Phytochemical evaluation and *In-vitro* Antibacterial Activity of *Curcuma longa*

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Abstract:
In the present investigation, we studied on the in-vitro antibacterial activity of *Curcuma longa* extracts in hexane, chloroform, and methanol as well as phytochemical screening. Steroids, terpenoids, glycosides, tannins, alkaloids, phenols, and carbohydrates were all identified in the data. Against the investigated bacterial strains, the chosen plant extracts generated a concentration-dependent zone of inhibition. Compared to gram positive species, the extracts demonstrated more potency against gram negative organisms. When the extracts were at their greatest concentrations, 500 and 250 µg/ml, they demonstrated better activity. At 500 µg doses, the chloroform extract of *Curcuma longa* exhibited superior effectiveness against gram negative bacteria compared to the other two extracts.

Keywords: *Curcuma longa*, Phytochemical screening, *In-vitro* Antibacterial activity.

Introduction

Medicinal plants are a valuable medicinal resource for a wide range of conditions and may be used to create new pharmacological molecules that have the potential to treat a wide range of illnesses. The earliest recorded scientific investigations on plant components' antibacterial qualities date back to the late 1800s[1]. Antimicrobial medications have now drastically altered human destiny in addition to the way infectious diseases are treated. Significant progress in antimicrobial chemotherapy led to the unduly optimistic belief that infectious illnesses will be eradicated in the near future. In actuality, though, re-emerging and newly developing infectious illnesses have left us counterattacking with infections. Drug-resistant organism infections continue to be a significant and challenging issue in clinical practise. The shortcomings of currently marketed medications encourage the search for novel pharmacotherapeutic agents in medicinal plants[2]. Consequently, we attempted to ascertain *Curcuma longa's* antimicrobial activity in hexane, chloroform, and methanol in the current study, and these extracts were discovered to exhibit strong antibacterial activity.
Curcuma longa is a perennial herb that has funnel-shaped blooms and oblong, pointed leaves. It is a member of the Zingiberaceae family and its rhizome is used medicinally. It is widely grown in China, Asia, and other tropical nations[3].

Materials and Methods

Chemicals:

Muller Hinton agar medium was obtained from Sisco Research Laboratories Pvt Ltd. in Mumbai. Analytical grade reagents and chemicals were all utilised.

Test Organisms:

There were five kinds of microorganisms used in the study. The National Collection of Industrial Micro Organisms (NCIM), located in Pune, is where the bacterial species were acquired. Using shakers in individual culture tubes for each species, the bacterial species were kept alive in the nutrient broth medium. Of the five, one is Gram positive (*S. aureus*) while the other four are Gram negative (*E. Coli, K. pneumoniae, Xanthomonas, and P. mirabilis*).

Culture Media:

Muller-Hinton Agar medium (Solid and Broth) was used to test for antibacterial activity. Nutrient both were employed to sustain the bacterial species.

Preparation of Extracts:

The plant material was gathered at Visakhapatnam, Andhra Pradesh. Plant material that had just been harvested was shade-dried and then ground into a coarse powder. The powdered substance was extracted separately in a Soxhlet

apparatus using hexane, chloroform, and methanol for six hours in succession. Rota-vapor was then used to concentrate the methanol to dryness under vacuum.

Phytochemical Analysis:
Using conventional techniques [4-6], phytochemical investigations were conducted on Curcuma longa extracts in hexane, chloroform, and methanol to identify the presence of several phytochemical elements such as steroids, terpenoids, tannins, flavonoids, saponins, glycosides, and amino acids.

Antibacterial Activity:
The drug potency cylinder plate assay measures the width of the zone where microbiological growth is inhibited around cylinders, or cups, that contain different test extract dilutions[7]. 50, 100, 250, and 500 µg/ml were the concentrations at which the dilutions were made. The 4 mm diameter cups were prepared using a sterile borer and placed in an agar medium containing 0.1 ml of inoculums and microorganisms. Using the spread plate method, these cups were distributed around the agar plate. Using a micropipette, precisely measured (0.05 ml) solutions of each concentration and reference standards were added to the cups. For two hours, all of the plates were stored in a refrigerator between 2 and 8°C to ensure that the test compounds and standards diffused properly. They were then incubated for 24 hours at 37°C. Antibacterial activity was shown by the existence of a distinct zone of inhibition surrounding the cup, regardless of size.

Results and Discussion

Phytochemical screening:
Different Curcuma longa extracts were subjected to a qualitative phytochemical screening process, which found the presence of steroids, terpenoids, glycosides, tannins, alkaloids, phenols, and carbohydrates. However, no extract included saponins, flavonoids, quinines, amino acids, or oils. Table 1 displayed the phytochemical screening findings.

Evaluation of antibacterial activity:
Chloroform and methanol extracts from *Curcuma longa* have demonstrated noteworthy antibacterial activity when compared to standard medication across all evaluated extracts. Compared to gram positive bacteria, the extracts demonstrated a better zone of inhibition against gram negative bacteria. At a dosage of 50 µg/ml, *Curcuma longa* extracts in hexane and methanol did not exhibit a zone of inhibition against several tested bacterial species. Comparing chloroform extract to other studied extracts, it demonstrated a good zone of inhibition at all dosages. At a dosage of 500 µg/ml, the chloroform extract demonstrated the largest zone of inhibition against both *P.mirabilis* and *Xanthomonas*. Table 2 displayed the antibacterial activity data.

Table 1: Phytochemical constituents present in different extracts of *Curcuma longa*

Name of the Phytochemicals	Extracts of *Curcuma longa*		
	Hexane extract	Chloroform extract	Methanol extract
Phenols	+	+	+
Phytosterols	+	+	+
Terpenoids	+	+	+
Glycosides	+	+	+
Saponins	-	-	-
Flavonoids	-	-	-
Tannins	-	+	+
Carbohydrates	+	+	+
Alkaloids	+	+	+
Amino acids	-	-	-
Oils	-	-	-
Quinones	-	-	-

+ = Present, – = Absent

Table 2: Antibacterial activity of *Curcuma longa* extracts

Name of the *Curcuma longa* Extract	**dose** (µg/cup)	**zone of inhibition[#] (diameter in mm)**				
		E.c	***Xa***	***P.m***	***S.a***	***K.p***
Hexane	50	-	5	-	-	-
	100	-	5	-	6	5
	250	-	6	6	6	6
	500	-	8	6	6	6
Chloroform	50	5	6	6	7	5
	100	7	6	7	8	6
	250	8	8	8	8	6

	500	8	9	8	8	6
Methanol	50	6	7	7	-	5
	100	7	8	7	-	5
	250	7	9	8	-	6
	500	8	9	8	-	6
Chloramphenicol		27	20	17	25	24
DMSO	-	-	-	-	-	-

X.c=Xanthomonas campestris; *P.m= P.mirabilis, S.a=Streptococcus aureus; E.c=Escherichia coli, K.p= Klebsiella pneumonia;* -No activity.

#Values are the average of triplicate; Includes the cup diameter (4mm)

Conclusion

The results unequivocally demonstrated that Curcuma longa's hexane, chloroform, and methanol extracts had strong antibacterial activity. The chloroform extract had the greatest zone of inhibition and the strongest activity against every tested organism of all the tested extracts.

Acknowledgment

The authors were thankful to Dr.V.S.Krishna Govt. College, Visakhapatnam for providing necessary laboratory facilities to carry out present research work.

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