Synthesis and in vitro SAR evaluation of natural vanillin-based chalcones tethered quinolines as antiplasmodial agents

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Abstract

In pursuit of antimalarial drug, to overcome the drug resistance and the risk of drug-drug interactions, the chalcone hybrids and their structure-activity relationship for the antimalarial activity studied against chloroquine sensitive as well as multi-drug resistant strain of Plasmodium falciparum. Our study has revealed that 7- Chloro quinoline groups on chalcone increase anti-malarial potency, while the positional interchange of these groups decreases the efficacy. Particularly, chloro-substituent provided potent analogues which were easily derived from naturally available vanillin. The most active compounds were relatively non-toxic and structure-activity relationship study suggested that 7-chloroquinoline group when attached with triazole linkage increased the antimalarial potential of the compound against both chloroquine-sensitive and multidrug resistant strain.

Introduction

Malaria is a mosquito-borne disease, which is associated with high morbidity and mortality [1]. Although, diverse potent antimalarial agents including quinolones [2] (e.g. chloroquine) and endoperoxides [3a] (e.g. Artemisinin and its derivatives) [3b] are available, emergence of resistance against front-line drugs has raised a major health concern. Thus, the search for newer efficacious drugs as well as new molecular frameworks possessing antimalarial activity remains a vital goal towards achieving control over malaria [4]. Among various antimalarial molecules, hybrid molecules also represent logical approach in designing and development of novel therapeutics that have the potential to overcome the rapid development of drug resistance, enhance patient difficulties, reduce both the cost and the risk of drug-drug interactions compared to cocktails or multicomponent drugs [5,6]. In view of their encouraging efficacies with good bioavailability and minimized toxicity; the next generation drugs may be hybrid molecules reducing the possibility of resistance towards them. This was demonstrated by the compounds such as tetraoxaquine **1a**, trioxaquine, trioxaferroquine and stilbene-chalcone hybrid **1b** [7-9, 26] (Figure 1).

Chalcone (1,3-diaryl-2-propen-1-one), coumarin, triazole, and quinoline are major classes of naturally occurring compounds which have been reported to possess many effective chemotherapeutic properties [10-13]. These pharmacophores that are derived from both natural and synthetic in a molecule, each with a distinct mechanism of action and could be beneficial in the malaria treatment. Ratifying this approach, various research groups have reported hybrid molecules by coupling of chalcone, coumarin, curcumin, flavone and quinoline with different other bioactive molecules like: resveratrol, maleimide and alpha-lipoic acid [14, 16] to address the problems with hybrid molecules and their solution-solubility with toxicity. Our group has reported chalcone, chalcone-stilbene hybrids and distyrylbenzenes for their antimalarial activities [17-18]. In synthetic perspective, chalcone, quinoline and coumarin derivatives are getting more attention due to their pivotal role in the organic as well as medicinal chemistry [19]. So, we designed some novel hybrid molecules and developed SAR (Structure Activity Relationship) against malaria parasite *Plasmodium falciparum.*

Results And Discussion

Chemistry

Earlier, we studied the anti-plasmodial activity of allylated chalcone based on the licochalcone **A** against chloroquine (CQ) sensitive Pf3D7 and CQ resistant PfINDO strains of *Plasmodium falciparum* [7]. Among them particular 1-(4-Chlorophenyl)-3-[3-methoxy-4-(prop-2-en-1-yloxy)phenyl]-prop-2-en-1-one was the most potent (IC_{50}: 3.0 μM) against Pf3D7 with resistance indices of 1.2 and 6.6 against PfDd2, and PfINDO strains, respectively. These results fascinated us to develop a SAR around this molecule by the synthesis of its derivatives with diverse functionalities to enhance the antiplasmodial activity. In this regard, a chalcone **3** was synthesized by the condensation of 4-Chloroacetophenone **2a** and vanillin **2b** in presence of base. Furthermore, Propargylated chalcone **3c** was obtained by the reaction of chalcone **3** with propargyl bromide. This further reacts with 4-azido-7-chloroquinoline **3d** and gives Compound **3e** copper catalyzed click chemistry (Scheme 1). However, allylated chalcone **4** was also synthesized from chalcone **3** by the reaction of allyl bromide. While, compound **3a** was synthesized from compound **3** following the Mannich reaction condition (Scheme 1).

In another attempts, our attention shifted to check the reliability of 4-chloro substituent in ring A (Figure 3). So we have replaced chloro substituent by 7-chloro quinoline **6b** in ring A and synthesized 1-(4-((7-chloroquinolin-4-yl)amino)phenyl)ethenone **7** and its derivatives **8-11**. These were synthesized from intermediate **6a** and 4,7-dichloroquinoline **6b** and fallowed by alkylation as shown in (scheme 2).

Notably that 7-chloroquinoline containing chalcone **3e** was most active but when we have attached 7-chloroquinoline **6b** in ring **A,** surprisingly total activity was lost. Furthermore, in another attempt, our attention shifted to replace 4-chloro ring with coumarin **12a** and **12b**, which has been reported to possess good pharmacokinetic properties. Henceforth, 3-acetylcoumarin **13b** was synthesized by the reaction of 2-hydroxybenzaldehyde **12b** and ethyl acetoacetate **12c** in presence of piperidine as a base, which was further reacted with vanillin **2b** via Claisen–Schmidt condensation reaction to get coumarin-chalcone **13b** (**scheme 3**) in acidic methanol (acetyl chloride in methanol), at room temperature for 3 h. After completion of the reaction, the mixture was filtered to collect the precipitates and purification by recrystallization afforded the pure hydroxylated and methoxylated coumarin-chalcone hybrids **14a** and **14b** in 52% and 75% yield respectively. Considering the significance of O-allyl groups, these coumarin-chalcone hybrids were O-allylated **15** and **16** by using allyl bromide and K_2CO_3 in DMF. Additionally, O-propargylated coumarin-chalcone **17a** was also synthesized by using propargyl bromide in presence of K_2CO_3 in DMF. Further keeping in mind, chalcone quinolone hybrid **3e** (**scheme 1**) was most active. So, we performed a reaction of propargylated coumarin-chalcone **17a** with 4-azido-7-chloroquinoline **17b** in presence of CuSO4, sodium ascorbate in DMF at 60°C to get triazole-linked coumarin-chalcone-quinoline hybrid **19**. On the other side chloroacetylchloride was reacted with coumarin-chalcone **14b** to get chloroacetylated coumarin-chalcone **18** which was further reacted with secondary amine piperidine to get the compound **20**. (**Scheme 3**).

Anti-malarial activity

All the compounds were tested for antimalarial activity against chloroquine sensitive *P. falciparum* 3D7 (*Pf*3D7) strain and chloroquine resistant *P. falciparum* K1 strain (Tables 1-2) by SYBR-Green-Iassay[20]. The fluorescence readout gives an indication of parasite growth in infected RBCs. SYBR green based fluorescence plotted with respect to drug concentration gives precise estimation of parasite inhibitory concentrations. In one of the recent works, the most active compound **3e** i.e. 4-chlorochalcone exhibited profound *in vitro* antimalarial activity (IC_{50} = 2.5 μM) which was comparable to Licochalcone and far superior to its well-reported analogue,4-dimethoxy-4'-butoxychalcone [21]. Thereafter, the structure activity relationship studies were carried out by changing the substitutions on ring B keeping 4-chloro substituent constant on ring A. Amino methyl, allyl and propargyl substituent on ring B (compound **3a-3c** and **4**) exhibited no activity against both the strains but compound **3e** was quite efficient in killing both chloroquine sensitive and resistant strain. This clearly shows the benefit of addition of triazole linked 7-Chloro quinoline to the chalcone. We next ventured to evaluate the positional importance of amino 7-chloroquinoline group on ring A

Table 1. Antimalarial activity and selectivity index of compounds (3-20)

Entry	Compound No	IC_{50} (μM) Pf3D7	IC_{50} (μM) PfK1	CC_{50} (μM)	Selectivity index Pf3D7	Selectivity index PfK1
1	**3**	>5	ND	ND	ND	ND
2	**4**	>5	ND	95.58	<19.1	ND
3	**3a**	>5	>5	ND	ND	ND
4	**3b**	>5	>5	ND	ND	ND
5	**3c**	>5	>5	90.26	ND	ND
6	**3e**	**4.12**	**3.55**	**46.18**	**11.2**	**13.0**
8	**7**	3.90	3.77	12.19	3.1	3.2
9	**8**	1.91	4.10	13.0	6.8	3.1
10	**9**	>5	ND	ND	ND	ND
11	**10**	4.13	>5	23.97	5.8	ND
12	**11**	>5	ND	ND	ND	ND
13	**14a**	>5	>5	ND	ND	ND
14	**15**	>5	ND	ND	ND	ND
15	**14b**	>5	ND	ND	ND	ND
16	**16**	>5	ND	ND	ND	ND
17	**17a**	>5	ND	95.58	<19.1	ND
18	**18**	>5	ND	ND	ND	ND
19	**19**	>5	>5	>200	ND	ND
20	**20**	>5	>5	ND	ND	ND
21	**CQ- diphosphate**	**0.005**	**0.258**	**125**	**25000**	**484**

(Table 1, **3-20**).

It is known that activity was markedly affected by *para*-substitution of *O*-allyl group [21]. So, allylated vanillin substitution at the ring **B** was kept unchanged and subsequently, the effect of changing the nature of the N-substituent (H, allyl, phenyl, C_4H_9 and $CH_2C_6H_4Br$) was evaluated and reduced activity was observed in each instance (Table **1**, **8** and **10-12**) except N-allylated chalcone (**9**) but lacked

selectivity. It is observed that there was no enhancement in antimalarial activity for any of the coumarin-chalcone hybrids (Table 2, **13-20**). Heteroaryl-substitution is an appealing strategy for desirable activity and several inspiring reports on the antimalarial activity of heterocyclic containing chalcone derivatives [22] boost up for us to synthesize such analogues. We designed chalcones by the condensation of benzaldehydes with different heterocyclic carbonyls like 7-chloroquinoline, coumarin [23]. However, in each case the antimalarial potential was not found, although, the 7-chloroquinoline is considered an excellent lead prototype for the development of antimalarial drugs [24, 25].

Microscopic examination of antimalarial activity

Compound 3e was studied for microscopic examination and pictures of control and treated samples revealed that the compound caused drastic effects on parasite growth. After 24 hours, ring stage parasite progressed into late trophozoite and schizont stage in control, whereas, the compound-treated sample showed delayed parasite growth and they were arrested in early trophozoite stage. After 48 hours, healthy schizonts in the control sample progressed into a new infection cycle and parasites were predominantly in ring stage. While, the treated parasite showed mainly stressed trophozoite with reduced staining of parasite DNA, probably due to excessive DNA damage. We also performed experiments to check whether parasite is able to recover after removal of compound 3e. Due to dramatic genotoxic effect of compound 3e, the parasite shows poor recovery from stress even after drug removal (Figure 2).

Conclusion

The study revealed that among all synthesized hybrid molecules vanillin-quinoline hybrid molecules show better antimalarial potency (maximum IC_{50} of 1.91 and 3.77 μM against *Pf*3D7 and *Pf*K1, respectively) in comparison to vanillin-coumarin hybrid molecules ((IC_{50} of greater than 5 μM) as well as natural licochalcone (IC_{50} of 4.1 μM). Microscopic examination studies of compound **3e** show drastic effect on parasite growth even after removal of compound. Some of these compounds revealed not only promising activities but are also easy and economical to prepare and thus might prove useful leads towards future antimalarial drug discovery.

Experimental Section

Chemistry

Chemical and reagents

All the reagents were obtained from commercial sources (Merck or Acros). The solvents used for isolation/purification of compounds were obtained from commercial sources (Merck) and used without further purification. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker Avance-400 spectrometer. TMS

was used as internal reference for ^{1}H NMR. HRMS-ESI spectra were determined using micro mass Q-TOF ultima spectrometer.

Procedure for the synthesis of 4-chloro substituted chalcones (3-4)

(*E*)-1-(4-Chlorophenyl)-3-(4-hydroxy-3-methoxyphenyl)prop-2-en-1-one.(**3**)

To the solution of vanillin **2b** (3 mmol) and 4-chloroacetophenone **2a** (3 mmol) in ethanol (20 mL), KOH (4 mmol) was added. Reaction progress was monitored with TLC. After completion reaction mixture was concentrated and washed with water, taken in ethyl acetate and dried over sodium sulfate. The desired compound was obtained after recrystallization in methanol and characterized by ^{1}H &^{13}C NMR and HRMS data. Bright yellow solid (Yield 60%) m.p. 110-115°C, ^{1}H NMR ($CDCl_3$, 400 MHz): *d* (ppm) 7.95 (d, *J* = 8.6 Hz, 2H), 7.75 (d, *J* = 15.6 Hz, 1H), 7.46 (d, *J* = 8.6 Hz, 2H), 7.32 (d, *J* = 15.6 Hz, 1H), 7.21 (dd, *J* = 8.2, 1.6 Hz, 1H), 7.12 (d, *J* = 1.6 Hz, 1H), 6.96 (d, *J* = 8.2 Hz, 1H), 6.09 (1H, s), 3.95 (3H, s); ^{13}C NMR ($CDCl_3$,100 MHz): *d* (ppm) 189.4, 148.6, 146.9, 145.8, 139.0, 136.8,129.9, 128.9, 127.3, 123.5, 119.2, 115.0, 110.2, 56.1. HRMS-ESI: m/z $[M+H]^+$ for $C_{16}H_{14}ClO_3$, calculated 289.0631; observed 289.0625.

(E)-1-(4-Chlorophenyl)-3-[3-methoxy-4-(prop-2-en-1-yloxy)phenyl]prop-2-en-1-one **(4)**.

Compound **3** (0.99 mmol) was treated with allyl bromide (1.05 mmol) in presence of K_2CO_3 (1.99 mmol) in DMF (5 mL) at rt. for 6 h. Reaction mixture was diluted with water and desired compound **4** was obtained by filtration and recrystallization with methanol and characterized by ^{1}H &^{13}C NMR and HRMS data. Pale yellow solid (Yield 82%) m.p. 90-93°C, ^{1}H NMR ($CDCl_3$,400 MHz): (ppm) 7.95 (d, *J* = 8.6 Hz, 2H), 7.75 (d, *J* = 15.6 Hz, 1H), 7.47 (d, *J* = 8.6 Hz, 2H), 7.33 (d, *J* = 15.6 Hz, 1H), 7.20 (d, *J* = 8.4, 1.9 Hz, 2H,), 7.16 (d, *J* = 1.9 Hz, 1H), 6.90 (d, *J* = 8.3 Hz, 1H), 6.14-6.04 (1H, m), 5.43 (dd, *J* = 17.2, 1.4 Hz, 1H,), 5.32 (d, *J* = 10.5, 1.3 Hz, 1H), 4.68 (m, 1H), 3.95 (3H, s); ^{13}C NMR ($CDCl_3$, 100 MHz): δ (ppm) 189.3, 150.7, 149.7, 145.5, 139.0, 136.8, 132.7, 129.9, 128.9, 127.9, 123.1, 119.6, 118.5, 113.0, 110.7, 69.8, 56.1. HRMS-ESI: m/z $[M+H]^+$ for $C_{19}H_{18}ClO_3$, calculated 329.0944 observed 329.0945.

(E)-1-(4-chlorophenyl)-3-(4-hydroxy-3-methoxy-5-(piperidin-1-ylmethyl)phenyl)prop-2-en-1-one (**3a**)

Compound **3** (0.69 mmol) was treated with paraformaldehyde (1.38 mmol), piperidine (1.38 mmol) in DMF (3mL) at 60° C for 20 hours. Reaction mixture was diluted with water and desired compound **3a** was obtained by filtration and recrystallization in methanol. This was fully characterized by ^{1}H &^{13}C NMR and HRMS data. Yellow solid (Yield 67%) m.p. 109-112°C, ^{1}H NMR ($CDCl_3$,400 MHz): δ (ppm) 8.17 (d, *J* = 8.0 Hz, 2H), 7.76 (d, *J* = 15.4 Hz, 1H), 7.68 (d, *J* = 15.2 Hz, 1H), 7.63 (d, *J* = 8 Hz, 2H), 7.47 (s, 1H), 7.26 (s, 1H), 3.87 (s, 3H), 3.65 (s, 2H), 3.61 (t, *J* = 4.0 Hz, 4H), 2.47 (m, 5H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ(ppm) 189.1, 150.2, 148.3, 145.7, 138.9, 136.8, 129.8, 128.8, 126.0, 122.7, 120.9, 118.8, 110.4, 66.1, 61.4, 56.0 and 52.8. HRMS-ESI: m/z $[M+H]^+$ for $C_{22}H_{25}ClNO_3$, calculated 386.1523 observed 386.1535.

(E)-3-(4-(allyloxy)-3-methoxy-5-(piperidin-1-ylmethyl)phenyl)-1-(4-chlorophenyl)prop-2-en-1-one (**3b**)

Compound **3a** (0.99 mmol) was treated with allyl bromide (1.05 mmol) in presence of K_2CO_3 (1.99 mmol) in DMF (5 mL) at rt. for 6 hours. Reaction mixture was diluted with water and desired compound **3b** was obtained by filtration fallowed by column purification with hexane: ethyl acetate (8:2) which was characterized by ^1H &^{13}C NMR and HRMS data.

Yellow oil (Yield 59%), ^1H NMR ($CDCl_3$,400 MHz): δ(ppm) 7.97 (d, *J* = 8 Hz, 1H), 7.72 (d, *J* = 16 Hz, 1H), 7.49 (d, *J* = 8 Hz, 2H), 7.37 (d, *J* = 16 Hz, 1H), 7.30 (s, 1H), 7.09 (s, 1H), 6.15-6.08 (m, 1H), 5.41-5.36 (m, 1H), 4.59 (d, *J* = 4.0 Hz, 2H), 3.92 (s, 3H), 3.65 (s, 2H), 3.72-3.70 (t, *J* = 4 Hz, 4H), 3.50 (s, 2H), 2.50 (m, 4H), 1.26 (s, 2H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ(ppm) 189.3, 150.2, 150.0, 145.9, 139.3, 137.2, 133.1, 130.2, 129.3, 128.3, 123.5, 119.9, 118.9, 113.4, 111.1, 70.2, 59.2, 57.0, 54.1, 28.7 and 25.4. HRMS-ESI: m/z $[M+H]^+$ for $C_{25}H_{29}ClNO_3$, calculated 426.1836 observed 426.1848.

(E)-1-(4-Chlorophenyl)-3-[3-methoxy-4-(prop-2-yn-1-yloxy)phenyl]prop-2-en-1-one **(3c)**.

Compound **3** (0.99 mmol) was treated with propargyl bromide (1.05 mmol) in presence of K_2CO_3 (1.99 mmol) in DMF (4 mL) at rt. for 6 hours. Reaction mixture was diluted with water and desired compound **3c** was obtained by filtration and recrystallization with methanol and characterized by ^1H &^{13}C NMR and HRMS data. Pale yellow solid (Yield 88%) m.p. 90-93°C, ^1H NMR ($CDCl_3$,400 MHz): δ (ppm) 7.95 (d, *J* = 8.6 Hz, 2H), 7.76 (d, *J* = 15.6 Hz, 1H), 7.47 (d, *J* = 8.6 Hz, 2H), 7.35 (d, *J* = 15.6 Hz, 1H), 7.24 (dd, *J* = 8.3, 1.9 Hz, 1H), 7.17 (d, *J* = 1.9 Hz, 1H), 7.06 (d, *J* = 8.3 Hz, 1H), 4.82 (d, *J* = 2.4 Hz, 2H), 3.95 (3H, s), 3.59 (t, *J* = 2.4 Hz, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ (ppm) 189.2, 149.8, 149.3, 145.3, 139.1, 136.7, 129.9, 128.9, 122.7, 120.0, 113.8, 110.8, 77.9, 76.3, 56.7, 56.1. HRMS-ESI: m/z $[M+H]^+$ for $C_{19}H_{16}ClO_3$, calculated 327.0788 observed 327.0781.

(E)-1-(4-Chlorophenyl)-3-(4-((1-(7-chloroquinolin-4-yl)-1H-1,2,3-triazol-4-yl)methoxy)-3-methoxyphenyl)prop-2-en-1-one **(3e)**

Compound **3c** (0.61 mmol) was treated with 4-azido-7-chloroquinoline **3d** (0.61 mmol) in presence of copper sulfate (0.12 mmol), sodium ascorbate (0.25 mmol) in DMF (5mL) at rt. for 15 hours. Reaction mixture was diluted with water and desired compound **3e** was obtained by filtration and recrystallization with chloroform/methanol and characterized by ^1H &^{13}C NMR and HRMS data. Pale yellow solid (Yield 62%) m.p. 90-93°C, ^1H NMR ($CDCl_3$,400 MHz): δ (ppm) 9.20 (s, 1H), 9.00 (s, 1H), 8.31 (s, 1H), 8.19 (d, *J* = 8.5 Hz, 2H), 8.02 (d, *J* = 9.1 Hz, 1H), 7.90 (dd, *J* = 7.7 4.2 Hz, 1H), 7.83 (dd, *J* = 6.0, 9.2 Hz, 2H), 7.76 (d, *J* = 15.6, 1H), 7.64 (d, *J* = 8.5 Hz, 2H), 7.60 (s, 1H), 7.47 (d, *J* = 6.4 Hz, 1H), 7.34 (d, *J* = 8.3 Hz, 1H), 5.41 (s, 1H), 3.89 (3H, s); ^{13}C NMR ($CDCl_3$, 100 MHz): δ (ppm) 188.5, 152.8, 150.4, 149.8, 145.4, 143.7, 140.8, 138.4, 136.9, 135.9, 130.8, 129.5, 129.3, 128.6, 128.6, 127.7, 125.9, 125.8, 124.4, 120.1, 117.7, 113.9, 111.7, 56.2. HRMS-ESI: m/z $[M+H]^+$ for $C_{28}H_{21}Cl_2N_4O_3$, calculated 531.0991 observed 531.0987.

Procedure for the synthesis of 7-chloroquinoline-chalcones and its derivatives (Compounds 6-11)

(E)-3-(4-(Allyloxy)-3-methoxyphenyl)-1-(4-((7-chloroquinolin-4-yl)amino)phenyl) prop-2-en-1-one **(7).**

To the solution of 4-allyloxyvanillin **5b** (3 mmol) and 4-aminoacetophenone **5a** (3 mmol) in ethanol (20 mL), KOH (4 mmol) was added and reaction mixture was stirred for 8 hours. After completion of reaction, reaction mixture was concentrated and washed with water and recrystallized in methanol to give compound **6**. Which was further treated with 4,7-dichloroquinoline in DMF at rt. for overnight to give compound **7**. Reaction progress was monitored with TLC. After completion reaction mixture was concentrated and washed with water, recrystallized in methanol and characterized by ^{1}H &^{13}C NMR and HRMS data. Yellow solid (Yield 76%) m.p. 168-171°C, ^{1}H NMR (DMSO-d_6, 400 MHz): (ppm) 8.78 (d, *J* = 9.0 Hz, 1H), 8.63 (d, *J* = 6.3 Hz, 1H), 8.31 (d, *J* = 8.4 Hz, 2H), 8.14 (s, 1H), 7.89 (d, *J* = 15.5 Hz, 1H) 7.82 (d, *J* = 9.0 Hz, 1H), 7.73 (d, *J* = 15.5 Hz, 1H), 7.65 (d, *J* = 8.4 Hz, 2H), 7.58 (s, 1H), 7.39(d, *J* = 8.3 Hz, 1H), 7.17 (d, *J* = 6.4 Hz, 1H), 7.04 (d, *J* = 8.4 Hz, 1H) 6.11-6.02 (m, 1H), 5.42 (dd, *J* = 17.3, 1.52 Hz, 1H), 5.28 (dd, *J* = 10.5, 1.2 Hz, 1H), 4.64 (d, *J* = 5.24 Hz, 1H), 3.89, (s, 3H); ^{13}C NMR (DMSO-d_6, 100 MHz): δ (ppm) 180.0, 150.6, 149.7, 147.0, 144.9, 143.3, 143.1, 137.6, 135.1, 134.0, 130.8, 128.2, 127.4, 126.3, 124.3, 123.2, 122.6, 120.0, 118.4, 117.9, 113.5, 112.6, 111.6, 103.0, 69.4, 56.3.HRMS-ESI: m/z [M+H]+ for $C_{28}H_{24}ClN_2O_3$, calculated 471.6449; observed 471.1464.

General procedure for the synthesis 7-chloroquinoline-chalcone derivative (8-11)

To the solution of compound **7** (2.7 mmol) in dry tetrahydrofuran (15 mL), potassium hydroxide (13.5 mmol), allyl/propargyl/benzyl/4-bromobenzyl bromide (5.5 mmol) and cetyltrimethylammonium bromide (CTAB) (0.7 mmol) was added. The contents were stirred at room temperature for 12-14 h till the starting disappeared on TLC. After the completion of reaction, the reaction mixture was partitioned between ethyl acetate (70 mL) and water (15 mL). The ethyl acetate layer was washed with water till neutral, dried over sodium sulfate and evaporated. The obtained residue was purified by column chromatography in hexane: ethyl acetate (7:3 v/v) to afford the desired compounds **(8-11)** whose structure was confirmed through NMR and mass spectrometry.

(E)-1-(4-(Allyl(7-chloroquinolin-4-yl)amino)phenyl)-3-(4-(allyloxy)-3-methoxy phenyl) prop-2-en-1-one **(8).**

Orange-yellow viscous liquid (Yield 61%), ^{1}H NMR ($CDCl_3$, 400 MHz): δ (ppm) 7.40 (d, *J* = 8.3 Hz, 2H), 6.92 (d, *J* = 7.8 Hz, 2H), 6.73 (s, 1H), 6.56 (s, 1H), 6.49-6.30 (m, 4H), 6.06 (s, 1H), 6.05 (d, *J* = 7.8 Hz, 1H), 5.90 (d, *J* = 4.9 Hz, 1H), 5.70 (d, *J* = 8.4 Hz, 1H), 4.80-4.75 (m, 2H), 4.19-4.02 (m, 1H), 3.40 (d, *J* = 5.1 Hz, 2H), 3.30 (d, *J* = 5.1 Hz, 2H), 2.60 (s, 3H); ^{13}C NMR ($CDCl_3$, 75.4 MHz): δ (ppm) 188.9, 151.4, 151.0, 150.0, 149.5, 147.7, 145.0, 144.7, 143.7, 137.6, 136.0, 135.0, 133.6, 130.7, 128.6, 127.7, 125.1, 124.0, 123.7, 122.7, 120.2, 118.4, 116.2, 113.7, 111.5, 103.4, 70.0, 56.3 and 44.5. HRMS-ESI: m/z [M+H]+ for $C_{31}H_{28}ClN_2O_3$, calculated 511.6759; observed 511.6782.

(E)-3-(4-(allyloxy)-3-methoxyphenyl)-1-(4-((7-chloroquinolin-4-yl)(prop-2-yn-1-yl)amino)phenyl)prop-2-en-1-one **(9)**.

Yellow solid (Yield 67%) m.p. 79-81°C, ^{1}H NMR ($CDCl_3$, 400 MHz): δ (ppm) 9.00 (d, *J* = 4.0 Hz, 1H), 8.20 (d, *J* = 2.0 Hz, 1H), 7.78 (s, 1H), 7.76 (d, *J* = 5.2 Hz, 1H), 7.48-7.38 (m, 1H), 7.21 (d, *J* = 8.0 Hz, 1H), 7.16 (d, *J* = 2.0 Hz, 1H), 6.92 (d, *J* = 8.0 Hz, 1H), 6.88 (d, *J* = 8.0 Hz, 1H), 6.11 (m, 1H), 5.46 (dd, *J* = 16.0, 1.2 Hz, 1H), 5.35 (dd, *J* = 10.5, 1.2 Hz, 1H), 4.68 (d, *J* = 5.4 Hz, 2H), 4.62 (d, *J* = 2.0 Hz, 2H), 3.94 (s, 3H), 2.39 (s, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ 188.3, 152.2, 151.1, 150.6, 150.4, 150.3, 149.6, 144.0, 136.0, 132.7, 130.8, 130.3, 129.1, 128.2, 128.1, 128.0, 125.1, 123.8, 122.7, 119.7, 118.5, 118.4, 116.0, 113.0, 110.5, 78.2, 74.1, 69.7, 56.0 and 42.5. HRMS-ESI: m/z $[M+H]^+$ for $C_{31}H_{26}ClN_2O_3$, calculated 509.1632; observed 509.1626.

(E)-3-(4-(allyloxy)-3-methoxyphenyl)-1-(4-(benzyl(7-chloroquinolin-4-yl)amino)phenyl)prop-2-en-1-one **(10).**

Yellow solid (Yield 66%) m.p. 224-226°C, ^{1}H NMR ($CDCl_3$, 400 MHz): δ (ppm) 8.8 (d, *J* = 6.0 Hz, 1H), 8.24 (d, *J* = 6.0 Hz, 1H), 8.06 (d, *J* = 16.0 Hz, 1H), 7.86 (s, 1H), 7.68-7.58 (m, 3H), 7.16-7.36 (m, 8H), 6.92 (d, *J* = 8.0 Hz, 1H), 6.79 (d, *J* = 8.0 Hz, 2H), 6.56 (d, *J* = 8.0 Hz, 1H), 6.09 (m, 1H), 5.42 (dd, *J* = 16.0, 1.2 Hz, 1H), 5.29 (dd, *J* = 10.5, 1.2 Hz, 1H), 4.68 (d, *J* = 2.0 Hz, 2H), 4.62 (s, 2H), 3.94 (s, 3H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ 188.7, 155.2, 152.5, 150.9, 149.6, 147.9, 145.1, 138.9, 135.1, 133.4, 130.8, 128.5, 127.5, 126.7, 124.1, 123.9, 122.7, 120.2, 118.6, 113.7, 111.5, 103.4, 70.1, 59.9 and 56.0. HRMS-ESI: m/z $[M+H]^+$ for $C_{35}H_{30}ClN_2O_3$, calculated 561.1945; observed 561.1932.

(E)-3-(4-(allyloxy)-3-methoxyphenyl)-1-(4-((4-bromobenzyl)(7-chloroquinolin-4-yl)amino)phenyl)prop-2-en-1-one **(11).**

Yellow solid (Yield 66%) m.p. 232-235°C, ^{1}H NMR ($CDCl_3$, 400 MHz): δ (ppm) 8.54 (d, *J* = 8.0 Hz, 1H), 8.11 (d, *J* = 8.0 Hz, 1H), 8.00 (d, *J* = 16.0 Hz, 1H), 7.71 (s, 1H), 7.52-7.46 (m, 6H), 7.35 (d, *J* = 8.0 Hz, 1H), 7.20-7.12 (m, 4H), 6.92 (d, *J* = 8.0 Hz, 1H), 6.72 (d, *J* = 8.0 Hz, 2H), 6.48 (d, *J* = 8.0 Hz, 1H), 6.08 (m, 1H), 5.40 (dd, *J* = 16.0 1.2 Hz, 1H), 5.24 (dd, *J* = 10.5, 1.2 Hz, 1H), 4.66 (d, *J* = 2.0 Hz, 2H), 4.61 (s, 2H), 3.81 (s, 3H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ 189.9, 156.3, 153.7, 151.0, 149.8, 147.7, 146.1, 138.0, 133.1, 131.2, 130.9, 128.9, 127.5, 124.1, 123.9, 122.7, 121.1, 118.6, 115.2 114.7, 111.5, 69.8, 57.0 and 56.3. HRMS-ESI: m/z $[M+H]^+$ for $C_{35}H_{29}BrClN_2O_3$, calculated 639.1050; observed 639.1039.

Procedure for the synthesis of coumarin-chalcones and its derivatives (Compounds 13-20)

(E)-3-(3-(4-hydroxy-3-methoxyphenyl)acryloyl)-6-methoxy-2H-chromen-2-one **(14a)**

Acetyl chloride (3 mL) was added drop wise to ice cold methanol (25 mL) with stirring and after 5 min 12a was added resulted 3-acetyl-6-methoxy-2H-chromen-2-one **13a** (0.01 mmol) was obtained. **13a** and vanillin **2b** (0.01 mmol) were stirred for 24 h at rt in the presence of NaOH in ethanol. Reaction mixture was concentrated and washed with water. The desired compound **14a** was recrystallized in methanol and characterized by ^{1}H &^{13}C NMR and HRMS data. Yellow solid (Yield 70%) m.p. 148-151°C, ^{1}H NMR ($CDCl_3$, 400 MHz): δ (ppm) 8.58 (s, 1H), 7.82 (d, J = 16.0 Hz, 1H), 7.69 (d, J = 8.0 Hz, 1H), 7.17 (m, 2H), 7.02 (d, J = 16.0 Hz, 1H), 6.99 (d, J = 8.0 Hz, 1H), 6.87 (s, 1H), 6.79 (d, J = 8.0 Hz, 1H), 3.98 (s, 3H), 3.91 (s, 3H); ^{13}C

NMR ($CDCl_3$, 100MHz): δ (ppm) 183.6, 159.8, 155.6, 149.9, 148.2, 145.7, 134.5, 126.0, 125.3, 124.1, 122.3, 119.0, 118.9, 117.0, 113.2, 111.1, 57.0 and 55.8. HRMS-ESI: m/z [M+H]+ for C20H17O6, calculated 353.1025; observed 353.1043.

(E)-3-(3-(4-(allyloxy)-3-methoxyphenyl)acryloyl)-6-methoxy-2H-chromen-2-one **(15)**

Compound **14a** (0.62 mmol) was treated with allyl bromide (0.62 mmol) in presence of K_2CO_3 (0.74 mmol) in DMF at rt. for 6 hours. Reaction mixture was diluted with water and desired compound was obtained by filtration and recrystallization with methanol/DCM and characterized by 1H &^{13}C NMR and HRMS data. Yellow solid (Yield 70%) m.p. 178-181°C, ^{1}H NMR ($CDCl_3$, 400 MHz): δ (ppm) 8.53 (s, 1H), 7.81 (d, J = 16.0 Hz, 2H), 7.79 (d, J = 8.0 Hz, 1H), 7.21-7.13 (m, 2H), 7.02 (d, J = 16.0 Hz, 1H), 6.99 (d, J = 7.4 Hz, 1H), 6.76 (m, 1H), 6.06 (m, 1H), 5.41 (dd, J = 17.2, 10.3 Hz, 2H), 4.68 (s, 2H), 3.95 (s, 6H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ (ppm) 182.0, 159.1, 156.6, 147.2, 145.7, 142.0, 134.5, 133.1, 127.9, 125.6, 123.3, 122.3, 118.7, 111.0, 70.1, 56.0 and 55.6. HRMS-ESI: m/z [M+H]+ for C23H21O6, calculated 393.1338; observed 393.1333.

(E)-3-(3-(4-Hydroxy-3-methoxyphenyl)acryloyl)-2H-chromen-2-one **(14b).**

Acetyl chloride (3 mL) was added drop wise to ice cold methanol (25 mL) with stirring and after 5 min. 3-acetyl-2H-chromen-2-one **13b** (0.01 mmol) was added and stirred for 4-5 min. To this solution vanillin (0.01 mmol) was added and stirred for 24 h at rt. Reaction mixture was concentrated and washed with water. The desired compound **14b** was recrystallized in methanol and characterized by 1H &^{13}C NMR and HRMS data. Yellow solid (58% yield), m.p 186-288°C, ^{1}H NMR ($CDCl_3$, 400MHz): δ (ppm) 8.58 (s, 1H), 7.86-7.77 (m, 2H), 7.66 (dd, J = 7.9, 10.2 Hz, 1H), 7.41-7.34 (m, 2H), 7.24-7.19 (m, 2H), 6.95 (d, J = 8.1 Hz, 1H), 5.97 (s, 1H), 3.97 (3H, s); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 186.3, 159.5, 155.2, 148.7, 147.8, 146.8, 145.6, 134.1, 130.0, 127.5, 125.6, 125.0, 124.6, 121.5, 118.7, 116.7, 114.8, 109.9, 56.1; HRMS-ESI: m/z [M+H]+ for $C_{19}H_{15}O_5$, calculated m/z 323.0919; observed 323.0909.

3-{(2E)-3-[3-Methoxy-4-(prop-2-en-1-yloxy)phenyl]prop-2-enoyl}-2H-chromen-2-one (compound 16)

Compound 15 (0.62 mmol) was treated with allyl bromide (0.62 mmol) in presence of K_2CO_3 (0.74 mmol) in DMF at rt. for 6 hours. Reaction mixture was diluted with water and desired compound was obtained by filtration and recrystallization with methanol/DCM and characterized by ^{1}H &^{13}C NMR and HRMS data. Yellow solid (Yield 70%) m.p. 148-151°C, ^{1}H NMR ($CDCl_3$, 400 MHz): δ (ppm) 8.57 (s, 1H), 7.82 (d, *J* = 2.2 Hz, 2H), 7.68-7.63 (m, 2H), 7.40 (d, *J* = 8.2 Hz, 1H), 7.35 (dd, *J* = 7.6, 7.6 Hz, 1H), 7.23 (dd, *J* = 8.3, 2.0 Hz, 1H), 7.20 (d, *J* = 1.9 Hz, 1H), 6.89 (d, *J* = 8.3 Hz, 1H), 6.14-6.04 (m, 1H), 5.43 (dd, *J* = 17.2, 1.4 Hz, 1H), 5.32 (dd, *J* = 10.5, 1.3 Hz, 1H), 4.67 (d, *J* = 5.4 Hz, 2H), 3.94 (s, 3H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ (ppm) 186.3, 159.4, 155.2, 150.8, 149.6, 147.8, 145.3, 134.1, 132.7, 130.0, 128.1, 125.6, 125.0, 123.8, 121.9, 118.6, 118.5, 116.7, 112.9, 110.8, 69.8, 56.0; HRMS-ESI: m/z $[M+H]^+$ for $C_{22}H_{19}O_5$, calculated 363.1232; observed 363.1220.

(E)-3-(3-(3-methoxy-4-(prop-2-yn-1-yloxy)phenyl)acryloyl)-2H-chromen-2-one **(17a)**

Compound **14b** (0.62 mmol) was treated with propargyl bromide (0.62 mmol) in presence of K_2CO_3 (0.74 mmol) at rt. for 6 hours. Reaction mixture was diluted with water and desired compound **17** was obtained by filtration and recrystallization with methanol and characterized by ^1H &^{13}C NMR and HRMS data. Yellow solid (Yield 78%) m.p. 185-187°C, ^1H NMR ($CDCl_3$, 400 MHz): δ (ppm) 8.58 (s, 1H), 7.83 (s, 2H), 7.68-7.64 (m, 2H), 7.40 (d, *J* = 8.2 Hz, 1H), 7.36 (dd, *J* = 6.6, 0.9 Hz, 1H), 7.24 (dd, *J* = 8.3, 2.0 Hz, 1H), 7.22 (d, *J* = 1.9 Hz, 1H), 7.06 (d, *J* = 8.32 Hz, 1H), 4.82 (d, *J* = 2.4 Hz, 2H), 3.94 (s, 3H), 2.55 (t, *J* = 2.4 Hz, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ (ppm) 186.3, 159.4, 155.2, 149.8, 149.4, 147.9, 145.1, 134.2, 130.0, 129.0, 125.5, 125.0, 123.4, 122.4, 118.6, 116.7, 113.6, 110.9, 78.0, 76.3, 56.6, 56.0; HRMS-ESI: m/z [M+H]+ for $C_{22}H_{17}O_5$, calculated 361.1076; observed 361.1074.

(E)-2-methoxy-4-(3-oxo-3-(2-oxo-2H-chromen-3-yl)prop-1-en-1-yl)phenyl 2-chloroacetate **(18).**

Compound **14b** (1.5 mmol) was drop wise treated with 1-chloroacetyl chloride (4.6 mmol) in presence of K_2CO_3 (6.2 mmol) in DMF (4 mL) at rt. for 7-8 hours. Reaction mixture was diluted with water and desired compound was obtained by filtration and recrystallization with methanol and characterized by 1H &^{13}C NMR and HRMS data. Yellow solid (Yield: 67%) m.p. 157-160°C, ^1H NMR ($CDCl_3$, 400 MHz): δ (ppm) 8.61 (s, 1H), 7.92 (d, J = 16.0 Hz, 1H), 7.83 (d, J = 16.0 Hz, 1H), 7.69 (t, J = 8.0 Hz, 1), 7.43-7.37 (m, 2H), 7.31-7.28 (m, 2H), 7.13 (d, J = 8.0 Hz, 1H), 4.37 (s, 2H), 3.92 (s, 3H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ (ppm) 186.3, 165.1, 159.4, 155.2, 151.1, 148.3, 145.6, 144.0, 141.3, 134.4, 130.1, 125.0, 124.4, 122.9, 122.1, 118.5, 116.7, 112.0, 56.0 and 40.5. HRMS-ESI: m/z $[M+H]^+$ for $C_{21}H_{16}ClO_6$, calculated 399.0635; observed 399.0613.

(E)-3-(3-(4-((1-(7-chloroquinolin-4-yl)-1H-1,2,3-triazol-4-yl)methoxy)-3-methoxyphenyl)acryloyl)-2H-chromen-2-one (compound 19)

Compound 17 (0.14 mmol) was treated with 4-azido-7-chloroquinoline (0.14 mmol) in presence of copper sulfate (0.028 mmol), sodium ascorbate (0.036 mmol) in DMF (5 mL) at rt. for 14 hours. Reaction mixture was diluted with water and desired compound was obtained by filtration and recrystallization with methanol and characterized by ^1H &^{13}C NMR and HRMS data. Yellow solid (Yield 55%) m.p. 222-224°C, ^1H NMR ($CDCl_3$, 400 MHz): δ (ppm) 9.07 (d, *J* = 4.6 Hz, 1H), 8.58 (s, 1H), 8.25 (d, *J* = 1.9 Hz, 1H), 8.16 (s, 1H), 7.97 (d, *J* = 9.1 Hz, 1H), 7.83 (s, 2H), 7.69-7.65 (m, 2H), 7.60 (dd, *J* = 9.1, 2.07 Hz, 1H), 7.50 (d, *J* = 4.6 Hz, 1H), 7.41-7.36 (m, 2H), 7.34-7.29 (m, 1H), 7.23 (d, *J* = 1.6 Hz, 1H), 7.15 (d, *J* = 8.3 Hz, 1H), 5.50 (s, 2H), 3.95 (s, 3H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ (ppm) 186.3, 159.5, 155.2, 151.4, 150.2, 150.0, 149.8, 148.0, 144.9, 144.7, 140.8, 137.0, 134.2, 130.0, 129.6, 129.1, 129.0, 125.4, 125.0, 124.9, 124.5, 123.6, 122.5, 120.6, 118.6, 116.7, 116.1, 113.7, 110.9, 62.9, 56.0; HRMS-ESI: m/z $[M+H]^+$ for $C_{31}H_{22}N_4ClO_5$, calculated 565.1279; observed 565.1288.

(E)-2-methoxy-4-(3-oxo-3-(2-oxo-2H-chromen-3-yl)prop-1-en-1-yl)phenyl 2-(piperidin-1-yl)acetate (compound 20)

Compound 18 (0.67 mmol) was treated with piperidine (1.2 equiv.) in DCM in presence of K_2CO_3 (2 equiv.) at rt. for 24 hours. Reaction mixture was concentrated and washed with water and recrystallized in methanol to give compound 20 which was characterized by 1H &^{13}C NMR and HRMS data. Yellow solid (Yield 70%) m.p. 148-151°C, ^{1}H NMR ($CDCl_3$, 400 MHz): δ (ppm) 8.61 (s, 1H), 7.94 (d, J = 16.0 Hz, 1H), 7.83 (d, J = 16.0 Hz, 1H), 7.69 (t, J = 8.0 Hz, 2H), 7.43-7.37 (m, 2H), 7.31-7.28 (m, 2H), 7.13 (d, J = 8.0 Hz, 1H), 3.95 (s, 3H), 3.37 (s, 2H), 2.45 (m, 4H) 1.53-1.59 (m, 6H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ (ppm) 186.6, 159.8, 155.6, 151.2, 149.9, 148.2, 145.7, 134.5, 133.1, 130.2, 128.5, 126.0, 125.3, 124.1, 122.3, 119.0, 118.9, 117.0, 113.2, 111.1, 70.1 and 56.4. HRMS-ESI: m/z $[M+H]^+$ for $C_{26}H_{26}NO_6$, calculated 448.1760; observed 448.1433.

Biology: Materials and Method

Evaluation of antimalarial activity

Chloroquine-sensitive strain 3D7 and multidrug-resistant strain K1 (resistant to chloroquine, sulfadoxine-pyrimethamine, chlorproguanil) of P. falciparum were maintained at 6-8% parasitemia and 2% hematocrit in RPMI complete medium (RPMI 1640 supplemented with HEPES, 0.2% sodium bicarbonate, 0.2% D-glucose, 0.5% albumax, 45 mg/L hypoxanthine, 0.25 mg/L fungi zone and 50mg/L gentamycin) at 37 °C in a humidified CO2 incubator. Antimalarial activity was determined using SYBR Green–I based fluorescence assay (Smilkstein et al., 2004). Chloroquine (C-6628, Sigma) was used as reference drug .Parasite inhibition experiments were conducted at 0.8% parasite maintained at 1% hematocrit in RPMI medium. Ring stage parasites were treated with different dilutions of compounds in a 96-well plate (37ºC, 72 h). Untreated parasite (infected-RBCs) and non-infected RBCs were used positive and negative control, respectively. In parallel, parasite culture was maintained in 60 mm dish without any drug to monitor the parasite growth (37 ºC, 72 h). After the 72 h, 100 µl lysis buffer [20 mM Tris-HCl(pH 7.5), 5 mM EDTA, 0.008% saponin, and 0.08% Triton X-100] containing 2× SYBR Green dye (S7585) was added in each well of 96-well plate and incubated (37ºC, 1 h). The fluorescence was recorded in an FLX800, BIOTEK instrument (excitation at 480 nm, emission at 520 nm). Data was analyzed to obtain inhibitory concentration (IC50) values.

Cytotoxicity was evaluated in VERO cells (C 1008; monkey kidney epithelial cells) using the MTT assay. VERO cells were maintained in RPMI media supplemented with HEPES, 0.2% sodium bicarbonate, 0.2% D-glucose, 10% FBS, fungi zone (0.25 mg/L) and gentamycin (50 mg/L) at 37 °C in a humidified CO2 incubator. VERO cells (104/well) were seeded in a 96 well plate and cells were treated with different dilutions of compounds (16-18 h post-seeding). Podophyllotoxin (P4405, Sigma) was used as the positive control. After 72 h, 25 µl of MTT (M2128, Sigma) (stock 5mg/ml) was added to each well and incubated for 2 h in CO2 incubator. Supernatant was removed carefully without disturbing the cells and 150µl DMSO was added in each well to dissolve the purple precipitate. Absorbance was recorded at 540

nm using ELISA plate reader and data was analyzed to determine 50% cytotoxic concentration (CC50). For microscopic examination, 3D7 was synchronized with 5% sorbitol and then treated with the compound 6 at 10μM concentration. After 24 h and 48 h of treatment, thin blood smears of both control and treated culture were prepared. Smears were fixed and stained with methanol and Giemsa, respectively. In parallel experiment, after 24 h of treatment, the culture was washed (twice) with RPMI media to remove drug and was further incubated without drug to check the revival of the parasite after drug removal.

Declarations

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References

1. Trampuz, A, Jereb, M, Muzlovic, I, Prabhu R. M, Clinical review: Severe malaria. Critical care 2003; 7: 1. https://doi.org/10.1186/cc2183
2. Baird J K, Resistance to chloroquine unhinges vivax malaria therapeutics. Antimicrob. Agents and Chemother. 2011; 55: 1827. https://doi.org/10.1128/AAC.01296-10
3. (a) Yáñez, J A, Remsberg, C M, Miranda N D, Vega-Villa K R, Andrews P K, Davies N M, Pharmacokinetics of selected chiral flavonoids: hesperetin, naringenin and eriodictyol in rats and their content in fruit juices. Biopharm.DrugDispos. 2008; 29: 63. https://doi.org/10.1002/bdd.588 (b) Sharma B, Singh P, Singh A K, Awasthi S K, Advancement of chimeric hybrid drugs to cure malaria infection: An overview with special emphasis on endoperoxides pharmacophores. Eur. J. Med. Chem. 2021; 219: 113408. https://doi.org/10.1016/j.ejmech.2021.113408.
4. Boonlaksiri C, Oonanant W, Kongsaeree P, Kittakoop P, Tanticharoen M, Thebtaranonth Y,An antimalarial stilbene from Artocarpus integer.Phytochem.2000; 54: 415. https://doi.org/10.1016/S0031-9422(00)00074-1.
5. Morphy R, Rankovic Z, Designed multiple ligands: An emerging drug discovery paradigm. J. Med.Chem. 2005; 48: 6523. https://doi.org/10.1021/jm058225d.
6. Muregi F W, Ishih A, Next-generation antimalarial drugs: hybrid molecules as a new strategy in drug design. Drug Dev.Res.2010; 71: 20. https://doi.org/10.1002/ddr.20345.
7. Sharma, N.; Mohanakrishnan, D.; Sharma, U. K.; Kumar, R.; Sinha, A. K.; Sahal, D. Design, Design, economical synthesis and anti-plasmodial evaluation of vanillin derived allylated chalcones and

their marked synergism with artemisinin against chloroquine resistant strains of Plasmodium falciparum. Eur. J. Med. Chem.2014, 79, 350. https://doi.org/10.1016/j.ejmech.2014.03.079.

8. Tripathi R, Mishra R, Dwivedi N, Tewari N, Verma S, Current status of malaria control. Curr.Med.Chem.2005; 12: 2643. https://doi.org/10.2174/092986705774370673.
9. Phillips R Clin, Current status of malaria and potential for control. Microbiol. Rev.2001; 14: 208.
10. Wernsdorfer W, The development and spread of drug-resistant malaria. J. Parasitol. Today 1991; 7: 297. https://doi.org/10.1016/0169-4758(91)90262-M.
11. Garcia Linares, G. E.; Rodriguez, J. B. Current status and progresses made in malaria chemotherapy. Curr.Med.Chem.2007; 14: 289. https://doi.org/10.2174/092986707779941096.
12. Newman, D. J.; Cragg, G. M.; Snader, K. M. Natural Products as Sources of New Drugs over the Period 1981–2002; J. Nat. Pro. 2003, 66, 1022. https://doi.org/10.1021/np030096l
13. Rogers D J, Randolph S E, The global spread of malaria in a future, warmer world. Science2000; 289: 1763. https://doi.org/10.1126/science.289.5485.1763.
14. (a) de Pilla Varotti F, Botelho A C C, Andrade A A, de Paula R C, FagundesE M, Valverde A, Mayer L M, Mendonça J S, de Souza M V, Boechat N, Synthesis, antimalarial activity, and intracellular targets of MEFAS, a new hybrid compound derived from mefloquine and artesunate. Antimicrob. Agents Chemother. 2008; 52: 3868. https://dx.doi.org/10.1128%2FAAC.00510-08. (b) Hernández C, Moreno G, Herrera-R A, Cardona-G W, New Hybrids Based on Curcumin and Resveratrol: Synthesis, Cytotoxicity and Antiproliferative Activity against Colorectal Cancer Cells. Molecule 2021; 26: 2661. https://doi.org/10.3390/molecules26092661. (c) Raiguru P B, Mohapatra S, Nayak S, Sahal D, Yadav M, Acharya N B, Flavone-stilbene hybrids: Synthesis and evaluation as potential antimalarial agents. Eur. J. Med. Chem. 2022; 4: 100029. https://doi.org/10.1016/j.ejmcr.2022.100029.
15. Kouznetsov V V, Gómez-Barrio A. Recent developments in the design and synthesis of hybrid molecules based on aminoquinoline ring and their antiplasmodial evaluation. Eur.J.Med.Chem.2009; 44: 3091. https://doi.org/10.1016/j.ejmech.2009.02.024.
16. de Pilla Varotti F, Botelho A C C, Andrade A A, de Paula R. C, Fagundes E M, Valverde A, Mayer L M, Mendonça J S, de Souza M V, Boechat N,Synthesis, antimalarial activity, and intracellular targets of MEFAS, a new hybrid compound derived from mefloquine and artesunate. Antimicrob. Agents Chemother. 2008; 52: 3868.
17. Sharma N, Mohanakrishnan D, Shard A, Sharma A, Sinha A K, Sahal D,Stilbene–chalcone hybrids: design, synthesis, and evaluation as a new class of antimalarial scaffolds that trigger cell death through stage specific apoptosis. J. Med. Chem.2012; 55: 297. https://doi.org/10.1021/jm201216y.
18. Sharma N, Mohanakrishnan D, Shard A, Sharma A, Sinha A K, Sahal D, Hydroxylated di-and tri-styrylbenzenes, a new class of antiplasmodial agents: discovery and mechanism of action. RSCAdv.2016; 6: 49348. https://doi.org/10.1039/c6ra06059e.
19. Pingaew R, Saekee A, Mandi P, Nantasenamat C, Prachayasittikul S, Ruchirawat S, Prachayasittikul V, Synthesis, biological evaluation and molecular docking of novel chalcone–coumarin hybrids as

anticancer and antimalarial agents. Eur. J. Med. Chem.2014; 85: 65. https://doi.org/10.1016/j.ejmech.2014.07.087.

20. Smilkstein M, Sriwilaijaroen N, Kelly J X, Wilairat P, Riscoe M,Simple and inexpensive fluorescence-based technique for high-throughput antimalarial drug screening. Antimicrob.Agents Chemother.2004; 48: 1803. https://dx.doi.org/10.1128%2FAAC.48.5.1803-1806.2004.
21. Sharma, N.; Mohanakrishnan, D.; Sharma, U. K.; Kumar, R.; Sinha, A. K.; Sahal, D. Design, economical synthesis and antiplasmodial evaluation of vanillin derived allylated chalcones and their marked synergism with artemisinin against chloroquine resistant strains of Plasmodium falciparum. Eur. J. Med. Chem 2014, 79, 350. https://doi.org/10.1016/j.ejmech.2014.03.079
22. Trivedi J C, Bariwal J B, Upadhyay K D, Naliapara Y T, Joshi S K, Pannecouque C C, De Clercq E, Shah A K, Improved and rapid synthesis of new coumarinyl chalcone derivatives and their antiviral activity. Tetrahedron Lett.2007; 48: 8472. https://dx.doi.org/10.1016/j.tetlet.2007.09.175.
23. Gummudavelly S, Ranganath Y, Bhasker S, Rajkumar N,Asian J. Res. Chem.2009; 2: 46.
24. Egan T J, Hunter R, Kaschula C H, Marques H M, Misplon A, Walden J,Structure−Function Relationships in Aminoquinolines: Effect of Amino and Chloro Groups on Quinoline−Hematin Complex Formation, Inhibition of β-Hematin Formation, and Antiplasmodial Activity. J. Med. Chem. 2000; 43: 283. https://doi.org/10.1021/jm990437l.
25. Kaur K, Jain M, Reddy R P, Jain R, Quinolines and structurally related heterocycles as antimalarials. Eur.J.Med.Chem.2010; 45: 3245. https://doi.org/10.1016/j.ejmech.2010.04.011.
26. Qin H.-L, Zhang Z-W, Lekkala R, Alsulami H, Rakesh K P, Chalcone hybrids as privileged scaffolds in antimalarial drug discovery: a key review. Eur. J. Med. Chem.2020; 193: 11221. https://doi.org/10.1016/j.ejmech.2020.112215.

Scheme

Scheme 1-3 are available in Supplementary Files section.

Tables

Table 2 is not available with this version

Figures

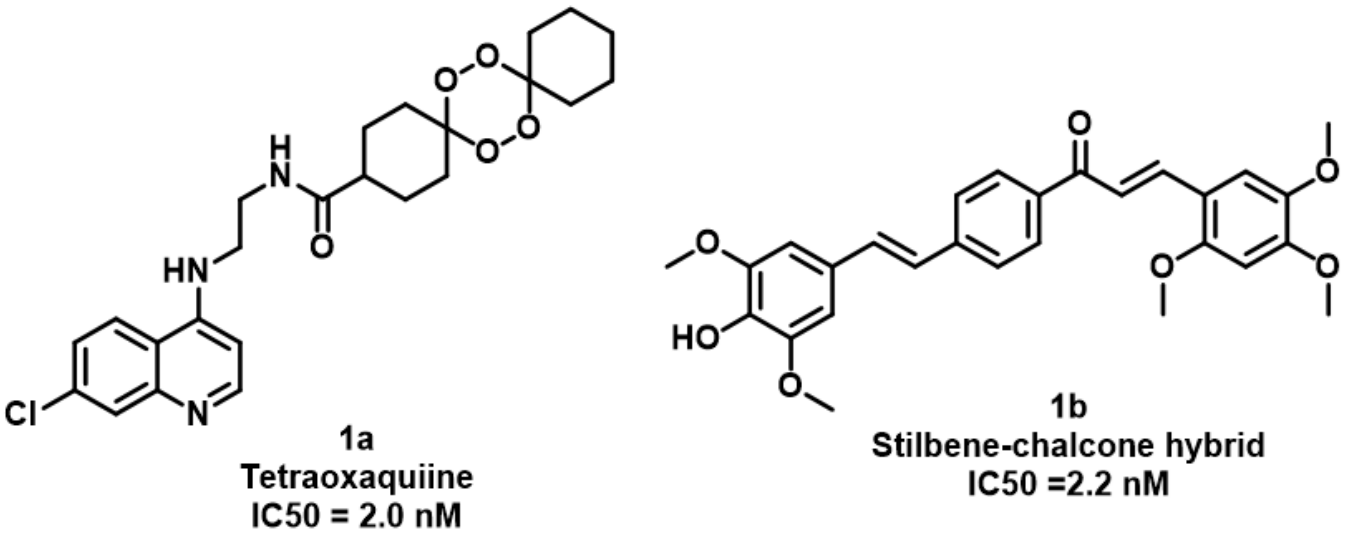

Figure 1

Hybrid molecules showing antimalarial potential.

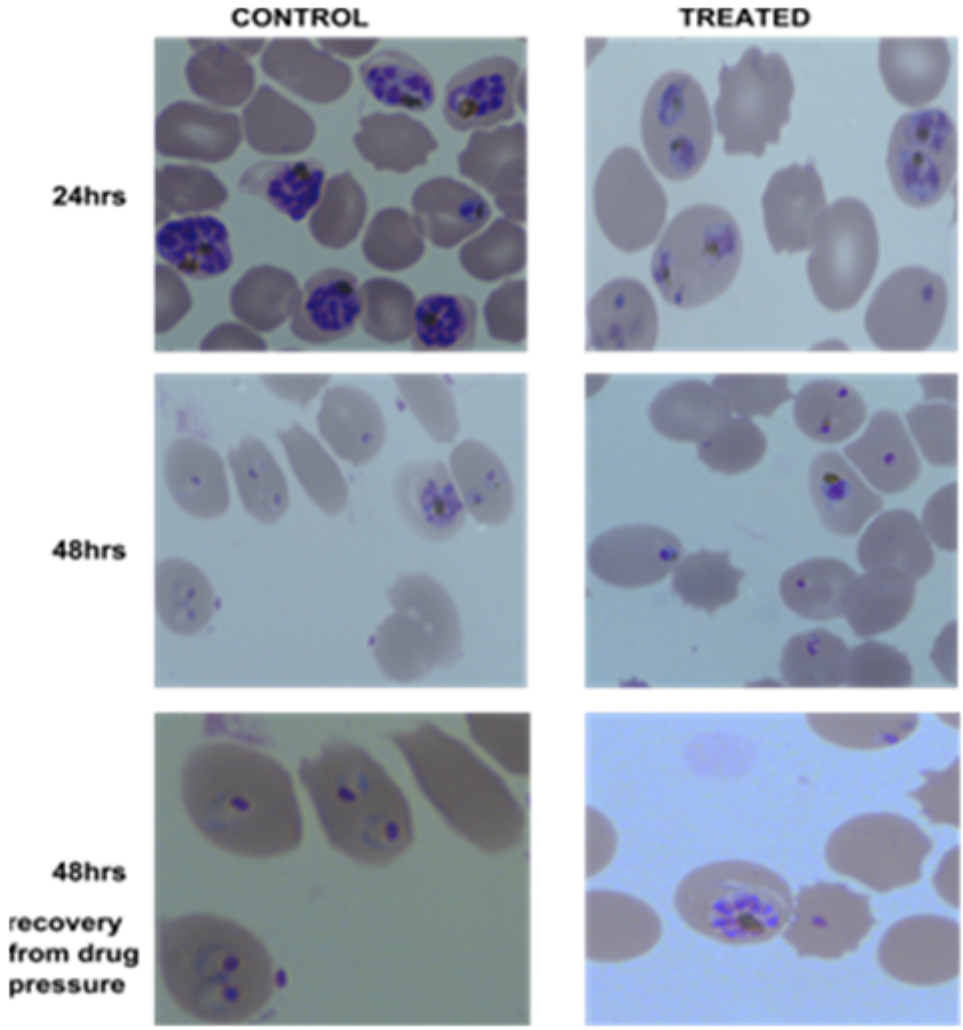

Figure 2

Microscopic examination of Giemsa-stained blood smears of P. falciparum (3D7) treated with 10 μM of compound 3e at 24 h and 48 h. After 24 h, both control and treated sample were washed and parasite was cultured without drug for another 24 h.

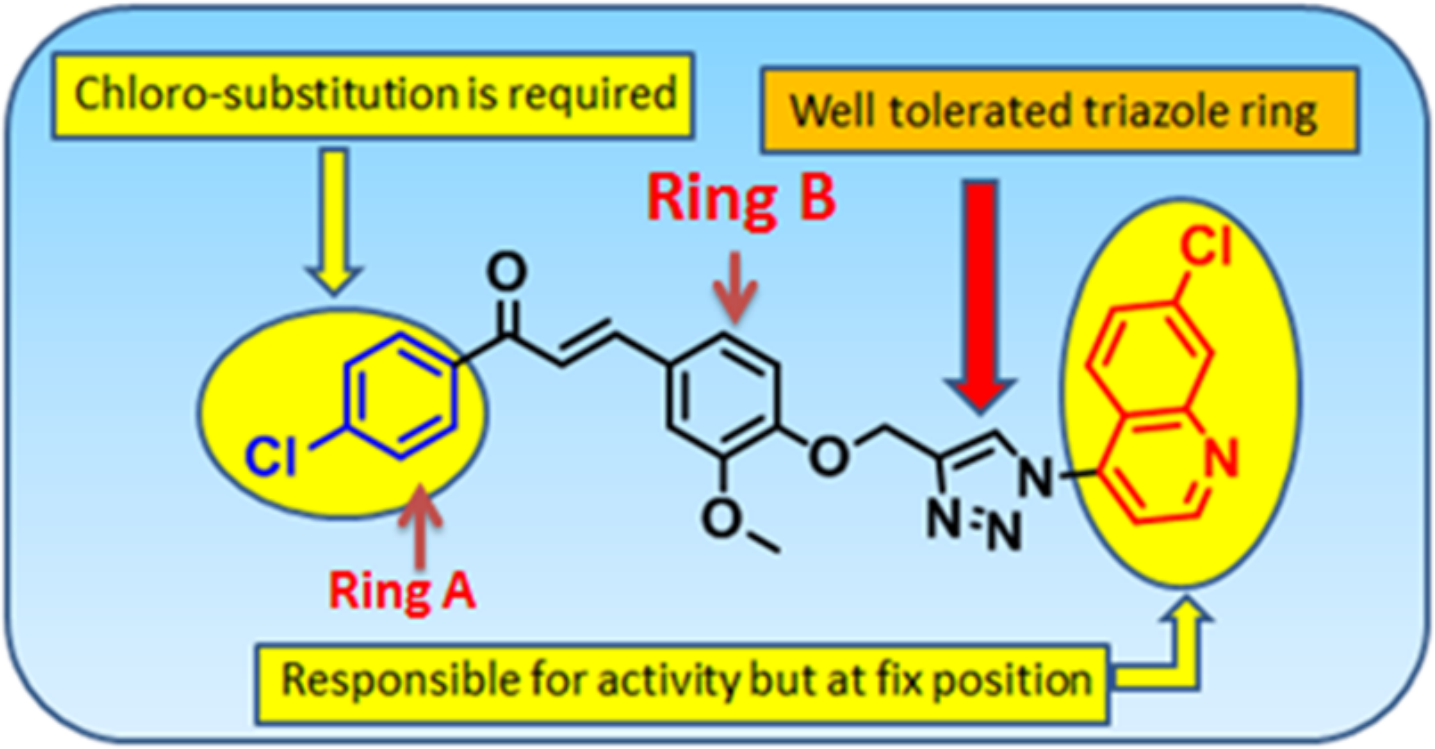

Figure 3

Represents structure activity relationship.

Supplementary Files

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