ARTICLE III

Synthesis, *In Vitro* Antimalarial and Cytotoxicity Of Artemisinin-Aminoquinoline Hybrids

The third article was also published in Bioorganic & Medicinal Chemistry Letters. The article was prepared according to the journal's instructions for authors (see article 3). The NMR spectra of the synthesised compounds of this article can be found in Annexure C.

Bioorganic & Medicinal Chemistry Letters 21 (2011) 1683–1686

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

journal homepage: www.elsevier.com/locate/bmcl

Synthesis, in vitro antimalarial and cytotoxicity of artemisinin-aminoquinoline hybrids

Marli C. Lombard [a], David D. N'Da [a,*], Jaco C. Breytenbach [a], Peter J. Smith [b], Carmen A. Lategan [b]

[a] Department of Pharmaceutical Chemistry, North-West University, Potchefstroom 2520, South Africa
[b] Department of Pharmacology, University of Cape Town, Groote Schuur Hospital, Observatory 7925, South Africa

ARTICLE INFO

Article history:
Received 7 October 2010
Revised 21 January 2011
Accepted 21 January 2011
Available online 27 January 2011

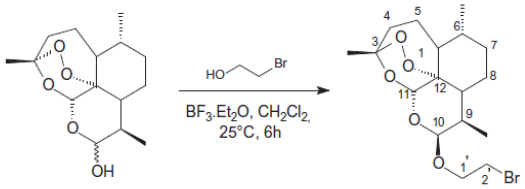
Keywords:
Artemisinin
Malaria
Hybrids
Quinoline

ABSTRACT

Dihydroartemisinin (DHA) was coupled to different aminoquinoline moieties forming hybrids **9–14**, which were then treated with oxalic acid to form oxalate salts (**9a–14a**). Compounds **9a**, **10a**, **12**, **12a**, and **14a** showed comparable potency in vitro to that of chloroquine (CQ) against the chloroquine sensitive (CQS) strain, and were found to be more potent against the chloroquine resistant CQR strain. Hybrids **12** and its oxalate salt **12a** were the most active against CQR strain, being 9- and 7-fold more active than CQ, respectively (17.12 nM; 20.76 nM vs 157.9 nM). An optimum chain length was identified having 2 or 3 Cs with or without an extra methylene substituent.

An emerging strategy within medicinal chemistry and drug discovery is the combination of two distinct pharmacophores into a single molecule. Hybrid molecules offer a simpler and more effective way to deliver these agents, especially when differences like elimination times occur.[1] The underlying mechanism behind the therapeutic effectiveness of hybrid molecules is that the artemisinin derivative are active on the young erythrocytic stages of *Plasmodium falciparum* and the chloroquine derivative is able to inhibit the polymerization of β-hematin.[2] Walsh et al. showed that a hybrid, artemisinin covalently linked to quinine via an ester linkage, had superior activity to that of artemisinin alone, quinine alone, or a 1:1 mixture of artemisinin and quinine. Given the lability of the ester linkage especially in vivo, it is expected that an ether/amine bond will be more stable.[3] The hybrid salt derived from artesunate and mefloquine (MEFAS) also showed to be more effective than the combination of the two antimalarial drugs and in addition exhibited lower toxicity against HepG2 hepatoma cells.[4] Dual molecules containing a trioxane moiety linked to an aminoquinoline entity showed efficient antimalarial activity without recrudescence.[5] A trioxaquine series (1,2,4-trioxolanes linked to quinoline) designed to incorporate the metabolically stable C-10 carbon linkage, showed that an increase in the linker length reduced the activity and that an additional protonation site within the hybrid drug has little impact on antimalarial activity.[6]

Although the mechanism of action of artemisinin is still being deciphered, Paitayatat et al.[7] showed that a number of artemisinin derivatives strongly interact with ferroprotoporphyrin IX. It is known that the artemisinin derivatives, artemether and arteether, are rapidly converted to DHA (**1**).[8]



1 2

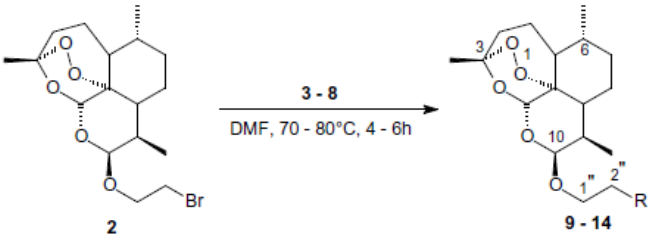
Scheme 1. Synthesis of 2-(10β-dihydroartemisinoxy) ethylbromide (**2**).

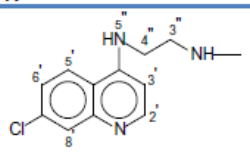
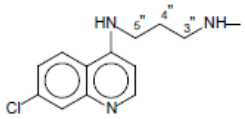
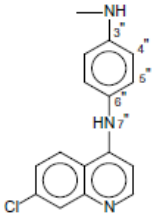
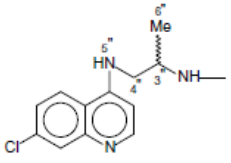
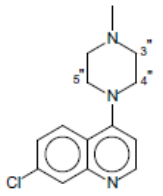
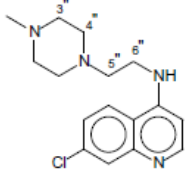
In this study, DHA (**1**) was coupled via an ether bond with an aminoquinoline entity, with the aim to prolong the half life of DHA by linking it with an ether/amine bond which is expected to be more stable than an ester bond, especially when going through the GI-track whereas an ether and amide will not be targeted by the esterase enzyme. It is also expected that these hybrids will be metabolized by the liver enzymes to release both DHA (**1**) and quinoline, giving rise to duel action. Hereby these novel artemisinin-quinoline hybrids synthesized (**13–19**) are anticipated to possess increased activity and prolonged half life, to avoid drug–drug interaction, and improve patient compliance.

The reaction of DHA (**1**) with bromoethanol in the presence of boron trifluoride etherate yielded **2** (Scheme 1),[9] which was converted to **9–14** (Scheme 2)[10] by treatment with various

* Corresponding author. Tel./fax: +27 18 299 2516.
E-mail address: david.nda@nwu.ac.za (D.D. N'Da).

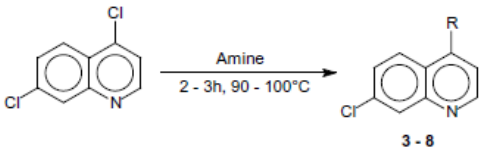
0960-894X/$ - see front matter © 2011 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2011.01.103

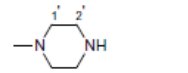
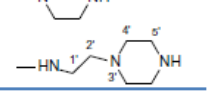


	R		R
9		10	
11		12	
13		14	

Scheme 2. Synthesis of DHA-quinoline hybrids (**9–14**).

quinoline based primary/secondary amines (**3–8**). These aminoquinolines (**3–8**) were obtained by the condensation of various diamines with 4,7-dichloroquinoline (Scheme 3).[11–13]



	R
3	$-NH(CH_2)_2NH_2$
4	$-NH(CH_2)_3NH_2$
5	$-NHC_6H_4NH_2$
6	$-NHCH_2CH(CH_3)NH_2$
7	
8	

Scheme 3. Synthesis of amino functionalized quinoline intermediates (**3–8**).

Only a few of the pure free base DHA-quinoline hybrids were solids, the rest were yellowish/brownish oils. These compounds were treated with oxalic acid to obtain the oxalate salts (**9a–12a**, and **14a**), primarily for solubility and stability reasons.

DHA was supplied as a mixture of epimers, but with the formation of 2-(10-dihydroartemisinoxy)ethylbromide (**2**) only the β isomer was obtained, therefore all synthesized hybrids were β isomers and were tested as such. This assignment was confirmed by X-ray analysis for compound **2**.[14] This configuration was also indicated by the small coupling constant ($J \approx 3$–4 Hz) between H-9 and H-10 in the ^{1}H NMR spectra. The OR-group at C-10 is *cis* to the CH_3-group at C-9 and ring D is in a chair conformation in contrast with that reported by Flippen-Anderson et al.[15] The two methylene protons on the carbon atom (C-1′) adjacent to the new ether oxygen atom are nonequivalent, due to the proximity to several asymmetric carbon centers of the artemisinin nucleus.[9]

In vitro antiplasmodial activity was determined against the chloroquine sensitive D10 strain and chloroquine resistant Dd2 strain of *P. falciparum* using a well established method.[16–18] In vitro cytotoxicity was conducted against a mammalian cell-line, Chinese Hamster Ovarian (CHO) using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazoliumbromide (MTT)-assay, using emetine as the reference.[16,18,19] Two reference antimalarial drugs, viz. CQ and DHA, were tested alongside the hybrids and their salts. The results showed that compounds **9a**, **10a**, **12**,

Table 1
In vitro antiplasmodial activity and cytotoxicity of synthesized artemisinin hybrids

Compound	D10: IC_{50} (nM)	SD	Dd2: IC_{50} (nM)	SD	RI	CHO: IC_{50} (μM)	SD	SI
9	84.59	2.22	152.80	2.06	2	33.04	3.24	391
9a	21.49	0.13	25.70	1.09	1	1.64	0.08	77
10	117.76	16.27	183.49	5.68	2	37.34	14.09	317
10a	14.27	2.65	19.75	0.25	1	0.17	0.01	12
11	30.39	2.71	69.21	2.02	2	ND	ND	ND
11a	17.25	1.06	30.22	12.22	2	35.18	20.14	2039
12[a]	12.18	1.21	17.12	0.44	1	3.39	0.83	279
12a	14.94	0.05	20.76	3.61	1	2.75	0.12	184
13[a]	30.72	1.85	68.49	4.19	2	5.92	0.88	193
14[a]	201.38	19.92	275.99	70.52	1	ND	ND	ND
14a	28.99	2.70	29.24	3.26	1	2.32	0.10	80
CQ (n = 3)	21.54	6.73	157.90	52.70	7	ND	ND	ND
DHA (n = 4)	5.11	0.64	2.09	0.33	0.4	ND	ND	ND
Emetine (n = 3)	ND	ND	ND	ND	ND	0.19	0.05	ND

[a] Tested as a suspension. n = Number of data sets averaged. Resistance index (RI) = IC_{50} Dd2/IC_{50} D10. Selectivity index (SI) = IC_{50} CHO/ IC_{50} D10. CHO = Chinese Hamster Ovarian. SD = standard deviation. ND = not determined.

12a, and **14a** displayed the best antimalarial activity (Table 1). These compounds showed comparable potency to CQ against the CQS strain, D10, and were found more potent than CQ (IC_{50} = 157.9 nM) against the CQR strain, Dd2, of *P. falciparum*, with IC_{50} ranging from 17.2 to 38.9 nM. Compounds **12** and **12a** displayed the best profile based on both antiplasmodial activity and cytotoxicity. Compounds **11**, **11a** and **13** showed good activity against the CQS strain, but were less active against the CQR strain of *P. falciparum*—indicated by the resistance index RI ⩾ 2. Compounds **12**, **13**, and **14** were tested as suspensions in DMSO, due to insolubility in the medium.

Overall the oxalates had better antiplasmodial activity than their free base hybrids, presumably due to their better aqueous solubility in the testing medium. Oxalic acid could also inherit antimalarial activity, but also add to the toxicity of a compound. Slight cytotoxicity was observed with the oxalate salt of compound **10** and **10a** with a selectivity index (SI) of 12, thus making it the most cytotoxic compound in this series. All other compounds showed good selectivity towards *P. falciparum* (SI ⩾ 20). The good activity of some of these compounds against the CQR strain is in agreement with the results from previous studies.[5,20–22] All compounds were less active than the antimalarial drug DHA irrespective of the *P. falciparum* strain making this a major drawback, but also merits further very essential investigation especially on how these hybrids will act in the body whereas the underlying motivation for a hybrid would come forth.

In conclusion, all of the compounds synthesized showed either higher or comparable potency to that of CQ, with the exception of hybrids **10** and **14** which were found with lower potency than CQ against the CQR strain of *P. falciparum*. Hybrid **12** and its oxalate salt (**12a**) possessed the highest antimalarial activity, even though hybrid **12** was tested as a suspension. These two compounds, respectively, showed 9- and 7-fold higher activity than CQ against CQR. The optimum linker could be identified based on the in vitro activity, as those hybrids inheriting a C-chain with 2/3 Cs and a C-chain with 3 Cs with a Me-substituent which is present in compound **9**, **10** and **12**. Therefore it could be deduced that no cyclic linkers should be included, only chains with 2/3 carbon atoms with or without Me-substituent.

Acknowledgements

The authors would like to thank the following: National Research Foundation (funding), North-West University (funding), Cipla Medpro (sample of primaquine), André Joubert (NMR) and Marelize Ferreira (MS). Special thanks to Drs. Attie Viljoen and Arina Lourens for their help and support.

References and notes

1. Kaur, K.; Jain, M.; Reddy, R. P.; Jain, R. L. *Eur. J. Med. Chem.* **2010**, *45*, 3245.
2. Meunier, B. *Acc. Chem. Res.* **2008**, *41*, 69.
3. Walsh, J. J.; Coughlan, D.; Heneghan, N.; Gaynor, C.; Bell, A. *Bioorg. Med. Chem. Lett.* **2007**, *17*, 3599.
4. Varotti, F. d. P.; Botelho, A. C. C.; Andrade, A. A.; De Paula, R. C.; Fagundes, E. M. S.; Valverde, A.; Mayer, L. M. U.; Mendonca, J. S.; De Souza, M. V. N.; Boechat, N.; Krettli, A. U. *Antimicrob. Agents Chemother.* **2008**, *52*, 3868.
5. Dechy-Cabaret, O.; Benoit-Vical, F.; Loup, C.; Robert, A.; Gornitzka, H.; Bonhoure, A.; Vial, H.; Magnaval, J.; Séguéla, J.; Meunier, B. *Chem. Eur. J.* **2004**, *10*, 1625.
6. Araújo, N. C. P.; Barton, V.; Jones, M.; Stocks, P. A.; Ward, S. A.; Davies, J.; Bray, P. G.; Shone, A. E.; Cristiano, M. L. S.; O'Neill, P. M. *Bioorg. Med. Chem. Lett.* **2009**, *19*, 2038.
7. Paitayatat, S.; Tarnchompoo, B.; Thebtaranonth, Y.; Yuthavong, Y. *J. Med. Chem.* **1997**, *40*, 633.
8. Gracе, J. M.; Aguilar, A. J.; Trotman, K. M.; Brewer, T. G. *Drug Metab. Dispos.* **1998**, *26*, 313.
9. Li, Y.; Zhu, Y.; Jiang, H.; Pan, J.; Wu, G.; Wu, J.; Shi, Y.; Yang, J.; Wu, B. *J. Med. Chem.* **2000**, *43*, 1635.
10. *General procedure for the synthesis of* **9–14**. The method as described by Li et al. was used. Particular attention was paid that heating temperature did not exceed 80 °C which could lead to disubstitution. The residue was extracted with CH_2Cl_2 and if any insolubility occurred, it was filtered off. Only the relevant peaks for hybrid **10–14** were notated and as the NMR data for the oxalate salts were a repetition to that of the free bases only the latter's were reported. In the MS spectra of compounds **9–14** the presence of one chlorine atom can be deduced by the presence of two peaks in a 3:1 ratio separated by 2 mass units. *1-(7-Chloro-4-(1,2-diaminoethyl)quinolyl)-2-(10β-dihydroartemisinoxy)ethane* (**9**). Yellowish oil. R_f = 0.46 (DCM/MeOH 9:1). Yield: 59%. δ_H ($CDCl_3$) 8.44 (d, J = 5.3 Hz, 1H, H-2′), 7.88 (d, J = 1.6 Hz, 1H, H-8′), 7.69 (d, J = 8.9 Hz, 1H, H-5′), 7.28 (dd, J = 8.9, 1.9 Hz, 1H, H-6′), 6.32 (d, J = 5.3 Hz, 1H, H-3′), 6.02–5.90 (br s, 1H, H-5″), 5.33 (s, 1H, H-12), 4.76 (d, J = 3.3 Hz, 1H, H-10), 3.92 (dt, J = 10.1, 5.0 Hz, 1H, H-1″α), 3.57–3.45 (m, 1H, H-1″β), 3.32–3.23 (m, 2H, H-4″), 3.00 (t, J = 5.6 Hz, 2H, H-3″), 2.87–2.75 (m, 2H, H-2″), 2.58 (dt, J = 7.7, 5.7 Hz, 1H, H-9), 1.38 (s, J = 12.9 Hz, 3H, 3-Me), 0.82 (d; J = 7.4 Hz, 3H, 9-Me), 0.80 (d; J = 5.5 Hz, 3H, 6-Me). δ_C ($CDCl_3$) 151.79 (C-2′), 149.87 (C-4′), 148.83 (C-8a′), 134.74 (C-7′), 128.34 (C-8′), 125.17 (C-6′), 121.39 (C-5′), 117.24 (C-4a′), 104.05 (C-3), 102.00 (C-10), 99.01 (C-3′), 87.77 (C-12), 80.84 (C-12a), 67.58 (C-1″), 52.30 (C-5a), 48.55 (C-2″), 47.11 (C-3″), 43.90 (C-8a), 42.00 (C-4″), 37.37 (C-6), 36.23 (C-4), 34.36 (C-7), 30.71 (C-9), 26.05 (3-Me), 24.48 (C-5), 24.42 (C-8), 20.15 (6-Me), 12.92 (9-Me). MS: m/z: 532 (M^+, 100%), 534 (M^++2, 30%). *1-(7-Chloro-4-(1,3-diaminopropyl)quinolyl)-2-(10β-dihydroartemisinoxy)ethane* (**10**). Yellow oil. R_f = 0.49 (DCM/MeOH 9:1). Yield: 35%. δ_H ($CDCl_3$) 5.38 (s, 1H, H-12), 4.84 (d, J = 3.4 Hz, 1H, H-10), 4.03 (dt, J = 10.4, 5.2 Hz, 1H, H-1α″), 3.60 (dt, J = 10.3, 5.1 Hz, 1H, H-1β″), 3.38 (t, J = 5.9 Hz, 2H, H-5″), 2.94 (t, J = 5.4 Hz, 2H, H-3″), 2.87 (t, J = 5.2 Hz, 2H, H-2″), 1.91 (dt, J = 11.2, 5.6 Hz, 2H, H-4″), 1.42 (s, 3H, 3-Me), 0.88 (d, J = 7.4 Hz, 3H, 9-Me), 0.87 (d, J = 5.8 Hz, 3H, 6-Me). δ_C ($CDCl_3$) 67.79 (C-1″), 49.19 (C-2″; C-3″), 43.88 (C-5″), 27.11 (C-4″). MS: m/z: 546 (M^+, 100%), 548 (M^++2, 30%). *1-(7-Chloro-4-(1,4-diaminophenyl)quinolyl)-2-(10β-dihydroartemisinoxy)ethane* (**11**). Brownish oil. R_f = 0.57 (DCM/MeOH 9:1). Yield: 35%. δ_H ($CDCl_3$) 7.08 (d, J = 8.6 Hz, 2H, H-4″), 6.78 (s, 1H, H-7″), 6.64 (d, J = 8.7 Hz, 2H, H-5′), 5.36 (s, 1H, H-12), 4.81 (d, J = 3.4 Hz, 1H, H-10), 4.01 (ddd, J = 10.3, 6.2, 4.1 Hz, 1H, H-1α″), 3.64 (ddd, J = 10.4, 6.5, 4.1 Hz, 1H, H-1β″), 3.38–3.25 (m, 2H, H-2″), 1.41 (s, 3H, 3-Me), 0.92 (s, 3H, 9-Me), 0.91 (d, J = 1.8 Hz, 3H, 6-Me). δ_C ($CDCl_3$) 146.45 (C-3″), 128.67 (C-6″), 126.64 (C-4′), 113.82 (C-5″), 66.95 (C-1″), 43.85 (C-2″). MS: m/z: 580 (M^+, 100%), 582 (M^++2, 30%). *1-(7-Chloro-4-(1,2-diaminopropyl)quinolyl)-2-(10β-dihydroartemisinoxy)ethane* (**12**). Unable to separate the two isomers, was therefore collected as a mixture. Fluffy light brown crystals. Mp: 67 °C R_f = 0.42 (DCM/MeOH 9:1). Yield: 45%. δ_H ($CDCl_3$) 5.99 (s, 1H, H-5″), 5.75 (dd, J = 36.6, 5.9 Hz, 1H, H-5″), 5.37–5.33 (m, 1H, H-12), 5.29 (s, 1H, H-12), 4.78 (dd, J = 7.4, 4.1 Hz, 1H, H-10), 4.76 (d, J = 3.4 Hz, 1H, H-10), 3.96 (ddd, J = 10.3, 7.1, 3.8 Hz, 1H, H-1α′), 3.90 (ddd, J = 10.5, 6.4, 4.3 Hz, 1H, H-1α″),

3.78 (dt, J = 11.8, 5.8 Hz, 1H, H-3″), 3.73 (dt, J = 11.6, 5.6 Hz, 1H, H-4′), 3.57–3.51 (m, 1H, H-1β″), 3.48 (ddd, J = 10.2, 6.6, 3.8 Hz, 1H, H-1β″), 3.30 (t, J = 15.4 Hz, 1H, H-4α″), 3.10 (tt, J = 12.9, 6.4 Hz, 1H, H-3″), 3.05–2.96 (m, 1H, H-4β″), 2.96–2.75 (m, 4H, H-2″), 1.40 (s, 3H, 3-Me), 1.38 (s, 3H, 3-Me), 1.27 (dd, J = 6.3, 4.0 Hz, 3H, H-6″), 1.20 (dd, J = 6.3, 3.5 Hz, 1H, H- 6″), 0.84–0.80 (m, 6H, 9-Me), 0.79 (dd, J = 6.8, 2.8 Hz, 6H, 6-Me). δ_C ($CDCl_3$) 67.80 (C-1″), 67.57 (C-1″), 54.12 (C-3″), 53.95 (C-3″), 49.28 (C-2″), 49.23 (C-2′), 47.59 (C-4″), 47.43 (C-4″), 18.06 (C-6′), 17.95 (C-6″). MS: m/z: 500 ($M^+-CH_2O_2$), 546 (M^+, 100%), 548 (M^++2, 30%). *1-(7-Chloro-4-(piperazin-1-yl)quinolyl)-2-(10β-dihydroartemisinoxy)ethane* (**13**). Fluffy light yellow crystals. Mp: 77 °C. R_f = 0.72 (DCM/MeOH 9:1). Yield: 39%. δ_H ($CDCl_3$) 5.47 (s, 1H, H-12), 4.82 (d, J = 3.3 Hz, 1H, H-10), 4.02–3.95 (m, 1H, H-1α″), 3.62–3.55 (m, 1H, H-1β″), 3.24–3.14 (m, 4H, H-3″), 2.84–2.77 (m, 2H, H-4″), 2.77–2.72 (m, J = 12.5, 6.8 Hz, 2H, H-5″), 2.71–2.65 (m, 2H, H-2″), 1.41 (s, 3H, 3-Me), 0.93 (d, J = 6.4 Hz, 3H, 9-Me), 0.90 (d, J = 7.4 Hz, 3H, 6-Me). δ_C ($CDCl_3$) 65.83 (C-1″), 57.86 (C-2″), 53.28 (C-3″), 52.26 (C-4″,C-5″). MS: m/z: 512 ($M^+-CH_2O_2$), 558 (M^+, 100%), 560 (M^++2, 30%). *1-(7-Chloro-4-(2-piperazinyl-ethylamino)quinolyl)-2-(10β-dihydroartemisinoxy) ethane* (**14**). Dark yellow oil. R_f = 0.51 (DCM/MeOH 9:1). Yield: 52% δ_H ($CDCl_3$) 5.33 (s, 1H, H-12), 4.75 (d, J = 3.0 Hz, 1H, H-10), 3.90 (dt, J = 9.9, 4.8 Hz, 1H, H-1α′), 3.48 (td, J = 10.8, 5.3 Hz, 1H, H-1β″), 3.18–3.10 (br s, 4H, H-3″), 2.76 (t, J = 5.1 Hz, 2H, H-2″), 2.72 (t, J = 5.9 Hz, 2H, H-5″), 2.66 (s, 4H, H-4′), 2.56 (t, J = 5.8 Hz, 2H, H-6″), 1.34 (s, 3H, 3-Me), 0.85 (s, 3H, 9-Me), 0.84 (s, 3H, 6-Me). δ_C ($CDCl_3$) 67.59 (C-1″), 57.65 (C-2″), 52.96 (C-3″), 52.00 (C-4″), 49.16 (C-5″), 46.05 (C-6″). MS: m/z: 555 ($M^+-CH_2O_2$), 601 (M^+, 100%), 603 (M^++2, 30%).

11. *Condensation of diamines with 4,7-dichloroquinoline* (**3**–**8**). A combined method as described by Biot et al. and N'Da et al. was used The residue was recrystallized form ethyl acetate. High yields were obtained (75–85%). *7-Chloro-4-(1,2-diaminoethyl) quinoline* (**3**). Off white crystals. δ_H (CD_3OD) 8.35 (d, J = 5.6 Hz, 1H, H-2), 8.10 (d, J = 9.0 Hz, 1H, H-5), 7.76 (d, J = 2.1 Hz, 1H, H-8), 7.39 (dd, J = 9.0, 2.1 Hz, 1H, H-6), 6.55 (d, J = 5.6 Hz, 1H, H-3), 3.43 (t, J = 6.4 Hz, 2H, H-1′), 2.97 (t, J = 6.4 Hz, 2H, H-2′). δ_C (CD_3OD) 152.83 (C-4), 152.47 (C-2), 149.68 (C-8a), 136.34 (C-7), 127.60 (C-5), 126.03 (C-6), 124.34 (C-8), 118.81 (C-4a), 99.70 (C-3), 46.34 (C-1′), 40.84 (C-2′). *7-Chloro-4-(1,3-diaminopropyl) quinoline* (**4**). Yellow white crystals. δ_H (CD_3OD) 3.40 (t, J = 7.0 Hz, 2H, H-3′), 2.79 (t, J = 7.1 Hz, 2H, H-1′), 1.86–1.92 (m, J = 7.0 Hz, 2H, H-2′). δ_C (CD_3OD) 41.69 (C-1′), 40.25 (C-3′), 32.02 (C-2′). *7-Chloro-4-(1,4-diaminophenyl) quinoline* (**5**). Dark brown crystals. δ_H (CD_3OD) 7.08 (d, J = 8.5 Hz, 2H, H-2′), 6.81 (d, J = 8.6 Hz, 2H, H-3′). δ_C (CD_3OD) 147.30 (C-1′), 130.83 (C-4′), 127.61 (C-2′), 117.30 (C-3′). *7-Chloro-4-(1,2-diaminopropyl) quinoline* (**6**). Off white crystals. δ_H (CD_3OD) 3.27 (ddd, J = 7.9, 5.8, 3.1 Hz, 2H, H-1′), 3.25 – 3.21 (m, 1H, H-2′), 1.19 (d, J = 5.7 Hz, 3H, H-3′). δ_C (CD_3OD) 51.77 (C-1′), 46.49 (C-2′), 21.09 (C-3′). *7-Chloro-4-(piperazin-1-yl) quinoline* (**7**). Off white crystals. δ_H (CD_3OD) 3.24–3.20 (m, 4H, H-1′), 3.12–3.08 (m, 4H, H-2′). δ_C (CD_3OD) 54.03 (C-1′), 46.40 (C-2′). *7-Chloro-4-(2-piperazinyl-ethylamino) quinoline* (**8**). Brown white crystals. δ_H (CD_3OD) 3.27 (s, 4H, H-4′), 2.81 (t, J = 6.5 Hz, 2H, H-2′), 2.76 (s, 4H, H-5′), 2.57 (t, J = 6.5 Hz, 2H, H-1′). δ_C (CD_3OD) 61.30 (C-2′), 54.16 (C-4′), 53.08 (C-5′), 39.01 (C-1′).

12. Biot, C.; Daher, W.; Ndiaye, C. M.; Melnyk, P.; Pradines, B.; Chavain, N.; Pellet, A.; Fraisse, L.; Pelinski, L.; Jarry, C.; Brocard, J.; Khalife, J.; Forfar-Bares, I.; Dive, D. *J. Med. Chem.* **2006**, *49*, 4707.
13. N'Da, D. D.; Breytenbach, J. C.; Smith, P. J.; Lategan, C. *Arzneim. Forsch–Drug Res.* **2010**, *60*, 627.
14. Lombard, M. C.; Fernandes, M. A.; Breytenbach, J. C.; N'Da, D. D. *Acta Crystallogr., Sect. E* **2010**, *E66*, 2182.
15. Luo, X.; Yeh, H. J. C.; Brossi, A.; Flippen-Anderson, J. L.; Gilardi, R. *Helv. Chim. Acta* **1984**, *67*, 1515.
16. Pillay, P.; Vleggaar, R.; Maharaj, V. J.; Smith, P. J.; Lategan, C. A. *J. Ethnopharmacol.* **2007**, *112*, 71.
17. Makler, M. T.; Ries, J. M.; Williams, J. A.; Bancroft, J. E.; Peper, R. C.; Gibbins, B. L.; Hinrichs, D. J. *Am. Soc. Trop. Med. Hyg.* **1993**, *48*, 739.
18. Trager, W.; Jensen, J. B. *Science* **1976**, *193*, 673.
19. Mosmann, T. *J. Immunol. Methods* **1983**, *65*, 55.
20. Grelepois, F.; Grellier, P.; Bonnet-Delpon, D.; Begue, J. *Chembiochem* **2005**, 648.
21. Bellot, F.; Cosledan, F.; Vendier, L.; Brocard, J.; Meunier, B.; Robert, A. *J. Med. Chem.* **2010**, *53*, 4103.
22. Dechy-Dabaret, O.; Benoit-Vical, F.; Robert, A.; Meunier, B. *Chembiochem* **2000**, 281.