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# Effects of *Maytenus senegalensis* (L) and *Cassia alata* (L) Extracts on the Kidney Function Test Parameters of Albino Rats, Infected with Schistosomes Cercariae

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**Abstract**
The aim of this investigation was to assess the kidney filtration as to ascertain the excretion of creatine and urea of albino rats which were subjected to the treatment with extracts of methanol and aqueous water extracts of *Maytenus senegalensis* and *Cassia alata* previously infected with schistosome cercariae. The normal values of creatine (160-1180µmol/l), and those of urea (10.7-27.1µmol/l) were not affected. This is an indication that the function of the kidney was not alterred neither by the infection with schistosome nor by the treatment with plant extracts with methanol or water, however some treatments such as 9g/kg body weight produced patechial haemorhage.

**Keywords**: Creatine, Urea, Albino rats, *Maytenus senegalensis*, *Cassia alata*

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## Introduction

Schistosomiasis is caused by *Schistosoma* spp. The mortality is low but it causes severe consequences including bladder cancer, renal failure as for *Schistosoma haematobium* infection and liver fibrosis and portal hypertension as in the case of *S. mansoni*. There are over 800 million people exposed to the risk of getting the infection spread over 77 countries (WHO, 2013). In the tropics, schistosomiasis constitutes an important public health problem. More than 682 million had experienced haematuria and 32 million others were with dysuria associated with *S. haematobium* infection (WHO, 2013). It was further estimated that 18 million people suffered bladder wall pathology and 10 million others were with hydronephrosis (WHO, 2013). In africa and other affected countries schistosomiasis is associated with agriculcural and other rural activities around the freshwater bodies such as swimming, fishhing, washing and bathing in ponds, rivers, streams or lakes where the snail intermediate hosts breed especially (John *et al*, 2008 and Shurrock, 2001). Schistosomiasis affects the health and the economy of infected population by reducing their ability to work (WHO, 2010) and infected people are generally poor and may not have the resources to afford the cost of modern treament and generally rely on traditional medicine (Mohammed *et al*., 2007).

Praziquantel (PZQ) is currently the drug of choice in the treatment of both intestinal and urinary schistosomiasis (Gonnert and Andrews, 1977) and several clinical important helminthic diseases in human (Archer, 1985). The potential for the development of resistance of schistosomes to PZQ was highlighted in 1995 by its apparently low efficacy when used to treat a newly established focus of *Schistosoma mansoni* in Senegal (Stelma *et al*.1995). The use of these plants by humans in curing severals ailments poses undoubtedly some problems since doses of such of preparations could not be determined. It is also necessary to investigate their therapeuthic effects as well as the level of their toxicity to humans. It is for this purpose that *Maytenus senegalensis* and *Cassia alata* had been selected, known as thorny staff and ringworm or yellow bush respectively.

This work is aimed at screening two medicinal plants as anti-schistosomal agents namely *Cassia alata* and *Maytenus senegalensis* and studying their effects on the body biochemicals. The specific objectives are to screen and evaluate the potential effects of these plants used by traditional herbalists in the body's kidney using laboratory rats as indicators. Also to compare the properties of the various extracts of the plants using methanol and water as the extracting solvents.

## Materials and Methods

### Collection of Plant materials, Snails and Schistosome eggs

The two plants *Maytenus senegalensis* and *Cassia alata*, were collected from Bauch and Mubi regions of Nigeria, and were identified based on the characteristics of the leaves, flowers, fruits, stem- bark, Stanfield and Hopkins (1966), Hutchinson and Dalziel (1968). The useful parts of each of the plants (leaves, seed and roots) were washed and dried under shade in the greenhouse. Each part was then separately pounded using a wooden mortar and pestle and then sieved through ordinary flour sieve of mesh size of 0.30 mm and the powder from each was separately stored in labelled air tight polyethene bags until required for use.

*Bulinus* snails were collected and kept into clean medium size aquarium, measuring 30 x 45 x 30 cm filled to 2/3 capacity with de-ionized water. The de-ionized water was changed forthnightly to reduce pollution. All the snails were fed with dry lettuce kept either in the oven or at ambient temperature as described by Madsen (1985).

Urine samples were collected from out patients clinics and examined for eggs of schistosomes. Stool samples were immediately processed by the keto-kartz method and before being filtered using Whatman No.2 filter paper

### Collection Rearing and Infection of Rats with Schistosome Cercariae.

Albino Winstar rats used in the experiments were purchased from (National Institute for Trypanosomiasis Research (NITR), Vom, Plateau State, and reared in well ventilated rat cages in the Animal House of the Biological Sciences Research Complex of, Adamawa State University, Mubi.

Infected snails were periodically exposed to strong electric light as from day 25 post infection to monitor the shedding of cercariae. The cercariae snails shedding were exposed to strong electric power light to induce the shedding of cercariae. Thereafter about 100 cercaraiae were transferred into a container each having a rat. The cercariae in the container were allowed to penetrate each of the rats, a method known as paddling After six (6) to seven (7) weeks post of infection, these were divided into groups of 5 rats each. Two replicates each of the infected rats served as experimental and control respectively.

### Formulation, Administration of Praziquantel and of Plant Materials

The standard anti-schistosomal drug Praziquantel tablets formulated 60mg, Batch No. 121257, Nafdac Reg. No. A4-4246 manufactured by Yanzhou Xier Kangtai Pharmaceutical Co., Ltd. China and marked by Chez Resources Pharm were purchased and used. The drug was administered orally as single dose of 600mg/kg of body weight as recommended by WHO (2003). The

quantity of Praziquantel given to each rat was obtained by using the following formulation:

$$\textbf{Weight of PZQ(mg)} = \frac{\textbf{600mg xWeight of Animal (mg)}}{\textbf{1000mg}} \quad \textbf{(1)}$$

After the treatment, a period of 10-14 days was accorded, after which animals were sacrificed for examination of the therapeutic effect of the drug.

Similarly, plant materials were weighted following the same procedure as above. Thereafter a single oral dose of various concentrations were administered to each rat using stomach gavage using the same formula:

$$\textbf{Weight of Plant extract (mg)} = \frac{\textbf{(A)mg xWeight of Animal (mg)}}{\textbf{1000mg}} \quad \textbf{(2)}$$

**(A)** represents the dose: 3, 6 or 9g/kg body weight

**Statistical Analysis**

Statistical analyzes were performed using the Fisher's test for contingency analysis and the student independent t-test. These analyses were performed using the Statistical Package for Social Sciences software (SPSS 17.0, Chicago, IL, USA). Continuous variables were expressed as means ± standard deviation (SD) of the median and range. P-values of less than 0.05 were considered statistically significant.

## Results and Discussion

The kidney function tests were performed based on instructions of the manufacturers as contained in the leaflet and the results are shown in the table 1 below.

***The Creatine Values***

Results for creatine values of rats in treated and control groups are shown in Tale 1. For the roots of *Maytenus senegalensis*, the highest average creatine count is 149 µm/l recorded in the group dosed 3g/kg body weight of methanol extracts of the plant., while the lowest average count 130 µm/l was found in the 6g/kg body weight rats. For the water extracts, the highest average value was 131 µm/l recorded in those dosed with 6g/kg body weight. The lowest value of 121 µm/l was the group given 9g/kg body weight.

For rats in the methanolic extracts of the leaves of *M. senegalensis*, the average highest creatine count was 140 µm/l recorded in those given 9g/kg body weight of extracts, with the lowest value of 127 µm/l obtained in 3g/kg body weight treated groups. The corresponding water extracts of the plant had a higher average value of 169 µm/l for rats dosed in the 3g/kg body weight. The lowest value of 122 µm/l was in the 9g/kg body weight treated group.

The methanolic extracts of the stem-bark of *M. senegalensis* recorded the highest average count of 206 µm/l for the groups dosed with 6g/kg body weight, while the lowest value of 161 µm/l was in those given 3g/kg body weight of extracts. The water extracts of the plant had the highest creatine average value of 176 µm/l in the 6g/kg body weight of treated rats. The average lowest value was 137 µm/l in rats given 3g/kg and 9g/kg body weight of extracts.

For methanolic leaf extracts of *Cassia alata*, the average highest creatine count was 197 µm/l, recorded in the 9g/kg body weight group. This decreased to its lowest value 179 µm/l in rats given treatments of 6g/kg body weight of extracts. Results for the water extracts revealed, the highest average count of 193 µm/l recorded in the 9g/kg body weight group which decreased to its lowest count of 172 µm/l in those treated with 3g/kg body weight.

Values for those in the methanolic extracts of the seeds of *Cassia alata*, revealed in the same Table 1, the highest average count value of 219 µm/l for groups of rats dosed with 3g/kg body weight, while the lowest value of 136 µm/l was found in the 6g/kg body weight group. The highest average count for water extracts was 163 µm/l in the 3g/kg body weight treated group, while the lowest count of 138 µm/l recorded in the 6g/kg body weight treated group.

Table 1: Result of kidney clearance test of rats treated with various doses of plant extracts and Praziquantel

| Plants | Parts | Doses | Methanol | | Water | |
|---|---|---|---|---|---|---|
| | | | Parameters | | Parameters | |
| | | | Creatinine (µm/l) | Urea (µm/l) | Creatinine (µm/l) | Urea (µm/l) |
| *Maytenus senegalensis* | Root | 3g/kg | 149 | 8.8 | 125 | 10.5 |
| | | 6g/kg | 146 | 7.2 | 131 | 8.4 |
| | | 9g/kg | 130 | 7.8 | 121 | 7.6 |
| | Leaves | 3g/kg | 127 | 8.8 | 169 | 9.6 |
| | | 6g/kg | 131 | 8.9 | 124 | 7.8 |
| | | 9g/kg | 140 | 10.0 | 122 | 7.4 |
| | Stem | 3g/kg | 16 1 | 8.1 | 137 | 8.0 |
| | | 6g/kg | 206 | 8.1 | 176 | 8.6 |
| | | 9g/kg | 164 | 8.4 | 137 | 8.0 |
| *Cassia alata* | Leaves | 3g/kg | 183 | 8.1 | 172 | 8.0 |
| | | 6g/kg | 179 | 9.5 | 175 | 9.0 |
| | | 9g/kg | 197 | 9.0 | 193 | 10.4 |
| | Seed | 3g/kg | 219 | 8.9 | 163 | 8.4 |
| | | 6g/kg | 136 | 8.5 | 138 | 8.1 |
| | | 9g/kg | 138 | 8.2 | 162 | 7.8 |
| Control | | PZQ | 277 | 13.9 | 277 | 13.9 |
| | | Not Infected | 197 | 18.4 | 197 | 18.4 |
| | | Inf. Not Treated | 193 | 28.1 | 193 | 28.1 |

The control groups treated with Praziquantel® had the highest average count of 277 µm/l, the infected non-treated rats recorded an average high count of 193 µm/l. The non-infected control group on the other hand recorded a count of 197 µm/l. ANOVA test revealed that these differences are not significant $P \leq 0.05$).

In the control groups, the average count recorded in the Praziquantel® treated group was (277 µmol/l), while the infected not treated recorded an average as high as (193 µmol/l). The non-infected control group recorded a count of (197 µmol/l). There was no significant difference statistically ($P \leq 0.05$) between the various doses of the methanol and the water extracts of the roots, leaves, stem-bark of *M. senegalensis*, the leaves, seed of *C. alata* and the Praziquantel® treated as well as the infected non-treated group.

The normal values of creatine in albino rats range from 160-1180µmol/l as per Giknis and Clifford (2008) works. Frances (2002) observed that decrease in creatine was linked to hepatic disease or renal failure. Frances (2002) observed also that the value of creatine could be affected by drugs such as steroids, the various extracts formulations given to the rats. However there was no significant variations in the values of the ceatine in all the rats in the experiment**.**

***The Values of Urea***

Value level of urea generated by the rats in treated and control groups are presented in Table 1. For the roots of *Maytenus senegalensis*, the highest average urea count is in rats treated with 3g/kg body weight of methanolic extracts was 8.8 µm/l while the lowest average count of 7.2 µm/l urea was found in the 6g/kg body weight treated rats. For the water extracts, the highest urea average value was 10.5 µm/l obtained in rats treated with the 3g/kg body weight group. The lowest count of 7.6 µm/l was recorded in the 9g/kg body weight group.

For the methanolic extracts of the leaves of *M. senegalensis*, the average highest urea count was 10.0 µm/l recorded in the 9g/kg body weight. This dropped to its lowest count of 8.8 µm/l in those treated with 3g/kg body weight of the extracts. Urea value for rats in water extracts had its highest urea average of 9.6 µm/l in rats dosed with 3g/kg body weight of extracts, while the lowest urea value was 7.4 µm/l obtained in the 9g/kg body weight treated rats.

For methanolic extracts from stem-bark of *M. senegalensis*, the average highest urea count of 8.4 µm/l was recorded in rats given 6g/kg body weight doses of extracts, while the lowest value of 8.1 µm/l was in the 3g/kg and 6g/kd body weight group. The urea values for rats in water extracts, was 8.6 µm/l in those dosed with 6g/kg body weight while the lowest value of 8.0 µm/l was in the 3g/kg and 9g/kg body weight groups.

The urea values for the methanolic extracts of leaves of *Cassia alata*, in Table 1, revealed that the average highest count of 9.5 µm/l was recorded in rats treated with the 6g/kg body weight of extracts, while the lowest value of 8.1 µm/l was in those treated with 3g/kg body weight. In water extracts of the same *C. alata*, the highest urea average count was 10.4 µm/l recorded in the 9g/kg body weight treated rats, while the lowest value of 8.0 µm/l was obtained in those given 3g/kg body weight of water extracts of the plant.

The methanolic extracts of seeds of the same plant used in treating these rats at 3g/kg body weight gave an average urea count of 8.9 µm/l. This decreased to its lowest level of 8.2 µm/l in rats treated with 9g/kg body weight for the extracts. For the water extracts, the highest average urea value was 8.4 µm/l in the 3g/kg body weight group, while the lowest value of 7.8 µm/l was in the 9g/kg body weight category.

The control groups treated with Praziquantel®, recorded urea value of 13.9 µm/l, while those infected non-treated rats recorded an average urea value of 28.1 µm/l, that was higher than the Praziquantel®.groups. Rats in the non-infected control group recorded an average count value of 18.4 µm/l. ANOVA test carried on the data showed that these variation are not significant ($P \leq 0.05$).

The serum urea of albino rats was also assessed. According to the works of Giknis and Clifford (2008), the standard values of urea range 10.7-27.1µmol/l. All the values recorded in this research work fall within the normal values except the value of urea recorded from the control group of infected non-treated. Walaa *et al*., (2015), in a separate investigation also observed some deviating results with some urea contents.

**Conclusion**

The crude methanol and water extracts from the roots, leaves, stem-bark of *Maytenus senegalensis* as well as that of the leaves and seed of *Cassia alata* showed varying levels of antischistosomal activities on schistosome infected winstar albino rats. The results obtained from the kidney parameters the treated rats showed that the 6g/kg body weight had a good potential :

a) The extracts from the roots of *M. senegalensis* at the dose of 9g/kg body weight and above should not be administered as antischistosomal agents due to its haemolytic effects and had side effects on the lungs by swallen it and showing signs of patechial haemorrhage.
b) The extracts from the stem-bark of *Maytenus senegalensis* had side effects on the lungs by having it swollen up.
c) The extracts from the leaves of *Cassia alata* can cause ulcers and tumors of the intestines of rats.
d) The extracts from the seed of *Cassia alata* can cause hepatomegaly in rats. Therefore no plant part of *C. alata* should be used as antischistosomal drug dispite some good result because of its side effects.

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