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ANTISPASMODIC EFFECT OF N-BUTANOL EXTRACT OF SOLENOSTEMMA ARGEL HYNE LEAVES ON ISOLATED RAT ILEUM.

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ABSTRACT

The present study was aimed to provide the pharmacological basis for medicinal use of *Solenostemma argel Hyne* leaves, as an antispasmodic agent using in-vitro pharmacological assay. Antispasmodic activity of n-butanol extract of Solenostemma argel Hyne leaves was evaluated for its antispasmodic effects against acetylcholine- induced contractile activity on isolated rat ileum preparations. The extract inhibited the contractile response elicited by acetylcholine on rat ileum significantly and in a concentration-dependent manner. The present study results revealed that, n-butanol extract of Solenostemma. argel Hyne leaves possesses a promising antispasmodic action confirming and justifying its use in folk medicine for the treatment of gastrointestinal disorders.

KEYWORDS: Spasmolytic effect; Solenostemma argel Hyne; Rat Ileum.

INTRODUCTION

Several plants are used in folk medicine for the treatment of gastrointestinal disorders such as spasm because of their antispasmodic activity. Solenostemma argel Hyne (Fam. Asciepiaceae) is one of those plants that has been used over centuries as a herbal medicine for the treatment of gastrointestinal disturbances. Phytochemical screening of the plant has revealed that, triterpenes, alkaloid, cuomarins, saponines, tannins and sterols are the main constituents of the plant. The present study aimed to evaluate n-butanol extract of Solenostemma. argel Hyne for its antispasmodic activity against acetyl choline induced

contraction on isolated rat ileum to provide a scientific evidence for the rational use of it in the treatment of gastrointestinal ailments.

MATERIALS AND METHODS

Plant: The plant was collected from the farm of Medicinal and Aromatic Plants and Traditional Medicine Research Institute (MAPTMRI) and authenticated by the harberium team of (MAPTMRI).

Extraction: Collected leaves of *Solenostemma argel Hyne* plant were shade dried at room temperature. The dried leaves were then grinded to get a course powder, then n-butanol Fraction was carried out according to method descried by Sukhdev *et. al.* (2008): 30 g of the ethanolic extract was dissolved in 500 ml of distilled water and shacked, three times with 100 ml of Petroleum ether each time using reparatory funnel. Petroleum ether layers were combined together and evaporated under reduced pressure using rotary evaporator apparatus. Aqueous layer was then re-shacked three times with 100 ml of chloroform in each time using reparatory funnel. Chloroform layers were combined together and evaporated under reduced pressure using rotary evaporator. Aqueous layer was then re-shacked, three times with 100 ml of ethyl acetate in each time using reparatory funnel. Ethyl acetate layers were combined together and evaporated under reduced pressure using rotary evaporator apparatus. Aqueous layer was finally shacked, three times with 100 ml of n-butanol in each time using reparatory funnel. Butanol layers were combined together and evaporated under reduced pressure using rotary evaporator apparatus. Then the yield was kept in the refiregarater for experimental use.

Animals: Albino Wister Rats (150–200 g) fed on standard diet and water ad libitum in the animal house of (MAPTMRI) have been used.

Methods: Adult male albino Wister rats weighing (200-250) g. were used for this study. Rats were fasted for 24 hr before the experiment. Sacrificing was done by cervical dislocation. The abdomens excised immediately, and the ileum was removed. The terminal portions, of about 10 to 20 mm in length, were taken after discarding the 15 cm portion nearest to the ileum-caecal junction (Schlemper, V., et al 1996). The intestinal content was eliminated by washing with Tyrode solution and the mesenteric residues were removed ,then each was mounted in a 25 ml tissue organ baths containing Tyrode solution at 37°C, continuously bubbled with 95% O2 and 5% CO2 under 1g of load, care should be taken that the lumen of the ileum is kept open. Then the tissues kept undisturbed for an equilibrium

period of 30- 45 minutes prior to drug addition. Transducers were connected to an amplifier to amplify the magnitude of contractions. Concentration dependent responses of acetylcholine were recorded with doses of (5ng, 10ng, 20ng, 40ng, 80ng, 160ng) using an oscillograph recording system. Contact time 60 sec. and base line of 30 sec. time cycle were adopted for proper recording of response. Sub maximal dose response (20ng) of acetylcholine was selected. Then the concentration dependent response of the extract followed by sub maximal dose of acetylcholine was recorded. Lastly the concentration dependent response of atropine followed by sub maximal dose of acetylcholine was recorded.

RESULTS

Acetylcholine induced contraction of excised rat ileum. The amplitude of the contraction increased due to increase of Ach dose till it reached the maximum response at a dose 40ng/ml. When Acetylcholine was given at a dose of 20ng/ml (sub maximal dose) in presence of n-butanol extract of *Solenostemma argel Hyne* at a dose 1mg/ml, the amplitude of the sub maximal dose of Ach has been markedly decreased. Moreover, The amplitude of the sub maximal dose of Ach has completely abolished at a dose 4mg/ml .Also the sub maximal dose of Ach has been markedly decreased, when given with atropine at a dose 8ng/ml and has completely diminished at a dose16ng/ml .

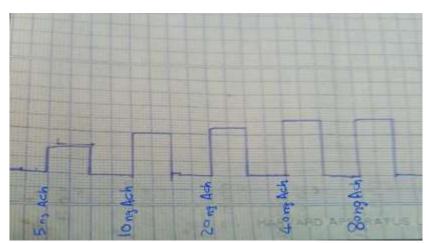


Fig 1: Response Curves of Acetylcholine on isolated rat ileum

Table 1: Dose Response Relationship of Acetylcholine on isolated rat ileum

Sr. No	Drug	Dose	Response (cm)
1		5 ng	0.9
2		10 ng	1.5
3	Acetylcholine	20 ng	1.8
4		40 ng	2.0
5		80 ng	2.0

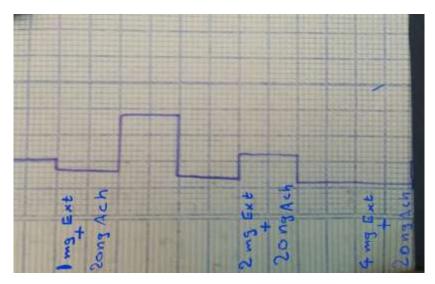


Fig 2: Response Curves of Acetylcholine + Leaves Extract on isolated rat ileum.

Table 2: Dose Response Relationship of Acetylcholine and Extract on isolated rat ileum.

Sr. No	Drug	Dose	Response (cm)
1		1mg extract+20 ng Ach.	1.1
2	Extract+Acetylcholine	2mg extract+20 ng Ach.	0.4
3		4 mg extract+20 ng Ach.	0.0

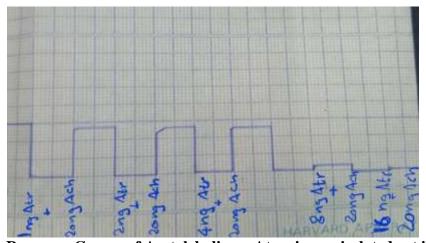


Fig 3: Response Curves of Acetylcholine + Atropine on isolated rat ileum.

Table 3: Dose Response Relationship of Acetylcholine and Atropine on isolated rat ileum.

Sr. No	Drug	Dose	Response (cm)
1		1ng Atropine+20ng Ach	1.4
2	Atropine +Acetylcholine	2ng Atropine+20ng Ach	1.3
3		4ng Atropine+20ng Ach	1.25
4		8ng Atropine+20ng Ach	0.2
5		16ng Atropine+20ng Ach	0.0

RESULTS DISUSSION

Intestinal motility is control by circulating hormons, intrinsic and extrinsic nerves that release a variety of transmitters. The most important excitatory transmitter in gastrointestinal tract is acetylcholine which act via muscarinic receptors in enteric smooth muscle cell membranes (Guyton,.) 2006. The results have showed that, acetylcholine elevated the amplitude of the base line contration in concentration dependent manner as shown in (Fig.-1.) However after pretreated the specimen with the extract, the sub maximal effect decreased as the extract dose increased till it has been completely abolished at a dose 4mg/ml of extract as shown in (Fig 2.) Also when the sub maximal dose of acetyl choline was given after atropine (as standard anti-spasmodic agent), it decreased as the dose of atropine was increasing till abolished at 16ng/ml atropine, shown in (Fig 3.).

Effect of Acetylcholine on isolated rat ileum reflected an increase in spasmodic activity (response) with an increase in dose. Different mechanisms are involved in gastrointestinal smooth muscle relaxation. These included the blocking action on excitatory agents and inducing anticholinergic (Unno, et al 2006) or antihistaminic (Sá-Nunes, et al 2003) effect. According to the findings of the present study concluded that, Solenostemma argel Hyne leaves extract possess a high degree of spasmolytic (anti-spasmodic) activity by blocking cholinergic receptors.

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