

# Pharmacological studies of *Gymnema sylvestre* leaves extract

## **Abstract-**

We have undertaken to evaluate the pharmacological studies of *Gymnema sylvestre* using pharmacological methods, disc diffusion, and anti-inflammatory methods. The Alkaloid, Carbohydrate, Protein, Resin, Saponin, and Glycosides were present whereas, steroid, flavanoid, and tannin were absent in phytochemical screening in *G. sylvestre* extract. The result showed induced paw edema using Carrageenan model. The dose-dependent antibacterial activity was also observed. The present study is important because *Gymnema sylvestre* plant is important to cure various ailments in traditional medicine.

**Key Words:** Antibacterial, Phytochemical, *Gymnema sylvestre*, Anti-inflammatory activity

## **INTRODUCTION**

The leaves of *Gymnema* reported a loss of sweet taste. The major chemical constituents of *Gymnema* are gymnemic acid and gurmarin. It was reported that gymnemic acids have antidiabetic, anti-sweetener, anti-inflammatory, antioxidant activities, and antidote properties. ((Gloria *et al* 2003, Dateo *et al*, 1973, Kanetkar *et al*, 2004, Persaud *et al*, 1999, Rachhet *et al*, 2009, Puratchimani). It helps to promote weight loss possibly through its ability to reduce cravings for sweets and control blood sugar levels. The gurmarin peptide blocks the ability to taste sweet or bitter flavors and thus reduces sweet cravings (Preuss *et al*, 2004). The wound healing activity in albino mice. *In vitro*, the inhibitory effects of DPPH radicals and LDL oxidation and muscle relaxant properties and anti-allergic activity were reported (Kiranmai *et al*, 2011, Luo *et al*, 1999, Tandon *et al*, 2010). The radio-protective and

immune modulatory effect of Gymnemic acid was reported on Swiss albino mice induced by radiation. The studies reported the anticancer activity of *Gymnemasylvestre* on MCF 7 (epithelial cells of human breast cancer and A 549 epithelial cells of human lung cancer under in vitro conditions by MTT assay method (Bhatia *et al*, 2008, Jitender *et al*, 2008 , 2009). Srikanth *etal*, 2010 and Sonam and Agrawal, 2018).

## **MATERIALS AND METHODS**

### **Extraction Process:-**

#### **Procedure:**

The *Gymnemasylvestre* leaves were collected from the local garden of Chitrakoot Uttar Pradesh in the month of April 2020 and were identified by competent Botanist Dr. Manoj Tripathi of DRI, Chitrakoot. The leaves were cleaned and dried for few days in shade. Then powder was made with the help of grinder. The extraction was done as per the method reported in earlier paper. On the day of experimentation, the desired amount of powder was suspended in double distilled water for the final administration. Phytochemical screening was done as per method reported by Agrawal, 2021a, b and 2022).

### **1.2 Antibacterial Activity:-**

The test organisms were obtained from the Department of Research, PBCRI Satna (M.P.). Antibacterial screening was done to find out the antibacterial properties of different concentration of 50% methanolic extract of *Gymnemasylvestre* leaves under study. Kirby-Bauer Method (Disc diffusion method) was followed to test the antibacterial activity of different concentration of leaves extract.

### **Evaluation of Anti-inflammatory Activity *Carrageenin-induced rat paw oedema***

The mice were divided into five groups ( $n = 3$ ). Acute inflammation was induced by the sub-plantar administration of 0.1 ml of 1% carrageenin in normal saline in the right hind paw of the mice. The paw volume was measured at 0 and 4 h after carrageenin injection, using Plethysmometer. The different groups were made as described in the Table 4. The mice were divided into five groups ( $n = 3$ ). After shaving the fur, the mice

were anaesthetized under light ether and 10 mg of sterile cotton pellets were inserted, one in each axilla of the mice. Extract (200, 300 and 500 mg/kg), Phenylbutazone (100 mg/kg) and to group control vehicle were administered orally for seven consecutive days from the day of cotton pellet implantation. The animals were anaesthetized on the eighth day and cotton was removed surgically. The pellets were dried at 60°C. The results were expressed as mean ± S.E.M. The significance statistical analysis was performed by test and  $P < 0.01$ , implied significance organism was recorded.

## RESULTS

The present study revealed the presence of medicinally important bioactive compound. The phytochemical screening of 50% methanolic extract of *Gymnema sylvestre* are depicted in table 1. The result showed the presence of Alkaloid, Carbohydrate, Protein, Resin, Saponin, Glycosides. Only Starch, steroid, flavanoid, tannin are absent in *G. sylvestre* extract.

**Table 1: Qualitative Phyto-chemical Screening of (*Gymnema sylvestre* (Retz.) Schult. (Leaf)**

S. No.	Name of Experiments	Observation	Result
1.	<b>Alkaloids</b>		
	a) Mayer's test	Yellow colour appear	Present
	b) Wagner's test	Brown colour appear	Present
	c) Dragendorff's test	Orange colour appear	Present
2.	<b>Carbohydrate</b>		
	a) Anthrone's test	Dark colour appear	Present
	b) Fehling's test	Green colour appear	Present
	c) Molisch's test	No red – violet ring disapper	Present
3.	<b>Proteins</b>		
	a) Biuret's test	Green colour appear	Present
	b) Millon's test	White ppt are not appear	Present
5.	<b>Resins</b>	Turbidity are seen	Present

<b>6.</b>	<b>Saponins</b>	Honey comb – like structure are form	Present
<b>7.</b>	<b>Starch</b>	Red colour is formed	Absent
<b>8.</b>	<b>Flavonoid</b>		Absent
	a) Ferric chloride test	Reddish pink colour is appear	Absent
	b) Alkaline reagent test	On addition of dilute acid yellow colour disappear	Present
<b>9.</b>	<b>Steroid</b>		
	a) Salkowski's reaction	A red colour is disappear in the chloroform layer	Absent
<b>10.</b>	<b>Glycoside</b>		
	a) Borntrager's Test	Colour is change	Present
<b>11.</b>	<b>Tannin</b>	Greenish colour appear	Absent
	a) Lead acetate Test	Reddish brown bulky ppt. are formed	Absent

## 2 Antibacterial assay

50% methanolic extract of *G. sylvestre* at the different concentration i.e. 25%, 50%, 75%, 100% exhibited antibacterial against *Bacillus subtilis*, *Staphylococcus aureus* but the lower activity was observed *E. coli* and *Pseudomonas aeruginosa* at 100%) the Minimum inhibitory concentration (MIC) of *Gym. Sylvestre* against gram positive bacteria i.e. *Bacillus subtilis*, *Staphylococcus epidermidis*, *Staphylococcus aureus* was 25% but against gram negative bacteria Zone of inhibition was observed only in 100% extract . Other concentration i.e. 25%, 50 %

**Table2 Antibacterial activity of *Gym. Sylvestre* against bacterial strains**

Name of microorganisms	% Concentration of Extract [zone of inhibition(mm)]			
	25	50	75	100
S.aureus	16	17	16	15
B.subtilis	10	12	13	15

<i>E. Coli</i>	13	14	13	15
<i>Ps. aeruginosae</i>	--	--	11	12



Figure1 (a) Zone of inhibition of *Gymnema sylvestre* extract against different strains of bacteria.

Table 3. Antibacterial activity of standard antibiotic (gram positive) against different bacteria.

Name of microorganisms	Name Standard antibiotics [zone of inhibition(mm)]			
	TE	OF	AZ	PC
S.aureus	15	16	16	14
B.subtilis	14	16	18	14
S.epidermidis	14	18	17	17

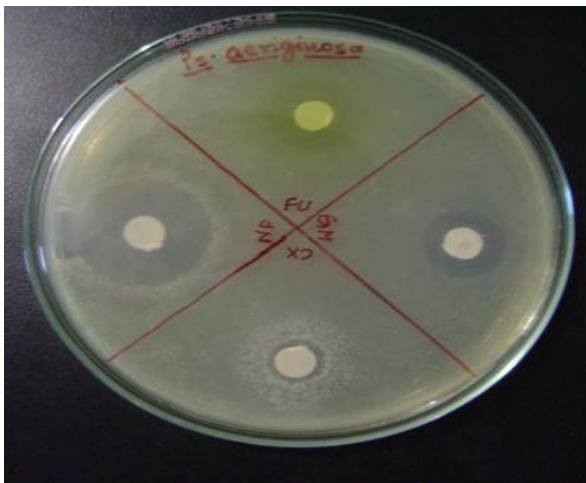
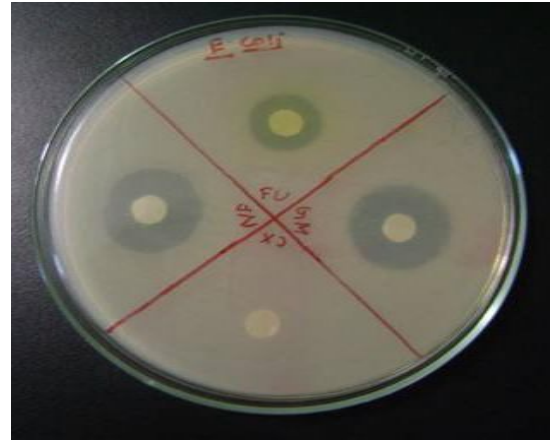
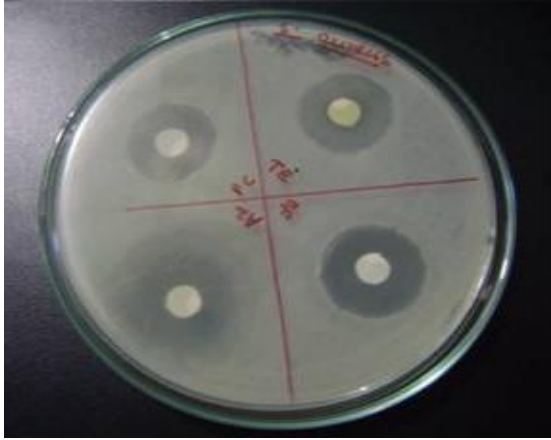
TE- Tetracycline, OF- Ofloxacin, AZ- Azithromycin & PC- Piperacillin

Table 4 Antibacterial activity of standard antibiotic (gram negative) against different bacteria.

Name of microorganisms	Name Standard antibiotics [zone of inhibition(mm)]			
	FU	GM	CX	NF
E.coli	12	16	8.0	16
Sh.flexineri	18	18	12	21
P.aeruginosa	14	13	18	20

FU- Nitrofurantoin, GM- Gentamicin, CX- Cefotaxime & NF- Norfloxacin

S. aureus – *Staphylococcus aureus*, B. subtilis – *Bacillus subtilis*, S. epidermidis – *Staphylococcus epidermidis*, E. coli – *Escherichia Coli*, Sh. Flexineri – *Shigella flexineri*  
P. Aeruginosa – *Pseudomonas aeruginosae*



**Figure 2(b) Zone of inhibition of standard antibacterial against different strains of bacteria**

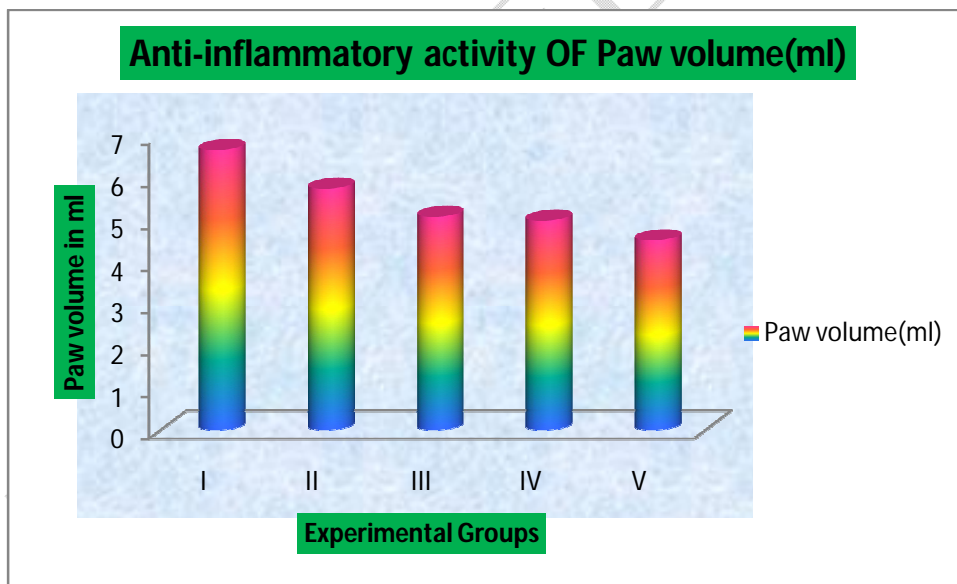
#### **4 Anti-inflammatory activity-**

The result of anti-inflammatory activity of methanol extract of *Gymnema sylvestre* on Carrageenan induced paw edema is shown in table 5. The methanolic extract of 400mg/kg

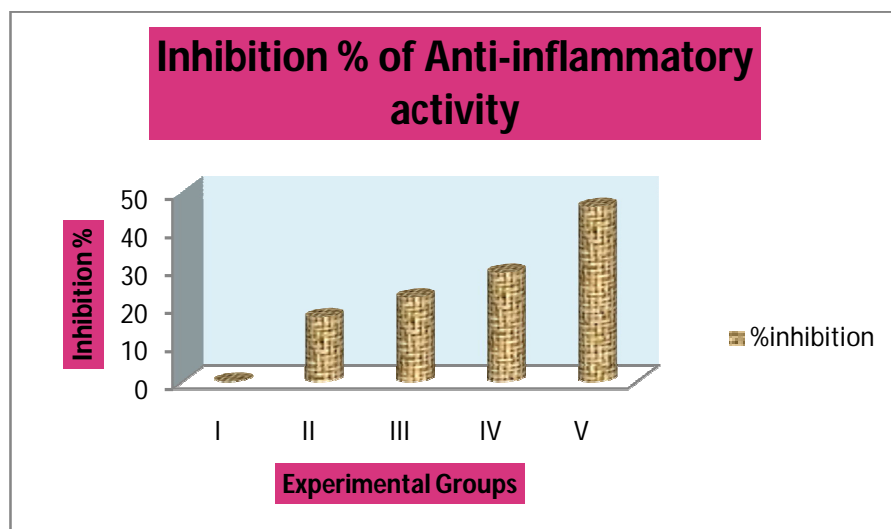
decreased the paw edema volume by 28.69% within 4 hour after administration, while standard drug decreased the paw edema volume by 46.05% when compared with the paw edema volume of control. The gymnemasylvestre extract at the dose of 200, 300, 400mg/kg produced significant reduction, when compared to the control group.

**Table 5. valuation of anti-inflammatory activity of *Gymnemasylvestre* leaves extract**

S.No.	Group	Paw volume(ml)	%inhibition
1.	Control	6.66±0.08	
2.	Gymnema sylvestre extract(200mg/kg)	5.74±0.11	16.95
3.	Gymnema sylvestre extract(300mg/kg)	5.07±0.05	22.16
4.	Gymnema sylvestre extract(400mg/kg)	4.98±0.08	28.69
5.	Diclofenac sodium(100mg/kg)	4.51±0.12	46.05



**Graph 1 Effect of *G.sylvestre* extract on Antiinflammatory activity of Paw volume(ml)**



**Graph2 Effect of *G.sylvestre* extract on inhibition % of Anti - inflammatory activity**

## DISCUSSION AND CONCLUSIONS

.Present study showed that the extract of *Gymnemasylvestre* extract caused antimicrobial activity against gram positive and gram negative bacteria. It also showed anti inflammatory activity in mice. It is also reported the significant anticancer activity and antimicrobial. The study is important for scientific community because this plant is used in traditional medicine to treat various diseases..

## REFERENCES

1. Agrawal, R.C. Pharmacological studies of *Moringa oleifera*. Asian Journal of current research 2021a, Vol 6 (3) 37-41
- 2 Agrawal, R.C. Antibacterial and Phytochemical studies of *Psidium guajava* leaf extracts, International Journal of Research- Granthalaya, 2021b, 9 (11) 1-4.
3. Agrawal, R.C. Pharmacological studies of *Azadirachta*

indica leaves extract. Open access research journal of Life Sciences.2022, 04 (01), 044-050.

4. Soni,,S and Agrawal R C : Antiproliferative effect of *Gymnemasylvestre* in in Vitro assays. International Journal of Research Publications. Vol 15 (1) October 2018

5.Bhatia AL, Raka K, Gulshan V, Sharma KV, Sharad V, Megha J (2008). Radio protective role of Gymnemic acid on mice: Study on Hepatic Biochemical Alterations. Asian J. Exp.Sci; 22(3):427-32.

6. Dateo G.P.and Long L. (1973), Gymnemic acid, the antisaccharine principle of *Gymnemasylvestre* Studies on isolation and heterogeneity of gymnemic acid A1. J. Agric. Food Chem.;21:899–903.

7.Gloria Y Yeh, David M. Eisenberg, Ted J. Kaptchuk, Russell S. Phillips (2003). Systematic Review of Herbs and Dietary Supplements for Glycemic Control in Diabetes. Diabetes Care; 26 (4): 1277-1294.

8.Jitender KM, Manvi FV, Alagawadi KR and Noolvi M (2008): Evaluation of anti-inflammatory activity of *Gymnemasylvestre* leaves extract in rats. Int. J. Green Pharmacy: 114-115.

9.Jitender KM, Manvi FV, Nanjwade BK, Alagawadi KR and Sanjiv S (2009): Immuno - modulatory activity of *Gymnemasylvestre* leaves extract on in vitro human neutrophils. J. Pharmacy

Res; 2(8):1284-6.

10.Kanetkar, P.V. Laddha, K.S. Kamat. M.Y (2004). Gymnemic acids: A molecular perspective of its action on carbohydrate metabolism. Poster presented at the 16th ICFOST meet organized by CFTRI and DFRL, Mysore, India.

11.Kiranmai M, Kazim SM and Ibrahim M (2011). Combined wound healing activity of *Gymnemasylvestre* and *Tagetes erecta* Linn. Int. J. Pharmaceu. Appl.; 2(2):135-140.

12.Luo H (1999). Possible participation of NO and EDHF in the 1.Agrawal,R.C. Pharmacological studies of *Moringa oliefera*. Asian journal of current Research .2021, Vol. 6 (3) 37-41.

UNDER PEER REVIEW

13. Omman, E., Shenoy, B.D., Vdupa, N., Kamath, R., Umadevi, P. (1999): Antitumour efficiency of cyclodextrin complexed and noisome encapsulated plumbagin in mice bearing melanoma B16F1. *Pharma pharmacol. Communication*, 5: 281-285.
14. Owens, C.W.I and belcher, R.V. (1965). Calorimetric micro method for the determination of glutathione. *Biochem. J.* 94(3): 705-711.
15. Paliwal, R Kathori, S Upadhyay. (2009). *B. Ethno-Med*, 3(2), 133-135.
16. Preuss, H.G. Bagchi, (2004). D Bagchi, M.. Rao, C.V. Dey, D.K Satyanarayana. *S. Diabetes ObesMetab*, 6(3), 171-80.
17. Puratchimani V and Jha S (2004b). HPTLC Standardization of *Gymnemasylvestre* R. Br. Using gymnestrogenin as reference, *Ind. J. Pharma. Sci.*, Vol. 66, pp. 164-166.
18. Rachh PR, Patel SR, Hirpara HV, Rupareliya MT, Rachh MR, Bhargava AS, Patel NM and Modi DC (2009): In-vitro evaluation of antioxidant activity of *Gymnemasylvestre* R.Br. leaf extract. *Rom. J. Biol. Plant Biol.*; 54(2):141-8.
19. Sinsheimer J.E., Subba R.G. (1970), Mc Ilhenny H.M. Constituents from *Gymnemasylvestre* Leaves V: Isolation and preliminary characterization of Gymnemic acids. *J. Pharm. Sci.*; 59:622-628.
20. Srikanth AV, Sayeeda M, Lakshmi NM, Ravikumar P and Madhava RB (2010): Anti cancer activity of *Gymnemasylvestre* R.Br. *Int. J. Pharmaceu. Sci. Nanotech*; 3(1):897-899.
21. Tandon Pankaj, Sirohi Anita (2010). Assessment of larvicidal properties of aqueous extracts of four plants against *Culex quinquefasciatus* larvae. *JJBS*; 3(1):1-6..

UNDER PEER REVIEW