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IN-VITRO ANTISPASMODIC EFFICACY OF ETHANOLIC EXTRACT OF LEAVES OF *SESBANIA GRANDIFLORA*

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ABSTRACT

The aim of this research was to use invitro pharmacological assay to provide the pharmacological basis for *Sesbania grandiflora* as an antispasmodic agent. It has been used to treat most illnesses in traditional medicine. The present study was undertaken to evaluate in-vitro antispasmodic activity of ethanolic extract of leaves *Sesbania grandiflora* by interpolation method on isolated chicken ileum. A kymograph reported the combined response of concentration to atropine and acetylcholine in the absence and presence of ethanolic extract. This revealed *Sesbania grandiflora's* ethanolic extract blocks the action of acetylcholine preventing impulses from the parasympathetic nervous system from entering smooth muscles and triggering contraction.

KEYWORDS: Antispasmodic Activity, In-vitro assay, *Sesbania grandiflora*, ethanolic extract.

1. INTRODUCTION

Spasm's definition is a sudden involuntary muscle contraction, a group of muscles, or a hollow organ like the heart. Many medical conditions, including dystonia, may induce a spasmodic muscle contraction. It is most often a muscle cramp that is followed by a sudden pain blast. In this disorder an Antispasmodics drugs that relax the smooth muscle of stomach, intestine, heart and bladder, used to treat indigestion not associated with peptic ulcers,

irritable bowel syndrome and of diverticular disease.[4] *Sesbania grandiflora* (also known as agate, syn. Aeschynomene grandiflora) or colibris / scarlet wisteria is a tiny tree in the *Sesbania* family. It is commonly referred to as caturay, corkwood, scarlet wisteria, sesban, colibris (English), hadga (hindi, Marathi).[2] Sesbania grandiflora belongs to the Fabaceae family and, because ofits medicinal qualities, is known in Bangladesh as "Bagful." Almost every part of S. Graniflora is a traditional medicine used to treat a wide range of diseases such as dysentery, stomatitis, fever, small pox, sore throat, migraine, etc.[1] The plant's methanol extract includes a variety of tanins, flavonoids, coumarins, hormones and triterpens. The herb used in the treatment of colic disease, jaundice, poisoning, smallpox, eruptive fever, epilepsy.[3] The dried leaves are often used to make tea and areconsidered antibacterial, anthelmintic, antitumor and contraceptive. Sesbania grandiflora leaves are highly nutritious and have been shown to contain significant quantities of triterpenoids, carbohydrates, vitamins, amino acids, proteins, tannins, saponins glycosides and steroids, fat, fiber and minerals such as steel, calcium and phosphorus.[8] The plant has also been reported to be a potent antidote for tobacco and smoking related disease.[1]

2. MATERIALS AND METHODS

2.1Plant Collection, Authentication and Extraction

The fresh leaf of *Sesbania grandiflora* was collected locally from Sangli district of Maharashtra, India and was authentified by Dr. S. M. Shendge at Balwant College, vita, India. A voucher specimen has been kept in the herbarium (Herbarium No 01 - 2018-19) at department of botany collected leaves were shade dried at room temperature for 7 days. The dried leaves of *sesbania grandiflora* then grinded to get fine powder using a grinder (Voltas-300). The grinded leaf powder was then processed with the aid of petroleum ether in a Soxhlet device to extract impurities, later powder was extracted from the Soxhlet device and dried for 5-10min for petroleum ether evaporation. After evaporation of petroleum ether, the powder samples were weighed and filled in clean extraction bag and placed in Soxhlet apparatus for extraction with the help of ethanol. This extraction process was repeated for 10 cycles. The extract were collected and dried in desiccators.[5]

2.2 Isolation of chicken Ileum

New whole good cock gastrointestinal tract was collected from an invita slaughterhouse. The caecum was lifted up, marking the ileocaecal junction. It was cut and removed a few centimeter of the ileum section and put in the watch glass containing physiological salt

solution immediately. The mesentery and adhering tissues were removed with gentle care. Almost care was taken to avoid any damage to the gut muscle. The ileum was cut into small segments of 2-3 cm long immersed in Tyrode's solution and cleaned off the mesentery. Respective segments of 2-3cm long were mounted in a 25ml tissue organ bath, filled with a mixture of 95% O2 and 5% CO2 and maintained at 37°C. The solution of tyrode (in Mm for lit.) was compared of 9 mg KCl, 0.1 mg NaCl, 0.1 mg NaHCO3, 0.42 mg NaH2PO4, 0.6 mg Glucose and 7.4 pH.[5][6]

2.3 Anti-Spasmodic Activity Assay Procedure

Secondly, concentration dependent acetylcholine responses (with a dosage of 0.1ml, 0.2ml, 0.4ml, 0.8ml, 1.6ml and 3.2ml) where recorded using Sherrington's frontal write lever recording drum. In the presence of plane Tyrode's solution as stock-I solution, contact time of 60 sec and base line of 30sec time cycle was chosen for proper recording of responses. Then same concentration dependent responses of acetylcholine (Ach) using same procedure for a mixture of Tyrode's solution+ *Sesbania grandiflora* extract (with a concentration of 1mg/ml) as a stock-II solution were recorded. Finally, the same concentration-dependent responses of Ach were reported as a stock-III solution for a mixture of Tyrode's solution + Atropine (as a generic anti-spasmodic agent).[5][7][9]

3. OBSERVATIONS AND RESULTS

Effect of standard drug (Ach) on excised chicken ileum reflected an increase in spasmodic activity (response) with an increase in dose as shown in Fig.-1.

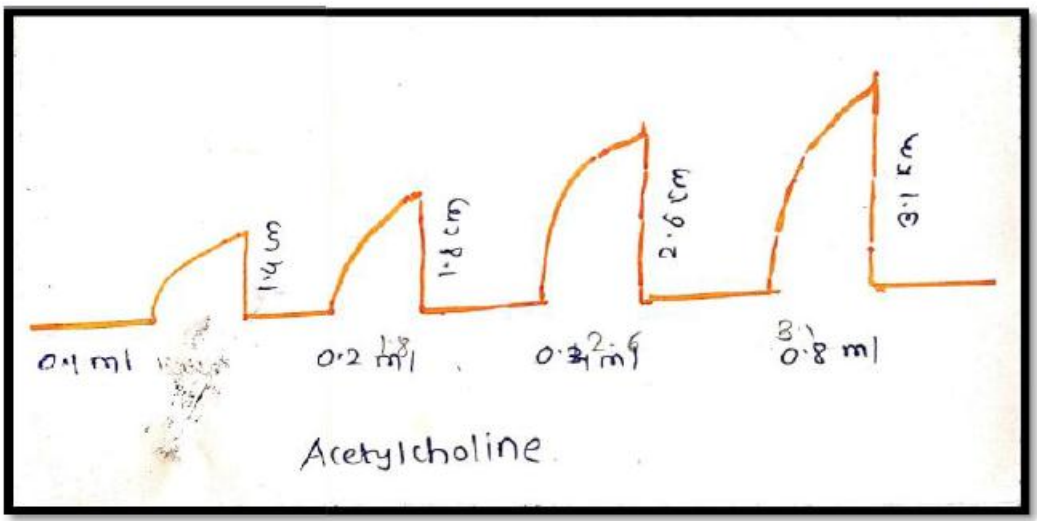


Fig no. 1: Response of curve acetylcholine.

Table No. 1: Dose response relationship observation of acetylcholine.

Sr. No.	Drug	Dose	Response in (cm)
1	Acetylcholine	0.1 ml	1.4 cm
2		0.2 ml	1.8 cm
3		0.3 ml	2.6 cm
4		0.4 ml	3.1 cm

Acetylcholine induced spasm followed by treatment of ethanolic extract of *Sesbania grandiflora* showed prominent antispasmodic activity as depicted in Fig.-2.

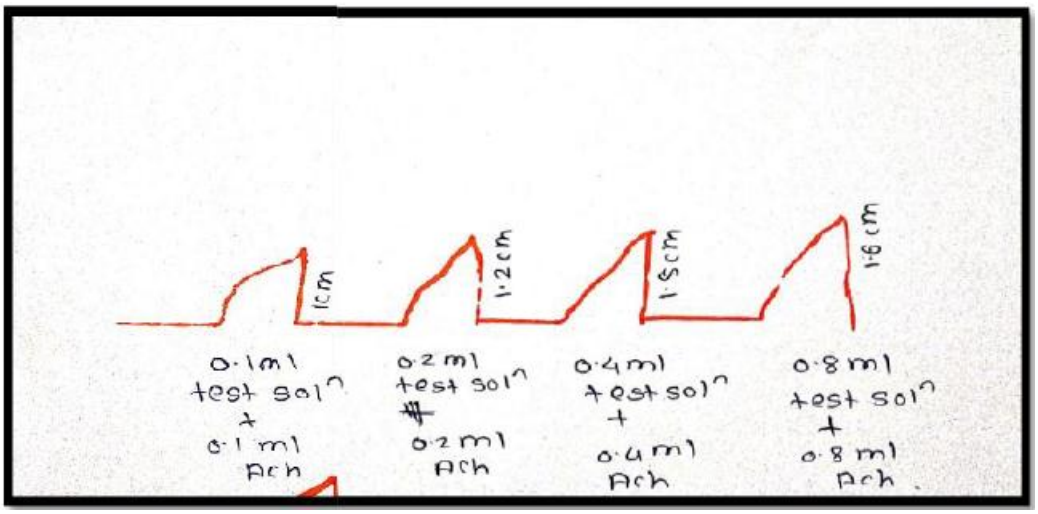


Fig 2: Response curves of Acetylcholine + Leaves extract.

Table 2: Dose response relationship observation of Acetylcholine and extract.

Sr. No.	Drug	Dose	Response in (cm)
1	Acetylcholine + Extract	0.1 ml + 0.1 ml	1 cm
2		0.2 ml +0.2 ml	1.2 cm
3		0.3 ml +0.3 ml	1.5 cm
4		0.4 ml + 0.4 ml	1.6 cm

While treatement of anticholinergic drug atropine (which is referred here as antispasmodic agent) showed expected receptor blocking action (antispasmodic) on isolated chicken ileum as shown fig. 3.

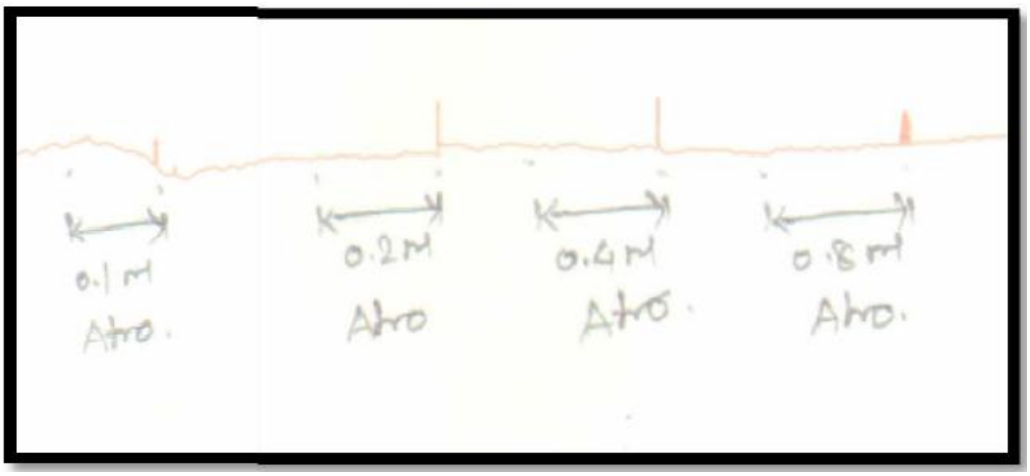


Fig. No. 3: Response Curve of Ethanolic Extract of *Sesbenia Grandiflora.*

Table no. 3: Dose response relationship of atropine.

Sr.No	Drug	Dose	Response in (cm)
1	Atropine	0.1 ml	-
2		0.2 ml	-
3		0.3 ml	-
4		0.4 ml	-

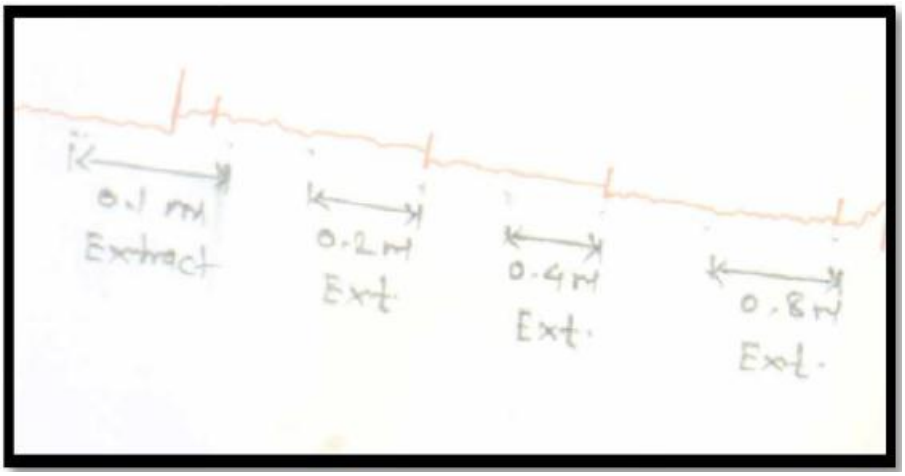


Fig No. 4: Response curve on ethanolic extract of *Sesbania grandiflora.*

Also treatment of ethanolic extract *of sesbania grandiflora* showed receptor blocking action (antispasmodic) as that of standard agent on isolated chicken ileum as shown in fig. no.4.

Table No. 4: Dose response relationship observation of *Sesbania grandiflora.*

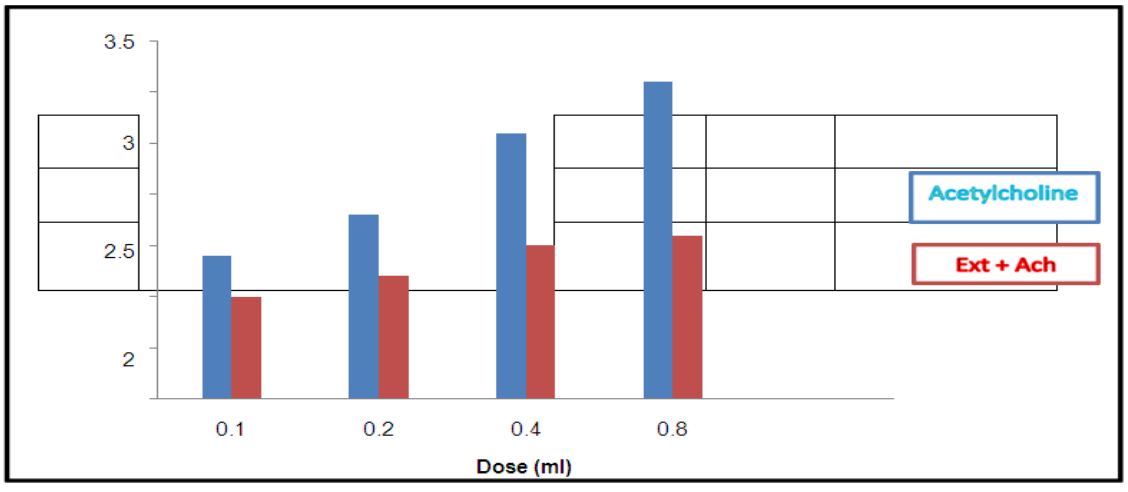
Sr. No.	Drug	Dose	Response in (cm)
1	*Sesbania grandiflora* ethanolic extract	0.1 ml	-
2		0.2 ml	-
3		0.3 ml	-
4		0.4 ml	-

4. DISCUSSION

From the current research observation and outcome, it was concluded that the ethanolic extractof *Sesbania grandiflora* exhibit promising antispasmodic activity. Standard drug (Ach) impact on excised chicken ileum reflected an increase in spasmodic activity (response) with an increase in additional dose i.e. the Acetylcholine (Ach) alone causes the contraction of excited chicken ileum, but a marked decrease in the contraction of ileum was noted when acetylcholine was administered in the presence of ethanolic leaves extracted from the *Sesbania grandiflora* linn. Plant. This reveled that ethanolic leaves extract of *Sesbania grandiflora* process of high degree of antispasmolytic activity by blocking cholinergic receptors.

Table No. 5: Comparative Dose Responses of Ach and Ach Followed by ethanolic Leaves Extract of *Sesbania grandiflora linn.*

Sr.no	Treatment given	Dose(ml)	Response	%Decreases in response
1	Acetylcholine	0.1	1.4	-
2		0.2	1.8	
3		0.4	2.6	
4		0.8	3.1	
5	Ethanolic extract + Acetylcholine	0.1+0.1	1	28.58
6		0.2+0.2	1.2	33.34
7		0.4+0.4	1.5	42.31
8		0.8+0.8	1.6	48.39



Graph no. 1: Graphical representation of response of Acetylcholine and Extract + Ach.

5. CONCLUSION

From all observations and results obtained for the present study it was concluded that ethanolic leaves extract of *Sesbania grandiflora* exhibits promising anti-spasmodic

activity. Also when compared with a standard anti-spasmodic agent (atropine), it was found that *Sesbania grandiflora* has spasmolytic activity like same as atropine atropine. Since many anti-spasmodic medications on the market have side effect as urinary delay, urinary retention, mydriasis, tachycardia, blurred vision and reactions to hypersensitivity; so, Sesbania *grandiflora* being an herbal origin drug with high degree of safety and efficacy could be a suitable alternative to existing drugs, as well as being a new antispasmodic familymember.

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