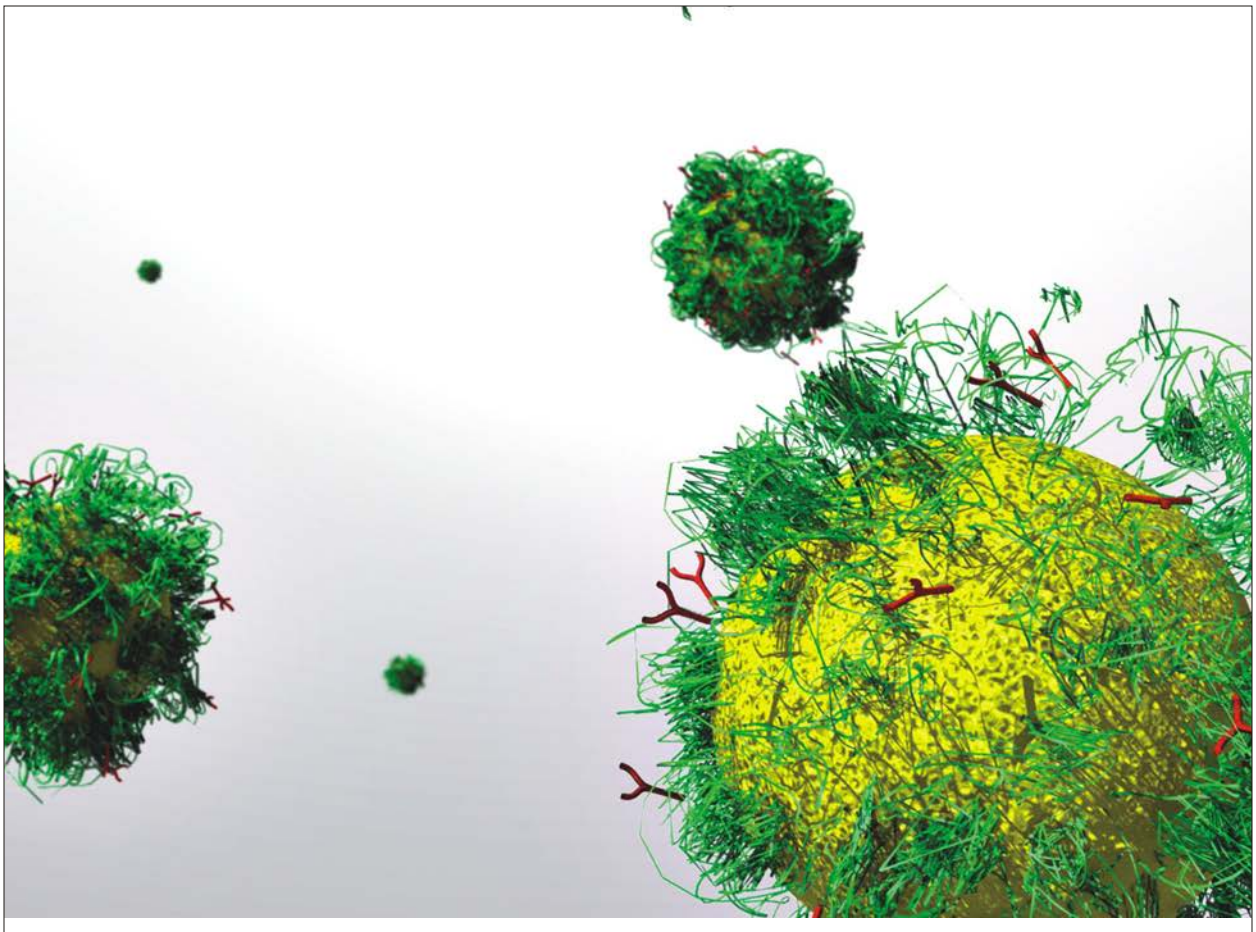

American Journal of Drug Discovery and Development

ISSN 2150-427X

Academic Journals Inc.

www.academicjournals.com

American Journal of Drug Discovery and Development 3 (3): 140-148, 2013
ISSN 2150-427x / DOI: 10.3923/ajdd.2013.140.148

Comparative Antimalarial and Safety Attributes of Methanolic Extract of Leaves of *Tithonia diversifolia* and *Morinda lucida* in Animal Models

[1]R.O. Arise, [1]S.O. Malomo and [2]O.A. Bolarinwa
[1]Department of Biochemistry,
[2]Department of Epidemiology and Community Health, College of Health Sciences, University of Ilorin, Kwara State, Nigeria

Corresponding Author: R.O. Arise, Department of Biochemistry, Faculty of Science, University of Ilorin, Kwara State, Nigeria

ABSTRACT

Anti-malarial property and toxicity of methanolic extract of leaves of *Tithonia diversifolia* and *Morinda lucida* were investigated in mice and rats, respectively. Percentage parasitemia and chemosuppresive effect of the extract on *Plasmodium berghei* infected mice were used to assess anti-malarial property while toxicity was evaluated by measuring the activities of alkaline phosphatase (ALP), alanine aminotransferase (ALT) and aspartate aminotransferase (AST) in serum and some selected tissues as well as evaluation of some hematological parameters, serum electrolytes, urea and creatinine in rats. The extract at 200 mg kg^{-1} b.wt. showed significant ($p<0.05$) chemo suppressive effect following five days administration in *P. berghei* established infection. There was a significant decrease ($p<0.05$) in the liver, kidney and serum activities of ALT following 7 and 14 days administration of the extract. Similar trends were observed in liver and serum ALP and liver AST. However, there was a significant increase ($p<0.05$) in kidney and serum AST activity for both periods of administration except for the significant decrease ($p<0.05$) in the kidney following the 14 days administration. The white blood counts showed a significant decrease ($p<0.05$) following administration of *T. diversifolia* for 7 and 14 days and *M. lucida* for 14 days while administration of both extract for 14 days revealed a significant increase ($p<0.05$) in platelet counts. Serum bicarbonate, chloride, sodium, potassium and urea decreased significantly ($p<0.05$) following administration of the extract for 7 and 14 days. This result therefore suggests that, methanolic extract of *T. diversifolia* and *M. lucida* possess considerable anti-malarial property but may potentiate some level of toxicity especially if administration is prolonged.

Key words: *Tithonia diversifolia*, *Morinda lucida*, *Plasmodium berghei*, anti-malarial

INTRODUCTION

Malaria is the world's most common tropical disease (Trampuz *et al*., 2003). At least two third of the world population live in malaria endemic areas and not less than 200 million people are infected each year of which estimated one to two million people succumb to the disease annually (Kochar *et al*., 2003) with about 40% of the world's population at risk (Palaniswamy *et al*., 2010). Around 800,000 children under the age of five die from malaria each year, making this disease one of the major causes of infant and juvenile mortality (WHO, 2005). Four species of plasmodium, namely *P. malariae*, *P. ovale*, *P. vivax* and *P. falciparum* may infest man of which *P. falciparum*

is responsible for the often fatal cerebral malaria (Phillipson and Wright, 1991). However, *P. berghei* have been used in many research institutes for studies aiming at the development of new drugs or vaccine against malaria. Like all malarial parasites of mammals, including the four human malaria parasites, *P. berghei* is transmitted by Anopheles mosquitoes (Phillipson and Wright, 1991).

In sub-saharan Africa, more than 80% of the population relies on traditional medicines and healers as the primary source of health care (WHO, 2005). This is mainly due to accessibility, affordability and cultural sensitivity of traditional medicines. Since time immemorial, plants have served as source of health management and prevention remedies as well as for the treatment of diseases. Medicinal plants have occupied an irreplaceable position in the discovery of pharmacologically active compounds including quinolines, endoperoxides, quinines which have been implicated in malaria treatment. Although there is a broad therapeutic arsenal against malaria, unfortunately, these arsenals present numerous draw back including several side effects and problems of drug resistance and cross resistance (Coker *et al*., 2001). Therefore, the need for continuous research and discovery of new, safe, effective and accessible malarial remedies cannot be overemphasized.

Morinda lucida (ML), a Rubiaceae, is a medium sized tree up to 15 m high with a characteristic yellow wood and a broad, elliptical leaves while *T. diversifolia* (TD) (marigold tree), a composite, is a shrub of about 2-3 m tall with a subvate, serrate and mostly lobed leaves. The leaves of *M. lucida* have been used in folkloric practice for making 'fever' tea used not only for the treatment of malaria but as a general febrifuge and analgesic (West and Sabin, 2012). *T. diversifolia* leaves have been implicated in the treatment of malaria and stomach ailments (West and Sabin, 2012). However, there is paucity of information regarding the anti-malarial properties and comparative efficacies of these plants. Thus, the objective of this study is to ascertain the folkloric practice of the use of these plants as anti-malarial and to compare their efficacy as well as ascertaining their safety.

MATERIALS AND METHODS

Chemicals and assay kits: AMP-buffered sodium thymolphthalein monophosphate substrate used for ALP was a product of Teco Diagnostics, Lakeview Avenue, Anaheim, Canada. Artemisinin was a product of Mekophar Chemical Pharmaceutical Joint-Stock Company Chi-Minh City, Vietnam. 2, 4-dinitrophenylhydrazin substrate and buffer were from Randox Laboratories Limited, UK. All other chemical used were of analytical grade obtained from BDH Chemical Limited, London.

Animals: Twenty five adult Swiss albino mice with an average weight of 20 g were obtained from the animal breeding units of the Department of Biochemistry, University of Ibadan, Oyo State, Nigeria. Thirty albino rats (*Rattus novergicus*) with an average weight of 148.35 g used for this study were obtained from the small Animal Holding Unit of the Department of Biochemistry, University of Ilorin, Ilorin, Nigeria. The animals were housed in plastic cages and maintained under standard laboratory conditions with free access to rat pellets and tap water *ad-libitum*. The studies adhere to the Principles of Laboratory Animal Care (NIH, publication number 85-23, revised in 1985).

Parasites: A dihydroartemisinin-sensitive strain of *Plasmodium berghei* was obtained from the Institute for Advanced Medical Research and Training (IAMRAT), College of Medicine, University of Ibadan, Oyo State, Nigeria.

Preparation of plants extract: Leaves of *M. lucida* and *T. diversifolia* were harvested within Ibadan City, Oyo State, Nigeria, in June, 2005. The plants were authenticated at the Department of Plant Biology, University of Ilorin, Ilorin, Nigeria. The leaves of the plants were sun-dried for 10 days. They were separately milled to powdered forms using a milling machine. Two hundrad gram of each of the crude powder were extracted after being soaked for 72 h in 70% methanol and thereafter filtered using Whatman filter paper. Each of the filtrate was then evaporated to dryness by heating on a water bath at 65°C.

Experimental design: Anti-malarial study was investigated in mice. The mice were grouped into five, each group consisting of five rats each. Group A consisted of uninfected mice and received 0.2 mL distilled water. The mice in group B consisted of infected mice and also received 0.2 mL distilled water. Group C, D and E were infected with *P. berghie* and treated with 200 mg kg^{-1} b.wt. of *T. diversifolia*, 200 mg kg^{-1} b.wt. *M. lucida* and 0.1 mg kg^{-1} b.wt. artesunate, respectively. Treatment was commenced 72 h after inoculation with the parasite and establishment of infection. The treatment lasted for 5 days.

Toxicological evaluation of the extract was carried out using thirty (30) rats (*Rattus norvegicus*). The rats were grouped into two major groups A and B and received treatment for 7 and 14 days respectively. Each group consists of three equal sub-groups of five rats each. Controls A and B served as controls for 7 and 14 days administration respectively and received 1 mL of distilled water daily while the remaining two groups (TD and ML) were administered 200 mg kg^{-1} b.wt. methanolic extract of *T. diversifolia* and *M. lucida* respectively.

Inoculation and confirmation of malaria parasite: The mice were screened for malarial parasites in their blood and only those that were free from malaria parasites were selected for this study. The selected mice were acclimatized for two weeks in the laboratory after which each mouse was induced with standard inoculums of 1 x 10^7 *Plasmodium berghei* via intra-peritoneal route. The parasite was then allowed to incubate in the mice for 72 h after which thin blood film was prepared from tail snip and stained using Giemsa stain and the parasitaemia confirmed by viewing using x100 objective of the light microscope. This method was also used for parasitaemia count throughout the period of study and percentage parasitaemia determined.

Biochemical assays: The procedure described by Roy (1970) was used to assay alkaline phosphatase (ALP) activity. Aspartate aminotranferase (AST) and alanine aminotranferase (ALT) activity was assayed using the method of Reitman and Frankel (1957). Serum urea and creatinine level was determined using the method described by Salisu (2004) while serum potassium, sodium and chloride level was determined by the method of Tietz *et al.* (1996). The method of Roth and Chan (2001) was used to estimate the level of bicarbonate in the serum.

Hematological parameters: Packed Cell Volume (PCV), Haemoglobin level (Hb), White Blood Cells count (WBC), Mean Corpuscular Hemoglobin Concentration (MCHC) and platelets count were determined using the methods of Adewuyi and Olatunji (1995).

Statistical analysis: Comparisons were made using Duncan's multiple range test (Duncan, 1955) and values were considered significant at $p<0.05$.

RESULTS

Figure 1 shows the activity of the extract and artemisinin on the parasite density following five days treatment of established *P. berghei* infected mice. Artemisinin reduced the level of parasite than either of the two extract. However, *M. lucida* reduced the level of parasitemia than did *T. diversifolia*.

The percentage chemosuppression of parasitaemia by the extract and artemisinin is represented in Table 1. The chemosuppressive properties is in the order of artemisinin>*M. lucida*>*T. diversifolia* with artemisinin having percentage chemosuppression of 96.60%, *M. lucida* with 81.18% and *T. diversifolia* being the least with percentage chemosuppression of 76.08%.

The effects of the extract on alkaline phosphatase, aspartate aminotransferase and alanine aminotransferase activities of liver, kidney and serum are presented in Table 2, 3 and 4, respectively. There was a significant reduction ($p<0.05$) in the liver ALP activity following 7 and 14 days administration of the extract while a significant decrease ($p<0.05$) was only observed in serum ALP activities following prolonged administration of the two extract. However, there was no significant difference ($p>0.05$) in the kidney activity of the enzyme following both 7 and 14 days administration. The liver activity of AST was significantly reduced ($p<0.05$) in a time-dependent manner for *T. diversifolia* while significant reduction ($p<0.05$) was observed following 14 days administration of *M. lucida*. However, kidney AST activity increased significantly ($p<0.05$) following 7 days administration of the extract but decreased significantly ($p<0.05$) afterwards. Similar pattern of alterations of AST activity was observed in *T. diversifolia* administered rats. Administration of the extracts also caused significant decrease

Table 1: Percentage chemo suppression of the extracts in *P. berghei* infected mice

	Chemosupression (%)				
Groups	Day 4	Day 5	Day 6	Day 7	Day 8
Artesunate	3.88+0.10[a]	52.55+0..67[a]	71.65+1.21[a]	83.67+0.23[a]	96.60+1.18[a]
M. lucida	1.55+0.12[b]	42.07+0.12[b]	58.71+0.04[b]	72.04+0.33[b]	81.18+0.24[b]
T. diversifolia	2.02+0.01[c]	37.12+0.02[b]	57.12+0.72[b]	69.96+0.04[b]	76.08+1.25[b]

Values are mean (n = 5)±SD, Values with different superscripts a, b, c in each column are significantly different

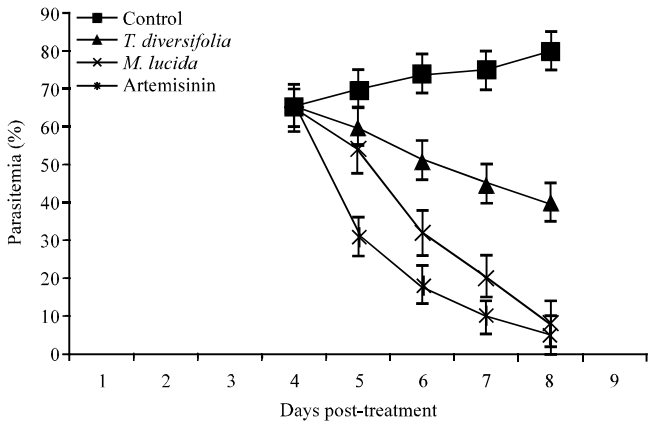

Fig. 1: Effect of artemisinin and methanolic extract of *Tithonia diversifolia* and *Morinda lucida* leaves on percentage parasitemia in *Plasmodium berghei* infected mice. (Each value is a mean of 5 determinations±SD)

Table 2: Effects of methanolic extract of *T. diversifolia* and *M. lucida* leaves on specific activity of alkaline phosphatase in the liver, kidney and serum of rat

	Enzyme activity (IU L^{-1})		
Groups	Liver	Kidney	Serum
Control A	171.59±1.51[a]	46.08±2.03[a]	7.56±0.37[a]
TDA	76.49±1.58[b]	42.50±1.12[a]	7.48±0.55[a]
MLA	97.79±1.36[c]	47.90±0.42[a]	7.63±0.03[a]
Control B	170.50±1.89[a]	46.12±1.46[a]	7.56±0.21[a]
TDB	61.72±1.07*[d]	47.00±2.15[a]	4.38±0.74[b]
MLB	66.76±0.93*[d]	47.25±0.61[a]	4.69±0.37[b]

Values are mean (n = 5)±SD. Values with different superscripts a, b, c in each column are significantly different, TDA: *T. diversifolia* administration for 7 days; TDB: *T. diversifolia* administration for 14 days. MLA: *M. lucida* administraton for 7 days; MLB: *M. lucida* administration for 14 days

Table 3: Effects of methanolic extract of *T. lucida* and *M. lucida* leaves on the specific activity of aspartate aminotransferase in the liver, kidney and serum of rat

	Specific enzyme activity (IU L^{-1})		
Groups	Liver	Kidney	Serum
Control A	39.54±0.50[a]	2.28±0.10[a]	0.14±0.08[a]
TDA	19.83±1.84[b]	4.24±0.01[b]	0.31±0.02[b]
MLA	41.32±0.35[a]	3.01±0.03[c]	0.15±0.08[a]
Control B	39.52±0.86[a]	2.27±0.20[a]	0.14±0.08[a]
TDB	17.70±0.57[b]	3.13±0.03[c]	0.18±0.01[a]
MLB	17.35±0.20[b]	1.99±0.09*[d]	0.15±0.01[a]

Values are mean (n = 5)±SD. Values with different superscripts a, b, c in each column are significantly different, TDA: *T. diversifolia* administration for 7 days; TDB: *T. diversifolia* administration for 14 days. MLA: *M. lucida* administraton for 7 days; MLB: *M. lucida* administration for 14 days

Table 4: Effects of administration of methanolic extract of *T. diversifolia* and *M. lucida* leaf on specific activity of alanine aminotransferase in liver, kidney and serum

	Specific enzyme activity (IU L^{-1})		
Groups	Liver	Kidney	Serum
Control A	297.20±1.30[a]	2.25±0.17[a]	0.12±0.02[a]
TDA	128.80±1.30[b]	0.60±0.02[b]	0.10±0.02[a]
MLA	164.00±4.06[c]	1.76±0.01*[c]	0.07±0.03[b]
Control B	296.42±1.42[a]	2.26±0.03[a]	0.11±0.01[a]
TDB	88.80±2.28*[d]	1.18±0.01[d]	0.05±0.02[b]
MLB	100.40±1.52*[e]	2.05±0.04[e]	0.05±0.02[b]

Values are mean (n = 5)±SD. Values with different superscripts a, b, c in each column are significantly different, TDA: *T. diversifolia* administration for 7 days; TDB: *T. diversifolia* administration for 14 days. MLA: *M. lucida* administraton for 7 days; MLB: *M. lucida* administration for 14 days

($p<0.05$) in the liver, kidney and serum ALT activity for both period of administration except for *T. diversifolia* which did not cause any significant alterations ($p>0.05$) when administered for 7 days.

The effect of the extracts on serum electrolytes, urea and creatinine level is presented in Table 5. Administration of the extract of *M. lucida* for 7 and 14 days caused a significant reduction

Table 5: Effects of administration of methanolic extract of *T. diversifolia* and *M. lucida* leaves on selected electrolytes, creatinine and urea in rat serum

Group	Bicarbonate (mmole L^{-1})	Chloride (mmole L^{-1})	Sodium (mmole L^{-1})	Potassium (mmole L^{-1})	Urea (mmole L^{-1})	Creatinine (mg dL^{-1})
Control A	3.40±0.55^{a}	20.20±1.92^{a}	28.00±0.70^{a}	1.90±0.16^{a}	2.30±0.83^{a}	14.62±3.40^{a}
TDA	3.60±0.54^{a}	10.60±3.84^{b}	23.40±3.59^{b}	1.42±0.15^{b}	1.18±0.09^{b}	14.64±0.11^{a}
MLA	1.80±0.83^{b}	14.00±1.00^{c}	18.00±1.85^{c}	1.38±0.08^{b}	1.12±0.08^{c}	16.14±0.40^{a}
Control B	3.40±0.45^{a}	20.22±1.79^{a}	28.10±0.42^{a}	1.90±0.34^{a}	2.30±0.50^{a}	14.62±3.70^{a}
TDB	3.60±1.14^{a}	15.80±1.79^{c}	17.40±3.54^{*c}	1.42±0.08^{b}	2.08±0.89^{d}	20.32±0.69^{b}
MLB	2.00±0.70^{b}	14.80±4.65^{c}	22.80±0.84^{b}	1.40±0.16^{b}	2.12±0.36^{d}	11.86±0.51^{a}

Values are mean (n = 5)±SD. Values with different superscripts a, b, c in each column are significantly different. TDA: *T. diversifolia* administration for 7 days; TDB: *T. diversifolia* administration for 14 days. MLA: *M. lucida* administraton for 7 days; MLB: *M. lucida* administration for 14 days

Table 6: Effects of administration of methanolic extract of *T. diversifolia* and *M. lucida* leaves on selected haematological parameters of rats

Group	Hb (g dL^{-1})	PCV (%)	WBC	MCHC (g dL^{-1})	Platelets (x10^{6} mL^{-1})
Control A	13.32±0.72^{a}	43.40±5.94^{a}	21.12±3.78^{a}	30.20±0.84^{a}	0.74±0.08^{a}
TDA	13.02±0.61^{a}	43.80±0.84^{a}	15.88±2.62^{b}	30.40±1.14^{a}	0.75±0.05^{a}
MLA	14.10±0.37^{a}	46.40±2.30^{a}	19.80±3.09^{a}	30.40±1.34^{a}	1.16±0.20^{b}
Control B	13.30±1.05^{a}	43.60±5.80^{a}	21.09±3.30^{a}	30.20±0.70^{a}	0.75±0.08^{a}
TDB	14.02±0.48^{a}	44.20±1.30^{a}	15.28±3.12^{b}	30.20±1.30^{a}	1.06±0.14^{c}
MLB	14.80±0.85^{a}	46.00±1.16^{a}	14.58±1.44^{b}	30.80±1.30^{a}	1.14±0.37^{b}

Values are mean (n = 5)±SD. Values with different superscripts a, b, c in each column are significantly different. TDA: *T. diversifolia* administration for 7 days; TDB: *T. diversifolia* administration for 14 days. MLA: *M. lucida* administraton for 7 days; MLB: *M. lucida* administration for 14 days

($p<0.05$) in serum bicarbonate level when compared with the control. Also there was significant decrease ($p<0.05$) in serum, chloride, sodium and potassium levels following 7 and 14 days of administration of the extract. However, 14 days administration of *T. diversifolia* significantly increased ($p<0.05$) serum creatinine level when compared with the control.

Table 6 shows the effect of the extracts on some haematological parameters. There was no significant difference ($p>0.05$) in the level of circulating haemoglobin, packed cell volume and MCHC in all the treatment goups when compared to the controls. However, 7 and 14 days administration of both extract led to a significant reduction ($p<0.05$) in the level of WBC. Also, 7 and 14 days administration of *M. lucida* extract and 14 days administration of *T. diversifolia* caused a significant increase ($p<0.05$) in platelet counts when compared to the controls.

DISCUSSION

Methanolic extract of the leaves of *T. diversifolia* and *M. lucida* showed significant anti-plasmodial activity as depicted by the percentage parasitaemia and chemosuppressive properties of the two extracts with *M. lucida* having a higher efficacy when compared with *T. diversivolia*. Although rodent models do not produce exactly the same signs and symptoms observed in the human plasmodial infection (Pedroni *et al.,* 2006) they, however, have been reported to produce disease features similar to those of human plasmodial infection, when infected with *P. berghei* (Alli *et al.*, 2011). Some of the phytochemicals identified in these plants have been established to have antiplasmodial and antiprotozoal activities (Arise *et al.*, 2012). Indeed, tannin C and some other sesquiterpene lactone isolated from *T. diversifolia* might be responsible for the anti-malarial property of the plant (Goffin *et al.*, 2002).

The measurement of the activities of various enzymes in tissues and body fluids plays a very significant role in disease investigation and diagnosis (Malomo, 2000). The choice of the enzymes studied was based on their locations, such that any change in their activities will give strong indication of cellular impairment. Ngaha (1981) reported that cell injury could be correlated and determined by assaying the level of activities of 'marker' enzymes in such cell. AST and ALT are important in assessing and monitoring liver cytolysis (Wada and Snell, 1962) while ALP has been used as an assessment of membrane integrity of most tissues (Akanji *et al.*, 1993). The reduction in ALP activity as a result of administration of the extracts may be adduced to either loss of membrane components into the extracellular fluid and serum (Malbica and Hart, 1971) or inactivation of the enzyme molecule. The lack of concurrent increase and significant reduction in serum ALP activity suggest that reduction in ALP activities observed during this study may be due to inactivation of the enzyme molecule. The reduction in liver AST activities with concurrent rise in serum activity of the enzyme may be an implication of loss of permeability function of the membrane, resulting in leakage of the enzymes from the cell. However, the significant decrease in liver and kidney AST activities following 14 days administration of *M. lucida* without a relative rise in serum activity of the enzyme may be attributed to reduced rate of synthesis of the enzyme. It further suggests that the extract may have inhibitory effect on the synthesis of the liver and kidney enzyme following prolonged administration. Similar reasons as with liver ALP can be attributed for the significant decrease in liver and kidney ALT activities with concurrent decrease in serum ALT activity.

White blood cells play important role in the immune system. The significant reduction observed in white blood cells following administration of the extracts for 7 and 14 days might be an indication that the immune system might be affected. Decreased white blood cell is an indication of reduced immunological action of the body (Qiao *et al.*, 1991). Thus, the result suggests immunosuppressive potential of the extracts. Platelets play major role in mediating blood clotting, modulating inflammatory and immune responses. This is achieved by the regulated expression of adhesive and immune receptors on the platelet surface and by the release of a multitude of secretory products including inflammatory mediators and cytokines, which can mediate the interaction with leukocytes and enhance their recruitment (Von Hundelshausen and Weber, 2007). The significant increase in the platelets counts observed following administration of *T. diversifolia* for 7 days and *M. lucida* for 7 and 14 days may be an indication that the extracts have stimulatory effect on megakaryocyte which regulates platelets production and release.

Renal function test are required either to demonstrate the presence or absence of active lesion in the kidney, or to assess the normal functioning capacity of the kidney (Guyton and Hall, 2000). Inorganic electrolytes which occur in large quantities in both extracellular and intracellular fluids mediate the transport of water and solute molecule across the kidney tubule. The significant decrease in sodium and potassium ions caused by the extract may be an indication that the functional capacity of the nephron has been compromised. This suggests a possible adverse effect of these extract on the normal functioning of the sodium pump. This is supported by significant decrease in serum chloride level since, Na^+/K^+ and Cl^- are co-transported. Thus, the extracts may potentiate some level of diuretic effects. The decrease in serum bicarbonate level observed following administration of *M. lucida* for both periods of administration is possibly due to increase in plasma acidity, a mechanism of buffering the plasma and possibly indicating systemic acidosis. Increase in plasma H^+ concentration results in lowered bicarbonate level which counteract the acidic condition of the plasma. Bicarbonate is one of the major inorganic buffers in serum. The kidney plays an

important role in homeostasis, secreting an excess of hydrogen ions which may result into increased efflux of sodium bicarbonate into the extracellular fluid (Guyton and Hall, 2000). The kidney plays an important role in clearing the product of metabolism of other tissues from the blood. Therefore, levels of some of these products in the serum are used as an assessment of kidney functionality such as creatinine, produced from muscle metabolism and urea, from liver metabolism. The drop in serum urea and creatinine level observed in this study could be traced to increase in functionality of the kidney at clearing metabolic byproducts.

CONCLUSION

Prolonged and repeated administration of methanolic extract of *Tithiona diversifolia* and *M. lucida* potentiated some level of toxicity. However, the extracts displayed considerable anti-malarial property with *M. lucida* having a higher efficacy and may be a template for short term anti-malarial drug development.

REFERENCES

Adewuyi, J.O. and P.O. Olatunji, 1995. Manual of Practical Haematology for the MBBS Course. University of Ilorin, Ilorin, Nigeria.

Akanji, M.A., O.A. Olagoke and O.B. Oloyede, 1993. Effect of chronic consumption of metabisulphite on the integrity of the rat kidney cellular system. Toxicology, 81: 173-179.

Alli, L.A., A.A. Adesokan, O.A. Salawu, M.A. Akanji and A.Y. Tijani, 2011. Anti-plasmodial activity of aqueous root extract of *Acacia nilotica*. Afr. J. Biochem. Res., 5: 214-219.

Arise, R.O., S.O. Malomo and M.M. Lawal, 2012. Comparative antimalarial and toxicological effects of artemisinin with methanolic extract of *Carica papaya* leaves and bark of *Alstonia broonai* in animal models. Adv. Natl. Applied Sci., 6: 116-123.

Coker, H.A., C.M. Chukwani, N.D. Ifudu and B.A. Awa, 2001. The malaria scourge concepts in disease management. Nig. J. Pharm., 32: 19-47.

Duncan, D.B., 1955. Multiple range and multiple F tests. Biometrics, 11: 1-42.

Goffin, E., E. Ziemons, P. De Mol, C. de Madureira Mdo and A.P. Martins *et al*., 2002. *In vitro* antiplasmodial activity of *Tithonia diversifolia* and identification of its main active constituent: Tagitinin C. Planta Med., 68: 543-545.

Guyton, A.C. and J.E. Hall, 2000. A textbook of Medical Physiology. 10th Edn., W.B. Saunders Co., Philadelphia, pp: 382-401.

Kochar, D.K., P. Singh, P. Agarwal, S.K. Kochar, R. Pokharna and P.K. Sareen, 2003. Malarial hepatitis. J. Assoc. Phys. India, 51: 1069-1072.

Malbica, J.O. and L.G. Hart, 1971. Effect of adenosine triphosphate and some anti-inflammatory agents on a purified lysosomal fraction having high acid phosphatase and labile-glucuronidase activity. Biochem. Pharmacol., 20: 2017-2026.

Malomo, S.O., 2000. Toxicological implication of triaxone administration in rats. Nig. J. Biochem. Mol. Biol., 15: 33-38.

Ngaha, E.O., 1981. Renal effects of potassium in the composition of excretion with corresponding tissue pattern. Gen. Pharmacol., 12: 291-358.

Palaniswamy, M., B.V. Pradaep, R. Sathya and J. Angayarkanni, 2010. *In vitro* anti-plasmodial activity of *Trigonella foenum-graecum* L. Evid. Compl. Altern. Med., 7: 441-445.

Pedroni, H.C., C.C. Bettoni, S.M. Spalding and T.D. Costa, 2006. *Plasmodium berghei*: Development of an irreversible experimental malaria model in Wistar rats. Exp. Parasitol., 113: 193-196.

Phillipson, J.D. and C.W. Wright, 1991. Medicinal plants against protozoal diseases. Trans. R. Soc. Trop. Med. Hyg., 85: 155-165.

Qiao, Z.S., H. Wu and Z.W. Su, 1991. Comparison with the pharmacological actions of *Morinda officinalis*, *Damnacanthus officinarum* and *Schisandra propinqua*. Zhong Xi Yi Jie He Za Zhi, 11: 415-417.

Reitman, S. and S. Frankel, 1957. Determination of serum transaminases. Am. J. Clin. Pathol., 28: 56-59.

Roth, K.S. and J.C. Chan, 2001. Renal tubular acidosis: A new look at an old problem. Clin. Pediatr., 40: 533-543.

Roy, A.V., 1970. Determination of alkaline phosphatase in human serum. Clin. Chem., 16: 431-431.

Salisu, S.O., 2004. Manual of Practical Chemical Pathology for M.B.B.S. Course. Unilorin Press, Ilorin, Nigeria, pp: 4.

Tietz, N., L.E. Pruden and S. Andersen, 1996. Electrolytes: In Tietz Fundamentals of Clinical Chemistry. 2nd Edn., W.B. Saunders Co., USA., pp: 721-738.

Trampuz, A., M. Jereb, I. Muzlovic and R.M. Prabhu, 2003. Clinical review: Severe malaria. Crit. Care, 7: 315-323.

Von Hundelshausen, P. and C. Weber, 2007. Platelets as immune cells: Bridging inflammation and cardiovascular disease. Circ. Res., 100: 27-40.

WHO, 2005. World malaria report. WHO Library Cataloguing-in-Publication Data, pp: 1-293.

Wada, H. and E.E. Snell, 1962. Enzymatic transamination of pyridoxamine: II. Crystalline pyridoxamine-pyruvate transaminase. J. Biol. Chem., 237: 133-137.

West, B.J. and R.A. Sabin, 2012. Efficacy of a *Morinda citrofolia* based skin care regimen. Curr. Res. J. Biol. Sci., 4: 310-314.