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Antiplasmodial and Antioxidant Activity of *Garcinia Bancana* Extract

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Abstract: Malaria caused by Plasmodium parasites is a significant public health issue, particularly in tropical and subtropical regions. There is also resistance to chloroquine-based therapy, which highlights the need for novel therapeutic agents. Therefore, our project in exploring antiplasmodial agents from *Garcinia* Indonesia continues. This study evaluates the phytochemical content of *G. bancana* by the total phenolic content (TPC) and total flavonoid content (TFC) and its *in vitro* antioxidant and antiplasmodial activities. The TPC and TFC values were determined using a UV-VIS spectrophotometer, while the antioxidant activity was determined using the DPPH, ABTS, and FRAP assays. Antiplasmodial activity against a chloroquine-sensitive strain 3D7 was evaluated using the Giemsa staining method. The highest TPC value of 195.75 ± 1.24 mg GAE/g was obtained from methanolic extract, while a TFC value of 82.79 ± 0.34 mg QE/g extract was found from dichloromethane extract. The methanolic extract exhibited the most potent antioxidant activity in the DPPH and FRAP assays with IC_{50} values of 6.07 ± 0.06 μg/ml and 74.35 ± 3.77 μM Fe^{2+}/g, respectively. The *n*-hexane extract was found to be the most potent on ABTS antioxidant and antiplasmodial assays with IC_{50} values of 1.22 ± 0.02 μg/ml and 0.23 ± 0.01 μg/ml, respectively. Furthermore, the DPPH antioxidant was negatively correlated with antiplasmodial significantly at 0.05. These findings suggest that the *n*-hexane extract of *G. bancana* has great potential as a source of antioxidant and antiplasmodial compounds. To the best of our knowledge, this study provides microscopic evidence in addition to the strongest antiplasmodial efficacy of *Garcinia* extract.

Keywords: *Garcinia bancana*, extract, antioxidant activity, antiplasmodial activity.

藤黄提取物的抗疟原虫和抗氧化活性

摘要：疟原虫寄生虫引起的疟疾是一个重大的公共卫生问题，特别是在热带和亚热带地区。对基于氯喹的疗法也存在耐药性，这凸显了对新型治疗药物的需求。因此，我们继续探索从印度尼西亚藤黄中提取抗疟原虫药物的项目。本研究通过总酚含量(台积电)和总黄酮含量(三氟化碳)及其体外抗氧化和抗疟原虫活性来评估班卡纳的植物化学成分含量。台积电和三氟化碳值是使用紫外-可见分光光度计测定的，而抗氧化活性是使用 DPPH、ABTS 和 FRAP 测定法测定的。使用吉姆萨染色法评估了针对氯喹敏感菌株 3 丁 7 的抗疟原虫活性。从甲醇提取物中获得了 195.75 ± 1.24 毫克通用电气工程师协会/克的最高台积电值，而从二氯甲烷提取物中发现了 82.79 ± 0.34 毫克量子效率/克提取物的三氟化碳值。甲醇提取物在

DPPH 和 FRAP 测定中表现出最强的抗氧化活性，我知道了 50 值分别为 6.07 ± 0.06 微克/毫升和 74.35 ± 3.77 μM Fe2+/克。发现正己烷提取物对 ABTS 抗氧化剂和抗疟原虫测定最有效，我知道了 50 值分别为 1.22 ± 0.02 微克/毫升和 0.23 ± 0.01 微克/毫升。此外，DPPH 抗氧化剂与抗疟原虫呈显着负相关，为 0.05。这些发现表明，班卡纳的正己烷提取物具有作为抗氧化剂和抗疟原虫化合物来源的巨大潜力。据我们所知，除了藤黄提取物最强的抗疟原虫功效外，这项研究还提供了微观证据。

关键词：藤黄，提取物，抗氧化活性，抗疟原虫活性。

1. Introduction

Despite various efforts to control and treat malaria, the disease remains a global issue, as evidenced by the increase in cases of deaths from 2019 to 2020 [1]. There is also resistance to the commercial antiplasmodial drugs that are currently in use [2]. Therefore, new antiplasmodial drugs are urgently needed. Natural products are a potential source of bioactive compounds because they have been used as a source of malaria drugs since their discovery [3]. The genus *Garcinia* contains phenolic compounds that have the potential as a source of antiplasmodial agents [4]. Dauphinols A, B, E, and F, and tocotrienol from *Garcinia dauphinensis* have good *in vitro* antiplasmodial activity against the Dd2 drug-resistant strain of *Plasmodium falciparum* with IC_{50} values ranging from 0.8 to 8.3 μM [5]. (+)-catechin from *G. celebica* also exhibited antiplasmodial activities against *P. falciparum* strain DD2 with an IC_{50} of 198 μM [6]. Furthermore, isoxanthochymol isolated from *G. celebica* showed activity against *the P. falciparum* 3D7 strain with an IC_{50} of 2.99 ± 0.20 μM [7]. A Phytochemical study of *G. forbesii* yielded 12b-hydroxy-des-D-garcigerrin A and subelliptenone H, which showed antiplasmodial activity against the 3D7 line of *P. falciparum* with IC_{50} values of 3.3 ± 0.04 and 5.0 ± 0.04 μM, respectively [8]. Wairata et al. [9] also reported good activity from the *n*-hexane, ethyl acetate, and methanol extract of *G. forbesii* against *P. falciparum* strain 3D7 with IC_{50} values ranging from 0.23 to 1.11 μg/ml. Additionally, an ethyl acetate extract of the stem bark of *G. husor* exhibited in vitro antiplasmodial activity against *the P. falciparum* 3D7 strain with an IC_{50} value of 0.31 ± 0.43 μg/ml. It also exhibited *in vivo* antiplasmodial activity against *P. berghei* with parasitemia suppression of 87.57 ± 1.41% compared to the negative control [10]. This study indicates that the genus *Garcinia* has great potential as antiplasmodial agents.

Garcinia bancana is endemic to peat forests in Southern Thailand, Malaysia, and Indonesia. The plant is locally called "Manggis hutan" and produces a bitter fruit with a dull orange-yellow color when ripe [11]. The leaves of *G. bancana* were traditionally used to treat fever. Furthermore, the Methanol extract of the stem and leaves of *G. bancana* showed moderate activity against H460 and MCF-7 cancer cells with IC_{50} values of 45 ± 5 μg/mL and 42 ± 4 μg/mL, respectively [12]. A phytochemical study of this plant revealed xanthone, polyprenylated benzophenones, flavonoid, coumarin, biphenyl, and terpenoid [13-14]. The isolated compound from this plant showed anti-inflammatory and immunoregulatory activities [15] and antimicrobial activity against methicillin-resistant *Staphylococcus aureus* [14].

Despite its traditional use as a fever remedy, there have been no reports of *G. bancana* having antimalarial activity. Therefore, based on ethnobotanical, *G. bancana* is selected as the research object. Additionally, *Garcinia* also possesses antiplasmodial activity. Here, the TPC and TFC contents of *n*-hexane, dichloromethane, ethyl acetate, and methanol extracts of *G. bancana* stem bark and their antioxidant (DPPH, ABTS, and FRAP) and antiplasmodial activities against *P. falciparum* chloroquine-sensitive strain 3D7 were reported. The correlation between TPC, TFC, antioxidant, and antiplasmodial activity was also studied.

2. Materials and Methods

2.1. General Experimental Procedures

The general experimental procedures for this study are shown in Fig. 1. The extraction solvents include *n*-hexane, dichloromethane, ethyl acetate, and methanol 99.9% and were obtained from Merck (Darmstadt, Germany) in analytical grade and used as received. Folin-Ciocalteu reagent, sodium carbonate solution, aluminum (III) chloride solution, 2,2”-azinobis (3-ethyl benzothiazoline-6-sulfonic acid) (ABTS), 1,1-diphenyl-2-picrylhydrazyl (DPPH), iron (III) chloride, 2,4,6-tris(2-pyridyl)-s-triazine (TPTZ), acetate buffer, gallic acid, and quercetin were purchased from Sigma (Sigma-Aldrich GmbH, Germany) and freshly prepared during the assays. Ethanol and dimethyl sulfoxide (DMSO) were also used. A Genesys UV-Vis

spectrophotometer (Thermo Fisher Scientific, Madison, WI, USA) was used for data collection from *in vitro* experiments. Furthermore, correlation studies were conducted using IBM SPSS Statistics Software Version 25.

2.2. Plant Material

The stem barks of *G. bancana* were collected from Tumbang Nusa peat forest, Central Borneo, Indonesia, at coordinates of 2°21'32.7"S, 114°05'05.6"E. The plant determination was conducted by the Herbarium Bogoriense, Research Center for Biology, Cibinong Science Center, Cibinong, Indonesia, with specimen voucher No. 2377.

2.3. Preparation of *G. Bancana* Extracts

The air-dried stem barks of *G. bancana* were ground into powder by the milling machine and subjected to extraction by maceration. About 10 g of stem bark powder was soaked in 100 mL of *n*-hexane, dichloromethane, ethyl acetate, and methanol, respectively, for 24 h. The extracts were then filtered and evaporated under reduced pressure using a rotatory evaporator (Büchi, Flawil, Switzerland) to obtain solid residues of extracts.

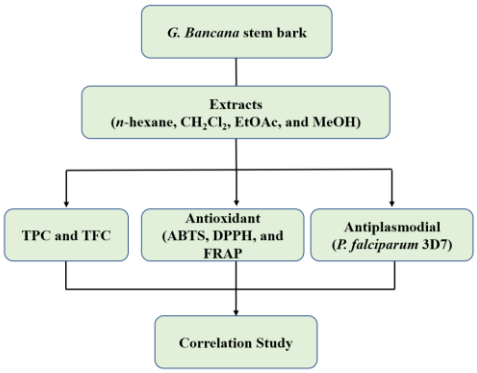

Fig. 1 Schema of the general experimental procedures (The data were developed by the authors)

2.4. Total Phenolic Content (TPC)

The TPC in *G. bancana* stem bark extracts was determined based on a method developed by Hossain et al. [16] with slight modification. About 0.5 mL of each extract at 1000 ppm in methanol was stirred with 2.5 mL of 10% Folin-Ciocalteu and left for five minutes. Subsequently, 2.0 mL of 7.5% Na_2CO_3 aqueous solution was added into the mixture. The mixture was then incubated at 40°C for one hour, and the absorbance was measured at 765 nm. Gallic acid was used as the standard for standard curve preparation at concentrations ranging from 0 to 200 mg/L. The TPC value of the extracts was reported as a gallic acid equivalent (mg GAE/g).

2.5. Total Flavonoid Content (TFC)

TFC was evaluated using a previously reported method with slight modifications [17]. A mixture of 0.5 mL of prepared extracts (100 ppm in methanol) and 0.5 mL 2% $AlCl_3$ (in methanol) was incubated at room temperature for one hour. The absorbance was then measured at 415 nm. Furthermore, the standard curve was made from quercetin at concentrations ranging from 0 to 50 mg/L in methanol. The TFC value was expressed as a quercetin equivalent (mg QE/g).

2.6. In Vitro Antioxidant Activities

2.6.1. ABTS Assay

The ABTS free radical activity of each extract was assayed according to a method by Jalloul et al. [18] with minor modifications, using gallic acid and quercetin as positive controls. The ABTS solution was prepared by combining 5 mL of 7 mM ABTS with 88 µL of 140 mM $K_2S_2O_6$ solution and incubating in the dark for 12 to 16 hours. Ethanol was used to dilute the ABTS solution, yielding an absorbance of 0.7 ± 0.02 at 734 nm. A series of samples was prepared, with concentrations of 99, 49.5, 24.75, 12.38, 6.12, and 3.10 µg/mL. The assay was conducted by mixing 10 µL of the sample with 1 mL ABTS solution and incubating for 4 minutes. The absorbance was measured at 734 nm with ethanol as the blank. Furthermore, the ability of the extract to scavenge the ABTS free radicals was calculated by the following equation:

$$\%\ Inhibition = \frac{Ab-As}{Ab} x100\% \quad (1)$$

where Ab - absorbance of the blank, As - absorbance of the sample.

2.6.2. DPPH Scavenging Assay

The DPPH extract activity was measured using the Brand-Williams method with minor modifications [19]. Quercetin and gallic acid were used as positive controls. Furthermore, 33 µL of the sample prepared in concentrations of 159.73, 79.86, 39.93, 19.97, and 9.98 µg/mL, respectively, were added to 1 mL of 6 x 10^{-5} M DPPH in methanol and incubated at 37°C for 20 minutes. The absorbance was measured at 517 nm with methanol as the blank, and the percentage inhibition was calculated by Equation 1.

2.6.3. Ferric Reducing-Antioxidant Power (FRAP) Assay

The FRAP assay was conducted as reported by Benzie et al. [20] with minor modifications. This assay measured the absorbance of strongly blue-colored ferrous complex Fe^{2+}-tripyridyltriazine before it was reduced to the colorless ferric complex Fe^{3+}-tripyridyltriazine at 593 nm. The FRAP reagents were prepared by mixing 20 mM $FeCl_3 \cdot 6H_2O$, 10 mM TPTZ solution in 40 mM HCl, and 300 mM acetate buffer of pH 3.6 in a 1:1:10 ratio. The assay was carried out by mixing 100 µL of the sample, 900 µL of distilled water, and 2 mL of FRAP reagent and then incubating for 30 minutes at 37°C in a dark room. The blank was

prepared from 2 mL of FRAP and 1 mL of water. The FRAP values were expressed as Fe^{2+}/g sample and calculated by FRAP Value of sample (μM) = abs (sample) × FRAP value of std (μM)/abs (std).

2.7. In Vitro Antiplasmodial Activity

The *in vitro* antiplasmodial activity against *Plasmodium falciparum* chloroquine-sensitive strain 3D7 was determined by Giemsa staining as previously reported [9]. The samples were prepared by dissolving 10 mg of the extract in 1000 μL DMSO. A serial dilution (1000, 100, 10, 1, 0.1, and 0.01 μg/ml) was made to determine the IC_{50} value (the half-maximum inhibitory concentration). The parasites used in this assay were synchronized (ring stage) with ± 1% parasitemia (5% hematocrit). The assay was performed by the addition of 2 μL of the sample to 198 μL of the parasite in 96 well plates and incubated at 37°C for 48 h in a 5% O_2, 5% CO_2, and 90% N_2 atmosphere. Furthermore, the culture was harvested, and the blood layer was thinned with 20% Giemsa staining. The number of infected erythrocytes per 1000 erythrocytes was counted under a microscope to determine the value in the blood smear. The growth percentages were calculated by % parasitemia – D0 (D0 = % growth at 0 h), and the percentage of inhibition was calculated using Equation 2.

$$\%\ Inhibition = 100\% - (\frac{Xu}{Xk} x 100\%) \quad (2)$$

where Xu - % growth of the test solution, Xk - % growth of the negative control.

Based on the inhibition data, statistical analysis was carried out using the probit analysis of the SPSS Version 25 program to determine the IC_{50} value or the concentration of the samples that inhibit parasite growth by 50%.

2.8. Statistical Analysis

The experimental test was carried out in triplicate, and the data from each experiment were presented with a mean and standard deviation, and analyzed by ANOVA. The IC_{50} values of DPPH and ABTS were calculated using a linear regression equation, and antiplasmodial activity was determined with probit analysis. The correlation of TPC, TFC, antioxidant, and antiplasmodial activities was studied using IBM SPSS Statistics (Version 25).

3. Results

3.1. TPC and TFC

The TPC and TFC values of *G. bancana* extracts are shown in Table 1. The methanol extract showed the highest TPC value of 195.75 ± 1.24 mg GAE/g extract, while dichloromethane extract exhibited the highest TFC value of 82.79 ± 0.34 mg QE/g extract.

Table 1 TPC and TFC values of *G. bancana* extracts (The data were developed by the authors)

Extracts	TPC (mg GAE/g extract)	TFC (mg QE/g extract)
n-hexane	160.73 ± 0.42	59.74 ± 0.12
CH_2Cl_2	126.71 ± 0.55	82.79 ± 0.34
EtOAc	174.78 ± 0.68	42.33 ± 0.17
MeOH	195.75 ± 1.24	32.05 ± 0.23

3.2. Antioxidant Activities

Table 2 shows the antioxidant activities of *G. bancana* extracts as evaluated by DPPH, ABTS, and FRAP assays. The highest DPPH and FRAP IC_{50} values of 6.07 ± 0.06 μg/ml and 74.35 ± 3.77 μM Fe^{2+}, respectively, were shown by methanol extract, while the *n*-hexane extract had the highest IC_{50} value of 1.22 ± 0.02 μg/ml for the ABTS assay.

Table 2 Antioxidant activities of *G. bancana* extracts (The data were developed by the authors)

Extracts	ABTS IC_{50} (μg/mL)	DPPH IC_{50} (μg/mL)	FRAP (μM Fe^{2+})
n-hexane	1.22 ± 0.02	23.31 ± 0.12	37.43 ± 1.37
CH_2Cl_2	4.30 ± 0.04	20.70 ± 0.31	24.35 ± 0.73
EtOAc	5.55 ± 0.03	24.00 ± 0.37	40.99 ± 0.98
MeOH	1.65 ± 0.02	6.07 ± 0.06	74.35 ± 3.77
Gallic acid	0.12 ± 0.01	0.55 ± 0.01	Nt
Quercetin	0.06 ± 0.00	1.34 ± 0.02	Nt
Ascorbic acid	Nt	Nt	30.62 ± 0.27

3.3. Antiplasmodial Activity

As shown in Table 3, the *n*-hexane extract displayed the most potent activity with an IC_{50} of 0.23 ± 0.01 μg/mL, followed by dichloromethane, ethyl acetate, and methanol extracts with IC_{50} values of 0.26 ± 0.01, 0.30 ± 0.02, and 3.36 ± 0.03 μg/mL, respectively.

At 100 μg/mL (Fig. 2), all extracts showed good antiplasmodial activity with a 100% inhibition of *Pf*3D7. Fig. 3 shows that there are no ring-shaped trophozoites of *Pf*3D7 on *n*-hexane, dichloromethane, ethyl acetate, or methanol extracts at 100 μg/mL, indicating a 100% inhibition. The ring stage of 3D7 trophozoites was observed at 1 μg/mL on *n*-hexane, dichloromethane, ethyl acetate, and methanol extracts, and inhibited in the range of 38.19 to 62.70%, as shown in Fig. 2. Furthermore, the ethyl acetate extract exhibited a consistent 100% inhibition until 10 μg/mL, with the highest inhibition at 1 μg/mL being 62.7%. The *n*-hexane extract showed higher activity at lower concentrations under 1 μg/mL and the highest antiplasmodial activity at 0.01 μg/mL, with a 29.6% inhibition.

Table 3 Antiplasmodial activity of *G. bancana* extracts (The data were developed by the authors)

Extracts	IC_{50} (µg/mL)
n-hexane	0.23 ± 0.01
CH_2Cl_2	0.26 ± 0.01
EtOAc	0.30 ± 0.02
MeOH	3.36 ± 0.03
Chloroquine	0.01 ± 0.01

(a) (b)

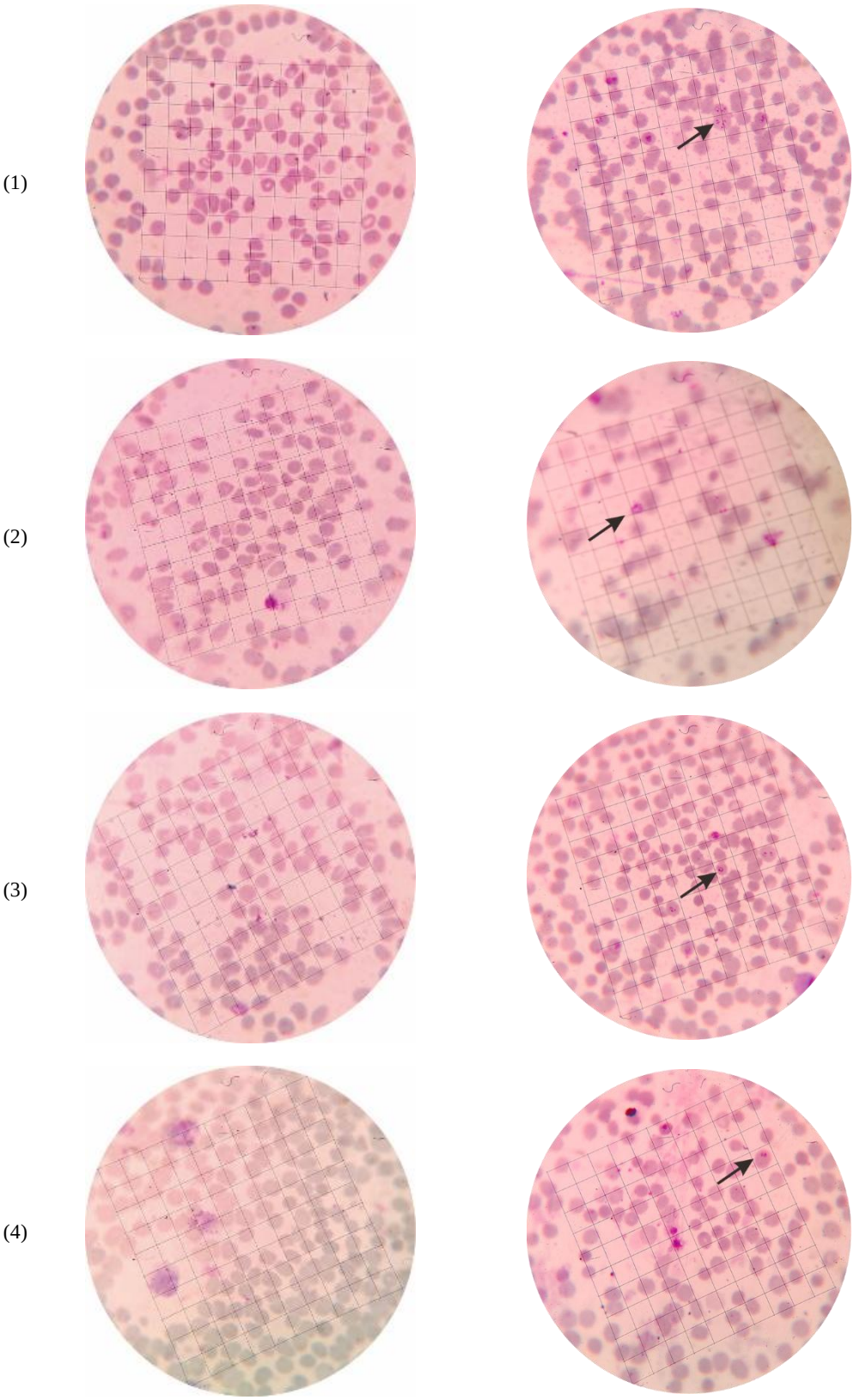

Fig. 3 Microscope evidence of blood smear with GIEMSA staining for *n*-hexane (1), dichloromethane (2), ethyl acetate (3), and methanol (4) extracts at 100 µg/mL (a) and 1 µg/mL (b) (The data were developed by the authors)

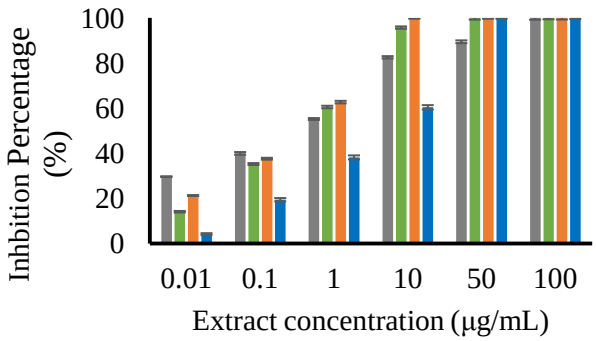

Fig. 2 Inhibition percentage of antiplasmodial activity of *n*-hexane (gray), dichloromethane (green), ethyl acetate (orange), and methanol extracts (blue) from the stem bark of *G. bancana* (The data were developed by the authors)

3.4. Correlation Studies

Pearson's correlation studies of TPC, TFC, antioxidant, and antiplasmodial activities are shown in Table 4.

Table 4 Pearson's correlation between TPC, TFC, antioxidant, and antiplasmodial activity (The data were developed by the authors)

	TPC[1]	TFC[2]	ABTS[3]	DPPH[3]	FRAP[3]	*Pf*3D7[4]
TPC	1					
TFC	-0.991**	1				
ABTS	-0.322	0.194	1			
DPPH	-0.591	0.534	0.476	1		
FRAP	0.910	-0.871	-0.471	-0.872	1	
*Pf*3D7	0.721	-0.671	-0.473	-0.985*	0.942	1

* Correlation is significant at the 0.05 level
** Correlation is significant at the 0.01 level
[1] Total phenolic content
[2] Total flavonoid content
[3] Coefficient of antioxidant activity
[4] Coefficient of antiplasmodial activity

4. Discussion

4.1. Total Phenolic and Flavonoid Contents

The TPC and TFC values of *G. bancana* stem bark extracts were evaluated using a colorimetric method and a calibration curve of gallic acid and quercetin, respectively [21]. A gallic acid linear standard curve ($y = 0.0042x + 0.0234$, $R^2 = 0.9901$) revealed that methanol extract contained the highest value of TPC, followed by ethyl acetate, *n*-hexane, and dichloromethane extracts. These results indicate that the phenolic compounds in the stem barks of *G. bancana* were mostly polar. The highest TPC in methanol extract was also reported previously from the leaves of *G. madruno* [22]. Another study found that methanol extract had higher TPC than other solvents in *R. tomentosa* leaves [23]. Meanwhile, the TFC values of *G. bancana* extracts were justified using a linear standard curve of quercetin ($y = 0.0337x$, $R^2 = 0.9973$). The data revealed that the flavonoid compounds in the stem barks of *G. bancana* were mostly non-polar and semi-polar. The methanol extract with the highest TPC value but the lowest TFC indicates that flavonoid is not the most abundant phenolic compound in *G. bancana* stem bark. Generally, the TPC will be higher than the TFC because flavonoid belongs to the phenolic class. Previously isolated compounds from *G. bancana*, such as garcinol, guttiferone F, isogarcinol, xanthochymol, and 30-epi-cambogin indicate that the major compounds of *G. bancana* belong to the polyprenylated benzophenones (PPB). Additionally, metabolomic studies of the phytochemical profiles of *G. bancana* extracts with ^{13}C-NMR showed other PPBs such as symphone A, garcinielliptone FC, cycloxanthochymol, guttiferone E, and 13,14-dedioxyisogarcinol [13-14].

4.2. Antioxidant Activities

The DPPH and ABTS assays are well-known methods for evaluating the scavenging antioxidant activity of natural product extracts [24]. The DPPH method involves a hydrogen atom transfer reaction, while ABTS involves a single electron transfer during the antioxidant activity evaluation. The single electron transfer is preferred due to the presence of ethanol and methanol in the antioxidant system [25]. Furthermore, the antioxidant evaluation using the ABTS assay exhibited a stronger activity than the DPPH assay. This is presumably due to the hydrophilic and lipophilic antioxidant systems of ABTS, while DPHH was limited to hydrophobic systems [26].

The stronger antioxidant activity in the ABTS assay than DPPH has also been reported in studies of *Garcinia subelliptica* branch extract [27], stem bark extract of *Dipterocarpus littoralis* [19], and date seed extract of *Phoenix dactylifera* L. [28].

The antioxidant mechanism in the FRAP assay was based on a single electron transfer in a redox reaction. The Fe^{3+}-TPTZ was reduced to blue-colored Fe^{2+}-TPTZ by an electron donated from the antioxidant agents [20]. The *G. bancana* extracts, excluding the dichloromethane extract, presented higher FRAP values than the positive control ascorbic acid. Furthermore, the methanol extract has more than double the FRAP value of ascorbic acid, and this value is proportional to the TPC. This result indicated that PPBs and the other phenolic compounds were responsible for its reducing power. Based on a previously reported study of the FRAP activity of PBBs from *G. celebica*, the hydroxyl substituent of PPBs possibly acts as an electron donor to the reactive oxygen species [7]. *G. bancana* extracts have higher antioxidant activity compared to *G. celebica, G. forbesii, and G. subelliptica* extracts [7, 9, 27]. This result signified the potential of *G. bancana* as an

antioxidant source.

4.3. Antiplasmodial Activity

As shown in Table 3, the extracts of *G. bancana* have potent antiplasmodial activity (IC_{50} < 5 μg/mL) based on the classification of the crude extract described earlier [29]. The potential activity of *G. bancana* extracts may be due to their high TPC. Isogarcinol and garcinol from *G. bancana* have activity against *P. falciparum* strain FcB1 with IC_{50} values of 3.5 ± 1.1 μM and 12.6 ± 4.8 μM, respectively [30]. Xanthones have also been predicted to contribute to antiplasmodial activity [31]. Furthermore, the antiplasmodial activity of the *n*-hexane extract of *G. bancana* is more potent than the ethyl acetate extract of *G. mangostana* (IC_{50} 0.42 μg/ml), ethyl acetate extract of *G. husor* (IC_{50} 0.31 μg/ml), and comparable with the *n*-hexane extract of *G. forbesii* (IC_{50} 0.23 μg/ml) [9, 10, 32]. This implies the potential of *G. bancana* extract as a source of antiplasmodial drugs.

4.4. Correlation Studies

Pearson correlations were broadly used for correlating natural products and their bioactivity [33]. As described in Table 4, TPC and TFC were significantly inversely correlated ($r = -0.991$, $p < 0.01$), indicating that the higher the concentration of phenolic content in the extract, the lower the flavonoid content. TPC was positively correlated in the FRAP assay, which means that phenolic compounds were responsible for reducing the Fe^{3+} ion. Antioxidant DPPH was inversely correlated with antiplasmodial activity at the 0.05 level ($r = -0.985$), indicating that extracts with lower DPPH antioxidant activity have a more potent antiplasmodial activity. Subsequently, FRAP was positively correlated with antiplasmodial activity with $r = 0.942$, indicating that more potent FRAP will have a greater chance of being active in antiplasmodial assays.

5. Conclusions

5.1. Main Findings of This Study

Based on the results, *G. bancana* stem bark extract showed favorable antiplasmodial activity against *Pf*3D7 and antioxidant activity in DPPH, ABTS, and FRAP assays. The total phenolic and flavonoid contents of *G. bancana* were first reported in this study, and the methanolic extract of *G. bancana* showed the highest TPC value and the highest antioxidant activity in DPPH and FRAP. However, the *n*-hexane extract of *G. bancana* exhibited the most potent antiplasmodial and antioxidant activity in ABTS. The GIEMSA method used in this study has presented the real and effective antiplasmodial activities, based on the microscopic evidence. At the 0.05 level, there was a significant negative correlation between DPPH and antiplasmodial *P. falciparum* 3D7.

5.2. Comparison with Other Studies

This study exhibits the potential of antioxidant and antiplasmodial properties from *G. bancana* compared to other *Garcinia* species.

5.3. Implications of the Study

This study implicates the continuation of exploration of an antiplasmodial agent from the natural resource.

5.4. Strengths and Limitations

This study provides microscopic evidence besides its IC_{50} antiplasmodial activity of *Garcinia bancana* extract. However, the biological activity in this study is not yet supported by a complete phytochemical analysis.

5.5. Recommendations and Future Research

Phytochemical analysis of *n*-hexane extract of the *Garcinia bancana* stem bark through LCMSMS can serve as a future recommendation. Furthermore, the bioguided isolation can also be an option for future research on *n*-hexane extract, to discover the potent compound with antiplasmodial properties.

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