EFFECT OF A SINGLE DOSE (600 MG) OF ALBENDAZOLE ON *LOA LOA* MICROFILARAEMIA

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Summary :

The problem of *Loa*-encephalopathy, which may occur after ivermectin treatment of patients harbouring high *Loa* microfilarial loads, might be solved if one could find a treatment regimen bringing about a significant but progressive decrease in the *Loa* microfilaraemia. A trial was performed in Central Cameroon, whose aim was to follow up for 10 months, and to compare the changes in the *Loa* microfilarial loads in two groups of patients, one treated with a single dose (600 mg) of albendazole (Alben®, SmithKline Beecham) given with fatty food, and the other treated with mebendazole (100 mg, twice a day, generic tablets) at a fasting state. The microfilarial loads remained stable in the mebendazole group, whereas a significant decrease in microfilaraemia was recorded in the albendazole group (initial median load: 230 microfilariae per 50 µl; median load ten months after: 84 microfilariae per 50 µl). This should encourage further trials to evaluate the effects and the safety of two- or three-day albendazole regimens in patients infected with *Loa loa*.

KEY WORDS : albendazole, *Loa loa*, loiasis, onchocerciasis, clinical trial, ivermectin, side effects.

Résumé : EFFET D'UNE DOSE UNIQUE D'ALBENDAZOLE (600 MG) SUR LA MICROFILARÉMIE À *LOA LOA*

La prise d'ivermectine peut induire, chez les sujets hypermicrofilarémiques pour Loa loa, *des réactions sévères à type d'encéphalopathie. Ce problème pourrait être résolu par l'administration préalable d'un traitement abaissant de manière progressive cette microfilarémie. Lors d'un essai mené au Cameroun, nous avons comparé l'évolution sur 10 mois de la microfilarémie à* Loa loa *dans deux groupes de patients, l'un traité par une dose unique (600 mg) d'albendazole (Alben®, SmithKline Beecham) associée à la prise d'aliments riches en graisse, et l'autre traité à jeun par mébendazole (100 mg deux fois par jour pendant trois jours sous forme de comprimés génériques). La microfilarémie, restée stable dans le groupe mébendazole, a en revanche significativement chuté dans le groupe traité par albendazole (médiane des charges initiales : 230 microfilaires par 50 µl ; médiane des charges 10 mois après traitement : 84 microfilaires par 50 µl). Ce résultat devrait conduire à évaluer l'effet et la tolérance de traitements de 2-3 jours par albendazole chez des patients parasités par* Loa loa.

MOTS CLÉS : *albendazole,* Loa loa, *loase, onchocercose, essai thérapeutique, ivermectine, effets secondaires.*

Large-scale ivermectin (Mectizan®) treatments against onchocerciasis are hampered in Central Africa by the fact that the drug may induce encephalopathy in those persons harbouring high *Loa loa* microfilaraemia (Gardon *et al.*, 1997; Boussinesq *et al.*, 1998). The pathological processes involved in these accidents are not well known, but ophthalmological examinations suggest that they may be related to a massive paralysis of the *Loa* microfilariae (mf) leading to obstructive phenomena in the capillaries of the brain (Fobi *et al.*, 2000). If this is so, a means of solving the problem would be to give, systematically to the whole population, a preliminary treatment that would bring about a slowly progressive decrease in the *Loa* mf counts. Should such a regimen be found, it would be possible, after a given interval, to administer Mectizan® at the standard dose without having to implement specific surveillance procedures to monitor the serious reactions. Our objective was to evaluate whether a single dose of albendazole could be used for such a preliminary treatment. The choice of this drug and of its dose was based on four reasons: a) the only study published so far on the effect of albendazole on *Loa* has shown that repeated treatments with 200 mg during 21 days lead to a decrease of 80 % in the *Loa* mf counts after six months (Klion *et al.*, 1993); b) trials have shown that a single dose of albendazole (400 or 600 mg) brings about a progressive decrease in the mf loads of *Wuchereria bancrofti* (Ismail *et al.*, 1998; Dunyo *et al.*, 2000a, 2000b); c) the decision by SmithKline Beecham Laboratories to donate albendazole for eliminating lymphatic filariasis (Sharp, 1998) suggests that the drug might also be made available free of charge to facilitate the development of onchocerciasis control programmes in areas where specific problems associated with co-endemic loiasis might be encountered; d) the choice of a single dose was based on the fact that it would be easily administered on a large scale.

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In a preliminary trial, a single dose (600 mg) of albendazole had been administered at a fasting state to 17 children with *Loa* mf in the blood. Repeated blood examinations up to 220 days after treatment did not show any decrease in the mf loads, when compared with the pre-treatment values. As it is known that a fatty meal increases markedly the absorption of albendazole (Lange *et al.*, 1988; Awadzi *et al.*, 1994), a second study was performed on two groups of loiasis patients, one treated with a single dose of 600 mg of albendazole given with fatty food, and the second receiving mebendazole in a fasting state. We report here the results of this study.

MATERIALS AND METHODS

The trial was conducted in Cameroon in two villages located about 60 km south of Yaounde. Standardized blood films (50 µl) were prepared between 11.00 a.m. and 1.00 p.m. from 277 persons aged 12 years and above. After Giemsa-staining, the *Loa* mf in the blood films were counted. A total of 45 persons with more than three *Loa loa* mf/50 µl blood accepted to participate in the trial. They were divided into two groups, after stratification for mf concentrations. One group (24 patients) was given, in a fasting state, a three-day course of mebendazole (100 mg, twice a day, in generic tablets). This drug was chosen because it was most unlikely to produce changes in *Loa* microfilaraemia, while bringing a benefit to the patients by eliminating their intestinal nematodes, which are highly prevalent in the study area. The second group (23 patients) received a single dose of 600 mg of albendazole (1.5 tablet of Alben®, SmithKline Beecham), which was given together with one wheat fritter of 45-50 g filled with some 25 g of butter. This meal was chosen because it is very commonly eaten by the local population, and it allowed us to standardize the quantity of fat taken by the patients. The *Loa* mf loads were re-measured, using the same methods as at the initial examination round, 1, 7, 15, 30, 60, 90, 120, 150, 180, and 300 days after treatment. The slides were examined after each examination, to determine whether a marked decrease in microfilaraemia would allow us to stop the trial; but for the final analyses, all the slides were re-examined at the end of the trial, after having been recoded so that the microscopist had no information on the patient's identity or the date of blood collection.

The analysis compared first the results obtained in the two groups, using the Wilcoxon-Mann-Whitney test. The Wilcoxon signed rank test was performed to evaluate the changes over time in the microfilaraemia. A 5 % significance level was used in all comparisons.

RESULTS

The two groups of patients did not differ significantly with regard to sex ratio, average age, and pre-treatment mf counts. The initial median mf loads in the albendazole and mebendazole groups were 230 mf/50 µl (range: 7-1377) and 199 mf/50 µl (range: 3-5535), respectively. The table I shows the number of patients present, and the medians, ranges and Williams geometric means (WGM) of the mf loads recorded at each examination; the WGM is the geometric mean calculated after having added 1 to each count, in order to take into account the negative values (Williams, 1937). As several patients missed some of the examinations, we compared, for each round, the pre-treatment mf counts of the patients of both groups present at that round. At all the examinations, the two groups remained comparable in terms of initial levels of microfilaraemia.

The comparisons between groups did not show any significant difference at any of the examination rounds (Table I). But when analysing each group separately, it was found that in the mebendazole group the mf loads recorded at each post-treatment round did not differ significantly when compared with the values at D0, whereas in the albendazole group the loads recorded at D7, D15 and D30 were similar to the initial values, but those found from D60 to D300 were significantly lower. From D30 to D180, the median load in the albendazole group decreased progressively from 304 to 86 mf/50 µl. Then, the loads seemed to remain stable, with a median value of 84 mf/50 µl at D300. Amongst the 23 patients who received albendazole, 14 presented mf counts below 50 % of the initial value at at least one follow-up examination. A fall of this magnitude was observed on only nine of the 24 persons treated with mebendazole.

DISCUSSION

It has been shown that albendazole is neither macronor microfilaricidal for *Onchocerca volvulus*, but that the drug has an embryotoxic effect on all the intra-uterine stages, including the stretched mf (Awadzi *et al.*, 1991). This would explain the progressive decrease in *O. volvulus* mf loads, which would be the result of natural death of mf and the interruption of production of new mf by the adult worms. With regard to *W. bancrofti*, it has been shown that daily doses of 800 mg repeated during three weeks have both micro- and macrofilaricidal effects (Jayakody *et al.*, 1993). Although little is known of the effect of a single dose of albendazole on *W. bancrofti*, some results suggest

Treatment		Day 0	Day 1	Day 3	Day 7	Day 15	Day 30	Day 60	Day 90	Day 120	Day 150	Day 180	Day 300
Albendazole + fatty food	No of patients	23	22	22	17	21	21	22	20	22	23	23	19
	Median	230.0	298.5	212.5	271.0	354.0	304.0	121.0	142.5	111.5	133.0	86.0	84.0
	1st-3rd quartiles	85.0-759.0	82.0-642.0	74.0-469.0	74.5-583.8	66.0-779.3	86.0-772.0	31.0-725.0	31.8-669.0	23.0-447.0	46.0-532.0	41.0-535.0	31.0-535.0
	WGM	180.3	170.4	109.2	206.2	225.9	193.5	117.3	108.4	95.9	130.3	107.6	102.4
	Range	7 – 1377	9 – 1187	8 – 1286	5 – 1208	11 – 1634	6 – 1580	0 – 1357	0 – 1129	0 – 1363	1 – 1063	0 – 1379	3 – 1474
	P^a	–	0.013	0.001	0.309	0.741	0.102	0.036*	$< 10^{-3}$*	0.001*	0.001*	0.001*	0.01*
Mebendazole (fasting state)	No of patients	24	24	24	20	22	19	22	20	23	22	23	22
	Median	199.0	254.5	227.5	232.5	266.5	249.0	266.0	293.0	312.0	217.0	231.0	231.0
	1st-3rd quartiles	66.8-625.3	86.0-629.3	80.3-599.0	96.0-758.5	89.0-645.0	53.0-534.0	101.0-567.0	94.3-669.0	78.0-632.0	88.0-780.0	81.0-620.0	67.0-546.0
	WGM	195.1	239.6	218.0	258.7	223.5	134.1	207.1	214.1	199.6	183.9	181.9	172.8
	Range	3 – 5535	8 – 4552	6 – 3978	5 – 4022	0 – 5194	0 – 3668	0 – 3307	0 – 4249	0 – 4802	0 – 4747	0 – 4242	0 – 5209
	P^a	–	0.067	0.153	0.433	0.148	0.205	0.475	1	0.903	0.884	0.976	0.833
P^b		0.550	0.974	0.676	0.819	0.903	0.936	0.307	0.341	0.242	0.388	0.307	0.403

[a] Comparison between initial microfilaraemia and the values recorded at each post-treatment examination (Wilcoxon signed rank test); * shows the significant results ($P < 0.05$).
[b] Comparison between the values recorded in the albendazole and the mebendazole groups at each examination (Wilcoxon-Mann-Whitney U test).

Table I. – Median, first and third quartiles, Williams Geometric mean (WGM) and range of *Loa loa* microfilaraemia (microfilariae per 50 µl) in the groups treated with albendazole (600 mg) + fatty food, or with mebendazole (600 mg) in a fasting state.

that it could have an effect on the production of mf by the adult worms (Dunyo *et al.*, 2000b).

In our study we decided, for ethical reasons, to use mebendazole as a "control" treatment, having little effect on the *Loa* microfilarial load; and the stability in the mf loads recorded in the mebendazole group showed that this decision was justified. The comparison between the two groups did not reveal a significant difference at any time after treatment, but this is probably related to the small number of patients enrolled in the study. In contrast, the fact that there were different patterns in the changes recorded in the two groups suggests that a single dose of albendazole given with a fatty meal brings about, in some, but not all patients, a marked decrease in the *Loa* microfilaraemia.

Striking differences in the responses were observed between individuals in the albendazole group. The mf densities of some of them decreased by more than 10-fold, as compared with the pre-treatment values, whereas others showed very stable microfilaraemias such as those demonstrated in untreated patients (Garcia *et al.*, 1995). These variations do not seem to be related to the sex, the age, or the initial level of infection of the patients; they could be due to differences in the absorption of the drug.

It is clear, from our results, that preliminary distributions of single doses of albendazole would not be a means to reduce, in a community, the *Loa* mf loads to such low values that ivermectin could be then given without risk. The inter-individual variability in the effect of the drug, and the relatively limited reductions observed in those patients who "responded" to treatment, are the main reasons for that. Although there was a significant decrease in the median of the *Loa* microfilaraemia in the albendazole group, the geometric means were not reduced markedly. This is probably related to the fact that there were still some very high values in the albendazole group at the end of the follow-up. However, the observations reported in the present paper should encourage further trials, where the effects of two- or three-day regimens of albendazole could be tested. During the present study, no reaction was recorded, even in patients with very high mf counts, but Blum *et al.* (2001) reported recently a case of encephalopathy which occurred on the third day of an albendazole treatment in a patient harbouring a low *Loa* microfilaraemia (152 mf/ml). This event should lead to further close monitoring in order to evaluate the safety of albendazole in patients infected with *Loa loa.*

The mechanisms by which albendazole reduces the *Loa* microfilaraemia is not known, but the slow rate of mf reduction observed in Cameroon and in Benin (Klion *et al.*, 1993), and the observations made on other filarial species, suggest that the drug is not

microfilaricidal, but acts either on the longevity, or on the reproductive capacities of the adult worms. This is an important point to consider because, in such a case, the decrease in the mf loads would then be perceptible only after a given delay, which, as emphasized by Cline *et al.* (1992), would mainly depend on the lifespan of the mf: the longer this is, the later would its effect be perceptible.

Concerning *Loa loa,* information on mf lifespan is very scarce. It is known that *Loa* mf can survive for more than 21 days in blood banked at 1-6° C (AuBuchon & Dzik, 1983). Besides this, several attempts have been made to transfer *Loa* mf to uninfected hosts: Fülleborn (1908) performed such experiments with dogs, and found that the mf had disappeared after one month, whereas Gönnert (1942), having injected *Loa* mf from a Cameroonian patient into himself, did not find the mf in his blood at the third day after injection. More interesting information could be drawn from the studies performed by Duke (1960), who followed up the course of infection in drills experimentally infected with human *Loa*, and then splenectomized. Following the removal of the spleen, an organ which suppresses the *Loa* mf from the peripheral blood in monkeys, the author observed a rapid rise in the mf densities, which reached a plateau some six months after the operation. One may assume that at that time, there was an equilibrium between the newly produced mf and those dying from old age, and that the lifespan of the *Loa* mf is in the order of six months. This estimate would be useful as part of the further trials to determine whether two- or three-day regimens of albendazole might finally solve the problem of ivermectin-induced *Loa* encephalopathy. Should the results be conclusive, they would give information on the interval of time to be respected between the albendazole and the ivermectin treatments.

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