

Study of antidiarrhoeal effect of *Datura innoxia* leave extract against diarrhoea induce by Castor oil and magnesium sulphate in mice.

Duraïd A. Abbas

Farrah R. Jaafar

Department of Physiology and Pharmacology, College of Veterinary Medicine, University of Baghdad, Baghdad, Iraq

Accepted – December – 2010

Summary

Aqueous extract of DI leaves was given to mice , at graded dose (100&200mg/kgB.W,P.O) to evaluate its antidiarrhoeal potential by using four experimentally induced diarrhoea treated groups each consist of five mice and one control group. Castor oil and magnesium sulphate were used to induce diarrhoea in mice, The extract at both doses showed a remarkable antidiarrhoeal activity manifested by significant delay in onset of diarrhea , decrease in number of wet stools , total number of stools and total weight of fecal output in 4 hours in both castor oil and magnesium sulphate induced diarrheal groups of mice. The results of this study reveal that The aqueous extract of *Datura innoxia* leaves contains pharmacologically active substance with antidiarrhoeal properties .These properties may explain the rational use of the plant as antidiarrhoeal agent in the traditional medicine.

Key word: antidiarrhoeal, *Datura innoxia*, leave extract, castor oil, magnesium sulphate.

Corresponding: Farrah R. Jaafar, Email: fajar@a2000

دراسة التأثير المضاد للاسهال لاوراق الداتورة انوكسيا ضد الاسهال المستحدث بأستخدام زيت

الخروج وكبريتات المغنسيوم في الفئران

فرح رسول جعفر

دريد عبد الهادي عباس

فرع الفلسفة والأدوية، كلية الطب البيطري، جامعة بغداد، العراق

الخلاصة

تم دراسة التأثير المضاد للاسهال للمستخلص المائي لاوراق نبات الداتورة *Datura innoxia* في العديد من النماذج التجريبية لدراسة الاسهال في الفئران . تم دراسة التأثير المضاد للاسهال للمستخلص المائي لاوراق نبات الداتورة وبجرعتي 100 و200 ملغم/كغم من وزن الجسم عن طريق الفم في الفئران من خلال استخدام طريقتين تجريبيتين لاحداث الاسهال لغرض تحديد الية فعل المستخلص المضاد للاسهال المستحدث في اربع مجاميع معالجة تحوي كل منها على خمس فئران ومجموعه مماثلة سيطرة . تم استخدام اختبار زيت الخروج واختبار سلفات المغنسيوم لاحداث الاسهال في الفئران . اظهر المستخلص وفي كلتا الجرعتين تأثيرا مميزا مضادا للاسهال بدا واضحا من خلال التاخير المعنوي للوقت اللازم لبدء الاسهال ، التقليل في عدد البراز الرطب ، العدد الكلي والوزن الكلي للبراز خلال (4) ساعات في كلتا الطريقتين . بينت النتائج في هذه الدراسة بان المستخلص المائي لاوراق نبات الداتورة يحتوي على العديد من المواد الفعالة دوائيا والتي لها تأثيرا مضادا للاسهال مما يفسر استعمال النبات كمضاد للاسهال في الطب الشعبي .

Introduction

The World Health Organization (WHO) has constituted a diarrhoeal disease program, which has been encouraged studies for treatment and prevention of diarrhoeal diseases using traditional medical practices (1). Medical plants are promising source of anti diarrhoeal drug (2). It is becomes important to identify and evaluate commonly available natural drug, which are not completely free from adverse effects. A range of medicinal plant with anti diarrheal properties has been widely used for traditional therapy; however, the effectiveness of many of these have not been scientifically evaluated, one of them is Datura, where the flower of Datura innoxia was traditionally used in treatment of diarrhea and gastrointestinal disturbances in infant. Datura plant is very important medicinal plant as it is well known source of the tropane alkaloids; hyoscyamine, atropine and scopolamine (hyoscine). The total alkaloid yield has been estimated to be between 0.06 and 0.5% (3), Hyoscine has anticholinergic, antispasmodic and antispasmodic effect. Hyoscyamine has similar chemical structure to hyoscine, but it is different only by a single molecule of oxygen. Hyoscyamine has the same pharmaceutical effects as hyoscine. Atropine is the racemic form of Hyoscyamine which affects the nervous system (4). Atropine is used in eye drop preparation. It is also used to treat nerve gas poisoning, Parkinson's disease, peptic ulcer, diarrhea and bronchial asthma. In addition to these alkaloids, the plant contains other minor tropane derivatives, as well as chlorogenic acid and lectins. (5) Since this plant is abundant in Iraq, information about its tropane antispasmodic and antidiarrhoea effects have been not well established. So present work took step forward to do study on Iraq local plant datura innoxia aiming the following:

- 1- Induce diarrhoea in mice by using magnesium sulphate or castor oil.
- 2- compare the antidiarrhoea effect of different doses of aqueous datura innoxia with loperamide.
- 3- Determining the mechanism of its anti diarrhoeal action.

Materials and Methods:

Plant materials : Crude aqueous extract of Datura innoxia leaves was prepared by using 60g of dried powdered of plant leaves. The dry powdered plant leaves were put in pyrex flask (500ml) containing (200ml) distilled water, mixed by magnetic stirrer at 40°C for 24 hours then filtered to get rid of residue and placed in an incubator at 40°C to produce a dried extract. The dried extract was weighed and kept at -20°C in sterile petridish. The net weight of dry extract of DI leaf was 3g making the yield 5%. Animals: Twenty five Albino swiss mice weighing 25-30g of either sex were used. The animals were grouped and kept in cage housed at standard condition of light and ventilation and have freely access to standard rodent diet (commercial feed pellets) and tap water. The animal were kept for a week for acclimatization. Experiment-1 Study of DI extract activity against diarrhea induced by castor oil : This was done according to the method, described by (6). The animals were all tested initially by giving them 0.5 ml of castor oil. Only those who showed diarrhea were selected for the final experiment. Twenty five mice were selected and divided equally into five groups which submitted for the following treatment : Negative control group (cont-ve) : normal mice received distilled water at a dose of 10ml/kg B.W.P.O. -positive control group (cont+ve): induced diarrhea mice received distilled water at a dose of 10ml/kg B.W.P.O. -loperamide treated group - induced diarrhea mice dosed with loperamide at a dose of 3mg/kg B.W.P.O. Two datura innoxia

treated groups- - induced diarrhea mice and received DI aqueous extract at a dose of 100 and 200 mg/kg B.W.P.O. respectively. Each mouse was placed over a glass funnel, the floor of which was lined with weighed paper, changed every hour. Diarrhoea was induced by oral administration of 0.5 ml of castor oil to each mouse, 45 minutes after the above treatment. The following parameters were observed for a period of 4 hours. The onset of diarrhea stool (first wet stool that leaves a halo on the weighed paper).

1. Number of wet stools.
2. Total number of stools.
3. Total weight of feces.

Experiment-2: Study of DI extract activity against diarrhea induced by magnesium sulphate: This was done according to the method, described by (7). Twenty five mice were divided equally into five groups as in the castor oil induced diarrhea experiment, with the same pretreatment measures before induction of diarrhea. After 30 minutes diarrhea was induced by oral administration of magnesium sulphate at a dose of 2g/kg B.W. to each mouse. Similar parameters for castor oil induced diarrhea experiment were measured. Statistics analysis: Results were expressed as means \pm standard error that subjected to statistical analysis using one-way analysis of variance (ANOVA) and LSD. The significance level considered was ($p < 0.01$).

Results and discussion

Four hours after castor oil administration, all the mice in control group produced copious diarrhoea. Pretreatment of mice with aqueous extract of *Datura innoxia* at doses of 100 or 200mg/kg B.W.P.O caused dose dependent significant ($P < 0.01$) delay in the onset of diarrhoea, decrease in the number of wet stools, total number of stools. Decrease in the total weight of fecal output also was noticed. However, the reduction in the above measured parameters that showed after administration of DI aqueous extracts at doses were comparatively less than that produced by loperamide (table,1-). It is well known that castor oil or its active component ricinoleic acid induced permeability change in mucosal fluid and electrolyte transport that result in hypersecretory response and diarrhoea (8). Castor oil broken down in the small intestine to ricinolic acid which is very irritating to gut promptly increases peristalsis(9). *Datura* plant is very important medicinal plant as it is well known source of the tropane alkaloids; hyoscyamine, atropine and scopolamine (hyoscyne). The total alkaloid yield has been estimated to be between 0.06 and 0.5%(3). Tropane alkaloids are commonly as anti-cholinergic compounds, due to their ability to bind to muscarinic acetylcholine receptors and hence acting as competitive antagonists at these receptors. Atropine is a non-selective antagonist of all classes of muscarinic receptors M_1, M_2, M_3, M_4 and M_5 . but known to have a stimulating effect on the central nervous system, whereas scopolamine is a depressant of the central nervous system (10). The result of antidiarrhoeal activity of DI extract against diarrhea induced by magnesium sulphate in mice were listed in table,2. In this model the aqueous extract of *Datura innoxia* at doses of 100 and 200 mg/kg B.O.W.P. significantly ($P < 0.01$) reduced the extent of diarrhoea in mice. Both doses were shown to delay in the onset of diarrhoea, decrease in the the number of wet stools, total number of stools and decrease in the total weight of fecal output significantly at ($P < 0.01$).

Table(1).Effect of Aqueous extract of datura innoxia and other treatment on castor oil induce diarrhea in mice

Group n=5	Dose	Onset of diarrhoea(minute)	Number of wet stools in four hour	Total number of stools in four hour	Total weight (g) of fecal output in four hour
Control negative (-ve)without diarrhea &treatment.D.W	10ml/kg BW,P.O	0	0	2±0.447 A	0.32±0.008 A
Control (+ve) positive induced diarrhea without treatment D.W	10ml/kg BW,P.O	74.5±9.107 A	8±0.988 A	11±1.224 B	0.873±0.139 B
Loperamide group induced diarrhea with treatment	3mg/kg BW,P.O	212.2±12.871 B	1.4±0.444 B	2.8±0.418 A	0.35±0.011 A
Low aqueous extract of DI treated group induced diarrhea with treated	100 mg/kg BW,P.O	149.8±8.603 C	3.4±0.672 C	5.4±0.898 C	0.444±0.053 C
High Aqueous extract of DI treated group induced diarrhea with treatment	200 mg/kg BW,P.O	154.4±8.148 C	2.6±0.470 C	4±0.820 C	0.320±0.019 A

Mean value different letter signnifigent value (P<0.01) F_test . DI=Datura innoxia leave aqueous extract.

However , loperamid also showed more reduction in the extent of diarrhoea when comparnd with DI extract effect in mice. Magnesium sulphate are non absorbable salts(Bulking agents) that hold in the water in the by osmosis, and distend the bowel, increasing intestinal activity and producing defication (11). It has also been demonstrated that it promotes the libration of cholecystokinin from the duodenal mucosa,which increases the secretion and motility of small intestine and thereby prevent the reabsorption of sodium chloriod and water(12 and13) have mentioned that alkaloids such as atropine and scopolamine constitute an important class of anticholinergic compounds derived from plants that occur in Datura species . clinically , they are used to block the muscarinic activity of acetylcholine showing antispasmodic and antisecretory effects in the treatment of spastic colitis , gastroenteritis and peptic ulcer.They are also useful pharmacological tools to discover new active principles with gastrointestinal tract action (2).The leaves and seed of Datura are widely used in herbal medicine as anaesthetic , antispasmodic, antitussive, and bronchodilator and as hallucinogenic(14).Aqueous extract of Datura innoxia leaves showed antispasmodic effect on isolated rabbit duodenum,That was attributed to trapane alkaloids , atropine and scopolamine identified by related standard in TLC (1).DI extract seem to have

effect against diarrhea induced by irritant and bulk diarrhea agent. DI aqueous extract showed dose dependent antidiarrheal effect but at dose 200mg/kg, antidiarrhea effect was even lesser but approximate to loperamide effect. It seems that the use of higher doses of DI extract may give similar antidiarrheal effect as loperamide.

Table(2). Effect of aqueous extract of datura innoxia on magnesium sulphate induced diarrhoea in mice

Group n=5	dose	Onset of diarrhoea (minute)	Number of wet stools in four hour	Total number of stools in four hour	Total weight of fecal output in four hour
Control negative (-ve) without diarrhea & treatment. D.W	10ml/kg BW,P.O	0	0	1.4±0.304 A	0.22±0.13 A
Control (+ve) positive induced diarrhea without treatment D.W	10ml/kg BW,P.O	89±9.32 A	7.4±0.31 A	10.6±0.165 B	0.745±0.082 B
Loperamide group induced diarrhea with treatment	3mg/kg BW,P.O	207.2±13.79 B	1.6±0.74 B	3.4±0.61 C	0.23±0.044 A
Low aqueous extract of DI treated group induced diarrhea with treated	100 mg/kg BW,P.O	123.2±12.852 C	3±0.992 C	4.2±0.86 D	0.363±0.070 C
High Aqueous extract of DI treated group induced diarrhea with treatment	200 mg/kg BW,P.O	151.2±13.07 D	2.4±0.56 D	3.6±0.51 C	0.341±0.077 D

Mean value diff. letter sign. Vale (P<0.01) F- test. DI= datura innoxia leave aqueous extract j

References

1. A bokataiffa MAG (2009). Acute toxicity and Relaxant activity of Extract of Local datura innoxia leaves smooth muscle of laboratory animal.
2. Rani S Ahmed N and Rajaram S (1999). Antidiarrhoeal evaluation of Clerodendrum Phlomidis Linn Leaf extract in rats. J Ethnopharmacol 68:315-9.
3. Basch EM Servoss J Cand Tedrow UB (2005). Safety assurances for dietary supplements policy issues and new research paradigms J Herb pharmacotherapy 5:3-15.
4. Ziegler J and Facchin PJ (2008). Alkaloid biosynthesis : metabolism and trafficking Annu Rev Plant Biol 59:735-769.
5. Boominathan R Devi BP Dewanjee S and Mandal SC (2005). studies on antidiarrhoeal activity of ionodium suffruticosam ging (Violaceae) extract in the rats phytotherapeutics 10:375-380.

6. Shoba F G and Thomas M (2001). Study of antidiarrhoeal activity of four medicinal plants in castor oil induced diarrhea. *J Ethnopharmac.* 76:73-76.
7. Dewick P M (1997) *Medicinal natural products: a biosynthetic approach* John Wiley and Sons Chichester. 8:274.
8. Mascolo N Izzo A A Autore G Barbato F and Capasso F (1994). Nitric oxide and castor oil –induce diarrhea. *J pharmaco experimen Therapeutic.* 268:291-295.
9. Ammon HV Thomas PJ and Philips S (1974). Effect of oleic and ricinoleic acid on Jejunal water and electrolyte movement. *J Clin Inves* 53:374_379
10. Brown JH and Taylor P (2006). *Goodman and Gilman's The pharmacological Basis of Therapeutics* 11 the Ed LL Brunton JS Lazo and KL Parker(Eds) McGraw-hill Med publ Div New York. Pp: 183-200 .
11. Mycek MJ Harvey RA and Champe PC (2000). *Pharmacology* 2nd ed Lippincott William and Williams and Wilkins Philadelphia London Hong Kong PP 46-139
12. Galves A Zarzuelo ME Crespo MD Lorente M Ocete A and Jimenez J (1993). Antidiarrhoeic activity of *Euphorbia hirta* extract and isolation of active flavonoid constituent. *Planta Medica.* 59:333_336.
13. Zavala MA Perez S Perez C Vargas R and Perez RM (1998). antidiarrhoeal activity Of americana *Commelina coelestis* and *Alternanthera respens.* *J Ethnopharmac* 61:41-47
14. Satyavaqti GV Rama Mk and Sharma M (1976). *Medicinal plant India* ICMR NewDelhi 1:333-334.