



Phytochemical screening and *in vitro* antifungal properties of *Fagara zanthoxyloides*

A. Banso¹ and Joshua E. Ngbede^{2*}

¹ Department of Science Laboratory Technology, The Federal Polytechnic, P.M.B. 55, Bida, Niger State, Nigeria. ² Bacteriology Division, P.O. Box 174, National Veterinary Research Institute, Vom, Jos, Plateau State, Nigeria. *e-mail: joshuangbede@yahoo.com

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Abstract

Fagara zanthoxyloides was screened for biologically active ingredients and its antifungal activities. The results revealed that the plant extract possessed the following active ingredients: alkaloids, saponins, tannins and glycosides. The extract exerted inhibitory effect on *Microsporum canis*, *Trichophyton rubrum* and *Trichophyton mentagrophytes*. The minimum inhibitory concentration of the extract ranged between 20 and 70% (w/v), while the minimum fungicidal concentration ranged between 30 and 80% (w/v). The results obtained suggest that this medicinal plant could be useful as an anti-fungal agent.

Key words: *Fagara zanthoxyloides*, phytochemical analysis, antifungal activities.

Introduction

Green plants possess the broadest spectrum of synthetic activity and have been the source of many useful compounds such as morphine, atropine, ergometrine, ergotamine and caffeine to mention but few¹⁵. Farnsworth and Bingel⁹ reported that about 80% of modern medicines are obtained from 15% of about 250,000 species of higher plants growing on earth. Also in 1982, Nwaiwu¹¹ reported that plants possess other natural compounds in addition to synthetic chemicals, and it has been proved that plant kingdom offers useful medicinal compounds^{6, 8, 14, 17}. In many tropical countries, especially in Africa, different plant parts are prepared and used for the treatment of illness as herbal medicine. Plant parts such as fruits, stems, leaves, seeds and roots are commonly employed for this purpose.

The preparation of the plant parts may be in powdered form, freshly boiled extract or as food condiment. Akpan and Akinremin² have showed that aqueous extract of *Fagara zanthoxyloides* exhibits antibacterial activity against *Streptococcus pyogenes*. The aim of this study was to screen *Fagara zanthoxyloides* for its biological active ingredients and its antifungal activities.

Materials and Methods

Plant materials and microorganisms: *Fagara zanthoxyloides* used in this study was obtained from Bida, Niger State. Identification was carried out at the Herbarium Unit of the Department of Biological Sciences, University of Ilorin, Nigeria. The representative microorganisms used were *Microsporum canis*, *Trichophyton rubrum* and *Trichophyton mentagrophytes*. The organisms were obtained from the Culture Collection Centre of the Department of Biological Sciences, University of Ilorin, Nigeria.

Preparation of plant extract: Ethanolic extract of the plant material was prepared according to standard methods^{4, 5, 16}. Five g of the plant material was air-dried, crushed and blended into powder using an electric blender (National MX 491 IV, Matsushita electric). The blended material was transferred into a beaker, and 10 ml of ethanol was added at room temperature. The mixture was extracted by agitation on a rotary shaker. The extract obtained was vacuum-dried, stored and later used for the tests.

Phytochemical screening: Root extract of *Fagara zanthoxyloides* was analyzed for the presence of alkaloids, saponins, tannins and glycosides according to standard methods^{12, 14, 15} for the presence of active ingredients in the extract.

Screening for alkaloids: From the previously dried plant material, 3 g was used for extraction with ethanol containing 3% tartaric acid. The filtrate was divided into three portions in beakers and tested for alkaloid as follows. To the first portion of the filtrate Mayer's reagent was added. To the second portion of the filtrate Haga's reagent was added and to the third portion Marqui's reagent was added. A precipitate in any of the three tests was regarded as positive test for alkaloid¹².

Screening for saponins: Five drops of olive oil was added to 30 ml of the ethanolic extract of the test plant in a test tube, and the mixture was vigorously shaken. Formation of soluble emulsion in the extract indicated the presence of saponin¹².

Screening for tannins: Into 10 ml of freshly prepared 10% potassium hydroxide (KOH) in a beaker, 10 ml of ethanolic extract of *Fagara zanthoxyloides* was added. A dirty precipitate observed in the extract indicated the presence of tannins^{12, 17}.

Screening for glycosides: One g of coarsely powdered *Fagara zanthoxyloides* plant leaves was added into two different beakers. To one of the beakers 5 ml of dilute sulphuric acid (5%) was added, and 5 ml of water was added to the other beaker. The two beakers were heated for 3-5 minutes and the contents filtered into labeled test tubes. The filtrate was made alkaline with 5% sodium hydroxide and heated with Fehlings solution for 3 minutes. The presence of reddish brown precipitate in the acid filtrate and absence of such a precipitate in the aqueous filtration was regarded as positive for glycosides^{12, 17}.

Antifungal test: The antifungal test was performed using the pour plate method⁷. Neat concentration of the plant extract was introduced into each labelled molten Sabroaud dextrose agar in bottles, mixed and poured into sterile Petri dishes evenly. The plates were then allowed to set on a flat surface, and then dried at 37°C incubator. The plates were inoculated using a sterile cork borer to cut the advancing edge of the test organisms and placed on the centre of the agar using sterile forceps. Growth diameters were measured after 18 days of incubation at room temperature (28±2°C).

Determination of minimum inhibitory concentration (MIC): Different quantities of *Fagara zanthoxyloides* fine powder were weighed and added to malt extract in the test tubes to make concentrations of 10, 20, 30, 40 and 60% (w/v). The contents were mixed thoroughly. Each tube was inoculated with 0.1 ml of spore suspension of *Microsporum canis*, *Trichophyton rubrum* and *Trichophyton mentagrophytes* to contain not more than 1x10⁶ cfu/ml³. The tubes were incubated at room temperature (28±2°C) and then examined for growth after 18 days. The least concentration of the plant extract that does not permit any visible growth of the inoculated test organism was regarded as the MIC in each case. Control experiments were performed without the plant extracts^{10, 13}.

Results and Discussion

The study showed that the extracts of *Fagara zanthoxyloides* possess alkaloids, saponin, tannin and glycosides. However, saponin was detected in trace amount (Table 1). Several plants that are rich in alkaloids, tannins and glycosides are known to possess antimicrobial activity against a number of microorganisms¹. The results on *M. canis*, *T. rubrum* and *T. mentagrophytes* to ethanolic extract of *F. zanthoxyloides* revealed that the extract exhibited inhibitory activity on the selected fungi. It was also noted that the higher the concentration of the extract, the more inhibitory effects were obtained which supports the works of other investigators on similar studies^{4, 10}.

The results of the minimum inhibitory concentration (MIC) of the extract ranged between 20 and 40% (w/v) (Table 2). The tested micro-organisms are disease causing fungi, therefore, we suggest that extract of *F. zanthoxyloides* could be useful as an antifungal agent. However, we recommend quantitative analysis of each component of this plant and their activities on each fungi tested. We recommend further work on preservation of each active ingredient for proper storage for its use as antifungal agent.

Table 1. Phytochemical screening of *F. zanthoxyloides* extract.

Chemical constituent	Results
Alkaloids	+
Saponins	±
Tannins	++
Glycosides	+

Key: + Positive, ± Trace, ++ Strongly positive.

Table 2. Minimum inhibition concentration of *Fagara zanthoxyloides* extracts (%w/v).

Microorganism	Extract concentration (% w/v)					
	10	20	30	40	50	60
<i>Microsporum canis</i>	-	-	-	+	+	+
<i>Trichophyton rubrum</i>	-	-	+	+	+	+
<i>Trichophyton mentagrophytes</i>	-	-	-	-	+	+

Key: - No growth (clear), + growth (turbid).

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