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RESEARCH PAPER

LIVER WEIGHT CHANGES IN WISTAR RATS TREATED WITH CRUDE AQUEOUS EXTRACTS OF *MANGIFERA INDICA* STEM BARK.

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ABSTRACT

This study was designed to determine the weight changes in the liver of wistar rats treated with crude aqueous extracts of *Mangifera indica* stem bark. Twenty Wistar rats (170-185g) were used for this study. They were sub-divided into four groups: A, B, C & D (n=5 each). Group A served as control, while B, C, and D served as tests. For 14 days, group A received normal feed mash and water only, while groups B, C and D received oral doses of 0.25ml (25mg), 0.5ml (50mg) and 1ml (100mg) of aqueous extract of *Mangifera indica* stem bark respectively. At the end of the experiment, the animals were weighed and sacrificed to harvest the liver for organ weight determination. Results showed no significant increase ($p>0.05$) in the body weight of the control rats compared to test group; but a significant decrease ($p<0.05$) in the weight of the liver in group D ($p< 0.05$) when compared with the control. It suggests that 100mg of *Mangifera indica* is above the safe dose level for rats and that crude AE of M. indica has no significant effect on somatic growth but caused a significant decrease in liver weight at higher doses.

Key Words: Aqueous extract, Liver, *Mangifera indica*, Weight changes,

INTRODUCTION

For several decades, the comparison of organ weights between treated and untreated groups of animals have conventionally been used to evaluate the toxic effect of the test article (Peters and Boyd, 1966; Pfeiffer, 1968). In recent times, organ weight changes have indeed become a major index in toxicological studies and there are recommendations that the approach to the evaluation and interpretation, must be done with appropriate scientific rigor and, to the extent possible, consistent with the regulatory guidance (Bindhu *et al*., 2007). Although, the approach in terms of methods of organ weighing and interpretation vary, organ weight changes remain an acceptable sensitive indicator for chemically induced changes to organs (Bindhu *et al*., 2007) and in that regard, potential changes in the weight of several organs have been recommended for toxicity studies (Roy and Andrews, 2004; Haley *et al*., 2005).

Specifically, liver weight changes have been considered useful by 81% of the pharmaceutical industry respondents and 100% of the respondents in other industries involved in a survey by the Society of Toxicologic Pathology (Bindhu *et al*., 2007). The factors cited for its usefulness included: its sensitivity to predict toxicity in toxicity studies; its usefulness to evaluate/support diagnosis of hepatocellular hypertrophy from hepatic enzyme induction, peroxisome proliferation or lipidosis; its status as a reflective medium for physiologic perturbations and metabolism; its correlative suitability for histopathological changes; its little animal-to-animal variability; the availability of historical control range data; and its importance as the primary detoxification organ (Bindhu *et al*., 2007).

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Of interest in this study, is the toxicity potentials of *Mangifera indica* (Mango) -a plant grown widely in different parts of Africa, especially in the southern part of Nigeria, where it is valued for its edible fruit (Nwinuka, *et al*., 2008). It is one of the several plants with potent therapeutic active ingredients as available literature indicates that *Mangifera indica* is used medicinally to treat ailments such as asthma, cough, diarrhea, dysentery, leucorrhoea, jaundice, pains, malaria (Madunagu *et al*., 1990; Gilles, 1992) and diabetes (Ojewole *et al*., 2005; Muruganandan *et al*., 2005; Perpetuo *et al*., 2003; Mahabir and Gulliford, 1997). Other therapeutic properties include analgesic, anti-inflammatory (Garrido *et al*, 2001), immune-stimulant (Makare, 2001; Garcia *et al*., 2002; Garcia *et al*., 2003a), antioxidant (Martinez *et al*., 2000; Sanchez et al., 2000; Sanchez et al., 2003), spasmolytic, antidiarrhea (Sairam *et al*., 2003), dyslipidemic (Anila and Vijayalakshmi, 2002), antidiabetic (Aderibigbe et al., 1999; Aderibigbe *et al*.,2001), antiamebic (Tona *et al*., 2000), antihelminthic, antiallergic (Garcia *et al*., 2003b) and antibacterial (Bairy *et al*., 2002).

However, beyond herbal drug-efficacy verification, there is a growing need to adequately subject herbal preparations to sound toxicity studies in order to understand the collateral systemic consequences of herbal preparations. Therefore, as organ weight evaluation has become an essential part of toxicologic and risk assessment of drugs, chemicals, biologics, food additives, and medical devices, this study examines the comparative liver weight changes in Wistar rats treated with crude aqueous extracts of *Mangifera indica* stem bark; judging by the well known strategic role of the liver in drug metabolism and its vulnerability to drug-pressure insults; with attendant consequences.

MATERIALS AND METHOD

Location and Duration of Study: This study was conducted at the histology laboratory of the Faculty of Basic Medical Sciences, Delta State University, Abraka. Preliminary studies, animal acclimatization, drug procurement, actual animal experiment and evaluation of results, lasted for a period of one month, while the actual administration of the drug to the test animals lasted for two weeks.

Animal Groupings: Twenty Wistar rats weighing between 170-185gm were procured and maintained in the Animal house of the Faculty of Basic Medical Sciences, Delta State University, Abraka. The experimental animals were divided into four groups A, B, C & D, with five rats each, and were housed in standard plastic cages. Group A serves as the control, while groups B, C, & D were used as the experimental animals. The rats were fed with grower's mash produced by Bendel Feeds and Flour Mills Limited, Ewu, (standard diet), and water was given ad libtium. They were allowed to acclimatize for one week before commencement of the study. Ethical approval was sought and received from the Department of Anatomy, Faculty of Basic Medical Sciences, DELSU, on the need to observe completely the rules guiding the employment of rats for scientific studies.

Preparation Of Aqueous Extract: *Mangifera indica* stem bark was obtained freshly from a farm in Ekpoma in Esan West L.G.A, Edo state of Nigeria. The plant was identified and authenticated at the Botany department of Ambrose Alli University, Ekpoma. The stem bark of Mangifera indica was cut into smaller pieces and sun-dried for two weeks. The dried sample was pulverized using mortar and pestle. The resulting powder material was used in the extraction process. Extraction was carried out using the method described by Harboone (1972) and Ekpe *et al*. (1990); Uhegbu *et al*. (2005); Nwinuka *et al*. (2008). Using distilled water as the solvent, 20 g of powdered sample of the herb was extracted by soaking in 200 ml of distilled water in a beaker, stirred for about 6 minutes and left overnight. Thereafter, the solution was filtered using filter paper (Whatman No. A-1) to remove cellulose fibers and extract stored in a refrigerator at 4°C.

Experimental Design: After acclimatization period, the rats were weighed and divided into groups A, B, C and D (n=5 each). Group A rats served as control while B, C and D served as the test groups that were treated with daily oral doses of 0.25ml (25mg), 0.5ml (50mg) and 1ml (100mg) daily of the aqueous extract for a period of 14 days. The 1ml of crude aqueous extract used in this study was based on the previous work done with this plant by Nwinuka *et al.* (2008). At the end of experimental period, the rats were weighed and anaesthetized with chloroform in order to dissect them and harvest the liver. The weight of the fresh liver was then taken and recorded accordingly.

Statistical Analysis: The data collected were analyzed using the Statistical Package for Social Sciences (SPSS for windows, version 12.0). Comparison were made between the control and experimental groups using student's T-test, with values of less than 0.05 ($P< 0.05$) were regarded as statistically significant.

RESULT

The mean weight and standard deviation of the liver in groups A, B, C and D are shown in table 1. Although there was no significant difference in the weight and gross anatomical features (*size, colour, and consistency*) of the liver in the control and test group B and C, group D however, showed marked decrease in the size of the liver with a significant reduction in organ weight ($p< 0.05$) when compared with the control.

Table 1: Effect of crude AE of *Mangifera indica* on body and liver weights in control and treated rat

Groups	A	B	C	D
Liver Weight (g)	7.92±0.77	7.44±0.54	7.39±0.67	*5.52±0.21

Values are expressed as mean ± SD; *significantly different from the control at $p<0.05$.

DISCUSSION

The observed decrease in the weight of the liver in group D, is an indication that crude AE of M. indica might have toxic effect on the liver at 1ml daily dose; contradicting the cytoprotective potentials (Sugikara *et al*., 1999; Lima *et al*., 2006; Yao *et al*., 2007; Zhao and Zhang, 2009) of the phenolic constituents, triterpenes, flavonoids, phytosterols, and polyphenols in *M. indica* (Saleh and El-Ansari, 1975; Anjaneyulu *et al*., 1994; Kharn *et al*., 1994; Selles and Castro, 2002; Singh *et al*., 2004), due to their antioxidant properties (Martinez et al., 2000; Sanchez *et al*., 2003) reported in several studies on *M. Indica*. Our observation however, is in line with the assertion by Simons *et al*. (1995) that an increase or decrease in either absolute or relative weight of an organ following the administering of a chemical or drugs, is an indication of the toxicity status of that chemical.

Obviously, the contention remains the high dose ingestion in group D, which incidentally, has become a characteristic feature of herbal medicine ingestion in populations and a source of public health concern. In fact, most of the readily ingested herbal therapies are yet to be subjected to sound toxicity studies to determine their effective (ED50) and lethal doses (LD50). Unfortunately, emphasis has been on efficacy and not on therapeutic safety. It is on the basis of this that Nwaopara (2013) opined that the advancement of herbal medicine is inextricably tied to the conscientious effort to constantly evaluate the therapeutic potentials of the abundant medicinal plants around us; and as such, cannot fathom why trained researchers should abandon herbal medicine in the hands of diviners and herbalist.

Overall, our findings suggest that at higher doses, *M. indica* is hepatotoxic and this is supported by the fact that polyphenols, at certain concentrations, have the capacity to cause oxidative stress and liver toxicity in vivo (Lambert et al., 2007). It suggests also that the 1ml dose administered to group D is apparently above the safe-dose level for rats, while 0.25ml and 0.5ml administered to groups B and C, apparently falls within the safety margin. It is our recommendation therefore, that further studies be conducted to determine the safe dose of *M. indica*, while indiscriminate consumption should be avoided.

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AUTHOR'S CONTRIBUTIONS

Oaikhena, G.A. and Izunya, A.M., conceptualized the research. Data analysis was done by Oaikhena, G.A, while Oaikhena, Izunya, Olugbenga and Ujaddughe, joined in writing and reviewing this paper. All authors funded it. No conflict of interest declared.