INTERNATIONAL JOURNAL OF CURRENT RESEARCH IN CHEMISTRY AND PHARMACEUTICAL SCIENCES
(p-ISSN: 2348-5213: e-ISSN: 2348-5221)
www.ijcrcps.com
Coden: IJCROO(USA) Volume 3, Issue 6 - 2016

Research Article

SOI: http://s-o-i.org/1.15/ijcrcps-2016-3-6-8

The Effect of Oak Fruit Husk Extract on Salivary Flow Rate in Rats

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Abstract

Background and Aim: Sialorrhea is a common clinical problem in children and adults that can have significant social and medical implications. The aim of this study was to evaluate the effects of extraction off oak fruit husk on amount saliva in rat. ***Materials and Method:*** In this experimental study, 18 rats were divided into three groups: Group A (n=6): Rats were injected with a single dose of 10mg.kg^{-1}Atropine. Group B (n=6): Rats were injected with a single dose of 10mg.kg^{-1} Oak Fruit husks. Group C (n=6): Rats received a single dose of 10mg.kg^{-1} saline. After the injection saliva volume was measured gravimetrically in four continuous seven minutes intervals. Results were analyzed by analysis of variance and tukey. (P<0.05 was considered significant). ***Results:*** The results showed that after injection of atropine salivation was significantly lower as compared to other groups. The peak of action was during the fourth 7 minute interval. In addition, the salivary flow rate decreased after injection of oak fruit husk extract but this decrease was not significant. Statistical analysis showed that the difference between atropine and two other matter, oak fruit husk extract and normal saline as significant (P<0.05). ***Conclusion:*** This study demonstrated that the extract of Oak fruit husk can decrease the rate of salivation in animal model.

Keywords: Sialorrhea , atropine , oak fruit husk .

Introduction

Sialorrhea is defined as an inability to control oral secretions which is manifested by drooling of saliva from the oral cavity and lips. Sialorrhea is induced by some factors such as medications, hyperhydration, and decreasein clearance due to a decrease in the esthesia of perioral muscles, open bite, and lip incompetence. The negative physical and social effects of sialorrhea on patients' lives cannot be ignoredwhich include aspiration pneumonia, partial or complete obstruction of airways, perioral ulcers, decrease in self-confidence,loneliness, depression[1, 2].

Behavior therapy, surgery and drug therapy are some treatment options that physicians take into account for sialorrhea[3]. Anticholinergic drugs, including scopolamine, glycopyrrolate and atropine, are used for the treatment ofsialorrhea. These medications block parasympathetic nerves to decrease salivary flow rate But are not appropriate for long-termuse because the high doses needed cause sideeffects such as urinary retention and visual disturbancesthat pose greater risks to the patient's health thansialorrhea[1,4].

The 21st century has been termed the era of herbal medicine. There is ever-increasing research into herbal medicines. One of these plants is theoak fruit 'husk' which has been reported to possess many therapeutic effects[5]. Tannin which hasastringent and antiseptic

effects is a chief component found in Iranian oak. It appears that the tannin in the oak fruit husk has a significant effect on decreasing salivary flow rate through its astringent effect[6,7,8].

Since no animal or human studies have been carried outon this subject, the present study was undertaken to evaluate the effect of oak fruit husk on the salivary flow rate in rats.

Materials and Methods

This experimental animal study was approved according to the guidelines of the ethics committee of Ahvaz Jundishapur University of Medical Sciences.

In this study, 18 adult female rats (weight: 200-300g) were used. The animals were kept in a well cross ventilated room with controlled temperature and humidity. Standard rodent food and tap water were available.Rats were divided randomly into three groups:Group A (n=6): Rats after anesthesia were injected with a single dose of 10 mg.kg-1 Atropine.Group B (n=6): Rats after anesthesia were injected with a single dose of 10mg.kg-1 Oak Fruit husks.Group C (n=6): Rats after anesthesia received a single dose of 10mgkg-1 saline.

Preparation of extracts:

Oak fruit husks were washed with distilled water to remove dirt andsoil, and shade dried. The dried materials submitted for a maceration process. The material was extracted twice with ethanol (80%). The extracts were filtered, pooled, and concentrated at high temperature (+50ºC) on a rotary evaporator (Heidolph, Germany). The extracts were suspended in normal saline with ethanol10% as cosolvent and stored in refrigerator within dark container.

Rats were anesthetized using a single intraperitoneal injection of 75 mg.kg-1 ketamine (Alfasan, Holland) and 5mg.kg-1 xylazine (Alfasan, Holland).

Technique IP injection:

Rats were held with their abdominal exposed in the left hand and the needle was inserted deeply into the abdominal cavity in the lower right quadrant. The needle angle was 15-20° and inserted approximately 5 mm.9

The unconscious rats were kept on a thermal pad to maintain their body temperature at the level of 37ºC. Before saliva collection, the oral cavity was wiped and dried with a cotton pellet and then 3 preweighed cotton pellets were inserted into the mouth of each animal: one cotton pellets underneath the tongue and one between the cheek and the teeth on either side. After seven minutes10, the cotton pellets were removed and weighed again on a precision weighing balance (Sartorius, Germany). The difference of the weight of the cotton pellets between two determinations was considered as the baseline weight of the saliva secreted.

The flow rate of saliva was determined gravimetrically, assuming that the specific gravity of saliva is 1 (i.e. 1gequals 1ml of saliva)

Following measurement of the baseline secreted saliva, the extracts were injected (10 mg.kg-1 bodyweight)10,intraperitoneally. The rate of saliva secretion was determined at four continuous seven-minute intervals. The investigator was blinded to all of the injected solutions in this study. Intraperitoneal injection was run in parallel in Group C (10ml.kg-1 normal saline mixed with cosolvent) and Group A (10mg.kg-1 atropine dissolved in distilled water and cosolvent). At the end of the experiments the rats were sacrificed by pentobarbital overdose.

Results

The results of the present study showed that salivary flow decreased significantly after injection of atropine with its peak effect at 21 minutes, i.e. the third 7-minute period. In addition, the results showed that the salivary flow rate decreased after injection of oak fruit husk extract with the maximum decrease during the last 7-minute period. . Statistical analysis showed that the difference between atropine and two other matter, oak fruit husk extract and normal saline was significant ($P<0.05$).

Table 1. Saliva secretion (Mean ± SD) before and after injection of herbal extract and atropine in four continuous seven-minute intervals

Drug	Time (Minutes)									
	0		7		14		21		28	
	Weight of the Saliva (mg)									
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Oak fruit husks	17	2.37	18.7	2.8	18.3	2.73	15.3	3.27	14.3	3.67
Atropine	23.2	4.59	11.3	2.94	9.3	2.94	8.8	2.79	10.5	2.07
Normal saline	17.5	3.08	17.5	3.08	17.3	3.08	17.3	3.08	17.3	3.2

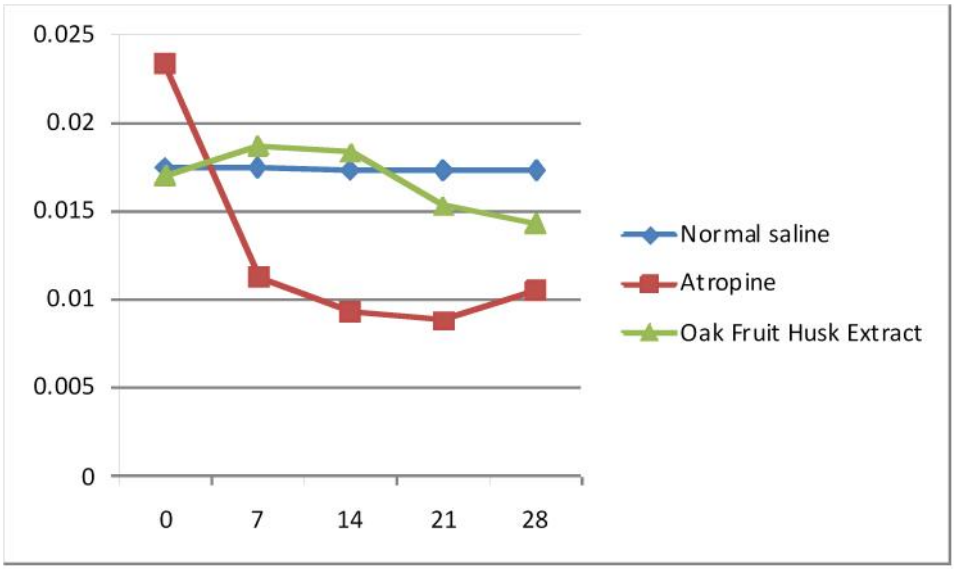

Figure 1. Saliva secretion before and after injection of three matter in four continuous seven-minute intervals

Discussion

Sialorrheais characterized by an inability to control the oral cavity secretionswith significant social and physical outcomes. The etiology of sialorrhea is the inability to swallow due to the poor control of oral and pharyngeal muscles rather than hypersalivation.Different treatment modalities are available for sialorrhea with varying success rates[11]. One of these treatments is drug therapy with anticholinergic agents. Atropine is one of these anticholinergic agentswhich decrease salivation by blocking muscarinicreceptors in salivary glands[12].

Based on the results of the present study the salivary flow rate decreased significantly after injection of atropine.The results of the present study are consistent with those of some other studies that evaluated the effect of atropine on salivation.

In this context, Hyson *et al.,* (2002) carried out a pilot study and evaluated the effect of sublingual atropine on sialorrhea secondary to Parkinson's disease, concluding that sublingual atropine resultedin objective and subjective improvements in sialorrhea[13]. Diamant*et al*., (1959) compared the activities of salivary glands during general anesthesia after injection of atropine, L-hyoscyamine (Bellafoline), scopolamine butylbromide (Buscopan), and oxyphenonium (Antrenyl).The result of their study reported that atropine was the best antisialagogue agent to decrease salivation during general anesthesia[14].

Rapoport(2010) evaluated the effect of sublingual atropine on sialorrhea in a 14-year-old boy with MLD and reported that it resulted in a subjective improvement in sialorrhea. However, no objective measurements were carried out in that study[15].

Mustafa *et al.,* (2013) evaluated the effect of sublingual atropine on the treatment of clozapine-induced sialorrhea and concluded that administration of sublingual atropine resulted in a significant decrease in salivation[16].

However, the results of some studiesdo not coincide with those of the present studywhich are mentioned here.

Desimone*et al*., (2006) reported no significant differences between atropine and placebo in the treatment of sialorrhea in patients with esophageal and gastric cancers. Althoughthe results of that study showed the efficacy of sublingual atropine, the results were not considered definite. The researchers attributed the negative results of the study to reasons such as small sample size, lack of homogeneity between the subjects, and more important than other reasons to the inadequate dose of atropine[17].

As well as Heisler*et al.,*(2013) evaluated the effect of sublingual atropine on the death rattle control in a double-blind study and reported no significant differences between placebo and atropine[18].

Despite the positive effect of atropine on the treatment of sialorrhea, its side effects have limited to its use which include visual disorders, urinary retention[4]. In addition, administration of anticholinergic agents is contraindicated in patients with glaucoma, obstructive uropathy, gastric disorders, and myasthenia gravis[3].

Another finding of the present study was a decrease in salivary flow rate subsequent to intraperitoneal injection of oak fruit husk extract in rats.

There is increasing research into herbal medicines and new herbal medicines are introduced regularly during this century which has been called the century to turn to nature and use of herbal medicines[5].

Different therapeutic effects have been attributed to the oak fruit husks. Ebrahimi*et al*, (2010) evaluated the antibacterial effect of hydroalcoholic extract of Iranian oak fruit and showed that different parts of oak tree exhibited antimicrobial effects and its antimicrobial effects increased withan increase in the concentration of the extract. In addition, the antimicrobial effects of the extract at high concentrations were similar to or even better than those of some antibiotics[19]. Sharifi*et al,* (2012) evaluated the antifungal effects of the hydroalcoholic extract of oak fruit husks and reported its effect on Saprolegnia fungal species. The extract of oak fruit husks prevented the growth of thisfungal species in a concentration-dependent manner and the antifungal effect increased with an increase in the concentration of the extract[20]. Nikrouz*et al.,*(2013) compared the effect of aqueous extract of oak fruit husk and silver sulfadiazine for healing of burn wounds in male rats and reported that the aqueous extract of oak fruit husks accelerated the healing of burn wounds and decreased the time necessary for complete healing of such wounds[21]. Since only one study is available on the effect of the extract of oak fruit husks on salivation, it is difficult to analyze the results due to a lack of similar studies.

One of the most important constituents of oak fruit husk is tannin which has astringent and antiseptic activities. Tannins are reported to have various physiological effects like anti-secratory, anti-irritant, anti-phlogistic, antimicrobial and anti-parasitic[22]. So, it seems reduce the amount of saliva after injection of oak fruit husk extract in rat because of anti-secratory effect of tannin. Furthermore the results of a study by Prinz*et al*.,(2000) on the effect of tannin on saliva showed that tannic acid (the common standard used for detection of tannin in tests) significantly decreased the lubricating effect of saliva by decreasing its viscosity and increasing friction, both of which highlight the astringent nature of saliva[8]. Therefore, it seems that topical use of oak fruit husk mouthwash can also decrease infected patients with sialorrhea.

Given that the present study is the first research has been done in this subject, more in vivo and in vitro studies is essential.

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How to cite this article:
Babadi F ,Hemmati AA, Davoudi AA, Hekmatinejad R,Kordian A. (2016). The Effect of Oak Fruit Husk Extract on Salivary Flow Rate in Rats. Int. J. Curr. Res. Chem. Pharm. Sci. 3(6): 51-55.