

## Research Article

# Concurrent Short Term Administration of Artesunate and Methanol Extract of *Ficus Platyphylla* Has No Hepato-Renal Consequences in Rats

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**Abstract.** Many believe that the use of orthodox drugs alongside herbal medications brings about enhanced efficacy. Thus, it is not uncommon to see these combinations in malaria treatment. However, this combination may lead to toxicity through drug-herb interaction. The liver and kidneys being important organs in metabolism and excretion of xenobiotics are potential target organs for the suspected adverse effects. This study hypothesized that the co-administration of artesunate and methanol stem bark extract of *Ficus platyphylla* may result in hepato-renal consequences. Twenty male wistar rats were divided into four groups of five rats each. Group one served as the normal control group and was treated with normal saline at a dose of 1 ml/kg. Rats in group two were treated with 300 mg/kg of *Ficus platyphylla* alone while rats in group three were treated with 2.9 mg/kg of artesunate alone. Furthermore, rats in group four were treated with *Ficus platyphylla* and artesunate at a dose of 300 mg/kg and 2.9 mg/kg respectively. All treatments were done orally for five continuous days within which body weight was determined. At the end of the treatment period, liver markers levels (alanine aminotransferase, aspartate aminotransferase and alkaline phosphatase) and kidney markers (urea, creatinine, uric acid, albumin and total protein) were determined. There were no statistically significant differences ( $p > 0.05$ ) in body weights, hepatic and renal biomarkers across all treated groups when compared to the control. These results may indicate the safety of this drug-herb combination when used in malaria therapy.

**Keywords:** malaria, artesunate, *Ficus platyphylla*, drug interactions

## 1. Introduction

Drug interaction may result when two or more drugs are concurrently administered [1]. Interactions could also result between drugs and foods (drug-food interactions), as well as between drugs and herbal medicines, also known as drug-herb interactions [2]. This may affect pharmacokinetic and pharmacodynamic outcomes of medications leading to

either therapeutic failure or toxicity [3]. In West Africa, co-administration of orthodox drugs and herbal medications is common and could be before, during or after treatment of malaria [4]. Malaria is a serious public health problem with increasing morbidity and mortality especially among developing countries. In 2017, about 219 million cases of malaria were reported with about 435,000 related deaths. Report suggested that Africa accounted for 92% and 93%

of the prevalence and mortality respectively [5]. Effective therapy is therefore required to reduce the morbidity and mortality as well as improve quality of life.

Artesunate, a derivative of artemisinin, has remained one of the drugs of choice used in the treatment of especially chloroquine and quinine resistant malaria and is highly effective against multidrug-resistant falciparum. It also provides an advantage over artemisinin combination due to a better absorption profile [6]. It is used for treatment of severe malaria and has proven effective with much faster fever resolution than occur with quinine [7][8]. Medicinal plants have received wide acceptability especially in low and middle income countries especially in Africa due to poor accessibility to orthodox drugs [9]. At the global stage also, majority of the world population still depends on traditional medicine such as herbs for treatment of various ailments [10]. *Ficus platyphylla* Del. Holl. (Moraceae) is a deciduous medicinal plant that is widely distributed throughout West Africa. It is widely distributed in Northern Nigeria and locally known as "gamji". It has gained a profound usage among native dwellers in Hausa communities and is used traditionally with widely acclaimed efficacy for the management of epilepsy, insomnia, psychosis, depression, pain and inflammation [11]. In addition, *Ficus platyphylla* has been scientifically validated to possess antimalarial activity [12] and a more recent study has established its safety in rats following 28 days oral administration [13]. This suggests the possibility of concurrent administration of artesunate and *Ficus platyphylla* for synergistic or additive effect in the treatment of malaria thus necessitating an investigation on the hepato-renal consequence of the combination.

## 2. Materials and Methods

**2.1. Plant Collection.** The stem bark of the plant was collected from Zaria, Kaduna State, Nigeria in May, 2018 and was identified by a taxonomist in the Department of Biological Sciences, Ahmadu Bello University Zaria by comparing with a specimen voucher number 900106 previously deposited in the herbarium.

**2.2. Preparation of Extract.** The collected plant materials were air-dried at room temperature for two weeks and size reduced with a mortar and pestle. The extraction was done by cold maceration using absolute methanol. 200 g of the powdered plant material was added into Erlenmeyer flask containing 1000 ml of methanol and kept for 48 hours with intermittent shaking, then filtered. The obtained filtrate was afterwards evaporated to dryness in an evaporator under reduced pressure at a temperature of 40-60°C. After evaporation, the extract was preserved in an air tight container for future use.

**2.3. Animals.** Male wistar rats weighing between 140-160 g were obtained and housed in the Animal House facility of the Department of Pharmacology and Therapeutics, Ahmadu Bello University, Zaria, Nigeria. They were maintained under natural day and night cycle and were allowed access to food and water *ad libitum*. The study was conducted according to ethical guidelines on laboratory animal use and care policy, which is in compliance with Ahmadu Bello University Research Policy.

**2.4. Drugs and Chemicals.** The chemicals and drugs used for the studies include; Normal saline (Dana Pharmaceuticals Limited, Nigeria), Artesunate (Mekophar Chemical Pharmaceutical Company, VD-10618-10) Chloroform (Sigma Chemical Co. USA), Methanol (Sigma Chemical Co. USA), Methanol extract of *Ficus platyphylla* (MEFP).

**2.5. Equipment and other Materials.** Animal cages, pestle and mortar, syringes, dissecting kits, mettler balance p165, measuring cylinders, separating funnel, beakers, sample bottles, funnel, water bath (Gallenkamp Cat No: H1054), water drinkers, evaporating dish, weighing balance, spectrophotometer, Randox<sup>®</sup> diagnostic kits.

**2.6. Acute Toxicity Studies ( $LD_{50}$ ).** The  $LD_{50}$  of the extract was determined according to the method in [14]. This was done in two phases, and in both cases, rats were fasted for 16-18 hours before administration. The extract was reconstituted in distilled water to obtain a stock solution before administration. In the first phase, three groups of three rats each were treated with the extract orally at a dose of 10 mg/kg, 100 mg/kg and 1000 mg/kg respectively. After the treatment the animals were observed for four hours for possible signs of toxicity. No death was recorded after 24 hours; the second phase was then initiated. In this phase, four rats were treated with the extract orally at a dose of 1200 mg/kg, 1600 mg/kg, 2900 mg/kg and 5000 mg/kg respectively. The rats were then observed for possible signs of toxicity for the first 4 hours and mortality for 24 hours. After phase two,  $LD_{50}$  was calculated as the arithmetic mean of the lowest dose that killed an animal and the highest dose that did not kill.

**2.7. Experimental design.** Twenty male albino wistar rats were divided into four groups of five rats each. Group one served as the normal control group and was treated with normal saline at a dose of 1 ml/kg. Rats in group two were treated with MEFP alone at a dose of 300 mg/kg while rats in group three were treated with artesunate alone at a dose of 2.9 mg/kg. Furthermore, rats in group four were treated with both MEFP at a dose of 300 mg/kg and artesunate at a dose of 2.9 mg/kg [25]. All treatments were done orally for five continuous days. At the end of the treatment

Table 1: Effect of 5-days concurrent administration of artesunate and *Ficus platyphylla* on body weight

Groups	Day 1	Day 2	Day 3	Day 4	Day 5
Normal control	150 ± 1.20	150 ± 0.90	151 ± 1.10	152 ± 1.00	153 ± 1.20
MEFP 300 mg/kg	150 ± 1.00	150 ± 0.70	152 ± 1.80	153 ± 1.20	153 ± 1.40
ASNT 2.9 mg/kg	150 ± 1.10	150 ± 0.90	150 ± 1.10	152 ± 1.00	152 ± 1.50
MEFP300mg/kg +ASNT 2.9mg/kg	150 ± 1.20	151 ± 0.89	152 ± 1.90	152 ± 1.20	153 ± 1.20

Values are Mean ± SEM, n= 5, oral route, ASNT 2.9 mg/kg = treated with artesunate at dose of 2.9 mg/kg, MEFP 300 mg/kg = treated with methanol extract of *Ficus platyphylla* at a dose of 300 mg/kg, No significant difference after analysis with one way ANOVA.

Table 2: Effect of concurrent administration of artesunate and *Ficus platyphylla* on liver function indices.

Groups	Alanine Transferase (U/L)	Aspartate Transferase (U/L)	Alkaline Phosphatase (U/L)
Normal control	30.36 ± 1.70	40.50 ± 0.30	41.01 ± 1.10
MEFP 300 mg/kg	31.31 ± 3.00	41.92 ± 1.90	41.22 ± 2.40
ASNT 2.9 mg/kg	32.21 ± 2.10	42.12 ± 1.20	43.32 ± 2.20
MEFP 300 mg/kg + ASNT 2.9 mg/kg	31.86 ± 1.20	41.25 ± 0.65	44.25 ± 1.95

Values are Mean ± SEM, n= 5, oral route, ASNT 2.9 mg/kg = treated with artesunate at dose of 2.9 mg/kg, MEFP 300 mg/kg = treated with methanol extract of *Ficus platyphylla* at a dose of 300 mg/kg, No significant difference after analysis with one way ANOVA.

period, the rats were euthanized under mild chloroform, and blood was collected from the jugular vein in plain blood sample containers. Blood samples obtained were centrifuged at 4000 rpm for 15 minutes and liver markers such as alanine aminotransferase (ALT), aspartate aminotransferase (AST) levels and alkaline phosphatase (ALP) levels as well as kidney markers (urea, creatinine, uric acid, albumin and total protein) were determined using Randox<sup>®</sup> diagnostic kits using the protocols described by manufacturer as adopted in [26].

**2.8. Data Analysis.** Data were analyzed using one way analysis of variance (ANOVA) for liver and kidney parameters followed by Dunnett post hoc test, and split plot ANOVA followed by Bonferoni post hoc test for body weight. Data were presented as mean ± SEM. Results were considered significant at P < 0.05.

### 3. Results

**3.1. Acute Toxicity Study.** The oral acute toxicity (LD<sub>50</sub>) was found to be greater than 5000 mg/kg body weight. No sign of toxicity was observed in both phases with lower and higher doses, as there were no changes in skin and fur, eyes and mucous membranes, motor activity and behaviour pattern, autonomic and central nervous systems. In addition, tremors, convulsions, salivation, diarrhoea, lethargy, sleep and coma were absent.

**3.2. Effect of 5-days concurrent administration of artesunate and *Ficus platyphylla* on body weight.** Data on body weight shows that there was no significant difference in body weight of rats treated with *Ficus platyphylla* and artesunate either singly or in combination, in a similar manner with rats in the control group throughout the duration the study. The result is shown on table 1.

**3.3. Effect of concurrent administration of artesunate and *Ficus platyphylla* on liver function indices.** The effects of concurrent administration of artesunate and *Ficus platyphylla* on liver function indices such as aspartate transferase, alkaline phosphatase, and alanine transferase are shown in table 2. There was no statistical significant difference in all the liver parameters assessed for all the treatment groups when compared to the normal control. The values obtained for the treatment group of rats were similar to that of the control group.

**3.4. Effect of concurrent administration of artesunate and *Ficus platyphylla* on renal function indices.** Data for serum renal markers such as urea, creatinine, uric acid, albumin and total protein are shown on table 3. The result reveals that there was no significant change in serum renal marker for the normal control when compared to the values of groups treated with artesunate or the methanol extract of *Ficus platyphylla*, either singly or in combination. Values for the serum renal

Table 3: Effect of concurrent administration of artesunate and *Ficus platyphylla* on renal function indices.

Groups	Urea (mg/dl)	Albumin (g/dl)	Total Protein (g/dl)	Creatinine (mg/dl)	Uric acid (mg/dl)
Normal control	31.38±0.54	4.04±0.50	4.57±0.93	1.09±0.23	1.90±0.33
MEFP 300 mg/kg	32.23±0.56	4.90±0.37	3.42±0.80	1.10±0.93	1.64±0.61
ASNT 2.9 mg/kg	31.85±0.61	4.38±0.72	5.02±0.48	1.22±0.41	1.91±0.71
MEFP 300 mg/kg + ASNT 2.9 mg/kg	32.86±0.90	4.37±0.17	3.70±0.89	1.23±0.56	1.97±0.13

Values are Mean ± SEM, n= 5, oral route, ASNT 2.9 mg/kg = treated with artesunate at dose of 2.9 mg/kg, MEFP 300 mg/kg = treated with methanol extract of *Ficus platyphylla* at a dose of 300 mg/kg, No significant difference after analysis with one way ANOVA.

marker for all the treatment groups are similar to the values obtained for the normal control group.

#### 4. Discussion

Severe malaria poses a serious health challenge in developing countries especially in the tropics [15]. With the short fall of health care facilities and poor standard of living in these countries, especially in the rural areas, traditional herbal medicine offers an affordable alternative. However, the concurrent use of herbal remedies with orthodox medications may increase the tendency for drug-herb interactions. This may enhance efficacy and/or predispose to toxicity. Hence, the study investigates the possible hepato-renal effects of concurrent administration of *Ficus platyphylla* and artesunate in rats.

In this study, the oral median lethal dose value for the methanol stem bark extract of *Ficus platyphylla* obtained in rats following oral administration was greater than 5000 mg/kg. This may suggest that the plant extract is non-toxic as no death nor sign of toxicity was recorded. This is consistent with previous findings [13], who reported that the methanol stem bark extract of *Ficus platyphylla* is practically non-toxic following acute toxicity studies. A substance is said to be very toxic if the value is less than or equal to 5 mg/kg, and toxic if the value is greater than 5 mg/kg but less 50 mg/kg. Furthermore, it is said to be harmful if the value is greater than 50 mg/kg but less 500 mg/kg, and non-toxic if the value is greater than 500 mg/kg or less than 2000 mg/kg [16]. Acute toxicity studies give an insight as to what dose of a substance or drug will cause deleterious effects and/or death. This helps in dose determination especially for an unknown substance in basic medical research [17].

Generally, weight loss associated with exposure to a potential toxicant serves as an index for toxicity. [18]. There was no significant change in body weights of all groups of animals throughout the duration of the study. This could be of clinical importance, as changes in body weight can have an effect on energy expenditure and has also been reported to be related to catecholamine and thyroid hormone levels [19]. This may suggest that overweight and/or insulin resistant

patients can take this drug-herb combination without risk of weight associated complications.

The liver and kidneys are the most important organs for both metabolism and excretion of xenobiotics, hence, they are predisposed to toxicity. Elevated levels of alanine transferase, aspartate, and alkaline phosphatase are indicative of abnormalities in liver function [20]. In this study, administration of *Ficus platyphylla* and artesunate either in combination or singly did not affect serum liver function markers. This data corroborates earlier findings [13] that reported no change in serum liver markers after 28 days of continuous oral administration. In addition, at low therapeutic doses, artesunate has been found not to interfere with liver functions in rats, however, prolonged administration for four weeks at 76 mg/kg has been found to produce hepatotoxicity [21].

Increased levels of serum creatinine, blood urea nitrogen and uric acid, and reduced levels of serum albumin and protein are common markers for renal toxicity [22, 23]. This present study suggests that both *Ficus platyphylla* and artesunate as single agents or combined have no adverse effect on renal function. This is because the renal function markers for all the groups did not differ significantly from the values of the normal control group. This is in agreement with a recent study where kidney function was left unaltered in rats after 28 days of treatment with methanol stem bark extract of *Ficus platyphylla* [13]. In addition, at therapeutic dose and duration, artesunate does not produce renal toxicity, as the serum levels of creatinine and urea were not affected after five days of oral administration [24], however, at higher doses and with prolonged duration of treatment, toxicity may occur.

#### 5. Conclusion

Concurrent short term administration of the methanol stem bark extract of *Ficus platyphylla* and artesunate does not affect body weight as well as liver and kidney functions in rats. These may indicate safety of this drug-herb combination. Thus, there is possibility of concurrent use of both agents in practice for the purpose of treating especially

complicated and chloroquine resistant malaria without affecting body weight and producing adverse effects on the liver and kidney. Further studies should however be carried out on concurrent administration of both agents to determine both the safety profile and efficacy of the combination in parasitized rats.

## Competing Interests

The authors declare no competing interests.

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