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"Antimalarial" medicinal plants and their impact on cell populations in various organs of mice

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Summary

We have investigated the effects of leaf and bark decoctions of *Ocimum gratissimum, Azadirachta indica, Morinda lucida* and *Enantia chlorantha* on (a) the course of *Plasmodium yoelii nigeriensis* malaria (b) reticulocyte and haematocrit values and (c) nucleated cell numbers in the spleen, bone marrow, peritoneum, liver and peripheral blood of Swiss albino mice.

Results obtained showed that normal mice infected with the parasite (10^4/mouse) suffered fulminant parasitaemia which resulted in death, 7-10 days later. All infected mice treated with chloroquine survived. On the other hand all infected mice treated with the medicinal plants exhibited varying percentages of chemosuppression of early parasitaemia which did not lead to their survival. The total number of nucleated cells in the liver, spleen and peripheral blood of malaria-infected mice increased enormously before the animals died. Such increases were maintained in other groups of mice treated with the medicinal plants.

In the non-infected mice, *O.gratissimum* and *E. chlorantha* administration increased the number of nucleated cells in the spleen, liver and peripheral blood. Chloroquine on the other hand decreased the number of nucleated cells in both the malaria-infected and un-infected mice. There was also a decrease in reticulocyte numbers in the blood of normal mice injected with chloroquine. Conversely reticulocyte numbers increased in normal mice administered with some medicinal plants. Acute and chronic toxicity tests revealed that some of the medicinal plants were much more toxic than others.

It is concluded that some "antimalarial" medicinal plants exhibit properties which could be deleterious to health if dosages are not well-controlled.

Résumé

Nous avons mené une enquêté sur les effets de feuille et d'écorce dans la décoction de *Ocimum gratissimum, Azadirachta indica, Morinda lucida* et *Enantia chlorantha* sur (a) la parti de *Plasmodium yoelii nigeriensis* paludisme (b) reticulocyte et less valeurs de d'haematocrite et (c) le nombre de cellule périphérique des souris-suisses-Albinos.

Les resultats obtenus ont montré que souris infectées par le parasite (10^4/un souris) ont été victime du parasitaemia fulminant qui a causé leur mort 7 – 10 jour aprés. Toutes les souris infectées aprés avoir recu le traitement de chloroquine ont survecu. D'autre part, toutes les souris infectées qu'ayant recut le traitement de plante medicinales ont montré pluisieurs varietés de pourcentages du suppression chemo de parasitaemia prematuré qui ne leur a pas permi de survivre.

Le nombre total des cellules nucleiques dans le foie et la rate et le sang périphérique des les souris infectées par le paludisme a augmenté enormément avant la mort de des animaux. De telles augementation ont été entretenues dans les autres groupes des souris ayant été traité par less plantes médicinales. Dans le cas des souris n'ayant pas été infectees l'injection de *O.gratissimum* et *E.chlorantha* a augmenté le nombre des cellules nucleiques dans la rate, le foie et le sang périphériques. La chloroquine de l'autre coté a fait diminuer le nombre des souris atteintes de malaria et telles qui ne le sont pas. Il ya aussi une diminution de reticulocyte dans le sang du souris normales traite a la chloroquine. Inversement le nombre de reticulocyte a augmenté chez les souris traitees de plantes medicinales.

Des tests de toxicité aigus et chroniques ont montré certaines plantes medicinales sont plus toxique que d'autres.

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Il est a noter comme conclusion que certaine plantes antipaludeemes presentent les propriétés qui sont nefaster a la santé si la dose n'est pas respectée.

Introduction

The use of herbal decoctions and extracts of medicinal plants for the cure of malaria could be traced to the early man [1]. Despite all efforts being made by both the orthodox and traditional medical practitioners to eradicate malaria, the infection has remained one of the greatest causes of debility and mortality, particularly in Africa. Sometime in 1983, there was an increased support for traditional healers by the Nigerian Government. This also brought about increased patronage by the general public. In most cases, decoctions of roots, leaves and barks were taken without reference to dosage limits.

A large number of plants claimed by traditional healers to be effective against malaria has been documented [2]. Many scientists have also carried out series of investigations to establish the authenticity of these claims [3-8]. Ekanem [3] found *Azadirachta indica* effective whereas other workers observed little or no chemosupression when the leaf extract was used either alone or in combination with other medicinal plants [1,4,5]. *Morinda morindoides* has also been reported to terminate the growth of *Plasmodium berghei* in mice [6]. However, Obi, Makinde and Laoye [7], working with the stem, bark and leaves of *Morinda lucida* found only the chromatographic fraction of the stem bark most promising. In a later report however, *Morinda lucida* leaf extract was found to exhibit schizontocidal and repository effects in mice infected with *Plasmodium berghei berghei* [8], a report that was not confirmed by others [1].

Enantia chlorantha has not been found effective against malaria [1]. Furthermore, the claimed antimalarial effect of *Ocimum gratissimum* has not been seriously investigated except in the present report. Investigations on the status of chloroquine in the host on the other hand have been carried out [9, 10] but not to the extent of studying their effect on cell proliferation changes in the bone marrow, liver, spleen and the peritoneum.

In our present work, we felt that there was the need not only to screen for the antimalarial effect of some of the medicinal plants but also to investigate biochemically, haematologically and immunologically, other possible effects of these decoctions in the organs of the host when administered orally, intramuscularly, subcutaneously and intraperitoneally. Our findings regarding oral administrations of leaf decoctions and their effect on the normal and malaria-infected hosts are presented here.

Material and methods

Parasites

Plasmodium yoelii nigeriensis [11] was maintained in our laboratory by blood passage. A standard dose of 10^4 parasitized red blood cells (RBC) was inoculated intraperitoneally and parasitaemia was assessed from Giemsa- stained thin blood films.

Host animals

Adult Swiss albino mice aged 6-8 weeks and weighing about 20gm were used. They were bred in our Institute and fed standard diets (Pfizer Livestock Feeds Ltd; Ikeja).

Preparation of cells from various organs, for nucleated cell counts.

A large pool of mice were used for these experiments since some of them had to be sacrificed at intervals. The mice to be used were anaesthetized with diethyl ether and then bled to death before extraction of the various organs. The cells from the peritoneum (PC), spleen (SPL), bone marrow (BM) and Liver (LIV) were extracted according to standard procedures [12]. Briefly, the spleen and liver were taken into petri dishes containing ice- cold sterile phosphate buffered saline (PBS). They were teased up with sterile scissors before the extracted cells were decanted into sterile, heparinized centrifuge tubes in ice buckets. The femur was used as a source of bone marrow cells. The muscles attached to the bone were removed with sterile cotton gauze swabs and each end of the femur bone was cut with sharp scissors. The marrow was then flushed out into sterile centrifuge tubes by thrusting a 23-25 gauge needle into the marrow cavity and pushing about 1 ml cold PBS through the cavity with a sterile syringe. Peritoneal cells were obtained by first injecting the cold sterile PBS into the peritoneum of each mouse and then recovering the liquid into sterile centrifuge tubes.

All cells were washed twice with cold PBS, centrifuged each time at 1000 rpm for 10 minutes at 4°C and re-suspended. Nucleated cell counts were estimated using the crystal violet-acetic acid- diluted specimens in a haemocytometer. At least 5 in-bred mice were used for each experiment and cells from each organ were pooled.

Haematocrit values

Venous blood from each mouse was collected in heparinized capillary tubes and the packed cell volumes

(PCV) were obtained by centrifuging the capillary tubes (sealed at one end) in a haematocrit centrifuge and reading off the values with a haematocrit reader.

Reticulocyte counts

Venous blood from each animal was diluted 1 in 2 with 1% brilliant cresyl blue in citrate-saline solution, incubated for 20 minutes at 37°C and smeared on a microscope slide. The number of reticulocytes was expressed as a percentage of the total red blood cells.

Preparation and administration of chloroquine and medicinal plants

Chloroquine was obtained from the National Institute for Medical Research (NIMR) clinic as chloroquine phosphate (250 mg tablet, containing 150 mg base). Each tablet was ground with sterile pestle and mortar and "dissolved" in 100ml sterile distilled water with gentle heating. A series of acute toxicity experiments had been performed in our laboratory over several months to determine both the minimum effective dose (MED) on malarial infection and the maximum tolerated dose (MTD) of chloroquine phosphate in groups of young mice. We confirmed that oral (force-feeding), intraperitoneal (IP) or intramuscular (IM) administration of chloroquine phosphate up to 80 mg/kg body weight of mouse caused no death whereas 90 mg/kg body weight or higher, was lethal 24 hours later. After these preliminary studies we de-

Table 1: Summary of our method of preparation and administration of the medicinal plants.

Drug or Medicinal Plant	Method of Preparation	Maximum tolerated dose (MTD) Per mouse/day	Dose administered/ mouse (4 consecutive days.)	Route of admin.	Wt. and age of mice	Acute toxicity as compared with H_2O or saline
Chloroquine phosphate (250mg or 150mg base)	Tablet ground with sterile pestle and mortar, "dissolved" in 100ml sterile distilled H_2O	0.65ml (approx. 80mg/kg)	0.5ml (62.5mg/kg or 37.5mg base/kg)	oral (force-feeding)	20mg (mean) 6-8 wks old	++
O.gratissimum	Reflux condensation method 100mg fresh, ground leaves, boiled in 200ml H_2O for 1 hr. Filtered hot and cooled	0.6ml stock decoction (i.e. 12,500mg/kg)	0.3ml stock (7,500mg/kg mouse)	"	"	++
A. indica	Fresh leaves (100gm) treated as in *O. gratissimum*	0.5ml of the twice concentrated stock solution (i.e. 25,000mg/kg)	0.3ml of twice concentrated stock (15,000mg/kg mouse)	"	"	+
M. lucida	Fresh leaves (100gm) treated as in *O. gratissimum*	0.7ml twice concentrated stock (i.e. 35,000mg/kg)	0.5ml of twice concentrated stock solution (25,000mg/kg)	"	"	+
E. chlorantha bark	Dry bark, ground with sterile pestle and mortar, 30gm bark extracted with 100ml H_2O.	0.3ml of 1:5 diluted stock solution (900mg/kg)	0.2ml of 1:5 dilution (600mg/kg)	"	"	+++

The doses shown in mg/kg are based on the wet wt. of fresh leaves and for *E. chlorantha*, on the dry wt.

cided to administer for 4 consecutive days, a safe volume of 0.5ml of the above preparation which in a 200gm mouse was equivalent to 62.5 mg/kg (or 37.5 mg base/kg).

Table 1 shows the method we used for the preparation and administration of chloroquine phosphate and the medicinal plants. The bark and fresh leaves of the medicinal plants were purchased from Iddo and Tejuosho markets. Some of the leaves were obtained from plants growing at the NIMR compound. They were identified as the correct species at the University of Lagos. The months (seasons) in which they were collected and used for each experiment were also noted. The decoctions were prepared to resemble the neat preparations consumed by some Nigerians for treatment of malaria. The MTD and the doses administered for 4 consecutive days are as shown in Table 1.

The 4-day suppressive test (4-DST)

The 4-DST as standardized by the World Health Organization [13] was applied in assessing the results. Here, each drug or decoction was given one hour after infection of the parasite. This was repeated daily for the next 3 days (D+0 – D+3), i.e, 4 consecutive administrations. Parasitaemia was monitored and the mean percentage suppression of early infection (day 6) was expressed as follows:

$$\text{Mean \% chemosuppression} = \frac{\text{Mean \% Parasitaemia in untreated controls} - \text{Mean \% Parasitaemia in treated groups}}{\text{Mean \% Parasitaemia in untreated controls}} \times 100$$

Similarly, normal mice without malarial infection were given the same treatment for 4 consecutive days.

Results

Effect of leaf and bark decoctions and chloroquine on parasitaemia.

The mean percentage chemosuppression of early (day 6) parasitaemia in treated mice is shown in Table 2. The chemosuppression by chloroquine was almost 100% as only one or two animals out of 10 exhibited transient parasitaemia of about 0.5% at this time. The parasitaemia eventually cleared and all animals survived. Conversely, parasitaemia in untreated mice increased linearly until all the animals died between days 7 and 10 (i.e. starting with 10^4 *P. yoelii nigeriensis* per mouse and not $2x10^7$ which some workers use [6]. Although *A. indica* and *M. lucida*

Table 2: Mean % chemosuppression (±SD) in *P.yoelii nigeriensis* infected mice treated with various decoctions of medicinal plants (day 6 after first treatment in the 4-day suppressive test).

	Normal mice	Chloroquine phosphate 62.5mg/kg	*O.gratissimum* 7500mg/kg	*A.indica* 15,000mg/kg	*M.lucida* 25,000mg/kg	*E.chloranthа* 600mg/kg
Mean % Parasitaemia on day 6	22.0±1.0	0.1±0.05	25.0±0.5	16.0±1.5	14.0±1.2	21.0±2.0
Mean % suppression of parasitaemia	0	99.5±2	−13.6±2.3	27.3±6.8	36.4+5.5	4.5±9.1
Mean % No of mice used	20	10	10	10	10	15
% of mice surviving on day 15	0	100	0	0	0	0

$$\text{Mean \% suppression of Parasitaemia} = \frac{\text{Mean \% Parasitaemia in untreated controls} - \text{Mean \% Parasitaemia in treated groups}}{\text{Mean \% Parasitaemia in untreated controls.}} \times 100$$

exhibited appreciable chemosuppression of parasitaemia (27.3% and 36.4%) respectively), the animals administered the decoctions subsequently died from fulminant *P. yoelii nigeriansis* infection. No suppression of parasitaemia was observed in *E. chlorantha* bark. Furthermore, *O.gratisssimum* on the other hand, seemed to encourage rather than suppress parasite growth (Table 2).

Total nucleated cell counts

1. *Malaria – infected mice.* The nucleated cell counts in the peripheral blood, spleen and liver of non- treated mice increased enormously until the animals died (Table 3). The administration of medicinal plants did not decrease the cell prolifereation. In fact, further increases were observed in some organs as a result of medicinal plant administration. The administration of chloroquine phosphate on the other hand decreased the spleen, liver and peripheral blood cell counts. Apart from *M. lucida* which increased the number of bone marrow cells, the counts from the bone marrow and peritoneum remained normal.

2. *Non-infected mice.* Chloroquine administration decreased the nucleated cell counts in the peripheral blood, the spleen and the liver and, to a certain extent, the bone marrow and the peritoneum of normal mice (Table 4). A follow-up however revealed that the total number of nucleated cells returned to normal within ten days, except in cases where chloroquine administration continued for more than 4 days. The administation of *Ocimum gratissimum* and *E. chlorantha* increased the cell counts in the liver. All the medicinal plants also caused slight increases in the number of nucleated cells in the peripheral blood. However, prolonged administration of the medicinal plants over and above the study period brought about a decrease in cell counts and subsequent anaemia (assessed by PCV measurements) in most of the mice.

Table 3: Malaria-infected mice: mean nucleated cell count (mL^{-1} ± SD) from various organs (day 6 after first treatment with chloroquine and decoctions in the 4-day suppresive test)

	Normal mice[1]	Infected mice[2]	Chloroquine phosphate (62.5mg/kg)	*O.gratissimum* (7500mg/kg)	*A. indica* (15,000mg/kg)	*M. lucida* (25,000mg/kg)	*E. chlorantha* (600mg/kg)
SPL	$95x10^6$ ± $1.5x10^6$	$190x10^6$ ± $4.1x10^6$	$82x10^6$ ± $1.92x10^6$	$180x10^6$ ± $3.2x10^6$	$170x10^6$ ± $4.5x10^6$	$165x10^6$ ± $3.1x10^6$	$175x10^6$ ± $2.9x10^6$
BM	$15.2x10^6$ ± $0.7x10^6$	$15.7x10^6$ ± $0.4x10^6$	$14.1x10^6$ ± $0.5x10^6$	$14.1x10^6$ ± $0.6x10^6$	$15.0x10^6$ ± $0.3x10^6$	$13.9x10^6$ ± $0.4x10^6$	$14.8x10^6$ ± $0.7x10^6$
PC	$0.5x10^6$ ± $0.02x10^6$	$0.68x10^6$ ± $0.05x10^6$	$0.45x10^6$ ± $0.03x10^6$	$0.54x10^6$ ± $0.04x10^6$	$0.54x10^6$ ± $0.04x10^6$	$0.62x10^6$ ± $0.03x10^6$	$0.7x10^6$ ± $0.06x10^6$
LIV	$5.3x10^6$ ± $0.4x10^6$	$16.3x10^6$ ± $0.5x10^6$	$4.0x10^6$ ± $0.2x10^6$	$15.4x10^6$ ± $0.6x10^6$	$13.5x10^6$ ± $1.0x10^6$	$14.0x10^6$ ± $0.9x10^6$	$15.9x10^6$ ± $0.7x10^6$
PBL	$15x10^6$ ± $0.8x10^6$	$28.7x10^6$ ± $0.8x10^6$	$12.8x10^6$ ± $1.0x10^6$	$25.2x10^6$ ± $0.9x10^6$	$23.8x10^6$ ± $0.5x10^6$	$21.3x10^6$ ± $0.7x10^6$	$26.5x10^6$ ± $1.2x10^6$

1. Normal, uninfected mice with no administration of chloroquine or decoction
2. (a) *P. yoelii nigeriensis* infected mice with no drug/decoction
   (b) Number of *P. yoelii nigeriensis* infected was $1x10^4$/mouse

KEY:

SPL: spleen cells; BM: bone marrow cells; PC : peritoneal cells
LIV: Liver cells; PBL: peripheral blood cells.
Note: The data presented are the mean counts/mL (± SD) from at least 5 experiments involving pooled organs (e.g. pooled BM) in each experiment.

2.1. *Peripheral blood reticulocyte counts and haematocrit values in non-infected mice.* Administration of chloroquine phosphate (62.5 mg/kg) for 4 consecutive days caused a decrease in both the reticulocyte counts (Table 5) and haematocrit values (Table 6). The values however returned to normal 10 to 15 days later. *E. chlorantha* bark on the other hand increased the reticulocyte count but decreased the haematocrit values. The administration of other medicinal plants did not seem to affect the reticulocyte counts and haematocrit values ($P = 0.1$). The interpretation of these results could not be made but further work is in progress to see if varying experimental conditions could change the results.

Table 4: Non-malaria infected mice: Mean nucleated cell counts ($mL^{-1} \pm SD$), day 6 after first administration with chloroquine and medicinal plants for 4 consecutive days

	Normal mice[1]	Chloroquine phosphate (62.5mg/kg)	*O.gratissimum* 7500mg/kg	*A. indica* 15,000mg/kg	*M. lucida* 25,000mg/kg	*E. chlorantha* 600mg kg
SPL	$95x10^6 \pm 1.5x10^6$	$80x10^6 \pm 1.2x10^6$	$102.3x10^6 \pm 2.1x10^6$	$98.5x10^6 \pm 1.1x10^6$	$87.6x10^6 \pm 1.8x10^6$	$105x10^6 \pm 3.0x10^6$
BM	$15.2x10^6 \pm 0.7x10^6$	$12.5x10^6 \pm 0.3x10^6$	$13.7x10^6 \pm 0.5x10^6$	$14.5x10^6 \pm 0.6x10^6$	$13.0x10^6 \pm 0.5x10^6$	$15.4x10^6 \pm 0.4x10^6$
PC	$0.5x10^6 \pm 0.02x10^6$	$0.45x10^6 \pm 0.03x10^6$	$0.55x10^6 \pm 0.05x10^6$	$0.48x10^6 \pm 0.06x10^6$	$0.44x10^6 \pm 0.02x10^6$	$0.67x10^6 \pm 0.04x10^6$
LIV	$5.3x10^6 \pm 0.4x10^6$	$4.9x10^6 \pm 0.2x10^6$	$8.4x10^6 \pm 0.5x10^6$	$6.7x10^6 \pm 0.7x10^6$	$5.9x10^6 \pm 0.3x10^6$	$8.3x10^6 \pm 1.1x10^6$
PBL	$15.0x10^6 \pm 0.8x10^6$	$13.0x10^6 \pm 0.5x10^6$	$18.9x10^6 \pm 0.7x10^6$	$19.2x10^6 \pm 0.4x10^6$	$17.9x10^6 \pm 0.5x10^6$	$20.8x10^6 \pm 0.6x10^6$

1. Normal mouse with no drug or decoction administration.

KEY:

SPL: spleen cells; BM: bone marrow cells; PC : peritoneal cells;
LIV: Liver cells; PBL: peripheral blood cells;
Note: The data presented are the mean counts/mL (± SD) from at least 5 experiments involving pooled organs (e.g. pooled BM) in each experiment.

Table 5: Mean reticulocyte values (% ± SD) of normal uninfected mice administered the medicinal plants*

	Days after first drug administration					
	0	1	3	5	10	15
Normal mice	4.8±0.4	4.7±0.6	4.9±0.8	5.0±0.5	4.8±0.7	4.9±0.9
Chloroquine	5.1±0.5	4.5±0.4	3.9±0.4	3.1±0.6	3.5±0.7	5.2±0.4
O.gratissimum	4.7±1.0	5.1±0.8	4.8±0.3	5.2±0.6	5.4±0.4	4.7±0.7
A. indica	4.9±0.6	5.5±0.4	5.8±0.6	5.7±0.7	5.8±0.4	5.9±0.6
M. lucida	5.5±0.8	6.0±0.4	6.2±0.5	6.5±0.4	5.9±0.6	6.1±0.5
E. chlorantha	4.5±0.7	5.7±0.9	5.7±0.4	6.4±0.3	6.7±0.2	6.0±0.5

* At least seven mice were used for each drug or medicinal plant.
* Drugs were administered for 4 consecutive days from day 0.

Table 6: Mean haematocrit values (% ± SD) of normal uninfected mice administered the medicinal plants*

	Days after first drug administration					
	0	1	3	5	10	15
Normal mice	44.5±0.3	45.8±4.0	45.0±0.3	44.1±0.4	45.2±0.2	44.9±0.4
Chloroquine	44.9±0.3	42.1±0.3	42.5±0.2	41.2±0.1	39.7±0.2	40.5±0.3
O.gratissimum	45.2±0.2	52.0±0.1	48.5±0.3	50.1±0.2	47.5±0.3	46.9±0.2
A. indica	45.0±0.2	43.1±0.4	45.8±0.5	45.6±0.3	44.5±0.4	43.8±0.5
M. lucida	44.6±0.3	43.9±0.5	43.0±0.2	43.6±0.4	44.0±0.2	43.9±0.5
E. chlorantha	44.9±0.4	45.0±0.4	41.2±0.3	40.9±0.5	40.2±0.2	41.5±0.4

* At least seven mice were used for each drug or medicinal plant.
* Drugs were administered for 4 consecutive days from day 0.

Toxicity of the medicinal plants

When we compared the effect of the minimum toxic volumes of our preparations with equivalent volumes of water or physiological saline, it was seen that *E. chlorantha* bark was the most toxic followed by chloroquine or *O. gratissimum*. Although *A. indica* and *M. lucida* are as bitter as *E. chlorantha* bark, they were less toxic than the rest of the "antimalarial" decoctions (Table 1).

Discussion

Many scientists have investigated the suppressive effects of *Azadirachta indica* [1, 3, 4], *Morinda morindoides* [6], *Morinda lucida* [1, 7, 8], *Enantia chlorantha* [1] and chloroquine [10] in human and in mouse malaria. Some workers have found the plants effective while others have not (see introduction). Our results on suppression however seem to agree with those of others [3, 8] who showed that *A. indica* [3] and *M. lucida* [8] exhibit some antimalarial activity. In our experiments however, all animals treated with the decoctions eventually died from the infection. The ability of others [1] to detect activity only in *Plasmodium gallinaceum* in chicks but not in mice could be as a result of differences in experimental protocol. We found that the 4 – day suppressive test was best studied when low concentrations of the parasites (1×10^4/mouse) were introduced. The suppressive effects of *M. lucida* and *A. indica* were non-existent with high concentrations of *P. yoelii nigeriensis* infection (e.g. 2×10^7/mouse) or, in established infections with high parasitaemia.

Our main objective in this work however was to determine the acute and cumulative effects of the medicinal plants in various organs of the host. The MTD results in this report (Table 1) were not based on the cumulative effect caused by the administration of the drug or decoctions for 4 consecutive days but on the single doses which did not kill the mice within 24 hours. The various doses chosen for the experiments (i.e. lower than the MTDs) were those found to be cumulatively safe for all mice after a 4–day administration.

The increases in the nucleated cell counts in the liver, spleen and peripheral blood during *P. yoelii nigeriensis* infection were consistent observations. Chloroquine administration abrogated this increase right from the outset although it also caused a decrease on continued administration. The depression of cell counts by chloroquine agrees with the *in vitro* work of others [9] who reported that chloroquine profoundly suppressed the proliferation of mitogen and antigen – stimulated cells as indicated by decreased ^{14}C – thymidine incorporation.

The increases in cell counts observed during *P. yoelii nigeriensis* infection were not diminished by decoctions of medicinal plants, thus confirming that parasites were not being eradicated. We could not interpret the increases in nucleated cell counts observed when some of the normal mice (i.e. without parasite infection) were given the required doses of the decoctions. Further work is in progress to see if varying the experimental conditions could alter the results.

Conclusion

Toxicity studies have been carried out on chloroquine and 4 medicinal plants used as antimalarials.

We have shown within this context, that *E. chlorantha* bark was the most toxic, followed by chloroquine and *O. gratissium*. *A. indica* and *M. lucida* were less toxic than the rest of the specimens. We have also shown that chloroquine and some of the "antimalarial" medicinal plants depressed normal cell proliferation and induced anaemia on continued administration. Some, on the other hand, abnormally increased cell counts. We are still investigating histologically and biochemically, the possibility of damage to the liver, spleen and other organs as a result of the frequent use of the "antimalarial" medicinal plants. Our results however warn that as orthodox medical profession keeps strictly to the right dosages for chloroquine administration, the need to regulate the intake of these medicinal plants cannot be overemphasized.

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