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Full Length Research Article

Effect of Halofantrine Administration on some liver and heart enzymes in Healthy Human Volunteers

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ABSTRACT

The effect of halofantrine on liver and the heart was evaluated in 15 healthy adult human volunteers (11 females and 4 males), after a single oral dose of halofantrine given to each volunteer. Blood samples were collected at predetermined time intervals and analysed for the various liver and heart enzyme activities {Aspartateaminotransferase (AST), Alanineaminotransferase (ALT), Alkalinephosphatase(ALP), Creatine phosphokinase (CK) and lactatedehydrogenase (LDH)}, before and after drug administration. Halofantrine administration caused a steady increase in the activities of all the enzymes with a peak at the 8th hour. These values were statistically significantly ($p < 0.05$) different from the base line (0 hr) especially at the 8th hour. The increase in enzyme activity corresponds to peak serum concentration of halofantrine as seen in earlier studies. It conclude that halofantine administration at therapeutic or higher doses could result in an increase in enzyme activities and since these vital organs, plays a significant role in halofantrine pharmacokinetics, may play a role in predicting halofantrine toxicity/and or effect. (Afr. J. Biomed. Res. 9: 31 – 35, 2006)

Keywords: Halofantrine, human, biochemistry, Hepato/Cardiotoxicity

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INTRODUCTION

The search for safe and effective anti malarial drug still remain a priority to health care givers in the sub-Saharan African, as malaria continues to threaten life through the bite of drug resistant parasite in the sub region (Fitch, 1983). More than two billion people (about 40% of the world population) in 100 countries are at risk of the disease (WHO, 1994). There are at least 300 million cases of acute malaria each year, most of whom are children under the age of 5 years. Other high risk groups include pregnant women, non-immune travellers/HIV patients, refugees, displaced persons, or labour forces entering into endemic areas (Phillips, 2001). Countries in tropical Africa bear the brunt of malaria, accounting for more than 90% of the 300 million cases of malaria. It is estimated to cost Africa more than 12 billion U.S dollars annually (Rathod *et al*; 1997)

Malaria can however be adequately and promptly treated through drug therapy (Peters, 1998, Olumese 1998) and over the years, various antimalarial drugs have been produced; The first widely used antimalarial drugs in the treatment of malaria is Quinine from Peruvian bark that were originally sensitive to *P. falciparum spp.* (Rathod *et al*; 1997). However strains resistant to this and other antimalarial drugs are now common (Warhurst, 2001, Salako *et al* 1987), hence the parasite is now able to multiply in the presence of the drugs (Chatterjee, 1976). The development of resistance to the various antimalarial drugs is causing a major draw back in the control of malaria (WHO, 1994).

Halofantrine, a phenanthrane-methanol, is highly effective drug for *P.falciparum* malaria (Ter Kule *et al* 1993) and remained effective in multi-drug resistant areas, has a simple dosage regimen and is relatively cheap, however consistent cardio toxicity reports has made its use on large scale difficult (Molta, 1995). moreover, most of the available options have their respective drawbacks, which is also a major considerable burden to the problem of drug resistance (WHO, 1994). Extensive work has been done to asses the cardiotoxicity effects of halofantrine both in healthy human volunteers and patients with malaria, (Sowounmi *et al* 1998, Margaretha B *et al*, Eric Monilun 1995), but one other area that needs evaluation is the hepatotoxicity of the drug, as this drug is metabolised in the liver (Robert *et al*; 1990) and therefore the liver carry a major burden of the effects of the drug. Enzymes have been used as effective tools in the diagnosis of various medical problems and they also serve as markers of hepatocellular damage (Afonja 1993 Ngana *et al* 1989, Briggs *et al* 1974). These enzymes are therefore likely to play a role in hepatic and cardiac toxicity. The study therefore evaluated, the role of cardiac and liver enzymes in the diagnosis of halofantrine toxicity, and also assessed

the changes of these enzymes before and after administration of halofantrine to apparently healthy human volunteers as well as compared these changes before and after the administration of halofantrine to the volunteers.

MATERIALS AND METHOD

Location of study

The study was carried out in the department of clinical pharmacology and therapeutic laboratory college of Medical Sciences University of Maiduguri. Ethical clearance was obtained from the joint ethical committee of the UMTH/College of Medical Science University of Maiduguri before conducting the study.

Chemical and Reagents

Methylated Sprite, (Radox laboratories Ltd U.K), Buffer/glucose (Radox laboratory Ltd U.K), NADH – kit (LD 526) (Radox Laboratories Ltd UK). Enzyme/Co enzyme/substrate – kit (Ck 110) (Radox Laboratories U.K), 2,4 – dinitrophenylhydroazine- (Radox laboratories U.K). Distilled water- (Ben Medical laboratory, Maiduguri). Chromogenic substrate- (Radox Laboratories Ltd U.K). Sodium Hydroxide – Avondate Laboratory (supplies) and services Ltd Banbury Oxon England.

Analytic equipment:

Semi Automated Chemistry Analyser-ERBA. CHEM PRO Transaia bio-medicals Ltd MUMBDI. Centrifuge Machine (Health University II England). Water bath (series 12790/38 Old England

Treatment of subjects:

A total of 15 healthy adult human volunteers (4 male and 11 females) after obtaining a written consent, were recruited into the study after medical history taking and clinical assessment. The subjects were between the ages of 18 and 25 years with an average body weight of 45kg. None of the volunteers had been on any medication in the previous two weeks preceding the study. All volunteers were students of the University of Maiduguri who reside in the hostels of the same institution. They all gave their written consent to participate in the study. Full Blood count, Packed Cell Volume, Liver function Test were all, done as part of clinical assessment, subjects were fed and kept in the laboratory for the first 12 hours of the study. Subsequently they were allowed to go to hostel which is about 500m from the study and reported everyday for assessment and sampling until the end of study. They were not allowed to engage in strenuous exercise or take other medication throughout the period of study. The analysis was carried out in Chemical pathology

department of the University of Maiduguri teaching hospital.

Drug administration

All subjects were administered a single dose of 500mg of halofantrine hydrochloride in the form of tablet (2 tablets of halfanR) Smithkline Beecham, Lagos, Nigeria) with 100ml of water.

Sample collection.

Blood (5ml) was collected at the enrolment from all subjects through venipuncture. The blood sample was taken for the determination of Full Blood Count and differentials, Packed Cell Volume and Liver Function Test. This served as a baseline data (i.e. 0 hour reading, control), before the administration of the drugs; it was also used for clinical assessment. The

Blood samples were taken at predetermined intervals of 1, 3, 6, 8, 12, 24, 48, 72, and 168hours after the administration of the drugs. These samples were placed in a centrifuge tubes and centrifuged at 500rpm for 15min in a centrifuge tubes to obtain serum, which was placed in a plain tubes well labelled bottles and were stored frozen at -20°C until analysis. Samples used for haematological analysis i.e. Packed Cell Volume, Full Blood Count were put into EDTA bottles. The serum collected was analysed for the following enzymes; ALP, ALT, AST, LDH and CK at Chemical pathology Dept. University of Maiduguri Teaching Hospital.

Data Analysis

Data from Case Report Form were entered into IBM compatible PC EPI –INFO software. Data analysis was performed using the EPI–INFO, and SPSS. Data were

compared using the student's t-test while means are expressed as mean standard deviation (\pm SD). P values of less than 0.05 were taken as statistically significant

RESULTS

Fifteen healthy adult volunteers were given a single dose (500mg) of halofantrine hydrochloride and their blood was taken at predetermined time intervals ranging from 0-168 hours. All the volunteers were monitored clinically through out the study. They all tolerated the study drug and there were no history of adverse drug reaction.

The data are presented on Table 2 as mean plus minus standard deviations (\pm SD). For the 0 hour parameters, the values are as follows AST 7.7 \pm 2.0 (range 7-13), ALT 6.1 \pm 1.9 (range 4.0 -12), ALP 35.3 \pm 7.0 (range 23 – 43), CK 44.0 \pm 21.6 (range 17 – 79), and LDH 152.0 \pm 37.6 (range 92 – 120) respectively. All these values falls within a normal laboratory reference range for adults.

Table 1

Anthropometric and Baseline parameters before administration of study drugs.

Parameter	Mean \pm SD	Range	
		Min	Max
Age (yrs)	22.20 \pm 2.04	18	25
Weight (g)	47.23 \pm 5.39	38.5	60
Pulse rate/min	72.73 \pm 11.07	52	91
Systolic B.P	109.23 \pm 13.00	90	130
Diastolic B.P	74.40 \pm 8.72	60	90
Body Temp (°C)	36.02 \pm 0.74	34.8	37.5
TWBC Count (10 ³ /mm ³)	6260 \pm 492.94	3600	10000
PCV (%)	45.73 \pm 5.48	37	52

Table 2:

Profile of the enzyme activity of the 15 volunteers before and after drug administration of 500 mg Halofantrine hydrochloride.

Hours/ Enzymes	0	1	3	6	8	12	24	48	72	168
AST	7.7 \pm 2.0 ^a	10. \pm 2.3 ^a	11.5 \pm 0.0- ^b	11.7 \pm 2.0 ^a	16.7 \pm 2.7 ^b	15.8 \pm 4.7 ^b	8.7 \pm 1.9 ^a	8.8 \pm 2.9 ^a	9.7 \pm 2.5 ^a	8.6 \pm 2.3 ^a
ALT	6.1 \pm 1.9 ^a	6.7 \pm 1.4 ^a	5.0 \pm 1.8 ^a	5.2 \pm 2.0 ^a	7.8 \pm 1.9 ^b	6.0 \pm 3.1 ^a	5.4 \pm 3.0 ^a	4.5 \pm 2.0 ^a	5.3 \pm 1.9 ^a	4.6 \pm 1.4 ^a
ALP	35.3 \pm 7.0 ^a	29.0 \pm 3.5 ^a	33.8 \pm 5.0 ^a	35.6 \pm 12.3 ^a	40.8 \pm 18.1 ^b	25.6 \pm 5.2 ^a	31.1 \pm 5.8 ^a	25.5 \pm 44 ^a	31.1 5.8 ^a	25.3 \pm 4.6 ^b
CK	44.0 \pm 21.6 ^a	41.3 \pm 21.0 ^a	40.2 \pm 16 ^a	51.3 \pm 15.0 ^a	63.8 \pm 23.7 ^b	61.8 \pm 23.7 ^b	58.3 \pm 23.4 ^a	54.2 \pm 23.2 ^a	47.2 \pm 22.7 ^a	36.4 \pm 16.2 ^a
LDH	151.0 \pm 37.6 ^a	140.0 \pm 39.4 ^a	146.6 \pm 4.1	184.7 \pm 53.0 ^a	198.7 \pm 70 ^b	145.7 \pm 39.7 ^a	115.3 \pm 17.9 ^b	139.2 \pm 44.0 ^a	155.5 \pm 42.1 ^a	154.4 \pm 28 ^a

N.B: The results are presented as means \pm standard error of mean. Values with different superscripts are significantly different while values with the same superscripts are not significantly different at P<0.05

The other baseline values measured e.g. PCV and WBC as well as clinical assessment was all normal before drug administrations. (see Table 1)

The highest enzyme activity was obtained at the 8th hour for both the enzymes with mean values of 16.7 ± 2.0 (10 – 23) and 7.8 ± 1.9 (4 – 12) IU/L respectively.

The enzyme activities for both groups are statistically significant ($p < 0.05$) at the 8th hour. AST also showed significantly ($p < 0.05$) different values at 3, 6 and 12 hour compared to the 0 hour (baseline values).

The Tables 1 and 2 also show the mean enzyme activities for ALP, LDH and CK before and after administration of the drugs. All the enzymes have their peak concentration at the 8th hour (40.8 ± 18 (23 =70), 63.8 ± 19.1 (44 – 92) and 198.7 ± 70 (123 – 371) IU/L respectively) with slight variations between 8 to 12 hours from the base lines. In the case of LDH there were significant ($P < 0.05$) changes at 6, 8, and 12 hours (184.7 ± 53.2 , 198.7 ± 70.7 and 155.5 ± 42.1 respectively). These hours' correlates with increase and decrease of plasma concentration of halofantrine, however the rise did not correspond to the normally expected rise of LDH as seen in myocardial damage, which usually (peak 24-48 hours). Though these early raise in LDH in these study, however, remains stable until the end of the study (168 hours).

DISCUSSION

The activities of some hepatic and cardiac enzymes in this study, after a single fed of healthy human volunteers were assessed. The results of this study shows that the enzyme activity decreases as the effect of the drug decreases and the peak enzyme activities correlate well the t_{max} of halofantrine as seen in earlier pharmacokinetic studies (Onyeji et al 1998, Bassi PU et al 2004).

The activities of liver transaminases (ALT & AST), the alkaline phosphatase and cardiac enzymes CK & LDH were significantly higher compared to baseline values. These values were also high between the 8-12 hours, which corresponds with the usual peak plasma concentration of halofantrine as seen in literature (Kwabang et al 1984, Milton et al 1985, Watkins et al 1995, Touze JE et al 1996 and Bassi et al 2004).

Halofantrine has a wide inter-individual variability, thus the fluctuation of the enzymes activities as seen in this subjects may be related to the inter individual variations of the oral bioavailability of halofantrine (Bassi et al, 2002 Milton et al; 1989) Sowunmi et al 1998, Eric Monlun 1995, Margretha et al 2004 all has demonstrated significant QTC prolongation in subjects after halofantrine administration. The QTC prolongation is also dose

dependent Milton et al 1989. This implies therefore that there may be a higher activity when full dose of the drug is administered.

CONCLUSION:

In conclusion, this study demonstrate from the results presented that there was peak enzyme level of the various enzymes as the drug reached its peak serum concentration i.e. 6-12 hours and the enzyme activity declined as the effect of the drug weaned off the system. This indicates that changes in the enzyme activities could occur at higher concentration of the drug or after a repeated dosing (it is concentration dependant) as the ECG changes in other studies. Thus it implies that halofantrine is not only cardiotoxic, it is also hepatotoxic, more so assessment of serum levels of this enzymes may have a place in the diagnosis and monitoring of patients with halofantrine toxicity.

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