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30	urnal or po OR	IGINAL RESEARCH PAPER	Chemistry				
Indian	ARIPET PHY OF E ANT MAC	TOCHEMICAL SCREENING AND EVALUATION EFFECT OF TEMPERATURE AND pH ON IBACTERIAL ACTIVITY OF CORDIAL ELEODII HOOK PLANT EXTRACT WITH SOME ECTED BACTERIA	KEY WORDS: Phytochemicals, Bacteria, chemotherapeutic agent, Plant extracts, Antibacterial activity, ZOI				
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IBSTRACT	antimicrobial activity of species at high temp phytochemicals in var acetate extract express aureus and <i>Bacillus ce</i> prevented the growth Pseudomonas aerugir	erimental study was carried out with an objective to evaluate of cordial macleodii hook plant leave extract at different pH and perature [75°c,105°c] Results: Preliminary screening ind ying degrees, which suggests the chemical diversity of this p sed a slight increment with increase PH (basic medium) for the reus increment in antibacterial activity found at low pH (acid of both Gram-positive S. aureus and <i>Bacillus cereus</i> and C nosa. The antibacterial activity (zone of inhibition) of aqueous the other solvent extracts against Gram-negative and Gram-	d temperature for selected bacterial licated the presence of different plant. Antibacterial activity of ethyl S.coli, while in case Staphylococcus dic medium). Although the extracts Gram-negative bacteria E coli and s leaves extract is maximum 19mm				

Pseudomonas aeruginosa. The antibacterial activity (zone of inhibition) of aqueous leaves extract is maximum 19mm higher than those of the other solvent extracts against Gram-negative and Gram-positive bacteria, respectively. **Conclusion:** We found that Cordia Macleodii Hook extracts showed antibacterial activity were maintained after treating them at higher temperature [75°c, 105°c]. The symbiotic effect from plant extracts against resistant bacteria leads to new options for the cure of dangerous diseases. Cordia Macleodii Hook possesses significant inhibitory and antimicrobial activities against select resistant strains of microbes.

1. INTRODUCTION

Cordia macleodii Hook belongs to family Boraginaceae, is an endangered medicinal plant, commonly known as "Dahiman" or "Dahipalas". It is distributed in moist and dry deciduous forests of India such as Chhattisgarh, Madhya Pradesh, Odisha, Chotanagpur, Maharashtra [1].it is reported for its ethnomedicinal use as a wound healing drug. Earlier study of *Cordia macleodii* has been studied included pharma cognostic evaluation of its leaf and presence of various phytoconstituents viz; alkaloids,

glycosides, phenols, flavonoids, terpenoids, tannin present in extract of *C. macleodii* leaves using various solvents.[2] The biological properties of the solvent extracts of leaf and other part of *cordia macleodii* have attracted us to study the antimicrobial properties against bacterial strains. The aim of this work to find out the optimum condition of temperature and pH at which Cordia macleodii Hook extract is more effective against Gram-positive S. aureus and *Bacillus cereus* and Gram-negative bacteria E coli and Pseudomonas aeruginosa..

2. Experimental Section 2.1 MATERIAL AND METHODS-

Fresh leaves of Cordia macleodii Hook were collected in month of july 2020 from the herbal garden of AKS university satna [M.P.]. the plant was carefully identified and authenticated by specialists in botany department.

2.2 Antibacterial Susceptibility Test (AST)-

The microorganisms employed in the current study were procured from clinical specimen, obtained from the Biotechnology Laboratory at AKS University. The organisms are Escherichia coli ATCC 25932, Staphylococcus aureus, ATCC 29213, Pseudomonas aeruginosa and Bacillus cereus ATCC 6633. The organisms were maintained on Nutrient agar slants at 4°C and sub-cultured for use in testing ofloxin and ampicillin 10 g/mL were used as positive control for the sensitivity test against bacterial strains.

2.3 Preparation of extract-

Fresh leaves were washed properly and air-dried for 2 weeks, then grinded into fine powdered form, by using a grinder, kept preserve in air tight plastic bags, for the further extraction. About 10 gm of powders were added to 300ml of solvents (double distilled water, methanol, ethyl acetate and

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n-hexane) and were exposed to hot extraction method for 6h using Soxhlet method of extraction for the period till the solvent in the siphon tube of extractor become colorless. After that extracts were filtered. The filtration process was carried out using whatman filter paper and filtrate were concentrated by evaporation to make the final volume one – fourth of the original volume and collected at air tight bottles used for further study of impact of temperature and pH in antibacterial activity of plant extract.[3] [4]

3. Quantitative Analysis

For Preliminary Phytochemical test different solvent extracts were tested to detect for the presence of different chemical groups of bioactive compounds as per the methods described in API. Test shows the presences of relatively high levels of diverse degree of flavonoid, alkaloids, steroids terpenoids, tannin, coumarines, etc.

3.1 Antimicrobial activity

Determination of zone of inhibition (ZOI) method

In vitro antimicrobial activity testing was carried out by using Agar disc diffusion method. The zones of inhibition around the disks were measured after 24 h of incubation at 37°C for bacteria. Control experiments were carried out under similar condition by using Ampicillin and ofloxin for antibacterial activity as standard drugs.[5]

Effect of Ph on antibacterial activity of extracts of Cordia macleodii Hook -

To find out the effect of ph the each extracts [water, ethanol, ethyl acetate and chloroform] having concentration 100 g/ml were taken in three set of test tubes and 1N HCl added drop wise until the ph of extract is 2 and 5 [ph is determined by ph meter] increment in pH in every extract is done by using 1N NaOH in three separate test tubes and extracts were then allowed to soaks for one 1 hr. after that acid base treatment the extracts were again neutralized with using 1N HCl and 1N NaOH and then every extracts were tested for antibacterial activity by using agar disc diffusion method.[6]

S.No	Phytochem	Name of	Aqueou	Ethan	Ethyl	Chloro
	icals/secon	test	s extract	ol	acetate	form
	dary			Extra	extract	extract
	metabolite			ct		
1	Carbohy	Molish	++	+		
	dates	Fehling's				

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2	Alkaloids	Mayer's Wagner's Dragen dorff's	++	+	-	
3	Glycoside		-	-	-	-
4	Tannin	FeCI3 Iodine Gelatin	++	+	-	-
5	Flavonoid	Lead acetate	-	+		
6	Saponin	Forth foam		-		
7	Resin	Copper acetate	-	-	++	+
8	Fats and fixed oil	Filter paper		-	++	+
9	Terpenoids,	Salkowski Liberman- burchard		-		++
10	Coumarines,	Alkaline	-	-		

Effect of temperature-

The antibacterial activity of Cordia macleodii Hook leave extract of 10 ml of 200mg/ml concentration of water, ethanol ethyl acetate and chloroform extracts were taken in test tubes and treated at 75°c and 105°c in a water bath for 1 hr. and then tested for antibacterial activity.

Table 1 – Table for antibacterial activity of water extract at different pH and temperature.

Aqueous Extract Zone Of Inhibition (m.m.)								
	Effect of	PH Effe	ect		Effect of Temperature			
Bacterial strain	Origina 1 PH=6.3	PH=2. 0	PH=5 0	PH= 8. 0	Original temperatu re 35°c	75° c	105° c	
Staphyloc occus aureus	14mm	18mm	13m m	15m m	14	13	15.5	
Escherichi a coli	10mm	8mm	9mm	19m m	9	10	11.5	
Pseudomo nas aeruginos a	9mm	12mm	llm m	6mm	7	8	9.0	
Bacillus cereus	7mm	10mm	13m m	14m m	8.5	9	10	

Table 2 – Table for antibacterial activity of ethanol extract at different pH and temperature.

Ethanol Extract Zone Of Inhibition (m.m.)								
	Effect of	f PH Ef	fect	PH=8	Effect of			
				0	temperature			
	Origina	Origina PH=2 PH=5.			Original	75°	105°	
Bacterial	1	.0	0		temperat	С	с	
strain	PH=6.3				ure			
					35°c			
Staphyloco	13mm	llm	12mm	15m	14	15	16.5	
ccus aureus		m		m				
Escherichia	13mm	llm	12mm	14m	12	12.	13	
coli		m		m		5		
Pseudomon	9mm	6mm	7mm	9mm	7	8	10.5	
as								
aeruginosa								
Bacillus	llmm	14m	12mm	10m	11	11.	12.0	
cereus		m		m		5		

 Table 3 – Table for antibacterial activity of ethyl acetate

 extract at different pH and temperature.

Ethyl Acetate Extract Zone Of Inhibition (m.m.)								
	Effect of	PH Effec	Effect of					
					temperature			
	Original	PH=2.0	PH=5.	PH=8.	Origin	75°c	105°	
Bacterial	PH=6.3		0	0	al		с	
strain					tempe			
					rature			
					35°c			
Staphylo	12mm	14mm	13mm	llmm	13	13.5	15.5	
coccus								
aureus								
Escheric	16mm	13mm	14mm	17mm	12	13	13.5	
hia coli								
Pseudom	7mm	10mm	9mm	6mm	6	8	9.5	
onas								
aerugino								
sa								
Bacillus	9mm	7mm	8mm	llmm	9	10	11.5	
cereus								

Table 4 – Table for antibacterial activity of chloroform extract at different pH and temperature.

Chloroform Extract Zone Of Inhibition (m.m.)								
	Effect of	PH Eff	ect	Effect of temperature				
	Original PH=6.3	PH=2 .0	PH=5. 0	PH= 8. 0	Original temperatu re 35°c	75°c	105°c	
Bacteria 1 strain								
Staphylo coccus aureus	12mm	14m m	13mm	llm m	11	11.5	12	
Escheric hia coli	15mm	12m m	13mm	16m m	12	13	13.5	
Pseudo monas aerugin osa	8mm	6mm	7mm	9mm	6	7	8.5	
Bacillus cereus	7mm	10m m	9mm	6mm	8	9	9.5	

4.RESULT AND DISCUSSION

Preliminary screening indicated the presence of different phytochemicals in varying degrees, which suggests the chemical diversity of this plant. Antibacterial activity of ethyl acetate extract expressed a slight increment with increase PH (basic medium) for E.coli, while in case Staphylococcus aureus and *Bacillus cereus* increment in antibacterial activity found at low pH (acidic medium). Although the extracts prevented the growth of both Gram-positive S. aureus and *Bacillus cereus* and Gram-negative bacteria E coli and Pseudomonas aeruginosa. The antibacterial activity (zone of inhibition) of aqueous leaves extract is maximum 19mm higher than those of the other solvent extracts against Gramnegative and Gram-positive bacteria, respectively.

5. CONCLUSION

We found that Cordia Macleodii Hook extracts showed antibacterial activity were maintained after treating them at higher temperature [75°c, 105°c]. The symbiotic effect from plant extracts against resistant bacteria leads to new options for the cure of dangerous diseases. Cordia Macleodii Hook possesses significant inhibitory and antimicrobial activities against select resistant strains of microbes even at different temperature and pH value

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