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Original Article

ANTIDIARRHEAL ACTIVITY OF *THYMUS ALGERIENSIS* BOISS AND REUT AQUEOUS EXTRACT ON RATS AND MICE

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ABSTRACT

Objective: *Thymus algeriensis* (Lamiaceae) used in Moroccan popular medicine has been investigated for its antidiarrheal effects.

Methods: Antidiarrheal effects of the aerial-part *Thymus algeriensis* aqueous extract was carried out by using two standard methods of bioassay: Castor oil-induced diarrhea and gastrointestinal transit test.

Results: The pre-treatment of rats with the 50 mg/Kg aqueous extract produced a significant inhibition against castor oil induced-diarrhea and fecal output; furthermore the extract significantly decreased the propulsive movement of the charcoal meal in the mice small intestines. The extract when administered at higher doses (100 and 200 mg/kg) caused increased peristalsis in charcoal fed animals and exhibited a less significant reduction in fecal output and castor oil-induced diarrheas than 50 mg/ml dose.

Conclusion: The antidiarrheal effects of the extract may explain the rationale for the use of the plant in traditional medicine as a popular antidiarrheal recipe.

Keywords: *Thymus algeriensis,* Antidiarrheal activity, Castor oil, Intestinal transit.

INTRODUCTION

Diarrhea is one of the most common disorders known to man. It is among the most prevalent causes of mortality in developing countries [1]. The World Health Organization (WHO) has encouraged studies for treatment and prevention of diarrheal diseases depending on traditional medicinal practices [2, 3]. In traditional medicine, different plants are used in order to control this disorder [4, 5]. Among these plants Thymus which is a large genus of Lamiaceae. The number of species within this genus is assumed to be more than 200 particularly prevalent in the Mediterranean area [6]. Many Thymus species are commonly used, dry or fresh, as herbal teas, condiments and spices, so as for various medicinal purposes [7]. Among these species, *Thymus algeriensis* is the most widespread North African species, endemic to Morocco, Algeria, Tunisia and Libya. It is largely used as a culinary herb, although results of its biological activity are still scarce. It is also used in traditional medicine in respiratory and digestive tube disorders and against abortion [8].

In view of the above information and folklore use of the aerial-part of this plant as an antidiarrheal agent, the present study was undertaken to evaluate the antidiarrheal activity of its aqueous extract *in vitro* and in a castor oil-induced experimental model of diarrhea in rats and mice.

MATERIALS AND METHODS

Plant material

The aerial-part of *Thymus algeriensis* was collected from the Oujda area in oriental Morocco in Mai 2014. A taxonomic identification was performed by Pr. B. Haloui from the Biology Department of Oujda Sciences Faculty (Morocco).

Preparation of the extract

Aqueous extract of the aerial part (stems, leaves and flowers) of *Thymus algeriensis* (AqTA) was decocted and evaporated to give a crude residue (yield: 18%).

Animals

Male and female Wistar rats (250-300g) and Swiss albino mice (20-25g) were obtained from our local colonies. They were kept under conditions of constant temperature (22±2°C) with a standard 12-h light: 12-h dark cycle and free access to food and water. All animals were cared for in compliance with the Guide for the Care and Use of Laboratory Animals, published by the US National Institutes of Health (NIH Publication 85-23, revised 1996; see (http: //grants. nih. gov/grants/olaw/olaw. htm).

Castor oil-induced diarrhea

The Animals were fasted for 18 h but allowed free access to water. They were randomized into five groups, control, positive control and test groups containing five rats each. The Control group received orally distillate water, 1 ml each rat. The positive control groups received loperamide hydrochloride at the dose of 10 mg/kg orally; test groups received the aqueous extract at the doses of 50, 100 and 200 mg/kg. Each animal was placed in an individual cage, the floor of which was lined with blotting paper. The floor lining was changed after every defecation. Diarrhea was induced by oral administration of 1 ml castor oil to each rat, 1 h after the above treatments. Fecal output was assessed by collecting fecal material for 4 h after the administration of castor oil, and this was dried at 70°C for overnight before weighing. The percentage fecal output (FOP) was calculated as follows:

$$\% FOP = (ft/fc)*100$$

Ft: mean fecal weight of each treatment group, Fc: mean fecal weight of the control group [9-11].

Small intestinal transit study

The effects on intestinal propulsion in Swiss albino mice were tested by using the charcoal method [12]. Animals were fasted for 18 h but allowed free access to water. They were randomized into five groups of five animals each. Group 1 (control) received 0.8 ml of distillate water by force-feeding; groups 2, 3 and 4 were treated with 50, 100 and 200 mg/kg of *T. algeriensis* extract, respectively; Group 5 was given orally with loperamide hydrochloride (10 mg/kg) as a standard. After 15 min, each mouse was administered orally by 0.5 ml-charcoal meal (3%), activated charcoal suspended in 0.5% of aqueous cellulose. All the mice were killed, 30 min later by cervical dislocation and bled, and the small intestine was rapidly dissected

out and placed on a clean surface. The intestine was carefully inspected and the distance traversed by the charcoal meal plug from the pylorus to caecum was measured. The length of the whole small intestine was also measured. The distance travelled by the charcoal plug from pylorus to caecum was expressed as a percentage of the total length of the small intestine [13].

Intestinal propulsion % = (A/B)*100.

Where 'A' is the distance moved by the suspended charcoal meal, 'B' is the whole length of small intestine. The percentage of inhibition compared with the control group was determined by using the following equation [14]

Inhibition % = {(E-C)/C}*100.

Where 'E' is the mean distance in the treated group, 'C' is the mean distance in the control group.

Statistical analysis

The results are expressed as mean ± S. E. M. Significance of differences between control and treated groups were determined using the Student's *t*-test.

RESULTS

The effect of AqTA on castor oil-induced diarrhea rats was assessed by using doses of 50-200 mg/kg in mice. The AqTA, at the dose of 50 mg/kg, reduced the total number of feces as well as of diarrhoeic feces, and the results were statistically significant (table 1).

At The AqTA dose of 100 mg/Kg exhibited a significant reduction but less than 50 mg/Kg. 200 mg/Kg slightly reduced diarrhea. This latter did not occur in mice treated with 10 mg/kg loperamide as a positive control group.

Table 1: Effect of aqueous extract of *Thymus algeriensis* on castor oil-induced diarrhea in rats

Treatment	Means of dry feces weight (g)[a]	% FOP	% of inhibition
Water 1 ml/kg	0,98 ± 0.08	-	-
Thymus algeriensis 50 mg/Kg	0.27 ± 0.13	28.09	71.90 ***
Thymus algeriensis 100 mg/Kg	0.30 ± 0.19	31.37	68.62 ***
Thymus algeriensis 200 mg/Kg	0.83 ± 0.06	84.64	15.35 *
Loperamide 10 mg/ Kg	0	0	100 ***

[a] Values are mean ± S. E. M. (n = 5), Results were analyzed by Student't-test. * p < 0.05; ** p < 0.01; *** p < 0.001 vs control.

Table 2: Effect of aqueous extract of *Thymus Algeriensis* on gastrointestinal transit in mice

Treatment	Distance traveled by charcoal as % of total lenght of small intestine[a]	% of inhibition
Water	57.09 ±3.08	0
*Thymus algeriensis 5*0 mg/Kg	23.76 ±3.77 ***	- 58.38
Thymus algeriensis 100 mg/Kg	28.56 ± 8.37 *	- 49.96
Thymus algeriensis 200 mg/Kg	37.59 ±1.28 ***	- 34.14
Loperamide 10 mg/ Kg	12.27 ± 0.78 ***	- 78.50

[a] Values are mean ± S. E. M. (n = 5), Results were analyzed by Student't-test. * p < 0.05; ** p < 0.01; *** p < 0.001 vs control.

Effect on gastrointestinal motility

In the gastrointestinal motility test, the AqTA, at the dose of 50 mg/kg, retarded significantly the intestinal transit of charcoal meal in mice as compared to the control. (table 2). More the AqTA concentration increase (100, 200), more the retarded intestinal transit decrease.

DISCUSSION

Diarrhea is considered as a consequence of altered motility and fluid accumulation in the intestinal tract [15]. It has been found that castor oil increases the peristaltic activity of the intestine and produces changes in the permeability of the intestinal mucosa to electrolytes and water [16] and also contributes to the pathophysiology of the gastrointestinal tract [17]. In this study, the antidiarrheal effect of *Thymus algeriensis* at a dose of 50 mg/kg was found to be similar to loperamide (10 mg/kg). The therapeutic effect of this latter is believed to be due to its antimotility and anti secretory properties [18]. Loperamide though appears to be at least 7 times more potent antidiarrheal agent that the AqTA, the low potency exhibited by *Thymus algeriensis* could be as a result of its crude nature. Loperamide, however, is used in its pure form. The exact mechanism by which the AqTA exerts this antidiarrheal effect in rats is not clear from this study. Numerous mechanisms have been proposed in the mediation of castor oil diarrhea involving platelet-activating factor [19], nitric oxide [20] and prostaglandins [12], it remains to be seen whether or not such mediators are inhibited in the antidiarrheal effect of the plant preparation.

Surprisingly, the extract when administered at higher doses (100 and 200 mg/kg) caused increased peristalsis in charcoal fed animals and exhibited a less significant reduction in fecal output than 50 mg/ml. The same kind of results was founded by Suleiman et al. [21] with the methanol stem-bark extract of *Annona senegalensis*. Similarly, morphine when administered at a lower dose cause decreased intestinal smooth muscle tone leading to constipation. However, when gave at relatively higher doses to a dog it causes a persistent spasmogenic effect upon the intestinal smooth muscle by direct action, partly by cholinergic and partly by histaminergic mechanisms [22]. Perhaps similar mechanisms could explain the action of the extract of *Thymus algeriensis*.

Also, some plants show antidiarrheal properties by their antimicrobial activities [23, 24]. The extract of *Thymus algeriensis* was shown to exhibit good antibacterial activity when tested against many pathological species [25, 26].

CONCLUSION

Antidiarrheal potential of the 50 mg/ml AqTA was evidenced by a significant reduction in fecal output and protection from castor oil-induced diarrhea in rats and decreased the propulsion of charcoal meal through the gastrointestinal tract in mice. These results may explain the rationale for the use of the plant in traditional medicine as a popular antidiarrheal recipe.

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CONFLICT OF INTERESTS

Declared None

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