

A COMPARATIVE EVALUATION OF CADMIUM AND LEAD IN SOME LAGOS DUMPSITES

ADELEKE ADENIYI
JUBRIL FASHOLA,
AND
OKON EKANEM

ABSTRACT

The Cd and Pb level profile of soils in some dumpsites in Ojo and Apapa areas in the outskirts of Lagos and non-dumpsites about 50 metres away from the dumpsites have been studied. The total Cd range between 0.52 to 1.96 ug/g with a mean of 1.13 ug/g for the dumpsites, while Cd level of the non-dumpsites range from 0.02 to 1.40 ug/g with a mean of 0.57 ug/g. The levels of Pb was in the range of 20.90 to 628.00 ug/g with a mean of 262.81 ug/g for the dumpsites, and the non-dumpsites range from 5.60 to 160.00 ug/g with a mean of 27.99 ug/g. The results indicate higher levels of these heavy metals in the dumpsites. Soil pH and moisture content were also determined. The pH values ranged between 6.8 to 7.9 (a mean of 7.3) in the dumpsites and 5.2 to 7.9 (mean of 7.0) in the non-dumpsites. Moisture content was in the range of 4.82 to 11.20 (mean: 8.62) and 3.71 to 11.70 (mean: 7.90) in the dumpsites and non-dumpsites, respectively.

INTRODUCTION

Soils are important to Man primarily because plants grow in them, and without plants there could be no food and fiber for animals and humans. It is therefore imperative that Man be concerned, among other things, about the heavy metal profile of soils since there is a relationship between the content of these elements in soil and their concentration in plant tissues (Demir et al (1990) and Maliugo (1941)). Davies (1980) proposed that pH, moisture, organic matter content, pore space, type and size of soil microbial life will have effects on the availability of metals to plants.

Some metals are essential for life, examples are Mn, Fe, Cu and Zn which are essential macronutrients, others such as Hg, Cd and Pb are not required even in small amount by living organism as they are known to be toxic to Man at relatively low levels (Tyler (1981)). Cd and Pb like other heavy metals, accumulate in the biological systems. Schroder (1965) in a study, linked Cadmium with hypertension. Whereas Ruthven and Cairns (1967) determined the minimal lethal concentrations and maximum tolerated concentrations of Pb as $PbNO_3$ for six species of freshwater algae and protozoan. The different organisms show different tolerance level; for *Paranema* (Mastigophora) and *Euglena gricitis* (100 mg/L) to *Chilomonas* (5.6 mg/L). Copper and Steinberg (1977) have demonstrated that both Cholinergic and adrenergic synaptic evoked transmitter release is inhibited by the presence of Pb.

The objective of this study is to compare the concentrations of Cd and Pb in the dumpsites and non-dumpsites with a view of estimating the extent of pollution caused by the waste materials dumped in these sites. It will also be of interest to know whether or not the levels of metals depend on the nature of waste materials dumped in these sites.

MATERIALS AND METHOD

The soil samples were collected from the various dumpsites and non-dumpsites with the aid of the soil auger. Each sample is a composite sample made up of five auger samples taken randomly within a depth of 15 cm from the soil surface.

Extraction of Cd and Pb from Soil Samples: 5g of air dried soil passed through 0.5 mm sieve was weighed into a 100 ml round bottom flask, 30 ml of 2N HNO₃ was added to the soil sample. The content of the flask was shaken vigorously for 5 minutes and immersed in a water bath (at 100°C) for two hours. The solution was then filtered into a 100 ml standard flask and made up to mark with deionised water. A blank containing deionised water was treated in the same manner as above without any sample.

Method of Determination by Atomic Absorption Spectrophotometer:

The instrument was adjusted to zero with the blank solution and then adjusted to read the concentration of the standard of the element being determined. The gas oxidant flow rates, flame conditions, wavelength and slit width were set according to the specification given in the instrument manual for Cd and Pb respectively.

Soil pH Determination: This was done in accordance with the method described by Schofield and Taylor (1955). To 10 g of air-dried soil (passed through a 2 mm sieve) in a 50 ml beaker was added, 0.01 M CaCl₂ and allowed to stand for 30 minutes with occasional stirring with a glass rod. The pH was then measured with a standardised Griffin pH meter, model 290.

Determination of Moisture Content: The moisture content was determined immediately the soil samples were collected from the sites, using 20 g of each wet sample at 110°C. the values were obtained from the relationship:

$$\text{Moisture content [\%]} = \frac{W_w - W_d}{W_d - W_c} \times 100$$

Where,

W_w = Weight of crucible and wet soil

W_c = Weight Crucible

W_d = Weight of Crucible and oven dried soil

RESULTS

The results of the analyses for cadmium and lead in the soils studied are listed in Table 1. While the pH values, percentage moisture of the soil samples are given in Table 2.

TABLE 1: CONCENTRATIONS OF Cd AND Pb IN THE SOIL SAMPLES

SAMPLE NO.	Cd[ug/g]	Pb [ug/g]
1.	1.30 [0.02]	628.00 [12.00]
2.	1.35 [1.01]	436.08 [8.20]
3.	1.00 [0.70]	146.04 [12.00]
4.	1.80 [1.30]	192.60 [5.60]
5.	1.32 [0.02]	332.00 [5.40]
6.	1.96 [1.40]	252.40 [160.00]
7.	0.72 [0.260]	226.00 [16.10]
8.	0.36 [0.25]	220.10 [24.60]
9.	1.00 [0.52]	20.90 [12.00]
10.	0.52 [0.23]	20.90 [12.00]
MEAN	1.13 [0.57]	262.81 [27.99]

Notes: Samples 1—5 are from Apapa sites, while 6—10 are from Ojo sites. The values in parentheses are for the non-dumpsites.

TABLE 2: SOIL pH VALUES AND MOISTURE CONTENT [%]

SAMPLE NO.	pH	% MOISTURE CONTENT
1.	7.2 [7.9]	8.02 [8.44]
2.	7.8 [5.2]	7.07 [3.71]
3.	6.9 [6.7]	9.03 [6.87]
4.	7.0 [6.6]	8.11 [7.08]
5.	6.8 [6.3]	4.82 [5.01]
6.	7.2 [7.4]	11.20 [9.30]
7.	7.7 [7.5]	8.10 [9.00]
8.	7.9 [7.6]	10.40 [8.00]
9.	7.0 [7.2]	8.60 [9.90]
10.	7.1 [7.3]	10.80 [11.70]
MEAN	7.3 [7.0]	8.67 [7.90]

Notes: Apapa sites [1—5]; Ojo sites [6—10] Figures in parentheses are for the non-dumpsites.

DISCUSSION

The Cd and Pb levels (Table 1) reveal that the concentrations of these heavy metals are as expected, higher in the dumpsites than the non-dumpsites. This is due to the presence of domestic and industrial waste materials at the dumpsites. Generally, the Pb levels are higher than the Cd levels, this may be related to the vehicular traffic density as shown by Harrison and Williams (1982), and Davies *et al* (1979). The Pb concentration in site 1 (Table 1) is particularly high when compared with others. A look at the history of the dumpsite reveal that it has been in use for about twenty years. Epstein [1965] showed that heavy metals accumulate on exchange sites and are very difficult to remove from the soil. They consequently deplete the soil of essential elements such as potassium and sodium, which are vital to plants. Plants also remove these toxic heavy metals when grown in these areas [Zimdahl *et al*, 1978]. The concentrations removed by the plants are relatively low, but since these metals have a very long half-life in the biological systems, they accumulate when these plants are consumed by animals and eventually by Man.

The pH values obtained [6.3 – 7.9], Table 2, are within the range predicted by Epstein [1965] for most soils i.e. between 4 to 8. Uptake of all metals except pB generally decrease with increase in soil pH [Sign and Narval (1984)].

From the values of the percentage moisture content [Table 21, it is evident that the soils were quite moist and had the effect of decreasing the pH due to the dissolution of SO₂ and CO₂ from the atmosphere [Tyler (1981)].

REFERENCES

- Anderson, A. [1976] *J. Agric. Res.* 6(1), 19.
- Cooper, G.P. and Steinberg, D. [1977], *Amer. J. Physiol.* 232, C, 128.
- Davies, B.E. [1980], "Applied Soil Trace Elements", John Wiley, New York, pp. 1-17.
- Davies, B.E. and Holmes, P.L. (1972), *J. Agric. Sci. Camb.*, 79, 890.
- Demir, M. Gucer, S. and Esen, T. [1990], *J. Agric. Food. Chem.* 38, 3, 726.
- Epstein, E. [1965] "Mineral Metabolism" Chp. 18 [pp. 438-466] in *Plant Biochemistry*. Eds. Bonner J. and Varner J.E. Academic Press, New York.
- Harrison, R.M. and Williams, C.R. (1982), *Atmospheric Environ* - 16(11), 2669.
- Maliuga H.J [1941]. *Compt. Rend. Acad. Sci. USSR* 31, 145. C.A. 284A [1941]

Ruthven, J.A. and Cairns, J. [1973], *J. Protozool.* 20, 127.

Scholfield, R.K. and Taylor, A.W. [1955],
J. Soil Sci. 6; 13.

Schroeder, H.A. [1957] "Mechanism of Hypertension"
Charles Thomas, Springfield, Illinois.

Schroeder, H.A., Nason, A.P. and Mitchener, M., [1981],
J. Physiol., 214, 796.

Signh, B.R. and Narval, R.P. (1984), *J. Environ. Qual.*, 13(3), 344.

Tyler, T.G. (1981) "Soil Biochemistry", Marcel Dekker, New York, pp. 372 –
414.

Zimdahl, R.L. [1978], *Bull. Environ. Contam. Toxicol.*, 19, 431.

AN IN-VIVO EVALUATION OF THE MUTAGENICITY OF SULPHAMETHOXYPRIDAZINE: PYRIMETHAMINE

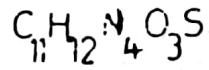
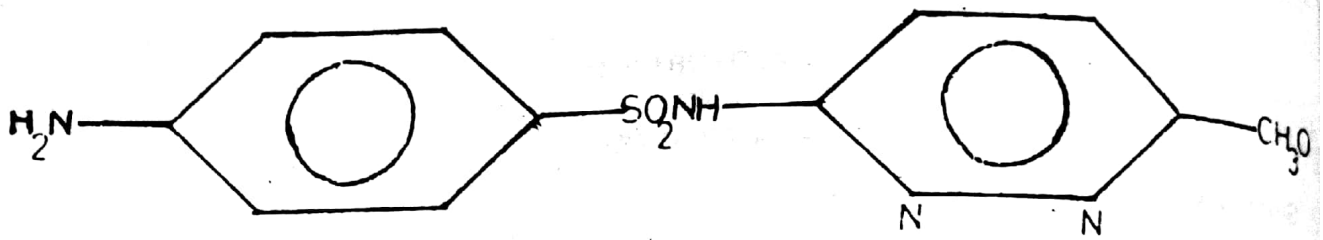
O. A. OTUBANJO
AND
A. A. MOSURU

Summary

Sulphamethoxypyridazine: Pyrimethamine (Metakelfin) is a newly developed antimalarial drug, whose effects on mouse sperm head morphology was evaluated in (U.I. Vet.) F1 mice. 5 different dose levels of 3.85: 0.19; 7.7: 0.38; 15.4: 0.77; 23.1: 1.13 and 30.8: 1.54 mg/kg body weight sulphamethoxypyridazine: pyrimethamine, respectively, were administered to the animals by a schedule of 5 consecutive daily intraperitoneal (.i.p.) injections. The sperm of the mice from the cauda epididymes were examined 5 weeks after treatment. Metakelfin induced sperm head abnormalities, however, the induction was not significantly elevated above the control value. Furthermore, the induction of the sperm head abnormalities was not strictly dose-dependent.

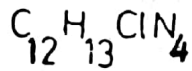
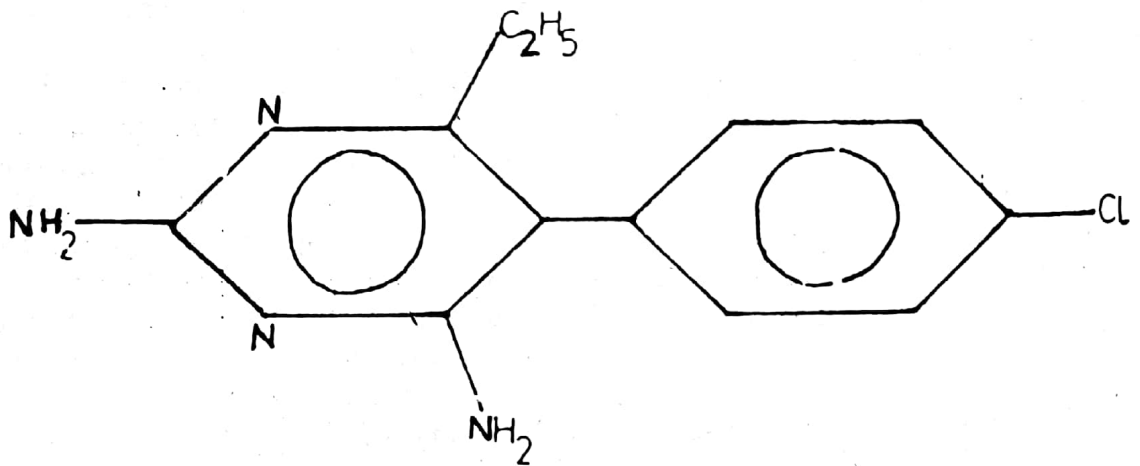
Introduction

Malaria continues to be the world's greatest disease in terms of morbidity and mortality, particularly in the underdeveloped third world tropical countries. Efforts have been made to control the disease. Chemotherapy has long been used as a means of control and many drugs have been developed that have potent antimalarial activities. Antimalarial chemotherapeutic drugs fall into two basic categories: the 4-aminoquinolines such as chloroquine and amodiaquine which are effective against blood parasites; and the antimetabolite antimalarials such as proguanil, pyrimethamine and sulpha drugs, which are effective against all other parasite stages except the blood phase. However, recent reports indicate parasite resistance to these drugs. Payne, (1987), reported that resistance of *Plasmodium falciparum* to chloroquine has now spread through Asia, Africa, South America and that resistance to other drugs is often present. A combination of sulphadoxine and pyrimethamine (Fansidar) has been used for chloroquine - resistant *falciparum* malaria, however, Fansidar - resistant *P. falciparum* now exist in a number of areas (Meek *et al*, 1986). This discovery of multi-drug resistance in various strains of *P. falciparum* led to the search for new potent antimalarial drugs and the search recently culminated in the development of a new combination drug containing sulphamethoxypyridazine and pyrimethamine (Metakelfin) Figs 1 and 2. Both anti-protozoal agents are particularly effective against *P. falciparum* as well as *P. malariae*, *P. vivax* and *P. ovale*. The reason for this is well understood. Pyrimethamine has been known to show strong potentiation when used in combination with sulphonamides due to the block at 2 points (double attack) lying in sequence along the same enzymatic metabolic pathway of the pathogen agent, (Rollo, 1955).



M.W. = 280.3

Fig. 1. Chemical structure of SULPHAMETHOXYPYRIDAZINE



MW = 248.7

Fig. 2. Chemical structure of PYRIMETHAMINE.

Sulphamethoxypyridazine is a long acting sulphomamide that is readily absorbed, distributed throughout the body and slowly but steadily excreted. The drug is protein - binding (Pharmaceutical codex, 1979). It is found in most body fluids, saliva, sweat, cerebrospinal, ocular, peritoneal, pleural, synovial and other fluids. Small amounts of the drug are taken up by the red blood cells. Pyrimethamine is also readily absorbed and slowly but steadily excreted from the body.

During the past two decades, increasing attention has been directed toward the determination of the mutagenic and carcinogenic potentials of drugs even though their useful therapeutic activities cannot be dissociated. It is desirable then that these indispensable agents be free from any deleterious effects. No reports exist of the mutagenicity and carcinogenicity of sulpha methoxypyridazine and pyrimethamine. (Metakelfin).

In this study, the sperm of metakelfin-treated mice were analysed to determine whether these drugs increased the incidence of abnormal sperm morphology (Wyrobek and Bruce, 1975).

Materials and Methods

Metakelfin (sulphamethoxypridazine: pyrimethamine) was supplied by Farmitalia Carla Erba, Italy.

U.I. Vet. F1 mice were obtained from the Veterinary Department breeding unit, University of Ibadan. The mice were housed in sterile animal cages in a pathogen-free environment, separated into test groups, kept at room temperature ($26 \pm 3^{\circ}\text{C}$) and were allowed food (Ladokun mouse cubes) and water *ad libitum*.

Assay of sperm abnormalities

Sulphamethoxypyridazine: pyrimethamine (Metakelfin) induction of sperm-head abnormalities was tested according to Wyrobek *et al*, (1983).

A suspension of metakelfin in normal physiological saline was freshly prepared before each experiment. Groups of 5 male mice, 12-14 weeks of age, received intraperitoneally one daily dose of the metakelfin suspension or of the vehicle solvent alone, as the negative control, for 5 days. The doses tested were 3.85: 0.19; 7.7: 0.38; 15.4: 0.77: 23.1: 1.13 and 30.8: 0.19: of sulphamethoxy pyridazine: pyrimethamine, respectively, / kg body weight/day for a period of 5 days. (The positive control was 100mg of methyl methanesulphonate/kg/day i.p. for a period of 5 days).

Single i.p. injections of 0.5ml of the suspensions or of the vehicle solvent alone were administered to the mice daily. After an exposure period of 5 weeks, the mice were sacrificed by cervical dislocation and sperm obtained from the cauda epididymes. The sperm were mounted on slides, fixed and stained with 1% Eosin Y. The slides were air dried and coded for subsequent microscopic 600 sperm were assessed for morphological abnormalities of the sperm-head according to criteria of Wyrobek and Bruce, (1978).

Doses used in this study correspond to the human therapeutic dose: the human therapeutic dose for sulphamethoxypyridazine is 15.4mg/kg and pyrimethamine is 0.77mg/kg. The highest daily dose utilised in this study corresponds to double (2x) the human therapeutic dose. The lowest dose corresponds to 0.25x. Other doses utilised are 0.5x 1.0x and 1.5 xHTD.

Statistical analysis

In the sperm head morphology test, differences between the control and experimental groups were analysed by means of the 't' test. The test was considered positive when the increase in abnormal heads was at least double the negative control level, with $P < 0.05$ as the criterion of significance. Furthermore, the test must have yielded statistically significant increases at a minimum of 2 consecutive dose levels, and show evidence of a dose-related increase in abnormalities.

Results

An increase in the percentage of sperm shape abnormalities was observed with metakelfin treated U.I. Vet. F1 mice after 5 weeks exposure. (Table 1). Of the 5 doses used only the 7.7mg 0.38mg/kg (0.5xHTD) gave a statistically significant increment. The negative control mice showed 2.53% abnormality. Furthermore, the induction of morphological aberrations in sperm heads by the different concentrations of metakelfin, after 5 weeks exposure, was not strictly dose-dependent. Figure 3 shows the dose-response relationship for the abnormal sperm in the mice.

The positive control gave a statistically significant elevation of abnormal sperm heads in the mice.

TABLE 1

Metakelfin (sulphamethoxypyridazine: pyrimethamine) effect on the sperm-head morphology in U.I. Vet F1 mice.

Daily dose (mg/kg)	Number of animals	Sperm with abnormal head shapes (5 weeks)
0	5	2.53
3.85: 0.19	3	2.83
7.7 : 0.38	5	7.87*
15.4: 0.77	5	3.50*
23.1: 1.15	5	4.90
30.8: 1.34	5	5.53

* significantly higher (P 0.05) than control, t test.

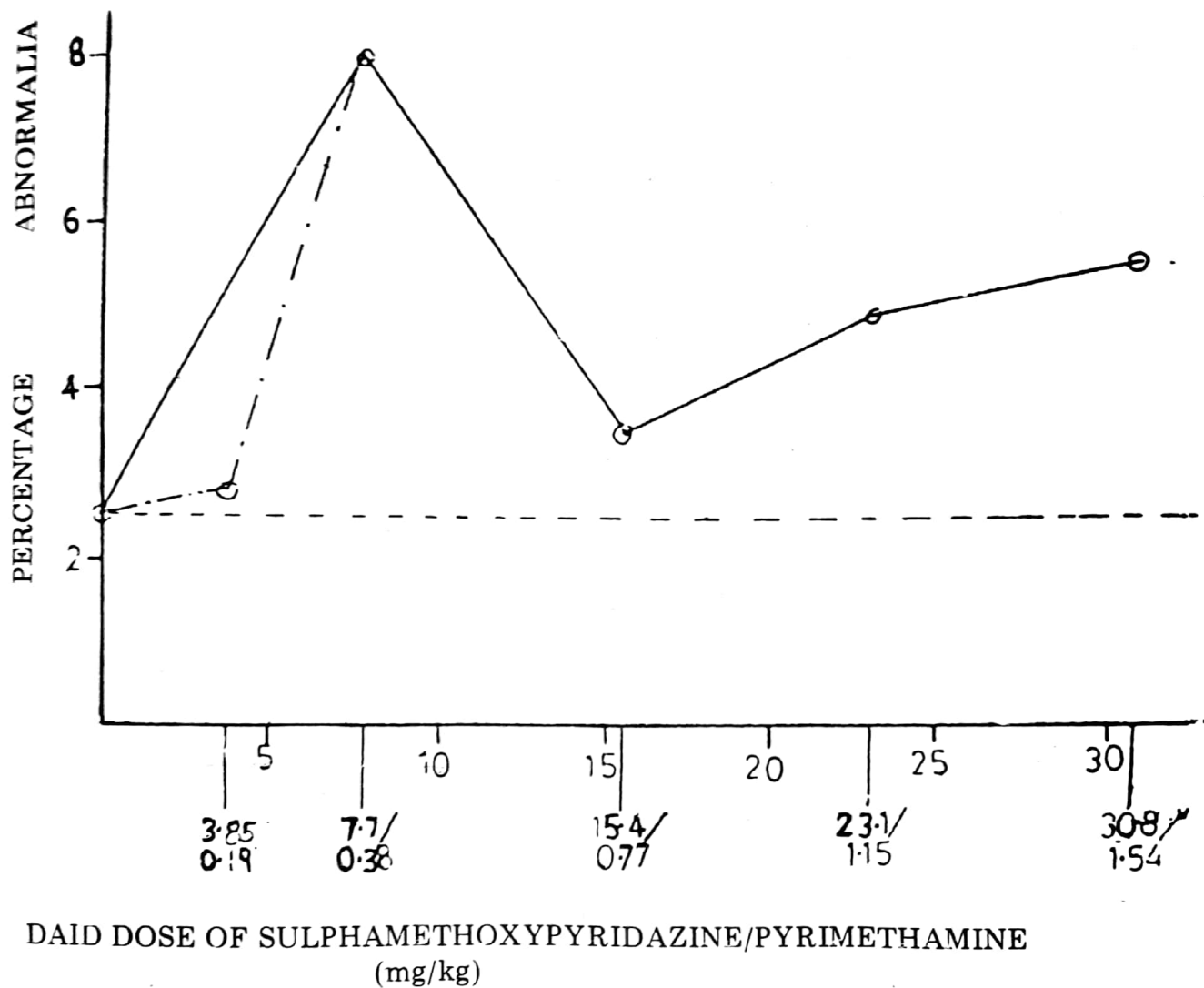


Fig. 3: Dose-response relationship for Sulphamethoxypyridazine/pyrimethamine (metakelfin) - induced morphologically abnormal sperm in albino mice. Each point represents the average percentage of abnormal sperm for 5 mice. Only at the 7.7/0.38 mg/kg dose level (x.0.5HTD) was the response statistically greater than the control value at $P < 0.05$. Hori-

Discussion

Malaria continues to be the world's greatest killer disease. In the absence of a potent antimalarial vaccine, the only other effective means of prevention and control lies in chemotherapy with potent antimalarial agents. It is therefore an issue of interest and consequently desirable that these agents be safe to use due to the widespread exposure to the agents, moreso when majority of the people in the under-developed countries of the world with teeming populations, do not have access to adequate trained medical personnel and consequently resort to

self medication with complete ignorance of the correct prescriptions. One cannot, therefore, rule out the possibilities of over exposure due to the indiscriminate use and abuse of these antimalarial drugs, as the drugs are over-the-counter products in such countries. The situation may be further worsened by the recent reports of the development of drug-resistance by the parasites (Meek et al, (1986); Payne, (1987). There are no available reports on the mutagenicity of sulphamethoxypyridazine and pyrimethamine. In this study, using the sperm head abnormality test a negative result was obtained for the combination antimalarial, sulphamethoxypyridazine and pyrimethamine (metakelfin).

One plausible reason for the inability of metakelfin to have induced statistically significant increases in abnormal sperm in mice is that the testes of the treated animals probably did not accumulate enough of the drugs, so as to alter the differentiation of spermatozoa. There have been no reports of the drugs binding to DNA which can lead to the faulty differentiation of spermatozoa, in the testes (Bruce *et al*, 1974)

Since mutagens are known to induce abnormal sperm heads, results obtained from the sperm head morphology tests may be useful in hazard assessment of a test compound. However, interpretations of finding obtained with the sperm morphology test must be made with caution until all available test systems have confirmed the test compound to be non-mutagenic.

References

- Bruce, W.R.: Furrer, R; Wyrobek, A.J (1974)
Abnormalities in the shape of murine sperm after acute testicular X-irradiation. *Mutation Res.* 23, 381-386.
- Meek, S.R.; Doberstyn, E.B.; Gaunere, B.A.;
Thanapanich, C.; Nordlander, E.; and Phiphaisan, S. (1986). Treatment of falciparum malaria with quinine and tetracycline combined mefloquine/sulfadoxine/Pyrimethamine on the Thai-Puchean border. *Amer. J. Trop. Med. Hyg.* 35, 246 - 250
- Payne, D. (1987) spread of chloroquine - resistance in *Plasmodium falciparum*
Parasitol. Today. 3, 241 - 246.
- Pharmaceutical Codex (1979).
- Rollo, I.M. (1955) Mode of action of sulphonamides, Proguanil and Pyrimethamine on *Plasmodium gallinaeum*, *Brit. J. Pharmacol.* 10, 208-214.
- Wyrobek, A.J. and Bruce, W.R. (1975) chemical induction of sperm abnormalities in mice. *Proc. Natl. Acad. Sci U.S.A.* 72, 4425-4429.
- Wyrobek, A.J. and Bruce, W.R. (1978). The induction of sperm-shape abnormalities in mice and humans, In *Chemical mutagens: Principles and*

methods for their detection, Hollaender, A; and de Serres, F.J. eds. (Vol.5). Plenum Press, New York pp 255 - 285.

Wyrobek, A.J.; Gordon, L.A.; Burkhardt, J.G.; Francis, M.W.; Kapp, R.W. (Jr.); Letz, G.; Malling, H.G.; Topham, J.C.; Whorton, M.D. (1983). An evaluation of the mouse sperm morphology test and other sperm tests in non human mammals. A report of the U.S. Environmental Protection Agency Gene Tox Programme *Mutation Res.* 115, 1-72.