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Original Article

Assessment of Phytochemical and Antiulcer activity of *Curcuma longa* leaves

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INTRODUCTION

Ulcer is a common disorder of the GIT system, which causes much discomfort to patients, disrupting their daily routines and causes mental agony [1]. Peptic ulcer is defined as a break off in the continuity of the mucosa of stomach or duodenum because of some medications like non-steroidal anti-inflammatory drugs (NSAIDS), gastric acids and pepsin finally causes lesions in intestinal mucosa [2].

The pathogenesis of peptic ulcer disease includes a complex imbalance between gastric offensive factors like acid, pepsin secretion, Helicobacter pylori (H. pylori), bile salts, ethanol, some medications like NSAIDS, lipid peroxidation, nitric oxide (NO) and defensive mucosal factors like prostaglandins (PG's), gastric mucus, cellular renovation [3]. Peptic ulcer is one of the world's major gastro-intestinal disorders, embracing both gastric and duodenal ulcers, and affecting 10% of the world population.

The patho-physiology of peptic disease is attributed to the imbalance between aggressive factors like acid, pepsin, and Helicobacter infection [4]. The suppression of stomach acid secretions is a key therapeutic target for ulcers and includes the use of antacids, specific muscarinic (M1) receptor antagonists, targeting gastrin receptors and histamine (H2) receptors [5], and the use of proton pump inhibitors

ABSTRACT: Peptic ulcer is one of the world's major gastro-intestinal disorders,

embracing both gastric and duodenal ulcers, and affecting 10% of the world population.

a common disorder of the GIT system, which causes much discomfort to patients,

disrupting their daily routines and causes mental agony. Here, present study was carried out to investigate antiulcer activity of methanol extract of Curcuma Longa

leaves (family Zingiberaceae) in pylorus ligated. The result of whole study has been

concluded that methanolic extract of curcuma longa leaves had two formulations which

are 20 mg/kg and 50 mg/kg both are effective against paracetamol induced ulcers. The evaluation of ulcer index was conformed the effectiveness of ulcer protective and

healing property of methanolic extract of curcuma longa.

Turmeric powder has beneficial effect on the stomach. It increases mucin secretion in rabbits and may thus act as gastroprotectant against irritants [6]. However, controversy exists regarding antiulcer activity of curcumin. Both antiulcer [7] and ulcerogenic [8, 9] effects of curcumin have been reported but detailed studies are still lacking.

Curcumin has been shown to protect the stomach from ulcerogenic effects of phenylbutazone in guinea pigs at 50 mg/kg dose [10].

Evaluation of turmeric has been done for gastric and duodenal antiulcer activity in rats *Curcuma longa* L. is commonly known as Turmeric and a household remedy for biliary disorders, anorexia, cough, diabetic wounds, hepatic disorders, rheumatism and sinusitis which belongs to the family Zingiberaceae. Curcumin inhibits the growth of *Helicobacter pylori*, which causes gastric ulcers and has been linked with gastric cancers.

110 species of the genus Curcuma L., only about 20 species have been studied phytochemically. *Curcuma longa* is the most chemically investigated species of *Curcuma*. Till date, at least 235 compounds, primarily phenolic compounds and terpenoids have been identified, including diary heptanoids (including commonly known as curcuminoids), diarylpentanoids, monoterpenes, sesquiterpenes, diterpenes, triterpenoids, alkaloid, and sterols, etc.

MATERIAL AND METHOD

Plant material collection and authentication:

Curcuma longa plant's leaves are collected at in the month of June 2016 from local area of Shubham nursery Bhopal M.P. India. The specimens submitted by Sandhya Sujane has been identified as leaves of Curcuma longa family belong to zingiberaceae. It's authenticated by Dr. Zea ul Hassan of the Department of Botany, Saifia science College, Bhopal. The authentication no. for the specimen is 438/BS/saifia/17 has been preserved for future identification.

EXTRACTION

Extraction The leaves were separated from the fresh and dried on filter paper sheets under shaded ried and powdered mechanically at room temperature until with changing of color of filter papers. The shade-dried, coarsely powdered leaves (500 g) were successively extracted with methanolic. The powdered plant material (500 g) was repeatedly extracted in a 2000 mL round bottomed flask with 1500 mL solvents of increasing polarity starting with methanolic solvent are used. The defatted marc was then subjected to soxhlet extraction with 95% methanol to obtain methanolic extract. The methanolic extract were evaporated under reduced pressure at low temperature (30°C) to dryness to yield brownish yellow color extracts of Curcuma longa 1. The extracts were cooled at room temperature. filtered, Preliminary Phytochemical screening methanolic extract of Curcuma longa leaf, stored in an airtight container in refrigerator for further experimental studies.

Experimental animals

Albino rats (Wistar strain) of either sex, weighing 180–200 g was procured from Javahar cancer hospital in Bhopal M.P. All the animals were placed in polypropylene cages in a controlled room temperature 22±1°C and relative humidity of 60–70% in registered animal house 1238/9/08/CPCSEA. The animals were maintained on standard pellet diet (Amrut Brand, Sangli, India) and water ad libitum. They were acclimatized to laboratory condition for seven days before commencement of the experiment. Ethical clearance was obtained from Institutional Animal Ethics Committee.

Preliminary Phytochemical Screening

Methanol extracts of turmeric leaf (METL) were screened for the presence of phytochemical constituents which may be the reason for the antimicrobial properties of Curcuma longa leaves for phytoconstituents like alkaloids, carbohydrates, glycosides, physterols, fixed oils and fats, saponins, phenolic compounds and tannins, proteins and amino acids, flavonoids using different phytochemical tests [11].

STUDY OF ULCER IN ANIMALS

Group 1. Six animals (rats) serves as control and were given only distilled water daily.

Group 2. Six animals drug induced diseased rats and serves as ulcer control and were given only distilled water.

Group 3. Six animals drug induced diseased rats and shall be treated with orally (leaf extract).20mg/kg

Group 3.1. Six animals drug induced diseased rats and shall be treated with orally (leaf extract).50mg/kg

Group 4. Six animals drug induced diseased rats and shall be treated with standard drug Ranitidine (2mg/kg).

Acute toxicity studies

Acute oral toxicity study of METL was carried out in RAT of both sexes (180-200g) according to OECD guidelines no 423. Extract at different doses up to limit dose 2000 to 5000 mg/kg was administered orally and animals were observed for behavioral changes, any toxicity and mortality up to 48 h [12].

PHAMACOLOGICAL SCREENING METHODS

Pylorus ligation in rats (SHAY rat)

Purpose and rationale

A simple and reliable method for production of gastric ulceration in the rat published by Shay *et al.*, (1945) based on ligature of the pylorus. The ulceration is caused by accumulation of acidic gastric juice in the stomach.

Procedure

The female Wistar rats weighing 180–200g are starved for 48 hrs. having access to drinking water ad libitum. During this time rats are housed single in cages with raised bottoms of wide wire mesh to avoid cannibalism and coprophagy. Ten animals are used per dose and as controls. Under ether anesthesia made a midline abdominal incision. The pylorus is ligated, care being exercised that neither damage to the blood supply nor traction on the pylorus occurs. Carefully grasping the stomach with instruments is to be avoided meticulously; else ulceration will invariably develop at such points. The abdominal wall is closed use sutures. The test compounds are given orally by gavage or injected subcutaneously.

The animals placed for 19 hrs in plastic cylinders with an inner diameter of 45 mm being closed by wire mesh on both ends. Afterwards, the animals sacrificed in CO_2 anesthesia. The abdomen opened, and a ligature placed around the esophagus close to the diaphragm. The stomach removed, and the contents drained in a centrifuge tube.

Along the greater curvature the stomach opened and pinned on a cork plate. In the rat, upper two fifths of the stomach form the rumen with squamous epithelium and possess little protective mechanisms against the corrosive action of gastric juice. Below a limiting ridge, in the glandular portion of the stomach, the medium two fifths of the stomach than in the lowest part, the protective mechanisms are better in the mucosa.

Therefore, lesions occur mainly in the rumen and in the antrum. The number of ulcers noted, and the severity recorded with the followed scores

- 0 = no ulcer
- 1 = superficial ulcers
- 2 = deep ulcers
- 3 = perforation.

The measured volume of the gastric content, after centrifugation, acidity is determined by titration with 0.1 n NaOH.

Evaluation:

An ulcer index UI is calculated:

 $UI = UN + US + UP \times 10 - 1$

UN= average of number of ulcers per animal

US= average of severity score

UP= percentage of animals with ulcers

The Ulcer index and acidity of the gastric content of treated animals compared with controls. Using various doses, establishes dose-response curves for ulcer formation and gastric acid secretion. $\rm ID_{50}$ values calculated by probity analysis, whereby 0% corresponds to no and 100% to maximal stimulated gastric acid output.

RESULTS

Results of Phytochemical Screening test

Methanol extracts of turmeric leaf (METL) were screened for the presence of phytochemical constituents which may be the reason for the antimicrobial properties of Curcuma longa leaves for phytoconstituents like alkaloids, carbohydrates, glycosides, physterols, fixed oils and fats, saponins, phenolic compounds and tannins, proteins and amino acids, flavonoids using different phytochemical tests [13].

Table 1: Methanol extracts of turmeric leaf (METL) were screened for the presence of phytochemical constituents

Phytochemical constituents	Name of the tests	Methanolic extract
	Mayer's test	-
Alkaloids	Dragendroff's test	+
Aikaioius	Wagner's test	-
	Hager's test	-
Glycosidos	Fehling's test	-
Glycosides	Legal's test	-
	Shinoda test	+
Flavonoids	Zinc chloride reduction	+
Travollolus	test	т
	Alkaline reagent test	+
Carbohydrate	Molisch's test	+
Carbonydrate	Fehling's test	+
Proteins	Millon's test	+
Amino acids	Ninhydrin test	+
Fats and fixed	Stain test	-
oils	Saponification test	-
Phenolic comp.	FeCl ₃ test	+
and tannins	Lead acetate test	+
Steroids	Liebermann-Burchard	
Steroids	test	-
Terpenoids	Salkowski test	-

Acute toxicity studies

Acute oral toxicity study of MECL was carried out in RAT of both sexes (180-200g) according to OECD guidelines no 423. It is found that MECL were safe at limit dose 1 mg/kg, 5 mg/kg, 10 mg/kg, 20 mg/kg and 50 mg/kg with no mortality in studied subjects.

1/10th of these doses i.e. 20 mg/kg and 50 mg/kg were used in the subsequent study respectively Extract at different doses up to limit dose 2000 to 5000 mg/kg was administered orally and animals were observed for behavioral changes, any toxicity and mortality up to 48 h [14].

Table 2: Acute oral toxicity of MECL was carried out in RAT of both sexes (180-200g)

S. No.	Dose (mg/kg) P.O.	No. of animals	Observation
1	1 mg/kg	06	All animals survived
2	5 mg/kg	06	All animals survived
3	10 mg/kg	06	All animals survived
4	20 mg/kg	06	All animals survived
5	50 mg/kg	06	All animals survived

 LD_{50} : 5000 mg/kg, ED_{50} : 500 mg/kg

PHAMACOLOGICAL SCREENING METHODS

Pylorus ligation in rats (SHAY rat) Purpose and rationale

Evaluation

severity of ulcers is registered with a stereo-microscope using the following scores:

- 0 = no ulcer
- 1 = superficial ulcers
- 2 = deep ulcers
- \blacksquare 3 = perforation

An ulcer index UI is calculated:

 $UI = UN + US + UP \times 10 - 1$

- UN= average of number of ulcers per animal
- US= average of severity score
- UP= percentage of animals with ulcers

Table 3: severity of ulcers is registered with a stereomicroscope using the following scores

S.	Group	Avg. No.	Severity of	% of animals
No.	No.	of Ulcers	scores	with ulcers
1	G-1	00	00	00
2	G-2	<u>±</u> 4	±2	±6
3	G-3A	±1	±1	±2
4	G-3B	±1	0	±1
5	G-4	0	0	±1

Group: 1 Normal Control

 $UI = 00+00+00 \times 10-1$

UI=00

Group: 2 Drug controlled (Disease induced)

UI= 4+2+6 x 10-1

UI= 119

Group: 3 Treated with Methanolic extract 20 mg/kg

 $UI = 1 + 1 + 2 \times 10 - 1$

III = 39

Group: 3 Treated with Methanolic extract 50 mg/kg

UI= 1+0+1 x 10-1

UI = 19

Group: 4 Standard drug treated (Ranitidine)

 $UI = 0 + 0 + 1 \times 10 - 1$

UI = 9

Table-4: Results of body weight and it's pH determination of animals from different dose groups

S.		WEIGHT OF RAT		
No.	GROUP	After	Before	pН
		dosing	dosing	
1	Normal group	180g	180g	3 ± 4
2	Disease induce group	159.4g	156.1g	1 ± 1.5
3	Treated group 20 mg	166g	158.4g	3 ± 3.1
4	Treated group 50 mg	185.2g	181.1g	3.2 ± 3.5
5	Ranitidine	179.4g	179.6g	5±4

Stomach weight: In paracetamol treated rats, enlargement of stomach was observed, which was evident of increase in the stomachs weight. The groups treated with *Curcuma longa* (250 and 500 mg/kg, p.o) and silymarin showed significant restoration of liver weight nearer to normal.

The stomach weight of paracetamol treated group was found to be $7.1\pm0.05 \, \mathrm{gm}$ b.w. whereas for *Curcuma longa* (250 and 500 mg/kg, po) and silymarin treated group, it was found to be 6.58 ±1.02 , 6.08 ± 0.81 and $6.02\pm0.46 \, \mathrm{gm}$ b.w. respectively. The results are shown in Table 5. Values are mean \pm SEM, n = 6. (One-way ANOVA Followed by Dunnette multiple comparisons test). Statistically significance of ** P<0.01, *** P<0.001, when compared with paracetamol induced group and * P<0.05, when compared with normal group.

Table 5: Effect of selected plant extracts stomachs ph and weight in paracetamol rats

Result of Histopathology study

S. No.	Treatment/ Dose	stomach weight (wt/gm)
1	Normal group	0.2 ± 0.05
2	Disease induce group	
3	Treated group 20 mg	
4	Treated group 50 mg	
5	Ranitidine	

DISCUSSION & CONCLUSION

The etiology of the peptic ulcer is unknown in most of the cases, it is generally accepted that it results from an imbalance between aggressive factors and the maintenance of mucosal integrity through the endogenous defence mechanisms. To regain the balance, different therapeutic agents including plant extracts may be used. Methanolic extract is one such herbal drug used in the present study primarily to evaluate the anti-ulcerogenic in paracetamol induced ulcers. The causes of gastric ulcer are believed to be due to increase in gastric hydrochloric acid secretion and/or stasis of acid and the volume of secretion is also an important factor in the formation of ulcer due to exposure of the unprotected lumen of the stomach to the accumulating acid.

Drug induced ulcers are due to auto digestion of the gastric mucosa and breakdown of the gastric mucosal barrier. These factors are associated with the development of upper gastrointestinal damage including lesions, ulcers and evaluation of anti-ulcer activity of methanolic extract of *Curcuma longa* leaves.

The result of whole study has been concluded that methanolic extract of *Curcuma longa* leaves had two formulations which are 20 mg/kg and 50 mg/kg both are effective against paracetamol induced ulcers. The evaluation of ulcer index was conformed the effectiveness of ulcer protective and healing property of methanolic extract of *Curcuma longa*.

REFERENCE

- 1. Sabiha S, Mohd AA, Asif M, Akhtar M. Role of phenolic compounds in peptic ulcer: An overview. *J Pharm Bioallied Sci* 2011; 3:361-7.
- 2. Jainu M, Devi CS. Antiulcerogenic and ulcer healing effects of *Solanum nigrum* (L.) on experimental ulcer models: Possible mechanism for the inhibition of acid formation. *J Ethnopharmacol* 2006; 104:156-63.
- 3. Bighetti AE, Antonio MA, Kohn LK, Rehder VL, Foglio M, Possenti A, et al. Anti-ulcerogenic activity of a crude hydroalcoholic extract and coumarin isolated from *Mikania laevigata* Schultz Bip. Phytomedicine 2005; 12:72-7.
- 4. Ode O.J. and Arya O.V. VS. Indian Medicinal Plants. 1st ed. Vol. 4. Hyderabad: *Orient Longman Ltd. Publication*; 1995.
- Bighetti AE, Antonio MA, Kohn LK, Rehder VL, Foglio M, Possenti A, et al. Anti-ulcerogenic activity of

- a crude hydroalcoholic extract and coumarin isolated from *Mikania laevigata* Schultz Bip. *Phytomedicine* 2005; 12:72-7. 5.
- Lee, C. J., Lee, J. H., Seok, J. H., Hur, G. M., Park, Y. C., Seol, I. C. and Kim, Y. H., Effects of baicalein, berberine, curcumin and hespiridin on mucin release from airway goblet cells. *Planta Med.*, 2003, 69, 523–52.
- 7. Sinha, M., Mukherjee, B. P., Mukherjee, B., Sikdar, S. and Dasgupta, S. R., Study of the mechanism of action of curcumin; an antiulcer agent. *Indian J. Pharmacol.*, 1975, 7, 98.
- 8. Prasad, D.N., Gupta B., Srivastava, R.K. and Satyavati, G.V., Studies on ulcerogenic activity of curcumin. *Indian J. Physiol. Pharmacol.*, 1976; 20: 92.
- 9. Gupta, B., Kulshrestha, V. K., Srivastava, R. K. and Prasad, D. N., Mechanisms of curcumin induced gastric ulcer in rats. *Indian J. Med. Res.*, 1980, 71, 806–814.
- Dasgupta, S. R., Sinha, M., Sahana, C. C. and Mukherjee, B. P., A study of the effect of an extract of *Curcuma longa* Linn. on experimental gastric ulcers in animals. *Indian J. Pharmacol.*, 1969, 1, 49–54.
- 11. Ghongane BB, Rahul K. Evaluation of anti-ulcer activity of *Curcuma longa* in rats. *Journal of Advances in Pharmacy and Healthcare Research* 2011; 1(2)
- 12. Ranajit K. Banerjee, *CURRENT SCIENCE*, VOL. 87, NO. 1, 10 JULY 2004; 44
- 13. Dashputre et al. / Evaluation of Anti-Ulcer Activity of Methanolic Extract of *Abutilon indicum* Linn. *IJPSDR* April-June 2011, Vol 3, Issue 2 (97-100) [35;1]
- 14. Rambir Singh., Ramesh Chandra., Mridula Bose., Prathibha Mehta Luthra. (2002) *Current Science*, 83(6): 737740.

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