# Preliminary Investigation on Anti Typhoid Properties of Acacia Nilotica Leaf Extract

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## Abstract

Anti typhoid properties of methanol leaves extract of Acacia nilotica was investigated. The phytochemical screenings for detection of the presence of bioactive constituents were carried out. Among other parameters investigated include LD50 of the leaves extract, inoculation of mice with Salmonella typhi parasites treating them with leave extract of Acacia nilotica followed by widal tests. The phytochemical screening of the methanol leave extract of Acacia nilotica revealed the presence of alkaloid, anthraquinone, carbohydrates, cardiac glycosides, flavonoid, saponin, tannins and terpens. The leaves extract of Acacia nilotica has possess some acute toxicity effect on animals (mice) at a dose of (LD50) 288.5mg/kg. The plant extract produced inhibitory activities against Salmonella typhi. It is interesting to note the widal test titer valve was found to be 10 for the treated group while the untreated group had titer value of 160, suggesting that the plant leaves extract was effective as anti typhoid agent against salmonella typhi on mice infected with typhoid parasites. The extract demonstrated high activity against Salmonella typhi, bacterial typhoid causing agent.

***Index terms***— typhoid fever, anti typhoid, acacia nilotica, salmonella typhi and ld.

## 1 Introduction

he emergence and spread of Salmonella typhi resistance to many commonly used antibiotics (Ampicillin, Chloromphenicol, Amoxicilllin is now a subject of international concern. The problem has become endemic in many developing countries, causing enormous childhood morbidity and high cost of treatment (Leume, 1999). Multidrug resistant Salmonella species are being increasingly reported from the developed world. There is therefore,the need for efficient and safe vaccine which can be used as a preventive public health tool (Leume, 1999).Thus the resistance of Salmanella typhi to these antibiotics couples with the high cost of treatment have prompted the present study of local herbs for

The treatment of typhoid fever. Acacia leaf is a plant used in Northern part of Nigeria and many have claimed to have gotten remarkable improvement in their condition after taking the preparation from the herbs **??**Aussie, 2006). The medicinal plant is used in various ways but commonly is boiled in water and then allowed Author ?: Department of Biochemistry, Modibbo Adama University Of Technology Yola Adamawa State Nigeria. Author ?: Department of Applied Science, Ka, duna Polytechnic, Kaduna Nigeria. e-mail: sarkiyayi_shehu@yahoo.com to cool before drinking. The patient may inhale the steam after which he or she drinks the herbal preparations.

Typhoid fever is an infectious disease cause by bacterial of salmonella group-Salmonella typhi and Salmonella para typhi A, B or C. The organism is Gram negative, flagellated, non encapsulated, non sporulating and facultative anaerobic bacillus. The strain differs from other salmonellas in that it does not produce gas from glucose and from little or no water. It has only one phase antigen and posses a capsular antigen called vi **??**Cook, 1988). Salmonella typhi causes typhoid (enteric) fever, the bacterial pass from the small intestine into the blood

through Lymphatic system. The reticulumendothelial system becomes infected as well as the gallbladder and kidneys. From the gallbladder, the organism invades the intestine causing inflammation and ulceration **??**Cook, 1988).

Typhoid fever is characterized clinically by continual high fever of 40 0 c and headache and the incubation period is normally two weeks, in the second or third week the organism becomes disseminated in the body and can be isolated from urine and feces. According to Cook (1988), Ivanoff et al, (1997) symptoms of infection includes fever with low pulse rate, headache, enlargement of the spleen and mental confusion. A rash (rose spots) may be seen on light colored skin. Epitasis intestinal hemorrhage and perforation may also occur. In uncompleted (Asymptomatic) typhoid, the total white cell count is low with a relative Lymphocytosis and there may also be anemia. The condition is an immune complex disorder of the kidney and is characterized by fever, edema and marked albuminuria. It also causes osteomyelltis (Inflammation of the bone marrow), especially in children with sickle cell disease and thalassaremia, typhoid nodules can be found in the bone marrow. Inflammation of the joints (Typhoid Arthritis) may also occur. Salmonella para typhi A and B causes paratyphoid (Enteric Fever) and the diseases are generally mild with Salmonella paratyphi A and B being less invasive than Salmonella typhi. These are usually characterized with diarrhea and vomiting and enteric intestinal treat may be inflamed especially in Salmonella paratyphi B infection.

In tropical and other developing countries paratyphoid is more commonly used by Salmonella paratyphi A than Salmonella paratyphi B (WHO). Before the early 20 th century, typhoid fever was a common disease that occurred in large epidemics everywhere. In countries where modern method of sanitation and sewage disposal is only rarely encountered but in parts of the world lacking good sanitary facilities, it continuous to represent a serious health problem.

There are two main diagnostic laboratory test for typhoid. These are the specimen cultured in which the organism is isolated from blood, bone marrow, urine or stool of the patient and the serological method, which is based on agglutination test with antisera from "O" and "H" antigen. The two main serological test are rapid slide (widal) test. The serological method were used to give fast results (Within an hour) in contrast to the culture method, which takes weeks . The widal test is based on the fact that the serum agglutination rise sharply during the second and third week of salmonella infection. High or rising titre of "O"antigen (1:160) suggest active infection. However, the result of serological test for salmonella infection must be interpreted cautiously because cross reaction antibodies can give a false positive result. Typhoid fever is now an endemic disease in our community.

Many medicinal plants are used to treat many health problems including internal and external forms depending on the region where people inhabit. Proven medical plant are used in the treatment of diseases either alone or in combination with other plants, they are used as anti-infections agent, anti-malaria, antihormonal and nerves remedy. Several laboratories have reported the effectiveness of common indigenous herbs against gram-negative and gram-positive microorganism. In clinical research, the extract of Acacia has been used in the treatment of inflammation in the respiratory ailment. It is also helpful for cough, sore throat, eye wash, diarrhea and dysentery.

This study, therefore intends to scientifically assess the effectiveness of Acacia leaf in the treatment of typhoid fever using experimental animals and also to substantiate the claims by the traditional medicine practitioners that Acacia leave extract is used for the treatment of patient caught with typhoid fever.

# 2 II.

# 3 Materials And Methods

# 4 a) Materials

Fresh leaves of Acacia nilotica were collected from Kaduna Polytechnic Main Campus Tudun Wada Kaduna. Experimental animal were obtained from animal house in the department of pharmaceutical science, Ahmadu Bello University Zaria. They were kept in clean cage and fed on chow diets and water for 2 weeks in order to be acclimatizing to room temperature before being exposed to the plant extract.

# 5 b) Preparation of Extract

The sample collected was air dried at room temperature, the dried leaves were then pounded to powder form using mortar and pestle. It was then properly store until required. Methanol extraction was carried out in a soxhlet apparatus .Preliminary phytochemical screening Extract obtained were screened for their phytochemical constituents using standard quantitative procedures (Harbane, 1973, Trease and Evans 1989, and Safowora, 1993). Alkaloid, anthraquinones, saponins, flavanoids, tannins, steroids ,cardiac glycosides, carbohydrate (reducing sugar), and terpenses

# 6 c) Acute toxicity test

Pilot study was carried out using Lorke's (1983) method to determine the LD 50 Value of the methanol extract of Acacia leaves. Pilot study was carried out using Lork's method to determine the LD 50 value of the methanol

extract of Acacia leaves. The pilot study was carried to determine the maximum dose that could not produce death and the minimum dose that could be lethal, the range of toxicity orally Lorkes method of determination of LD 50 is carried out in two stages (Lorke, 1983).

## 7 d) First stage of Lorke'test

Three groups I, II and III of albino-mice for each of the extract used weighed separately and placed cages. The groups were given 10mg/kg, 100mg/kg and 1000mg/kg of doses of the extract respectively. The mice were observed for 24hours and all symptoms of intoxication and number of dead mice were recorded.

## 8 e) Second stage of Lorke's test

Based on the result obtained from the first stage of pilot studies, dose were chosen from the extracts and further experiment were carried out as detailed below Four groups I, II, III and IV of one mice each was weighed and placed in separate cage, the mice were given the extracts at doses of 140mg/kg, 225m/kg, 370mg/kg and 600mg/kg respectively. For the methanol extracts they were then observed closely for 24hours for signs and symptoms of intoxication and death (Lorke 1983).

## 9 First stage of

## 10 f) Parasite inoculation

Salmonella typhi obtained from 44 Army Hospital was maintained by serum passing in albino mice. Nine mice were distributed into 3 groups with three mice in each group. These were kept in separate apartments. The mice in each group I, II and III were injected intravenously with typhoid fever bacterial causative organism (Salmonella typhi). Group I and II were however placed on oral treatment with 125mg/kg and 150mg/kg of the extract for 3days, while group III was served as control. After which a widal test was carried out on them to confirm if they were infected with typhoid fever. g) Widal test 2 drops of serum to be tested is place on a while tile, the antigen suspension was shaken and 1 drop of the antigen was added. It was then mix over an area of 3cm, rock gently and examine for agglutination after 1 minute

## 11 h) Determination of Minimum Inhibitory Concentration

The minimum inhibitory concentration (MIC) of the methanol Extract was determined by method described by Akinpelu and Kolawole (2004).

## 12 III.

## 13 Results And Discussion

The result of anti-typhoid properties of Acacia is presented as follows. **??**)

## 14 Acute toxicity test

The LD 50 of the plant extract was 288.5mg/kg as calculated using Lorke's method.

## 15 b) Widal Test

Significant titer value for widal test range from 1/160 and above while non-significant valve range from 1/40 and below. The widal test titer valve was found to be 1\40 which is not significant for the treated groups Suggesting that the leaves extract has some anti typhoid properties. Our findings revealed that methanol extract of Acacia n. plant material has some antimicrobial activity. The minimum inhibitory concentration of the plant was at 2mg/ml suggesting that the leaves methanol extract of Acacia n. possess some anti typhoid properties.

## 16 IV.

## 17 Discussion

According to the centre for disease control, approximately 5% of people who effectedwith typhoid continues to carry the disease after they recover. The world Health Organization (WHO) identified typhoid as a serious public health problem. Its incidence is highest in children and young between 5 and 19 years old **??**WHO ,2008). According Ivanoff et al (1997) , symptoms of infection include fever with low pulse rate, headache, enlargement of the spleen and mental confusion.

From the analysis carried out, it was observed that the methanol extract of Acacia nilotica have some phytochemicals like Alkaloids, Anthraquine, Glycoside, Flavanoids and Tannins . The presence of these compounds promises it potential application in the treatment of microbial ailment. While the acute toxicity test showed that the LD 50 of Acacia nilotica was 288.5mg/kg. Also the group of the experimental animal (mice) treated with the methanol extract of Acacia nilotica after infection with Salmonella typhi, showed total clearance and 100% protection from Salmonella typhi. A similar studies was conducted by Vivek et al (2010)

which showed that aqueous extract of fruits of Citrus sinensis confer anti typhoid activity against Salmonella typhi . The results in table 2 revealed that the mice in group I, and II that were placed on oral treatment with 125mg/kg and 150mg/kg of the extract respectively had recovered, suggesting that Acacia nilotica has some anti typhoid activity.While the mice in group 3 had died. Similarly, Sarkiyayi et al., (2011) reported that aqueous methanol leaves extract of Albazia ferruginea plant was effective as anti typhoid agent against Salmonella typhi on mice infected with typhoid parasites. Furthermore, our findings revealed that methanol extract of Acacia n. plant material has some antimicrobial activity with a minimum inhibitory concentration of 2mg/ml, suggesting that the leaves methanol extract of Acacia n. Possess some anti typhoid properties. In a related development, the ethanol extract of ginger at 0.8g/ml concentration produced higher inhibition zone diameter than the garlic extract. The inhibitory property of ginger against S. typhi, E. coli and B. subtilis has been demonstrated by Azu and Onyeagba (2007). Ayogu and Amadi (2009) reported that raw Allium sativum and ethanolic extracts of Allium sativum and Zingiber officinale have inhibitory activity against the test organism.

V.

# 18 Conclusion

From the analysis carried out, it was observed that the methanol extract of Acacia nilotica showed antityphoid activities against the species of organism (Salmonella typhi) used for this study. As such, the antityphoid activities of the extract promises it potential application in the treatment of typhoid fever.

# 19 VI.

# 20 Recommendation

Since the presence of some phytochemical compounds (such as alkaloids, tannins, saponin, flavanoid and so on) were detected, further work is recommended in order to identify the bioactive components of the plant extract for the purpose of drug development. [1] [2] [3] [4]


Figure 1:

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[4] ( )

a) Phytochemical composition of the extract

The phytochemical screening of the methanol extract revealed the presence of Anthraquinone, Terpenoids, Saqorin, Tannin, Alkaloid, Flavanoid and Cardiac glycoside (Table

| | Minimum dose that Does not produce death | Maximum dose that produce death D D D D ) F ( |
|---|---|---|
| Phytochemicals | | Methanol extract |
| Alkaloids | | + |
| Anthraquinones (Free state) | | + |
| Anthraquinonea (Combined States) | | + |
| Cardiac Glycooide | | + |
| Flavanoids | | + |
| Saponins | | + |
| Steroids | | - |
| Tannins | | + |
| Terpenoids | | + |

Key: + = Present, –= Absent

Figure 2:

**2**

Three days inhibitory effect of plant extract on Salmonella typhi growth

| | Day I | Day II | Day III |
|---|---|---|---|
| Group I (125mg/kg of Acacia leaf) | H= | | |

Figure 3: Table 2 :

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