Pergamon

Bioorganic & Medicinal Chemistry Letters 9 (1999) 2969–2972

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

STRUCTURE AND ANTIMALARIAL ACTIVITY OF ADDUCTS OF 11-AZAARTEMISININ WITH CONJUGATED TERMINAL ACETYLENES

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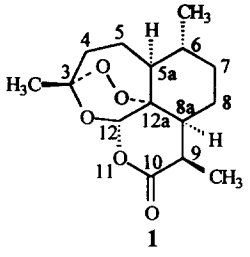
Received 7 July 1999; accepted 8 September 1999

Abstract: Several N-substituted 11-azaartemisinins were prepared from 11-azaartemisinin in high yield by the DMAP catalyzed addition of terminal acetylenes conjugated with electron-withdrawing groups. Their antimalarial activities against two drug-resistant strains of *Plasmodium falciparum* were determined.

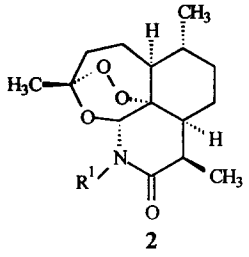
The discovery by Chinese investigators of artemisinin (**1**) a sesquiterpene lactone endoperoxide which was an effective antimalarial against drug-resistant strains of *Plasmodium falciparum*, stimulated a host of studies to prepare more active drugs[1] and to investigate their mechanism of action.[2] As part of a program to prepare new, more active artemisinin derivatives, we synthesized[3] 11-azaartemisinin, **2a** and a series of N-substituted 11-azaartemisinins, e.g. **2b**. by reacting a variety of alkyl, aryl and heterocyclic amines with **1** followed by an acid catalyzed cyclization. Also, Avery et al.[4] prepared and tested a series of N-substituted 11-aza-9-desmethylartemisinin derivatives. The two studies[3,4] established that replacing the lactone oxygen in the artemisinin structure by a nitrogen does not reduce their antimalarial activities and in fact several of the compounds are more active than artemisinin. In vitro and in vivo studies reveal that the presence of a polar substituent on the β-carbon (e.g., **2c**) significantly increases the molecule's activity. In another study, we employed **2a** as a starting material in base-catalyzed reactions with olefins conjugated to electron-withdrawing groups (EWG).[5] We sought to prepare a cyclic derivative containing a carbonyl group by the addition of α,β-unsaturated cyclic ketones. Unfortunately, we were consistently unable to obtain the desired products. As an alternative, we decided to rigidify the structure by introducing a double bond in the substituent

0960-894X/99/$ - see front matter
PII: S0960-894X(99)00506-5

between the polar group and the lactam nitrogen. We wish to report the syntheses and antimalarial activities of those derivatives against two drug-resistant clones of *Plasmodium falciparum*.



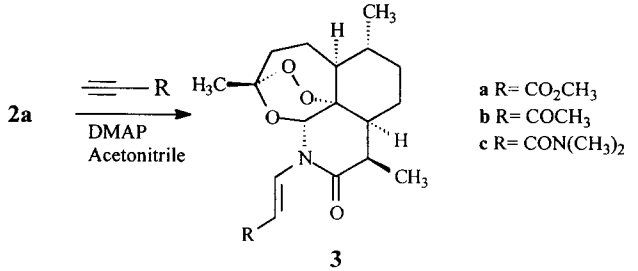
1



2

a R^1 = H
b R^1 = alkyl, aryl, or heterocyclic substituents
c R^1 = CH_2CHO

There are relatively few reports of the addition of amides to terminal acetylenes conjugated to EWGs. Johnson et al.[6] identified and synthesized propargyl aldehyde adducts of each of the purine and pyrimidine bases obtained by bleomycin-induced strand-scission of DNA. The adducts, which possessed an E-sterochemistry about the double bond, were prepared using triethylamine-catalyzed additions to propargyl aldehyde. Faja et al.[7] recently reported the use of DMAP (dimethylaminopyridine) catalyzed additions of methyl propynoate to the amides of several uridines and thymidines. The latter report as well as the cytotoxic activity of the purine and pyrimidine adducts described by Johnson et al.[6] prompted us to investigate the reaction of **2a** with methyl propiolate[8], 3-butyn-2-one[8] and dimethylpropiolamide[9] as well as the antimalarial activity of the resulting adducts. While reaction of **2a**, methyl propiolate and DMAP in acetonitrile proceeded rapidly at room temprature and yielded **3a** in high yield,[10] the reaction of **2a** with 3-butyn-2-one was slower and incomplete after standing overnight at room temperature. Reaction of **2a** with *N,N*-dimethyl propiolamide proceeded very much slower than the above reaction, considerable 11-azaartemisinin was still present after a week at room temperature at which time the reaction was stopped. The products was isolated by column chromatography. An E-stereochemistry was assigned to the adducts based on the magnitude (14~16 Hz) of $J_{13,14}$ cis couplings are 6~9 Hz.[9]



3

a R= CO_2CH_3
b R= $COCH_3$
c R= $CON(CH_3)_2$

In vitro antimalarial activities of **3a**, **3b**, and **3c** against two drug-resistant clones (W-2 and D-6) of *Plasmodium falciparum* were determined by the semi-automated micro-dilution technique.[11] The W-2 clone is

resistant to chloroquine, quinine, sulfadoxine, and pyrimethamine but sensitive to mefloquine. The D-6 clone is resistant to mefloquine but sensitive to chloroquine, quinine, sulfadoxine and pyrimethamine. For the two clones, the ratio of the IC_{50} value of artemisinin divided by that of the compound is given in Table 1. Values greater than one indicate that the compound was more effective than artemisinin. Two derivatives, **3a** and **3b**, were more active than artemisinin. Data on the ^{13}C NMR spectral assignments are summarized in Table 2. Mass spectra (CI and EI) of **3 a - c** described in this report are in agreement with the assigned structures.

Table 1

Relative antimalarial activities against two *Plasmodium falciparum* clones

Compound	R	D-6 IC_{50} **1** / IC_{50} **3**	W-2 IC_{50} **1** / IC_{50} **3**
3a	$^{13}CH=^{14}CH^{15}CO_2{}^{16}CH_3$	2.0	1.2
3b	$CH=CHCOCH_3$	2.54	1.25
3c	$CH=CHCON(CH_3)_2$	0.39	0.35

The IC_{50} value of artemisinin is 1.15 ng/mL for W-2, and 4.19ng/ml for D-6.

Table 2

Summary of ^{13}C assignments

Carbon	**2a**	**3a**	**3b**	**3c**
3	104.8	105.4	105.6	105.3
3-methyl	25.1	25.0	25.1	25.1
4	36.5	36.55	36.5	36.5
5	25.3	25.3	25.3	25.4
5a	51	51.5	51.6	51.5
6	37.6	37.5	37.5	37.6
6-methyl	19.0	19.7	19.9	19.7
7	33.8	34.0	34.1	34.1
8	23.0	22.7	22.8	22.7
8a	46.0	45.4	45.1	45.4
9	32.8	33.6	33.7	33.7
9-methyl	12.1	13.0	13.2	13.1
10	173.0	172.7	172.1	172.3
12	75.6	77.9	78.0	78.8
12a	79.9	79.9	79.9	80.1
13		139.8	139.8	137.9
14		114.3	102.9	104.2
15		199.3	168.5	168.2
16		26.4	51.5	
16a				35.9
16b				29.8

Acknowledgments: We express our appreciation to Mr. Noel Whittaker for mass spectral data and acknowledge the generosity of WHO/CHEMAL and Col. John Scovill of the Walter Reed Institute of Research for providing the artemisinin used in these studies.

References and Notes

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10. Typical experimental procedure: To a solution of **2a** (27 mg, 0.1 mmol) in anhydrous acetonitrile (2 mL) were added at room temperature DMAP (20 mg) and methyl propiolate (30 mg, 0.36 mmol). Color developed immediately (yellow to red). After 15~20 min at room temperature, an examination of the reaction by thin layer revealed that **2a** had completely disappeared and a less polar product had formed. The reaction mixture was concentrated on a rotovap and the residue purified by column chromatography on silica gel.
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