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## Simple TLC-densitometric method for the quantification of asiaticoside in *Centella asiatica* from different origins for its standardization FREE

K. Kartini ■; A. Rosidah; R. Budiono



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# Simple TLC-Densitometric Method for the Quantification of Asiaticoside in *Centella asiatica* from Different Origins for Its Standardization

K Kartini<sup>1,a)</sup>, A Rosidah<sup>1,b)</sup> and R Budiono<sup>2,c)</sup>

<sup>1</sup>Department of Pharmaceutical Biology, Faculty of Pharmacy, University of Surabaya, Jl. Raya Kalirungkut 60293 Surabaya, Indonesia

<sup>2</sup>Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Surabaya, Jl. Raya Kalirungkut 60293 Surabaya, Indonesia

a) Corresponding author: kartini@staff.ubaya.ac.id
b) farmasi@unit.ubaya.ac.id
c) farmasi@unit.ubava.ac.id

Abstract. Gotu kola (*Centella asiatica* (L.) Urb.) is one of the "Jamu Saintifik" ingredients that contains various active compounds, including asiaticoside. Asiaticoside content depends on several factors, such as the plant's origin, age, and harvesting season. This study was conducted to analyze the concentration of asiaticoside in gotu kola crude drug obtained from nine different planting locations using a TLC-densitometer. The TLC used silica gel 60 F<sub>254</sub> as the stationary phase, ethyl acetate-methanol-water (10:2.5:1) as the mobile phase, and anisaldehyde sulfuric acid as a derivatizing agent. The developed method met the specificity parameters, as characterized by the identical visible spectrum of the asiaticoside in the sample extract to that of the asiaticoside standard, with a maximum wavelength of 594 nm. The standard curve showed good linearity in the concentration range of 1.05–10.5 μg/band with an r-value of 0.989437. The limits of detection (LOD) and quantification (LOQ) were 0.48820 and 1.47939 μg/band, respectively. This method was repeatable and precise, with relative standard deviations between 2.72 and 7.16% for intraday precision and 2.82% for interday precision. Except for two samples, all the asiaticoside concentrations were quantifiable and ranged between 0.53933 and 3.46866 mg/g of crude drugs. In conclusion, they vary across the *C. asiatica* samples collected from different origins in Indonesia. The gotu kola crude drugs obtained from Lombok Barat, Magelang, and Hulu Sungai Selatan have the highest asiaticoside content among the nine samples studied.

Keywords: Asiaticoside, Centella asiatica, Gotu Kola, TLC-Densitometer

#### INTRODUCTION

Centella asiatica (L.) Urban, also known as Gotu Kola, is a plant with herbaceous habitus belonging to the Apiaceae family. In Indonesia, it is known by various regional names, including pegagan, kaki kuda, and antanan gede. C. asiatica grows wild throughout the country and other tropical regions in general (India, Sri Lanka, China, Malaysia, Thailand, etc.) at varying altitudes, from lowlands to 2500 m above sea level [1]. C. asiatica is a perennial herb that is stemless but has short rhizomes and creeping stolons, 10–80 cm long, and has 2–10 single leaves arranged in a rosette.

Traditionally, *C. asiatica* is used to treat problems related to skin, nerves, and metabolic processes [2]. Previous research at the clinical trials level has also confirmed the effectiveness of *C. asiatica* and its compounds as anti-inflammatory, antioxidant, anti-apoptotic, and in improving mitochondrial function [3].

C. asiatica contains mainly pentacyclic triterpene compounds, such as asiatic acid, asiaticoside, madecassic acid, and madecassoside. It also contains flavonoids (quercetin and kaempferol), phytosterols (campesterol, sitosterol, and

stigmasterol), gulonic acid, ferulic acid, and chlorogenic acid. However, the pharmacological activity of the herb and its derivative products is particularly ascribed to triterpenes, especially asiaticoside. Various standardized *C. asiatica* extracts traded in the market are also standardized with the concentrations of this marker compound [2]. Monograph of *C. asiatica* in various herbal pharmacopeias (Ayurvedic Pharmacopeeia of India, European Pharmacopeeia 9.0, Hong Kong Chinese Materia Medica Vol. 7, Indian Pharmacopeeia 2018, Malaysian Herbal Monograph 2009 Vol. 2, Pharmeuropa, Thai Herbal Pharmacopeeia 2018, USP41-NF36 S1, Vietnamese Pharmacopeeia 2017, and Indonesian Herbal Pharmacopeeia 2017) also use asiaticoside as a marker for analytical purposes.

Having a high-quality and uniform supply of raw materials is a problem faced by herbal medicine industries worldwide, including in Indonesia. It is in part because medicinal plants are grown or cultivated in various geographies across which temperature, humidity, rainfall, sunlight intensity, soil types, and other ecological factors may differ. These factors result in diverse biomass and concentration of chemical compounds in a plant. For instance, variations in asiaticoside contents of *C. asiatica* samples collected from locations with different geographical conditions have been previously reported [4–6]. Also, there have been accounts of variations in biomass due to different cultivation treatments [7]. However, variations in asiaticoside contents of *C. asiatica* samples from various locations in Indonesia remain under researched [8].

Several methods for detecting and measuring asiaticoside contents are TLC-densitometry [8–10], HPTLC-densitometry [4,11,12], and HPLC [13–15]. TLC is a separation method that has been used to analyze medicinal plants qualitatively and quantitatively for many years. Although other more sophisticated techniques have been developed, TLC remains the method of choice for herbal analysis and is adopted by many pharmacopeias because of its many advantages: multiple samples, including comparison samples, that can be analyzed simultaneously, selection of stationary and mobile phases that can be performed flexibly, simple equipment, ease of use, high reproducibility, and relatively low cost of analysis [16].

Considering the high industrial demand for raw *C. asiatica*, this research was conducted to develop and validate the TLC-densitometric method for identifying and measuring asiaticoside concentration of *C. asiatica* (i) and to determine the asiaticoside contents of *C. asiatica* samples from various locations in Indonesia using the validated TLC-densitometry (ii). It is expected that with a simple analytical method, as derived in this study, the standardization or quality control of *C. asiatica* as raw materials and their derivative products can be conducted easily.

#### **MATERIAL AND METHODS**

#### **Chemicals and Reagents**

The asiaticoside standard was obtained from Sigma-Aldrich (USA). Precoated TLC Si Gel 60  $F_{254}$  and proanalytical grade solvent were from Merck (Darmstadt, Germany).

#### **Collection of Samples**

Centella asiatica herbs were collected in July-September 2020 from four islands in Indonesia: Jawa (Tawangmangu, Batu, Probolinggo, and Magelang), Bali (Bangli), Lombok (Mataram, Lombok Barat, and Lombok Timur), and Kalimantan (Hulu Sungai Selatan). Details of the sampling locations are provided in Table 1. All the samples have been authenticated by the Center for Information and Development of Traditional Medicines (PIPOT), University of Surabaya.

**TABLE 1.** Locations of the *Centella asiatica* samples

Codes	Cities	Coordinates (Latitude, Longitude)	Elevation (masl)
a	Tawangmangu	7°39'49" S, 111°08'06" E	1,123
b	Batu	7°52'02" S, 112°31'11" E	895
c	Probolinggo	7°57'38" S, 113°23'00" E	550
d	Magelang	7°29'44" S, 110°08'10" E	714
e	Mataram	8°33'58" S, 116°07'27" E	33
f	Lombok Barat	8°34'17" S, 116°13'31" E	193
g	Lombok Timur	8°22'16" S, 116°32'09" E	1,166
h	Bangli	8°21'53" S, 115°20'44" E	850
i	Hulu Sungai Selatan	2°47'05" S, 115°15'37" E	23

#### **Extraction**

One gram of *C. asiatica* powder was first extracted with 7 ml of methanol using the ultrasonication method for 15 minutes at room temperature. Then, the extraction results were filtered and put into a 10 ml volumetric flask. The residue was rinsed with 3 ml of methanol and put into the same volumetric flask while filtered, then methanol was added until 10 ml.

#### **Preparation of the Standard Solutions**

To prepare the standard solutions, approximately 1.00 mg of asiaticoside standard was weighed, then put into a 1.5 ml microtube and added with 1000 µl of methanol using a micropipette.

#### **TLC System**

Samples of the *C. asiatica* extract and asiaticoside standard were spotted on the silica gel 60  $F_{254}$  TLC plate in the form of 6 mm bands using a CAMAG 100-microliter sample syringe (Hamilton, Switzerland). The spotting was conducted automatically using a Linomat 5 sample applicator (CAMAG, Switzerland) with the help of  $N_2$  gas. Then, elution was performed in a twin-through chamber (CAMAG, Switzerland) saturated with the mobile phase (ethyl acetate-methanol-water = 10:2.5:1 v/v/v) for 30 minutes. It was set in an ascending mode, with an elution distance of 80 mm from the spotting site. Afterward, the plate was dried in a fume hood, sprayed with anisaldehyde sulfuric acid reagent, and heated in an oven at  $110^{\circ}$ C for 10 minutes. Then, the TLC plate was documented in UV 254 nm, UV 365 nm, visible light and scanned using the TLC Scanner 4 (CAMAG, Switzerland) equipped with winCATS software, a scanning slit of 4 mm  $\times$  0.3 mm, and a data resolution of 1 nm/step at a scanning speed of 100 nm/s.

#### **Method Validation**

#### Specificity Determination

First, 4  $\mu$ l of the standard asiaticoside solution and 6  $\mu$ l of the *C. asiatica* extract sample were spotted on the TLC plate and then eluted and documented according to the designed TLC system. Then, to determine the specificity of the method, the bands of the asiaticoside standard and sample were compared to observe their Rf values, the shape of the visible spectrum, and maximum wavelength value using a densitometer. In addition, the densitometer was also used to determine if the asiaticoside band contained only one compound or more by measuring the visible spectrum at the beginning, apex, and end of the peak [17].

 $Determination\ of\ Linearity,\ Limit\ of\ Detection\ (LOD),\ and\ Limit\ of\ Quantification(LOQ)$ 

Linearity was determined by spotting the standard asiaticoside solution at various volumes (1, 2, 4, 6, 8, 10 µl). For each volume, spotting was replicated three times on different plates. After elution and derivatization, the asiaticoside band was scanned with a TLC-scanner at the maximum wavelength obtained in the previous step, specificity determination. Afterward, the derived asiaticocide area (as y) was plotted against the mass of the spotted

asiaticoside (as x) to create a standard curve. The curve's linearity was determined from the correlation coefficient (r-value) of the formed linear regression equation (y = bx + a). The limit of quantification (LOQ) and detection (LOD) was each calculated from the standard deviation (SD) of the mean intercept and slope ( $\bar{x}$ ) in the linear regression equation of the standard curve. The equations used were LOD = 3.3 (SD/ $\bar{x}$ ) and LOQ = 10 (SD/ $\bar{x}$ ) [12].

#### Precision Determination

Intraday precision was determined by spotting 4  $\mu$ l of the standard asiaticoside solution onto a plate six times, creating six replicates. The eluted TLC plate was analyzed using a densitometer, then the mean, standard deviation, and relative standard deviation (%RSD) of the asiaticoside area were calculated. To determine the interday precision, 4  $\mu$ l of the standard asiaticoside solution was spotted onto three different plates on three days. Afterward, the eluted plate was analyzed using a densitometer, then the mean, standard deviation, and relative standard deviation (%RSD) of the detected asiaticoside area were calculated [17].

#### Determination of the Asiaticoside Contents of *C. asiatica* Samples

To calculate the asiaticoside concentration in the C. asiatica herbs from nine (9) different locations, 8  $\mu$ l of each extract sample was spotted onto a TLC plate and analyzed by the validated method. The analysis was performed in three replications. For each sample, the asiaticoside area detected by the densitometer was substituted into the standard curve's regression equation, and the asiaticoside content was measured in mg/g dry weight.

#### **Data Analysis**

Statistical analyses were conducted to distinguish the asiaticoside concentration of the *C. asiatica* herbs obtained from various locations. One-way ANOVA ( $\alpha$ = 0.05) and a post hoc Tukey test were processed in GraphPad Prism Version 5.01.

#### RESULTS AND DISCUSSION

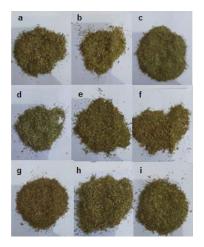
#### Physical Characteristics of the Sample Powder

The *Centella asiatica* herbs investigated in this study were collected from four major islands in Indonesia to represent areas with varying geographical conditions at different elevations, from 33 to 1,166 masl. The crude drugs produced from the samples shared similar organoleptic properties: greyish-green, dry, and brittle leaves (easily crumbled when crushed). Pictures of the nine powdered crude drugs are shown in Fig. 1.

#### **Specificity**

Specificity was determined by comparing similarities in color and Rf value between the bands of the asiaticoside standard and sample. In addition, their visible spectra were also overlaid to see any shared similarities. In the specificity analysis, the maximum wavelength of the asiaticoside standard was scanned within the range of 200–700 nm.

The chromatograms of the asiaticoside standard and sample shows that the band of the suspected asiaticoside in the sample had the same color as that of the asiaticoside standard, i.e., greenish blue in visible light (Fig. 2). The position of the two compound bands was also parallel.



**FIGURE 1.** Organoleptic characteristics of the *Centella asiatica* powder from Tawangmangu (a), Batu (b), Probolinggo (c), Magelang (d), Mataram (e), Lombok Barat (f), Lombok Timur (g), Bangli (h), and Hulu Sungai Selatan (i)

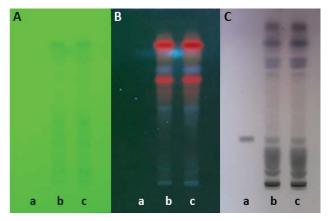


FIGURE 2. TLC chromatograms of the asiaticoside standard (a) and the *Centella asiatica* extract (b, c) with anisaldehyde sulfuric acid as the derivatization reagent, detected at 254 nm (A), 365 nm (B), and in visible light (C)

Results of specificity analysis shows the overlaid UV-vis spectra of the asiaticoside standard and the sample (Fig. 3). It indicates that the spectrum profile of the sample coincided with that of the standard, with a maximum wavelength of 594 nm. This result corresponds to a previous study that used maximum wavelengths of 595 nm [12] and 600 nm [4] for asiaticoside analysis.

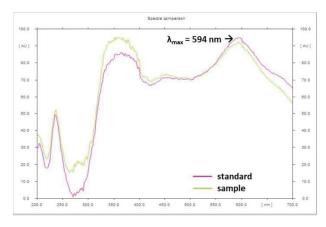


FIGURE 3. Overlaid UV-Vis spectra of the asiaticoside standard and the asiaticoside sample

Densitograms of the asiaticoside standard and sample shows that both of them had the same Rf value, 0.34 (Fig. 4). The peak purity of the sample (Table 2) is indicated by the values of r (s, m) and r (m, e), i.e., 0.998946 and 0.995208; both were > 0.99 [12]. From these data, it can be inferred that the TLC method designed for measuring the asiaticoside contents of *C. asiatica* has good specificity.

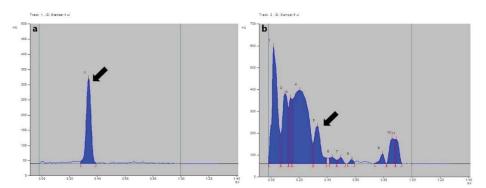


FIGURE 4. Densitograms of the asiaticoside standard (a) and Centella asiaticosida extract (b)

**TABLE 2.** Rf and peak purity parameter values of the asiaticoside standard and sample

Tracks	Identities	Assigned substances	Rf	r (s, m)	r (m, e)
1	Standard	Asiaticoside	0.34	0.999556	0.999006
2	Samples	Asiaticoside	0.34	0.998946	0.995208

#### Linearity, Limit of Detection (LOD), and Limit of Quantification (LOQ)

Linearity was determined by calculating the correlation coefficient (r) of the standard curve's linear regression equation. The standard curve was obtained from spotting different volumes of the standard asiaticoside solution (1.05  $\mu$ g/ $\mu$ l), i.e., 1, 2, 4, 6, 8, and 10  $\mu$ l or equivalent to 1.05, 2.1, 4.2, 6.3, 8.4, and 10.5  $\mu$ g/band. The TLC chromatogram and two-dimensional densitogram of the standard asiaticoside solution are shown in Fig. 5, while the standard curve formed is in Fig. 6.

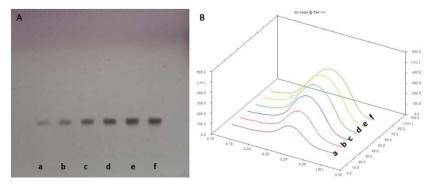


FIGURE 5. TLC chromatogram (A) and 2D densitogram (B) of the standard asiaticoside solution

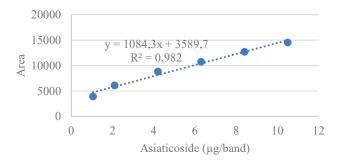


FIGURE 6. The curve of the linear regression equation between asiaticoside concentration and area

The final standard curve's equation, formulated from three replications, was y = 1084.3x + 3589.7. The LOD and LOQ calculated using the equation were  $0.48820 \,\mu\text{g/band}$  and  $1.47939 \,\mu\text{g/band}$ , respectively. The *r*-value obtained was 0.989437, indicating a linear relationship between concentrations in the range of  $1.05-10.5 \,\mu\text{g/band}$  and each of the asiaticoside areas [4,12]. Previous research using a combination of TLC and densitometry also found LOD and LOQ nearly identical to the findings in this study, i.e.,  $0.72 \,\mu\text{g}$  and  $1.2 \,\mu\text{g}$  [9]. However, in another study using HPTLC-densitometer, the derived LOD and LOQ of asiaticoside are lower [4,12,18]. LOD and LOQ are defined as the smallest amount of analyte that can be detected or quantified with reasonable statistical certainty, depending on the analytical method used. HPTLC is distinguished from TLC in that the former uses a stationary phase with a smaller particle size than the latter, thus providing better analytical performance, e.g., a smaller volume of the mobile phase, better precision, shorter analysis time, and increased sensitivity [19,20].

#### Precision

To determine intra- and interday precisions, the spotting of the asiaticoside standard onto the TLC plate was replicated six times and repeated on three different days. The asiaticoside areas read after the TLC-densitometer analysis are shown in Table 3.

**TABLE 3.** Intraday precision and interday precision test results

Donlination		Areas	
Replication	Day 1	Day 2	Day 3
1	13300.32	14416.70	13458.23
2	13683.41	13861.42	13748.83
3	13962.02	13588.83	13539.71
4	13327.32	13305.22	13683.28
5	12444.93	13085.16	13725.45
6	11433.20	12655.40	14505.11
Mean±SD	$13025.20\pm932.63$	$13485.46\pm616.60$	$13776.77 \pm 374.25$
RSD (%)	7.16	4.57	2.72
Intraday precision (%	(6RSD, n = 6) = 2.72 - 7.16		
Interday precision (%	6RSD, $n = 3$ ) = 2.82		

Based on Table 3, it can be concluded that the TLC-densitometry method designed for detecting and measuring the asiaticoside contents of *C. asiatica* has good intra- and interday precision, with %RSD of 2.72–7.16 and 2.82, respectively [4,12].

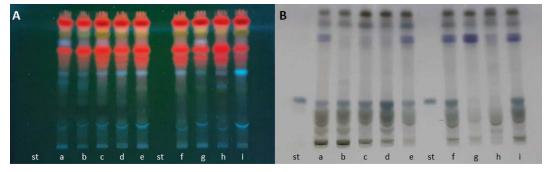
#### Determination of Asiaticoside Concentration in Centella asiatica Samples

As seen in Table 4, the samples obtained from Lombok Timur and Bangli had minute asiaticoside contents (below the LOQ) that were not quantifiable. On the contrary, the other seven samples had varying concentration of asiaticoside, from 0.5393 to 3.4687 mg/g. Figure 8 shows that samples from Magelang, Lombok Barat, and Hulu Sungai Selatan had the highest asiaticoside content, while those from Batu, Probolinggo, and Mataram had the second-highest. The asiaticoside of the Tawangmangu sample was significantly lower than the *C. asiatica* collected from other regions (p<0.05), making it the smallest concentration detected in the study. However, although the samples from Tawangmangu, Lombok Timur, and Bangli had low asiaticoside, they contained other metabolites in a quite substantial or even higher amount than the other samples. These results are evident in the TLC profile shown in Fig. 7.

TABLE 4. Asiaticoside concentrations of the Centella asiatica samples obtained from nine locations

Sample codes	Sample origins	Asiaticoside concentration (mg/g)
a	Tawangmangu	0.5393±0.1172
b	Batu	1.0295±0.6675
c	Probolinggo	1.2104±0.3513
d	Magelang	3.2774±1.0630
e	Mataram	1.5700±0.6121
f	Lombok Barat	3.4687±0.9992
g	Lombok Timur	*
h	Bangli	*
i	Hulu Sungai Selatan	2.3390±1.2291

<sup>\*:</sup> asiaticoside concentrations are not quantifiable ( $\langle LOQ \rangle$ ; mean $\pm \overline{SD}$  (n = 3)



**FIGURE 7.** TLC chromatograms of the asiaticoside standard (st) and *Centella asiatica* samples from nine different locations (a-i), with anisaldehyde sulfuric acid as the derivatization reagent, detected in UV light at 365 nm (A) and visible light (B)

As seen in Table 4, the samples obtained from Lombok Timur and Bangli had minute asiaticoside contents (below the LOQ) that were not quantifiable. On the contrary, the other seven samples had varying concentration of asiaticoside, from 0.5393 to 3.4687 mg/g. Figure 8 shows that samples from Magelang, Lombok Barat, and Hulu Sungai Selatan had the highest asiaticoside content, while those from Batu, Probolinggo, and Mataram had the second-highest. The asiaticoside of the Tawangmangu sample was significantly lower than the *C. asiatica* collected from other regions (p<0.05), making it the smallest concentration detected in the study. However, although the samples from Tawangmangu, Lombok Timur, and Bangli had low asiaticoside, they contained other metabolites in a quite substantial or even higher amount than the other samples. These results are evident in the TLC profile shown in Fig. 7.

The study results proved that the asiaticoside concentration of the sampled *C. asiatica* are influenced by the plant's geographical location. However, whether the difference is due to the location's elevation or other factors remains unknown and, thus, requires further investigation. Therefore, before cultivating *C. asiatica*, it is necessary to consider the local geographical conditions so as to achieve harvests with a standard and consistent asiaticoside content.

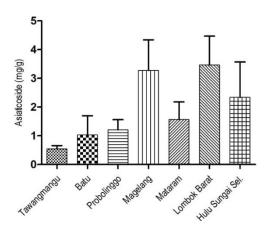


FIGURE 8. Asiaticoside concentrations of seven samples of Centella asiatica

#### **CONCLUSION**

The TLC-densitometry method designed is relatively simple but meets all the validation parameters. As a result, it can be used for analyzing asiaticoside in *Centella asiatica* both qualitatively and quantitatively. Furthermore, this study has found that the extracts of the *C. asiatica* collected from various locations and geographic characteristics in Indonesia contain different concentration of asiaticoside. In the future, the research can be developed to obtain data from unexplored regions in the country as well as other factors affected the different content of asiaticoside.

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#### ZnO's effect on Portland cement's antibacterial properties ₩

Randy Trafino; Muhammad Thoriq Albari; Muhammad Arya Ghifari; Muhammad Raffi Ghifari; Devi Purnamasari; Riso Sari Mandeli

AIP Conf. Proc. 3001, 030002 (2024) https://doi.org/10.1063/5.0184160



#### Effect of adding CuO as antibacterial to Portland cement ₩

Rida Oppi Ramadhani; Muhammad Thoriq Albari; Muhammad Arya Ghifari; Muhammad Raffi Ghifari; Devi Purnamasari; Riso Sari Mandeli

AIP Conf. Proc. 3001, 030003 (2024) https://doi.org/10.1063/5.0184162



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Wulandari Agustin; Muhammad Thoriq Albari; Muhammad Arya Ghifari; Muhammad Raffi Ghifari; Devi Purnamasari; Riso Sari Mandeli

AIP Conf. Proc. 3001, 030004 (2024) https://doi.org/10.1063/5.0184163



#### MgO's effect on wall paint's antibacterial properties ₩

Sintia Noveliza; Muhammad Thoriq Albari; Muhammad Arya Ghifari; Muhammad Raffi Ghifari; Devi Purnamasari; Riso Sari Mandeli

AIP Conf. Proc. 3001, 030005 (2024) https://doi.org/10.1063/5.0184164





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The impact of the home environment and community behavior of dengue hemorrhagic fever (DHF) in Rimbo Tengah district, Bungo Regency in 2022 ☑
Naldi Candra; Eri Barlian; Abdul Razak; Linda Handayuni; Aidil Onasis; Cica Ramadani
AIP Conf. Proc. 3001, 030006 (2024) https://doi.org/10.1063/5.0184152
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Ongoing attempt on the incorporation of pyrazine as pillar in the construction of Zn(II)-tartrate polymeric complex for renewable porous materials ⋤
Yana Yunita; Rachmat Triandi Tjahjanto; Danar Purwonugroho; Yuniar Ponco Prananto AIP Conf. Proc. 3001, 030007 (2024) https://doi.org/10.1063/5.0184577
Abstract ∨ View article
Simple TLC-densitometric method for the quantification of asiaticoside in Centella asiatica from different origins for its standardization ≒
K. Kartini; A. Rosidah; R. Budiono
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Abstract ✓ View article
Jawa, Indonesia ☐   M. Afiyanti; E. L. Arumingtyas; R. Azrianingsih; S. Indriyani   AIP Conf. Proc. 3001, 030009 (2024) https://doi.org/10.1063/5.0184949   Abstract ✓ View article ☐ PDF
Study of <i>Phytoene synthase 1 (PSY1)</i> and <i>capsanthin-capsorubin synthase (CCS)</i> gene profile in chili peppers ( <i>Capsicum frutescens</i> L.)  Cakra Hijau, G1/M8 line, and HV-149 🖫
A. S. Zakiyah; D. Siswanto; E. L. Arumingtyas  AIP Conf. Proc. 3001, 030010 (2024) https://doi.org/10.1063/5.0189450
Abstract ✓ View article D PDF
The effect of auxin 2,4-dichlorophenoxyacetic acid and explant type on red spinach ( <i>Amaranthus gangeticus</i> . Sp) callus induction ≒
Alif Rofiqotun Nurul Alimah; Estri Laras Arumingtyas; Retno Mastuti  AIP Conf. Proc. 3001, 030011 (2024) https://doi.org/10.1063/5.0185965
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Mycelium-based leather potential for sustainability: A review ₩
N. R. Ariyani; S. P. Wijayanti; N. G. Putra; M. B. Kusumawati; A. A. R. Setiawan; F. Isharyadi; N. Widyastuti; I. N. Djarot; T. Handayani  AIP Conf. Proc. 3001, 030012 (2024) https://doi.org/10.1063/5.0184187
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The potential use of *Arbuscular mycorrhizal Fungi* (AMF) to improve cocoa seedlings growth performance under environmental stress

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Propagation of Habenaria beccarii Schltr. by in vitro culture in purwodadi botanic garden ₩ E. E. Ariyanti; D. Mudiana; N. D. Yulia AIP Conf. Proc. 3001, 030021 (2024) https://doi.org/10.1063/5.0183879 Abstract ✓ View article PDF The fish diversity in code river, Bantul Regency, Yogyakarta special region ₩ A. Budiantoro; L. R. Sari; N. Suwartiningsih AIP Conf. Proc. 3001, 030022 (2024) https://doi.org/10.1063/5.0189449 Abstract ∨ View article 🔁 PDF Screening of mung bean genotypes for their resistance against legume pod borer, Maruca testulalis ≒ N. Nuryati; T. Trustinah; M. S. Y. I. Bayu AIP Conf. Proc. 3001, 030023 (2024) https://doi.org/10.1063/5.0184828 Abstract ✓ View article 🖪 PDF Ecosystem assessment of several springs in UB forest (UBF) based on phytoplankton and vegetation community ₩ V. Vidayanti; C. Retnaningdyah; E. Arisoesilaningsih AIP Conf. Proc. 3001, 030024 (2024) https://doi.org/10.1063/5.0184966 Abstract ✓ View article 🛚 PDF Studies on the yield rate, nutrient composition, and simple bulk handling methods to maintain the nutritional value of stored rice straw \ Khalil; Roni Pazla; Andri; Hermon AIP Conf. Proc. 3001, 030025 (2024) https://doi.org/10.1063/5.0183885 Abstract ∨ 🖪 PDF View article Relationship between geographical factors and feed carrying capacity with the existence of the endemic butterfly Ornithoptera croesus on Bacan Island ₩ A. Mas'ud; A. R. Tolangara; B. K. Lahati; Sundari AIP Conf. Proc. 3001, 030026 (2024) https://doi.org/10.1063/5.0198958 Abstract ∨ PDF View article Structure and carbon stock of vegetation in the arboretum area of the Purwodadi Botanic Garden ₩ F. Fauziah; Muhammad Fuad Fajrushobah; Deva Eka Ramadhani; Titut Yulistyarini; Abban Putri Fiqa AIP Conf. Proc. 3001, 030027 (2024) https://doi.org/10.1063/5.0184039

Isolation and characterization of *Salmonella* typhimurium lytic bacteriophages from fermented shrimp paste (*Terasi*) ≒

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A. Gaffar; Y. D. Jatmiko; A. A. Prihanto

Abstract ✓

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Orchid diversity in Kayan Mentarang National Park, North Kalimantan   A. R. U. Wibowo; R. K. Wati; G. A. E. Oktavia; D. Lestari; I. K. Toya; I. G. Tirta  AIP Conf. Proc. 3001, 030029 (2024) https://doi.org/10.1063/5.0184101
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Preliminary investigation of buried caves in Alas Kasinan, Pesanggrahan village, East Java based on Georadar ⋤
S. Maryanto; G. Al-Atqiya; R. P. H. Sari
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Efforts for selection of the <i>Lenggek</i> crowing number of Kokok Balenggek chicken based on qualitative phenotypic characteristics ₩
F. Arlina; Rusfidra; Y. Safitri; T. Rafian  AIP Conf. Proc. 3001, 030031 (2024) https://doi.org/10.1063/5.0185980
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potential pan-PI3K inhibitor ♀  A. H. Ahkam; N. M. Maghfiroh; F. N. Sari; M. Rifa'i  AIP Conf. Proc. 3001, 030032 (2024) https://doi.org/10.1063/5.0189093  Abstract ∨ View article
Comparative morphology of cerithioidean radula (Mollusca: Gastropoda) observed using scanning electron microscope ♀
Y. Apriyanti; N. R. Isnaningsih
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Abstract ✓ View article
Effect of <i>Trichoderma asperellum</i> application on growth and total phenolic content in soybean seedlings ₩  E. Yusnawan; E. Uge; A. Inayati  AIP Conf. Proc. 3001, 030034 (2024) https://doi.org/10.1063/5.0184209  Abstract ∨ View article
Comprehensive application to grow <i>Calliandra calothyrsus</i> by using cow dung biochar and solid waste from silica sand purification on critical mining land   Bernadetha Susianti; I. D. A. A. Warmadewanthi; Bieby Voijant Tangahu  AIP Conf. Proc. 3001, 030035 (2024) https://doi.org/10.1063/5.0194966
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Interaction of epiphyte *Polysiphonia sp* with *Kappaphycus alvarezii* host based on anatomical structure and phytochemical profile 🖫

Morphological characterization of *Phalaenopsis* spp. for the servation

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Sahyadrassus malabaricus (Moore) stem borer attacks on <i>Trema orientalis</i> L. Blume plant ∖ U. W. Darmawan; Y. Lisnawati  AIP Conf. Proc. 3001, 030052 (2024) https://doi.org/10.1063/5.0189417
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Leaf sheaths micromorphology of four <i>Glomera</i> species from new Guinea ₩
R. K. Wati; I. A. Fijridiyanto
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Firmansyah Rangga; Nazlina Shaari; Sumarni Ismail; Nensi Golda Yuli; Dinda Salmarisha AIP Conf. Proc. 3001, 040001 (2024) https://doi.org/10.1063/5.0184126
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I ne impact of the coal stockpile plan at Loktuan Port - Bontang on the environment and public health levels \ M. Musharyanto; Eri Barlian; Heldi; Mulya Gusman AIP Conf. Proc. 3001, 080005 (2024) https://doi.org/10.1063/5.0185867 View article 🔁 PDF Abstract ✓ Comparison of the environmental impacts of aquaponics and hydroponics using the LCA method in Bogor, Indonesia ₩ Fikri Dinegoro; Djoko M. Hartono; Dwica Wulandari; Dwi Ajeng Sarasputri; Dasi Agung Ospaman AIP Conf. Proc. 3001, 080006 (2024) https://doi.org/10.1063/5.0184149 Abstract ✓ View article PDF Study of carrying capacity and capacity of Lawai river and Kiahan river, Muara Enim Regency ₩ Yulkar Pramilus; Eri Barlian; Iswandi Umar AIP Conf. Proc. 3001, 080007 (2024) https://doi.org/10.1063/5.0184142 Abstract ∨ View article PDF Evaluation of the water environment quality of several rivers in Lahat Regency based on plankton composition \₩ Andri Wijaya Kesuma; Muhammad Agus; Rahman Arif; Raja Muhammad Faisal; F. Firmansvah AIP Conf. Proc. 3001, 080008 (2024) https://doi.org/10.1063/5.0185864 Abstract ∨ View article PDF The role of extrinsic and intrinsic factors on successful tuberculosis treatment that impacts on sustainable development in Bukittinggi A. Athosra; M. Maisyarah; Indang Dewata; Iswandi Umar AIP Conf. Proc. 3001, 080009 (2024) https://doi.org/10.1063/5.0184141 Abstract ∨ View article 🖪 PDF The analysis management and potential cultural tourism objects (case study of Makam Syuhada Lapan cultural tourism object in Bireuen distric, Aceh Province, Indonesia) ₩ M. Jamil Husen; Rahmi Novalita; Sri Wahyuni AIP Conf. Proc. 3001, 080010 (2024) https://doi.org/10.1063/5.0184168 Abstract ∨ View article 🛚 PDF Health environmental policy formulation in overcoming the impact of forest and land fires in the city of Payakumbuh, West Sumatra ₩ Linda Handayuni; Ririn Afrima Yenni; Eri Barlian; Abdul Razak; Ratna Wilis AIP Conf. Proc. 3001, 080011 (2024) https://doi.org/10.1063/5.0184958 Abstract ✓ View article PDF

Sustainability analysis of Rumah Gadang in Nagari Tuo Pariangan, Tanah Datar Regency using multidimensional scaling (MDS)

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Developing strategies to promote Pantai Lango "the boat craftsmanship" village as a destination of Balikpapan Bay integrated ecotourism ₩

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Support vector machine classification datamining on the predicted value of construction completed by construction companies  $\Xi$ 

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Mapping evaluation using data mining in the number of villages with sports fields  $\mbox{\ensuremath{\square}}$ 

Alex Wenda; Heri Purwanto; Muhammad Najamuddin Dwi Miharja; Ratna Puspita Sari; Riesta Ayu Oktarina

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