

EVALUATION OF ANTI-SPASMODIC EFFECT OF *MANGIFERA INDICA* STEM BARK USING ISOLATED CHICKEN ILEUM

**Jasmine Shalet D. Souza^{*}, Aswathi Nambiar M. P., T. M. Fathima,
Nabeesa Shahla M. M., Zainabath Wafa and Abdul Basith**

^{*}Assistant Professor, Department of Pharmacology, Malik Deenar College of Pharmacy,
Kasaragod, Kerala Malik Deenar College of Pharmacy, Seethangoli, Kasaragod, Kerala
671321.

Article Received on
13 October 2022,

Revised on 02 Nov. 2022,
Accepted on 23 Nov. 2022

DOI: 10.20959/wjpr202216-26287

***Corresponding Author**

Jasmine Shalet D. Souza

Assistant Professor,
Department of
Pharmacology, Malik
Deenar College of
Pharmacy, Kasaragod,
Kerala Malik Deenar
College of Pharmacy,
Seethangoli, Kasaragod,
Kerala 671321.

ABSTRACT

The plant *Mangifera indica* belongs to the family Anacardiaceae. The plant *Mangifera indica* is used in Indian ethnomedicine for its anti-diabetic, anti-inflammatory, anthelmintic and spasmolytic properties. The main active constituent is Mangiferin. Phytochemical tests revealed the presence of alkaloids, carbohydrates, saponins, glycosides, terpinoids. The present study deals with the evaluation of spasmolytic activities of aqueous extract *Mangifera indica* stem bark in isolated chicken ileum. Atropine is used as standard drug. Antispasmodic effect was studied against contraction produced by Acetylcholine in the chicken ileum. The results were compared with atropine. The aqueous extract of *Mangifera indica* stem bark showed a decrease in Ach induced spasm.

KEYWORDS: Anti-spasmodic, *Mangifera indica*, Stem bark, Aqueous extract, Chicken ileum, Acetylcholine.

INTRODUCTION

A muscle spasm, or muscle cramp, is an involuntary contraction of a muscle. Muscle spasms occur suddenly, usually resolve quickly, and are often painful. Antispasmodics are widely used to treat conditions involving impaired contraction and relaxation of smooth muscles in order to relieve muscle spasms, breathing problems, gastrointestinal cramps, movement disorders, etc.^[1] Concerning gastrointestinal smooth muscle contractility, it represents the key mechanism involved in the pathophysiology of several gastrointestinal disorders including

irritable bowel syndrome (IBS).^[2] Therefore, antispasmodics play an important role in the pharmacotherapy of such diseases. Considering the development of tolerance to present drugs and their potential adverse effects, the search for new antispasmodics that can act as bronchodilator agents and/or gastrointestinal smooth muscle relaxants are of great interest.^[3,4] The traditional Medicine is used in all parts of the world and has a rapidly growing economic importance, mainly by the use of medicinal plants that have a respectable position today, especially in the developing countries, where the modern health service is limited and represent the only accessible treatment.^[5] According to the World Health Organization (WHO, 1999), the current estimative suggests that many developed countries have a great proportion of the population making use of traditional practice of health, especially the use of the medicinal plants. Medicinal plants represent an important health and economic component of biodiversity and also conservation and sustainable use.^[6]

Mangifera indica(MI), is commonly known as mango. It has been an important herb in the Ayurvedic and indigenous medical systems for over 4000 years. Mangoes belong to genus *Mangifera* which consists of about 30 species of tropical fruiting trees in the flowering plant. They belong to the family Anacardiaceae. According to Ayurveda, varied medicinal properties are attributed to different parts of mango tree. Mangiferin, being a polyphenolic antioxidant and a glucosyl xanthone, it has strong antioxidant, anti-lipidperoxidation, immunomodulation, cardiogenic, hypotensive, wound healing, antidegenerative and antidiabetic activities.^[7]

2. MATERIALS AND METHODS

2.1 Collection and authentication of plant material

The stem bark of the plant *Mangifera indica* of the family Anacardiaceae was collected from Kasaragod district in the month of December. The plant material was taxonomically identified by the Botanist. The collected stem barks were cleaned to remove the adhered dust particles and were then dried in shade at room temperature. The dried plant materials were powdered by using a Mechanical grinder, weighed and stored in an air tight container till use.

2.2 Extraction

Extraction of dried powder of the stem bark of *M. indica* was carried out by maceration technique. Extraction is done using chloroform water.^[8] Around 20g of dried powder was weighed and dissolved in 180ml of chloroform water. Maceration was carried on a heavy

rotary shaker for 7 days. The extract was filtered and solvent was removed and finally the dried extract was obtained.^[9]

2.3 Phytochemical screening

Phytochemical screening of aq. Extract of *M. indica* was done using various phytochemical tests.^[10,11,12]

2.4 Anti-spasmodic activity

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 aqueous extract of *Mangifera indica* (100 µg/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.^[13] The percentage inhibition of extract and standard drug was calculated and graph was plotted by taking dose verses % decrease in response.^[14]

3. RESULT

3.1 Extraction

The dried stem bark of *Mangifera indica* was powdered and subjected to extraction by maceration. The extraction is carried out by using aqueous solvent. After extraction, the percentage yield of aqueous extract was calculated with reference to air-dried drug used in the study. The percentage yield of the extract was tabulated below.

Table 1: Percentage yield of extraction by water.

SL. NO	SOLVENT USED	PERCENTAGE YIELD (%W/W)
1	WATER	4.7

3.2 Phytochemical screening of extract

The phytochemical studies on stem bark of *M. indica* was carried out and the details are enlisted below. The aqueous extract shows the presence of alkaloids, carbohydrates, saponin, phenolic compounds, tannins, steroids, glycosides and terpenoids.

Table 2: Phytochemical constituents of various extracts of *M. indica*.

SL. NO	TEST	RESULT
1.	Alkaloids	Mayer's test
		Wagner's test
		Hager's test
		Dragendorff's test
2.	Carbohydrates	Molish test
		Fehling's test
		Benedict's test
3.	Saponins	Froth test
4.	Phenolic compounds and Tannins	Ferric chloride test
		Gelatin test
		Lead acetate test
		Decolorization test
5.	Flavonoids	Aq. NaOH test
		Ammonia vapour test
6.	Steroids	Liebermann-Burchard's test
		Salkowski test
7.	Glycosides	Legal's test
		Baljet's test
		Liebermann-Burchard's test
		Borntrager's test
8.	Terpenoids	Salkowski test
9.	Proteins and Amino acids	Million's test
		Biuret test
		Ninhydrin test

3.3 Pharmacological Screening

3.3.1 Antispasmodic study

Table 3: Dose response relationship observations of Ach, Atropine and Aq. extract of *M. indica* stem bark on chicken ileum.

Sl. No.o.	DRUG	DOSE (ml)	HEIGHT OF RESPONSE(cm)	% DECREASE IN RESPONSE
1	ACETYL CHOLINE	0.1	0.9	-
2		0.2	1.0	-
3		0.4	1.1	-
4		0.8	1.2	-
5	ATROPINE + ACETYL CHOLINE	0.1+0.1	0.1	88.88
6		0.1+0.2	0.2	88.00
7		0.1+0.4	0.4	63.63
8		0.1+0.8	0.6	50.00
9	ACETYL CHOLINE	0.1	1.1	-
10		0.2	1.2	-
11		0.4	1.3	-
12		0.8	1.4	-
13	AQ. EXTRACT of	0.1+0.1	0.3	72.72

1414	<i>M. indica</i>	0.1+0.2	0.5	58.33
1515	+ ACETYL	0.1+0.4	0.7	46.15
1616	CHOLINE	0.1+0.8	0.9	35.71

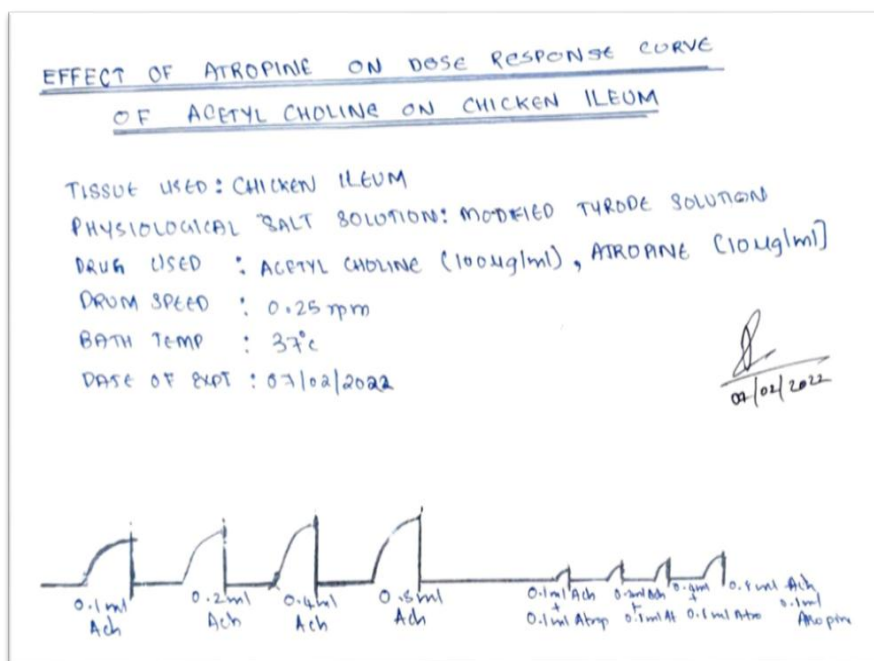


Fig 1: Response curve of Ach and Atropine.

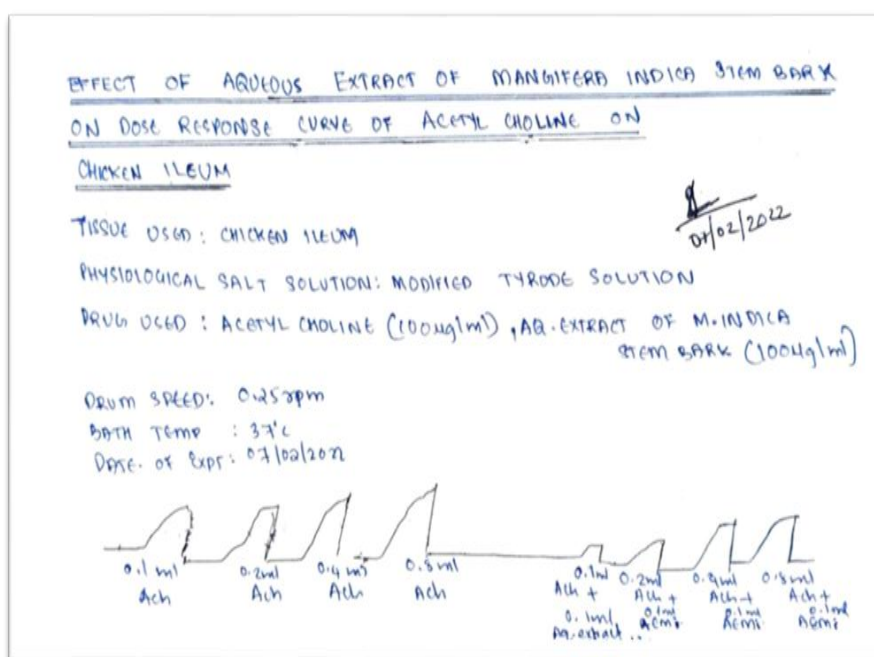
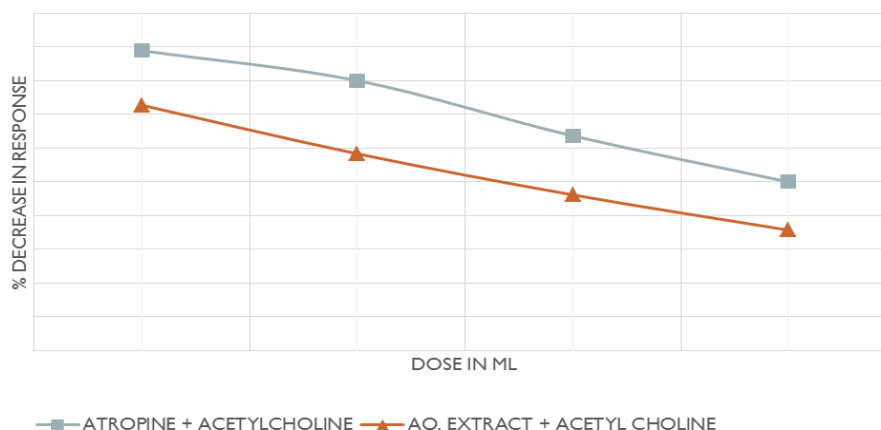


Fig 2: Response curve of Ach and Aq. Extract of *M. indica* stem bark.



Comparative dose response relationship of Ach, aq. Extract of *M. indica* stem bark and Atropine on chicken ileum.

The graph shows dose-dependent increase in spasm in presence of Ach whereas aq. Extract of *M. indica* gives a marked decrease in spasm induced by Ach. It is compared with antispasmodic effect produced by the standard drug atropine.

4. DISCUSSION

The dried stem barks of *M. indica* was powdered and subjected to extraction by maceration technique. The phytochemical screening of the aqueous extract of *M. indica* stem barks shows the presence of saponin, alkaloids, carbohydrates, phenols, tannins, steroids, glycosides and terpenoids. Antispasmodic activity of *M. indica* stem bark is carried out by rotating drum method by using chicken ileum. For this study we have used aqueous extract of *M. indica* stem bark. Dose response curve of Acetylcholine was recorded on the kymograph first. Then cumulative concentration-effect curves were recorded on kymograph for Acetylcholine (100 µg/ml) in absence and presence of aqueous extract of *Mangifera indica* (100 µg/ml) on Kymograph by using Sherrington's Recording Drum. The same procedure was carried for concentration effect curve of Ach in the presence of Atropine sulphate as a standard drug. The percentage inhibition of extract and standard drug was calculated and graph was plotted by taking dose verses % decrease in response. Antispasmodic study on aqueous extract of *M. indica* stem bark, indicate that in the presence of the extracts, chicken ileum shows decrease in Ach induced spasm. The effects were compared with the standard drug atropine, an antispasmodic drug. Aqueous extract of *M. indica* possess comparatively lesser spasmolytic activity. A number of investigators have reported that terpenoids^[15], alkaloids^[16] and phenolic

compounds^[17] are known to possess antispasmodic activity. It is therefore to speculate that the phytoconstituents present in these plant extract might be responsible for the observed antispasmodic activity. They directly or indirectly modulate the contraction of smooth muscles. Thus, from the study, it can be concluded that 100µg/ml of aqueous extract of *M.indica* possess significant antispasmodic activity.

REFERENCES

1. Randall, O.L. Spasmolytic Activity of Synthetic Drugs. J. Am. Pharm. Assoc. (Pract. Pharm. Ed.), 1948; 9: 572–574.
2. Ford A.C, Talley N.J, Spiegel B.M.R, Foxx-Orenstein A.E, Schiller L, Quigley E.M.M, Moayyedi P. Effect of fibre, antispasmodics, and peppermint oil in the treatment of irritable bowel syndrome: Systematic review and meta-analysis. BMJ, 2008; 337: a2313.
3. Clyde B.L, Firlik A.D, Kaufmann A.M, Spearman M.P, Yonas H. Paradoxical aggravation of vasospasm with papaverine infusion following aneurysmal subarachnoid hemorrhage. Case report. J. Neurosurg, 1996; 84: 690–695.
4. Haney S, Hancox R.J. Recovery from bronchoconstriction and bronchodilator tolerance. Clin. Rev. Allergy Immunol, 2006; 31: 181–196.
5. Agra M.F, Freitas P.F, Barbosa-Filho J.M. Synopsis of the plants known as medicinal and poisonous in Northeast of Brazil. Rev Bras Farmacogn, 2007; 17: 114-140.
6. Rahman M.A, Mossa J.S, Al-Said M.S, Al-Yahya M.A. Medicinal plant diversity in the flora of Saudi Arabia 1: a report on seven plant families. Fitoterapia, 2004; 75: 149–161.
7. Shah KA, Patel MB, Patel RJ, Parmar PK. *Mangifera indica* (mango). Pharmacogn Rev, 2010 Jan; 4(7): 42-48.
8. Pharma E-Library, Pharma Knowledge Portal. [Internet]. 2021 [cited 01 february 2022]. Available from: <https://pharmalibrary.blogspot.com/2021/03/preparationofchloroform-waterbysimplesolutionmethod.html>.
9. Nwibani NM, Monanu, O Michael, and Barine IN. Effects Of Aqueous Extract Of *Mangifera indica* L. (Mango) Stem Bark On Haematological Parameters Of Normal Albino Rats. PJN, 2008; 7(5): 663-666.
10. Bag GC, Devi PG, Bhaigyabati. Phytochemical Screening and Antioxidant Activity of *Meyna laxiflora* Species Found in Imphal West District of Manipur. Int. J. Pharm. Sci. Rev. Res, 2016; 36(1): 137-143.

11. Obouayeba AP, Diarrassouba Moussa, Francis ES, Kouakou TH. Phytochemical Analysis, Purification and Identification of Hibiscus Anthocyanins. J Pharm Chem Biol Sci, June-August 2015; 3(2): 156-168.
12. Suman KR, Venkateshwar C, Samuel G, Gangadhar Rao S. Phytochemical Screening of some compounds from plant leaf extracts of *Holoptelea integrifolia* (Planch.) and *Celestrus emarginata* (Grah.) used by Gondu tribes at Adilabad District, Andhrapradesh, India. International Journal of Engineering Science Invention, 2013; 2(8): 65-70.
13. Motghare VM, Nandeshwar MB, Bajait CS, Pimpalkhute SA, Sontakke SD. Chicken ileum: a better option for conducting isolated tissue experiments and bioassay. Indian Journal of Pharmacy and Pharmacology, 2017; 4(2): 110-113.
14. Ghodake, Prasanna and Kulkarni, Ajit and Aloorkar, Nagesh and Osmani, Riyaz and Harkare, Bhargav and Kale, Birudev. In-vitro Antispasmodic Activity Analysis of Methanolic Leaves Extract of *Lantana camara* Linn. on Excised Rat Ileum. Journal of Pharmacognosy and Phytochemistry, 2013; 2(3): 66-71.
15. Martinez-Perez F, Juarez Z.N, Hernandez L.R, and Bach H. Review article, Natural antispasmodics: Source, Stereochemical Configuration, and Biological Activity. Hindawi, BioMed Research International, 2018; 8: 1-32.
16. Rauf A, Akram M, Semwal P, A.H Adil, M Naveed, Riaz Z, et al., Antispasmodic Potential of Medicinal Plants: A Comprehensive Review. Oxid Med Cell Longev, 2021; 4889719: 1-12.
17. Ventura-Martinez R, Esther A.G, Eva G.M, Carrasco O.F, DC Myrna. Study of Antispasmodic and Antidiarrheal Activities of *Tagetes lucida* In Experimental Models and Its Mechanism of Action. Hindawi, Evidence-Based Complementary and Alternative Medicine, 2020; 1-10.