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**Original Research Article** 

# Curcuma longa leaves exhibits a potential antioxidant, antibacterial and immunomodulating properties

Ritwiz S Bhardwaj<sup>1</sup>, Kusha S Bhardwaj<sup>1</sup>, Ranjeet D<sup>1</sup>, Ganesh N.<sup>1</sup>\*

#### \*Corresponding author:

#### Ganesh N.

 Department of Research, Jawaharlal Nehru Cancer Hospital & Research Centre, Bhopal.
 <sup>®</sup>Both have contributed equally.

## Abstract

The use of herbs to treat illness has its roots in an ancient holistic healing tradition that originated in Asia more than 3000 years ago. Plants rich in a wide variety of secondary metabolites like tannins, terpanoids, alkaloids, alkaloids and flavonoids, which have been found in vitro to have antimicrobial and antioxidant properties. Turmeric, a representative of plant genus Curcuma, is the member of ginger family, Zingiberaceae. There are five different varieties we have taken Curcuma amada, Curcuma aromatica, Curcuma longa, Curcuma zeodaria, Curcuma caesia. The active substance of turmeric is the polyphenol curcumin, also known as natural yellow 3. It exists at atleast 2 tautomeric forms, keto and enol. It has aroma and flavour and it is used in cosmetics and food additive besides medicinal properties. The present study is a new attempt in leaves segments of the plant Curcuma longa to prove the medicinal potential of the whole plant. This study also includes the comparison of the different species of Curcuma. The active constituents of turmeric are the flavonoid curcumin and volatile oils including turmeron, atlanton and gingibaron. Our experiments are based on 50% methanolic extract of Curcuma leaves to perform anti bacterial, anti oxidant and immunomodulating properties. The antibacterial activity has been done by the help of Disc Diffusion Susceptibility Testing (Kirby-Bauer Method) against four species of bacteria which include Bacillus cereus, Diploccocus pneumoneae, Streptococcus pyrogens & Micrococcus glutamicus. Antioxidant activity has been detected by the Fenton's reaction. Immunomodulation activity has been seen in curcuma leaves. The results of the present study reveal that all the five species of curcuma exhibit an antioxidant activity and antibacterial activity on different concentration of leave extract. The extract was found to increase the phagocytic activity of macrophages against yeast cells. However, the best activity has been revealed by curcuma longa among the all species of curcuma. In near future the pharmaceuticals studies can be extended to prove its tremendous medicinal properties in case of drug preparation for immune cell properties enhancement and to prove its potential value unlike Curcuma rhizome.

**Keywords**—Curcuma Longa, Curcuma Amada, Curcuma Aromatica, Immounomodulation, Curcumin.

# Introduction

any of the 20<sup>th</sup> century practitioners of western medicine, incorporating herbal remedies in healing practices, such as traditional Chinese medicine, Japanese kampo and Indian ayurveda, which are rapidly gaining acceptance in the west development The of science of phytopharmaceuticals and the hopes of the remedies chronic diseases generated in enthusiasm in the researchers to develop herbal medicines(1). The driving factor for the renewed interest in plant antimicrobials in the past 20 vears has been the rapid rate of species of extinction (2). This is the feeling among naturalproducts chemists and microbiologists alike that the multitude of potentially useful phytochemical which could be synthesized structures. chemically, is at risk of being lost irretrievably (3). There is a scientific discipline known as ethnobotany, whose goal is to utilize the impressive array of knowledge assembled by indigenous peoples about the plant and animal products they have used to maintain health (4,5,6,7). Plants have an almost limitless ability to synthesize aromatic substances, most of the phenols or their oxygen-substituted derivatives (8). They are widely used as ingredients in dietary supplements used for health purposes such as attempting to prevent cancer and heart diseases. The term antioxidant (also anti-oxygen) originally referred specifically to a chemical that pretended the consumption of molecular oxygen (9). The possible mechanisms of action of antioxidants were first explored thoroughly by

Moreau and Dufraisse (1926), who recognized that a substance with antioxidative activity is likely to be one that is itself a target for oxidation (10). The normal antioxidant defence system in biological systems consists of both enzymatic and non enzymatic systems. Although both are important in biological systems, the nonenzymatic anti-oxidant systems which include substances such as tocopherol (Vitamin E), ascorbic acid (Vitamin A precursor), uric acid and plasma proteins such as albumin. ceruloplasmin, transferrin, metelothionein, etc. (11).Water and fat soluble extracts of turmeric and its curcumin component exhibit strong antioxidant activity, comparable to vitamins C and E(Toda S et al 1985)(12). A review of recent reports on immunomodulatory effects attributed to mind early studies are quite challenging (Alper J.1998, Amoros M .et al 1992)(13-14)Many herbal preparations alter immune function and had an amazing array of immunomodulatory effects attributed to them (15). Modulation of immune functions by using medicinal plants and their products as a possible therapeutic measure has become fundamental principles of theraputical approach for the treatment of many ailments and diseases. Immunomodulation is a change in the body's immune system that caused by agents leads to activate or suppress its function(16). Thus, medicinal herbs have played a major role in the development of modern medicine and continue to be widely used in their original form(17).

	B. cereus	D. pneumoneae	M. glutamicus	S. pyogens
Curcuma aromatica	$14.32 \pm 0.39$	$13.25 \pm 0.29$	$12.32 \pm 0.42$	$12.21 \pm 0.34$
Curcuma caesia	$14.95 \pm 0.71$	$14.65 \pm 0.71$	$12.50 \pm 0.24$	$13.71 \pm 0.41$
Curcuma longa	$12.50 \pm 0.41$	$13.75 \pm 0.21$	$12.78 \pm 0.61$	$13.5 \pm 0.71$
Curcuma amada	$14.75 \pm 0.31$	$12.75 \pm 0.21$	$12.77 \pm 0.41$	$13.55 \pm 0.41$
Curcuma zedoary	$14.78 \pm 0.41$	$12.80 \pm 0.21$	$12.94 \pm 0.41$	$13.80 \pm 0.31$

Table 1. Comparative table of five different Curcuma leaf extracts (concentration 100%) against different microorganisms.

# **Material And Method**

Following steps were used in the method:-

- 1. Leaves of Curcuma Species were collected locally (Bhopal) in India. They were dried, powdered finely, weighted and soaked with solvent (50% methanol).
- 2. A powder bed and solvent layer was separated in separating funnel and both layers were agitated at regular interval for 72 hours
- 3. The filtrate was eluted and fresh solvent was added again to the pretreated powder.
- 4. Ether defatted filtrate was concentrated at 40° C to dry , powder and finally packed in an air tight container.
- 5. Antibacterial activity of the powder with its different concentration (25%, 50%75%, and 100%) was assessed against Diplococcus pneumoneae, Streptococcus pyogens, Micrococcus glutamicus, and Bacillus cereus using the Disc Diffusion Susceptibility Method (Kirby-Bauer Method) (18).
- 6. Antioxidant activity of the test powder was comapared with DMSO (positive control) against PBS (blank) by making dilution series and optical density of the final colour was measured on spectrophotometery at 532 nm.
- 7. Immunomodulation was measured by macrophage activity. Doses were administrated for 5 days, peritoneal fluid was isolated and centrifuged at 1000rpm, pellet was suspended in RPMI media, culture were co-incubated for 24 hrs with heat inactivated veast. After 1hr it was centrifuged at 1000rpm for 20 mins, smear were prepared and fix it with fixative, air dried and stained, observed under light microscope. The % age of inhibition of yeast digest /macrophage index was calculated and graph was plotted. The data was analysed by ANOVA.

Table 2.	Antioxidant activity of five different species of
Curcuma	a longa leaves.

Curcuma longa leaves.						
Antioxidant activit	ty of Curcu	ma amada				
Concentration(µg/ml)	DMSO	Curcuma				
10	$70 \pm 0.38$	$58.7 \pm 0.35$				
25	$75.2 \pm 0.56$	$64.0\pm0.40$				
50	$79.4 \pm 0.45$	$69.1 \pm 0.52$				
150	$87.2 \pm 0.29$	$72.0 \pm 0.52$				
250	$94.5 \pm 0.36$	$80.3 \pm 0.035$				
300	$91.1 \pm 0.42$	$69.5 \pm 0.26$				
500	$92.9 \pm 0.58$	$64.3 \pm 0.41$				
Antioxidant activ	ity of Curcur	na longa				
Concentration(µg/ml)	DMSO	Curcuma				
10	$70 \pm 0.38$	$59.24 \pm 0.32$				
25	$75.2 \pm 0.56$	$60.24 \pm 0.24$				
50	$79.4 \pm 0.45$	$65.34 \pm 0.3$				
150	$87.2 \pm 0.29$	$70.26 \pm 0.75$				
250	$94.5 \pm 0.36$	$81.58 \pm 0.36$				
300	$91.1 \pm 0.42$	$79.0 \pm 0.05$				
500	$92.9 \pm 0.58$	$78.42 \pm 0.06$				
Antioxidant activity	of Curcuma	a aromatica				
Concentration(µg/ml)	DMSO	Curcuma				
10	$70 \pm 0.38$	$56.24 \pm 0.32$				
25	$75.2 \pm 0.56$	$59.24 \pm 0.24$				
50	$79.4 \pm 0.45$	$61.34 \pm 0.3$				
150	$87.2 \pm 0.29$	$66.26 \pm 0.75$				
250	$94.5 \pm 0.36$	$75.58 \pm 0.36$				
300	$91.1 \pm 0.42$	$69.0 \pm 0.05$				
500	$92.9 \pm 0.58$	$77.42 \pm 0.06$				
Antioxidant activi	ty of Curcur					
Concentration(µg/ml)	DMSO	Curcuma				
10	$70 \pm 0.38$	$47.24 \pm 0.25$				
25	$75.2 \pm 0.56$	$51.64 \pm 0.28$				
50	$79.4 \pm 0.45$	$54.44 \pm 0.45$				
150	$87.2 \pm 0.29$	$62.26 \pm 0.29$				
250	$94.5 \pm 0.36$	$71.58 \pm 0.41$				
300	$91.1 \pm 0.42$	$73.3 \pm 0.45$				
500	$92.9 \pm 0.58$	$65.6 \pm 0.22$				
Antioxidant activit						
Concentration(µg/ml)	DMSO	Curcuma				
10	$70 \pm 0.38$	$51.64 \pm 0.25$				
25	$75.2 \pm 0.56$	$51.44 \pm 0.65$				
50	$79.4 \pm 0.45$	$61.2 \pm 0.29$				
150	$87.2 \pm 0.29$	$65.46 \pm 0.52$				
250	$94.5 \pm 0.36$	$71.58 \pm 0.35$				
300	$91.1 \pm 0.42$	64.2± 0.39				
500	$92.9 \pm 0.58$	$70.52 \pm 0.22$				

#### **Observations and results** Antimicrobial activity-

Our present study shows that antimicrobial activity of 50% methanolic extract of Curcuma amada leaves against Bacillus cereus is the best in 100% concentration in between the 8<sup>th</sup>-10<sup>th</sup> hrs of incubation, as 14.32mm zone of inhibition has observed, although 75% concentration been presented mild effect (zone of inhibition = 12.32mm). In Curcuma longa, 100% conc. of leaf extract shows maximum activity with maximum zone of inhibition as 14.95mm between 10<sup>th</sup>-12<sup>th</sup> hrs of incubation, although 75% conc. is having mild effect as 12.11mm zone of inhibition. Curcuma aromatica leaves are having best effect at100% con between 10<sup>th</sup>-12<sup>th</sup> hrs as 12.50mm zone of inhibition, although 75% con is having mild effect as 11.50mm zone of inhibition.. Curcuma caesia, at 100% conc. shows 14.75mm zone of inhibition in 10<sup>th</sup>-12<sup>th</sup> hrs, although 75% conc. shows 13.75mm zone of inhibition. Curcuma zedoary at 100% concentration between 10<sup>th</sup>-12<sup>th</sup> hrs shows maximum effect with zone of inhibition of 14.78mm and has a mild effect as 13.60mm zone of inhibition.

Table 3. Showing the % of yeast digestion of differentCurcuma leaf species extract.

S. No	Extract	Total no of monocytes	Macrophage showing yeast digestion	% of yeast digestion
1.	Curcuma amada	100	18	18
2.	Curcuma longa	100	30	30
3.	Curcuma aromatica	100	25	25
4.	Curcuma caesia	100	15	15
5.	Curcuma zeodary	100	10	10

Curcuma amada leaves against Diplococcus pneumoneae is the best in 100% concentration in between the  $8^{th}$ -10<sup>th</sup> hrs of incubation, as 13.25mm zone of inhibition has been observed, although 75% concentration presented mild effect

(zone of inhibition = 12.12mm). In Curcuma longa, 100% conc. of leaf extract shows maximum activity with maximum zone of inhibition as 14.65mm between 10<sup>th</sup>-12<sup>th</sup> hrs of incubation, although 75% conc. is having mild effect as 12.72mm zone of inhibition. Curcuma aromatica leaves are having best effect at 100% con between 10<sup>th</sup>-12<sup>th</sup> hrs as 13.75mm zone of inhibition has been observed, although 75% con is having mild effect as 12.26mm zone of inhibition.. Curcuma caesia, at 100% conc. shows 12.75mm zone of inhibition in 10<sup>th</sup>-12<sup>th</sup> hrs, although 75% conc. shows 10.50mm zone of Curcuma zedoary at inhibition. 100% concentration between 10<sup>th</sup>-12<sup>th</sup> hrs shows maximum effect with zone of inhibition of 12.80mm and has a mild effect as 12.65mm zone of inhibition

S. No.	Extract	No. of sample	Mean S.D
1.	Control	4	Nil
2.	Curcuma amada	4	$18.25 \pm 2.22$
3.	Curcuma longa	4	$30.33\pm0.54$
4.	Curcuma aromatica	4	$25.12 \pm 1.36$
5.	Curcuma caesia	4	$15.06\pm0.82$
6.	Curcuma zeodary	4	$10.36\pm0.63$

Table 4. Showing the microphage activity of differentsample.

Curcuma amada leaves against Streptococcus pyogens is the best in 100% concentration in between the 8<sup>th</sup>-10<sup>th</sup> hrs of incubation, as 12.21mm zone of inhibition has been observed, although 75% concentration presented mild effect (zone of inhibition = 11.31mm). In Curcuma longa, 100% conc. of leaf extract shows maximum activity with maximum zone of inhibition as 13.70mm between 10<sup>th</sup>-12<sup>th</sup> hrs of incubation, although 75% conc. is having mild effect as 12.71mm zone of inhibition. Curcuma aromatica leaves are having best effect at 100% con between 10<sup>th</sup>-12<sup>th</sup> hrs as 13.50 mm zone of inhibition has been observed, although 75% conc. is having mild effect as 12.50 mm zone of inhibition. Curcuma caesia, at 100% conc. shows 13.55 mm zone of inhibition in 10<sup>th</sup>-12<sup>th</sup> hrs. although 75% conc. shows 12.65mm zone of Curcuma zedoary at 100% inhibition.

concentration between 10<sup>th</sup>-12<sup>th</sup> hrs shows maximum effect with zone of inhibition of 13.80 mm and has a mild effect as 12.mm zone of inhibition.

Curcuma amada leaves against Micrococcus glutamicus is the best in 100% concentration in between the  $10^{\text{th}}$  -12th hrs of incubation, as 12.32mm zone of inhibition has been observed, although 75% concentration presented mild effect (zone of inhibition = 11.31mm). In Curcuma longa, 100% conc. of leaf extract shows maximum activity with maximum zone of inhibition as 12.50mm between  $10^{\text{th}}$ -12<sup>th</sup> hrs of

incubation, although 75% conc. is having mild effect as 11.40mm zone of inhibition. Curcuma aromatica leaves are having best effect at 100% con between 10<sup>th</sup>-12<sup>th</sup> hrs as 12.78mm zone of inhibition has been observed, although 75% con is having mild effect as 11.60mm zone of inhibition. Curcuma caesia, at 100% conc. shows 12.77mm zone of inhibition in 10<sup>th</sup>-12<sup>th</sup> hrs. although 75% conc. shows 11.75mm zone of inhibition. Curcuma zedoary at 100% concentration between 10<sup>th</sup>-12<sup>th</sup> hrs shows maximum effect with zone of inhibition of 12.94mm and has a mild effect as 11.77mm zone of inhibition.

Table 5. Antibacterial activity of Curcuma amada against four bacteria.

S. No.	Conc.	Zone of inhibition at different incubation Period{in mm}					
		4hrs	6hrs	8hrs	10hrs	12hrs	
		Antibacteri	al activity of Di	iplococcus pneun	ioneae		
1	75%	$9.25 \pm 0.28$	$9.32 \pm 0.24$	$12.12 \pm 0.25$	$12.12 \pm 0.25$	$12.12 \pm 0.25$	
2	100%	$10.25 \pm 0.28$	$11.12 \pm 0.25$	$13.25 \pm 0.29$	$13.25 \pm 0.29$	$13.25 \pm 0.29$	
		Antiba	cterial activity of	of Bacillus cereu	IS		
1	75%	$8.37 \pm 0.48$	$10.12 \pm 0.25$	$12.37 \pm 0.48$	$12.37 \pm 0.48$	$12.37 \pm 0.48$	
2	100%	9 0±.41	$11.35 \pm 0.51$	14.32 ±0.39	$14.32 \pm 0.39$	$14.32 \pm 0.39$	
		Antibacter	rial activity of S	Streptococcus py	ogens		
1	75%	$8.28 \pm 0.35$	$9 \pm 0.31$	11.22 0.26	$11.31 \pm 0.29$	$11.31 \pm 0.19$	
2	100%	$8.48 \pm 0.71$	$10.27 \pm 0.31$	$12.21 \pm 0.24$	$12.21 \pm 0.34$	$12.21 \pm 0.24$	
	Antibacterial activity of Micrococcus glutamicus						
1	75%	$8.12 \pm 0.35$	$9.23 \pm 0.34$	$11.25 \pm 0.52$	$11.31 \pm 0.44$	$11.31 \pm 0.44$	
2	100%	8.65 ± 0.33	$10.41 \pm 0.28$	$12.52 \pm 0.71$	$12.32 \pm 0.42$	12.32 ±0.52	

Table 6 Antibacterial activity of Curcuma longa against four bacteria.

S. No.	Conc.	Zone of inhibition at different incubation Period{in mm}						
		4hrs	6hrs	8hrs	10hrs	12hrs		
		Antibacte	erial activity of D	iplococcus pneur	noneae			
1	75%	$9.23 \pm 0.26$	$9.26 \pm 0.21$	12.41± .61	$12.72 \pm 0.31$	$12.72 \pm 0.45$		
2	100%	$10.35 \pm 0.24$	$11.50 \pm 0.31$	$14.20 \pm 0.41$	$14.65 \pm 0.52$	$14.65 \pm 0.71$		
		Ant	ibacterial activity	of Bacillus cerei	15			
1	75%	$9.20 \pm 0.55$	$10.25 \pm 0.31$	$12.62 \pm 0.61$	12.11 ±0.36	$12.11 \pm 0.71$		
2	100%	$9.21 \pm 0.31$	$11.50 \pm 0.41$	14.32 ±0.71	$14.95 \pm 0.41$	$14.95\pm0.71$		
		Antibac	terial activity of	Streptococcus py	ogens			
1	75%	$8.65 \pm 0.72$	$9.0 \pm 0.31$	$11.50 \pm 0.51$	$12.71 \pm 0.71$	$12.71 \pm 0.61$		
2	100%	$8.11 \pm 0.21$	$10.0 \pm 0.51$	$13.70 \pm 0.31$	$13.70 \pm 0.37$	$13.71 \pm 0.41$		
	Antibacterial activity of Micrococcus glutamicus							
1	75%	$9.25 \pm 0.62$	$10.21 \pm 0.41$	$11.11 \pm 0.44$	$11.40 \pm 0.31$	$11.40 \pm 0.71$		
2	100%	$9.26 \pm 0.71$	$11.0 \pm 0.31$	$12.25 \pm 0.46$	$12.50 \pm 0.75$	$12.40 \pm 0.24$		

# **Antioxidant Activity**

The five different species of Curcuma leaves extract in different concentration show varying antioxidant activity.

**Curcuma amada:-**The % inhibition of Curcuma amada is found to be the highest at  $250\mu$ g/ml, i.e- $80.3\pm0.35\%$  and % inhibition of Curcuma amada is best responded than  $50\mu$ g/ml % inhibition of DMSO.

**Curcuma longa:** - The % inhibition of Curcuma longa is found to be the highest at  $250\mu$ g/ml, i.e- $81.58\pm0.36\%$  and % inhibition of Curcuma longa is best responded than  $50\mu$ g/ml % inhibition of DMSO.

**Curcuma aromatica:** - The % inhibition of Curcuma aromatica is found to be the highest at  $250\mu$ g/ml, i.e- $77.42\pm0.06\%$  and % inhibition of Curcuma aromatica is best responded than  $50\mu$ g/ml % inhibition of DMSO.

**Curcuma caesia:** - The % inhibition of Curcuma caesia is found to be the highest at  $250\mu$ g/ml, i.e-73.3±0.45% and % inhibition of Curcuma caesia is best responded than  $50\mu$ g/ml % inhibition of DMSO.

**Curcuma zedoary:** - The % inhibition of Curcuma zedoary is found to be the highest at  $250\mu$ g/ml, i.e- $71.58\pm0.35\%$  and % inhibition of Curcuma zedoary is best responded than  $50\mu$ g/ml % inhibition of DMSO.

 Table 7. Antibacterial activity of Curcuma aromatica against four bacteria.

S. No.	Conc.	Zone of inhib	Zone of inhibition at different incubation Period{in mm}					
		4hrs	6hrs	8hrs	10hrs	12hrs		
		Antibacterial	activity of Dip	lococcus pneum	oneae			
1	75%	$9.28 \pm 0.41$	$10.50 \pm 0.11$	$11.11 \pm 0.41$	$12.26 \pm 0.71$	$12.26 \pm 0.64$		
2	100%	$9.60 \pm 0.27$	$10.11 \pm 0.41$	$12.26 \pm 0.31$	$13.75 \pm 0.41$	$13.75 \pm 0.21$		
	Antibacterial activity of Bacillus cereus							
1	75%	$9.27 \pm 0.31$	$10.62 \pm 0.41$	$11.11 \pm 0.71$	$11.50 \pm 0.74$	$11.50 \pm 0.41$		
2	100%	$9.50 \pm 0.62$	$11.65 \pm 0.84$	$12.31 \pm 0.64$	$12.50 \pm 0.31$	$12.50 \pm 0.41$		
		Antibacteria	al activity of St	reptococcus pyo	ogens			
1	75%	$8.62 \pm 0.71$	$9.24 \pm 0.31$	$11.10 \pm 0.41$	$12.50 \pm 0.71$	$12.50 \pm 0.31$		
2	100%	$8.20 \pm 0.62$	$10.26 \pm 0.61$	$13.0 \pm 0.41$	$13.50 \pm 0.21$	$13.50 \pm 0.71$		
	Antibacterial activity of Micrococcus glutamicus							
1	75%	$8.70 \pm 0.27$	$10.22 \pm 0.31$	$11.20 \pm 0.44$	$11.60 \pm 0.21$	$11.60 \pm 0.29$		
2	100%	$8.30 \pm 0.61$	$10.26 \pm 0.34$	$12.10 \pm 0.21$	$12.78 \pm 0.31$	$12.78 \pm 0.61$		

#### Table 8. Antibacterial activity of Curcuma caesia against four bacteria.

S. No.	Conc.	Zone of inhibition at different incubation Period{in mm}						
		4hrs	6hrs	8hrs	10hrs	12hrs		
	Antibacterial activity of Diplococcus pneumoneae							
1	75%	$9.30 \pm 0.22$	$9.11 \pm 0.25$	$10.50 \pm 0.34$	$10.50 \pm 0.21$	$10.50 \pm 0.41$		
2	100%	$10.20 \pm 0.41$	$11.52 \pm 0.51$	$12.25 \pm 0.46$	$12.75 \pm 0.25$	$12.75 \pm 0.21$		
		Antib	acterial activity	of Bacillus cer	eus			
1	75%	$9.35 \pm 0.65$	$10.36 \pm 0.65$	$13.65 \pm 0.69$	$13.75 \pm 0.71$	$13.75 \pm 0.21$		
2	100%	$9.25 \pm 0.31$	$13.50 \pm 0.58$	$14.11 \pm 0.64$	$14.75 \pm 0.41$	$14.75 \pm 0.31$		
		Antibacte	erial activity of	Streptococcus p	yogens			
1	75%	$7.31 \pm 0.31$	$9.31 \pm 0.61$	$11.50 \pm 0.31$	$12.65 \pm 0.65$	$12.65 \pm 0.21$		
2	100%	$8.32 \pm 0.25$	$10.05 \pm 0.21$	$13.25 \pm 0.44$	$13.55 \pm 0.21$	$13.55 \pm 0.41$		
	Antibacterial activity of Micrococcus glutamicus							
1	75%	$8.35 \pm 0.31$	$10.50 \pm 0.41$	$11.64 \pm 0.21$	$11.75 \pm 0.34$	$11.75 \pm 0.61$		
2	100%	8.25 ± 0.68	$10.11 \pm 0.35$	$12.25 \pm 0.24$	$12.77 \pm 0.51$	$12.77 \pm 0.41$		

## Immunomodulatory activity

Fight against the disease is well define by immune system .when antigen over produce, the immunity reduced and so we required a modulator which possesses immunomodulating factor/character. Our studies have shown that the Curcuma longa shows maximum 30% macrophage yeast digestion, Curcuma amada, Curcuma aromatica, Curcuma caesia, Curcuma zedoary has shown the 18%, 25%, 15% and 10% macrophage veast digestion respectively. Immunomodulating activity was also asseted by serum electrophoresis and followed by serum protein bands. It was found that albumin, alpha-1, alpha-2, beta and gamma globulin is maintained and slightly raised in all the five samples as compared with normal control.

## Conclusion

To anticipated the antibacterial activity of five different species of Curcuma leaves, our data revealed the sensitizing quality of extract against Bacillus cereus, Diplococcus pneumoneae, Streptococcus pyrogens , and Micrococcus glutamicus 75% and 100% concentration were having good activity, showing zone of inhibition from 12mm to 14mm in 6<sup>th</sup> to 12<sup>th</sup> hour time interval.

As Streptococcus pyrogens & Micrococcus glutamicus are gram positive bacteria and Bacillus cereus & Diplococcus pneumoneae are gram negative, our extract is having good activity against all four. So the extract can be a good alternative for the broad spectrum antibiotics in near future. Although work is required to be commenced on the broad range of microbial strain and use of specific isolated constituent for future studies.

The % TBARS (Thio Bartutric Acid Reactive Substance) inhibition of five different species of Curcuma leaves has compared with known antioxidant DMSOby different taking concentration  $5\mu g/ml$  to  $500 \mu g/ml$  the % inhibition of Curcuma amada was shown by maximum conc. i.e-250  $\mu$ g/ml where as in case of known antioxidant DMSO% inhibition was obtained in 50  $\mu$ g/ml, on the other hand % inhibition of Curcuma longa was shown by maximum conc. i.e-250  $\mu$ g/ml where as in case of known antioxidant DMSO% inhibition was obtained in 50 µg/ml, in Curcuma aromatica % inhibition of Curcuma aromatica was shown by maximum conc. i.e-250 µg/ml where as in case of DMSO% inhibition was known antioxidant obtained in 50 µg/ml, in Curcuma caesia % inhibition of Curcuma aromatica was shown by maximum conc. i.e-300  $\mu$ g/ml where as in case of known antioxidant DMSO% inhibition was obtained in 25 µg/ml, in case of Curcuma zedoary % inhibition of Curcuma zedoary was shown by maximum conc. i.e-250 µg/ml where as in case of known antioxidant DMSO% inhibition was obtained in 25  $\mu$ g/ml, on the other hand. So being an antioxidant the extract can be used as easily accessible source of natural antioxidants and as a possible food supplement or in pharmaceutical industry.

In present studies the extract targeted has been found to increase phagocytic activity of macrophage against yeast. It also activates the macrophage to secret a large number of molecules such as substance including a colony stimulating factor and interleukin.

Comparing the electrophoretic bands of the sample and the control, albumin, alpha-1, alpha-2, beta and gamma globulin was found to be raised /modulated in all the five samples.

This study clearly revealed, that the extract has a very good immunomodulating potential in application to the animal model system.

S. No.	Conc.	Zone of inhibit	Zone of inhibition at different incubation Period{in mm}					
		4hrs	6hrs	8hrs	10hrs	12hrs		
		Antibacterial	activity of Diple	ococcus pneum	oneae			
1	75%	$8.20 \pm 0.26$	$9.60 \pm 0.71$	$12.51 \pm 0.64$	$12.65 \pm 0.58$	$12.65 \pm 0.24$		
2	100%	$8.65 \pm 0.58$	$10.20 \pm 0.64$	$12.25 \pm 0.46$	$12.80 \pm 0.35$	$12.80 \pm 0.21$		
	Antibacterial activity of Bacillus cereus							
1	75%	$9.80 \pm 0.61$	10.10 ±0.27	$13.20 \pm 0.31$	$13.60 \pm 0.41$	$13.60 \pm 0.50$		
2	100%	$9.60 \pm 0.31$	13.20 0.28	$14.50 \pm 0.21$	$14.78\pm0.61$	$14.78\pm0.41$		
		Antibacteria	al activity of Str	eptococcus pyo	gens			
1	75%	7.29± 0.38	9.31± 0.52	$11.11 \pm 0.41$	$12.0 \pm 0.21$	$12.0 \pm 0.41$		
2	100%	$8.0 \pm 0.18$	$10.80 \pm 0.21$	$13.36 \pm 0.54$	$13.80 \pm 0.28$	13.80 0.31		
	Antibacterial activity of Micrococcus glutamicus							
1	75%	$8.52 \pm 0.25$	$10.11 \pm 0.21$	$11.54 \pm 0.24$	$11.77 \pm 0.41$	$11.77 \pm 0.28$		
2	100%	$8.26 \pm 0.38$	$10.36 \pm 0.41$	$12.58 \pm 0.71$	$12.94 \pm 0.29$	$12.94 \pm 0.41$		

 Table 9. Antibacterial activity of Curcuma zeodary against four bacteria.

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