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EFFECT OF ADDITION OF SELENIUM TO KURDI SHEEP AND ITS INTERACTIONS WITH SOME NECESSARY AND TOXIC ELEMENTS ON HEALTH AND THE ENVIRONMENT

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Abstract

This study was conducted to investigate the effect of the addition of selenium in inorganic form which is Sodium Selenite, and its effects on animal health and the environment and its effect on age by measuring some necessary and toxic elements in the muscles, liver and dung of Kurdi lambs and rams. Six (6) lambs were taken and separated into two groups with three (3) lambs in each group, the first group standard (control) group that fed on regular feed, while the second group fed on feed with added selenium with a concentration of 0.5 mg/kg of feed. In addition, Six (6) rams were taken and split into two groups with three (3) rams in each group. The first group of rams where control that fed on regular feed, whilst the second group fed on feed that supplied selenium with a concentration of 0.5 mg/kg of feed, in a 90-day trial. Organic matter (OM), carbohydrates (CHO), nitrogen (N), iron (Fe), zinc (Zn), manganese (Mn), copper (Cu), Molybdenum (Mo) and selenium (Se) were measured. The results showed that selenium supplementation had a significant effect ($P \leq 0.05$) in changing the elements that has been analyzed, and differences in some elements between lambs and rams. Moreover, Selenium supplementation enhances animal health, and there was no high level of Se in muscle and liver in concentration that causes damage to health or negatively affect the environment. The levels of the elements in the dung were significant ($P \leq 0.05$) and did not have an adverse effect on the environment. Furthermore, Concentrations of elements were all within the permitted limits of ANZFA, RDA, NIST, SRM, WHO, and Institute of Medicine (US). It concludes from these results that selenium supplementation with a concentration of 0.5 mg/kg of feed improves animal health and has no detrimental effect on the environment. Sheep dung can be used as fertilizer for plants which has an environmental benefit.

Key words : Selenium, Toxic Elements, environment and nutrients

Introduction

The pollution of the environment and nutrients causes the accumulation of heavy metals in human and animal bodies, which leads to inhibition of chemical reactions and enzymes in the cells, also causes malfunction in the organs and tissues of the body because of their toxic effect. The assessment of the pollution of heavy metals in the environment and food is to observe the harmful effects on human and animal health (Oymak *et al.,* 2017). Environmental pollution is one of the issues that threaten human and animal health around the world. More recently, pollution has increased due to the continuous and rapid growth in the population and the random exploitation of natural materials (Yabe *et al.,* 2011). Poisoning with heavy metals is one of the major health and environmental problems and it is dangerous due to bioaccumulation through nutrient substances (Ayciek *et al.*, 2008). This causes generally serious dangerous effects on human and animal wealth through these toxic elements and this depends on the level of available ingredients and absorption in the food (Aschner *et al.*, 2002). Selenium (Se) is an important nutrient for human and animal health (Preda *et al.*, 2016); because of extraordinary benefits such as: hormonal regulator, anti-carcinogen, and improves of the situation of antioxidants, and some serum biochemical indicators (Palani *et al.,* 2018 and Weeks and Hanna, 2012). Nevertheless, selenium can be toxic

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if it was consumed more than its adequate amount, and it can be one of the most toxic nutrients (Preda *et al*., 2015 and Zarczynska *et al.*, 2013). Selenium toxicity was discovered in animals; cattle grazing on plants that grown in seleniferous soil developed a peculiar condition called blind staggers, or alkali disease that could finally lead to death (Watts, 1994; Spallholz, 1994 and Spallholz, 1997). According to Preda *et al.,* (2015) and Bem (1981) selenium is present in the environment (soil, air, and water) in very low concentrations (<1µg/g). In addition, human activities (Agricultural and industrial utilize, agricultural irrigation of seleniferous soil in arid and semi-arid area, and disposal of fossil fuels wastes) can result in distributing the selenium compounds through the environment and poising wildlife, and fishes and threatened public health (Bueno *et al.*, 2007 and Lemely, 1997). Se considered as an environmental pollutant which obtained widespread attention among research scientists, natural resource managers, and regulatory agencies in the United State (Lemely, 1997). In several environmental pollutant circumstances, selenium has become the main element of concern owing to its bio-accumulative nature in food webs (Hamilton, 2004). It is one of the 129 priority pollutants that listed by Environmental Protection Agency (EPA), it is a toxic pollutant designated pursuant to section 307 (a) (1) of Clean Water Act (Irwin, 1997). An ecosystem can be disrupted by chemical pollutants; however, exact impacts could not be expected when the processes poorly known for example, food web dynamics are influential, food preferences, and uptake via diet (Stewart *et al.*, 2004). Selenium can enter the food chain through plants and aquatic organisms (Saranac *et al.*, 2011). The main passes into the food web are a combination of selenium by micro-heterotrophs and phytoplankton and uptake from sediment by benthic organisms. In either these two ways can convert selenium to new forms, or through being eaten it pass to greater trophic level (Preda *et al.*, 2015). Human health could be in danger if people consume Se contaminated wildlife and fish (Lemely, 1997). Besides, Soil is the basic resource of dietary selenium for animal and human. Se exists in the soil in different forms: selenates, selenides, elemental Se, and organic selenium compounds (Hall *et al.*, 2013). The typical total concentration of Se in soil globally is 0.01–2.0 mg/kg, with an overall mean of 0.4mg/kg (Fairweather-Tait *et al.*, 2011). The average of selenium intake for adult can differ depending on geographic area. For example, acidic soils release less selenium less than alkaline; moreover, Se concentration tends to be high in the driest regions (Joy *et al.*, 2015 and Mehdi *et al.*, 2013). While, Middle- East, China, India and some European countries have low content of selenium in the soil that leading to selenium deficiency. Se is a promising nutrient in prohibition and curing of several diseases, including cancer (Combs, 2005). Zarczyñska *et al.,* (2013) found that the selenium level in their cancer patients blood is very low; furthermore, in the soil of the region with Se deficiency, the incidence of neoplastic diseases is great. Se has an inhibitory or a stimulatory influence on the tumor growth in animal that are sensitive to the cytotoxic action of NK cells. It prevents angiogenesis and prompts the apoptosis of cancer cells, it also encourages the producing of anti-neoplastic metabolites (Zarczyñska *et al.*, 2013). Hence, there are varying kinds of supplements of selenium around the world with different species and concentrations. Moreover, it is frequently available in mineral/multivitamin supplements. These supplements generally utilize in the Europe, USA, Uinted Kingdom, Finland, Denmark, Spain and Poland (Fairweather-Tait *et al.*, 2011). In addition, because the intake of Selenium by human and cattle has been very low in Finland, and around the world Sodium Selenite has been added to multimineral fertilizers to raise the dietary intake of selenium (Parkman and Hultberg, 2002). Nonetheless, Se play a vital role in protective effect against toxicity of heavy metals; Se reduce the negative effects of excess intake of iron (Hosnedlova *et al.*, 2017). Recent studies have shown that pollution of the environment and food with toxic metals has reached unprecedented levels over the past decade and that the exposure of humans to toxic metals has become a serious health hazard on the continent (Yabe *et al.*, 2010). The European Food Safety Authority (EFSA, 2014) has decided to allow the addition of selenium to the feed of about 0.2 to 0.5 mg/kg of dry matter to avoid toxins and environmental pollution. This study aims to investigate the effect of selenium as supplements to feed sheep, and illustrates environmental and health effects by evaluating some necessary and toxic elements in the liver, muscles and dung.

Materials and Methods

The present study was conducted during May to August 2017 in summer season at University of Sulaimani, College of Agricultural Sciences, Animal Production Department. 6 Kurdish rams were taken with age 16-18 months, then the rams spread into two groups with three rams in each group. The first group control (0), the second group added Selenium (Sodium Selenite) Na_2Seo_3 with concentration of 0.5 mg/kg of feed, Moreover, 6 Kurdish lambs were taken in 3-4 months aged, these split into two groups with 3 lambs in each group. The first group control (0), the second one applied Selenium (Na_2Seo_3)

with a dose of 0.5 mg/kg of feed. After that, all rams and lambs randomly spread and put each one in a single cage with area of 1*1.5 m^3 for 90 days. The animal feed was consisted of 60% Barley, 12% Soybean which did not contain any amount of Selenium; beside that, the feed contained 26% Wheat bran that consist of 0.039% Se, 1% salt, 0.5% limestone, and 0.5% mixed of minerals and vitamins that had 0.01% Se. Capsule used to apply Selenium for animals, in which amount of Se has been taken and weighted by sensitive balance, and these amounts was according to amount of consumed pasture for each animal. Where the Se mixed with corn powder and put in empty capsule. Then the capsule has given daily to the animals through their month in the morning with their feed. Samples of dung have been collected in the early morning before feed them, and put it in special plastic bags for a week. Then, the dung dried in semi-open building, and the dung for each animal that had been taken during the week mixed all together. In addition, the dung of each animal put in pasteurized (sterilizer) closed container and saved in the refrigerator (frozen) to be prepared for chemical analysis. After that, the animals slaughtered and took samples of their liver and muscles and then dried in oven at 150ºC and grinding them. ICPE-9000 from Shimadzc Japanese made use to evaluate the minerals content in liver and muscle. Finally 200 mg samples were taken after diluting with 1-4 Sulphuric acid (H_2SO_4), Perchloric acid $Hclo_4$ for 16 hours, and added to 50 ml deionized water. Chemical analyzes of nitrogen, carbohydrates and organic matter were carried out by method of A.O.A.C (2005). The design of experiments was a Factorial Complete Randomized Design (CRD) to determine the effects of Selenium on age. The analysis is conducted by using XLstat (2017) according to this equation: M= mean, Ai= effect of apply Se factor, Bj: effect age factor, ABij= effect of interactions between two factors, eijk= standard error, means compared according to Dunken (1955) within the program.

Results and Discussion

This table 1 demonstrates that there were significant differences between the treatments. Moreover, there was significant improvement in organic matter (OM), carbohydrate (CHO), nitrogen (N) as affected by Se addition, compared to control in the dung of ram. While, there was significant decrease in organic matter and nitrogen of lamb's dung in Se treatment compared with the control.

In addition, there was a significant increase in organic matter for Se treatment of ram compare to the Se treatment of lamb. However, there were no differences between Se treatments of both ram and lamb in carbohydrate and nitrogen content concentration. Animal manures are a crucial source of nutrients for grasslands and crops, an adequate approach or application them, it could minimize environmental and human health impacts and maximize agricultural value (Shober and Maguire, 2018). In addition, use animal manure as a compost could replace the require for commercial chemical fertilizer, and it can improve profitability by meeting the crops need from nutrients and minimize the threat of polluting the water (Sutton, 1994). Furthermore, the animal manures have a nutrient and economical profit (Brown, 2013). Because these nutrients are necessary for plants; for example, Nitrogen is an essential nutrient for plant growth and the plant take it from the soil. Moreover, a typical plant contains 1.5% of N on dry weight; however, this rate relying on plant species that can range from 0.5-5.0% (Mahler, 2004). Additionally, using manures and sewage sludge are helpful in arid and semiarid soils, because organic matter acts as a reservoir for Selenium and preventing leaching and bioaccumulation. It also works as a source of nutrients (Moreno *et al*., 2013; Moral, 2008; and Moreno-Caselles, *et al.,* 2002). In the research of Angers and N'dayegamiye (1991) established that carbohydrates of both microbial origin and plant were improved upon application of dung to the soil.

Table 1: Effect of Selenium application on some Chemical Characterizes in Kurdi Sheep's Dung.

Type of animal	Treatment	%OM	%CHO	%N
Ram	Control	95.167c	6.517c	2.167b
	Selenium	96.603a	8.143ab	3.517a
Lamb	Control	95.900b	7.447b	4.010a
	Selenium	93.990d	8.627a	3.740a
SEM		0.122	0.176	0.109
LS		**	**	**

Means with different letters within each column differ significantly (Pd ≤ 0.05) according to Duncan's test.

The data in (Table 2) illustrates that there were decreases in each Fe, Cu, and Mo in selenium treatment compared to control; while, the level of Mn, Zn and Se increased in ram's dung. However, in case of lamb's dung there were a significant rise in Mn, and Zn of selenium treatment; whereas, no significant differences for Fe, Cu, Mo, Se. In addition, compare between dungs in treatments of selenium application of ram and lamb, there were a significant rise in Mn, and Zn levels; while, there were no significant differences in Fe, Cu, Mo, and Se contents. Zhang *et al*., (2012) mentioned that dung is a crucial source of heavy metal in the environment. The results of the study of Li *et al.,* (2009) showed that the contents of

Table 2: Effect of Selenium application on Micronutrients Content of Kurdi Sheep.

Organs	Type of Animal	Treatment	Fe ppm	Cu ppm	Mn ppm	Zn ppm	Mo ppb	Se ppb
Dung	Ram	Control	0.371b	26.433a	0.407f	0.292e	109.333d	176.333cd
		Selenium	0.291de	20.867bc	0.499c	0.549a	102.000e	207.000ab
	Lamb	Control	0.339bc	22.500b	0.562b	0.300e	139.667b	211.667ab
		Selenium	0.328cd	20.767bc	0.584a	0.493b	110.333d	215.333ab
Muscle	Ram	Control	0.307cd	19.033bcd	0.498c	0.293e	143.067b	170.667d
		Selenium	0.491a	15.033de	0.465d	0.461c	87.967g	183.667c
	Lamb	Control	0.257ef	16.433d	0.362g	0.234f	150.667a	203.333b
		Selenium	0.471a	17.067cd	0.561b	0.424d	109.000d	217.667a
Liver	Ram	Control	0.212g	21.567b	0.340h	0.174g	96.267f	188.000c
		Selenium	0.245fg	18.567bbcd	0.346gh	0.291e	97.400ef	203.000b
	Lamb	Control	0.289de	18.900bcd	0.430e	0.292e	89.867g	134.667e
		Selenium	0.456a	12.300e	0.453d	0.417d	120.833c	189.333c
SEM			0.009	0.937	0.004	0.006	1.330	3.127
LS			**	**	**	**	**	**

Means with different letters within each column differ significantly (Pd ≤0.05) according to Duncan's test

N, P, Zn, Cu were noticeable lower in sheep and cattle manures than chicken and pig manures; while, having the same amount of K. Besides, the average content of N, Zn and Cu in sheep manure were 1.31%, 88.9 and 23.5 mg.kg^{-1} respectively. According to Mahler (2004) micronutrients are essential for plant growth; however, increase amount of metal will lead to toxicity which causes multiple direct and indirect influences in plants that concern, especially all physiological functions (Barcelo and Poschenrieder, 1990). In case of ram's muscle, Fe, Zn, and Se recorded the highest level; while, the Cu, Mn, and Mo concentrations declined significantly in selenium application treatment. In lamb's muscle all micronutrient levels have increased in selenium treatment compared to control. Furthermore, treatments of selenium application in Ram and Lamb recorded the significant rise in Cu, Mo, and Se contents. Moreover, there were a significant increase in the amount of Zn, and Se; whilst, there was no difference in level of Fe, Cu, Mn, and Mo in liver of ram compared to control. In addition, the concentrations of Fe, Mn, Zn, Mo, and Se significantly increased; while, the level of Cu decreased significantly in lamb's liver in the selenium addition treatment. The contents of Fe, Mn, Zn, and Mo increased significantly in liver of lamb compared to the ram's liver in selenium application treatment; while, the other nutrients Cu and Se significantly reduced. The liver accumulates the greatest amount of Se, which is the reason to make it the typical sample for laboratory testing, followed by kidney and heart (McKenzie and Al-Dissi, 2017). In a study carried out by Zahrana and Hendyb (2015) results showed that when evaluating meat in Egypt, the iron level was 190.2, copper was 3.18, zinc was 137.4, manganese was 1.95 and selenium was 1.42 ppm. However, Oymak *et al.,* (2017) discovered that in evaluating cattle tissue, the level of manganese was 8.80, copper 386.4, molybdenum 4.88, selenium 2.32 mg/kg of dry matter in the liver; however, in the muscle manganese, copper, molybdenum, and selenium were (1.32, 6.90, 0.28, 1.15) mg/kg of dry matter respectively. The investigation of Shen *et al*., (2018) about the evaluation of liver and muscle of sheep in China, which is growing up nearly the zinc laboratory, proved that the concentrations of zinc, copper, manganese, and molybdenum in the liver were (233.9, 258.1, 4.7 and 1.4)

Table 3: Effect of Selenium application on Micronutrients in total of Dung, Muscle, and Liver in Ram and lamb of Kurdi Sheep.

Organs	treatment	Fe/ppm	Cu/ppm	Mn/ppm	Zn/ppm	Mo/ppb	Se/ppb
Dung	control	0.355b	24.467a	0.541a	0.296d	124.500b	194.000bc
	Selenium	0.310c	20.817b	0.513b	0.521a	106.167c	211.167a
Muscle	control	0.282d	17.733cd	0.485c	0.264e	146.867a	187.000c
	Selenium	0.481a	16.050d	0.430d	0.443b	98.483d	200.667b
Liver	control	0.251e	20.233bc	0.400e	0.233f	93.067e	161.333d
	Selenium	0.351b	15.433d	0.385f	0.354c	109.117c	196.167b
SEM		0.007	0.663	0.003	0.004	0.940	2.211
LS		**	**	**	**	**	**

Means with different letters within each column differ significantly (Pd≤0.05) according to Duncan's test.

mg/kg of body weight respectively; while, in the muscles were (137.9, 9.5, 2.5 and 0.79) mg/kg of body weight respectively. According to Badis *et al*., (2014) investigation, in Algeria sheep meat the level of iron was 70.36, copper 2.56 and zinc 39.6 microgram/g of dry matter. Nevertheless, Seiyaboh *et al*., (2018) found that the level of iron was 654.6, zinc 53.4, copper 140.05, manganese 2.062 mg/kg in the liver of Nigerian cows; whilst, in the muscle the level of iron, zinc, copper and manganese were (43.8, 38.0, 1.27 and 0.87) mg/kg.

The data in (Table 3) present that the level of Zn, and Se rose significantly, while Fe, Cu, Mn, and Mo decreased significantly in the total dung of ram and lamb. In case of total muscle of ram and lamb, the Fe, Zn, and Se recorded the highest level with selenium addition compared to control; while, there were significant declines for other (Mn, and Mo) except Cu slightly reduced. In addition, there were significant differences in the amount of micronutrients in the total liver. There was a significant increase in Fe, Zn, Mo, and Se content in selenium treatments compared to control; whilst, the level of Cu, and Mn decreased significantly in the total liver. Low level of Se, Co, Cu, and vitamin E in cattle decrease the capacity of separated neutrophils to kill bacteria and or yeast. Moreover, small amount of Cu decreases the production of antibody; however, it does not affect cell-mediated immunity. Nevertheless, low level of Cu can decrease interferon production and tumour necrosis factor via mononuclear cells (Spears, 2000).

Table 4 shows that there was a great increase in the content of Fe, Mn, Zn, and Se in selenium applications compared to control for ram; however, the level of Cu, and Mo decreased significantly. In addition, the concentration of Fe, Mn, Zn, and Se in lamb were increased significantly; while, the content of Cu, and Mo reduced in selenium treatment compared to the control. Mineral elements such as zinc, lead, copper, cadmium, chromium, iodine, and selenium are vital to animal health, production and survival as they are part of structural, physiological, catalytic and regulatory organism roles (Ries *et al.*, 2010). In addition, farm animals are greatly relying on their nutritional status (micronutrients) for their reproductive and performance., intracellular detoxification of free radials (Smith and Akinbamijo, 2000). Moreover, Soetan *et al.,* (2010) illustrated that copper, magnesium, selenium, iron, zinc, molybdenum and manganese are crucial co-factors found in some enzyme structure and are indispensable in abundant biochemical pathways. Nevertheless, Selenium has an antagonistic relationship with some importance or toxic element, such as: Cu, Co, As, Cd, Zn, Mn, Cu, Ni, Sn, Au, Bi, Ag, Pb, Hg, Mo and S. For instance, take a high level of Sulfur reduce its bioavailability in the organism and reduce the plasma Se level (Hosnedlova *et al*., 2017). The levels of toxic elements in this study were lower than in previous studies and were also lower than recommended levels in sources and organizations of food, environment and health. Where the level of iron allowed for children and adults from 40 to 45 mg/day (Institute of Medicine, 2003). Furthermore, the level of copper for adults is 900 to 1,300 micrograms/day (Stern, 2010). However, according to the World Health Organization (WHO) the toxicity of copper is 100 mg/kg. On the recommendation of NIST SRM (Standard Reference Material) was determined that the level of Mangis is 33, copper 4.02 and selenium 1.80 (mg/kg). When the levels that recommended by Recommended Dietary Allowance (RDA) the National Academy of Medicine in the United States of America set the iron level from 10 to 15, copper from 1.5 to 3.2, manganese from 300 to 500 and zinc from 12 to 15 (mg/day). While, the National Research Council of Canada identified the level of manganese for young people from 2 to 5 mg and for adults 10 mg; furthermore, manganese limits are 2.9, zinc 150 and copper 200 (ppm) according to ANZFA recommendations. Overall, this study found that all Rams and Lambs look healthy when use Selenium at ratio 0.5 mg\kg of feed. Palani (2019) pointed out that the low level of selenium in the blood of Kurdi sheep is due to its low level of Se in plants and the soil of Sulaimani governorate which is in Iraq Kurdistan Region. The differences in the concentration of selenium in the tissues of animale can be depended on environmental conditions (Kadim, 2013). In contrast, the research that's done by

Table 4: Effect of Selenium application on Micronutrients for total of Dung, Muscle, and Liver of Kurdi sheep.

Type of animal	Treatment	Fe/ppm	Cu/ppm	Mn/ppm	Zn/ppm	Mo/ppb	Se/ppb
Ram (total of dung, muscle and liver)	Control	0.297c	22.344a	0.415d	0.253c	116.222b	178.333c
	Selenium	0.342b	18.156bc	0.437c	0.434a	95.789c	197.889b
Lamb (total of dung, muscle and liver)	Control	0.295c	19.278b	0.451b	0.275b	126.733a	183.222c
	Selenium	0.418a	16.711c	0.533a	0.445a	113.389b	207.444a
SEM		0.005	0.541	0.002	0.003	0.768	1.805
LS		**	**	**	**	**	**

Means with different letters within each column differ significantly (Pd≤0.05) according to Duncan's test.

Kyle and Allen (1990) discovered that the rams received around 0.38 mg/kg of selenium survived; as well as, the ewes received about 0.50 mg/kg; while, the lambs received roughly 0.45 mg/kg of selenium of body weight died. Furthermore, Tiwary *et al*., (2006) established that using Sodium Selenite at concentrations of 2, 3 and 4 mg/kg resulted in respiratory distress and/or tachypnea.

Conclusion

Sodium Selenite supplementation with a concentration of 0.5 mg / kg of feed in Kurdi sheep has led to improve some important elements in the dung of Kurdi lambs and rams. This is considered as an adequate for use it as fertilizer for plants and which has no harmful effects on the environment and their levels within the limits allowed from ANZFA, RDA, NIST, SRM, WHO and Institute of Medicine (US). Moreover, there are no adverse effects of metal elements in muscle and liver, which is important for animal health as well as for human consumption.

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