



EVALUATION OF ANTI-SPASMODIC ACTIVITY OF PEPPERMINT OIL IN AN ISOLATED CHICK ILEUM

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ABSTRACT:

Background

Mentha plants are one of the most popular plants used since ancient times for their medicinal and aromatherapy properties. The essential oil of peppermint, which is the most important Mentha specie, has been reported to have antioxidant, antidiabetic and anticarcinogenic effects⁽¹⁾.

Method

The antispasmodic activity of Peppermint oil (PO) was checked *in vitro* on chick ileum against spasmogen, acetylcholine. The activity was compared with standard antispasmodic agent, atropine. The effectiveness of various doses of PO that cause inhibition in isolated chick ileum contractions was evaluated.

Result

The study showed that the PO dose-dependently decreased the amplitude of acetylcholine-induced contractions. The cumulative application of PO at different concentrations caused inappropriate relaxation to the gradual concentration response in the ileum.

Conclusion

The study demonstrated that PO were able to prevent contraction induced by acetylcholine in isolated chicken ileum. The *in vitro* results tend to support the use of PO as an antispasmodic agent.

Keywords - Acetylcholine, Antispasmodic agent, Atropine, Contraction, Spasmogen

INTRODUCTION

Antispasmodic compounds are currently used to reduce anxiety, emotional and musculoskeletal tension, and irritability. Although most of the available antispasmodic compounds are synthetic or semisynthetic, traditional uses of this group of compounds are still popular. Mentha plants are one of the most popular plants used since ancient times for their medicinal and aromatherapy properties. The essential oil of peppermint, which is the most important Mentha specie, has been reported to have antioxidant, antidiabetic, antibacterial, antimutagenic, antifungal, and anticarcinogenic effects⁽¹⁾. It is also stated that peppermint has a beneficial effect on the absorption surface area by improving the small intestine length and villi structure in quails. These plants are used for the treatment of gastrointestinal (GI) system diseases. The present study was aimed to investigate the anti-spasmodic effect of PO of the family Lamiaceae on isolated chicken ileum by an *in vitro* pharmacological assay. PO is available throughout India with countless hidden

therapeutic values. It is used in traditional system for antidiabetic, irritable bowel syndrome, anticarcinogenic and as an antioxidant properties. Clinically, PO other postoperative adjuvant therapy. Due to the phytochemical property of the oil, the extraction and isolation of PEO are quite tricky but important for the research and development of PEO. Transdermal absorption following topical administration of PEO is critical, which influences the effectiveness of PEO applications in clinical settings, particularly in dermatologic disease, such as burn wounds and psoriasis⁽²⁾.

MATERIALS AND METHODS

Materials

Atropine (ATR), Acetyl choline (Ach), Peppermint oil (PO), Freshly prepared Tyrode solution

Experimental Protocol

Isolated chicken ileum preparation: The fresh chicken ileum was collected from local slaughter house in Tyrode solution and cleaned off the mesentery. The segment of 2cm long was mounted in a 20ml tissue organ bath and maintained at 37°C. The tissue was allowed to equilibrate for 30 min, during which, the bathing solution was changed at every 10 min. Contact time of 60 sec, and base line of 30sec time cycle were opted for proper recording. Dose response curve of acetyl choline was recorded on the kymograph first. Then cumulative concentration-effect curves were recorded on kymograph for Acetyl choline (100µg/ml) in absence and presence of peppermint oil (100µg/ml) on Kymograph by using Sherrington's Recording Drum. The same procedure was carried for concentration-effect curve of Ach in presence of Atropine sulphate as a standard drug. The percentage inhibition of PO and standard drug was calculated and graph was plotted by taking log dose verses height of response curve.

RESULT AND DISCUSSION

Evaluation of Antispasmodic activity

Dose response relationship observations of Ach, Peppermint oil (PO) and atropine on chicken ileum.

DRUG	DOSE	PERCENTAGE RESPONSE
Acetylcholine	0.1	68.5%
	0.2	85.93%
	0.4	93.75%
	0.8	100%
Atropine + Acetyl choline	0.1+0.1	22.22%
	0.1+0.2	33.33%
	0.1+0.4	44.44%
	0.1+0.8	55.55%
PEO+Acetyl choline	0.1+0.1	18.75%
	0.1+0.2	17.18%
	0.1+0.4	12.5%

	0.1+0.8	9.3%
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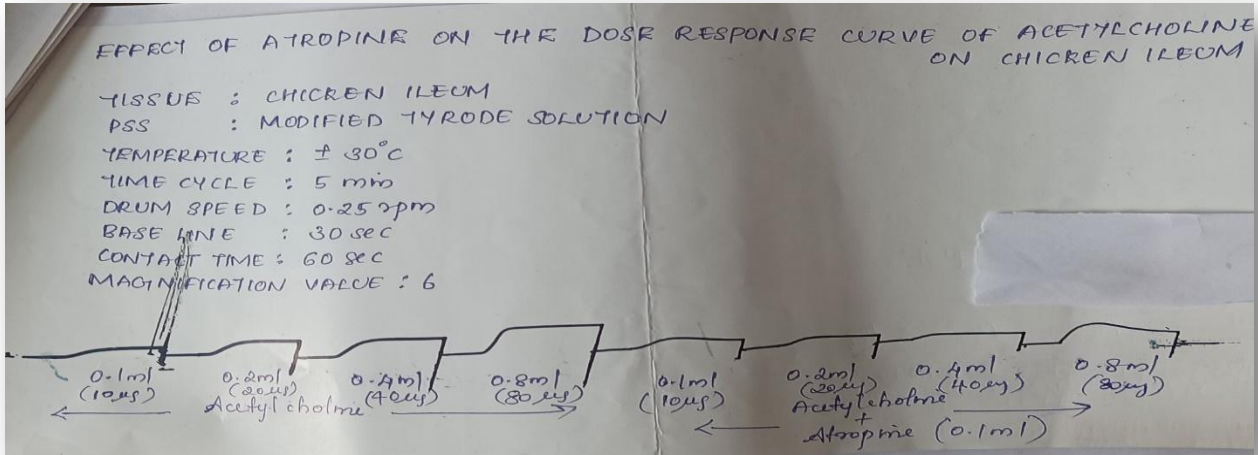


Fig. 1: Dose response curve of Ach and Atropine.

Effect of Ach on chicken ileum reflected an increase in spasmodic activity (response) with an increase in the dose. DRC of Ach in presence of atropine was taken as shown in figure.

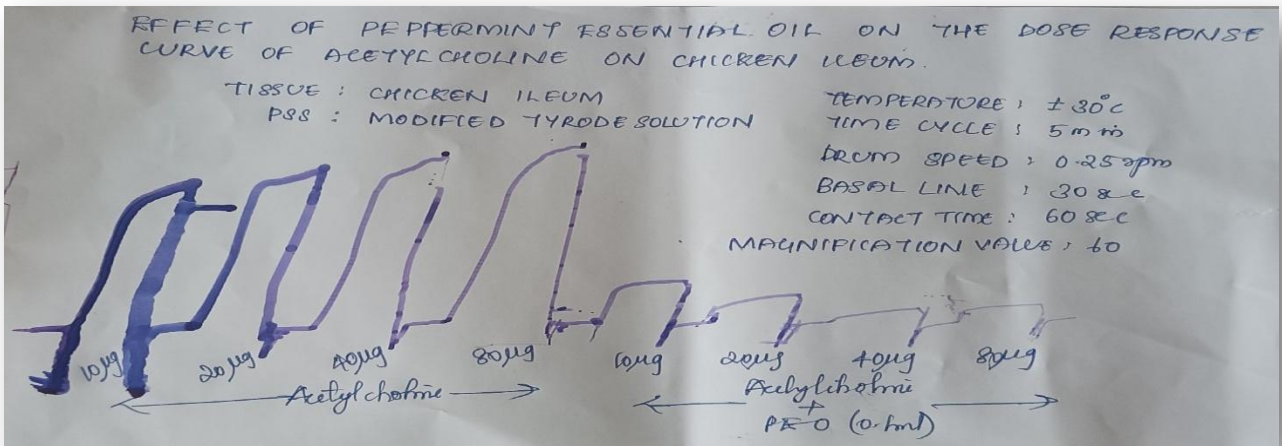
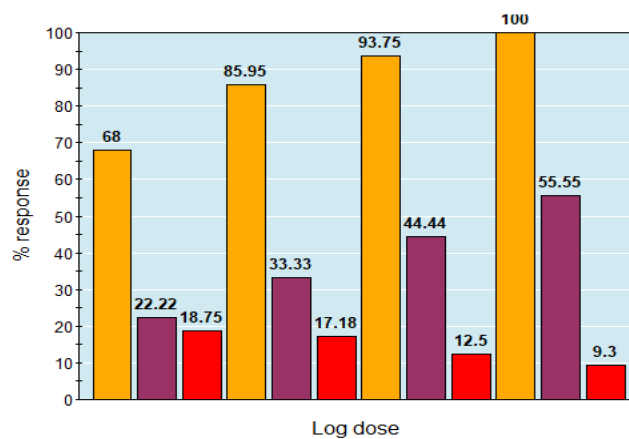


Fig. 2: Response curve of Ach+ peppermint oil.

Ach induced spasm followed by the treatment of showed prominent antispasmodic activity as shown in the figure.

Effect of PEO on DRC of Ach & Atropine on Chicken ileum



Results were compared with standard drug atropine, an anti-spasmodic drug. But there is no dose dependent increase in antispasmodic activity is seen in the 3rd and 4th treatment of PO.

DISCUSSION

For screening the activity of a drug on intestinal smooth muscles, chick ileum preparations can be used. Chicken intestine is easier to acquire, handle and easier to dissect and has the same reactions to spasmogenic and spasmolytic drugs. Cholinergic agonists like acetylcholine elicit a contractile response in isolated chicken ileum⁽³⁾. M₃ receptor, a subtype of cholinergic (muscarinic) receptor activation causes contraction of intestinal smooth muscle. The M₃ receptor function through Gq protein and trigger membrane bound phospholipase C (PLC) provoking inositol triphosphate (IP₃) and diacylglycerol (DAG) which in succession release Ca²⁺ intracellularly leads to actin-myosin phosphorylation causing increased smooth muscle tone. Thus, the contraction of intestinal smooth muscle in vitro has often been utilized for the study of contractile/dilator responses of agonists as well as antagonist. In current investigation, acetylcholine showed greater contraction while peppermint oil significantly inhibited the acetylcholine induced contraction on isolated chicken ileum preparation. The parallel shift of graph towards right side in acetylcholine dose-response curves in the presence of increasing concentrations of PO indicating that there was competitive antagonism between acetylcholine and PO for M₃ receptors present on the smooth muscle. This effect may be due to its anti muscarinic or antispasmodic activity⁽⁴⁾. Acetylcholine elicited contractions in ileum are believed to be mediated through M₃ receptors present on ileum. It was found that peppermint oil produced dose dependent inhibition of ileum contractions induced by acetylcholine. The parallel rightward shift in agonist concentration response curves of acetylcholine in presence of increasing concentrations of PO was indicating antispasmodic activity (anti-muscarinic). The inhibition may be due to the antagonism of cholinergic-muscarinic receptors or nonspecific spasmolytic action of PO.

Conclusion

It can be concluded from the study that the PO possesses significant anti-spasmodic activity that may be due to the M₃-antagonism (cholinergic-muscarinic-antagonism) on smooth muscles of chicken ileum. This may substantially benefit in the treatment of spasm and other intestinal muscular disorders.

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