Studies on the resistance to single and combined antimalarials in the "Plasmodium berghei" mouse model

Autor(en): **Merkli, B. / Richle, R.W.**

Objekttyp: **Article**

Zeitschrift: **Acta Tropica**

Band (Jahr): **37 (1980)**

Heft 3

Persistenter Link: https://doi.org/10.5169/seals-312652

Acta Tropica *37*, 228–231 (1980)

Pharmaceutical Research Division, F. Hoffmann-La Roche & Co. Ltd., Basel, Switzerland

Studies on the resistance to single and combined antimalarials in the *Plasmodium berghei* mouse model

B. MERKLI, R. W. RICHLE

Using the method of increasing drug pressure during repeated weekly passages in mice (Peters, 1965), lines of *Plasmodium berghei* K 173 resistant to pyrimethamine (P), mefloquine (M), chloroquine (C), and to the combination of sulfadoxine plus pyrimethamine (S-P) and of mefloquine plus sulfadoxine plus pyrimethamine (M-S-P) were developed. At the first passage, treatment was started with the "maximum dose tolerated by the parasite" (mtD: still permitting weak parasitaemia at the end of the passage), and during the further passages mtD was gradually increased with increasing resistance of the parasite. The number of passages necessary to achieve maximum resistance (mtD equal to the highest doses still tolerated by the murine host), or the mtD after a maximum of 60 passages, were considered parameters of the risk of resistance existing with extensive, prolonged use of the drugs in human malaria.

With P as a single drug mtD (expressed as weekly total dose) increased from initially 8 mg/kg to the maximum of 400 mg/kg after only 13 passages. With M alone mtD rose from 15 mg/kg in the first to the maximum of 550 mg/kg in the 45th passage, and in the mice receiving C alone mtD increased from 10 to 350 mg/kg (maximum) after 44 passages. With the combination S-P (1 part of P plus 2 parts of sulfadoxine) mtD was, after 60 passages, raised from initially 2.5 mg/kg to 500 mg/kg which were still tolerated by the host. When the triple combination M-S-P was used, development of resistance was markedly delayed. After the maximum of 60 passages mtD had risen from initially 10 mg of the M plus 1 mg/kg of the S-P component to only 50 mg/kg of M plus 15 mg/kg of S-P (Fig. 1 and Table 1; cf. Merkli et al., 1980).

The cross-resistance pattern of the resistant lines of *P. berghei* resulting from the above-mentioned experiments was checked by infecting mice and determining the chemotherapeutic response to each of the other drugs and drug combinations. The response to quinine (Q) was also included in this study.

Correspondence: Dr. Bernhard Merkli, Pharmaceutical Research Division, F. Hoffmann-La Roche & Co. Ltd., Grenzacherstrasse 124, CH-4002 Basel, Switzerland

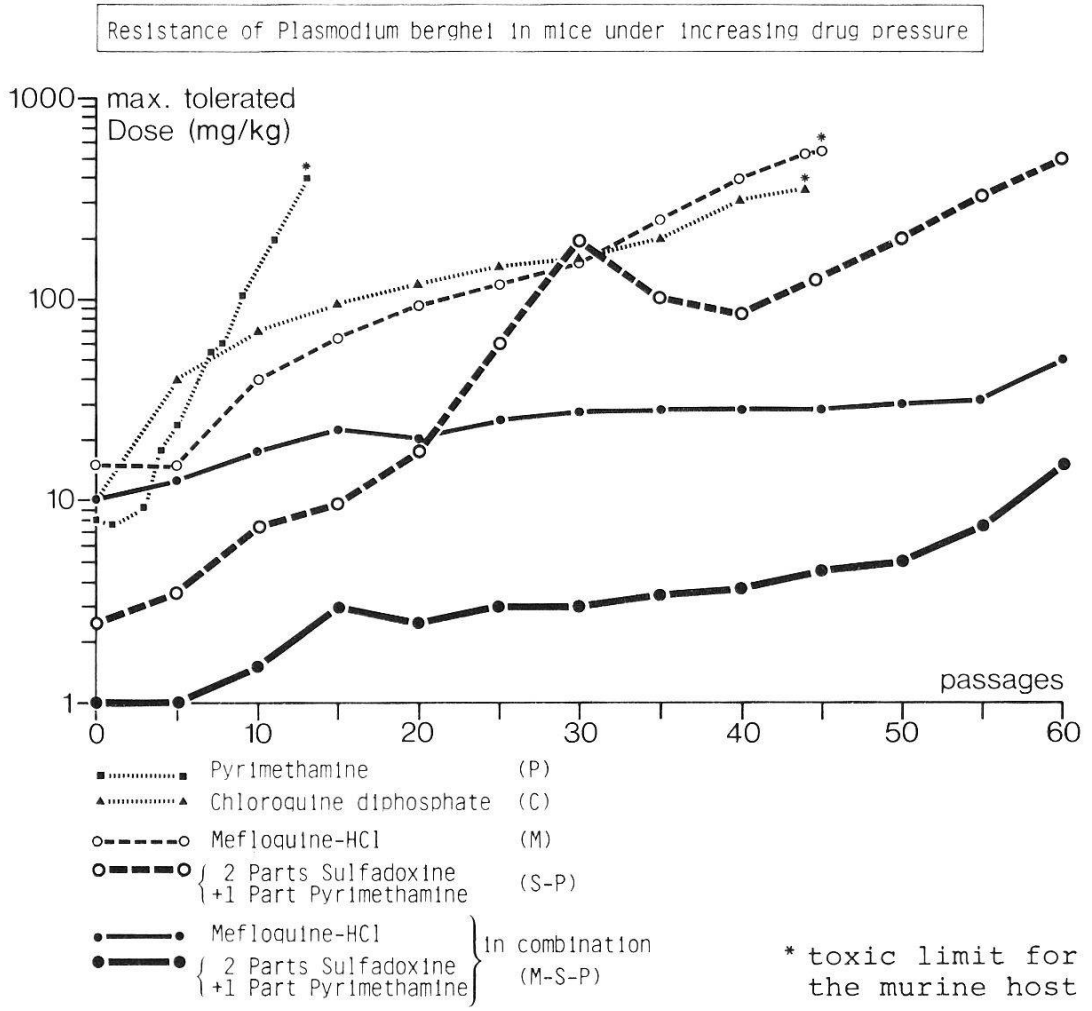

Fig. 1

Table 1. Maximum tolerated dose (mtD)* of some antimalarial drugs alone and in combination in mice infected with resistant lines of *Plasmodium berghei* K 173**

Line	Substance	Initial mtD (mg/kg)	Final mtD (mg/kg)	Number of passages (60 = maximum)
NP	Pyrimethamine	8	400***	13
NC	Chloroquine diphosphate	10	350***	44
NM	Mefloquine-HCl	15	550***	45
NS-P	2 parts sulfadoxine + 1 part pyrimethamine	2.5	500	60
NM-S-P	(10–30) parts mefloquine + 2 parts sulfadoxine + 1 part pyrimethamine	10 / 1	50 / 15	60

* Maximum tolerated dose by the parasite: total dose of the drug(s) (given on 4 to 5 days per week) just permitting weak parasitaemia at the end of each of the weekly passages. All the drugs were administered orally.

** For details of the method, see Peters (1965), and Merkli et al. (1980).

*** Highest doses still tolerated by the murine host.

Table 2. Cross resistance pattern of the resistant lines from *Plasmodium berghei* K 173* in chemotherapeutic experiments**

Drug	*P. berghei* K 173; ED 90 (mg/kg)	Resistant lines***						*P. yoelii nigeriensis*
		NP	NC	NM	N/1100****	NS-P	NM-S-P	
Chloroquine diph.	6.2	S	$^{++}$R	S	$^{+}$R	S	HS	$^{++}$R
Mefloquine-HCl	5	S	$^{++}$R	$^{++}$R	$^{++}$R	S	$^{+}$R	S
Pyrimethamine	1.5	$^{+++}$R	S	S	S	$^{+++}$R	$^{+++}$R	S
Sulfadoxine	61	S	S	HS	HS	$^{++}$R	S	HS
2 parts sulfadoxine + 1 part pyrimethamine	1.3	–	S	S	–	$^{+++}$R	$^{++}$R	HS
Quinine-HCl	153	S	S	$^{+}$R	$^{+}$R	S	S	S

* Cf. Fig. 1 and Table 1. The lines were used when they had reached maximum resistance and drug pressure was maintained up to the test.

** "4-day chemotherapy test" according to Peters (1965). The chemotherapeutic response was expressed as ED_{90}, i.e., the daily dose of the drug(s) reducing parasitaemia by 90% (with the parasitaemia of untreated controls taken as 100%). All drugs were given orally.

*** Resistance was measured by calculating the "Resistance factor"

$$I_{90} = \frac{ED_{90} \text{ resistant line}}{ED_{90} \text{ parent line}}$$

**** Produced by W. Peters, London, from the same parent strain.

The following symbols were used in the table:

HS = "hypersensitive": $I_{90} < 1$
S = "sensitive": $\cong 1$
$^{+}$R = "slightly resistant": 1–10
$^{++}$R = "moderately resistant": 10–100
$^{+++}$R = "strongly resistant": > 100

Table 3. Virulence of the resistant lines from *Plasmodium berghei* K 173

Line	Parasitaemia Day 4 post inf. (%)*	Mean survival time (days)	Number of mice studied
P. berghei K 173	52.1	6	143
NP	66.1	6.5	65
NC	4.1	>18.8 (11% surviving)**	71
NM	1.2	>28.4 (18% surviving)**	131
N/1100***	7.3	25	57
NS-P	26.5	7.7	118
NM-S-P	18.8	8.7	70
P. yoelii nigeriensis	51	>8.9 (3% surviving)	127

* As determined in the untreated control mice used in the chemotherapeutic experiments (Table 2), parasitaemia is expressed in % of parasitized erythrocytes.

** Mice still alive on day 42 *post infectionem* (end of the experiments) were considered as surviving.

*** Mefloquine-resistant line supplied by Prof. W. Peters, London.

The P-resistant line proved also resistant to S-P but was sensitive to M, C and Q. The line resistant to M was sensitive to P, C and S-P but moderately resistant to Q. The C-resistant line was sensitive to P, Q and S-P but moderately resistant to M. The S-P-resistant line was sensitive to M, C and Q, and the line (moderately) resistant to the triple combination M-S-P was sensitive to C and Q. Another M-resistant *P. berghei* line kindly supplied by Prof. Peters, London (Peters, 1977), was tested in the same manner, and in contrast to our M-resistant line showed moderate cross-resistance to C (Table 2).

Most of the resistant lines proved less virulent to the mice than the parent strain of *P. berghei*, and all lost their resistance when passaged without drug pressure (Table 3).

Merkli B., Richle R., Peters W.: The inhibitory effect of a drug combination on the development of mefloquine resistance in *Plasmodium berghei*. Ann. trop. Med. Parasit. *74*, 1–9 (1980).

Peters W.: Drug resistance in *Plasmodium berghei*, Vincke and Lips, 1948. I. Chloroquine resistance. Exp. Parasit. *17*, 80–89 (1965).

Peters W., Portus J., Robinson B. L.: The chemotherapy of rodent malaria XXVIII. The development of resistance to mefloquine (WR 142,490). Ann. trop. Med. Parasit. *71*, 419–427 (1977).