Monoterpenic fragment analogues of Aplasmomycin as potential anti-malarials

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Malaria has become a serious disease throughout recorded history and the protozoan parasite of the family Plasmodidae is known to be the etiologic agent for this. The dangerously improved immunological status of various parasites (especially human malaria parasite P. falciparum) against conventional drugs such as chloroquine necessitates the development of new agents having optimum efficacy.

Aplasmomycin 1, one of the metabolites isolated from the marine fungus, Streptomyces griseus is a boron containing ionophore with significant antibiotic and anti-malarial properties. Several cyclic sesquiterpene peroxides (such as artemisinin, 2) also exhibit anti-malarial activity against a variety of parasite strains. The presence of the terpenoidal moiety in 1 and 2 led us to investigate the structure-anti-malarial activity relationship of monoterpenic fragment analogues of aplasmomycin. In our earlier studies we have observed in vivo anti-malarial activity of the diene ester 3 and in vitro test results of compounds 3-8. We report herein the in vivo anti-malarial activity study of the acids 4, 7 and 9 (Figure 1). Compound 9 was prepared from acid 4 on treatment with thionyl chloride followed by reacting with proline in the presence of potassium carbonate in acetone.

All the compounds exhibited very good activity against P. berghei in mice. On comparison of activities of the compounds with that of artemisinin (40 mg/kg), acid 4 was found to have the highest potency and superior (showed activity at 20 mg/kg dose level) to artemisinin and aplasmomycin (Table I).

Experimental Section

Preparation of 5,9-dimethylnona-2,4,8-trienoic-1-(2-carboxypyrrolidine)amide 9. Acid 4 (500 mg,
Table 1—Anti-malarial activity of compounds 4, 7 and 9 against *P. berghei* in vivo

<table>
<thead>
<tr>
<th>Compd</th>
<th>Untreated control (days)</th>
<th>Mean survival time (days)</th>
<th>Remarks</th>
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<tbody>
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<td>20</td>
<td>40</td>
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<tr>
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<td>4</td>
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<td>13.9</td>
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<tr>
<td>7</td>
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<td>8.40</td>
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<tr>
<td>9</td>
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<td>6.0</td>
<td>11.8</td>
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*Compounds 2 and 3 are tabulated for comparison (ref. 5a).

2.57 mmoles) and thionyl chloride (450 mg, 3.85 mmoles) were heated on water-bath for 1.5 hr. After cooling, the reaction mixture and removal of excess thionyl chloride in vacuo provided the acid chloride 10. To a solution of 10 in acetone (6 mL) was added a mixture of proline (300 mg, 2.60 mmoles) and potassium carbonate (150 mg, 1.30 mmoles), acetone (6 mL) and the resultant reaction mixture stirred at room temperature for 2 hr. The work-up followed by removal of acetone in vacuo and purification of the residue by column chromatography over alumina using pet. ether-ethyl acetate (50%) gave the amide 9 as a viscous liquid (150 mg, 20%).

**IR (CDCl3)**: 3460 (b), 2960, 1715, 1630 cm⁻¹. **H NMR** (60 MHz, CDCl₃): δ 7.30 (m, 1H, olefinic H), 5.90 (m, 1H, olefinic H), 5.40 (m, 1H, olefinic H), 3.20 (m, 3H, -NCH₂-), 2.20 (m, 4H, allylic H), 1.90 (s, 3H, -CH₃), 1.60 (s, 3H, -CH₃), 1.50 (s, 3H, -CH₃), 1.20 (m, 4H, -CH₂).

**Biological Method**: *In vivo Rane's Test*: The compounds 4, 7 and 9 were evaluated for their activity against virulent strains of *P. berghei yoelli* (NK 65) using Rane's schizontocidal method described by Osdene *et al*. Four weeks old mice weighing 2-25 g each received an intraperitoneal inoculum of 1 × 10⁶ parasitized *P. berghei* red cells. The test solutions of synthesized compounds in distilled water were prepared by homogenization with 2 drops of 1% Tween-80 and injected once subcutaneously 72 hr post-infections. A control group of infected mice was not administered any drug was kept as untreated control. The dose range selected was 20, 40, 80 and 160 mg/kg and a minimum of five mice per dose were used. Artemisinin (20 and 40 mg/kg), cycloguanil hydrochloride (25 mg/kg) and DDS (20 mg/kg) were kept as standard drugs in trial for comparison. Deaths occurring within 24 hr of treatment was classified as death due to toxicity. All mice receiving drug showed a survival time of 12-18 days. Testing was evaluated by calculating mean survival time (MST) of the treated and controlled group of mice.

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**References**

2. (a) Quinghaosu Antimalarial Coordinating Research Group, *Chin Med J.*, 92, 1979, 811