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# Antiplasmodial activity of chalcone derivatives compound through phagocytosis of kupffer cells in experimental malaria hosts



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Lilik Wijayanti<sup>1</sup>, Paramasari Dirgahayu<sup>2</sup>, Yulia Sari<sup>3</sup>, Danus Hermawan<sup>4</sup>, Ida Nurwati<sup>5</sup>

## ABSTRACT

**Introduction:** Malaria is still one of the major health problems, specifically due to drug resistance in Plasmodium, which encourages extensive research to find effective alternatives. One of the new antimalarial compounds is chalcone-derivative compound (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one. However, its potency is still needed to be evaluated. Therefore, this study aimed to determine the efficacy and identify the pharmacological mechanism of this chalcone-derivative.

**Methods:** An experiment using post-test only with control group design was conducted from May 2016 to July 2017 in the laboratory of Parasitology and Clinical Pathology Faculty of Medicine, Universitas Gadjah Mada. Swiss mice were used and infected with Plasmodium before being divided into nine groups with different concentrations of (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one. Histological examination was conducted

to count the number of Kupffer cells and the proportion of Kupffer cells that had phagocytosed the Plasmodium.

**Results:** Both doxycycline and (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one decreased the parasitemia in the tested mice with higher efficacy was observed in the doxycycline group. Likewise, both compounds also increase the number of Kupffer cells which become phagocytes of erythrocytes containing Plasmodium was higher in the treatment group than in the control group (p 0.000 < 0.05). Substance (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl) prop-2-en-1-one may act as an antimalarial by phagocytes of erythrocytes containing Plasmodium.

**Conclusion:** In conclusion, the (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one exhibited potent antimalarial activity via the phagocytic activity of Kupffer cells. This compound may be developed into a new antimalarial drug.

**Keywords:** Antiplasmodial, Chalcone, Kupffer Cells, Phagocytosis

**Cite this Article:** Wijayanti, L., Dirgahayu, P., Sari, Y., Hermawan, D., Nurwati, I. 2020. Antiplasmodial activity of chalcone derivatives compound through phagocytosis of kupffer cells in experimental malaria hosts. *Bali Medical Journal* 9(3): 912-917. DOI: [10.15562/bmj.v9i3.2021](https://doi.org/10.15562/bmj.v9i3.2021)

## INTRODUCTION

Malaria is a global infectious disease caused by the Plasmodium and transmitted by the Anopheles mosquito. Malaria still posed a considerable burden on global health, especially in Africa and Asiatic countries. In Indonesia, there was a decreasing trend of malaria morbidity in 2009-2016 from 1.8/1,000 at risk population in 2009 to 0.84/1,000 at risk population in 2016. However, some provinces in Indonesia still have a considerable burden from malaria. For example, Papua is one of the provinces with the highest Annual Parasite Incidence (API) with 45.85 per 1,000 inhabitants.<sup>1</sup> The main problem of malaria is not only because of its mortality and impact on national productivity but also due to the drug resistance found in *P. falciparum* and *P. vivax*. Furthermore, vaccine development is also hampered by the complex life cycle of *P. falciparum*.<sup>2,3</sup> This phenomenon has ignited extensive research to find an effective alternative of current antimalarials. The ideal antimalarial drugs should fulfill several criteria, including low toxicity and high efficacy in combating the major species of Plasmodium.

Fortunately, there have been several promising

compounds that had been evaluated for their antimalarial properties. Chalcones (1,3-diaryl-2-propen-1-ones) are secondary metabolites of flavonoids found in several plant species and have antimalarial activity.<sup>4,5</sup> Antimalarial activity of chalcones was investigated after a report on the research results stating potent antimalarial activity in vitro and in vivo from a compound, namely Licochalcone A, an isolated compound from the root of Chinese licorice.<sup>6,7</sup>

Suwito *et al.* had designed several chalcones derivatives as an inhibitor in ferredoxin (Fd) interaction with ferredoxin-NADP<sup>+</sup> reductase (NFR), which is a crucial redox system in *P. falciparum*'s survival.<sup>8</sup> The result indicated that one of the synthesized compounds (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl) prop-2-en-1-one had a remarkable inhibitory activity against Plasmodium through molecular docking. They also found that the amino group of the amino-methoxy derivative of chalcones played an essential role in inhibition through electrostatic interactions and can form a more stable complex with NFR compared to Fd. Likewise, an in vitro study found that (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy

<sup>1</sup>Department of Clinical Pathology, Study Program of Medicine, Faculty of Medicine, Universitas Sebelas Maret, Surakarta, Indonesia

<sup>2</sup>Department of Parasitology, Study Program of Medicine, Faculty of Medicine, Universitas Sebelas Maret, Surakarta, Indonesia

<sup>3</sup>Department of Parasitology, Study Program of Medicine, Faculty of Medicine, Universitas Sebelas Maret, Surakarta, Indonesia

<sup>4</sup>Department of Biochemistry, Study Program of Medicine, Faculty of Medicine, Universitas Sebelas Maret, Surakarta, Indonesia

<sup>5</sup>Department of Biochemistry, Study Program of Medicine, Faculty of Medicine, Universitas Sebelas Maret, Surakarta, Indonesia

\*Corresponding to:

Lilik Wijayanti; Department of Clinical Pathology, Study Program of Medicine, Faculty of Medicine, Universitas Sebelas Maret, Surakarta, Indonesia; [lilikwijayanti@staff.uns.ac.id](mailto:lilikwijayanti@staff.uns.ac.id)

Received: 2020-10-17  
Accepted: 2020-11-26  
Published: 2020-12-01

phenyl)prop-2-en-1-one had good antiplasmodial activity, selectivity index, and plasmodial growth-inhibiting effect.<sup>9</sup> Finally, the in vivo study also confirmed the efficacy of this substance, in which it was shown that the effective dose (ED50) for this substance was at 17.36 mg/kgBW/day.<sup>10</sup> Overall, the initial evidence showed that (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one has a potent antimalarial ability.

Other studies had also revealed that (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one can alter the formation of hemozoin and stomatocytes.<sup>11</sup> However, this compound's effect on host-immunity is not yet investigated, especially immunity mediated by Kupffer cells. On the other hand, it has been known that plasmodium infection can induce host immunity and one of the immune responses involved is mediated by Kupffer cells in the liver.<sup>12</sup> Therefore, this study was aimed to evaluate the effect of (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one toward Kupffer cells phagocytosis in the presence of plasmodium infection.

## MATERIALS AND METHODS

### Tested Compounds

The tested compound, (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl) prop-2-en-1-one, was synthesized by Dr. Hery Suwito from the Department of Chemistry, Faculty of Science and Technology, Universitas Airlangga, Surabaya. The chemical structure of this compound is depicted in Figure 1. Doxycycline was used as a positive control and obtained from Sigma Chemical Co. (St. Louis Mo.).

### In vivo antiplasmodial activity testing

The in vivo antiplasmodial activity was evaluated using Swiss Mice infected with *Plasmodium berghei* by the classical 4-day suppressive test. The research protocol had been approved by the Health Research Ethics Committee, Dr. Moewardi General Hospital/ Faculty of Medicine, Universitas Sebelas Maret, Surakarta. *P. berghei* strain was obtained from the

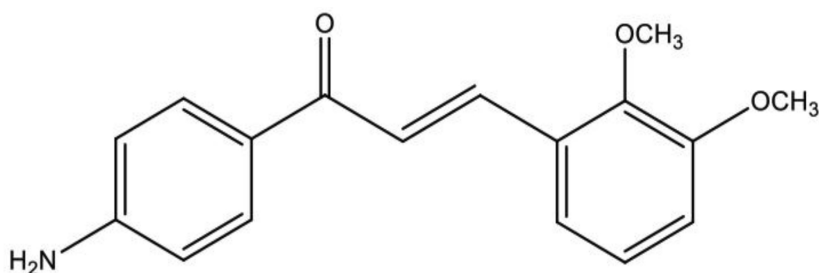
Department of Parasitology, Faculty of Medicine, Universitas Sebelas Maret, Surakarta. The Swiss mice were obtained from the Integrated Research and Testing Laboratory, Universitas Gadjah Mada, Yogyakarta. *P. berghei*-infected mice erythrocytes were obtained from donor mice and resuspended in RPMI 1640 medium to a volume of 0.2 ml a day before inoculation. Ninety male mice (20-25 g and 6-8 weeks) were inoculated intraperitoneally with  $10^7$  *P. berghei*-infected mice erythrocytes. The mice were then divided into nine groups, with ten mice in each group. The first four groups received 10, 20, 40, and 80 mg/kgBW/day of the tested compound. The second four groups were treated with 0.25, 0.5, 1, and 2 mg/kgBW of doxycycline as the positive control. Another group (negative control) was only received aquadest. Each dose of tested compound or doxycycline was given to the mice daily for four consecutive days, starting two hours after inoculation until the third day. A day after the last treatment, a Giemsa-stained thin blood smear from the tail vein was prepared. Parasitemia level was then determined microscopically by counting the number of parasitized erythrocytes out of 200 erythrocytes in the microscope's random fields.

### Histological preparations

After treatment, the mice were sacrificed on a determined day by neck dislocation. The liver organs were then taken for histological preparations using the paraffin block method and stained with hematoxylin and eosin (HE). The right lobe of the liver was taken and sliced at the middle of the lobe to get a uniform preparation. Three slices were made from each right lobe of the liver with a thickness of 3-8  $\mu$ m. The distance between slices with each other was about 25 slices. Three preparations were made from each experimental host and Kupffer cells were counted (400X magnification). Additionally, the number of Kupffer cells that had phagocytosed the infected erythrocytes was also calculated.

### Data analysis

The data were compiled and the study groups were compared by using the ANOVA test. Then, a post-hoc multiple comparisons test (Tamhane) was also performed to compare the groups against each other. Finally, the number of Kupffer cells were compared using Kruskal Wallis and Man Whitney statistical tests. The p-value  $\leq 0.05$  was considered significant.



**Figure 1:** Chemical structure of (E)-1-(4-aminophenyl)-3-(2,3-dimethoxyphenyl) prop-2-en-1-one

**Table 1.** Antiplasmodial activity of chalcone derivatives compared to doxycycline toward Plasmodium parasitemia

Tested compound			Doxycycline			P-Value*
Dose (mg/kgBW)	Parasitemia (%)	± SD	Dose (mg/kgBW)	Parasitemia (%)	± SD	
80	11.33	2.39	2	11.33	2.39	<0.05
40	55.50	7.09	1	9.67	2.33	
20	65.83	11.99	0.5	12.33	3.04	
10	138.17	7.39	0.25	15.67	3.61	
Aquadest	191.00	13.01	Aquadest	191.00	13.01	

\*P-Value of ANOVA; P-value was considered as significant at <0.05

**Table 2.** The effect of chalcone derivative on the average number of Kupffer cells

Tested compound			Doxycycline			P-Value*
Dose (mg/kgBW)	Average Kupffer Cells (%)	± SD	Dose (mg/kgBW)	Average Kupffer Cells (%)	± SD	
80	16.8	2.53	2	21.2	2.78	<0.05
40	15.4	2.32	1	17.7	2.11	
20	12.2	2.30	0.5	15.3	2.45	
10	6.3	1.57	0.25	10.2	2.57	
Aquadest	3.2	1.4	Aquadest	3.2	1.4	

Source: primary data (2019)

## RESULTS

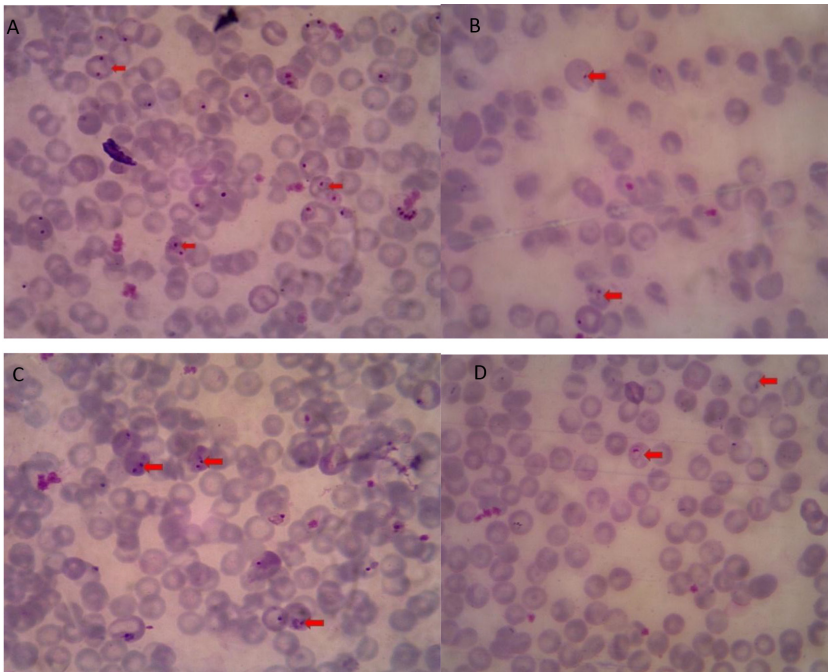
### Antiplasmodial activity of chalcone and doxycycline derivatives with parasitemia

According to the comparison between tested groups, it showed that parasitemia was decreased in both doxycycline and (E)1-(4 aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one. The parasitemia level in (E)1-(4 aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one was significantly lowered to less than 100 at the dose 20 mg/kgBW and approaching 10% at dose 80 mg/kgBW. However, doxycycline had a much higher efficacy against Plasmodium, which was reflected by the sharply decreased parasitemia pattern even from the dose of 0.25 mg/kgBW. The parasitemia level did not change much at higher tested dosage (0.5, 1, and 2 mg/kgBW). As depicted in Table 1, the tested compound has only matched the doxycycline at 80 mg/kgBW dose. The ANOVA test showed a significant result ( $p < 0.05$ ), which indicated that there were significant differences between (E)1-(4 aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one and doxycycline. Post-hoc analysis showed that (E)1-(4 aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one at dose 80 mg/kgBW had comparable effect to doxycycline at dose 0.5, 1, and 2 mg/kgBW. The visual comparison of the blood smear is presented in Figure 2.

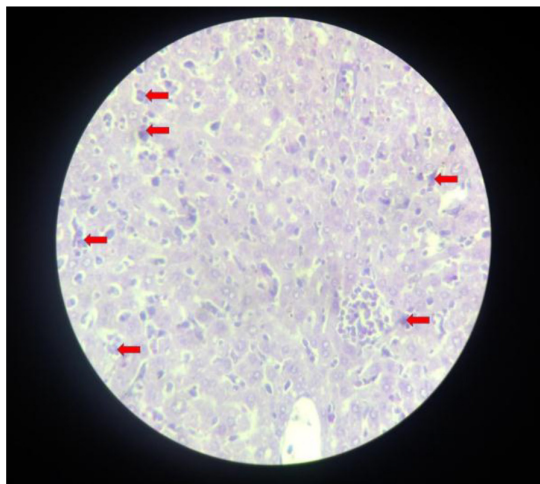
### The Relationship between the test substance and the number of Kupffer cells

After analyzing the effect of the chalcone derivative (E)1-(4 aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one toward parasitemia, we assessed the effect of the compound toward Kupffer cells number within the liver. As depicted in Table 2, the number of Kupffer cells was doubled in the mice, which received chalcone derivative at 10 mg/kgBW, compared to the negative control group (Aquadest). The Kupffer cells were doubled again at 20 mg/kgBW and begin to plateau at the higher dosage. Comparably, doxycycline was also effective in increasing the number of Kupffer cells and it seemed to have higher efficacy than the tested chalcone derivative. The number of Kupffer cells was tripled at dose 0.25 mg/kgBW and increased at a higher dose. The ANOVA showed a significant result, which indicated that there were significant differences between the study groups. However, in-depth analysis using a post-hoc test revealed that the chalcone derivative at dose 20 mg/kgBW had a comparable effect to 0.25 mg/kgBW doxycycline. Also, 80 mg/kgBW of chalcone derivative did not significantly differ from 40 mg/kgBW in increasing the number of Kupffer cells. 40 mg/kgBW of chalcone derivative was also had comparable effect with 0.5 and 1 mg/kgBW doxycycline while 2 mg/kgBW doxycycline proved to be superior to even 80





**Figure 2.** Erythrocytes infected with *P. berghei* in microscopic examination of thin blood smear preparations in the negative control group (A), chalcone derivatives at dose 80 mg/kgBW (B), chalcone derivatives at dose 10 mg/kgBW (C), and doxycycline at dose 2 mg/kgBW (D).



**Figure 3.** Kupffer cells that phagocytosed infected erythrocytes within the mouse liver.

mg/kgBW. The visual depiction of the Kupffer cells in the liver slice is presented in Figure 3.

## DISCUSSION

Malaria is a disease caused by plasmodium parasites and is transmitted by the *Anopheles* mosquito. Malaria can cause anemia, reduce productivity, and cause death, especially in groups with high-risk factors, namely infants, toddlers, and pregnant women. Many deaths occur in children in

Africa, and it is estimated that every minute one child dies of malaria. The main problem in the treatment or eradication of malaria to date is the increased resistance of *P. falciparum* and *P. vivax* to antimalarial drugs. Meanwhile, the development of malaria vaccines is hampered due to the complexity of *P. falciparum*'s life cycle, and therefore efforts need to be made to develop multistage vaccines.<sup>2</sup>

Drug resistance in Plasmodium is continuously reported from various parts of the world. Consequently, the World Health Organization (WHO) has instructed to stop the monotherapy protocol and recommended Artemisinin Combination Therapy (ACT) to improve treatment outcomes and reduce mortality. Meanwhile, rigorous research in drug discovery is continuously conducted to find new antimalarial agents as current agents' alternatives. The ideal antimalarial drug should meet several criteria in which one of which is to have mild side effects with low toxicity. Chalcones (1,3-diaryl-2-propen-1-ones) are secondary metabolites of flavonoids that can be found in several types of plants.<sup>13</sup> Chalcones and their derivatives are known to have various interesting biological activities such as antiviral, anti-inflammatory, antimicrobial, antitumor, cytotoxic, analgesic, antifungal, antioxidant, anticancer, and antiplasmodial.<sup>14,15</sup>

Following previous studies, this study found that chalcone derivative effectively reduced parasitemia and increased the number of stomatocytes-containing Kupffer cells. Compared to doxycycline, the tested compound was still inferior and only the highest dosage (80 mg/kgBW) was comparable to 0.5 mg/kgBW doxycycline. Likewise, 40 mg/kgBW of E)1-(4 aminophenyl)-3-(2,3-dimethoxy phenyl) prop-2-en-1-one was also comparable to 0.5 mg/kgBW doxycycline and 1 mg/kgBW doxycycline while the highest dose of the compound was comparable to 40 mg/kgBW. Nevertheless, we found a novel compound that has a promising prospect as a new alternative antimalarial agent.

Although the mechanism of action of chalcone was not part of this study, it had been proposed that chalcone and its derivatives act as an inhibitor of ferredoxin (Fd) and ferredoxin-NADP<sup>+</sup> reductase (NFR) interaction. The presence of inhibitors inhibits electron addition to Lyt B resulting in a disruption of the synthesis of isoprenoid and isoprenoid precursors.<sup>8</sup> Additionally, chalcone inhibits hemoglobin digestion by binding to and inhibiting falcipain, an enzyme that plays a role in hemoglobin digestion in Plasmodium's food vacuole.<sup>16,17</sup> The free heme, which is released as the byproduct of hemoglobin digestion, is usually aggregated into hemozoin by the Plasmodium to

avoid its toxic effect. Chalcone was also known to inhibit hemozoin synthesis and enhanced its anti-plasmodial effect through heme intoxication of the Plasmodium.<sup>18</sup> Also, the degradation of hemin that leaked out into the cytoplasm will inhibit chalcone through the inhibition of glutathione (GSH).<sup>18</sup>

Other reports also showed that chalcone attacks the bc1 complex and complex II (succinate ubiquinone reductase) of Plasmodium's mitochondria.<sup>19</sup> This effect will completely disrupt the electron transport chain, which is highly lethal for Plasmodium. Other effects reported include inhibition of Plasmodial development from ring form into a schizont form, inhibiting sorbitol-induced erythrocytes hemolysis infected by parasites, altering the ultrastructure of parasitic mitochondria and inhibiting mitochondrial function.<sup>6,16</sup>

Additionally, our study also showed the immunological effect of chalcone. This study proved that chalcone administration significantly increased the number of Stomatocyte-containing Kupffer cells, which indicated increased phagocytic activity. However, this immunological effect is contrary to previous reports regarding the role of chalcone in host immunity. Chalcone contains two aromatic rings connected by an  $\alpha$ ,  $\beta$  unsaturated ketone, and reactive keto-ethylonic groups (-CO-CH = CH-) responsible for anti-inflammatory and antimalarial effects.<sup>20,21</sup> The anti-inflammatory effect of chalcone was reported by Arya et al., who showed that where phenyl-sulfonyl-uranyl-chalcone derivatives inhibited PGE2 production in LPS-induced RAW 264.7 macrophages through selective inhibition of COX-2 activity. Additionally, Singh et al. also reported that 2',5'-dimethoxy-4-hydroxy chalcone and 3,4-dichloro-2',5'-diethoxychalcone inhibited NO production in RAW 264.7 macrophages and LPS-induced microglial cells.<sup>22</sup> Therefore, further studies are needed to analyze the difference between our substance with previous studies and the detail of chalcone's immunological effects so the complete pictures of its effect can be unveiled and the treatment strategy can be devised for the clinical trial.

## CONCLUSION

In conclusion, the (E)-1-(4-aminophenyl)-3-(2,3-dimethoxy phenyl)prop-2-en-1-one exhibited potent antimalarial activity, possibly via enhancement of the phagocytic activity of Kupffer cells. Further studies are needed to assess the efficacy and the toxicity of this compound in vivo and to evaluate the immunological effect in detail so the complete pharmacological picture of this compound can be obtained.

## AUTHOR CONTRIBUTION

Danush Hermawan: processed the ethical clearance, prepared materials in this research, and doing the statistical test; Yulia Sari and Ida Nurwati: performing the research' procedures; Lilik Wijayanti: performing statistical analysis and preparing the manuscript; Paramasari Dirghayu: finalization of the manuscript.

## CONFLICT OF INTEREST

The author declared that there is no conflict of interest regarding all aspects of the study.

## ETHICS APPROVAL

This study has been approved by the Health Research Ethics Committee, Dr. Moewardi General Hospital/ Faculty of Medicine, Universitas Sebelas Maret, Surakarta, with letter number 104/II/HREC/2016.

## ACKNOWLEDGMENTS

Authors acknowledge the Institute of Research and Community Services' contribution, Universitas Sebelas Maret, Surakarta, through the Maintenance Research Group Funding scheme.

## FUNDING

This study was funded by the Institute of Research and Community Services, Universitas Sebelas Maret, Surakarta, through the Maintenance Research Group Funding scheme.

## REFERENCES

1. Papua DKP. *Profil Kesehatan Provinsi Papua Tahun 2017*. Dinas Kesehatan Provinsi Papua; 2017. [https://www.kemkes.go.id/resources/download/profil/PROFIL\\_KES\\_PROVINSI\\_2017/34\\_Papua\\_2017.pdf](https://www.kemkes.go.id/resources/download/profil/PROFIL_KES_PROVINSI_2017/34_Papua_2017.pdf)
2. Septiawan M, Budayatin B, Wiyono HT, Senjarini K. Immunogenicity of Protein Extract from Salivary Gland of *Anopheles aconitus* in Malaria Endemic Area. *J ILMU DASAR*. 2017;18(1):25. doi:10.19184/jid.v18i1.2372
3. Amato Roberto, Pharath Lim, Olivo Miotto, Chanaki Amaratunga, Dalin Dek, Richard D Pearson, Jacob Almagro-Garcia ATN, Sokunthea Sreng, Seila Suon, Eleanor Drury, Dushyanth Jyothi, Jim Stalker, Dominic P Kwiatkowski RMF. Genetic markers associated with dihydroartemisinin-piperaquine failure in Plasmodium falciparum malaria in Cambodia: a genotype-phenotype association study. *Lancet Infect Dis*. 2017;17(2):164-173. doi:10.1016/S1473-3099(16)30409-1.Genetic
4. Guantai EM, Ncoakazi K, Egan TJ, et al. Design, synthesis and in vitro antimalarial evaluation of triazole-linked chalcone and dienone hybrid compounds. *Bioorganic Med Chem*. 2010;18(23):8243-8256. doi:10.1016/j.bmc.2010.10.009
5. Pingaew R, Saekee A, Mandi P, et al. Synthesis, biological evaluation and molecular docking of novel chalcone-coumarin hybrids as anticancer and antimalarial agents. *Eur J Med Chem*. 2014;85:65-76. doi:10.1016/j.ejmech.2014.07.087

6. Go M, Liu M, Wilairat P, Rosenthal PJ, Saliba KJ, Kirk K. Antiplasmodial Chalcones Inhibit Sorbitol-Induced Hemolysis of. 2004;48(9):3241-3245. doi:10.1128/AAC.48.9.3241
7. Yadav N, Dixit SK, Bhattacharya A, et al. Antimalarial Activity of Newly Synthesized Chalcone Derivatives In Vitro. *Chem Biol Drug Des.* 2012;80(2):340-347. doi:10.1111/j.1747-0285.2012.01383.x
8. Suwito H, Jumina, Mustofa, et al. Design and synthesis of chalcone derivatives as inhibitors of the ferredoxin-ferredoxin-NADP+ reductase interaction of Plasmodium falciparum: pursuing new antimalarial agents. *Molecules.* 2014;19(12):21473-21488. doi:10.3390/molecules191221473
9. Setyarini II. Antiplasmodial activity and selectivity of chalcone derivatives. Published online 2015.
10. Wijayanti L, Purwanto B, Mudigdo A, Suwito H, Dirgahayu P. The Antiplasmodial Activity of Chalcone Derivatives Through the Inhibition of Hemozoin Formation And the Increased Number of Stomatocytes. 2018;(January):251-251. doi:10.26911/mid.icph.2018.05.13
11. Wijayanti L, Yarso KY, Purwanto B, et al. The antiplasmodial activity of chalcone derivative through the inhibition of haemozoin formation and the induction of stomatocytes formation. *Bali Med J.* 2019;8(1):365. doi:10.15562/bmj.v8i1.1196
12. Lee JS, Bukhari SNA, Mohd Fauzi N. Effects of chalcone derivatives on players of the immune system. *Drug Des Devel Ther.* 2015;9:4761-4778. doi:10.2147/DDDT.S86242
13. Parlapalli A, Manda S. *Pharmacophore.* 2015;6(5):243-248.
14. Hans RH, Guantai EM, Lategan C, et al. synthesis, antimalarial and antitubercular activity of acetylenic chalcones. *Bioorganic Med Chem Lett.* 2010;20(3):942-944. doi:10.1016/j.bmcl.2009.12.062
15. Tadigoppula N, Korhikunta V, Gupta S, et al. synthesis and insight into the structure-activity relationships of chalcones as antimalarial agents. *J Med Chem.* 2013;56(1):31-45. doi:10.1021/jm300588j
16. Park P-H, Kim HS, Jin XY, et al. KB-34, a newly synthesized chalcone derivative, inhibits lipopolysaccharide-stimulated nitric oxide production in RAW 264.7 macrophages via heme oxygenase-1 induction and blockade of activator protein-1. *Eur J Pharmacol.* 2009;606(1):215-224. doi:https://doi.org/10.1016/j.ejphar.2008.12.034
17. ParkSH, KwakJA, JungSH, et al. Piperidylmethoxychalcone improves immune-mediated acute liver failure via inhibiting TAK1 activity. *Exp Mol Med.* 2017;49(11):1-10. doi:10.1038/emmm.2017.156
18. Frölich S, Schubert C, Bienzle U, Jenett-Siems K. In vitro antiplasmodial activity of prenylated chalcone derivatives of hops (*Humulus lupulus*) and their interaction with haemin. *J Antimicrob Chemother.* 2005;55(6):883-887. doi:10.1093/jac/dki099
19. Mi-Ichi F, Miyadera H, Kobayashi T, et al. Parasite mitochondria as a target of chemotherapy: Inhibitory effect of licochalcone A on the Plasmodium falciparum respiratory chain. *Ann N Y Acad Sci.* 2005;1056:46-54. doi:10.1196/annals.1352.037
20. Chu J, Guo CL. Design and Discovery of Some Novel Chalcones as Antioxidant and Anti-Inflammatory Agents via Attenuating NF-κB. *Arch Pharm (Weinheim).* 2016;349(1):63-70. doi:10.1002/ardp.201500349
21. Sarda SR, Jadhav WN, Bhusare SR, et al. Solvent-free NaOH-Al<sub>2</sub>O<sub>3</sub> supported synthesis of Ar. 2009;1(2):265-269.
22. Singh H, Sidhu S, Chopra K, Khan MU. Hepatoprotective effect of trans-Chalcone on experimentally induced hepatic injury in rats: Inhibition of hepatic inflammation and fibrosis. *Can J Physiol Pharmacol.* 2015;94(8):879-887. doi:10.1139/cjpp-2016-0071



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