

Antimicrobial Activity of *Polyalthia longifolia* (Sonn.) Thw. var. Pendula Leaf Extracts Against 91 Clinically Important Pathogenic Microbial Strains

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Abstract

The methanol, acetone and 1,4-dioxan fractions of leaves of *Polyalthia longifolia* (Sonn.) Thw. were evaluated for antibacterial and antifungal activity. 91 clinically important strains were used for the study which were both clinical isolates as well as identified strains. Piperacillin and gentamicin were used as standards for antibacterial assay, while nystatin and flucanazole were used as standards for antifungal assay. The antibacterial activity was more pronounced against gram positive bacterial and fungal strains. Poor activity was shown against gram negative bacterial strains studied.

Keywords: Antibacterial, Antifungal, Polyalthia longifolia, Clinical Isolates, Organic Solvent Extracts

1. Introduction

Due to the increasing development of drug resistance in human pathogens as well as the appearance of undesirable effect on certain antimicrobial agents, there is a need to search for new agents. The world health organization in 1997 suggested that effective locally available plants be used as substitutes for drugs. Research work on medicinal plants be intensified and information on these plants be exchanged. This thought will go a long way in the scientific exploration of medicinal plants for the benefit of man and is likely to decrease the dependence on importance of drugs [1]. Polyalthia longifolia (Annonaceae) is a tree, which is widely distributed in Bangladesh, Srilanka and throughout the hotter parts of India [2]. In India, the seeds of this plant were used as febrifuge [3]. Literature survey revealed that most of the plants of annonaceae family contain antitumor and anticancer principles [4,5]. The bark is also used as a febrifuge in the Balasore district of Orissa [6]. The extract of stem bark and the alkaloids isolated from this were found to demonstrate a good antibacterial and antifungal activities [7]. In the present study, antimicrobial potentiality of the P. longifolia leaves was investigated against a few clinically isolated as well as standard microbial cultures.

2. Materials and Methods

2.1. Plant Material

Polyalthia longifolia (Sonn.) Thw. (Annonaceae) leaves were collected in May, 2004 from Rajkot in the State of Gujarat (Western India) and identified by comparison with specimens (PSN 4) available at the Herbarium of the Department of Biosciences, Saurashtra University, Rajkot, Gujarat, India.

2.2. Extraction

Leaves of *P. longifolia* were collected, air dried and then powdered in a homogenizer and 10 gm was used for different solvent extractions (Methanol, Acetone, N, N-dimethylformamide); the sample was extracted in solvent kept on a rotary shaker overnight, and then the filtrate was collected and centrifuged at 5000 rpm. The solvent was then evaporated to dryness under reduced pressure and the extracted compound left was used for the antimicrobial assay. The percentage yield of 1, 4-dioxan, methanol and acetone extracts were 20.56, 29.30 and 13.52 respectively.

Microorganisms Studied 91 clinically important microbial strains which included 23 gram positive, 56 gram

32 Pseudomonas spps. [43]

33 Pseudomonas spps. [46]

34 Pseudomonas spps. [49]

35 Pseudomonas spps. [50]

36 Pseudomonas fluorescence [59]

Pus

Sputum

Sputum

Urine

Tracheal secretion

negative and 15 fungal strains were studied for the antimicrobial activity. These strains included both clinical isolates as well as identified strains. The details of the microorganisms used are shown in **Table 1**.

Table 1. List of bacterial and fungal strains studied for antimicrobial assay.

tim	nicrobial assay.		
	<u> </u>		37 Pseudomonas aeruginosa ATCC 27853 -
Sr.	Strain	Specimen	38 Pseudomonas testosteroni NCIM 5098 -
	Gram Positive bacteria		39 Pseudomonas pseudoalcaligenes ATCC
1	Staphylococcus spps. [10]	Sputum	40 <i>E.coli</i> [14] Pus
2	Staphylococcus aureus [11]	Pus	41 <i>E.coli</i> [16] Urine
_	Staphylococcus aureus [13]	Urine	42 <i>E.coli</i> [21] Urine
3			43 <i>E.coli</i> [22] Urine
4		Pus	44 <i>E.coli</i> [24] Urine
5	Staphylococcus spps. [26]	Pus	45 <i>E.coli</i> [28] Pus
6	Staphylococcus aureus [34]	Sputum	46 <i>E.coli</i> [31] Urine
7	Staphylococcus aureus [35]	Tracheal	47 <i>E.coli</i> [32] Stool
8	Staphylococcus aureus [36]	Tracheal	48 <i>E.coli</i> [33] Pus
9	Staphylococcus spps. [44]	Sputum	49 <i>E.coli</i> [41] Urine
10	Staphylococcus aureus [47]	Ear swab	50 <i>E.coli</i> [45] Pus
11	Staphylococcus aureus [48]	Sputum	51 <i>E. coli</i> [51] Urine
12	Staphylococcus aureus [55]	Pus	52 <i>E. coli</i> [58] Vaginal swal
13	Staphylococcus aureus [56]	Pus	53 <i>E. coli</i> [60] Urine
14	Staphylococcus aureus ATCC 25923	-	• •
15	Staphylococcus epidermidis ATCC 12228	-	54 <i>E. coli</i> [61] Blood
16	Staphylococcus subfava NCIM 2178	-	55 E. coli ATCC 25922 -
17	Bacillus cereus ATCC 11778	-	56 Enterobacter spps. [1] Tracheal
18	Bacillus subtilis ATCC 6633	-	57 Enterobacter spps. [8] Tracheal
19	Bacillus megaterium ATCC 9885	-	58 Enterobacter aerogenes ATCC 13048 -
20	Micrococcus flavus ATCC 10240	-	59 Klebsiella spps [6] Urine
	Gram negative bacteria		60 Klebsiella spps [19] Sputum
21	Pseudomonas spps. [15]	Sputum	61 Klebsiella aerogenes [52] Pus
22	Pseudomonas spps. [17]	Pus	62 Klebsiella spps. [54] Urine
23	Pseudomonas fluorescence [18]	Pus	63 Klebsiella aerogenes [57] Urine
24	Pseudomonas spps. [25]	Urine	64 Klebsiella pneumoniae NCIM 2719 -
25	Pseudomonas spps. [27]	Pus	65 Proteus mirabilis [4] Wound swab
26	Pseudomonas aeruginosa [30]	Sputum	66 Proteus spps. [53] Pus
	Pseudomonas spps. [37]	Tracheal	67 Proteus mirabilis NCIM 2241 -
	Pseudomonas aeruginosa [38]	Pus	68 Proteus vulgaris NCTC 8313 -
	Pseudomonas spps. [39]	Wound swab	69 Proteus morganii NCIM 2040 -
	Pseudomonas fluorescence [40]	Tracheal	70 Providencia rettgeri [5] Pus
31	Pseudomonas spps. [42]	Pus	71 Citrobacter spps. [20] Pus

72 Citrobacter freundii [29]	Pus
73 Citrobacter freundii ATCC 10787	-
74 Alcaligenes fecalis ATCC 8750	-
75 Salmonella typhimurium ATCC 23564	-
Fungus	
76 Candida albicans [1]	Urine
77 Candida albicans [2]	Sputum
78 Candida spps. [3]	Sputum
79 Candida spps. [4]	Sputum
80 Candida spps. [5]	Urine
81 Candida albicans ATCC 2091	-
82 Candida albicans ATCC 18804	-
83 Candida glabrata NCIM 3448	-
84 Candida tropicalis ATCC 4563	-
85 Candida apicola NCIM 3367	-
86 Cryptococcus neoformans ATCC 34664	-
87 Cryptococcus luteolus ATCC 32044	-
88 Trichosporan beigelii NCIM 3404	-
89 Aspergillus flavus NCIM 538	-
90 Aspergillus candidus NCIM 883	-
91 Aspergillus niger ATCC 6275	-

2.3. Preparation of Samples

Methanol, acetone and 1,4-dioxan extracts were dissolved in 100% DMSO at a concentration of 25 mg/ml and 12.5 mg/ml and were used as working stocks respectively. Sterile discs (Hi-media Labs) were impregnated with 20 μ l of the stock solution. Gentamicin (10 μ g/disc) and Piperacillin (100 μ g/disc) for bacteria; nystatin (100 units/disc) and flucanazole (10 μ g/disc) (Himedia Labs) for fungus were used as positive control and pure DMSO was used as a negative control.

2.4. Antimicrobial Study

Antimicrobial activity was performed by agar disc diffusion method [8,9]. The bacterial strains were grown in nutrient broth while fungal strains were grown in MGYP (Malt glucose yeast peptone) broth. Mueller Hinton agar no. 2 was the media used to study the antibacterial susceptibility while Sabroaud agar was used to study the antifungal susceptibility test. The cultures were grown for 24 hours, and the turbidity of the culture was maintained according to the 0.5 MacFarland standards. The inoculum's size was 1×10^8 cells/ml.

2.5. Agar Disc Diffusion

The media (Mueller Hinton Agar No.2 and MRS media) and the test bacterial cultures were poured into Petri dishes (Hi-Media). The test strain (200 µl) was inoculated into the media (inoculums size 10⁸ cells/ml) when the temperature reached 40-42°C. The test compound (20 µl) was impregnated in to sterile discs (7 mm) (Hi-Media) and was then allowed to dry. The disc was then introduced into medium with the bacteria. The plates were incubated overnight at 37°C for bacterial strains and 28°C for fungal strains. The experiment was performed under strictly aseptic conditions. Microbial growth was determined by measuring the diameter of the zone of inhibition. The experiment was performed in triplicates and the mean values of the result are shown in **Table 2**.

3. Results and Discussion

Herbal medicine in developing countries is commonly used for the traditional treatment of health problems [10]. In recent years multiple drug resistance in human pathogenic microorganisms has developed due to the indiscriminate use of commercial antimicrobial drugs commonly used in the treatment of infectious diseases, making it a global growing problem [11-13]. In addition to this problem antibiotics are sometimes associated with adverse effects on host including hypersensitivity, immune suppression and allergic reactions [14]. Therefore there is a need to develop alternative antimicrobial drugs for the treatment of infections obtained from various sources such as medicinal plants [15,16]. In the present study, P. longifolia leaf extracts extracted in 1, 4-dioxan (PDE), methanol (PME) and acetone extracts (PAE) were investigated at two different concentrations for their antimicrobial potentiality against 91 clinically important microbial strains. All the three extracts (PDE, PME and PAE at 500 µg/disc concentration) were active against 95% of the total gram positive bacterial strains studied. PDE was active against 18.18% of the total gram negative bacterial strains studied (active against 21% of Pseudomonas spps., 33.3% of Enterobacter spps., 16% of Klebsiella spps., 33.3% of Proteus spps. and 66.6% of Citrobacter spps.). PME and PAE were active against 12.72% of the total gram negative bacterial strains studied. P. aeruginosa is most common pathogen of immuno-compromised individuals [17]. Infections caused by Pseudomonas spps. are among the most difficult to treat with conventional antibiotics. Both PME and PAE were active against 5.26% of the Pseudomonas spps. and 66.6% of Enterobacter spps. PME was active against 33.3% of Klebsiella spps. and Proteus spps., while PAE was active against 66.6% of Klebsiella spps. and Proteus spps. studied. Salmonellosis is an important public

Table 2. Antimicrobial activity of *Polyalthia longifolia* against 91 clinically important microbial strains (inhibition zone in mm).

Sr.	a	Control Extracts							Antibiotics				
No.	Strain	DMSO	PDE-500	PME-500	PAE-500	PDE-250	PME- 250	PAE- 250	G	Pc	Fu	Ns	
	Gram Positive bacteria						230	230					
1	Staphylococcus spps. [10]	-	15 ± 0.58	12.67± 0.33	10 ± 0.58	10 ± 0.58	11 ± 0.58	14.67± 0.88	-	-	NT	NT	
2	Staphylococcus aureus [11]	-	13 ± 0.58	12 ± 0.58	10 ± 0.58	11 ± 0.58	12 ± 1.15	12 ± 0.58	18.67± 0.33	17.33 ± 0.33	NT	NT	
3	Staphylococcus aureus [13]	-	12 ± 1.15	12 ± 2.31	9 ± 0.58	-	-	-	-	-	NT	NT	
4	Staphylococcus aureus [23]	-	11 ± 0.58	12 ± 1.73	9 ± 0.58	12 ± 1.73	9 ± 1.15	-	-	-	NT	NT	
5	Staphylococcus spps [26]	-	16.5 ± 0.28	11± 0.58	13± 0.58	15± 0.58	13 ± 1.73	14 ± 1.73	-	-	NT	NT	
6	Staphylococcus aureus [34]	-	15.5 ± 0.28	9 ± 0.58	13 ± 0.58	14 ± 0.58	9 ± 0.58	10 ± 1.15	-	-	NT	NT	
7	Staphylococcus aureus [35]	-	22± 0.28	12 ± 0.28	14 ± 0.58	17± 0.58	8 ± 0.58	11 ± 0.58	-	-	NT	NT	
8	Staphylococcus aureus [36]	-	13 ± 0.58	9.67 ± 0.33	13 ± 0.58	12.67 ± 0.88	-	0.38 8.67± 0.88	_	-	NT	NT	
9	Staphylococcus spps [44]	-	13 ± 0.58	10.33 ± 0.33	12.33 ± 0.33	18 0.58	11 ± 0.58	10.67 ±	14.67± 0.33	-	NT	NT	
10	Staphylococcus	-	12 ±	10.67 ±	11 ±	9 ±	8.67 ±	0.66 12 ±	-	_	NT	NT	
11	aureus [47] Staphylococcus	-	3.21	2.03	2.31	1.15	0.88	2.89	20.67±	-	NT	NT	
12	aureus [48] Staphylococcus aureus [55]	-	13.67 ± 0.33	12.67 ± 0.33	13.67 ± 0.33	11.67 ± 0.33	-	-	0.33	-	NT	NT	
13	Staphylococcus aureus [56]	-	15.67 ± 0.33	10 ± 1.53	11.67 ± 0.88	12.33 ± 0.33	10.33 ± 1.76	12.67 ± 0.33	10.33± 0.33	-	NT	NT	
14	Staphylococcus aureus ATCC 25923	-	13 ± 0.58	8 ± 0.58	9 ± 0.58	14.33 ± 0.88	9.5 ± 0.28	9 ± 0.58	-	-	NT	NT	
15	Staphylococcus epidermidis ATCC 12228	-	14.5 ± 2.60	16 ± 2.69	13 ± 0.58	13.5 ± 0.87	13 ± 0.57	12 ± 1.73	-	-	NT	NT	
16	Staphylococcus subfava NCIM 2178	-	10.5 ± 0.29	11.5 ± 1.44	12.5 ± 0.28	13 ± 2.31	9.5 ± 0.28	9.5 ± 0.28	-	20.17 ± 0.44	NT	NT	
17	Bacillus cereus ATCC 11778	-	29.5 ± 0.28	21.5 ± 0.28	25. ± 0.58	25 ± 2.31	21 ± 0.58	25 ± 0.58	20.17 ± 0.16	18.83 ± 0.16	NT	NT	
18	Bacillus subtilis ATCC 6633	-	26.5 ± 1.44	21.5 ± 1.44	23.5 ± 0.28	25 ± 0.58	21 ± 0.58	21 ± 0.58	18.33 ± 0.33	17.83 ± 0.93	NT	NT	
19	Bacillus megaterium ATCC 9885	-	14 ± 0.58	10.5 ± 0.28	12.5 ± 0.28	13 ± 0.58	11 ± 0.58	10.5 ± 0.28	-	-	NT	NT	
20	Micrococcus flavus ATCC 10240	-	12.5 ± 0.28	10.5 ± 0.28	11 ± 2.31	11.5 ± 0.28	9 ± 0.58	9 ± 0.58	27.67 ± 0.33	12.67 ± 0.33	NT	NT	
	Gram negative bacteria										NT	NT	
21	Pseudomonas spps. [15]	-	-	-	-	-	-	-	14 ± 0.58	-	NT	NT	
22	Pseudomonas spps. [17]	-	8 ± 0.58	-	-	-	-	12 ± 2.89	-	-	NT	NT	
23	Pseudomonas fluorescence [18]	-	8± 0.58	-	-	-	-	12± 2.89	-	-	NT	NT	
24	Pseudomonas spps. [25]	-	-	-	-	-	-	-	-	-	NT	NT	

25	Pseudomonas spps. [27]	-	-	-	-	-	-	-	-	-	NT	NT
26	Pseudomonas	_		_	-	_	_	_	16.67±	_	NT	NT
27	aeruginosa [30] Pseudomonas								0.67		NT	NT
	spps. [37] Pseudomonas	-		-	-	-	-	-	- 19.67±			
28	aeruginosa [38]	-	-	-	-	-	-	-	0.33	-	NT	NT
29	Pseudomonas spps. [39]	-	-	-	-	-	-	-	-	-	NT	NT
30	Pseudomonas fluorescence [40]	-	-	-	-	-	-	-	-	-	NT	NT
31	Pseudomonas spps. [42]	-	-	-	-	-	-	-	-	-	NT	NT
32	Pseudomonas	-	-	_	-	_	_	_	-	_	NT	NT
33	spps. [43] Pseudomonas	_	_	_	_	_	_	_	_	_	NT	NT
34	spps. [46] Pseudomonas		8 ±					8 ±	20 ±	_	NT	NT
	spps. [49] Pseudomonas	-	0.58	-	-	-	-	0.58	0.58	-		
35	spps. [50] Pseuodmonas	-	-	-	-	-	-	-	-	-	NT	NT NT
36	fluorescence [59]	-	-	-	-	-	-	-	-	-	NT	NI
37	Pseudomonas aeruginosa ATCC 27853	-	-	-	-	-	-	-	17 ± 1.15	12.33 ± 0.66	NT	NT
38	Pseudomonas testosteroni NCIM 5098	-	-	-	-	-	-	-	22.33 ± 0.66	-	NT	NT
39	Pseudomonas pseudoalcaligenes ATCC 17440	-	8.5 ± 0.86	14 ± 1.73	10.5 ± 0.86	-	-	-	19.33 ± 0.6	-	NT	NT
40	E.coli [14]	-	-	-	-	-	-	-	-	-	NT	NT
41 42	E.coli [16]	-	-	-	-	-	-	-	-	-	NT NT	NT NT
43	E.coli [21] E.coli [22]	_	-	-	-	-	-	-	-	-	NT	NT
44	E.coli [24]	-	-	-	-	-	-	-	-	-	NT	NT
45	E.coli [28]	-						17± 0.33			NT	NT
46	E.coli [31]	-	-	-	-	-	-	-	-	-	NT	NT
47	E.coli [32]	-	-	-	-	-	-	-	21± 0.58	-	NT	NT
48	E.coli [33]	-	-	-	-	-	-	-	-	-	NT	NT
49	E.coli [41]	-	-	-	-	-	-	-	18.67 ± 0.33	-	NT	NT
50	E.coli [45]	-	-	-	-	-	-	-	20.22.	-	NT	NT
51	E. coli [51]	-	-	-	-	-	-	-	20.33 ± 0.33	-	NT	NT
52	E. coli [58]	-	-	-	-	-	-	-	-	-	NT	NT
53	E. coli [60]	-	-	-	-	-	-	-	-	-	NT	NT
54	E. coli [61] E. coli ATCC	-	-	-	-	-	-	-	17.83	14.5	NT	NT
55	25922	-	-	-	-	-	-	-	± 0.16	± 0.50	NT	NT
56	Enterobacter spps. [1]	-	-	-	-	-	-	-	-	-	NT	NT
57	Enterobacter spps. [8]	-	15 ± 0.58	12 ± 0.58	14.33 ± 1.20	13 ± 0.58	12.33 ± 0.88	12 ± 1.15	19.67± 0.88	-	NT	NT
58	Enterobacter aerogenes ATCC 13048	-	-	8.5 ± 0.86	15 ± 0.58	-	-	-	-	-	NT	NT
59	Klebsiella spps [6]	-	-	-	-	-	-	-	22± 0.58	-	NT	NT
60	Klebsiella spps [19]	-	-	-	-	-	-	-	-	-	NT	NT
61	Klebsiella aero- genes [52]	_	-	_	8 ± 0.58	13 ± 1.73	11 ± 2.08	_	_	_	NT	NT

62	Klebsiella spps. [54]	-	-	-	-	-	-	-	-	-	NT	NT
63	Klebsiella aero- genes [57]	-	-	-	-	-	-	-	-	-	NT	NT
64	Klebsiella pneu- moniae NCIM	-	12 ± 0.58	12 ± 0.58	10.5 ± 0.28	10.5 ± 0.86	10.5 ± 0.86	11 ± 0.58	-	24.67 ±	NT	NT
65	2719 Proteus mirabilis	_	-	-	-	-	-	-	_	0.33 14±	NT	NT
66	[4] Proteus spps. [53]	_	_	_	_	_		_	_	0.58	NT	NT
	Proteus mirabilis	-	10.5 ±	10.5 ±	9.5 ±	-			18.67			
67	NCIM 2241	-	0.86	0.28	0.86	-	-	-	± 0.33	-	NT	NT
68	Proteus vulgaris NCTC 8313	-	-	9 ± 1.15	-	-	-	-	18 ± 1.00	-	NT	NT
69	Proteus morganii NCIM 2040	-	9 ± 0.58	-	-	8 ± 0.58	-	-	-	-	NT	NT
70	Providencia rett- geri [5]	-	-	-	-	-	-	-	-	-	NT	NT
71	Citrobactor spps [20]	-	8 ± 0.58	8 ± 0.58	8 ± 0.58	-	-	-	-	-	NT	NT
72	Citrobactor freundii [29]	-	-	-	-	-	-	-	12.33± 0.33	-	NT	NT
73	Citrobactor freundii ATCC	-	11 ± 0.58	-	-	11.5 ± 0.28	10 ± 0.58	9.5 ± 0.28	-	_	NT	NT
74	10787 Alcaligenes fecalis ATCC 8750	-	-	-	-	-	-	-	18.33 ± 0.66	-	NT	NT
75	Salmonella ty- phimurium ATCC 23564	-	-	-	-	-	-	-	18.5 ± 0.28	-	NT	NT
	Fungus											11.22
76	Candida albicans [1]	-	7.5 ± 0.29	8 ± 0.58	-	7.5 ± 0.29	10.5 ± 0.29	10 ± 0.58	NT	NT	-	11.33 ± 0.33
77	Candida albicans [2]	-	-	-	10 ± 0.58	13.33 ± 0.88	9 ± 0.58	-	NT	NT	-	18 ± 0.58
78	Candida spps. [3]	-	-	-	9.5 ± 0.29	14.33 ± 0.66	12.5 ± 0.86	8 ± 0.58	NT	NT	-	14 ± 0.58
79	Candida spps. [4]	-	11 ± 2.13	10.5 ± 2.02	11.5 ± 2.06	8 ± 0.58	8.5 ± 0.29	12.5 ± 0.86	NT	NT	-	14 ± 0.58
80	Candida spps. [5]	-	7.5 ± 0.29	8.5 ± 0.29	9.5 ± 0.29	7.5 ± 0.29	-	-	NT	NT	-	10 ± 0.58
81	Candida albicans ATCC 2091	-	11.5 ± 2.60	11 ± 2.31	8 ± 0.58	7.5 ± 0.29	7.5 ± 0.29	10.5 ± 2.02	NT	NT	17.67 ± 0.33	13 ± 0.58
82	Candida albicans	_	10.5 ±	8 ±	_	_	11 ±	15 ±	NT	NT	-	14.33 ±
	ATCC 18804		0.29	0.58			0.58	1.15			20.65	0.33
83	Candida glabrata NCIM 3448	-	-	-	-	-	-	-	NT	NT	39.67 ± 0.88	22 ± 0.58
84	Candida tropicalis ATCC 4563	-	-	-	7.5 ± 0.29	11 ± 0.58	12 ± 0.58	9.5 ± 0.29	NT	NT	-	8.33 ± 0.33
85	Candida apicola NCIM 3367	-	23 ± 3.60	26 ± 0.58	28 ± 1.15	25.33 ± 0.88	24 ± 0.58	21.66 ±	NT	NT	-	21.33 ±
	Cryptococcus				1.10	0.00	0.50	0.33			21.33	0.88
86	neoformans ATCC 34664	-	7.5 ± 0.29	8 ± 0.58	-	-	-	9.5 ± 1.4	NT	NT	± 0.33	17 ± 0.58
87	Cryptococcus luteolus ATCC	-	14 ± 0.58	11.5 ± 0.86	11 ± 1.15	9.5 ± 1.44	8.5 ± 0.86	8.5 ± 0.88	NT	NT	23.66 ±	17.66 ±
00	32044 Trichosporan		12 ±	13 ±	10.5 ±				NIT	NIT	0.88	0.88
88	beigelii NCIM 3404	-	0.58	1.73	2.02	-	-	-	NT	NT	-	-
89	Aspergillus flavus NCIM 538	-	-	-	-	14.67 ± 4.34	22 ± 0.58	10.33 ± 2.02	NT	NT	-	-

90	Aspergillus can- didus NCIM 883	- 10. 0.2	$9 \pm 9 \pm 9 \pm 115$	11 ± 0.58	-	-	-	NT	NT	-	-
91	Aspergillus niger ATCC 6275			-	11 ± 2.31	-	-	NT	NT	-	-

Mean ± SEM, n = 3, zone includes disc diameter 7 mm; G – Gentamicin (10 μg/disc), Pc – Piperacillin (100 μg/disc), Ns – Nystatin (100 units/disc), Fu – Fluconazole (10 μg/disc); PME – Methanol extract, PAE – Acetone extract, PDE – Dioxan extract, DMSO – Dimethylsulphoxide.

health problem worldwide. Salmonella infection is primarily associated with gastroenteritis. This illness poses a more serious health risk to sensitive populations in the community such as the elderly, young and the immunocompromised, where hospitalization may be required. All the three extracts were inactive against E. coli, A. fecalis and S. typhimurium. Several antimycotic drugs are available at present, its use is limited by a number of factors such as low potency, poor solubility, emergence of resistance strains and drug toxicity. Therefore there is distinct need for the discovery of new, safer and more effective antifungal agents. Candida species have become a common cause of hospital acquired infections and a large number of patients die as a result of invasive Candidal infections [18]. All the three extracts were active against 62.5% of the total fungal strains studied. The three extracts were active against A. candidus while it was inactive against the remaining two moulds (A. flavus and A. niger) studied. The details of the results are given elaborately in Table 2. From the results obtained, it seems that the antibacterial action of the extracts is more pronounced on gram positive than on gram negative bacteria and these findings correlate with the observations of previous screenings of medicinal plants for antimicrobial activity, where most of the active plants showed activity against gram positive strains only [19-21]. This difference in susceptibility is because of the difference in cell wall structure of gram positive and gram negative organisms. The lipopolysaccharide content of gram negative bacteria makes them resistant to plant extracts while the peptidoglycan layer of gram positive bacteria is not an effective permeability barrier.

4. Conclusions

All the extracts of *P. longifolia* exhibited the highest rates of antimicrobial activity against gram positive and fungal strains studied. Therefore, it is concluded that *P. longifolia* extracts should further be studied phytochemically to elucidate the active principle in the leaf, which can be used as a leading antibacterial (specific for gram positive) and antifungal agent.

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6. References

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