In Vitro and *In Vivo* Antimalarial Activities of the Monoglycoside Polyether Antibiotic, K-41 against Drug Resistant Strains of *Plasmodia*

Sir:

In the course of our screening program to discover antimalarial antibiotics from soil microorganisms which are active against drug resistant parasites *in vitro* and *in vivo*, we previously reported that the polyether antibiotic X-206 exhibited potent antimalarial activities *in vitro* and *in vivo*[1]. Now, we have found that a substance produced by an actinomycete strain KP-4050 has selective and potent activities. It was identified as a known monoglycoside polyether antibiotic, K-41 (Fig. 1)[2]. We report here *in vitro* and *in vivo* antimalarial activities of K-41 as compared with clinically used antimalarial drugs.

In vitro antimalarial activities for *Plasmodium falciparum* strains K1 (drug resistant) and FCR3 (drug sensitive) and cytotoxicity for human diploid embryonic cell line MRC-5, were described previously[1]. Rodent malaria-derived strains for *in vivo* testing, *P. berghei* strain N (drug sensitive) and *P. yoelii* ssp. strain NS (chloroquine resistant), were a generous gift of Dr. W. PETERS (Northwick Park Institute for Medical Research, Middlesex, UK). *In vivo* oral antimalarial activities were determined using these strains according to the 4-days suppressive test of PETERS *et al.*[3] and our previous method[1] with some modifications. Test compounds were dissolved in 10% DMSO aqueous solution and administered orally (p.o.) to the mice two hours after infection with parasites (Day 0). Test compounds were successively administered (p.o.) to the mice once a day for 3 days (Days 1~3). The day after the last treatment (Day 4), thin blood films were made from the tail blood of the infected mice, and the parasitaemia was determined with the image analysis equipment (software: Win ROOF, Mitani Co., Ltd., Japan).

K-41 was purified in our institute from the culture broth of an actinomycete strain KP-4050.

The *in vitro* antimalarial activities of K-41 and the standard antimalarial drugs are presented in Table 1. K-41 showed similar antimalarial activity against the drug resistant K1 strain of *P. falciparum* to artemether and artesunate, and was more potent than the clinically used antimalarial drugs, artemisinin, chloroquine and pyrimethamine. Furthermore, the IC_{50} value of K-41 against the K1 strain was 3.6-fold more potent than the FCR3 strain. We then investigated the cytotoxicity of K-41 against MRC-5 cells. The IC_{50} value of K-41 was 690 nM. K-41 showed moderate selectivity indexes with the ratios of 81 and 22 for the MRC-5 cells/K1 strain and MRC-5 cells/FCR3 strain, respectively.

The results of the *in vivo* oral antimalarial activities of K-41 and the standard antimalarial drugs are shown in Table 2. K-41 had oral antimalarial activity against both rodent malaria-derived *P. berghei* strain N and *P. yoelii* ssp. strain NS. Against the drug sensitive N strain of *P. berghei*, K-41 showed more potent oral antimalarial effects than artemether and artesunate. The ED_{50} value of K-41 against the chloroquine resistant *P. yoelii* ssp. strain NS was only

Table 1. Antimalarial activities of K-41 and the antimalarial drugs against K1 and FCR3 strains of *Plasmodium falciparum*

Compound	IC_{50} (nM)	
	K1 strain	FCR3 strain
K-41	8.5	31
Artemether	7.6	2.2
Artesunate	11	2.7
Artemisinin	24	18
Chloroquine	357	29
Pyrimethamine	>100,000	7.8

Fig. 1. Structure of K-41.

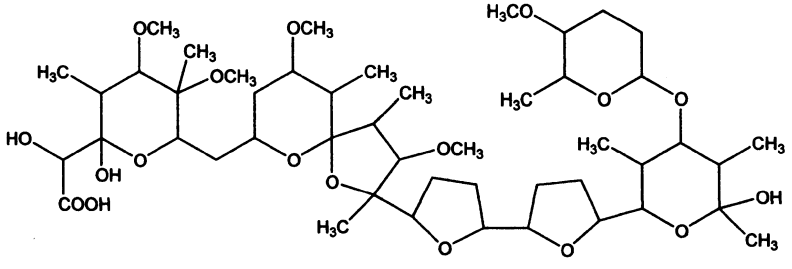

Table 2. *In vivo* oral antimalarial activities of K-41, artemether, artesunate and chloroquine against *P. berghei* strain N and *P. yoelii* ssp. strain NS.

Parasite	Compound	ED_{50} (mg/kg)	ED_{90} (mg/kg)
P. berghei strain N*	K-41	1.9	12.0
	Artemether	2.8	>10.0
	Artesunate	4.0	>10.0
	Chloroquine	1.0	2.5
P. yoelii ssp. strain NS**	K-41	7.0	16.0
	Artemether	5.0	40.0
	Artesunate	4.0	>50.0
	Chloroquine	4.5	>100.0

* drug sensitive strain ** chloroquine resistant strain

slightly greater than those of artemether, artesunate and choloroquine. However, the ED_{90} value of K-41 against *P. yoelii* ssp. strain NS was >2.5-fold higher than these three drugs. These observations are the first report of antimalarial activity of K-41.

In general, polyether compounds exhibit selective toxicity against coccidia and are toxic and poorly absorbed from the intestinal tract of the host. Their interperitoneal LD_{50} values range from 1 to 60 mg/kg[4]. The LD_{50} (i.p. in mice) of K-41 was 48 mg/kg, which has been reported by TSUJI, *et al.*[2]. We then investigated the preliminary acute toxicity of K-41 using mice. The LD_{50} (p.o. in mice) value of K-41 was >100 mg/kg (data not shown). Currently, monensin and salinomycin are on the market as anticoccidal agents, being administered as food supplements. *In vivo* oral antimalarial activities of polyether antibiotics have been reported by RAETHER *et al.*[5], SCHILDKNECHT *et al.*[6] and GUMILA *et al.*[7] using different test conditions. Salinomycin and lasalocid A were shown to be inactive against the drug resistant strain of *P. berghei* in rats after oral administration[5]. Three synthetic monensin urethane derivatives were reported moderately active against *P. berghei* in mice after oral administration[6]. Monensin A and 5-bromo lasalocid A were found to be active against *P. vinckei petteri* or *P. chabaudi* in mice after oral administration, with the ED_{50} values of 10.1 mg/kg and 11 mg/kg, respectively[7], using a slightly modified version of the 4-day suppressive test of PETERS *et al.*[3]. Present data indicate that the levels of oral absorption for K-41 is high as compared with those of other polyether antibiotics.

These results support the view that K-41 is a promising lead compound for a new type of oral antimalarial drug. We are investigating the antimalarial potential of K-41 further.

Acknowledgments

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