WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 7.523
Volume 6, Issue 4, 163-172. Research Article ISSN 2277– 7105

EVALUATION OF ANTI-DIARRHOEAL AND APPETIZER EFFECTS OF *NIGELLA SATIVA* SEEDS

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Article Received on
26 Jan. 2017,

Revised on 18 Feb. 2017,
Accepted on 08 March. 2017

DOI:10.20959/wjpr20174-7987

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ABSTRACT

This study aims to investigate the anti-diarrheal and the appetizer activities of *Nigella sativa* seeds. To evaluate the anti-diarrheal effect using castor oil induced diarrhoea in albino wistar rats, crude extract of seeds was given to rats, at graded dose of 100, 250 & 500mg/kg by oral gavage, by using four experimentally induced diarrhoea treated groups each consist of five rats and one control group. Loperamide (3mg/kg p.o) were used as standard drugs. The crude extract at 500mg/kg showed a remarkable antidiarrhoeal activity manifested by significant decrease in number of wet stools and total number of stools, output in 4 hours. The results of this study indicate the presence of some active principles in the crude extract of *Nigella sativa* seeds possessing anti-diarrhoeal effect. To investigate appetizer effect of oral administration of *Nigella sativa* seeds, 36 adult healthy volunteers of either sex, of age 28, 44 ± 10, 53 years and mean BMI 24, 242 ± 3,107 kg/m2, were divided into four groups. The subjects were administered orally respectively at 1 tablet based on Fenugreek (600 mg), 1g, 2g and 3g of *Nigella sativa* seeds powder. After 20 min of treatments, we observed after 6 hour, the subjective appetite of study participants using a visual analog scale. The results show that oral administration of powder of *Nigella sativa* seeds significantly increased subjective appetite. These results may explain the rational uses of this seeds as antidiarrhoeal and appetizer agent in the traditional medicine.

KEYWORDS: *Nigella sativa*, antidiarrhoeal, crude extract, castor oil, appetizer, appetite.

INTRODUCTION

Diarrhoea can be defined as the increased frequency of bowel movements accompanied by a loose consistency of stools.[1] It results from an imbalance between the absorptive and secretory mechanisms in the intestinal tract, accompanied by intestinal hurry, resulting in poor absorption of water, nutritive elements and electrolytes producing abnormal frequent evacuation of watery stools.[2,3] Secretory diarrhoea is the common form of acute diarrhoea which occurs as a result of increased intestinal secretion or decreased intestinal absorption of fluid and electrolytes or by altered motility of gastro-intestinal tract.[4] Diarrhoeal diseases are one of the leading causes of morbidity and mortality in developing countries and are responsible for the death of millions of people each year.[5] Available medications such as loperamide may be beneficial; however they may be contraindicated in certain situations[6], or it suffer from adverse effects like the induction of bronchospasm, vomiting by racecadotril; intestinal obstruction and constipation by loperamide.[7] The World Health Organization has encouraged studies for treatment and prevention of diarrhoeal diseases using traditional medical practices.[8] Medical plants are promising source of anti diarrhoeal drug.[9] It is becomes important to identify and evaluate commonly available natural drug. A range of medicinal plant with anti-diarrheal properties has been widely used for traditional therapy; however, the effectiveness of many of these has not been scientifically evaluated, one of them is *Nigella sativa*. In addition, this seeds is used in traditional medicine to treat loss of appetite.[10] Anorexia is defined as involuntary loss of appetite that is associated with decreased oral intake.[11] It is common in patients of advanced age and can lead to drastic weight loss. Anorexia and weight loss also complicate diseases such as cancer, AIDS, and cardiac failure, regardless of age. Consequences of weight loss associated with anorexia can be devastating in all age groups and constitute a special problem in older adults.[12, 13] Early detection and treatment of anorexia may prevent weight loss, improve health outcomes, and reduce mortality.[14] Hence, the present study was under taken to evaluate the antidiarrhoeal activity of crude extract of *Nigella sativa* seeds in Albino Wistar rats model and appetizer effect in healthy human subjected to its traditional claims.

Nigella Sativa Linn belongs to family Ranulaceae. The herb is widely known in different parts of the world and its seeds are used as condiment. In subcontinent it is known as '*kalonji*' and its Arabic name is '*Haba Sauda*'. In the west it is known as "Black Cumin".[15]

The seed of this plant is referred to by the prophet Mohammed as having healing powers.[16]

MATERIAL AND METHODS

Plant materials

Crude aqueous extract of *Nigella sativa* seeds was prepared by using 60g of powder of seeds in 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 -4° C in sterile petridish.

Experimental animals

Twenty five adult Wistar rats of the either sex weighing (120-220 g) were used. The animals were kept in standard environmental conditions (at 24.0 ± 4°C temperature, 55-65% relative humidity and 12 hour light/12 hour dark cycle) for at least one week for adaptation.

Antidiarrhoeal activity study by castor oil induced diarrhea

The method, described by Dangi *et al.*[17], was followed for this study with slight modification. Rats of either sex were fasted for 18 h. They were divided into five groups (n=5). The first group of animals, which served as control was administered with distilled water. The second group received standard drug, loperamide (3 mg/kg) orally as suspension. The extract was administered by oral gavage at 100, 250 and 500 mg/kg dose to third, fourth and fifth group respectively. After 60 min of drug treatment, the animals of each group received 1ml of castor oil orally and the watery faecal material and frequency of defecation were recorded during 4 h in the transparent metabolic cages with pre weighed plastic dishes placed at the base. Weight of plastic dish before and after defecation was noted and compared to control.

Subjective appetite measurement

The study was conducted on 36 adult, healthy volunteers of either sex. They were divided into four groups. The first which served as control was administered with 1 tablet based on *Trigonella foenum-graecum* (Fenugreek) (600 mg) which increases the appetite for food (18, 19). The *Nigella sativa* seeds powder was administered orally at 1g, 2g and 3g to second, third and fourth group respectively. 20 min after the above treatments the spontaneous food consumption and the subjective appetite of study participants were evaluated during an observation period of 6 hour. They were assessed using a visual analog scale (VAS) (20). Which is a very sensitive and reliable tool designed to determine subjective appetite (21).

Appetite was assessed subjectively on a ten-point scale. Mean appetite scores were calculated for each treatment.

Statistical Analysis

All the data obtained were expressed as the mean ± standard error of mean (SEM). Statistical differences between the treatments and the controls were estimated by Minitab (Version 17) by the student's t-test. The minimum level of significance was set at $p<0,05$.

RESULTS

Anti-diarrhoeal effect

In a model of castor oil-induced diarrhoeal experiment in rats, at all the doses tested, a decrease in the frequency of defecation, the frequency of watery faecal material, and total weight of stools were observed. Statistical analysis show that pretreatment of rats with the crude extract of *Nigella sativa* at doses of 100 mg/kg caused a significant decrease ($p<0,05$) of total number of wet faeces. At the dose of 500 mg/kg, the crude extract reduced a decrease in mean frequency of the total number of faeces as well as the mean frequency of total number of diarrhoeic faeces (Table. 1). These results were shown to be statistically significant ($p< 0,05$). The anti-diarrhoeal activity of the crude extract at a higher dose (500 mg/kg) was better than that of loperamide at a dose of 3 mg/kg.

Table 1: Effect of crude extract of *Nigella sativa* and Loperamide on castor oil induced diarrhea in rats.

Treatment	Dose (mg/kg)	Total number of faeces in 4 h	Total number of wet faeces in 4 h	Mean wet defecation (g)
Distilled water	-	3,2 ± 0,86	2,600 ± 0,510	4,048 ±0,545
Loperamide	3	1 ±0,316	0,800±0,374	2,842 ±0,523
Crude extract *(NS)*	100	2,400 ± 0,812	1,60 ±0,678 *	4,60 ±1,62
Crude extract *(NS)*	250	2,400 ±0,812	2,400 ±0,812	5,54 ±1,02
Crude extract *(NS)*	500	0,600 ±0,400 *	0,400 ±0,245 **	3,32 ±1,00

* $p<0,05$, ** $p<0,01$ vs control.

Appetizer effect

The mean age of the subjects was 28, 44 ± 10,53 years and the mean BMI was 24,242 ± 3,107 kg/m2. The results show that oral administration of powder of *Nigella sativa* seeds, at all doses, significantly ($p < 0,001$) increases mean appetite scores comparing with those noted

in subjects treated with fenugreek. At all doses *Nigella sativa* seeds effect was higher to which of fenugreek tablet. *Nigella sativa* seeds showed dose dependent appetizer effect. At a dose of 2g and 3g we recorded the highest mean appetite score (Table. 2).

Table 2: Mean appetite scores for each treatment.

Treatment	Mean appetite scores
Fenugreek	2,777 ± 1,787
NS 1g	6,333 ± 2,91***
NS 2 g	7,777 ± 2,333***
NS 3 g	7,333 ± 1,870***

*** p< 0,001 vs control.

DISCUSSION

The seeds of *Nigella sativa* L. have been used in traditional medicine by many Asian, Middle Eastern and Far Eastern Countries to treat headache, coughs, abdominal pain, asthma, rheumatism and other diseases.[10] These seeds have been widely used as antidiarrheal, digestive and appetite stimulant. They are used in indigestion, dyspepsia and sour belching.[22, 23, 10] A number of studies have explained some effects of these seeds on the digestive tract. The volatile oil and ethanolic extract of *N. sativa* inhibited spontaneous movements of the rabbit jejunum as well as agonist-induced contractions and the spasmolytic effect involved calcium channel blockade.[24] The aqueus-methanolic extract of Nigella seeds also showed spasmolytic effect mediated through calcium antagonist effect thus providing scientific basis for its traditional use in diarrhoea.[22]

Several mechanisms have been proposed to induce the diarrhoeal effect of castor oil.[25] Among which, 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 diarrhea.[26] Castor oil broken down in the small intestine to recinolic acid which is very irritating to gut promptly increases peristalsis.[27] In this study the crude extract of *Nigella sativa* seeds successfully inhibited the castor oil-induced diarrhoea; the extract might have exerted its antidiarrhoeal action by antisecretory mechanism. This was also evident from the reduction of total number of stools and total number of wet faeces in the test group at a dose of 500 mg/kg.

Other studies showed that anti-dysenteric and anti-diarrhoeal properties of medicinal plants were due to tannins, alkaloids, saponins, flavonoids, sterol and/or triterpenes and reducing

sugars.[28, 29] *Nigella sativa* seeds contain all this variety of component.[30] This constitution may be responsible for the mechanism of action of antidiarrhoeal activity. This can be due to the fact that the extract increased the reabsorption of water by decreasing intestinal motility. In addition, Flavonoids possess a wide range of activities *in vitro* including antidiarrhoeal activity[31, 32, 33] may have contributed to this activity, but further studies are required.

Anorexia is a fairly common symptom in many diseases. Age-related homeostatic compromise increases the likelihood of subsequent weight loss and death in older anorectic patients. Anorexia-related weight loss can have devastating consequences on quality-of-life, morbidity, and mortality.[14]

In this study, we examined the effect of *Nigella sativa* seeds per oral on subjective appetite in healthy volunteers. Because it have been used in traditional medicine as appetite stimulant.[10] The effect of *Nigella sativa* seeds was compared with which of fenugreek in control group. Fenugreek *(Trigonella foenum-graecum* L.) is an erect annual herb of the leguminous family.[34] It increases the appetite for food.[18] After administration of *Nigella sativa* seeds significantly ($p < 0,001$) increases of the motivation to eat and subjective appetite was noted, consequently it increase food intake. Fenugreek seeds are traditionally assumed to have restorative properties. It was shown that the treatment with steroid saponins, extracted and purified from the seeds of fenugreek, significantly increased food consumption and induced hypocholesterolemia in normal rats.[35] Many saponins were determined from *Nigella sativa* seeds.[36, 37, 38, 39, 40, 41, 42] These data suggest that the content of black seed in saponins may be responsible for its appetizer effect.

Furthermore, *Nigella sativa* seeds have odor slightly aromatic and taste bitter.[43] Bitter substances form a very diverse group of components whose common point is the bitterness of their taste. This bitterness stimulates the secretions of the salivary glands and digestive organs. These secretions increase appetite and improve digestion.[44]

The present study provided a support for the traditional use of *Nigella sativa* seeds as an antidiarrhoeal remedy and an appetite stimulant. However, further studies will be necessary to isolate, fractionate and purify the extracts and characterize the active principles which are responsible for the antidiarrhoeal and appetizer effects and to understand exact its mechanisms of actions.

CONCLUSIONS

In conclusion, the present study demonstrates that the crude extract of *Nigella sativa* seeds exhibited antidiarrheal activity in a model of castor oil-induced diarrhoeal in rats. The extract could be useful as a nonspecific treatment for diarrhea. It is also reasonable to suppose that this crude extract might be effective in secretory diarrhea. On the basis of these findings, it is can be assumed that the *Nigella sativa* seeds could be potential source for an antidiarrheal drug in combination with other drugs.

On the other hand, the appetizer effect was significantly most important in subjects treated with *Nigella sativa* seeds than in those who received fenugreek tablet. These results suggest that the administration of *Nigella sativa* seeds enhances motivation to eat and appetite in healthy human. These results support the traditional use of *Nigella sativa* seeds as anti-diarrheal and appetizer.

CONFLICT OF INTEREST

The authors declare that they have no conflicts of interest related to this article.

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