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BIOLOGICAL ACTIVITIES OF CHAMPACA (*MAGNOLIA CHAMPACA*) PLANT – A REVIEW

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ABSTRACT
Magnolia champaca (Family name: Magnoliaceae) is a medicinal plant which is traditionally used against a number of diseases. The flowers are used in Southeast Asia for several purposes. Especially in India, they are primarily used for worship at temples whether at home or out, and more generally worn in hair by girls and women as a means of beauty ornament as well as a natural perfume. The extracts of champaca plant were investigated for antibacterial activity through disc diffusion method against four different bacterial strains using ampicillin as control. Carrageenan has been widely used as a noxious agent able to induce experimental inflammation for the screening of compounds possessing anti-inflammatory activity. Anti-ulcerogenic activity of flower and leaves are evaluated against aspirin induced gastric ulcers in pylorus ligated rats. The anti-ulcer potential is determined by using gastric juice volume, total acidity, pH and ulcer index. Both aqueous and alcoholic extracts of flower and leaves are evaluated for their anti- ulcer potential.

KEYWORDS: Champaca plant; Biological activities; Anti- bacterial; Anti- inflammation; Anti- ulcer.

INTRODUCTION
Magnolia champaca is a member of family Magnoliaceae. It is well known and widely used in traditional medicine such as fever, colic, leprosy, post partum protection (Perry, 1980), eye disorder and many more. This plant was claimed possesses various pharmacological properties such as antipyretic, anti-inflammatory (Vimala *et al.,,* 1997), insecticidal, antimicrobial and etc. Furthermore, Atjanasuppat *et al.,* (2009) reported that this plant can be as remedy of anti-uretic, carminative and anti-dinic. Several compounds of this plant were also characterized and identified such as alkaloids, saponins, tannins, sterols, flavonoids and triterpenoids in the study of Khan *et al.,* (2002). The increasing of incidence of antibiotic resistance case among pathogenic bacteria lead to the most of commercial antibiotics were no longer effectively in controlling bacteria disease. As oxidative stress may lead to many human diseases, the use of antioxidants in pharmacology is intensively studied, particular as treatment for stroke and neurodegenerative diseases.

The acute inflammatory response elicited upon invasion of microorganisms or upon tissue damage is associated with the release of pro-inflammatory mediators by tissue macrophages, mast cells, or other tissue cells such as damaged fibroblasts. Soluble mediators like Histamine, Interleukin 1, and TNF-a activate the endothelium lining post capillary venules, i.e., they induce the expression of adhesion molecules and the secretion of soluble mediators, which in turn allow the leukocyte-endothelial cell interactions.

Carrageenan has been widely used as a noxious agent able to induce experimental inflammation for the screening of compounds possessing anti-inflammatory activity. It is well known that carrageenan-induced edema is characterized by biphasic effects. The first phase begins immediately after injection which further produces inflammation due to chemical mediators such as histamine and serotonin and diminishes within 1 h. The second phase begins at 1 h and remains through 3 h with metabolism of arachidonic acid by both cyclooxygenase and lipoxygenase enzyme pathways.

Biological activity Anti- bacterial assay
The extracts of *Magnolia champaca* flower and the isolated compounds were tested against two Gram positive and two Gram negative bacteria namely *Bacillus subtilis, Staphylococus aureus, Salmonella typhi* and *Shigella dysentriae.* Bacterial strains were maintained on Nutrient Agar slants at 4°C. The agar disc diffusion method was employed for the determination of antimicrobial activity of the extracts. Briefly a suspension of the microorganism was spread on the solid media plates. Whatman filter paper discs (6 mm in diameter)

were soaked with 10 µl of the test sample and placed on the inoculated media. Plates were kept at 4°C for 2 hours and then incubated at 37°C for 24 hrs. The diameter of the inhibition zones were measured in mm.

The increasing failure of chemotherapeutics and antibiotic resistance exhibited by pathogenic microbial infectious agents has led to the screening of several medicinal plants for their potential antimicrobial activity. The present study was targeted to screen the antibacterial activity and free radical scavenging activity of the different extracts of the flowers of *M. champaca*. The IC50 values calculated for scavenging DPPH free radical of hexane, ethyl acetate extracts and the three isolated compounds 1-3 were found to be 250 µg/ml, 160 µg/ml, 200 µg/ml, 220 µg/ml and 150 µg/ml, respectively. Percentage scavenging of DPPH radical was found to rise with increasing concentration of the crude extracts and the compounds (Figure 1).

The antibacterial activity of hexane and ethyl acetate extracts and the three isolated compounds 1-3 against the different strains namely *B. subtilis, S. aureus, S. typhi, S. dysentry* is measured in diameter of zone of inhibition (in mm) and the values are given in (Table 1) Ethyl acetate extract of *M. champaca* showed the highest activity against *S. aureus, B. subtilis, S. typhi* and *S. dysentry* (zones of inhibition: 12, 12, 14 and 8 mm) whereas compound-2 showed the lowest activity against *S. aureus* and *S. dysentry* (zones of inhibition:10 and 7 mm).While the hexane extract, compound-1 and compound-3 showed good to moderate activity against all the used bacterial strains with different zones of inhibition (Figure 2) (Umadevi Parimi and Deephi Kolli 2012).

Table 1: Antibacterial activity of *M. champaca* flower extracts and diameter of zones of inhibition (in mm).

Name of the organism	Hexane extract	Ethyl acetate extract	Compound-1	Compound-2	Compound-3	Ampicillin (30 µg/ disc)
Gram +ve						
Staphylococcus aureus	11	12	11	10	11	18
Bacillus subtilis	10	12	8	9	12	16
Gram -ve						
Salmonella typhi	13	14	13	12	10	NE
Shigella dysentriae	8	8	8	7	7	NE

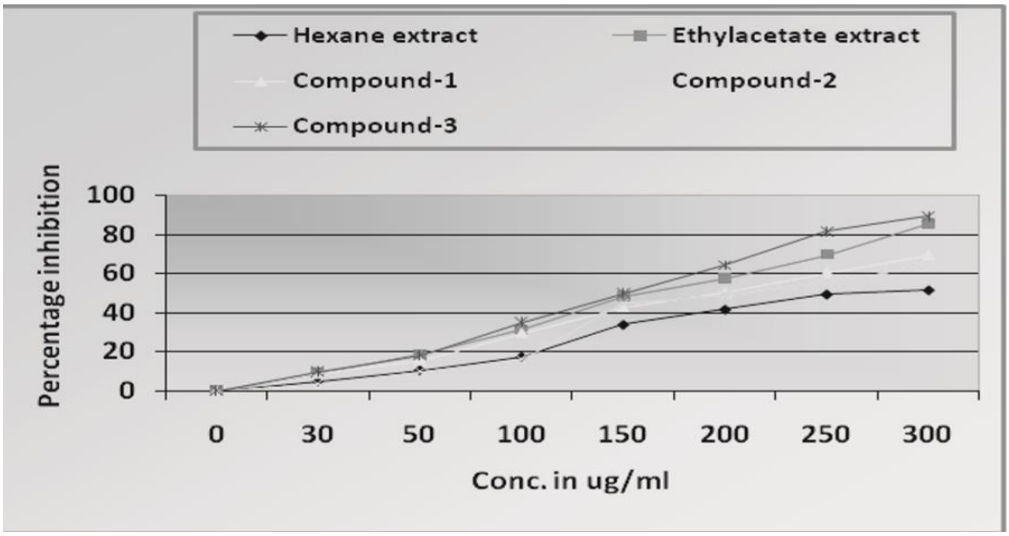


Figure 1: DPPH radical scavenging efficacy of different organic extracts of *Magnolia champaca*.

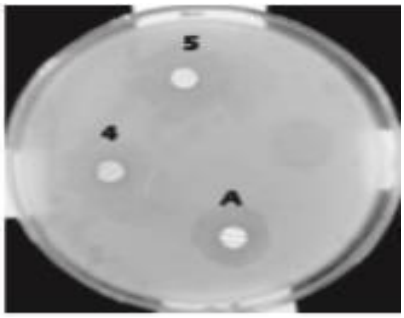
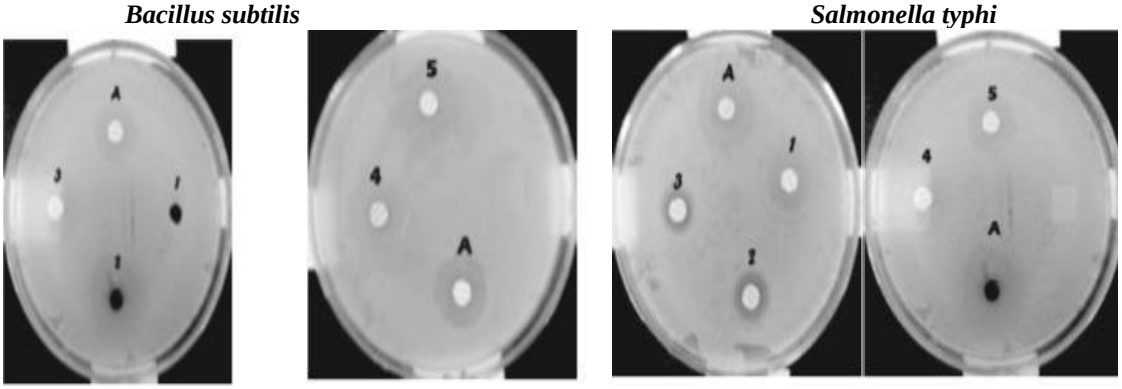


Figure 2: Zone of inhibition of the extracts and isolated compounds against the tested micro- organisms.

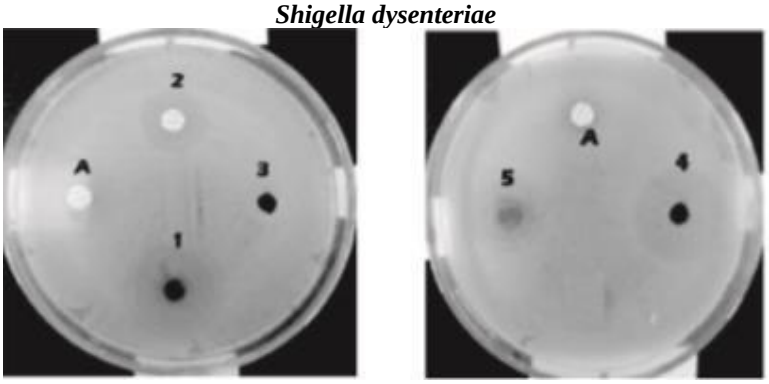
Staphylococcus aureus

1. Hexane extract
2. Ethyl acetate extract
3. Compound- 1
4. Compound- 2
5. Compound- 3
6. A- amphicillin

Bacillus subtilis ***Salmonella typhi***



Shigella dysenteriae



Anti- inflammation assay Preparation of plant extract

The dried powdered of leaves (2 kg) was successively Soxhlet extracted using petroleum ether (60-80°), chloroform, acetone, and ethanol for 72 h each. Crude aqueous extract of these leaves were prepared separately by maceration for 24 h. The last trace of solvent was removed by reduced pressure distillation and then vacuum dried. A dark semi solid mass was obtained. It was stored below 4 ºC until further used. When needed, the extract was suspended/dissolved in desired solvent and used. The extracts were concentrated by performing the qualitative chemical tests to determine the presence of sterols, phenol compounds, flavonoids and saponins, respectively.

Paw edema induced by Carrageenan

The animals received sub plantar injection of 0.1 ml of carrageenan (100 µg/rat) in 0.9 % NaCl into the right hind paw 30 min after dosing with the vehicle (0.2 ml of 2 % w/v carboxy methyl cellulose with 2 % Tween 80). Extracts were suspended in 0.2 ml of 2 % w/v carboxy methyl cellulose with 2.0 % Tween 80 and administered orally (200 mg/kg) to rats. The dose of extracts was selected on the basis of acute toxicities studies 10. Diclofenac sodium (10 mg/kg) was given to standard group. Animals were divided into 6 groups, each group composed by 6 rats: Group 1: Positive Control, carrageenan (C); Group 2: Standard group, carrageenan + diclofenac sodium 10 mg/kg b.w p.o. (S); Group 3: Test group, carrageenan + pet ether extract 200 mg/kg b.w p.o (Ptet200); Group 4: Test group, carrageenan + chloroform extract 200 mg/kg b.w p.o (Chl200); Group 5: Test group, carrageenan + ethanol extract 200 mg/kg b.w p.o (Et200); Group 6: Test group, carrageenan + aqueous exract 200 mg/kg b.w p.o (Aq200). The paw volume was measured with a digital plethysmometer (model 7140, UGO Basile, Italy) at first at zero hour and then at 30 min, 1, 2 and 3 h after administration of drugs. The percentage inhibition of edema compared with that of the control was taken as anti-inflammatory activity. The percentage inhibition of edema was calculated by the formula 13.

Percentage inhibition of edema = (A-B)/A x 100

where, A represents the paw volume of the control group and B represents the paw volume of the test drug treated group.

Table 2: Effects of different extract of leaves *Magnolia champaca* on carrageenan induced rat paw edema model. Each value is the Mean ml ± S.E.M. for 6 rats. **P < 0.01; *< 0.0001, ns: non-significant compared with control group after 3hrs (Tukey's t- test). Percentage (%) inhibition of paw volume at different intervals.**

Group	Dose (mg/kg)	Carrageenan induced rat paw edema (Mean (ml) + SEM)			
		30 Mins	1 h	2 h	3 h
Control (G1)	–	0.651±0.078	1.281±0.052	1.800± 0.210 ns	2.046±0.0352
Standard (G2)	10 mg/kg	0.644 ± 0.080 ns (1.075%)	0.570±0.036*** (55.50%)	0.364±0.060*** (↑79.77%)	0.185 ± 0.047 *** (↑90.95%)
Pet ether (G4)	200 mg/kg	0.625 ±0.061ns (3.99%)	1.225±0.054 ns (4.37%)	0.995±0.062*** (44.72%)	0.829±0.11*** (59.48%)
Chloroform (G6)	200 mg/kg	0.610±0.068 ns (6.29%)	1.105±0.075*** (13.73%)	1.038±0.075*** (42.33%)	0.985±0.030*** (51.85%)
Ethanol (G8)	200 mg/kg	0.662 ±0.107 ns (-1.68%)	0.929±0.080*** (27.47%)	0.739±0.125*** (58.94%)	0.647±0.068*** (68.42%)
Aqueous (G10)	200 mg/kg	0.691± 0.049 ns (-6.14%)	1.119 ±0.065** (12.64%)	0.843±0.017*** (53.16%)	0.799±0.030*** (60.99%)

The results of qualitative analysis indicated that pet ether and chloroform extract is rich in steroids while ethanolic and aqueous extract is rich in alkaloids and saponins (data not shown). In acute inflammation model, the carrageenan induced paw edema was significantly reduced by all the extracts as shown in Table 1. The result obtained indicates that the extracts found to have significant (P < 0.0001) anti-inflammatory activity against to positive control group in rats. The chloroform, ethanol, pet ether and aqueous extracts at a dose of 200 mg/kg b.w reduced the edema induced by carrageenan by 51.85 %, 68.42 %, 59.48 % and 60.99 %, respectively at 3 h, whereas the standard drug (10 mg/kg b.w) showed maximum inhibition 90.95,% against to positive control group. Among these, ethanolic extract showed maximum anti-inflammatory activity at every hour interval.

Based on the results of this study, the research work come to the conclusion that the *Magnolia champaca* leaves has potential anti-inflammatory activity, and thus provides a scientific basis for the utilization of this plant in traditional medicine for the treatment of pro-inflammatory diseases. Further studies will be carried out on pharmacodynamics pattern to establish the mechanism of action of the plant extracts (Sumeet GUPTA *et al.*, 2010).

Anti- ulcer assay

Aspirin induced Gastric Ulcers in rats: 36 healthy male Albino rats were selected and randomly distributed into 6 groups of six animals each Group I and II served as induction and positive control groups and received 200mg/kg b. wt of aspirin in 1% CMC solution followed by 0.9% saline by oral route for 5 days and the standard drug cimetidine 50mg/kg b.wt followed by 200mg/kg b.wt of aspirin in 1%CMC solution by oral route for 5 days respectively. Whereas, groups III, IV, V, VI received the treatment along with the aspirin induction. Flower aqueous and flower alcoholic 300mg/kg b.wt was given for group III and IV and group V and VI received leaf aqueous and leaf alcoholic 300mg/kg b.wt.

On the sixth day pylorus ligation was performed for each animal. The animals were sacrificed after four hours using diethyl ether anaesthesia, abdomen was opened and the stomach was isolated. The gastric juice was collected in a measuring cylinder and the stomach was opened along the curvature. The mucosa was washed with 1ml distilled water and the washings were added to the gastric juice, the gastric content was centrifuged at 1000 rpm for 10mins. 1ml of supernatant was diluted with 9 ml of distilled water. The solution was titrated against 0.01N Sodium Hydroxide using phenolphthalein as indicator and the end point was noted 7, 8, 9.

Each stomach was examined grossly and the ulcer was graded using the followingscoring system;
0= normal mucosa 0.5=red coloration 1=spot ulcers
1.5 = heamorrhagic streaks
2=ulcers >3 but <5
3=ulcers > 5

The mean of the ulcer scores in each group was taken as ulcer index

Result of this study shows that antiulcer property of *Magnolia champaca* Linn flowers and leaves were evaluated by employing pylorusligation and aspirin induced ulceration models in albino rats. Aspirin is a known ulcerogenic agent which significantly increases the free and total acidity with a decrease in hexose, hexosamine and sialic acid levels. It is found to be one of the notable agents which cause ulceration in human

beings. Factors such as gastric acid output, gastric wall mucin depletion and vascular injury are associated with ulcerogenesis caused by aspirin 10. Gastric acid and Pepsin are important factors for the formation of ulcers in pylorus-ligated rats.

Pylorus ligation may also attribute to increased synthesis of nucleic acid metabolism of carbohydrates. Aspirin along with pylorus ligation is found to be a very effective model for induction of ulceration in rats. Aspirin causes mucosal damage by interfering with prostaglandin synthesis, increasing acid secretion and back diffusion of H+ ions 3. The inhibition of mucosal prostaglandin production occurs rapidly following oral administration of aspirin. This is correlated with rapid absorption of these drugs through the mucosa. In pylorus ligation, the digestive effect of accumulated gastric juice and interference of gastric blood circulation are responsible for the induction of ulceration.

Magnolia champaca Linn. flowers and leaves were evaluated for the antiulcer potential in the current study. Both alcoholic and aqueous extracts were tried against the model using cimetidine as a reference drug. All the four extracts significantly decreased the gastric juice volume as compared to the induction group and the results are exhibited in (Table 3) (M. Surendra Kuma *et al.*, 2011).

Table 3: Effect of various extracts of *Magnolia champaca* and cimetidine in pyloric ligated rats.

Group no	Treatment	Volume of gastric juice (in ml)	pH	Total acidity (mEq/lit)	Ulcer index
I	Aspirin+0.9%Saline	4.5±0.095	2.0±0.049	34±0.045	3.0±0.002
II	Cimetidine + Aspirin	1.6±0.1687***	4.0±0.090***	14±0.098***	0.1±0.105***
III	Flower aqueous	2.5±0.089***	3.5±0.182***	23±0.005***	0.6±0.154***
IV	Flower alcoholic	2.96±0.032***	3.0±0.238**	31±0.028**	1.2±0.211***
V	Leaf aqueous	3.2±0.104**	2.5±0.234	32±0.004	1.5±0.154***
VI	Leaf alcoholic	2.77±0.0460***	3.5±0.422***	28±0.547**	1.0±0.146***

All the values are mean ± SEM; n=6; *** P<0.0001; **P<0.001 as compared to the group 1.

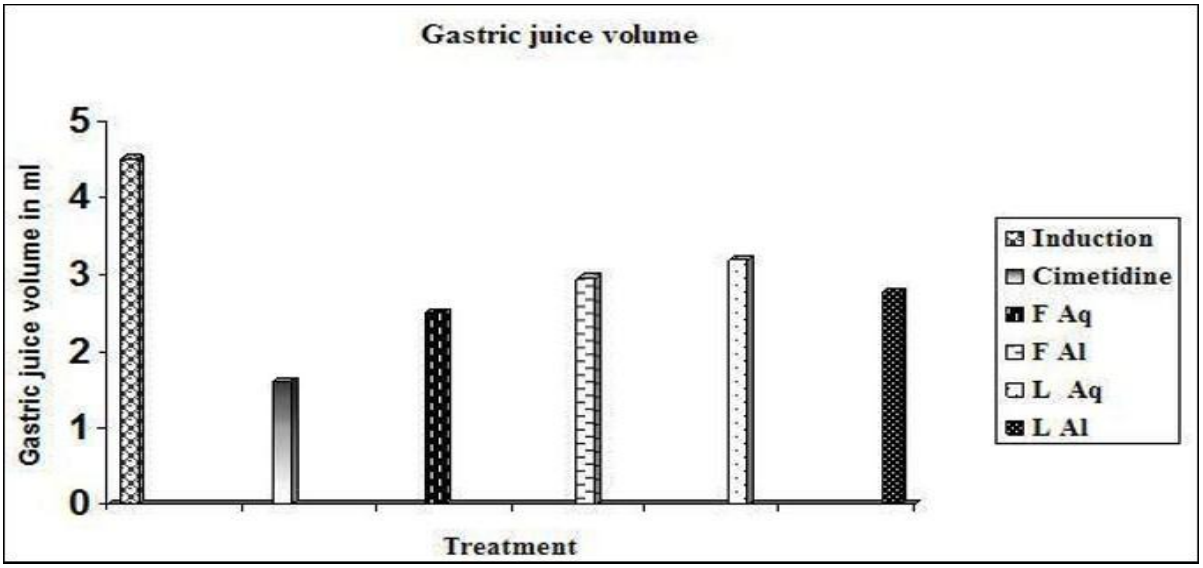


Figure 2: clearly depicts the decrease in gastric juice volume was at its maximum level for the reference standard group followed by flower aqueous extract treatment group. The least level of reduction was seen in leaf aqueous extract treated group.

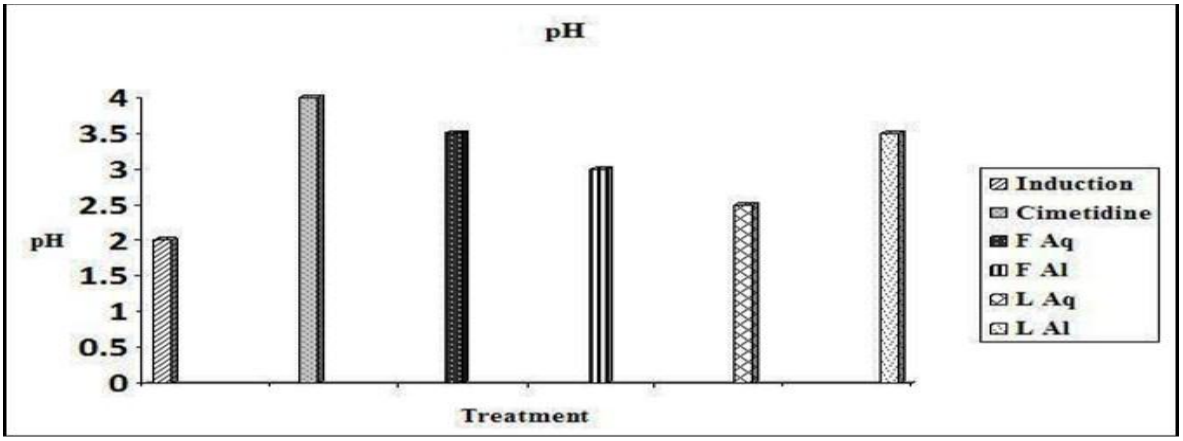


Figure 3: shows the increase in pH was at its maximum level for the reference standard group followed by flower aqueous extract treatment group. The least level of increase was seen in leaf aqueous extract treated group. The excessive secretion of hydrochloric acid in the stomach was considered to be an important factor in the formation of peptic ulcer. Hydrochloric acid is known to produce ulceration and digestion of the stomach tissues as well as to reduce the neutralizing capability of the stomach mucus secretions.

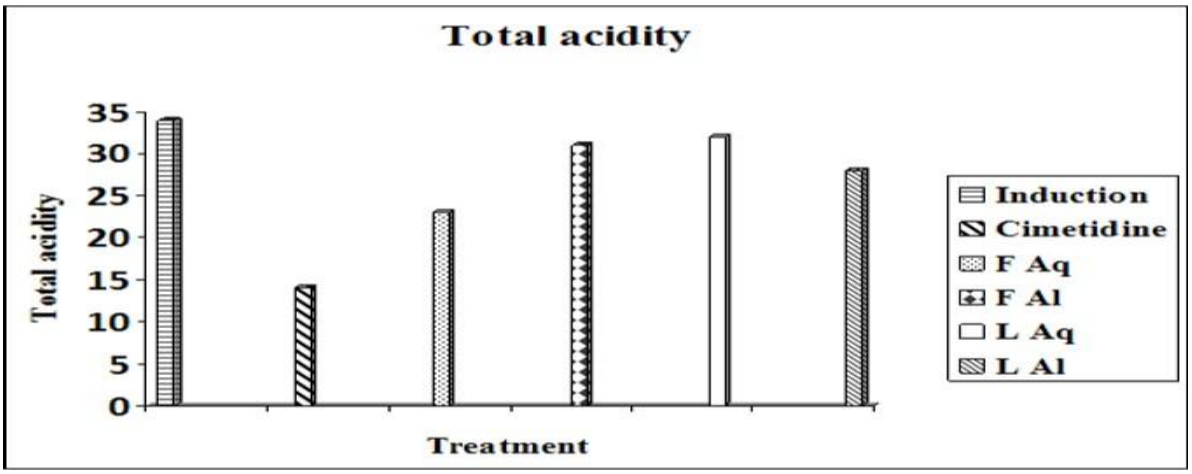


Figure 4: depicts the decrease in acidity was at its maximum level for the reference standard group followed by flower aqueous extract treatment group. The least decrease in acidity was shown by leaf aqueous extract treated group.

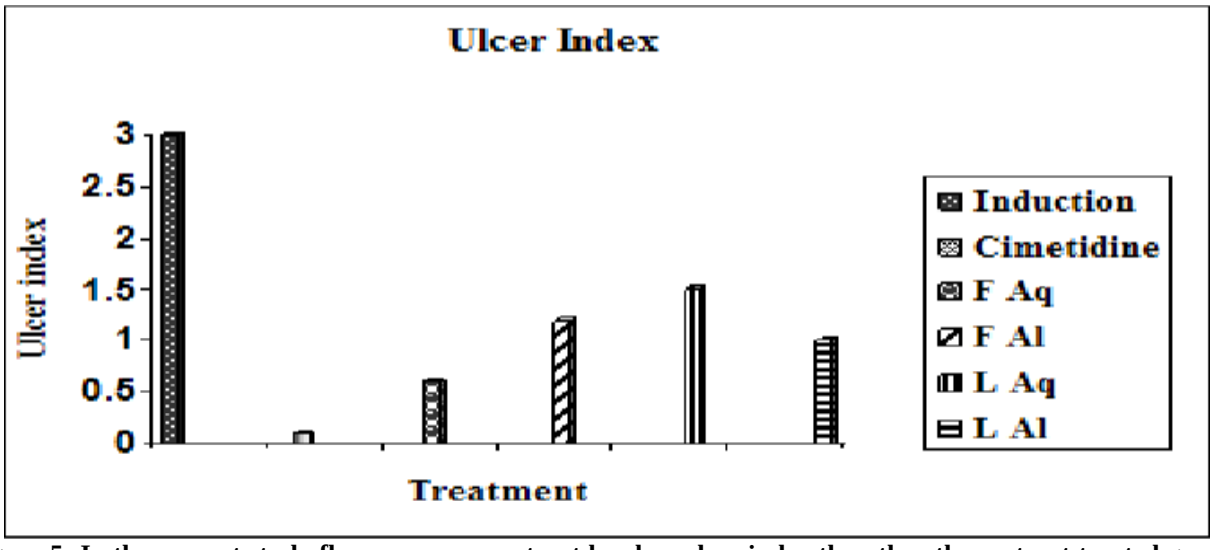


Figure 5: In the present study flower aqueous extract has less ulcer index than the other extract treated groups and high ulcer index was exhibited by leaf aqueous extract. This least ulcer index of the flower aqueous extract showed that, it was a better drug of choice than the other three extracts.

CONCLUSION

This study concludes that the extracts and the isolated compounds are good to antibacterial activity. This is the first report of the antimicrobial properties of the individual pure compounds isolated from *Magnolia champaca* Linn. Flowers (Umadevi Parimi and Deephi Kolli 2012). Based on the results of anti- inflammatory study, they came to the conclusion that the *Magnolia champaca* leaves has potential anti-inflammatory activity, and thus provides a scientific basis for the utilization of this plant in traditional medicine for the treatment of pro-inflammatory diseases(Sumeet GUPTA *et al.*, 2010). It is also concluded that the flower aqueous extracts of *Magnolia champaca* Linn. possess significant antiulcer activity than the other three extracts (leaf alcoholic, flower alcoholic, leaf aqueous) (M. Surendra Kuma *et al.*, 2011).

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