Diarrhea Associated with Ingestion of *Zygophyllum coccineum* (Tartir) in Camels in the Al-Najaf Desert in Iraq

Karima Akool Al-Salihi[1*] and Hayder M. Al-Rammahi[2]

[1]Faculty of Veterinary Medicine, Kufa University; currently Guest Fellowship, School of Veterinary Medicine and Science, University of Nottingham, Sutton Bonington Campus, Leicestershire, UK
[2] College of Veterinary Medicine, Al-Qassim Green University, Babylon, Iraq

*Corresponding author: Karima Akool Al-Salihi, kareema.nassrullah@nottingham.ac.uk

Abstract

Zygophyllum coccineum is a common plant found in areas of the Eastern (Arabian) Desert. It is rich in saponins, and its extracts have a variety of pharmacological actions, including acting as a diuretic and antipyretic. After several years of drought, during which many forbs and shrubs disappeared, heavy rains in 2012 and 2013 resulted in profuse growth of *Z. coccineum* (called "tartir" in Arabic). Field outbreaks of diarrhea in camels due to ingestion of *Z. coccineum* were reported in the Al-Najaf desert in Iraq. The clinical signs observed in the field cases resembled those reproduced in experimental rats and were consistent with acute diarrhea due to poisoning. Relocation of the camels to a different grazing area led to remission of the clinical signs.

Keywords: Al-Najaf desert, camel, Iraq, poisoning, *Zygophyllum coccineum*

Introduction

Camels *(Camelus dromedarius)* are important and versatile animals found in arid and semi-arid areas of the world. According to statistics from the Food and Agriculture Organization of the United Nations (FAO), there are approximately 25 million camels in the world. There were a total of 58,000 camels in Iraq, according to the FAO statistics of 2011 (Omer 2011). This estimation agrees with the statistical information of the Iraqi Veterinary General Company, which in 2008 had recorded a total of 58,893 camels in Iraq. This is an increase of 21.28% when compared to the Comprehensive Agrarian Census Data from 2001, which had recorded 23,413 camels.

The split upper lip of camels means that they can graze selectively up to a recorded height of 3.5 meters and utilize thorny plants. Camels have a reported preference for plants that have high water and salt content. It has been recorded that camels eat toxic plants avoided by other mammals (Dörges and Heucke 1997, Lapidge et al. 2008).

The Al-Najaf desert suffered from an absence of rainfall for several years, which led to the absence of pastures and disappearance of many grasses and other desert plants. But at the end of 2012 and in early 2013, all of Iraq's provinces experienced heavy rainfall, which led to growth of shrubs and forb plants in most desert areas. Consequently, many of the Bedouins moved to the areas flourishing with grass and green plants to graze their camels.

Z. coccineum has rarely grown in the past few years in the Iraqi desert due to the scarcity of rain. However, there was profuse growth of *Z. coccineum* after the heavy rain in 2012 and 2013 in the Al-Najaf desert. The plant is very common in the limestone "wadis" and plains of the Eastern (Arabian) Desert and is tolerant of saline soils. In the Egyptian deserts, *Z. coccineum*, from the family Zygophyllaceae, grows wild (Batanouny and Ezzat 1971, El-Hadidi 1972) and has been traditionally used in medicines as an antihelminthic, diuretic, antidiabetic, antiasthma, antigout, antirheumatic, and

antihypertensive agent (Saber and El-Moghazy 1960, Gibbons and Oriowo 2001).

Morphologically, *Z. coccineum* appears as a low shrub, perennial herb, or desert succulent undershrub, is up to 75 cm high, and is characterized by numerous stems and erect, young, green branches (Soliman 1939, Reed 1997). The leaves are 2-foliolate, over 10 mm long, cylindrical, bright green, glabrous, fleshy, and carried on a fleshy, long petiole. The plant is common, but it is unpalatable, and animals generally do not graze on it. It does not make good fuel. The plant is, therefore, neither used for grazing nor is it cut for fuel.

Many studies have shown the pharmacological action and toxicity of *Z. coccineum*. The aqueous extract of the plant is documented to cause lowering of blood pressure, and it acts as a diuretic, antipyretic, and local anesthetic. It has antihistamine activity and helps in the stimulation of an isolated amphibian heart, stimulation of a guinea pig's intestine, relaxation of an isolated intestine, contraction of the uterus, and vasodilation. The extract antagonized acetylcholine's action on skeletal muscle and acted additively with the muscle-relaxant effect of d-tubocurarine (Saad et al. 1967a,b, Aclinou et al. 1988, Gibbons and Oriowo 2001, Doligalska et al. 2011, Bhattacharya and Haldar 2012, Das et al. 2012).

Z. coccineum is a saponin-rich plant. Phytochemical investigations have revealed that the major secondary metabolites in *Z. coccineum* are a class of quinovic acid compounds belonging to the ursane-type triterpene saponins, including zygophylloside S, together with a known flavonoid glycoside and a sterol glycoside (Ghafoor 1974, Ahmad et al. 1990, 1992, 1993, Ghazala 1992, Elgamal et al. 1995, Amin et al. 2011).

The authors report here severe watery diarrhea, polyuria, and conjunctivitis in a herd of camels that grazed accidentally on *Z. coccineum*.

History of Outbreak

In May 2013, a herd of 40 adult camels and 10 young camel calves was moved to a new, flourishing grazing area in the Al-Hidaya/Al-Najaf/Iraqi desert. Green plants and low shrubs (figure 1) were growing in this area, and the camels readily grazed them. After 24 hours, all the adult camels started showing signs of severe diarrhea (figure 2A), excessive voiding of urine, restlessness, severe lacrimation, and photosensitization. One pregnant female died after severe bloating, respiratory distress, and polyuria with dark red urine. The diarrhea became worse as the camels continued to graze, and most animals became dehydrated. A few camels were in sternal recumbency and expressed high-pitched bleats, loud bellows, and roars in addition to straining and suffering during the passing of soft-formed green feces (figure 2B). Fecal samples and swabs were collected and sent for bacterial and parasitic investigations. Relocation of the camels to a different grazing area led to remission of these clinical signs.

Figure 1. *Zygophyllum coccineum.*

Figure 2. A: Tail and legs contaminated by feces in a camel with diarrhea. B: Camel passing soft green feces, which contaminate the surrounding thighs and tail.

Materials and Methods

In order to do the preliminary evaluation of the plant, a pilot study was designed. The plant is well known in pastoralism and is locally called "tartir". The plant was sent to the Herbal Center for scientific identification. Scientifically, the plant is called *Z. coccineum*. Soon after collection, 100 g of the fresh plant were washed thoroughly with water, separated into pieces, and immediately crushed thoroughly in tepid water (~ 40 °C) using a mechanical grinder.

After cooling to room temperature (24 ± 2 °C), the extract was separated from the remaining vegetable debris by pressing the material through a muslin cloth. The resulting liquid was filtered and kept as aliquots and considered as water suspension of the fresh *Z. coccineum* (WSFZC).

Four adult Sprague-Dawley male rats weighing 180 to 250 g were used in this study. The rats were obtained from the Laboratory Animal Unit, Department of Physiology and Pharmacology, Faculty of Veterinary Medicine (FVM), Kufa University (KU), Iraq. The rats were grouped randomly into 2 groups, housed in polyacrylic cages, and maintained under standard laboratory conditions (temperature 25 ± 2 °C with a dark/light cycle of 12/12 hours). They were allowed free access to the standard dry pellet diet and water ad libitum. The rats were acclimatized to laboratory conditions for 3 days before starting the experiment. All the procedures described were reviewed and approved by the Kufa University Research and Animal Ethical Committee (KU.FVM.AEC number 0706-2013). All rats were fasted for 12 hours with water ad libitum and placed in individual observation cages. The first group (treatment) was orally gavaged 5 mL WSFZC/rat for 2 days; the second group (control) was orally gavaged distilled water. The rats were observed to see whether they produced formed or unformed stool. Formed stool referred to stool that was in a pellet form or a rugby-ball shape. Unformed stool referred to feces that were muddy or watery, and this was judged as diarrhea. After 3 days, the rats were euthanized via an intraperitoneal injection of ketamine:xylazine. Subsequently, the rats were necropsied, and the liver, gastrointestinal tract, spleen, kidney, pancreas, and lymph nodes were collected. All the tissues were fixed in neutral buffer formalin (10%) and submitted to the Anatomy and Histology Department, Faculty of Veterinary Medicine, Kufa University, for histopathology processing.

Results

Testing of fecal samples from the camels revealed neither pathogenic bacteria nor parasitic infestation.

Two hours after oral gavage with WSFZC, the treatment group of rats showed diarrhea and passing of unformed stool, which was soft and green. Severe diarrhea continued the 2nd day, and the rats became dehydrated. They developed rough coats, aggressive behavior, and polyuria. They had an abnormally high consumption of water in comparison to the control group, which continued to pass formed stools.

Histopathological analysis did not reveal any specific pathological changes in the intestine or other tissues from the rats.

Discussion

Before the occurrence of diarrhea, the herd of camels was allowed to graze on dry grass shrubs. Different green desert plants grew in the Al-Najaf desert after a heavy rain, which encouraged the Bedouins to move their herds of camels to these areas. The camels grazed readily and were observed eating toxic plants normally avoided by other mammals (Dörges and Heucke 1997). The abundant presence of *Z. coccineum* in the region encouraged the camels to consume it quickly, which led to the occurrence of diarrhea, polyuria, and lacrimation in all adult animals. The young animals did not show any clinical signs, because they were suckling milk and did not eat the plant.

The clinical signs that appeared in these animals were compatible with the pharmacological and toxic effects of the *Z. coccineum* saponin compound that was identified in previous studies (Saad et al. 1967a,b, Johnson, et al. 1986, Aclinou et al. 1988, Gibbons and Oriowo 2001, Doligalska et al. 2011, Bhattacharya and Haldar 2012, Das et al. 2012). Ghazala (1992) isolated 5 triterpenoidal saponins from *Z. coccineum*. Several studies agree that saponin compounds affect animals and humans in both a positive and negative manner. Saponins play a biological role as they have membrane-permeabilizing, immunostimulant, and hypocholesterolemic properties, and they have been found to significantly affect growth and feed intake in animals. These compounds have been observed to kill protozoa, impair protein digestion and the uptake of vitamins and minerals in the gut, and act as hypoglycemic agents (Das et al. 2012).

A triterpenoid was found to be the most active compound and caused diarrhea in mice due to an increase in the motor activities of the normal bowel (Bhattacharya and Haldar 2012). In the present study, diarrhea was found to be one of the very prominent clinical signs in the camels as well as in the experimental rats. The symptom of diarrhea was compatible with the previous studies that demonstrated the gastrointestinal effects of the triterpenoidal saponin extract; it was a remarkable stimulant laxative that increased the motor activities of a normal bowel along with having prokinetic effects in normal as well as constipated Swiss albino mice, while inducing diarrhea with higher doses (Bhattacharya and Haldar 2012). It was due to this

fact that the severity of diarrhea was dose-dependent and some camels in the present outbreak showed severe diarrhea, as they had consumed large quantities of *Z. coccineum*, which contained triterpenoidal saponin. The diarrheal action due to the triterpenoidal compound could cause toxic effects when consumed in appreciable amounts and could be life-threatening in contraindicated subjects (Pasricha 2006).

Bhattacharya and Haldar (2012) investigated the gastrointestinal effect of triterpenoids on normal nonconstipated (naive) mice, to determine the increase in motor activities of the normal bowel, and in drug-induced constipated mice, to determine the excretory bowel activities to counteract the constipation. They found that oral administration of a triterpenoid produced a laxative effect in both naive and constipated mice in a dose-dependent manner. Triterpenoids modified the excretory bowel activities, as evidenced by an increase in fecal output, number of wet feces excreted, frequency and weight of stools, fecal water content, and diarrhea episodes in both naive and constipated mice. They agreed that triterpenoids accelerated the gastrointestinal propulsion rate and acted as stimulants for gastrointestinal motility, as it occurred in the case of putative stimulant laxatives, thereby confirming its action as a stimulant laxative. They also found that triterpenoids elicited gastrointestinal activity similar to that affected by the reference drug castor oil. An accelerated transit of the liquid digestive contents through the small intestine and colon did not permit adequate time for reabsorption of water and electrolytes and resulted in watery stool and diarrhea.

In the present outbreak, 1 pregnant female camel died after severe bloating, respiratory distress, polyuria, and dark red urine. These physiological disorders might be due to a *Z. coccineum* saponin compound. This was compatible with a previous observation, which found that saponin could play a role in the pathophysiology of bloating and was associated with bloat in ruminants (Wilkes and Godwin 1995). Saponin had the ability to form stable foams in low concentrations that led to reduced microbial protein production and protozoal numbers (Wilkes and Godwin 1995). In addition, saponins caused severe physiological disorders, such as hemolysis, gastroenteritis, paralysis, and death (Bondi et al. 1973). Other researchers found that saponins reduced the digestibility of forage in ruminants (Oleszek 1996, Small 1996, Oleszek et al. 1999).

Rats were used in the pilot investigation to determine the effects of *Z. coccineum*. Rats are more sensitive to plant compounds and toxins and have been used in screening techniques for detection of the toxicity of plant compounds. Several workers have used a rat bioassay to test for the presence of toxins in tropical pasture legumes (Strickland et al. 1987, Bindon and Lamond 1966). The treated rats showed diarrhea; however, neither a histopathological change in the intestine nor in any other tissue of the experimental rats was seen. These results supported the stimulant laxative mechanism of diarrhea that led to an increase in the motor activities of the digestive tract. The polyuria that appeared in the experimental rats as well as in the camels confirmed that *Z. coccineum* had a diuretic effect, as had been shown previously (Amin et al. 2011).

The severe lacrimation and photosensitization that has appeared in all camels may be due to the hypersensitivity reaction and the effect of the saponin compound on the immune system. Oda et al. (2000) demonstrated that saponin has the unique ability to stimulate the cell-mediated immune system as well as to enhance antibody production. The other advantage is that only a low dose of saponin is required for adjuvant activity (Oda et al. 2000). Saponins reportedly induce the production of cytokines, such as interleukins and interferons, that might mediate immunostimulant effects (Kensil 1996). The camels in this case were exposed to a high dose of saponins, which might have increased the uptake of other antigens from the gut and other membranes, leading to stimulation of the cell-mediated immune system, which further led to the development of lacrimation and photosensitization.

This is the first reported case of acute poisoning diarrhea in a herd of camels due to *Z. coccineum* with a high concentration of the saponin compound. The prominent clinical signs were severe diarrhea, polyuria, and photosensitization, which occurred due to the pharmacological action and toxicity effects of *Z. coccineum* saponins. A similar clinical presentation was reproduced in rats by giving them an oral gavage of the plant extract in water. This field outbreak showed the toxicity of *Z. coccineum*, although this plant has been traditionally used as a hypertensive, anthelminthic, diuretic, and for reducing blood sugar.

Precautions must be taken to prevent misuse by humans when making traditional medicines. More studies are highly recommended for further investigation.

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