A. SATIVUM IN THE PREVENTION OF SCHISTOSOMIASIS IN SOKOTO, NIGERIA: EVALUATION OF PHYTOCHEMICAL CONTENTS, ACUTE TOXICITY, AND EFFECT ON SOME KIDNEY FUNCTION PARAMETERS IN RATS

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Abstract

Schistosomiasis is an infectious disease of public health importance in the African countries. Due to poor prevention, poverty, and poor settings; areas like Sokoto, Nigeria are battling with the disease. Consequently, people are compelled to use preventive measures locally. There is recorded use of A. sativum for the prevention of schistosomiasis without enough scientifically supported information in that regards. Therefore, this work evaluated phytochemicals, acute toxicity, and subchronic toxicity (effects on some kidney indices). Standard methods and reagents of analytical grade were utilized. The work reveals the presence of some phytochemicals, no acute toxicity (at 4000 mg/kg body weight), as well as no major change in the urea, uric acid, and creatinine levels. 1.111 mg/dl to 1.118 mg/dl creatinine, 1.011 mg/dl to 1.025 mg/dl urea, and 1.120 mg/dl to 1.140 mg/dl uric acid were determined in control and highest dose (4000 mg/kg body weight) (p<0.05). This indicates that, the plant is likely to elicit little effects on the experimental animals. Much work is important to evaluate sufficiently the safety of the A. sativum in prevention of Schistosomiasis in the Sokoto, Nigeria.

Keywords: A. Sativum; Schistosomiasis; Phytochemicals; Liver Function
INTRODUCTION

Life on earth is not possible without the presence of plants, because they have the ability to make their own food from sunlight and other simple organic molecules. On the other hand, the humans (animals) practice a heterotrophic mode of nutrition by depending on plants for source of organic substances. Additionally, humans depend on plants since from time immemorial for their healthcare needs (Mana et al., 2022; Hamza et al., 2023). Nowadays, the modern medicine is redirecting its searchlight on the plant-based materials for the therapeutic purposes to curb the growing trend of antimicrobial resistance, pollution, and other uncertainties (Mana et al. 2022; Hamza et al., 2023). The need for alternative plant-based materials like A. sativum is more aggravated in places like Sokoto State, Nigeria where there are rural settings with poor health care provisions, poor health education, and preventive measures are insufficient (Mohammed et al., 2022). Meanwhile, Schistosomiasis is more prevalent among rural people living near water sources (such as pond, and lake), people living with poor sanitation, and people exhibiting poor preventive measures (Muhammad et al., 2011; Mohammed et al., 2022).

Nigeria is the country having highest prevalence of the disease in the year 2010. Thus, preventive measures of various means are employed. Synthetic chemicals are used. However, people in most rural areas or poor settings are unable to access chemotherapy due to poverty or lack of education (Aula et al., 2021; Anyolitho et al., 2022). Likewise, the synthetic chemicals utilized in prevention against the intermediate host (snail) are scarce and pollutants (Sady et al., 2015; Dawaki et al., 2016; Mana et al., 2022). Thus, it is good to seek for a better alternative that is biofriendly, cheap, and available. People search for better alternatives and utilized A. sativum as an alternative preventive measure (in chemotherapy and molluscicidal activity) against Schistosomiasis in Sokoto, Nigeria (Gamde et al., 2022; Mana et al., 2022). However, there is scarce information about the possible effect of A. sativum on humans and the phytochemical constituents of the plant (Lawal et al., 2016; Suleiman et al., 2018a; Johnbull et al., 2021). Kidney is very important, when it is affected, the biochemical balance of the body is easily distorted and results in morbidity and even death (Muhammad et al., 2011). Therefore, the objective of this work was to carry out a phytochemical analysis, acute toxicity evaluation, and sub-chronic toxicity evaluation of A. sativum in Sokoto, Nigeria.
METHODS

Preparation of Plant Materials Used in the Research

The bulb of *Allium sativum* were purchased from Sokoto market, Nigeria and identified and confirmed by a senior plant taxonomist from Biological Sciences Department, Usmanu Danfodiyo University Sokoto. Powder of *A. sativum* bulbs were prepared by peeling and slicing healthy cloves into 3mm thick, air dried and pulverizing into a mortar and pestle. The powder was kept dry, stored in air tight container in refrigerator and tested for toxicity on rats and other steps of the study.

Mthanolic Extraction

Five hundred grams (500g) of air-dried *A. sativum* (bulbs), was extracted with 1.5 liters of Methanol. The extraction was kept in orbital shaker for 30 minutes. The extracts were filtered, using muslin cloth and concentrated to dry under reduced pressure in a rotary evaporator at 40°C which yield ethanolic extract of *A. sativum*. The extracts were kept in fridge in Laboratory for further use.

Animals: Albino rats (males and females) weighing 165 to 300 g were purchased from the Department of Biological Sciences, Usmanu Danfodiyo University, Sokoto, Nigeria kept at the animal house of the department in a wire mesh cages fed with grower’s feed and tap water *ad libitum* for two weeks to aclimatize before starting the experiment. Animal treatment and handling were done according to the standard ethical guideline procedures (Muhammad et al., 2011).

Acute toxicity studies (Determination of LD<sub>50</sub>): A 1cm<sup>3</sup> extract of the sample (3000mg/kg body weight) was administered to 5 groups of one rat each (one after the other at a grace observation period of 24 hrs) in a single oral dose using a feeding needle. Another (control group) received distilled water. Observation for toxic symptoms was made and recorded systematically at 1, 2, 4 and 6 hrs after administration. Finally, the number of survivors was noted after 48hrs for each animal (Muhammad et al., 2011).

Subchronic toxicity: As shown in Muhammad et al., (2011), a total of thirty albino rats were divided into five groups of six each. The animals in groups 2, 3, 4 and 5 were orally administered with (1cm<sup>3</sup> of 1000, 2000, 3000 and 4000mg/kg body weight) of the extract once daily for 28 days respectively. Animals in group 1 served as the control group (i. e. 0.00mg/kg) and received only drinking water by the same route. The body weights of all
the animals before and within 28 days (weekly) of treatment were recorded. The animals were sacrificed 24 hours after the last treatment after which blood samples were collected, allowed to clot and then centrifuged at 3000rpm for 10 minutes to obtain sera. The biochemical parameters of urea, uric acid, and creatinine were determined (Muhammad et al., 2011).

**Phytochemicals determination**

Phytochemicals in *A. sativum* were determined using standard procedures mentioned in Mana et al., (2022).

**RESULTS AND DISCUSSION**

The results for this work was shown in tables 1-3.

**Table 1.** Determination of phytochemicals in *A. sativum* from Sokoto, Nigeria

<table>
<thead>
<tr>
<th>Phytochemicals</th>
<th>Observation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flavonoids</td>
<td>Present</td>
</tr>
<tr>
<td></td>
<td>Present</td>
</tr>
<tr>
<td></td>
<td>Present</td>
</tr>
<tr>
<td>Saponins</td>
<td>Present</td>
</tr>
<tr>
<td>Tannins</td>
<td>Present</td>
</tr>
<tr>
<td>Steroids</td>
<td>Present</td>
</tr>
</tbody>
</table>

**Table 2.** Showing the results of LD$_{50}$ (acute toxicity) due administration of *A. sativum* in rats

<table>
<thead>
<tr>
<th>Group</th>
<th>Body weight</th>
<th>Temperature</th>
<th>Food intake</th>
<th>Breathing</th>
<th>Coma</th>
<th>Drowsiness</th>
<th>Death</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Absent</td>
<td>Absent</td>
<td>Absent</td>
</tr>
<tr>
<td>Group 2</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Absent</td>
<td>Absent</td>
<td>Absent</td>
</tr>
<tr>
<td>Group 3</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Absent</td>
<td>Absent</td>
<td>Absent</td>
</tr>
<tr>
<td>Group 4</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Absent</td>
<td>Absent</td>
<td>Absent</td>
</tr>
<tr>
<td>Group 5</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Absent</td>
<td>Absent</td>
<td>Absent</td>
</tr>
</tbody>
</table>
The use of plants by humans has been recorded from the time immemorial. Likewise, plant-based materials are out there to sustain life on earth due to their valuable chemical constituents. In rural and poor settings, where provisions of healthcare are low, health education is poor, and preventive measures are poor, diseases such as urinary Schistosomiasis are rampant and communicable (Sarkingobir et al., 2023ab). The situation forced the public to seek for alternatives. Hence, the use of *A. sativum* to treat urinary Schistosomiasis. Similarly, modern medicine and cities are now turning proper attention to the use of alternative medicines especially with a view to affordable and biofriendly healthcare to the door steps of all, to combat the spread of antimicrobial resistance and reduce healthcare cost (Noorshilawati et al., 2020; Sarkingobir et al., 2022). On the other hand, the scientists shall work to expunge the empirical evidence for the use of plant-based materials to treat or prevent diseases. Thus, phytochemicals in *A. sativum* were determined as shown in Table 1. These kinds of metabolites are exhibited by the plant *A. sativum* as shown in some other studies (Suleiman et al., 2018). The presence of metabolites in *A. sativum* was also found in the past studies such as Mikhail (2010); Lawal & Matazu (2015); Lawal (2016); and Suleiman et al., (2018b).

Verily, urinary Schistosomiasis, a popular disease otherwise known as bilharzia is a thing of great concern in the African countries, more especially in the areas of Sokoto State living nearby water sources such as ponds, rivers and lakes (Rinaldo et al., 2021; Suleiman et al., 2022). The disease is endemic in the region and results in abnormalities of great public health concern (Akinneye et al., 2018; Anyolitho et al., 2019). Due to factors such as poverty, poor environmental sanitation, poor healthcare provision, and lack of prevention interventions; the disease is increasingly transmitted through the critical role of the snail.
(which act as intermediate host) (Nelwan, 2019; Umoh et al., 2020). However, because most of the victims are from poor setting, and due to current rise in alternative medicine; the public resorts to the use of alternative traditional medicinal plants such as *A. sativum* for the treatment and prevention of bilharzia in Sokoto State, Nigeria (Jibia & Sani, 2019; Onyeyemi et al., 2020; Harmburg et al., 2021). Likewise, substantiated few information obtained through scientific works have demonstrated the effectiveness of using the *A. sativum* to tackle the snail host of bilharzia. This action is considered a helping hand in the prevention of the disease (Suleiman et al., 2018ab). Meanwhile, it is now imperative to provide a baseline information of phytochemicals, acute toxicity, and subchronic toxicity of *A. sativum* to safeguard public health in the course of using *A. sativum* as an alternative preventive measure of bilharzia in Sokoto State, Nigeria.

Meanwhile, the acute toxicity study of the *A. sativum* methanolic extract in Table 2, that is an initial step to evaluate hazard or toxicity due to plants extract revealed that abnormal changes regarding the general behavior of experimental animals was not observed. There is no recorded mortality; thus, the LD$_{50}$ of the plant might be elevated above 4000 mg/kg concentration of the extract.

Forsooth, in the course of studying toxicity, the activity of kidney is studied because kidney is a vital metabolic organ that when disturbed a great consequence occurs. Kidney is responsible for the elimination of urea, uric acid, and creatinine among others. These compounds when left uncleared turn to hazard in the body (Lawal et al., 2016; Dikko et al., 2020). Therefore, an effect of *A. sativum* on kidney was studied and result in Table 3 revealed that no major change was found between control and treated animals. Thus, this result likely indicates that, the normal function of the kidney about these metabolites (urea, creatinine, and uric acid) are not disturbed by the *A. sativum* (Mikhail, 2010; Lawal et al., 2016). Meanwhile, it is recommended that further works shall be dine revalidate the use of *A. sativum* in prevention of schistosomiasis to curtail public health implications.

**CONCLUSION**

*A. sativum* is a new prevention against Schistosomiasis in Sokoto State, Nigeria; thus, it is pertinent to evaluate the plant scientifically. This study evaluates the phytochemical content, LD$_{50}$, and subchronic toxicity due to the *A. sativum* methanolic extract. It was observed that, the plant has no acute toxicity, and contains phytochemicals that are behind
it’s roles. Likewise, there was little changes in to the creatinine, urea, and uric acid of the kidney due to the *A. sativum* administration in rats, which is an indication of little effect on the kidney. More works are needed to clear the plant for complete safety pertaining utilization in the prevention of schistosomiasis.

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