



ORIGINAL RESEARCH

Antimicrobial activities of *Dillenia indica* Linn. (Dilleniaceae) and *Spondias mombin* Linn. (Anacardiaceae) extracts on selected pathogenic organisms

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ABSTRACT

Background: The prevalence of increasing bacterial resistance against antibacterial agents in the past decade has spurred the search for substitutes. This necessitates the need to identify new compounds as alternatives to common synthetic antimicrobial compounds.

Objectives: To establish and evaluate the antimicrobial potentials of *Dillenia indica* and *Spondias mombin* against strains of some selected pathogens.

Method: Concentrations of the extract (methanol and dichloromethane) were prepared and the susceptibility of the test organisms to the antimicrobial effect of the plant extracts was determined using the agar well diffusion method. Standard antibiotics were used as positive control. Various culture media were used for this experiment and the minimum inhibitory concentration (MIC) was determined using the agar dilution method.

Results: Extracts of *D. indica* fruit, leaves and stem bark as well as *S. mombin* leaf and stem bark showed significant antimicrobial activity. Activity was dose dependent and the methanol extracts had better and wider activity than the dichloromethane extracts. *Spondias mombin* bark had the greatest activity against most of the test organisms while the positive control drugs had better activity than the extracts.

Conclusion: The leaves and stem bark of *Spondias mombin* and the leaves, fruit and stem bark of *Dillenia indica* have antimicrobial properties thus justifying their ethnopharmacological use in prevention and treatment of infections.

Keywords: Antimicrobial, *Dillenia indica*, *Spondias mombin*, *Helicobacter pylori*, plant extracts, pathogens

INTRODUCTION

New antibiotic resistance mechanisms are emerging and spreading globally, threatening our ability to treat common infections¹. Plant-derived antimicrobials have several advantages over synthetic antibiotics making the discovery of novel antimicrobials from plants expedient².

Some natural compounds may exhibit antimicrobial properties that differ mechanistically from other antimicrobials thus minimizing resistance and providing cross protection³. Traditionally, whole plant of *Dillenia indica* is used in cases of fever, as an aphrodisiac and also promotes virility. A study on fruit of *D. indica* confirms its anti-proliferation activity⁴.

Aqueous leaf extract of *Spondias mombin* showed significant antisecretory and gastric cytoprotective effects⁵ and tea made from the flowers and leaves is taken to relieve stomach ache, urethritis, cystitis and eye and throat inflammations⁶.

Due to the reported activities of *Dillenia indica* and *Spondias mombin* against species of Bacillus, Staphylococcus, Escherichia, Pseudomonas and Salmonella^{6, 7}, their antimicrobial properties would be evaluated in this study. The aims and objectives of this study were to evaluate the antimicrobial potentials of *Dillenia indica* and *Spondias mombin* against strains of *Helicobacter pylori* and some selected pathogens by authenticating plant samples, carrying out antimicrobial susceptibility testing of the extracts against selected microorganisms and determining the minimum inhibitory concentrations of active extracts.

MATERIALS AND METHODS

Plant collection and identification

Fresh leaves stem bark and fruits of *Dillenia indica*, together with leaves and stem bark of *Spondias mombin* were collected from the University of Ibadan, authenticated at the University's Herbarium and assigned voucher specimen number UIH- 22427 and UIH- 2228 respectively. The plants were air dried, pulverised, weighed and stored for the study. The fruits of *Dillenia indica* were both oven-dried and sun-dried due to high moisture content before air-drying.

Materials

Distilled methanol, Soxhlet extractor, distillation flask, electrically heated bath, reflux condenser, round bottom flask, Mettler balance, dichloromethane, retort stand, separating funnel, sterile distilled water, bottles, test tubes, test tube holder, electrically heated bath, Pasteur pipettes, Fehling's solution, 1% aqueous hydrochloric acid, 1% hydrochloric acid, Dragendorff's reagent, 5% ferric chloride, sulphuric acid, chloroform, dilute sodium

hydroxide, copper acetate solution, glacial acetic acid, benzene, 10% ammonia solution, distilled water.

Extraction procedure

The powdered plant was loaded into the Soxhlet extractor. The side arm was lagged with glass wool and the methanol was heated. The resulting extract was collected, evaporated to dryness, and the weight determined.

The crude extracts were defatted using distilled n-hexane. Each extract was reconstituted with 50 mL of methanol and 30 mL of sterile distilled water. 50 mL of n-hexane was poured into the individually reconstituted extracts in a separating funnel fixed on a retort stand. The content was shaken to ensure thorough partitioning into the two solvents. Cork of the funnel was removed at intervals to release the pressure while shaking. The methanol fraction was collected.

Each of the defatted extracts was poured into a separating funnel fixed on a retort stand. 50mL of dichloromethane was added and the contents shaken for thorough partitioning of the extracts. Cork was removed at intervals. The resulting fractions were collected, concentrated, dried and weighed.

Antimicrobial agents used as positive control

Rifampicin (30µg/ml and 20µg/ml), Ciprofloxacin (20µg/ml and 10µg/ml), Gentamicin (10µg/ml), Ketoconazole (20 µg/ml)

Culture media

Tryptic Soy Broth (OXOID), Mueller Hinton Agar (OXOID), Sensitivity Test Agar (OXOID). Each media was prepared according to the manufacturer's specification.

Antimicrobial screening of plant extracts

Mycobacterium fortuitum ATCC684, *M. smegmatis* ATCC 19420, *M. abscessus*, *M. phlei* ATCC 19240, *M. smegmatis*, *Helicobacter pylori* BA 26, *H. pylori* BA

36, *H. pylori* BA 38, *H. pylori* BA 42, *H. pylori* BA 504, *Staphylococcus aureus* ATCC 29213, *Pseudomonas aeruginosa* ATCC 27853, *Escherichia coli* ATCC 25922, *Klebsiella Pneumonia* ATCC 35675 and *Candida albicans* ATCC 90029 were used in this study.

Overnight bacteria culture was obtained by sub culturing from stored slopes. The stored bacteria slopes were streaked with a sterile inoculating loop and inoculated into 5ml Tryptic Soy broth in test tubes. These were then incubated for 18 hours at 37°C and 25°C for bacteria and fungi respectively. Strains of Mycobacteria and Helicobacter were incubated for 18 hours at 37°C in a microaerophilic environment (5% O₂, 10% CO₂ and 85% N).

Preparation of plant crude extracts

As extract stock solution, concentrations of 1mg/mL, 2mg/mL, 10mg/mL and 20mg/mL were prepared. Each extract (0.3g) was weighed and dissolved in 15 mL of reconstituting solvent mixture (5.7 mL of methanol and 9.3 mL of sterile distilled water). This was prepared to obtain the 20mg/mL concentration. Other concentrations were prepared by dissolving the appropriate amount of dried extract with reconstituting solvent mixture.

Susceptibility studies

This was determined using the agar diffusion method⁸.

Preparation of bacterial seeded agar plates

A 1 in 100 mL dilution of the overnight culture of each bacterium in appropriate broth medium was prepared by adding 0.1 mL of the overnight culture into 9.9ml of sterile distilled water. Using a sterile pipette, 0.2 mL of the 1 in 100 mL dilution of the overnight culture of the test organism was seeded into 20 mL of melted and cooled agar medium, mixed thoroughly, poured into sterile Petri-dishes and allowed to set. Equidistant wells were bored into the solidified agar using sterilized cork borer (8mm).

Preparation of fungal seeded plates

A 1 in 100 mL dilution in sterile distilled water of the overnight culture of the fungi, prepared in tryptic soy broth, was made. Then, 0.2 mL of the 1 in 100 mL dilution of the overnight culture of the test organism was added into 20 mL of melted and cooled agar medium, mixed thoroughly, poured into sterile Petri-dishes and allowed to set. Equidistant wells (8mm) were bored into the solidified agar.

Screening of the plant extract

Volumes (100µL) of individual extracts (1mg/mL, 2mg/mL, 10mg/mL and 20mg/mL) were introduced into the wells using sterilized Pasteur pipette. Ciprofloxacin (10µg/mL and 20µg/mL) was introduced into the well as positive control for Helicobacterium species, Rifampicin (20µg/mL and 40µg/mL) as positive control for Mycobacterium species and Gentamycin (10µg/mL) for the others. Ketoconazole (20 µg/mL) was used as positive control for fungus. Distilled methanol solution (40%) was used as negative control. The plates were left for about an hour to allow pre-diffusion of the extracts and control in the well through the agar after which they were incubated at 37°C for 24 hours (for bacteria) and 25°C for 48 hours (for fungus). After incubation, the zones of inhibition were measured and recorded. All procedures were performed in duplicates.

Determination of minimum inhibitory concentration (MIC)

The MIC was determined using the agar dilution method². Concentrations of the extract were prepared (20mg/mL to 1.25mg/mL). To 14 mL of the melted and cooled agar, 1 mL of the extract was added, shaken vigorously and poured into the petri dishes and allowed to set. The surface of the agar was dried at low temperature in the oven and streaked with the overnight culture of the susceptible organism. The plates were incubated at 37°C for 24 hours (for bacteria) and 25°C for 48 hours (for

fungus) and examined for growth. The lowest concentration at which there was absence of growth was taken as the minimum inhibitory concentration of the extract. All procedures were performed in duplicate.

RESULTS

Table 1 below shows that the percentage yield of methanol fraction was higher than the dichloromethane fraction for both *Dillenia indica* and *Spondias mombin* extracts.

The result obtained shows that that *Dillenia indica* leaf shows activity against *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli* (Table 2).

The result shows that *Dillenia indica* stem bark has no activity against Mycobacterium but activity exists against strains of other organisms (Table 3).

Table 4 also shows that *Dillenia indica* fruit has no activity against Helicobacteria and Mycobacterium strains but active against other test organisms.

Table 1: Yield and macroscopical characteristics of extracts of *Dillenia indica* and *Spondias mombin*

Plant	Plant part	Weight of ground sample (g)	Yield				Macroscopical Characteristics		
			MeOH	DCM	MeOH	Dichloro.	MeOH	DCM	
<i>Dillenia indica</i>	Leaf	300	23.0	2.0	7.6	0.67	Dark greyish congealed extract	Greyish green congealed extract	
	Stem bark	300	22.9	3.1	7.6	1.03	Shiny reddish brown and powdered extract	Dark brown congealed extract	
	Fruit	300	11	1.8	3.7	0.60	Dark brown oily extract	Dark brown congealed extract	
<i>Spondias mombin</i>	Leaf	300	2.3	1.0	0.7	0.33	Dark brown congealed extract	Greyish green congealed extract	
	Stem bark	300	5.7	0.9	1.9	0.3	Reddish brown congealed extract	Dark brown congealed extract	

Key: RM = Rifampicin; Cipro = Ciprofloxacin; Ket = Ketoconazole; Gent = Gentamycin; dichloro = Dichloromethane; ND = not determined; R = resistant; (-) = absent; MeOH = Methanol; DMSO = Distilled methanol solution; R = Resistance; ND = not determined; DCM – Dichloromethane

Table 5 shows that the extract of *Spondias mombin* leaf shows no activity against Mycobacterium but is active against strains of Helicobacteria, Staphylococcus, Escherichia and Candida The result further shows that the extract of *Spondias mombin* stem bark has activity against strains of Mycobacterium, Helicobacter,

Staphylococcus, Pseudomonas, Escherichia, Klebsiella and Candida (Table 6).

Table 7 shows the minimum inhibitory concentration (MIC) range of various extracts of the two agents tested. Values ranged from 1.25mg/ml to 20mg/ml for the extracts evaluated.

Table 2: Antimicrobial screening of the extracts of *Dillenia indica* leaf

Extract (mg/ml)	Methanol				Dichloromethane				RMP ($\mu\text{g}/\text{mL}$)		Cipro. ($\mu\text{g}/\text{mL}$)		Gent. ($\mu\text{g}/\text{mL}$)	Keto. ($\mu\text{g}/\text{mL}$)	DMSO
	1	2	10	20	1	2	10	20	20	40	10	20	10	20	40%
<i>M. fort.</i> ATCC 684	-	-	-	-	-	-	-	12 \pm 0.5	24 \pm 0.0	30 \pm 0.0	ND	ND	ND	ND	-
<i>M. smeg.</i> ATCC 19420	-	-	-	-	-	-	-	15 \pm 0.5	30 \pm 0.5	31 \pm 0.5	ND	ND	ND	ND	-
<i>M. abs.</i> <i>M. smeg.</i>	-	-	-	-	-	-	-	12 \pm 0.0	26 \pm 0.5	27 \pm 0.5	ND	ND	ND	ND	-
<i>M. phlei</i> ATCC 19240	-	-	-	-	-	-	-	18 \pm 2.5	22 \pm 0.5	22 \pm 0.5	ND	ND	ND	ND	-
<i>H. pylori</i> BA 26	-	-	-	-	-	-	-	-	-	-	14 \pm 0.0	19 \pm 1.5	ND	ND	-
<i>H. pylori</i> BA 36	-	-	-	-	-	-	-	-	-	-	R	23 \pm 1.0	ND	ND	-
<i>H. pylori</i> BA 38	-	-	-	-	-	-	-	-	-	-	15 \pm 0.0	21 \pm 0.0	ND	ND	-
<i>H. pylori</i> BA 42	-	-	-	-	-	-	-	-	-	-	R	23 \pm 0.5	ND	ND	-
<i>H. pylori</i> BA 504	-	-	-	-	-	-	-	-	-	-	R	20 \pm 1.5	ND	ND	-
<i>S. aureus</i> ATCC 29213	-	-	14 \pm 2.5	18 \pm 0.5	-	-	-	-	-	-	ND	ND	25 \pm 0.5	ND	-
<i>P. aeruginosa</i> ATCC 27853	-	-	10 \pm 0.0	17 \pm 0.5	-	-	-	-	-	-	ND	ND	23 \pm 0.0	ND	-
<i>E. coli</i> ATCC 25922	-	16 \pm 0.0	17 \pm 0.0	19 \pm 1.5	-	-	-	-	-	-	ND	ND	32 \pm 1.5	ND	-
<i>Kleb. Pneu</i> ATCC 35657	-	-	-	-	-	-	-	-	-	-	ND	ND	16 \pm 2.0	ND	-
<i>C. albicans</i> ATCC90029	-	-	-	-	-	-	-	-	-	-	ND	ND	ND	R	-

Key: RM = Rifampicin; Cipro = Ciprofloxacin; Ket. = Ketoconazole; Gent. = Gentamycin; dichloro = Dichloromethane; ND = not determined; R = resistant; (-) = absent; MeOH = Methanol; DMSO = Distilled methanol solution; R = Resistance; ND = not determined; DCM – Dichloromethane

Table 3: Antimicrobial screening of the extracts of *Dillenia indica* stem bark

Extract (mg/mL)	Methanol				Dichloromethane				RMP ($\mu\text{g}/\text{mL}$)		Cipro. ($\mu\text{g}/\text{mL}$)		Gent. ($\mu\text{g}/\text{mL}$)	Keto. ($\mu\text{g}/\text{mL}$)	DMSO
	1	2	10	20	1	2	10	20	20	40	10	20	10	20	40%
<i>M. fort.</i> ATCC 684	-	-	-	-	-	-	-	-	24 \pm 0.0	30 \pm 0.0	NDs	ND	ND	ND	-
<i>M. smeg.</i> ATCC 19420	-	-	-	-	-	-	-	-	30 \pm 0.5	31 \pm 0.5	ND	ND	ND	ND	-
<i>M. abs.</i> <i>M. smeg.</i>	-	-	-	-	-	-	-	-	26 \pm 0.5	27 \pm 0.5	ND	ND	ND	ND	-
<i>M. phlei</i> ATCC 19240	-	-	-	-	-	-	-	-	18 \pm 2.5	22 \pm 0.5	ND	ND	ND	ND	-
<i>H. pylori</i> BA 26	-	-	10 \pm 0.0	11 \pm 0.0	-	-	-	-	-	-	14 \pm 0.0	19 \pm 1.5	ND	ND	-
<i>H. pylori</i> BA 36	-	-	-	11 \pm 0.0	-	-	-	-	-	-	R	23 \pm 1.0	ND	ND	-
<i>H. pylori</i> BA 38	-	-	-	-	-	-	-	-	-	-	15 \pm 0.0	21 \pm 0.0	ND	ND	-
<i>H. pylori</i> BA 42	-	-	11 \pm 0.0	13 \pm 0.0	-	-	-	-	-	-	R	23 \pm 0.5	ND	ND	-
<i>H. pylori</i> BA 504	-	-	-	13 \pm 0.5	-	-	-	-	-	-	R	20 \pm 1.5	ND	ND	-
<i>S. aureus</i> ATCC 29213	-	-	-	11 \pm 1.0	-	-	-	-	-	-	ND	ND	25 \pm 0.5	ND	-
<i>P. aeruginosa</i> ATCC 27853	-	-	11 \pm 0.5	12 \pm 0.0	-	-	-	-	-	-	ND	ND	23 \pm 0.0	ND	-
<i>E. coli</i> ATCC 25922	-	-	11 \pm 0.0	16 \pm 0.5	-	-	-	12 \pm 0.0	-	-	ND	ND	32 \pm 1.5	ND	-
<i>Kleb. Pneumonia</i> ATCC 35657	-	-	-	-	-	-	-	-	-	-	ND	ND	16 \pm 2.0	ND	-
<i>C. albicans</i> ATCC 90029	-	-	16 \pm 1.0	20 \pm 0.0	-	-	-	-	-	-	ND	ND	ND	R	-

Table 4: Antimicrobial screening of the extracts of *Dillenia indica* fruit

Extract (mg/mL)	Methanol				Dichloromethane				RMP (µg/mL)		Cipro. (µg/mL)		Gent. (µg/mL)	Keto. (µg/mL)	DMSO
	1	2	10	20	1	2	10	20	20	40	10	20	10	20	40%
<i>M. fort.</i> ATCC 684	-	-	-	-	-	-	-	-	24±0.0	30±0.0	ND	ND	ND	ND	-
<i>M. smeg.</i> ATCC 19420	-	-	-	-	-	-	-	-	30±0.5	31±0.5	ND	ND	ND	ND	-
<i>M. abs.</i>	-	-	-	-	-	-	-	-	26±0.5	27±0.5	ND	ND	ND	ND	-
<i>M. smeg.</i>	-	-	-	-	-	-	-	-	25±0.5	26±0.0	ND	ND	ND	ND	-
<i>M. phlei.</i> ATCC 19240	-	-	-	-	-	-	-	-	18±2.5	22±0.5	ND	ND	ND	ND	-
<i>H. pylori</i> BA 26	-	-	-	-	-	-	-	-	-	-	14±0.0	19±1.5	ND	ND	-
<i>H. pylori</i> BA 36	-	-	-	-	-	-	-	-	-	-	R	23±1.0	ND	ND	-
<i>H. pylori</i> BA 38	-	-	-	-	-	-	-	-	-	-	15±0.0	21±0.0	ND	ND	-
<i>H. pylori</i> BA 42	-	-	-	-	-	-	-	-	-	-	R	23±0.5	ND	ND	-
<i>H. pylori</i> BA 504	-	-	-	-	-	-	-	-	-	-	R	20±1.5	ND	ND	-
<i>S. aureus</i> ATCC 29213	-	-	14±1.0	17±0.0	-	-	-	-	-	-	ND	ND	25±0.5	ND	-
<i>P. aeruginosa</i> ATCC 27853	-	-	10±0.0	16±2.0	-	-	-	-	-	-	ND	ND	23±0.0	ND	-
<i>E. coli</i> ATCC 25922	-	-	16±0.0	19±1.5	-	-	-	-	-	-	ND	ND	32±1.5	ND	-
<i>Kleb. Pneumonia</i> ATCC 35657	-	-	-	13±0.0	-	-	-	-	-	-	ND	ND	16±2.0	ND	-
<i>C. albicans</i> ATCC 90029	-	16±1.0	20±0.0	23±0.5	-	-	-	-	-	-	ND	ND	ND	R	-

Key: RM = Rifampicin; Cipro = Ciprofloxacin; Ket. = Ketoconazole; Gent. = Gentamycin; dichloro = Dichloromethane; ND = not determined; R = resistant; (-) = absent; MeOH = Methanol; DMSO = Distilled methanol solution; R = Resistance; ND = not determined; DCM – Dichloromethane

Table 5: Antimicrobial screening of the extracts of *Spondias mombin* leaf

Extract (mg/mL)	Methanol				Dichloromethane				RMP (µg/mL)		Cipro. (µg/mL)		Gent. (µg/mL)	Keto. (µg/mL)	DMSO
	1	2	10	20	1	2	10	20	20	40	10	20	10	20	40%
<i>M. fort.</i> ATCC 684	-	-	-	-	-	-	-	-	24±0.0	30±0.0	ND	ND	ND	ND	-
<i>M. smeg.</i> ATCC 19420	-	-	-	-	-	-	-	-	30±0.5	31±0.5	ND	ND	ND	ND	-
<i>M. abs.</i>	-	-	-	-	-	-	-	-	26±0.5	27±0.5	ND	ND	ND	ND	-
<i>M. smeg.</i>	-	-	-	-	-	-	-	-	25±0.5	26±0.0	ND	ND	ND	ND	-
<i>M. phlei.</i> ATCC 19240	-	-	-	-	-	-	-	-	18±2.5	22±0.5	ND	ND	ND	ND	-
<i>H. pylori</i> BA 26	-	-	-	-	-	-	-	10±0.0	-	-	14±0.0	19±1.5	ND	ND	-
<i>H. pylori</i> BA 36	-	-	-	13±0.5	-	-	-	-	-	-	R	23±1.0	ND	ND	-
<i>H. pylori</i> BA 38	-	-	-	-	-	-	-	-	-	-	15±0.0	21±0.0	ND	ND	-
<i>H. pylori</i> BA 42	-	-	-	-	-	-	-	-	-	-	R	23±0.5	ND	ND	-
<i>H. pylori</i> BA 504	-	-	-	15±0.0	-	-	-	-	-	-	R	20±1.5	ND	ND	-
<i>S. aur.</i> ATCC 29213	-	-	13±0.0	15±0.5	-	12±0.0	13±1.5	17±1.5	-	-	ND	ND	25±0.5	ND	-
<i>P. aer.</i> ATCC 27853	-	-	-	-	-	-	-	11±0.0	-	-	ND	ND	23±0.0	ND	-
<i>E. coli</i> ATCC 25922	-	-	-	15±0.0	-	-	-	10±0.0	-	-	ND	ND	32±1.5	ND	-
<i>Kleb. Pneu</i> ATCC 35657	-	-	-	-	-	-	-	14±1.5	-	-	ND	ND	16±2.0	ND	-
<i>C. albicans</i> ATCC 90029	-	-	14±0.0	16±0.0	-	-	-	-	-	-	ND	ND	ND	R	-

Table 6: Antimicrobial screening of the extracts of *Spondias mombin* stem bark

Extract (mg/mL)	Methanol				Dichloromethane				RMP (µg/mL)		Cipro. (µg/mL)		Gent. (µg/mL)	Keto. (µg/mL)	DMSO
	1	2	10	20	1	2	10	20	20	40	10	20	10	20	40%
<i>M. fort.</i> ATCC 684	-	-	-	-	-	-	-	-	24±0.0	30±0.0	ND	ND	ND	ND	-
<i>M. smeg.</i> ATCC 19420	-	-	-	-	-	-	-	-	30±0.5	31±0.5	ND	ND	ND	ND	-
<i>M. abs.</i>	-	-	-	-	-	-	-	-	26±0.5	27±0.5	ND	ND	ND	ND	-
<i>M. smeg.</i>	-	12±0.0	15±0.5	18±0.5	-	-	-	-	25±0.5	26±0.0	ND	ND	ND	ND	-
<i>M. phlei</i> ATCC 19240	-	-	-	-	-	-	-	-	18±2.5	22±0.5	ND	ND	ND	ND	-
<i>H. pylori</i> BA 26	-	-	-	-	-	-	-	10±0.0	ND	ND	14±0.0	19±1.5	ND	ND	-
<i>H. pylori</i> BA 36	-	-	10±0.0	12±0.0	-	-	-	-	ND	ND	R	23±1.0	ND	ND	-
<i>H. pylori</i> BA 38	-	-	-	-	-	-	-	-	ND	ND	15±0.0	21±0.0	ND	ND	-
<i>H. pylori</i> BA 42	-	-	-	13±0.0	-	-	-	-	ND	ND	R	23±0.5	ND	ND	-
<i>H. pylori</i> BA 504	-	-	10±0.0	13±0.0	-	-	-	-	ND	ND	R	20±1.5	ND	ND	-
<i>S. aur.</i> ATCC 29213	-	-	10±0.0	12±0.0	-	-	-	-	ND	ND	ND	ND	25±0.5	ND	-
<i>P. aer.</i> ATCC 27853	-	10±0.0	16±1.5	19±1.5	-	-	-	-	ND	ND	ND	ND	23±0.0	ND	-
<i>E. coli</i> ATCC 25922	-	16±0.0	18±0.0	20±0.0	-	-	-	-	ND	ND	ND	ND	32±1.5	ND	-
<i>Kleb. Pneu</i> ATCC 35657	-	-	16±0.0	12±0.0	-	-	-	-	ND	ND	ND	ND	16±2.0	ND	-
<i>C. albicans</i> ATCC 90029	-	-	10±0.0	20±1.0	-	-	-	-	ND	ND	ND	ND	ND	R	-

Key: RM = Rifampicin; Cipro = Ciprofloxacin; Ket = Ketoconazole; Gent = Gentamycin; dichloro = Dichloromethane; ND = not determined; R = resistant; (-) = absent; MeOH = Methanol; DMSO = Distilled methanol solution; R = Resistance; ND = not determined; DCM – Dichloromethane

Table 7: Minimum inhibitory concentration of bioactive extracts

Extracts (mg/ml)	Dillenia leaf MeOH	Dillenia stembark MeOH	Dillenia fruit MeOH	Spondias leaf MeOH	Spondias stembark MeOH	Cipro (µg/mL)	Gent. (µg/mL)	Ket. (µg/mL)
<i>H. pylori</i> BA 26	ND	10	ND	ND	ND	20	ND	ND
<i>H. pylori</i> BA 36	ND	20	ND	20	5	20	ND	ND
<i>H. pylori</i> BA 38	ND	ND	ND	ND	ND	20	ND	ND
<i>H. pylori</i> BA 42	ND	10	ND	ND	20	20	ND	ND
<i>H. pylori</i> BA 504	ND	20	ND	20	10	20	ND	ND
<i>S. aureus</i> ATCC 29213	5	20	10	10	1.25	ND	10	ND
<i>P. aeruginosa</i> ATCC 27853	2.5	2.5	5	ND	1.25	ND	10	ND
<i>E. coli</i> ATCC 25922	2.5	2.5	5	10	2.5	ND	10	ND
<i>K. Pneumonia</i> ATCC 35657	ND	ND	20	ND	5	ND	10	ND
<i>C. albicans</i> ATCC 90029	10	10	2.5	10	5	ND	10	R

DISCUSSION

Antimicrobial resistance has become a significant threat to the prevention and treatment of bacterial infections globally⁹. Antimicrobial activities of *Dillenia indica* and *Spondias mombin* has been reported¹⁰,¹¹ thus the need to compare their antimicrobial effects to standard antimicrobials.

Methanol was shown to be the optimal solvent for obtaining high yield of the extracts due to similarity in polarity to the plant materials corresponding to results obtained in other studies^{12,13,14}.

The susceptibility test result showed that methanol extracts of *Spondias mombin* leaf, stem bark and *Dillenia indica* bark showed activity against the Helicobacter strains thus supporting use in treating stomach bleeding caused by Helicobacter pylori¹⁵. *Spondias mombin* bark was the most active plant extract as it demonstrated a broad spectrum of activity; having greatest activity against *Escherichia coli* and similar activity against *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Helicobacter pylori*¹⁶. It also had activity against *Mycobacterium smegmatis* even at a concentration of 2mg/ml thus can exhibit the same degree of effect when compared with standard drugs against respiratory infection¹⁷. All methanol extracts of the plant extracts had activity against *Candida albicans* hence supporting their use in treatment of skin infections and refractory oral candidiasis¹⁸. The dichloromethane fraction of *Dillenia indica* leaf was active against all Mycobacterium species except *M. smegmatis* and *M. phlei*; also supporting its use in managing respiratory infections¹⁷. The minimum inhibitory concentration of the bioactive plant extracts investigated in this study ranged from 1.25mg/ml to 20mg/ml. *Spondias mombin* stem bark showed the lowest MIC value of 1.25mg/ml inferring it had the highest potency. The MIC of all the plant samples showed varied concentrations against organisms and this gives a clue to their effective pharmacological concentration. A research

suggested that the MICs of various plants differ significantly according to extraction solvent¹⁹ due to solvent polarity and this is evidenced by the greater activity of the methanol extracts compared to the dichloromethane extracts.

In summary, this research justifies the ethnopharmacological use of these plants in the treatment of respiratory, gastrointestinal and skin infections^{15,17,19}.

Limitations of the study

Although *Spondias mombin* is indigenous to Nigeria, *Dillenia indica* is not but can be cultivated; this made collection difficult and increased research cost.

Also, extraction process of the fruits was tedious.

CONCLUSION

The leaves and stem bark of *Spondias mombin* and the leaves, fruit and stem bark of *Dillenia indica* possess significant antimicrobial properties. More research can be done to isolate and commercialize the active ingredients responsible for this activity in order to minimize the incidence of antimicrobial resistance and broaden antimicrobial treatment options.

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Preparations for FIP's 80th World Congress of Pharmacy and Pharmaceutical Sciences, in Seville, Spain, continue in partnership and collaboration.

We will, of course, continue to assess the situation and will advise on any developments in the coming weeks and months.

In order to give everyone more time in these extraordinary circumstances, the deadlines for this congress have been extended by one month:

- The deadline for abstract submission has been moved from 15 April to 15 May 2020;
- The deadline for early bird registration has been moved from 10 June to 10 July 2020.

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