

The Bioactivity of *Parinari curatellifolia*

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Abstract: The methanol extract of the leaves, stem and root of *parinari curatellifolia* Benth, showed moderate cytotoxicity to brine shrimp with LC_{50} values of $1412 \mu\text{g mL}^{-1}$, $414 \mu\text{g mL}^{-1}$ and $724 \mu\text{g mL}^{-1}$ respectively, indicating the presence of bioactive compounds in the extract of this plant. Screening of the stem and root bark extracts for antimicrobial activity against *staphylococcus aureus*, *streptococcus pyogenes*, *corynebacterium ulceran*, *Escherichia coli*, *Salmonella Typhi*, *candida albicans* and *Aspergillus niger* revealed the extracts to inhibit all pathogens examined except *Aspergillus niger*. This result implies that these extracts can be used to treat wounds, boils, stomach trouble, diarrhea, food poisoning and fevers. These results further show that the principles of *parinari curatellifolia* Benth are excellent candidates for additional *in vitro* pharmacological and efficacy studies.

Key words: *Parinari curatellifolia*, cytotoxicity, antimicrobial activity, *in vitro* pharmacological effect

INTRODUCTION

Infectious diseases account for approximately one-half of deaths in tropical Africa and is the third leading cause of death in U.S.A.^[1,2]. Microorganisms which cause many of these disease conditions are yet to be properly checked despite the advancement in medicine. Control of diseases is primarily via chemotherapeutic interventions and many of the drugs available are generally unaffordable for most people due to poverty.

There is great scope for new drug discovery based on medicinal plants^[3-5]. Indigenous people of Africa have profited from plant extracts and have used a wide range of plants to sustain their health. These medicinal plants used in various traditional systems need to be subjected to phytochemical analysis with the aim of isolating the bioactive compounds.

The search for bioactive compounds, which can inhibit the growth of pathogens at low concentration, with fewer side effects at low cost from the flora of Nigeria has become paramount. This search requires identification of new biochemical targets for drug development. The objective of the present study was to determine the bioactivity of the methanol extract of the stem and root barks of *parinari curatellifolia* Benth using the brine shrimp lethality bioassay and antimicrobial screening. Since the interest in bioactive compounds depends upon lethal concentrations and the spectrum of activity, this will be determined in our investigations.

MATERIALS AND METHODS

General: *Artemia salina* Leach (Aquarium system U. S. A) was used for brine shrimp lethality bioassay while *candida albicans* ATCC10231, *Escherichia coli* NCTC10418; *Salmonella typhi* NCTC5231, *staphylococcus aureus* NCTC 6571, *Aspergillus niger* Ls, *corynebacterium uiceran* Ls and *streptococcus pyogenes* Ls were used for antimicrobial test using Nutrient agar as medium.

Plant material: The plant material was collected from Kaduna State of Nigeria in January 2004 and properly identified at the herbarium of the Biological Sciences Department of Ahmadu Bello University Zaria where a voucher specimen has been deposited. Voucher No 903.

Extraction: The powdered plant material (1000 g) of leaves, stem and root were separately packed into the thimble of a Soxhlet extractor and extracted with concentrated *in vacuo* and the residue washed with petroleum ether (60-80), chloroform, Ethyl acetate and methanol. The organic extracts were dried and evaporated *in vacuo* to give the following residue: petroleum ether (60-80) (0.80 g), chloroform (3.24 g), Ethyl acetate (0.42 g) and methanol (39.56 g).

Brine shrimp lethality bioassay: This was conducted according to standard protocols^[6,7].

Table 1: Result of sensitivity test of *parinari curatellifolia* ex benth

Test of organism	Extract	
	Stem	Root
<i>Staphylococcus aureus</i>	S	S
<i>Streptococcus pyogenes</i>	S	S
<i>Corynebacterium ulceran</i>	S	S
<i>Escherichia coli</i>	S	S
<i>Salmonella typhi</i>	S	S
<i>Candida albicans</i>	S	S
<i>Aspergillus niger</i>	R	R

Table 2: Result of M. I. C test for *parinari curatellifolia* ex benth

Test organism	Zone of inhibition (MM) at different concentration ($\mu\text{g mL}^{-1}$) OD extract							
	Stem				Root			
	1×10^4	2×10^4	3×10^4	4×10^4	1×10^4	2×10^4	3×10^4	4×10^4
<i>Staphylococcus aureus</i>	0	8	14	20	0	0	4	16
<i>Streptococcus pyogenes</i>	0	4	10	18	0	0	6	17
<i>Corynebacterium ulceran</i>	0	10	16	24	0	4	10	18
<i>Escherichia coli</i>	0	10	18	22	0	0	6	12
<i>Salmonella typhi</i>	0	12	19	27	0	4	10	18
<i>Candida albicans</i>	0	0	0	12	0	0	8	16
<i>Aspergillus niger</i>	0	0	0	0	0	0	0	0

Antimicrobial screening: The paper disc. Diffusion method was used^[8,9] to determine the anti-bacterial activity of the extracts. Dilution susceptibility testing method was used to determine the minimum lethal concentrations.

Solutions of 0.5 g mL^{-1} , 0.6 g mL^{-1} , 0.7 g mL^{-1} and 0.9 g mL^{-1} concentration of plant extracts using the pure extracting solvent in each case were prepared.

Petri dishes were washed and sterilized in an autoclave at about 120°C for 25 minute and allowed to equilibrate to $48\text{-}50^\circ\text{C}$ before use. They were labeled to indicate the concentration, test organism and type of extract.

Nutrient agar (28 g) was dissolved in a Litre of distilled water a 2L conical flask capped with a cotton wool plug. This was sterilized at 120°C for 15 minutes and allowed to cool. The sterilized medium (20 mL) was poured into the sterilized petri dishes, covered and allowed to solidify. The plates were seeded with test microorganisms by the spread plate technique. This was allowed to dry for 30 min. Filter paper was cut, sterilized, soaked in the solution of the extract and allowed to dry. The dried discs were then planted on the nutrient agar seeded with the test microorganism. The plates were incubated at 37°C for hrs, after which the zones of inhibition of growth were measured and recorded in millimeter of their diametrical section.

A control experiment was also set up using only the extracting solvent for each of the test organism.

RESULTS AND DISCUSSION

Table 1 show clearly that *parinari curatellifolia* Benth contains bioactive principles. The extracts of this plant were active against Brine shrimp at moderately low concentration. It has been demonstrated that higher plants with moderate toxicity to Brine shrimp may contain bioactive compounds^[10,11].

The selection of pathogens used for the antimicrobial screening was guided by folklore claims of therapeutic principles. The extracts suppressed the pathogens used for the screening except *Aspergillus niger* but was particularly highly active against *salmonella typhi*. The extract could be expected to be effective for fever treatment (Typhoid and scarlet) since it inhibited the growth of *salmonella typhi* and *streptococcus pyogenes*. Various wounds infections and skin problems are caused by *staphylococcus aureus*, *candida albicans* where as *streptococcus pyogenes* causes erysipela^[12]. The extracts were active against all these organisms with m.i.c value of $2 \times 10^4 \mu\text{g mL}^{-1}$ Table 2. This result lends credence to the traditional use of thus plant for wounds, malarial, typhoid fever, washing fractures and internal troubles. This result further supports the report that the plant has antiplasmodic activity^[13].

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