d'aisance, les puits perdus, etc.. Les méthodes de lutte génétique — lâchers de mâles stérilisés par radiations — n'ont pas donné de résultats concluants en Inde.[q] Mais ces essais ne ferment pas la porte à tout espoir, d'autant que les stérilisants chimiques lèsent moins les moustiques que les rayons gamma. Une autre méthode de lutte génétique, basée sur les incompatibilités cytoplasmiques, a été envisagée. On sait en effet que certaines souches de *C. p. fatigans*, bien que s'accouplant entre elles, ne sont pas interfertiles. C'est ainsi que des souches de Californie sont incompatibles avec celles de Rangoon;[r] des incompatibilités de même ordre existent d'ailleurs entre souches d'Afrique de l'Ouest. En introduisant en nombre suffisant des mâles incompatibles avec la souche locale, on devrait obtenir les mêmes résultats qu'en lâchant des mâles stériles. D'ailleurs on a pu expérimentalement, en cage, anéantir une population de *C. p. fatigans* de Rangoon en lâchant des mâles de Californie.[s] Bien sûr, de telles méthodes restent encore dans le domaine expérimental mais elles sont significatives de la volonté des chercheurs d'aborder le problème de la lutte contre *C. p. fatigans* sous tous ses angles et il n'est pas exclu que la solution ne consiste en l'intégration de différents procédés mécaniques, chimiques et biologiques de contrôle de ce vecteur.

[q] Krishnamurthy, B. S., Ray, S. N. & Joshi, G. C. (1962) *Indian J. Malar.*, **16**, 365-373.

[r] Laven, H. (1967) *Bull. Org. mond. Santé*, **37**, 263-266.

[s] Comité OMS d'Experts de la Filariose (1967) *Org. mond. Santé Sér. Rapp. techn.*, **359**.

Conclusion

Actuellement, en Afrique de l'Ouest, *C. p. fatigans* est simplement une gêne pour la population, par suite de sa pullulation et de la multiplicité de ses agressions. Bien qu'on ne soit pas encore certain qu'il transmette *W. bancrofti*, il n'est pas exclu que, dans un proche avenir, il se révèle un vecteur de cette filaire. C'est donc dès maintenant qu'il faut entreprendre la lutte contre ce moustique, car ultérieurement on risque d'avoir à combattre et le vecteur et la maladie elle-même, ce qui deviendrait beaucoup plus difficile.

* * *

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Studies on Alleged Chloroquine-Resistance of Malaria Parasites in Axim and Obuasi, Ghana *

by E. G. BEAUSOLEIL, *Senior Medical Officer, Malaria Service, Ministry of Health, Accra, Ghana*

Studies in Axim

A report of a strain of *Plasmodium falciparum* apparently resistant to suppressive and therapeutic doses of chloroquine in Axim, Ghana,[a] led to further investigations carried out in the months of March, April and May 1966, following the procedure recommended by the WHO Scientific Group on Resistance of Malaria Parasites to Drugs.[b]

* Fuller details of the results of these studies are given in unpublished mimeographed documents WHO/Mal/67.601 and WHO/Mal/67.622, which are available, on request, from Research and Technical Intelligence, Division of Malaria Eradication, World Health Organization, Geneva, Switzerland.

[a] Schwendler, H. P. (1965) *Ghana med. J.*, **4**, 20.

[b] WHO Scientific Group on Resistance of Malaria Parasites to Drugs (1965) *Wld Hlth Org. techn. Rep. Ser.*, **296**.

Methods. All the subjects were resident in Axim and selected from children 5–17 years old attending primary and secondary schools in Axim. On the day prior to treatment, designated Day −1, the children were weighed and screened for the presence of malaria parasites in the peripheral blood and for the presence of chloroquine in the urine. The examination of the blood for malaria parasites and of the urine for evidence of chloroquine was repeated on the day of treatment, designated Day 0. Only subjects with malaria infection and no trace of chloroquine in the urine on Day 0 were selected for the investigation.

Examination for malaria was made daily on thick and thin blood films, stained by the standard Giemsa method. A film was declared positive for *P. falciparum* only if trophozoites were present, and negative only if parasites were not seen in 400 oil-immersion fields (thick film).

The parasite density was estimated by counting the parasites against 400 leucocytes and multiplying the figure obtained by 20 (taking 8000 as the average leucocyte count per mm³ of blood); parasite densities were classed according to the scale proposed by Bruce-Chwatt.[c] From Day 0, films were first screened by 1 of the 4 technicians and then checked individually by the author. A subject was dropped from the group after the first negative film. If parasites were present on Day 5, the blood was tested again on Days 6 and 11.

Urine samples were collected from each subject daily and tested for chloroquine by the method described by Wilson & Edeson.[d] All tests were carried out by the author within 3 hours of the collection of the samples. Although there was no proof that every sample of urine tested actually belonged to the child who brought it, there is no reason to believe that some of the children collected urine from other members of the group. Whenever the result of the test was questionable, the test was repeated using the method described by Haskins.[e]

On Day 0 each subject was given an oral dose of chloroquine corresponding as nearly as possible to 10 mg of chloroquine base per kg of body-weight, according to a standard dosage schedule.

In each case, the drug was swallowed with a draught of water and the mouth was inspected by the author to ensure actual ingestion. If a child chewed the tablet (a frequent occurrence), it was asked to rinse the mouth thoroughly with water and then swallow the water. If visual inspection showed debris of the tablet between the teeth, the process was repeated until no drug remained; the child was given a toffee to promote co-operation.

If parasites were present on Day 5, treatment was continued as follows. On Day 6, after testing of the blood for malaria parasites and of the urine for the presence of chloroquine, 2 doses of chloroquine were administered orally. The first dose corresponded to 10 mg of chloroquine base per kg of body-weight and the second dose, given 6 hours later, corresponded to 5 mg of chloroquine base per kg of body-weight. On Day 7, a single dose of chloroquine corresponding to 5 mg/kg body-weight was given orally. This dose was repeated on Day 8.

Results. The crude parasite rate among the population of schoolchildren aged 5–17 years was found to be 70%. Most infections (over 90%) were due to *P. falciparum.*

P. malariae accounted for about 15% of infections, about 60% of which occurred as mixed infections with *P. falciparum.* No *P. ovale* infection was observed during the investigations.

Altogether, 141 subjects with *P. falciparum* infections, 14 of whom had mixed infections with *P. malariae*, and 9 subjects who had a single infection with *P. malariae* were followed up after treatment to determine the clearance of parasites from the peripheral blood.

In the 141 cases with *P. falciparum* infection with an over-all parasite density index of 3.4 (see the accompanying table), parasite clearance occurred in all but 9 cases (6.4%) by Day 5. In most of the cases (about 70%), parasites were cleared by Day 3. In 14 cases the first day of negative blood could not be determined because of absenteeism. In 3 children who were absent on Day 1, the blood was negative on Day 2; and in 9 children who were absent on Days 1 and 2, there were no parasites on Day 3. In 2 children who were absent for the first 3 days or longer, no parasites could be seen on Day 5.

In about 80% of those cases with initial parasite densities up to 1600/mm³, parasite clearance occurred by Day 3; in cases with initial parasite densities of over 1600/mm³, parasite clearance occurred by Day 3 in about 70% of cases. Parasite clearance times of 1 day occurred only in some of the cases with initial parasite densities of up to 200/mm³.

[c] Bruce-Chwatt, L. J. (1958) *Trans. roy. Soc. trop. Med. Hyg.*, **52**, 389.

[d] Wilson, T. & Edeson, J. F. D. (1954) *Med. J. Malaya*, **9**, 1150.

[e] Haskins, W. T. (1958) *Amer. J. trop. Med. Hyg.*, **7**, 199.

RESPONSE OF *P. FALCIPARUM* INFECTIONS AT AXIM (GHANA) TO STANDARD DOSES OF CHLOROQUINE

Age-group (years)	Sex	Mean weight (kg)	Mean dosage of chloroquine base (mg)	No. with parasitaemia							Mean parasite density [a]						
				Day −1	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day −1	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5
5-7	M	16.6	170	11	11	9	7	5	3	2	5.2	5.1	4.3	1.4	1.4	1.0	1.5
	F	17.6	175	7	7	7	6	6	0		5.1	5.2	2.4	1.1	1.0	0	
8-10	M	19.7	200	26	26	20	9	1	0		2.8	2.6	1.8	1.0	1.0	0	
	F	21.0	200	17	17	14	8	4	3	1	4.2	4.3	2.4	1.6	1.6	1.0	1.0
11-14	M	29.0	260	35	35	31	6	6	5	2	3.0	3.4	2.1	1.2	1.2	1.0	1.0
	F	32.5	350	14	14	10	5	3	2	2	3.3	3.3	2.6	2.8	2.3	3.0	3.0
15+	M	46.5	465	23	23	15	10	5	3	2	2.6	2.7	1.8	1.6	1.8	1.6	2.0
	F	51.0	510	8	8	7	6	4	2	0	3.8	4.0	2.7	1.6	1.0	1.0	0

[a] According to the scale proposed by Bruce-Chwatt, L. J. (1958) *Trans. roy. Soc. trop. Med. Hyg.*, **52**, 389.

The initial parasite densities of the 9 cases followed in the second stage of the investigation ranged from under 100/mm³ to 1600/mm³.

Rather surprisingly, parasite clearance occurred by Day 5 in all the cases with high initial parasite densities. In 8 out of the 9 cases where parasite clearance had not occurred by Day 5, it occurred by Day 11.

In the only case (No. 16) in which parasite clearance did not occur by Day 11, only 4 disintegrating tropozoites were seen in more than 400 oil-immersion fields. When seen 4 days later, the blood was negative. The case was followed up daily for 2 weeks a month later (from Day 46 to Day 59), the blood remaining negative throughout the whole period of observation.

In all but 3 of the 23 cases of mixed or single infections with *P. malariae* which were observed daily without interruption, parasite clearance occurred by Day 3. Only 1 case of single infection with *P. malariae* remained positive on Day 5; complete parasite clearance had however occurred by Day 11.

Excretion of chloroquine occurred in all cases after its administration by mouth. Although no quantitative estimations were made, it was quite obvious from the intensities of the qualitative reactions that there was considerable variation in the rate of excretion of chloroquine in the urine. In 20 cases that were observed for 5 days after treatment, 17 samples of urine were positive on Day 5, 1 was negative and 2 were doubtful.

An entomological study showed the principal vectors to be *Anopheles gambiae* and *An. funestus*, *An. gambiae* being the predominant species during the period of observations. The room densities were 5.4 and 0.7, and the sporozoite rates 1.9% and 0%, for *An. gambiae* and *An. funestus* respectively.

Discussion. A critical review of the study carried out previously[a] shows many sources of error in the method of investigation. Although there is no doubt that the nurses administered the drugs, there is no evidence that tablets were actually ingested and retained. Ingestion and absorption were not checked by testing for urinary excretion of chloroquine. There is also the possibility that diarrhoea, which is a common complication of malaria in infants and children, could have reduced the absorption of the drug in some cases.

The method of diagnosis could have introduced false positives, and errors could have been introduced by variation in the rates of absorption and excretion of chloroquine and in the parasite clearance time. In particular, it was observed that in all cases the second course of therapy was started after 72 hours without waiting to see if clearance would occur by the fifth day after treatment.

These points emphasize the need for the application of rigorous criteria in the assessment of the response of malaria parasites to drugs.

In the present investigations, *P. falciparum* and *P. malariae* infections treated with a single dose of chloroquine administered orally at the rate of 10 mg of chloroquine base per kg of body-weight appeared to respond normally to the drug.

About 77% of the cases with *P. falciparum* infections were cleared of parasitaemia by the third day after treatment, and 94% by the fifth day. Of the 9 cases which were not cleared by the fifth day, 8 were cleared of parasitaemia by the eleventh day, i.e., 3 days after the completion of a second course of therapy.

In the single case with persistent parasitaemia on the eleventh day, only 4 trophozoites were seen in over 400 oil-imersion fields and even then the parasites appeared to be in a process of disintegration.

However, the fact that in all cases with initial parasite densities of over 1600/mm^3 parasite clearance occurred within 5 days after treatment, and that the only cases in which parasite clearance did not occur within 5 days after a single dose of chloroquine had initial densities of less than 1600, does not exclude the possibility that there may be some strain or strains of *P. falciparum* that are less responsive to the standard dosage of chloroquine.

P. malariae showed normal response to chloroquine. Only one child had persistent parasitaemia on Day 5, and response to the second course of treatment was normal. Unfortunately, prevailing conditions did not permit the proper follow-up of acute cases to assess the response to chloroquine.

Thus, the results of the present investigation failed to establish the existence of strains of *P. falciparum* resistant to chloroquine in Axim.

Studies in Obuasi

Another investigation was carried out in Obuasi, Ghana, where 2 children died early in 1966 from a complaint initially diagnosed as malaria, which apparently failed to respond to chloroquine or parenteral quinine. The same treatment and test procedure was followed as in Axim.

The subjects were selected from schoolchildren aged 8–18 years, all of whom were resident in Obuasi. Preliminary screening of 336 children showed a parasite rate of 50% and a *P. falciparum* gametocyte rate of 5.4%.

One hundred children with *P. falciparum* were selected for study of the response of *P. falciparum* to chloroquine (base) given at 10 mg/kg. By Day 3, 68% of the subjects were free from parasites, and by Day 5, 85% were cleared. Of the 15% with persistent parasitaemia on Day 5, about 50% had initial parasite densities of under 401 parasites per mm^3.

Parasites were cleared in all subjects by Day 11, 3 days after the second course of chloroquine. Fifteen subjects with single or mixed infections of *P. malariae* were free from parasites by Day 5.

The investigations failed to establish the existence of chloroquine resistance in both *P. falciparum* and *P. malariae*. However, in view of the high proportion of subjects with parasitaemia on Day 5 and the initial low parasite densities of many subjects with persistent parasitaemia on Day 5, there is need to reassess the situation in due course.

* * *

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