Research Article

Evaluation of Antispasmodic Potential of Polyherbal Formulation

Prashant Shamkuwar

Department of Pharmacognosy, Government College of Pharmacy, Vedant Road, Aurangabad, Maharashtra, INDIA.

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ABSTRACT

The aim of the present study was to investigate the antispasmodic efficacy of polyherbal formulation prepared using extract of *Apium graveolens*, *Helicteres isora* and *Mentha piperita* with excipients. Antispasmodic effectiveness of polyherbal formulation was assessed by inhibition of the spasams induced by different spasmogens. The spasmolytic activity of polyherbal formulation was studied in isolated guinea pig ileum model using acetyl choline, nicotine and histamine as agonist. Effect on gastrointestinal motility was assessed by charcoal meal test in mice. The results of the study demonstrated that polyherbal formulation showed significant reduction of spasm induced by acetyl choline, nicotine and histamine in isolated guinea pig ileum. Polyherbal formulation has also inhibited the intestinal motility in mice. Polyherbal formulation was found to be the potent spasmolytic medication. Mechanism of the antispasmodic activity of polyherbal formulation is non-specific and may be mediated by inhibiting the muscarinic, nicotinic and histamine receptors.

Key words: Antispasmodic, Acetyl choline, Histamine and Polyherbal formulation, Nicotine.

Correspondence:
Dr. Prashant Shamkuwar,
Associate Professor, Government College of Pharmacy, Vedant Road, Aurangabad-431005, Maharashtra, INDIA.
Phone no: +91-2346820

Email id: shamkuwarp@rediffmail.com

INTRODUCTION

Spasms are continuous smooth muscle contraction induced by endogenous histamine and acetyl choline.[1] It causes discomfort and uneasiness which may further leads to irritation and inflammation of gastrointestinal tract, colic, abdominal pain and it could even result in to threatening condition such as gastritis, colitis and irritable bowel syndrome.[2] It is well know that excessive infant crying may be due to abdominal pain on account of spasms, cramping, gas and abnormal intestine contraction.[3]

An antispasmodic is a drug that aims to control gut spasm by reduction of excessive smooth muscle contraction causing discomfort and cramping in the abdominal area. It gives relief not only for spasm, but also for bloating and abdominal pain and limits the movement of the intestine.[4] Despite the clinical efficacy of variety of synthetic antispasmodic drugs which have been prescribed worldwide, the use of these medicines is often restricted by the occurrence of unpleasant and sometimes severe side effects which may turn down the patient compliance and diminish the effectiveness of treatment. There is a great demand for the development of new, more efficacious, low-cost and safe antispasmodic medicine to overcome the constraints of existing therapy.[5,6]

Apium graveolens, *Helicteres isora* and *Mentha piperita* are the medicinal plants used in Ayurveda for abdominal discomfort, colic pains, indigestion, vomiting, hepatobiliary disorders, diarrhea and dysentery.[7-9] Hence further characterization of antispasmodic activity is needed on scientific basis to evolve medicinal preparation from combined extracts of these plants. Thus the present study was undertaken to investigate the spasmolytic effects of medicinal plants extracts together as polyherbal formulation in solution form in isolated guinea pig ileum model using acetyl choline, nicotine and histamine as spasmogens and charcoal meal test in mice.

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MATERIALS AND METHODS

Drugs and chemicals

Acetyl choline (Sigma Chemicals Ltd.), Nicotine (Sigma Chemicals Ltd.), Histamine (Sigma Chemicals Ltd.), Atropine sulphate (Sigma Chemicals Ltd.) and Activated Charcoal (E. Merck). All other chemicals and reagents used in the studies were of analytical or laboratory grade.

Plant collection and identification

The selected crude plant materials *Apium graveolens* (seeds), *Helicteres isora* (fruits) and *Mentha piperita* (leaves) were procured from local market and same were authenticated by Herbarium in charge, Department of Botany, Dr. Babasaheb Ambedkar Marathwada University, Aurangabad (M.S.), India.

Preparation of extracts

The plant materials were cleaned, shade dried and powdered by mechanical means. 100 gm standardized powder of each *Apium graveolens* (seeds), *Helicteres isora* (fruits) and *Mentha piperita* (leaves), were subjected to extraction by maceration with hydroalcohol (60%). Thereafter the extract was filtered and concentrated at room temperature.

Development of Polyherbal formulation

Oral solution containing hydro alcoholic extract (6% w/w) with suitable excipients like Sorbic acid (0.2 %), butylated hydroxyanisole (0.2 %), sodium saccharin (0.1 %), Chocolate flavor (q.s.) was prepared by dissolving all these ingredients in water up to 100 ml. Polyherbal formulation in solution form was standardize by using different organoleptic characters such as color, odor and taste as well as physicochemical parameters like pH, visibility in light and gas evolution studies.

Animals

Guinea pigs (300- 400 gm each) of either sex was used for *in vitro* antispasmodic study. Albino mice of either sex, weighing between 25-35 gm were used to study the effect of the polyherbal formulation on gastrointestinal motility in mice. Animals were kept in standard environmental condition, fed standard laboratory diet and water *ad libitum*. All experiments were performed after an overnight fast. The protocol of study was approved by the institutional animal ethical committee (GCPA/IAEC/2011/235).

Acute toxicity study

Polyherbal formulation studied for acute oral toxicity as per revised OECD guidelines number 423. Polyherbal formulation was devoid of any toxicity up to 20 ml/kg in albino mice by oral route.

Experimental procedures for antispasmodic activity

Effect of polyherbal formulation on stimulant effect of acetyl choline, nicotine and histamine in isolated guinea pig ileum

The *in vitro* method was conducted to study the effect of polyherbal formulation on acetyl choline, nicotine and histamine induced contractions in isolated guinea pig ileum. Guinea pigs were sacrificed by a cervical blow and dislocation. A piece of ileum was removed and suspended under a constant tension of 1 g in organ baths containing 10 ml Tyrode solution at 37°C. Composition of Tyrode solution solution in mili mole was KCl 2.68, $NaCl$ 136.89, $CaCl_2$ 1.36, $MgCl_2$ 1.05, $NaHCO_3$ 11.90, NaH_2PO_4 0.32 and glucose 5.55. Tissue was aerated and allowed to stabilize for 30 min with an initial load of 0.5 g before starting of the experiment. Normal response of the ileum was recorded on a student's physiograph through isometric force transducer. Effect of the polyherbal formulation was tested on the contractions of guinea pig ileum induced by acetyl choline (1μM), nicotine (2 μg/ml) and histamine (1μg/ml).[10,11]

Effect of polyherbal formulation on gastrointestinal motility

In an *in vivo* method, effect of polyherbal formulation on the motility of intestine is evaluated by charcoal meal test in mice. Charcoal meal test is the passage of charcoal meal along the small intestine. Albino mice were divided in to five groups containing six animals each. Vehicle control group I received 20 ml/kg, p.o. of normal saline. Polyherbal formulation was given to test group II, III and IV at a dose of 2, 4 and 8ml/kg p.o. Standard Group V received Atropine sulphate5 mg/kg, p.o. After 30 min, mice of all the groups received 0.2 ml of charcoal meal (10% charcoal suspension in 5% gum acacia). Mice were sacrificed 30 min after the administration of charcoal meal and the intestine was removed. Percentage distance traveled by the charcoal meal relative to the total length of the small intestine in all groups was determined.[12,13]

Statistics

The results of all experiments were expressed as mean ± standard error of mean. Data analysis was carried out using Students '*t*'-test. A level of significance of $P < 0.05$ was considered as statistically significant.

RESULTS

Effect of polyherbal formulation on stimulant effect of acetyl choline, nicotine and histamine in isolated guinea pig ileum.

Polyherbal formulation inhibited the contractile response of guinea pig ileum induced by acetylcholine, nicotine and histamine. Polyherbal formulation (0.3 ml/ml) decreased the stimulant effect of acetyl choline (1μM), nicotine (2 μg/ml) and histamine (1μg/ml) on isolated guinea pig ileum by 82.99% and 71.07% and 81.01%, respectively (Table 1).

Effect of polyherbal formulation on gastrointestinal motility.

The results of charcoal meal test indicated that polyherbal formulation reduced intestinal transit time in mice dose dependently by decreasing gut motility. Polyherbal formulation produced significant inhibition of the distance moved by the charcoal meal by 25.39%, 33.05% and 56.36% at doses of 2 ml/kg, 4 ml/kg and 8 ml/kg, respectively. Atropine sulphate at a dose of 5 mg/kg produced inhibition of intestinal transit by 60.43% as shown in Table 2.

DISCUSSION

Currently researchers are trying to explore development of new polyherbal therapy because of its medicinal and therapeutic application. The results of this study indicated that the prepared polyherbal formulation is an effective antispasmodic remedy. Antispasmodic activity of polyherbal formulation was evaluated by using isolated guinea pig ileum model and charcoal meal test.[14,15]

Table 1: Effect of polyherbal formulation on stimulant effect of acetyl choline, nicotine and histamine on guinea pig ileum.

Group	Dose (/ml)	Effects on tissue	Contraction (gm)	Inhibition (%)
Normal			1.13 ± 0.03	
Acetyl choline	1 μM	Contraction	2.47 ± 0.04	
Formulation + Acetyl choline	0.3 ml 1 μM	Relaxation	0.42 ± 0.02	82.99
Nicotine	2 μg	Contraction	2.04 ± 0.03	
Formulation + Nicotine	0.3 ml 2 μg	Relaxation	0.59 ± 0.02	71.07
Histamine	1 μg	Contraction	2.16 ± 0.04	
Formulation + Histamine	0.3 ml 1 μg	Relaxation	0.41 ± 0.03	81.01

Values are mean ± standard error of mean. Each value represents average of six determinations. $P < 0.05$ vs. control, student's 't' test.

Table 2: Effect of polyherbal formulation on intestinal transit in mice.

Group	Dose (mg/kg)	Percent intestinal transit	% Inhibition
Control		62.38 ± 1.86	
Formulation	2 ml	46.54 ± 1.27	25.39
Formulation	4 ml	41.76 ± 1.52	33.05
Formulation	8 ml	27.22 ± 0.91	56.36
Atropine sulphate	5 mg	24.68 ± 0.88	60.43

Values are mean ± standard error of mean. Each value represents average of six determinations. $P < 0.05$ vs. control, student's 't' test.

Parasympathetic stimulation increase the activity in the entire enteric nervous system causing increasing overall blood flow to the gut as well as increasing secretions and general gut activity. Parasympathetic nervous system, mediates its action in the gut via stimulation of muscarinic receptors and nicotinic receptors.[16,17] In the gastrointestinal tract acetyl choline produces its stimulant effect through muscarinic receptors while nicotine produce stimulant effect by nicotinic receptors.[18,19] The contractile effects of acetylcholine and nicotine on isolated guinea pig ileum were inhibited by polyherbal formulation.

Histamine is synthesized in the mast cells and occurs throughout the gastrointestinal tract. It is released during the inflammatory processes causing increase capillary permeability and stimulates the secretion of gastric juice. It causes contraction of smooth muscle of the alimentary tract by binding to histamine receptors.[20,21] Polyherbal formulation produced relaxation against histamine induced contraction in guinea pig isolated ileum.

Gastrointestinal motility is the result of coordinating contractions and relaxations of the smooth muscles in the gut. Peristalsis and segmentation are the two distinct patterns of gastrointestinal contraction. The regulation of gastrointestinal motility is complicated and involves enteric nervous system, hormones, paracrine substances and inflammatory mediators.[22,23] Effect of polyherbal formulation on the motility of intestine is evaluated by charcoal meal test in mice. Polyherbal formulation was found to be the inhibitor of intestinal motility.

CONCLUSION

In present study polyherbal formulation has shown comparable antispasmodic activity to the standard drug atropine by inhibiting the gastrointestinal motility in mice. Antispasmodic activity of polyherbal formulation is non-specific and may be mediated by inhibiting the muscarinic, nicotinic and histamine receptors.

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CONFLICT OF INTEREST

The author declare no conflict of interest.

SUMMARY

This study reports on the antispasmodic efficacy of the polyherbal formulation prepared using extract of *Apium graveolens*, *Helicteres isora* and *Mentha piperita*. The results of the spasmolytic activity of polyherbal formulation studied in isolated guinea pig ileum model using acetyl choline, nicotine and histamine as spasmogens and charcoal meal test in mice, showed the significant reduction of spasm and inhibition of the intestinal motility.

REFERENCES

1. Saini N, Singh GK. Pharmacological evaluation of a polyherbal formulation for its antispasmodic activity. International Journal of Green Pharmacy. 2015;9(2):104-9.
2. Scarpignato G, Pelosini I. Management of irritable bowel syndrome: Novel approaches to the pharmacology of gut motility. Can J Gastroenterol. 1999;13(Suppl A):50A-65A.
3. Mamadou G, Meddah B, Nzouzi I, Nicolas SA, Haj A, *et al*. Antispasmodic phytomedicine, from traditional utilization to rational formulation: Functional approach. Phytopharmacology. 2011;1(3):20-35.
4. Heghes SC, Vostinaru O, Rus L, Mogosan C, Adela C, Lorena F. Antispasmodic Effect of Essential Oils and Their Constituents: A Review. Molecules. 2019;24(9):1675-9.
5. Koloski NA, Talley NJ, Boyse P. The impact of functional gastrointestinal disorders on quality of life. The American Journal of Gastroenterology. 2000;95(1):67-71.
6. Sisay M, Bussa N, Gashaw T. Evaluation of the Antispasmodic and Antisecretory Activities of the 80% Methanol Extracts of *Verbena officinalis* L: Evidence From *in vivo* Antidiarrheal Study. Journal Evidance Based Integrative Medicine. 2019;24:1-7.
7. DeSousa A, Soares P, Almida A, Alana M, Souza E, Maria A. Antispasmodic effect of *Mentha piperita* essential oil on tracheal smooth muscle of rats. Journal of Ethnopharmacol. 2010;130(2):433-6.
8. Pohocha N, Grampurohit ND. Antispasmodic activity of the fruits of *Helicteres isora* Linn. Phytotherapy Research. 2001;15(1):49-52.
9. Al-Asmari AK, Athar MT. An updated phytopharmacological review on medicinal plant of arab region: *Apium graveolens* Linn. Pharmacognosy Review. 2017;11(21):13-8.
10. Seung D, Chang S, Baek H, Baek S, Sung Y, Chung W. Antidiarrhoeal and spasmolytic activities and acute toxicity study of Soonkijangquebo, a herbal antidiarrhoeal formula. Journal of Ethanopharmacology. 2004;91(1):75-80.
11. Jung H, Wen-Yuan G, Ling N, Liu C. Antidiarrhoeal and intestinal modulatory activities of Wei-Chang-An-Wan extract. Journal of Ethanopharmacology. 2009;125(3):450-5.
12. Shamkuwar P, Shahi S. Evaluation of antidiarrhoeal activity of Mebarid: An ayurvedic formulation. International Journal of Pharmacy and Pharmaceutical Sciences. 2012;4(2):714-6.
13. Vareishang T, Yadav A. Antidiarrhoeal activity of *Rhus javanica* ripen fruit extract in albino mice. Fitoterpia. 2004;75(1):39-44.
14. Kumar D, Ganguly K, Hegde H, Patil P, Kholkute S. Spasmolytic effect of traditional herbal formulation on guinea pig ileum. Journal of Ayurveda and Integrative Medicine. 2015;6(3):194-7.
15. Adeyemi O, Akindele A, Ogunleye E. Evaluation of Antidiarrhoeal effect of *Sanseviera liberica* Geerome and Labroy (Agavaceae). Journal of Ethanopharmacology. 2009;123(3):459-63.
16. Chiwororo WD, Ojewole JA. Spasmolytic effect of *Psidium guajava* Linn. (Myrtaceae) leaf aqueous extract on rat isolated uterine horns. J Smooth Muscle Res. 2009;45(1):31-8.
17. Kamble R, Sathaye S, Shah D. Evaluation of antispasmodic activity of different shodhit guggul using different shodhan process. Indian Journal of Pharmaceutical Sciences. 2008;70(3):368-72.
18. Anwar HG, Samra B, Khalid H, Abdul J. Presence of cholinergic and calcium channel blocking activities explain the traditional use of *Hibiscus rosasinensis* in constipation and diarrhoea. Journal of Ethanopharmacology. 2005;102(2):289-94.
19. Uchiyama T, Chess-Williams R. Muscarinic receptor subtypes of the bladder and gastrointestinal tract. Journal of *Smooth Muscle Research.* 2004;40(6):237-47.
20. Arul V, Miyakazi S, Dhananjayan R. Mechanisms of the contractile effect of the alcoholic extract of *Aegle marmelos* Corr. On isolated guinea pig ileum and tracheal chain. Phytomedicine. 2004;11(7-8):679-83.
21. Rang H, Dale M, Ritter J, Moore P. The gastrointestinal tract. In: Pharmacology, 5th edition, Churchil Livingstone, Edinburgh. 2003;367-79.
22. Chatterjee CC. Human Physiology. 10th edition, Medical Allied Agency, Calcutta. 1984;428-42.
23. Abdulkarim A, Sadiq Y, *et al.* Evaluation of five medicinal plants used in diarrhoea treatment in Nigeria. Journal of Ethanopharmacology. 2005;101(1-3):27-30.

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