furfural	2.16	Furan	Antimicrobial, Antiviral, Antioxidant, Antitumor	[17]
Methylene cyclopropane carboxylic acid	0.65	Alkanoic acid	Antibacterial	[18]
2-Furancarboxaldehyde, 5-methyl	0.71	Furan	Antioxidant, Antibacterial	[19]
2,4-Dihydroxy-2,5-dimethyl-3(2H)-furan-3-one	0.51	Flavonoid	Antioxidant	[20]
D-Limonene	4.57	Monoterpene	Antioxidant, Antimicrobial, Antifungi, Antiarrhythmic	[21]
2-Furancarboxylic acid, hydrazide	2.04	Furan	Antioxidant, Anticancer, Antibacterial	[22]
Ethanamine, N-ethyl-N-nitroso	0.85	Amino acid	Antioxidant, Anticancer	[23]
4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl	8.37	Ketone	Antioxidant, Antidiabetic	[24]
1-Octanol, 2-nitro	1.28	Cinnamic acid	Antimicrobial	[25]
5-Hydroxymethylfurfural	48.96	Furan	Antioxidant, Antiproliferative	[26]
5-Hydroxymethylfurfural	2.25	Furan	Antioxidant, Antiproliferative	[26]
2,6-Octadien-1-ol, 3,7-dimethyl-, acetate	11.89	Monoterpene	Antimicrobial	[27]
2,3-Anhydro-d-mannosan	1.02	Pentosans	Antiproliferative	[28]

1,4-Cyclohexanedimethanol	1.96	Cyclic Hydrocarbon	Antioxidant, Antimicrobial	[5]

The metabolite compounds found in Minnieroot leaves consist of various organic compounds classified into categories such as alkanoic acid, carboxylic acid, cinnamic acid, aldehyde, alkaloic acid, flavonoids, monoterpenes, phenols, primary amines, benzoids, carbohydrates, and cyclic hydrocarbons, with cyclic hydrocarbons being the dominant class. According to [29], different parts of the *Ruellia tuberosa* L. plant contain varying contents and amounts of bioactive compounds. The leaves are notably rich in apigenin, luteolin, alkaloids, flavonoids, triterpenoids, steroids, and saponins.

The phytochemical properties of Minnieroot leaves offer numerous medicinal benefits. The leaves are particularly known for their high antioxidant content, which [30] identify as a strong attribute of the extract. This quality can be valuable in pharmacology for developing herbal medicines and combating free radicals. Additionally, the roots and flowers of the Minnieroot plant contain compounds with antibacterial properties.

In summary, 17 active compounds have been identified in the extract from Minnieroot leaves, categorized into three primary biosynthesis pathways. The comparison of these biosynthesis pathways for the active compounds present in Minnieroot leaves is depicted in Figure 2.

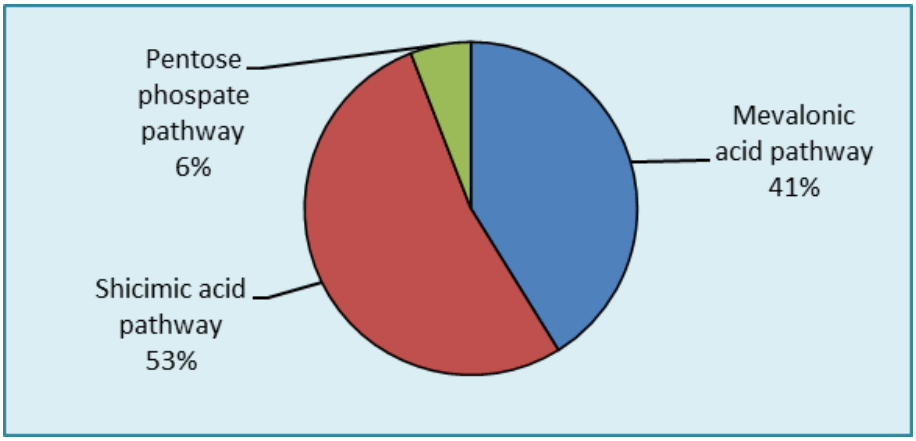

Figure 2 Metabolite Compounds Based on Biosynthesis Pathway in Minnieroot (*Ruellia tuberosa* L.) Leaves

Comparison of metabolite compounds from Minnieroot leaves based on biosynthesis pathways as shown in Figure 2 reveals that out of 17 active compounds, 6% are from the pentose phosphate pathway biosynthesis, while a larger proportion, 53%, is from the shikimic acid pathway, and 41% from the mevalonate pathway. According to [31] Silalahi (2017), groups of secondary metabolite compounds are formed from biosynthesis pathways with each precursor. Secondary metabolites in plants can originate from the shikimic acid pathway, malonic acid pathway, mevalonic acid pathway, and the methylerythritol phosphate pathway.

Based on the 17 biologically active compounds identified in Minnieroot leaves and traced through biosynthesis pathways, 11 types of metabolite compound classes were obtained. A comparison of the classes of metabolite compounds from Minnieroot leaves is presented in Figure 3.

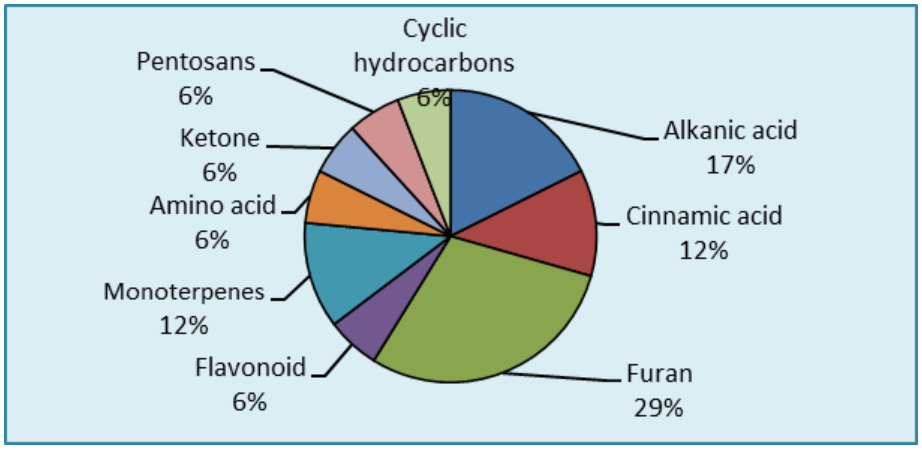

Figure 3 Metabolite Class of Metabolite Compounds in Minnieroot (*Ruellia tuberosa* L.) Leaves

Figure 3 shows the classification of various identified compounds into different classes. The distribution is as follows: amino acid class 6%, monoterpene class 12%, flavonoids 6%, alkanoic acids 17%, cinnamic acids 12%, cyclic hydrocarbons 6%, pentosans 6%, amino acids 6%, ketones 6%, and furan classes dominate at 29%.

Based on the analysis of active compounds present in Minnieroot leaves, they can be grouped as follows:

- **Furan Compounds Group**. The largest proportion of secondary metabolites in Minnieroot leaves belongs to the furan group, accounting for 29%. Active compounds classified in this group include *Furfural*, *2-Furancarboxaldehyde*, *5-methyl-2-Furancarboxylic acid*, *hydrazide*, and *5-HMF*. These compounds are derived from the biosynthesis of the shikimic acid pathway and exhibit various biological activities, including antioxidant, antimicrobial, anticancer, antitumor, antiviral, and antiproliferative effects [17].
- **Alkanoic Acids Group.** The second largest group of secondary metabolites in Minnieroot leaves comes from the alkanoic acid group, which comprises 17%. Active compounds in this class include *acetic acid*, *acetic anhydride*, and *methylene cyclopropane carboxylic acid*. These compounds are synthesized through the mevalonic acid pathway and demonstrate antibacterial activity [18].
- **Cinnamic Acids Group.** Secondary metabolites derived from the cinnamic acid group account for 12% of the total compounds found in Minnieroot leaves. The active compounds included in this group are *2-Propenoic acid, ethenyl ester*, and *1-Octanol, 2-nitro*. Like the furan compounds, these also arise from the shikimic acid pathway, displaying a range of biological activities, such as antimicrobial, antibacterial, anesthetic, and anti-inflammatory properties [16].
- **Monoterpenes Group**. Compounds from the monoterpene group are 12%, and the active compounds of this group are *D-Limonene* and *2,6-Octadien-1-ol, 3,7-dimethyl-*, and acetate. Monoterpene compounds come from the biosynthesis of the mevalonic acid pathway which has biological activities including antimicrobial, antioxidant, and antiarrhythmic [21].
- **Flavonoid Group.** Secondary metabolite compounds of Minnieroot leaves from the flavonoid group were obtained as much as 6%. The active compound component of the flavonoid class, namely *2,4-Dihydroxy-2,5-dimethyl-3(2H)-furan-3-one* comes from the biosynthesis of the shikimic acid pathway. This compound has biological activity as an antioxidant [20].
- **Amino Acid Group.** The secondary metabolites of Minnieroot leaves are amino acid groups, namely *Ethanamine, N-ethyl-N-nitroso* compounds as much as 6% and are amino acids derived from the biosynthesis of shikimic acid pathways. *Ethanamine, N-ethyl-N-nitroso* compounds function as antioxidants and anticancer [23].
- **Ketone Group.** The ketone group comprises metabolites that also account for 6%. Active compound components in the ketone class include *4H-Pyran-4-one* and *2,3-dihydro-3,5-dihydroxy-6-methyl*. These compounds are derived from the mevalonic acid biosynthesis pathway and have biological activities as antioxidants and antidiabetics [24].
- **Pentosan Group.** Minnieroot leaf secondary metabolites include the pentosan group, specifically glucose *2,3-Anhydro-d-mannosan*, which constitutes 6% of the total. This glucose compound is derived from the pentose phosphate biosynthesis pathway and exhibits antiproliferative activity [28].

- **Cyclic Hydrocarbon Group.** The compound *1,4-Cyclohexanedimethanol* is a secondary metabolite compound of Minnieroot leaves including the cyclic hydrocarbon group as much as 6%. This compound comes from the biosynthesis of the mevalonic acid pathway with biological activity as an antioxidant and antimicrobial [5].

4. Conclusion

The metabolite profile of the Minnieroot leaf (*Ruellia tuberosa* L.) reveals 17 active compounds derived from three biosynthesis pathways: the pentose phosphate pathway (6%), the shikimic acid pathway (53%), and the mevalonate pathway (41%). The proportion based on the classification consists of amino acid class (6%), monoterpene (12%), flavonoid (6%), alkanoic acid (17%), cinnamic acid (12%), cyclic hydrocarbon (6%), pentosan (6%), ketone (6%), and the most furan class (29%). Among these, nine dominant compounds have been identified, including *5-Hydroxymethylfurfural* (48.96%), *2,6-Octadien-1-ol, 3,7-dimethyl-, acetate* (11.89%), *Acetic acid* (11.58%), *4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl* (8.37%), *D-Limonene* (4.57%), *2-Furancarboxylic acid, hydrazide* (2.04%), *1,4-Cyclohexanedimethanol* (1.96%), *1-Octanol, 2-nitro* (1.28%), *2,3-Anhydro-d-mannosan* (1.02%) These compounds exhibit a variety of biological activities, including antioxidant, antimicrobial, antibacterial, anesthetic, anti-inflammatory, antiviral, antitumor, anticancer, antiarrhythmic, antidiabetic, antifungal, and antiproliferative properties. This suggests significant potential for developing treatments based on these metabolites.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declared no conflict of interest.

References

[1] Afzal, K., Uzair, M., Chaudary, B. A., Ahmad, A., Afzal, S., and Saadullah, M. (2015). Genus Ruellia: Pharmacological and Phytochemical Importance in Ethnopharmacology. *Acta Poloniae Pharmaceutica: Drug reaserch,* 77(5): 821-827.

[2] Matos P, Batista MT, and Figueirinha A. (2022). A review of the ethnomedicinal uses, chemistry, and pharmacological properties of the genus Acanthus (Acanthaceae). *J Ethnopharmol* 293: 115271. DOI: 10.1016/j.jep.2022.115271.

[3] [3] Intan, A. E. K., Jannah, N., and Septiana, S. (2020). Pharmacological activities of *Ruelia tuberosa*. *Infokes*, 10(1): 239-243.

[4] Khan I, Jan SA, Shinwari ZK, Ali M, Khan Y, and Kumar T. (2017). Ethnobotany and medicinal uses of folklore medicinal plants belonging to family Acanthaceae: An updated review. *MOJ Biol Med* 1 (2): 34-38. DOI: 10.15406/mojbm.2017.01.00009.

[5] Hu, Y., Zhao, Z., Liu, Y., Li, G., Wang, A., Cong, Y., Zhang, T., Wang, F. and Li, N., (2018). Synthesis of 1, 4-Cyclohexanedimethanol, 1, 4-Cyclohexanedicarboxylic Acid and 1, 2-Cyclohexanedicarbo xylates from Formaldehyde, Crotonaldehyde and Acrylate/Fumarate. *Angewandte Chemie International Edition*, 57(23): 6901-6905. https://doi.org/10.1002/anie.201801287

[6] Vitalia, N., Ahmad N., and Aktsar R. A., (2016) Toxicity Test of Pletekan Leaf Extract (*Ruellia tuberosa* L.) using the Brine Shrimp Lethality Test (BSLT) method (Ina). *Jurnal Fitofarmaka Indonesia*, 3 (1): 124-12

[7] Safitri, A., Roosdiana, A., Rosyada, I., Evindasari, C.A., Muzayyana, Z. and Rachmawanti, R. (2019.) Phytochemicals screening and anti-oxidant activity of hydroethanolic extracts of *Ruellia tuberosa* L. In *IOP Conference Series: Materials Science and Engineering* . 509(1): 012-017.

[8] Nopiari, I. A., Adriani A. N P., and Intan W. N. (2016). Identifikasi senyawa aktif daun pletekan (*Ruellia tuberosa* l.) dengan menggunakan GC-MS. *SIMBIOSIS.* 4(2): 55-57. https://ojs.unud.ac.id/index.php/simbiosis/article/view/28755/17873

[9] Dorcas, O. M., and Sherifat, A. A. (2015). Composition of volatile oils from leaf, stem, root, fruit, and flower of *Ruellia tuberosa* L. (Acanthaceae) from Nigeria. *Journal of Medicinal Plants Research,* 9(41): 1031-1037.

[10] Ida, N.P. and Intan, N. (2016). Identification of Active Compounds in Pletekan Leaves (*Ruellia tuberosa* L) Using GC-MS (Ina). *Jurnal Simbiosis IV*. 2(1): 250-259.

[11] Putiani, K., Juswardi and Harmida (2022). Differences and Abundance of Moringa Leaf Metabolites (Moringa oleifera Lam.) Based on Altitude in South Sumatra. *Proceedings of SNPBS (National Seminar on Biology and Science Education)*: 205-212. https://proceedings.ums.ac.id/ snpbs/article/view/1760

[12] Al-Rubaye, A. F., I. H. Hameed, and Moh. J. Kadhim. (2017). A Review: Uses of Gas Chromatography-Mass Spectrometry (GCMS) Technique for Analysis of Bioactive Natural Compounds of Some Plants. *International Journal of Toxicological and Pharmacological Research*. 9(1): 81-85.

[13] Kartika, L., Mirhansyahardana, and Rolan, R. (2020). Antioxidant Activity of Plants of the Genus Artocarpus (Ina). *Jurnal Farmasi*. 3(1): 237-244.

[14] Fraise AP, Wilkinson MA, Bradley CR, Oppenheim B, and Moiemen N. (2013). The antibacterial activity and stability of acetic acid. *J Hosp Infect*. 84(4): 329-31.

[15] Dawood, A.A., Mohammed, S.R. and Mahmoud, M., (2020). Synthesis, identification and biological activity of new heterocyclic compounds from reaction of new schiff-bases with phathalic anhydride. *Science Journal of University of Zakho*, 8(1): 12-18.

[16] Kiswandono, A.A., Iswanto, A.H., Susilowati, A., and Lumbantobing, A.F. (2016). Analisis Kandungan Asam Sinamat dan Skrining Fitokimia Getah Kemenyan Jenis Bulu (*Styrax benzoine* var *Hiliferum*) dari Tapanuli Utara. *Prosiding Seminar Nasional Kimia Lombok* Aug 10-11 2016.

[17] Donlawson, C., Nweneka, D.O., Orie, K.J. and Okah, R. (2020). Synthesis and bioactivity of 1-((2-carbamoylguanidino) (furan-2-ylmethyl) urea. *American Journal of Analytical Chemistry*, 11(7): 280-288.

[18] Sato, H., Macchiarulo, A., Thomas, C., Gioiello, A., Une, M., Hofmann, A.F., Saladin, R., Schoonjans, K., Pellicciari, R. and Auwerx, J., (2008). Novel potent and selective bile acid derivatives as TGR5 agonists: biological screening, structure-activity relationships, and molecular modeling studies. *Journal of medicinal chemistry*, 51(6): 1831-1841.

[19] Fagbemi KO, Aina DA, Adeoye-Isijola MO, Naidoo KK, Coopoosamy RM, and Olajuyigbe OO. (2022). Bioactive compounds, antibacterial and antioxidant activities of methanol extract of *Tamarindus indica* Linn. *Sci Rep*. 8. 12(1):9432. doi: 10.1038/s41598-022-13716-x.

[20] Majumder, S., Saha, S., Ghosh, A. *et al*. (2021). Production of fermented tea petal decoction with insights into in vitro biochemical tests, antioxidant assay and GC-MS analysis. *Food Prod Process and Nutr*. 3:32. https://doi.org/10.1186/s43014-021-00075-9

[21] Ribeiro AD, Cardoso MNA, Freire JCP, Figueirêdo Jr EC, Costa MMA, Silva PG, Gomes DQC, Brito EMM, and Pereira JV. (2023) Antimicrobial activity of limonene: Integrative review Bol Latinoam Caribe *Plant Med Aromat* 22 (5): 581-593. https://doi.org/10.37360/blacpma.23.22.5.42

[22] Sivanandhan, M., Seeman, U. and Parasuraman, A. (2024). *In-silico* molecular docking, ADMET and DFT evaluation of piperidin-4-one furoic hydrazone derivatives as antimicrobial, antioxidant and anticancer agents. *J IRAN CHEM SOC* .21: 463-478.

[23] Sambath Kumar, R. (2008). The Antioxidant Defense System Induced by Methanol Extract of Careya arborea in N-Nitroso-diethylamine (NDEA) Induced Hepatocarcinogenesis. *Journal of Complementary and Integrative Medicine*, 5(1). doi.org/10.2202/1553-3840.1125

[24] Özdemir Ö, Çöl Z, and Ertürk Ö. (2023) Efficacy of Bee Products (Anzer Honey, Pollen and Propolis) in Detection and Healing of Damage Induced by Antidiabetic Drug Vildagliptin/ Metformin Hydrochloride in Healthy Human Pancreatic Cells: Cytotoxic, Genotoxic and Biochemical Studies. *Curr Med Sci*. 43(6):1173-1182.

[25] Muthulakshmi, A.R.J.M. and Mohan, V.R., (2012). GC-MS analysis of bioactive components of *Feronia elephantum* Correa (Rutaceae). *Journal of Applied Pharmaceutical Science*, 2(2): 69-74.

[26] Zhang, H., Jiang, Z., Shen, C., Zou, H., Zhang, Z., Wang, K., Bai, R., Kang, Y., Ye, X.Y. and Xie, T., (2021). 5-hydroxymethylfurfural alleviates inflammatory lung injury by inhibiting endoplasmic reticulum stress and NLRP3 inflammasome activation. *Frontiers in Cell and Developmental Biology*, 9: 782427. doi: 10.3389/fcell.2021.782427.

[27] Hasan, M.M., Al Mahmud, M.R. and Islam, M.G., (2019). GC-MS analysis of bio-active compounds in ethanol extract of *Putranjiva roxburghii* Wall. fruit peel. *Pharmacognosy Journal*, 11(1): 146-149. http://dx.doi.org/10.5530/pj.2019.1.24

[28] Loganathan, S., Selvam, K., Sivasakthi, V., Prakash, P., Yamuna, M., Lalitha, K., Shivakumar, M.S. and Senthil Nathan, S., (2021). Phytochemical and pharmacological evaluation of methanolic extract of *Knoxia sumatrensis* leaves. *Journal of Herbs, Spices & Medicinal Plants*, 27(2): 200-217.

[29] Chaitanya, B.Khrisna, Atigari, Diana, Vivian, Babu, S.Ravindra, Ravella, Alekhya, Vardhan, and Jayasree. (2012). Hypolipidemic and Anti Oxidant Activity of *Ruellia tuberosa* Linn. *International Journal of Pharmacy an Biological Science*. 2(3): 63-72

[30] Mayangsari, E., Kalsum, U., and Pragiwaksana, R. G. A. (2020). The effect of Kencana Ungu (*Ruellia tuberosa*) leaf extract on intestinal malondialdehyde (MDA) levels in rats induced by indomethacin (Ina). *Majalah Kesehatan FKUB*. 7(2): 97-101.

[31] Silalahi, M., (2017). Secondary metabolite compounds in *Etlingera elatior* (Jack) RM Smith (Ina). *Jurnal Nasional Pendidikan Biologi dan Saintek*. 2(3): 41-47. https://proceedings.ums.ac.id/snpbs/article/download/357/355/361.