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Research Article

Antimicrobial Assay of Extracts of *Cassia Siamea (Lam.)* and *Cassia Javanica (Linn.)*

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ABSTRACT

Extracts from *Cassia siamea (Lam.)* and *Cassia javanica (Linn.)* were evaluated for antimicrobial assay using Broth dilution method at a concentration ranging from 1.0×10^3 mg/L to 100 mg/L. Total eight solvents were used to extract various secondary metabolites from leaves bark and flowers of *Cassia siamea*, leaves, and seeds of *Cassia javanica*. Antibacterial assay of eight extracts was done against gram-positive bacterial strains of *Staphylococcus aureus* and *Streptococcus pyogenes* and gram-negative *Escherichia coli* and *Pseudomonas aeruginosa*. The results were compared with standard antibiotics ciprofloxacin and norfloxacin. Similarly, antifungal assay of eight extracts was done against fungi strains *Candida albicans, Aspergillus niger* and *Aspergillus clavatus* and results were compared against standard antifungal agent K. nystatin. The disc dilution method was used for evaluation of antimicrobial assay successfully. Leaf extract of *Cassia siamea* in acetone showed good activity against *E. coli* bacteria with minimum inhibitory concentration (MIC) at 25.0 mg/L. Leaf extract of *Cassia siamea* in chloroform was excellent against fungal pathogen *C. albicans* with the MIC at 250 mg/L. While leaf extract in 95% ethanol was found active against *A. niger* with the MIC at 250 mg/L. Acetone and water extracts of *Cassia siamea* flowers were found active against *C. albicans* with the MIC at 250 mg/L respectively.

Keyword: *Cassia siamea; Cassia javanica;* antibacterial assay; antifungal assay; broth dilution method.

INTRODUCTION

The importance of natural plants as alternative medicines to treat various diseases has been reported very well. Within this context, many traditional claims on the value of natural products of healthcare were reported [1]. The genus *Cassia*, comprising of 600 species widely distributed worldwide and is well known for its diverse biological and pharmacological properties [2].

Cassia siamea (Lam.) and *Cassia javanica (Linn.)* are distributed in various countries including Asia, South Africa, Mexico, China, West Indies, East Africa and Brazil. These plants are widely used by tribal people to treat various ailments including ringworm and other fungal skin infections [3]. Malayalis tribe in

India to treat a nasal infection [4] uses *Cassia* genus. The pulp of the ripe fruits has a mild, pleasant purgative action and is used as an antifungal drug [5]. Indian people are using the leaves to treat inflammation, the flowers as a purgative, the fruit as anti-inflammatory, antipyretic, abortifacient, demulcent, purgative, refrigerant, the plant is good for chest complaints, eye ailments, flu, heart and liver ailments and rheumatism [6-9]. It is useful in treating haematemesis, pruritus, leucoderma and diabetes [10-11]. Besides its pharmacological uses, its extract is also recommended for pest and disease control [12-14]. *Cassia siamea* and *Cassia javanica* exhibited significant antimicrobial activity and showed properties that support folkloric use in the treatment of some diseases as broad-spectrum antimicrobial agents [15]. The whole plant is used to treat diarrhea; seeds are used to treat skin diseases, traditional people [16-17] use flowers and fruits to treat skin diseases, fever, abdominal pain, and leprosy.

Infectious diseases are the world's leading cause of premature deaths, killing almost 50,000 people every day. In recent years, drug resistance to human pathogenic bacteria has been commonly reported from all over the world [18-20]. However, the situation is alarming in developing as well as in developed countries due to indiscriminate use of antibiotics. The drug-resistant bacteria and fungal pathogens have further complicated the treatment of infectious diseases in immune compromised, AIDS and cancer patients. In the present scenario of the emergence of multiple drug resistance to human pathogenic organisms has necessitated a search for new antimicrobial substances from other sources including plants.

Traditionally used medicinal plants produce a variety of compounds of known therapeutic properties. The substances that can either inhibit the growth of pathogens or kill them and have no or least toxicity to host cells are considered candidates for developing new antimicrobial drugs. In recent years, antimicrobial properties of medicinal plants are being increasingly reported from different parts of the world [21-26]. It is expected that plant extracts showing target sites other than those used by antibiotics will be active against drug-resistant microbial pathogens. However, very little information is available on such acts of medicinal plants [27-28].

In the present study, we focused on two plants of *Cassia* genus viz. *Cassia siamea (Lam.)* and *Cassia javanica (Linn.)* to be screened against multi-drug resistant bacteria including *S. aureus, S. pyogenes, E. coli, P. aeruginosa,* and against fungi pathogens of *C. albicans, A. niger,* and *A. clavatus*. However, most of these plants were not previously screened against multi-drug resistant pathogenic organisms. Phytochemical analysis of active plant extracts for their major group of phytoconstituents and the active group of certain extracts is reported here.

METHODOLOGY

Plant material

Leaves bark and flowers of *Cassia siamea* were collected from University campus of Bhavnagar University Bhavnagar, India. Leaves and seeds of *Cassia javanica* were collected from Gandhinagar City, India. Voucher specimens of *Cassia siamea* (BU/ PG/ CH/ HERBS/ 01/11) and *Cassia javanica* (BU/ PG/ CH/ HERBS/ 02/11) were deposited. Both species were authenticated at Botany Department, Sir P.P. Institute of science, Bhavnagar, India.

Preparations of extracts

The leaves bark and flowers of *Cassia siamea,* leaves, and seeds of *Cassia javanica* were cleaned with deionized water, oven dried at 50 ^{0}C for 48 h and powdered in a grinder. The plant material (100 g) was sequentially extracted with different solvents (petroleum ether, acetone, chloroform, ethyl acetate, n-hexane, methanol, 95% ethanol and water) (1500 mL) according to their increasing polarity using Soxhlet apparatus for 24 h at a temperature not exceeding the boiling point of the respective solvents. The obtained extracts were filtered using Whatman filter paper No. 1 and concentrated under vacuum at 40°C using a rotary vacuum evaporator *(Büchi Laboratoriums, Switzerland) to dryness*. The extractive value of the extracts (percentage yield, water-soluble extractive, and alcohol soluble extractive) was calculated (Fig. 1). The residual extracts were stored in a refrigerator at 4°C in small and sterile plastic bottles. For antibacterial and antifungal assays, all extracts

were dissolved in DMSO to a concentration 2.0 × 10^3 mg/L.

Microorganisms and in vitro antimicrobial assays

Bacteria

The bacteria used in this investigation *S. aureus* (MTCC 96), *S. pyogenes* (MTCC 442), *E. coli* (MTCC 443) and *P. aeruginosa* (MTCC 1688) strains were clinical isolates, which were obtained from Institute of Microbial Technology, Chandigarh. The control tube containing no antibiotic is immediately subcultured (before inoculation) by spreading a loopful evenly over a quarter of a plate of medium suitable for the growth of the test organism and incubated at 37 ^{0}C overnight on Mueller-Hinton Agar (MHA) (Hi-media, Mumbai).

Antibacterial Assay

For the antibacterial assay, the MIC of all the extracts was determined by various amounts, (2.0 × 10^3 mg/L, 1.0 × 10^3 mg/L, and 500 mg/L) of extracts using disc-diffusion method (Murray et al., 1995). The tests were conducted at three different concentrations with three replicate measurements. The test cultures were swabbed on the top of the solidified media and allowed to dry for 10 min. The loaded discs were placed on the surface of the medium and left for 30 min. at room temperature for compound diffusion. The bacterial colonies were transferred into the sterile screw-capped round tubes with glass beads to which 5 mL of saline (0.9% NaCl) was added for achieving ciprofloxacin and norfloxacin standard (10^8 CFU/mL) [29]. A hundred microlitre of each suspension was introduced on petri-dish containing the extracts and controls. The plates were then allowed to incubate at 37 ^{0}C for 24 h and served as a positive control, while respective solvents mixed with nutrient agar served as negative controls. The highest concentration of respective solvents (4%) did not affect the growth of any of the organism.

Fungi

The fungal pathogens used in the study, *C.albicans* (MTCC 227), *A.niger* (MTCC 282) and *A.clavatus* (MTCC 1323) were from culture collection at the Institute of Microbial Technology, Chandigarh. Each fungus was maintained on Sabouraud Dextrose Agar (SDA) (Hi-Media, Mumbai) at 25 ^{0}C ± 2 ^{0}C for 72 h. The SDA reference strains of fungi were sub-cultured regularly and stored at -80 ^{0}C by preparing suspensions in 10% glycerol.

Antifungal Assay

For the antifungal assay, the required amount of extracts in respective solvents were added to sterile SDA in 5 mL petri-dish containing to yield a final concentration of 2.0 × 10^3 mg/L, 1.0 × 10^3 mg/L, and 500 mg/L. SDA plates with respective solvents alone incubated with fungi served as growth controls. Once the agar has solidified, a 5 mm plug of 72 hold fungal culture was placed in the center of the petri-dish containing the extract amended and un-amended SDA plates. Each solution of the extract was introduced with five µL of a suspension containing 10^4 spore/mL fungi [30].

The plates were sealed with paraffin and placed in a 25 ^{0}C ± 2 ^{0}C incubator. Fungal growth was measured on two diametric lines after 24 h, 48 h, and 72 h at 25 ^{0}C up to 9 days for dermatophyte strains. Each treatment was replicated three times and results expressed as the mean of three replicates. The results of six days growth were statistically analyzed using analysis of variance (ANOVA). The antifungal agent Fluconazole was incubated in the assay as positive controls and was added to agar plates with a final concentration of each solvent (4%) did not affect any of the organism.

RESULT AND DISCUSSION

The total percentage of secondary metabolites of *Cassia siamea* and *Cassia javanica* extracted by eight solvents viz. 95% ethanol, methanol, ethyl acetate, n-hexane, chloroform, petroleum ether, acetone, and water were depicted in the Fig. 1. The maximum percentage yield was found in leaf extract of *Cassia javanica* in 95% ethanol. The optimum condition for percentage yield of extraction was also defined.

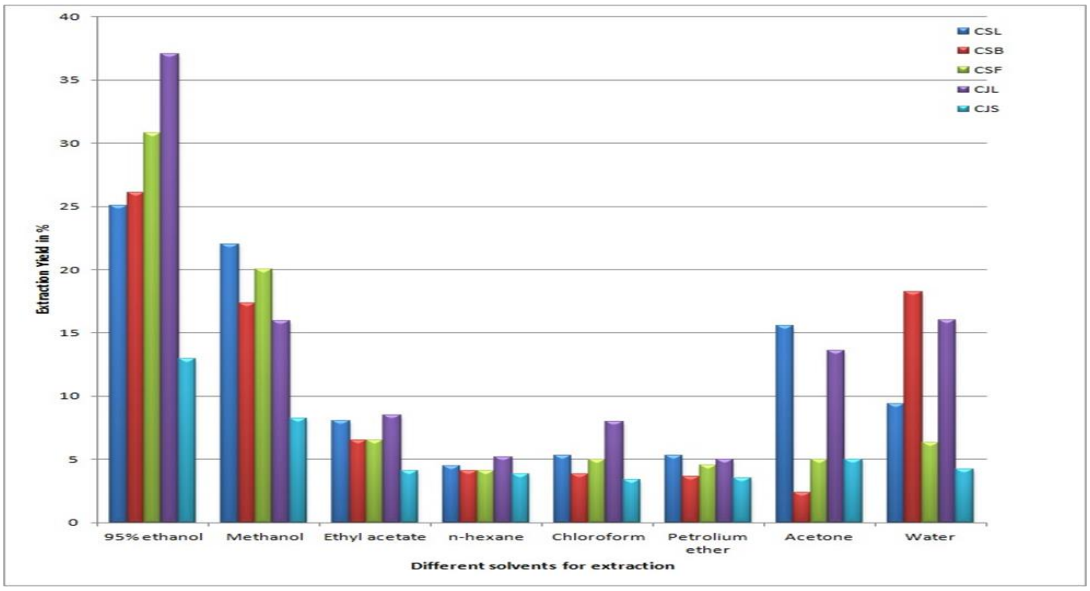

Fig. 1: Total percentage yield of extracted components from *Cassia siamea* and *Cassia javanica* plant parts are shown here. The bar chart shows that maximum percentage yield was obtained in 95% ethanolic leaf extract of *Cassia javanica*.

Antibacterial Assay

The results of antibacterial assay showed that the leaf extract of *Cassia siamea* in acetone was found excellent active against gram-negative bacteria strain of *E.coli* with 25 mg/L MIC, while n-hexane extract of the leaf was found good active against gram-negative bacteria strain of *P. aeruginosa* with 125 mg/L MIC. It was also observed from results that methanolic and petroleum ether extracts of *Cassia siamea* leaves were found excellent active against gram-positive bacteria stain of *S. aureus* with 62.5 mg/L MIC.

Chloroform extract of *Cassia siamea* flowers was found excellent active against gram-negative bacteria strain of *E. coli* with 62.5 mg/L MIC, while petroleum ether extract of the flower was found excellent active against the gram-negative bacterial strain of *P. aeruginosa*. Similarly, acetone extract was found excellent active against gram-positive bacteria strain of *S. aureus*. The results also indicate that chloroform and petroleum ether extracts of *Cassia siamea* flowers were found excellent active against all four bacterial strains with 62.5 to 100 mg/L MIC, while n-hexane extract of *Cassia siamea* bark was found excellent active against all four bacterial strains with 62.5 to 100 mg/L MIC.

Leaf extract of *Cassia javanica* in n-hexane was found excellent active against gram-negative bacterial strains of *E. coli* and *P. aeruginosa* with 62.5 mg/L and 100 mg/L MIC respectively. While methanolic extract of the leaf was found excellent active against the gram-positive bacterial strain of *S. aureus*. Acetone extract of *Cassia javanica* leaves was found excellent active against gram-positive bacteria strain of *S. pyogenes* with 100 mg/L MIC.

Ethyl acetate and chloroform extracts of *Cassia javanica* seeds were found excellent active against gram-positive bacterial strains of *S. aureus* and *S. pyogenes* with 62.5 mg/L and 100 mg/L MIC respectively, while n-hexane and water extracts of seeds were found excellent active against gram-negative bacterial strain of *E. coli* with 100 mg/L MIC. Therefore, it is evident from our results that all the extracts of both species were found fairly well to excellent activity against all four bacterial strains used for the present study. Therefore, it is suggestive that extracts were multi-resistance in nature as both gram-negative and gram-positive bacterial strains was inhibited successfully at very low concentration range, if compared with standard antibiotic like ciprofloxacin with 25mg/L to 50 mg/L MIC, and norfloxacin with 10 mg/L MIC for all the four bacterial strains. The results of the antibacterial assay were summarized in Table 1.

Anti fungal Assay

Results of the antifungal assay were also encouraging. Chloroform and 95% ethanolic extracts of *Cassia siamea* leaves were found good activity against fungal pathogens *C. albicans* and *A. niger* with 250 mg/L MIC, while acetone

and water extracts of *Cassia siamea* flowers were found good active against *C. albicans* with 250 mg/L MIC. Ethyl acetate and n-hexane extracts of *Cassia siamea* bark were found good active against *C. albicans* with 250 mg/L MIC, while acetone extract was found active against fungal pathogen *A. niger* with 250 mg/L MIC. Chloroform extract of *Cassia javanica* seeds was found good active against fungal pathogen *A. niger* with 250 mg/L MIC. The results of the antifungal assay were summarized in Table 1.

Table 1: Antibacterial assay of *Cassia siamea* and *Cassia javanica* plant parts in various solvents with their minimum inhibitory concentration (MIC)

	S. aureus	***S. pyogenes***	***E. Coli***	***P. aeruginosa***	***C. albicans***	***A. niger***	***A. clavatus***
				***C. siamea* leaves**			
95% ethanol	100±1.03**	50±0.87**	200±0.93	250±1.12	>1000	250±1.11*	500±0.78
Methanol	62.5±1.49***	100±2.78*	250±1.97	250±0.54	>1000	500±2.37	500±1.89
Ethyl acetate	250±1.89	250±2.2	62.5±1.52***	100±0.47**	>1000	500±2.1	500±1.8
n-hexane	250±1.5	250±0.8	125±1.6*	125±1.2**	500±2.44	1000±2.3	1000±2.1
Chloroform	100±0.8**	100±0.95**	125±0.78*	500±1.23	250±1.5**	>1000	>1000
P. Ether	62.5±0.9*	200±2.1	200±1.97	250±1.56	500±2.42	>1000	>1000
Acetone	125±1.6*	100±2.1**	25±0.58***	250±1.68	500±2.65	1000±2.1	1000±2.5
Water	250±2.3	500±2.8	125±1.52**	200±2.9	1000±3.2	500±1.88	500±1.54
				***C. siamea* flower**			
95% ethanol	250±1.15	100±0.59*	200±0.73	250±1.19	1000±2.35	>1000	>1000
Methanol	200±2.15	100±0.82**	250±	200±	>1000	1000±3.54	1000±2.57
Ethyl acetate	250±1.47	250±1.99	250±2.03	200±2.11	>1000	>1000	>1000
n-hexane	100±0.84**	100±0.57*	62.5±1.1***	100±0.9*	500±1.35	1000±1.82	>1000
Chloroform	250±0.8	200±0.73	100±1.1*	125±1.3*	1000±2.3	>1000	>1000
P. Ether	200±2.31	250±1.80	250±1.94	200±1.4	500±1.92	>1000	>1000
Acetone	100±1.9*	100±1.8**	200±0.76	125±0.81**	250±2.4*	1000±2.21	1000±
Water	100±0.94**	125±1.65*	250±2.6	200±2.1	250±1.5**	1000±2.6	1000±2.15
				***C. siamea* bark**			
95% ethanol	250±1.94	250±2.07	250±1.93	100±0.87**	>1000	>1000	>1000
Methanol	500±1.15	500±1.25	100±0.89**	125±2.19*	>1000	>1000	>1000
Ethyl acetate	125±0.57*	100±0.67**	200±0.95	250±1.2	250±1.3**	1000±1.41	1000±2.32
n-hexane	200±1.35	250±2.4	125±0.85**	250±2.72	250±0.56*	1000±2.55	>1000
Chloroform	100±1.2**	100±0.8**	62.5±1.2***	100±1.45**	500±2.1	>1000	>1000

P. Ether	100±0.9**	100±1.43**	100±0.94*	62.5±1.42**	1000±1.45	1000±1.56	1000±1.83
Acetone	62.5±0.95**	200±1.35	200±2.1	250±2.53	>1000	250±1.8**	500±2.8
Water	200±2.15	100±1.69**	125±1.5*	125±0.9*	500±1.73	1000±2.1	1000±2.16
***C. javanica* leaves**							
95% ethanol	200±1.37	250±0.85	200±2.14	125±0.63**	500±1.87	>1000	>1000
Methanol	100±0.55**	125±1.57*	100±2.4*	200±2.18	>1000	500±1.49	500±1.78
Ethyl acetate	200±1.68	200±0.85	200±0.9	250±1.1	>1000	1000±2.4	1000±1.91
n-hexane	250±1.48	250±0.67	62.5±1.23**	100±0.83**	500±1.4	1000±2.44	1000±1.49
Chloroform	125±0.9*	200±1.7	100±1.2*	125±1.9*	500±1.8	>1000	>1000
P. Ether	250±0.86	200±0.75	100±1.34*	125±1.69*	1000±2.8	>1000	>1000
Acetone	125±1.6*	100±2.2**	200±1.9	125±0.9*	500±2.7	>1000	>1000
Water	200±1.56	250±1.79	200±1.1	200±1.5	>1000	1000±3.15	1000±3.05
***C. javanica* seeds**							
95% ethanol	100±1.56**	100±0.82**	125±1.64**	250±2.47	>1000	>1000	>1000
Methanol	125±0.85*	125±0.76*	200±1.58	200±2.11	>1000	>1000	>1000
Ethyl acetate	62.5±0.67***	100±2.2**	250±1.5	250±2.7	>1000	500±1.23	500±0.82
n-hexane	200±1.45	200±1.67	100±1.25**	200±2.1	500±0.8	1000±0.9	500±1.15
Chloroform	62.5±0.5**	100±0.84*	250±1.26	250±2.3	1000±2.42	250±2.6**	500±1.64
P. Ether	100±1.5*	250±1.9	200±1.24	125±1.87*	1000±2.6	500±2.2	1000±1.96
Acetone	125±0.75**	200±2.4	125±1.2*	250±1.5	1000±2.4	>1000	>1000
Water	200±2.15	250±2.4	100±0.89**	125±1.34**	500±1.9	1000±1.77	1000±2.35

Note: * = $p < 0.05$ moderately significant; ** = $p < 0.01$ significant; *** = $p < 0.001$ extremely significant *S. a. = Staphylococcus aureus, S. p. = Streptococcus pyogenes, E. c. = Escherichia coli, P. a. = Pseudomonas aeruginosa, C. a. = Candida albicans, A. n. = Aspergillus niger, A. c. = Aspergillus clavatus*

Zone of Inhibition

The results of antibacterial activity were further supported by the diameter of disc diffusion (in mm) with five different doses were selected for this study ranged from 5 µg/mL to 250 µg/mL and compared with standard antibiotics (Table 2). Initially, no disc diffusion was observed for a dose of 5 µg/mL for each extract. The 95% ethanol and acetone extracts of *C. siamea* leaves have largest disc diffusion ranged from 13 mm to 21 mm for all of the bacteria tested, especially for *E. coli*., while same leaf extract in ethyl acetate had large disc diffusion for *P. aeruginosa* ranged from 13 mm to 21 mm. 95% ethanol extract of *C. siamea* flowers had a good impact on growth inhibition of *E. coli* and *P. aeruginosa* with 13 mm to 21 mm disc diffusion respectively. Bark extract of *C. siamea* in n-hexane had a good impact on *P. aeruginosa* with 13 mm to 20 mm disc diffusion. Ethyl acetate extract of *C. javanica* leaves had a good impact on growth inhibition of *P. aeruginosa* with 14 mm to 21 mm disc diffusion, while seeds extract of same species in ethyl acetate had an excellent impact on *E. coli* with 15 mm to 21 mm disc diffusion.

The results of antifungal activity was further supported by diameter of disc diffusion (in mm) with five different doses were selected for this study ranged from 5 µg/mL to 250 µg/mL and compared with standard fungal agent. All the extracts of different parts of *C. siamea* and *C. javanica* have very good impact on both fungal pathogens used for present study with effective disc diffusion between 18 mm to 26 mm. Ethyl acetate extract of *C. siamea* bark had an excellent results of growth inhibition for *A. clavatus* with disc diffusion between 21 mm to 26 mm.

Table 2: Zone of inhibition [in mm] using disc dilution method with disc dilution dose for 250 µg/mL for each solvents

	S. aureus	***S. pyogenes***	***E. Coli***	***P. aeruginosa***	***C. albicans***	***A. niger***
			***C. siamea* leaves**			
95% Ethanol	17.66±0.94	17±0.81	20.33±0.57	17±0.81	24.33±1.24	25.66±0.47
Methanol	17±0.81	16±0.81	18.33±0.47	19.66±0.47	23.33±1.24	25.66±0.47
Ethyl acetate	16.33±0.47	18.33±0.47	19.33±0.47	20.33±0.47	24.33±1.24	24.66±0.47
n-hexane	15.66±0.47	17.33±0.47	17.66±0.57	17.33±0.47	26±0.81	24.66±0.47
Chloroform	16.66±0.94	16.33±0.47	20.33±0.47	19.33±0.47	24±0.81	24.66±0.47
Petrolium ether	15±0.81	17.33±0.57	20±0.81	17.33±0.47	25.33±1.24	25.66±0.47
Acetone	17.33±0.47	19.33±0.57	20.33±0.47	17.33±0.47	22.66±1.24	23.66±0.47
Water	16±0.81	15.33±0.47	18.33±0.47	18.33±0.47	24.33±1.24	25.66±0.47
			***C. siamea* flower**			
95% Ethanol	17.33±0.94	18.33±0.47	20.33±0.47	20.33±0.47	23±0.81	23±0.81
Methanol	17±0.81	15.33±0.47	20.33±0.47	21.66±1.24	22±0.81	22±0.81
Ethyl acetate	18±0.81	16±0.81	17.33±0.47	19±0.81	22±0.81	22±0.81
n-hexane	18.66±0.47	20±0.81	17.66±0.47	18±0.81	22±0.81	24±0
Chloroform	16.66±0.47	17.66±0.47	20±0	18±0.81	22.66±0.47	24.66±0.47
Petrolium ether	15.33±0.47	16.66±0.47	18.66±0.47	20±0.81	24.66±0.47	24.66±0.47
Acetone	18.66±0.47	18±0.81	17.66±0.47	19±0.81	24±0.81	22.66±0.47
Water	16±0.81	18±0.81	20.33±0.47	16.66±0.47	22±0.81	24.33±0.47
			***C. siamea* bark**			
95% Ethanol	16.33±0.47	17±0.81	17±0.81	18.33±0.47	21.66±1.24	20.66±0.47
Methanol	15.333±0.47	17±0.81	18.33±0.47	15.33±0.47	23±0.81	23.66±0.47
Ethyl acetate	15.33±0.47	17.66±0.47	15±0.81	16.33±0.47	24.33±0.47	25±0.81
n-hexane	18±0.81	16±0.81	16.33±0.47	18.33±0.47	25±0.81	21.66±0.47
Chloroform	16.33±0.47	16±0.81	16.33±0.47	19±0.81	22.66±0.47	21.66±0.47
Petrolium ether	17±0.81	17.66±0.47	18.33±0.47	16±0.81	22±0.81	22±0.81
Acetone	16.33±0.47	16.33±0.47	21±0.81	18±0.81	23.33±0.47	23.33±0.47
Water	17.33±0.47	18.33±0.47	18.33±0.47	14.33±0.47	23.33±0.47	21.66±0.47
			***C. javanica* leaves**			
95% Ethanol	17.66±0.47	17.66±0.47	19±0.81	16±0.81	22±1	23±0.81
Methanol	18.33±0.47	18.33±0.47	19.33±0.47	18.33±0.47	23.66±0.57	23±0.81
Ethyl acetate	16.33±0.47	16.33±0.47	20.33±0.47	16.33±0.47	24.33±0.57	23.66±0.47
n-hexane	19.33±0.47	15±0.81	18.66±0.47	18.33±0.47	21±1	24±0.81

Chloroform	15.33±0.47	16.33±0.47	20.33±0.47	18.33±0.47	22±1	21±0.81
Petrolium ether	17.33±0.47	18.33±0.47	18.33±0.47	17.66±0.47	23±1	23.33±0.47
Acetone	18.33±0.47	17.33±0.47	18.33±0.47	19.33±0.94	22±1	21±0.81
Water	16.33±0.47	16.33±0.47	20.33±0.47	16±0.81	22±1	23±0.81
***C. javanica* seeds**						
95% Ethanol	15.33±0.47	18.33±0.47	20.33±0.47	18.66±0.47	24±1	23.66±0.47
Methanol	16.33±0.47	18.33±0.47	17.33±0.47	17.66±0.47	23±1	24.66±0.47
Ethyl acetate	18.33±0.47	18.33±0.47	20.33±0.47	14.33±0.47	24.33±0.57	23.33±0.47
n-hexane	18.33±0.47	16.33±0.47	19.33±0.47	18.33±0.47	23±1	22±0.81
Chloroform	16.33±0.47	15.33±0.47	20.33±0.47	14.33±0.47	24.33±0.57	23.33±0.47
Petrolium ether	18.33±0.47	17.66±0.47	16.33±0.47	17.33±0.47	23.33±0.57	24.33±0.47
Acetone	18.33±0.47	19.66±0.47	19.66±0.47	18.33±0.47	23.33±0.57	23±0.81
Water	18.33±0.47	19.33±0.47	21±0.81	16±0.81	22±1	21.33±0.47

S. a. = Staphylococcus aureus, S. p. = Streptococcus pyogenes, E. c. = Escherichia coli, P. a. = Pseudomonas aeruginosa, C. a. = Candida albicans, A. n. = Aspergillus niger, A. c. = Aspergillus clavatus

CONCLUSION

It is evident from our results of an antibacterial and antifungal assay that all the eight extracts from *Cassia siamea (Lam.)* and *Cassia javanica (Linn.)* species were found fairly well to excellent activity against various bacterial and fungal pathogens. It was reported that gram-negative bacteria are more resistance in nature but our results were found encouraging with leaf extract of *Cassia siamea* in acetone was found excellent active against *E. coli* bacteria, which is gram-negative in nature with the MIC at 25.0 mg/L. This suggests that the leaves extract of *Cassia siamea* in acetone may be exerting a metabolic interference for *E.coli* and may possibly inhibit *E. coli* growth. This finding also suggests that the potential bioactive compound(s) in *Cassia siamea* leaf extract have distinct influence on *E. coli* cell growth and function by interfering with any of steps involved in the growth of *E. coli* bacterial strain. Similarly, n-hexane extract of *Cassia siamea* bark was found excellent active against all four bacterial strains with 62.5 to 100 mg/L MIC. All eight extracts were found fairly well to excellent active against multi-drug resistant pathogenic organisms if compared with two antibiotics ciprofloxacin and norfloxacin using disc dilution method.

Results of the antifungal assay were also found in good agreement with antibacterial assay and MIC for all three fungal pathogens was 250 mg/L, which was considered fairly well antifungal activity of each extract. It was found from the results that the more than one extracts of *Cassia Siamea* were fairly well active against fungi pathogen *C. albicans*. The findings suggest that the potential bioactive compound(s) in *Cassia siamea* may be exerting a metabolic interference for *C. albicans* and have the divergent influence of *C. albicans*. If we compared all the results of antibacterial and antifungal assay suggest that the leaves extract of *Cassia siamea* was found excellent active against the gram-negative bacterial strain of *E. coli* and equally active against fungi pathogen *C. albicans*. This can be attributed that leaves of *Cassia siamea* may contain multi-resistance bioactive compound(s), which inhibit the growth of bacteria and fungi effectively.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this paper.

REFERENCES

1. Nair R, Kalariya T, Sumitra C. Antibacterial activity of some selected Indian Medicinal flora. Turkey J Biol 2005; 29: 41–47.

2. Claudio VJ, Bolzani VS, Maysa F, Barreiro EJ, Maria CM. Young, Daniela T, Eberlin MN. Further Bioactive Piperidine Alkaloids from the Flowers and Green Fruits of *Cassia spectabilis*. J Nat Prod 2004; 67(5): 908-910.
3. Rajan S, Baburaj DS, Sethuraman M, Parimala S. Stem and stem bark used medicinally by the Tribals Irulas and Paniyas of Nilgiri District, Tamilnadu. Ethnobot 2001; 6: 19-24.
4. Perumal SR, Ignacimuthu S, Sen A. Screening of 34 medicinal plants for antibacterial properties. J Ethnopharmacol 1998; 62: 173-182.
5. Kasuko I, Nagayo O. Effects of vegetable drugs on pathogenic fungi *I*. Effect of anthraquinone-glycoside containing crude drugs upon the growth of pathogenic fungi. Bullet Pharm Res Inst 1951; 2: 23-29.
6. Patel D, Karbhari D, Gulati D, Gokhale D. Antipyretic and analgesic activities of *Aconitum spicatum* and *Cassia fistula*. Pharm Biol 1965; 157: 22-27.
7. Biswas K, Ghosh AB. Bharatia Banawasadhi, Calcutta University. Calcutta India: Advancement of learning; 1973, p 336.
8. Kirtikar KR, Basu BD. Indian Medicinal Plants. New Delhi, India: Jayyed Press; 1975.
9. Satyavati GV, Sharma M. Medicinal Plant in India. New Delhi, India: ICMR; 1989.
10. Alam MM, Siddiqui MB, HussianW. Treatment of diabetes through herbal drugs in rural India. Fitoterpia 1990; 61: 240-242.
11. Asolkar LV, Kakkar KK, Chakre OJ. Second supplement to a glossary of Indian medicinal plant with active principles. New Delhi, India: Publication and Information Directorate, CSIR; 1992, p 177.
12. Jaipal S, Sing Z, Chauhan R. Juvenile hormone-like activity in extracts of some common Indian plants. Indian J Agricul Sci 1983; 53: 730-733.
13. Sharma BK, Basandrai AK. Efficacy of some plant extracts for the management of Karnal bunt [Neovossia (Tilletia) indica] of wheat *Triticum aestivum*. Indian J Agricul Sci 1999; 69: 837-839.
14. Raja N, Albert S, Ignacimuthu S. Effect of solvent residues of *Vitex negundo Linn*. and *Cassia fistula (Linn*.) on pulse beetle, *Callosobruchus maculates* Fab. and its larval parasitoid, *Dinarmus vagabundus* (Timberlake). Indian J Exper Biol 2000; 38: 290-292.
15. Prashanth Kumar V, Chauhan NS, Padh H, Rajani M. Search for antibacterial antifungal agents from selected Indian medicinal plants. J Ethnopharmacol 2006; 107: 182-188.
16. Perry LM. Medicinal plants of East and South East Asia. Cambridge: MIT Press; 1980.
17. Hua YC, Chien MY, Lin TC, Shieh DE, Chun CL. Epiafzelechin-(4aR8)-epiafzelechin extracted from *Cassia javanica* inhibits herpes simplex virus type 2 replication. J Med Microbiol 2006; 55: 201-206.
18. Piddock KJV, Wise R. Mechanisms of resistance to quinolones and clinical perspective. J Antimicrob Chemother 1989; 23: 475-483.
19. Mulligen ME, Murry-Leisure KA, Ribner BS, Standiford HC, John JF, Karvick JA, Kauffman CA, Yu VL. Methicillin-resistant Staphylococcus aureus. American J Med 1993; 94: 313-328.
20. Robin EH, April W, Alexander M, Loeto M, Keith K. Nasopharyngea carriage and antimicrobial resistance in isolates of *Streptococcus pneumonia* and *Heamophilus influenzae* Type-B in children under 5 years of age in Botswana. Int J Infect Dis 1998; 3: 18-25.
21. Grosvenor PW, Supriono A, Gray DO. Medicinal plants from Riau Province, Sumatra, Indonesia. Part 2, Antibacterial and antifungal activity. J Ethnopharmacol 1995; 45: 97-111.
22. Ratnakar P, Murthy PS. Purification and mechanisms of action of antitubercular principle from garlic (Allium sativum) active against isoniazid susceptible and resistant *Mycobacterium tuberculae* H37RV. Indian J Clin Biochem 1995; 10: 14-18.
23. Silva O, Duarte A, Cabrita J, Gomes E. Antimicrobial activity of Guinea-Bissau traditional remedies. J Ethnopharmacol 1996; 50: 55-59.
24. David M. Antimicrobial activity of garlic. Antimicrob Agent Chemothe 1997; 41: 2286.

25. Saxena K. Antimicrobial Screening of Selected Medicinal Plants from India. J Ethnopharmacol 1997; 58: 75-83.
26. Saxena VK, Sharma RN. Antimicrobial activity of essential oil of *Lantana aculeate*. Fitoterapia 1999; 70: 59-60.
27. Hasegawa H, Matsumya S, Yamasak K. Reversal of efflux-mediated tetracycline resistance in *Staphylococcus aureus* clinical isolates by Ginseng prosaponenins. Phytother Res 1995; 9:260-263.
28. Lee CK, Kin H, Moon KH, Shun KH. Screening and isolation of antibiotic resistance inhibitors from herb materials-resistance inhibition of volatile components of Korean aromatic herbs. Arch Pharm Res 1998; 21: 62-66.
29. Anja K, Piskernik S, Barbara JS, Smole M. Evaluation of diffusion and dilution methods to determine the antibacterial activity of plant extracts, J Microbiol Meth 2010; 81: 121-126.
30. Duraipandiyan V, Ignacimuthu S. The antibacterial and antifungal activity of *Cassia fistula* L.: An ethnomedicinal plant. Ethnopharmacol 2007; 112(3): 590-594.

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