Ecological evaluation of microelements in Astrakhan region and the dynamics of microelements in organs and tissues of Soviet Merino sheep

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ABSTRAK

Unsur mikro penting untuk menstabilkan struktur sel, tetapi dalam kondisi yang kurang baik elemen dapat merangsang jalur alternatif dan menyebabkan penyakit. Penelitian ini bertujuan untuk menyajikan data pemantauan tentang situasi biogeokimia ekosistem padang rumput di wilayah Astrakhan, Rusia selatan. Unsur mikro dalam sampel dikumpulkan dari ekosistem padang rumput, serta organ dan jaringan domba Merino Soviet, yang dianalisis dengan metode serapan atom. Hasil pengamatan menunjukkan bahwa tingkat mikro yang rendah di tanah, tanaman dan hijauan ekosistem di wilayah Astrakhan. Selain itu, konsentrasi mikroelemen yang rendah (selenium, yodium, dan kobalt) juga ditemukan di organ dan jaringan domba Merino. Hipomikroelementosis pada domba menyebabkan stres oksidatif pada hewan, menurunkan produktivitas dan menurunkan imunitas hewan, yang dapat menjadi predisposisi penyakit tertentu

Kata kunci : ekosistem, tanah, pakan, hipomikroelemntosis, stres oksidatif

ABSTRACT

Microelements are important for stabilizing cell structures, but in deficient conditions they can stimulate alternative pathways and cause disease. This study was aimed to presents the monitoring data on the biogeochemical situation of pasture ecosystems in the Astrakhan region, southern Russia. Microelements in the collected samples from the pasture ecosystem, as well as the organs and tissues of Soviet Merino sheep, were determined by atomic absorption method. It was found a low level of microelements in soil, plants and forages of the ecosystem in the Astrakhan region. In addition, it was found a low level of microelements (selenium, iodine, and cobalt) in the organs and tissues of Merino sheep. Hypomicroelementosis in sheep leads to oxidative stress in animals, lower productivity and decrease the immunity of animals, which can be a predisposing for other diseases.

Keywords: ecosystem, soil, forage, hypomicroelemntosis, oxidative stress

INTRODUCTION

Soviet Merino is one of the best fine-fleeced sheep breeds, which is successfully bred in the extremely arid zone of the Lower Volga region, including in the Astrakhan region, southern Russia. This breed, by its physical and mechanical properties, is suitable for extremely thin wool production. Living organisms require microelements for normal functioning. Mineral imbalances in soils and forages were considered the cause of low productivity and reproductive problems of grazing ruminants (Gorlov *et al.,* 2016; Vorobev, 2014; Riaz and Muhammad, 2018; Amer *et al.,* 2020). An analysis of the microelement profile of a particular area is mandatory to evaluate its microelement status in order to compare the presence of the level of microelements in grazing animals. Soil is one of the main sources of microelements for plants and forages. The availability of these elements depends on the concentration present in the soil and the nature of the soil. Animal nutrition depends on the soil-plant-animal complex and season, which can also affect the nutritional needs of trace elements (Syso *et al.,* 2017).

In addition to water and soil, forage is the main source of microelements for animals. The level of microelements in forage depends on the type and stage of feeding, season and soil type (Khan *et al.,* 2004). To assess the mineral status of animals, analysis of forages for the presence of microelements should be carried out regularly (Ogebe *et al.,* 1995). Deficiency of the microelements in the soil and forages adversely affect animal productivity (Vorobiov *et al.,* 2018). In animals, level of microelement deficiency affects the physiological process differently as these elements are involved in metabolic activities related to growth, reproduction and health. Sub-clinical deficiency of micronutrients may not affect growth and feed efficiency; this may cause impairment in reproduction and immunity (Shukla *et al.,* 2018). The presence of microelements in the organs and tissues of animals is one of the important diagnostic indicators. A latent syndrome of hypomicroelementosis is found in ruminants in regions where a low level of microelements was determined (Vorobiov *et al.,* 2017; Vorobiov *et al.,* 2018). Previous studies have shown that microelements contents of soils in the Astrakhan region are low and corresponding deficiencies in plants appear to reduce the plant communities'

productivity. Plants must maintain sufficient microelement contents and relatively stable microelement ratios (stoichiometric balances) for healthy growth (Vorobev, 2014; Vorobiov *et al.,* 2017). Meeting nutrient needs is essential in order to be free from malnutrition, but in many countries, the current local diet of vulnerable groups does not provide the nutrients required, resulting in a "nutrient gap". The aim of the work was to study the biogeochemical situation of microelements in the Astrakhan region and the level of microelements, for the first time, in the organs and tissues of Soviet Merino sheep.

MATERIALS AND METHODS

Study area and Experimental Design

For determination the levels of microelements in the ecosystem, this study was conducted during the period from April 2019 to June 2020 in the Astrakhan region, southern Russia, at a latitude and longitude of 46°20'58.85"N, 48°2'26.74"E. Astrakhan region is located in the Caspian Lowland where the Volga empties into the Caspian Sea. Its surface is flat and lies primarily below sea level with elevations ranging from -2.7 m in the north to -27.5 m in the south. The relief of the region is flat with saline-dome uplifts in the Caspian Lowland. The Astrakhan Region is located in the semi-desert zone. The climate is sharply continental and arid. The coldest month is January (temperatures average –10°C), and the warmest is July (temperatures average +26°C). Astrakhan region is characterized by sand dunes and ridges, salt soil, clay deserts (takyrs) and in places solonchaks (shors) or salt pans of 30–40 cm thickness devoid of vegetation. The samples from the ecosystem (soil, forages and water) were collected from the grazing areas of Astrakhan region. Moreover, Different tissues and organs from 30 Soviet Merino sheep were collected to determine the levels of microelements inside it. Experimental analysis was done at the Faculty of Veterinary Medicine, Astrakhan State University.

Sampling and Microelements Analysis

Medium samples of soils, water, plants, forages and different organs of 30 Soviet Merino sheep were selected for microelement analysis according to the procedure (Ermakov *et al.,* 2007; Khismetov and Vorobev, 2015). Soil samples from different depths from the grazing fields were taken with the help of a sampling auger. Five

Table 1. The Average Content of Microelements (Co, Se, I_2, Mn, Zn and Cu) in Soils and Plants in the Astrakhan Region, mg/kg

Microelement	Soil (n=40)	Plant (n=20)
Cobalt	7.3 ± 1.02	2.02 ± 0.13
Selenium	0.07 ± 0.021	0.13 ± 0.002
Iodine	0.4 ± 0.02	0.05 ± 0.005
Manganese	135.4 ± 8.6	38 ± 7.13
Zink	45.7 ± 3.8	33 ± 7.6
Copper	12.4 ± 0.25	8.52 ± 0.58

samples were collected from each of the selected grazing fields and a representative sample. Accurately weighed one gram of air-dried soil was processed following the method of Amacher (1996). Forage species in triplicate were collected from the grazing sites. The forage samples were collected after careful observation of grazing pattern of sheep. The leaves of forages were washed with 1% HCl followed by washing with distilled water. Air dried samples were again dried in drying oven (65±5 °C). The dried leaves were ground to powder by an electric grinding machine and subjected to wet digestion (Miller, 1998).

Microelements in the collected samples were determined by Atomic Spectrophotometry Absorption method on a CHITAHI 180-50 spectrophotometer (Japan). The samples was aerolized and mixed with gases (acetylene and air) after burned in flame. Individual atoms released which converted to excited state by passing UV light. On absorbing UV light the region of the spectrum to be measured. The detector and the output is amplified and sent to a computer data processing system and processed through software.

Animal Research Ethics

Analysis of microelements in different tissues and organs of sheep was carried out in accordance with the standards of humane treatment of animals and is based on the EU directive (86/609/EEC) and the Helsinki Declaration.

Statistical Analysis

The research data were processed statistically on a computer using SPSS computer 89 software v.20 (IBM, USA). Standard statistical characteristics were determined: arithmetic mean, coefficient of variation, errors of arithmetic mean and correlation coefficient. The significance of the differences was determined by Student's t test with a significance level of $P<0.01-0.05$.

RESULTS AND DISCUSSION

It was found that was found that various types of soils in the Astrakhan region contained low ranges of microelements (cobalt, selenium, iodine, manganese, zinc and copper) (Table 1). These results and ranges had close similarity with the ranges observed by (Vorobiov *et al.,* 2017; Vorobyov *et al.,* 2018). Soil is a direct and indirect source of microelements for livestock. The bioavailability of these microelements for livestock is related to their soil level (Galindo *et al.,* 2014), soil quality, lime, pH, electrical conductivity, plant species, and seasonal changes (Khan *et al.,* 2004). Selenium in soils depends on the geological parent material, among other factors. It is known that sandy soils have lower Se contents compared to organic and calcareous soils (Kabata-Pendias, 2011). Besides other variables, such as clay and organic matter contents, the major factors that control Se forms in soil are pH and redox potential (Eh) (El-Ramady *et al.,* 2014), this was in agreement with our results that recorded a low levels of Se in the soil. Se contents in plants are related with Se contents of the soils where plants grow. In the case of Se, it is well known its essentiality for animals and humans (Christophersen *et al.,* 2013). Reproductive problems and poor growth are observed in animals in areas with soil with a low level of microelements (Shukla *et al.,* 2018). Soil analysis to determine the level of microelements is

Table 2. Concentration of Microelements (Mn, Co, Cu and Se) of Forages in Astrakhan Region, mg/kg (n=10), Mean±SE

Forage species	Microelement			
	Mn	Co	Cu	Se
Lucerne hay	45.2 ± 2.10*	0.6 ± 0.01*	8.8 ± 1.07*	0.04 ± 0.002
Hay meadow	44.1 ± 2.03*	0.3 ± 0.005	6.4 ± 0.37*	0.01 ± 0.001
Bread crumbs	48.0 ± 1.62	0.38 ± 0.005	4.4 ± 0.25	0.02 ± 0.002
Delphinium consolida	69.1 ± 4.12*	0.04 ± 0.006	4.5 ± 0.28	0.07 ± 0.0006
Sudanese grass	91.1 ± 5.69*	0.05 ± 0.003	4.9 ± 0.27	0.08 ± 0.14
Birch leaf	41.1 ± 3.05	0.64 ± 0.02	5.9 ± 0.98	0.04 ± 0.007
Chenopodium quinoa	23.0 ± 2.10	-	2.8 ± 0.15	0.05 ± 0.006
Sandy oats	61.7 ± 1.39*	0.36 ± 0.05	5.5 ± 0.27	0.03 ± 0.009
Prickly thorn	25.1 ± 1.7	-	3.3 ± 0.69	0.04 ± 0.003
Camelthorn	16.2 ± 0.71	-	6.6 ± 0.08	0.05 ± 0.005
Nettle	129.2 ± 8.76*	-	6.7 ± 0.98	0.88 ± 0.059*
Wormwood sand	61.2 ± 3.54	0.49 ± 0.08	7.5 ± 0.87	1.06 ± 0.028*
Potatoes	61.3 ± 5.06	0.28 ± 0.03	7.6 ± 0.72	0.02 ± 0.004
Barley grain	7.1 ± 0.53	0.73 ± 0.02*	6.4 ± 0.18	0.04 ± 0.005
Aristida	26.6 ± 1.84	0.67 ± 0.35*	6.6 ± 0.19*	0.05 ± 0.005
Bran	15.1 ± 2.2	0.14 ± 0.02	11.1 ± 0.95	0.08 ± 0.0013
Anisantha tectorum	19.3 ± 1.05	0.03 ± 0.08	7.5 ± 0.97	1.67 ± 0.39*
Phragmites australis	14.5 ± 6.03	0.06 ± 0.07	8.5 ± 0.13*	0.18 ± 0.04
Compound feed	91.3 ± 16.0*	1.6 ± 0.05	7.7 ± 0.76	1.02 ± 0.07*
Astragalus	89.0 ± 6.3*	5.3 ± 0.15*	26.3 ± 2.93*	11.9 ± 3.14*

* (P<0.05 relative to other forage species

Table 3. Nutritional Value of Forages Consumed by Soviet Merino Sheep in the Geochemical Conditions of the Astrakhan Region, mg/kg (n=10), Mean±SE

Type of Forage	Carotene	Digestible Protein	Ca	P	Co	Cu	Se	I_2	Mn
Natural hay	5.2	36	2.1	2.2	0.3	7.2	0.04	0.06	91.3
Alfalfa hay	6.1	35	7.2	2.6	0.7	8.2	0.13	0.02	75
Barley	25.2	20	5.5	2.5	0.8	3.4	0.03	0.02	35.2
Oatmeal	23.3	22	2.6	4.7	0.7	9.3	0.15	0.18	43.1
Wheat bran	18.7	21	2.7	7.6	0.6	5.8	0.11	0.05	36.3
Birch leaf	15	22	1.7	1.4	0.03	1.9	0.24	0.018	23.4

Table 4. Levels of Some Microelements in Organs and Tissues of Soviet Merino Sheep Biogeochemical Conditions of Astrakhan Region, mg/kg (n=10), Mean±SE

Name	Se	Cu	Co	Mn	Zn	I_2
Muscles	0.03 ± 0.005	5.7 ± 0.33	0.05 ± 0.003	21.6 ± 1.13	$75.2 \pm 6.36*$	0.04 ± 0.003
Liver	0.32 ± 0.06	$17.1 \pm 0.21*$	$2.21 \pm 0.64*$	$45.5 \pm 7.12*$	114 ± 11.2	$0.25 \pm 0.002*$
Spleen	0.28 ± 0.003	$14.6 \pm 1.04*$	0.7 ± 0.02	35.6 ± 1.05	38.2 ± 3.21	0.07 ± 0.003
Blood	0.03 ± 0.002	$13.5 \pm 1.91*$	1.26 ± 0.05	$48.5 \pm 3.13*$	32.5 ± 2.10	0.23 ± 0.023
Lung	0.06 ± 0.002	24.2 ± 0.15	$0.73 \pm 0.004*$	26.5 ± 3.03	$108 \pm 4.17*$	0.22 ± 0.024
Kidney	$0.52 \pm 0.003*$	$12.7 \pm 0.05*$	0.64 ± 0.03	$48.4 \pm 2.14*$	$86 \pm 7.65*$	0.27 ± 0.003
Abomasm	$0.32 \pm 0.005*$	$15.3 \pm 0.04*$	0.94 ± 0.06	44.2 ± 1.25	$122 \pm 8.74*$	0.31 ± 0.07
Small intestine	0.43 ± 0.005	$21.2 \pm 0.03*$	0.96 ± 0.04	28.6 ± 2.03	76.4 ± 11.4	0.26 ± 0.018

* $P<0.05$ relative to other organs and tissues

important because it allows recommending mineral supplements for grazing animals (Vorobiov *et al.,* 2017). Adsorption of trace elements by roots is controlled by the concentration of other elements in soil solution (Violante *et al.,* 2010). Synergistic or antagonistic effect between soil trace elements should be considered while determining their bioavailability. High Zn, Fe, and P concentration in soil can inhibit Cu absorption by plant root system (Havlin *et al.,* 2005). It was established that the level of microelements, physiologically important for the body, in plants directly depends on their content in the soil, the nature of the mobility of chemical elements, pH, plant species and their physiological state (Vorobyov *et al.,* 2018). The levels of microelements (Co, Se, I_2, Mn, Zn, and Cu) were also below normal levels in plants. In the Volga water, the levels of Co, Se, I_2, Mn, Zn and Cu were recorded 0.01 ± 0.003, 0.02 ± 0.007, 0.004 ± 0.0003, 0.15 ± 0.009, 0.03 ± 0.003 and 0.19 ± 0.007 mg/L, respectively, which was consistent with the results obtained by studying the microelements in the main components of the ecosystems of the Lower Volga (Vorobiov *et al.,* 2017; Vorobev *et al.,* 2014). In the natural grazing system, the available feed is considered the main source of microelements for livestock. In addition to the nutritional value, these feeds can contain many substances that are beneficial for animals. Feed analysis should be a common practice for determining microelements, since the microelements in the feed consumed reflect the mineral content of grazing sheep (Ogebe *et al.,*

1995).

Table 2 shows that the low level of cobalt in the feed species was well comparable with the documented level (Polkovnichenko *et al.,* 2019). Manganese is found in plants in optimal amounts close to its level in similar macrophytes from the chernozem region (Lukin *et al.,* 2019). Basically, the level of Mn in feed depends on the level of Mn in the soil, but it has been reported that livestock receives a sufficient amount of Mn even in soil with a deficiency of Mn (Underwood, 1999). The concentration of Co in the forages depends on the concentration of Mn. The higher the Mn level in the soil, the lower the Co absorption in the feed. In soil, the presence of antagonistic microelements may decrease or increase the absorption of other microelements. Factors responsible for the presence of microelements in plants are land topography, plant species, and seasonal variability (Khan *et al.,* 2004). From Table 2, the concentration of Cu in the identified forage species was at the lower boundary of the normal level, which is consistent with the data of other authors (Vorobev, 2014; Vorobiov *et al.,* 2017). Low soil pH affects the concentration of Cu in plants, because the low pH increases the solubility of Fe, which reduces the absorption of Cu (Ginocchio *et al.*, 2002). The presence of Mo, Ca, and S acts as a Cu antagonist and shows that a higher level of these elements in the soil reduces the level of Cu in the feed (McDowell, 2003).

Given the nutritive value of feed for sheep in the biogeochemical conditions of the Astrakhan

region Table 3, we can note a low level of carotene in all the studied feed objects. We found low levels of selenium in natural hay and barley. A low level of iodine was also recorded in alfalfa hay, barley and birch leaf. Microelements are lost during harvesting and storing concentrated feed (Polkovnichenko *et al.*, 2019).

The level of microelements in the organs and tissues of the Soviet Merino sheep varied significantly, Table 4. The dynamic of the content of selenium in sheep was arranged in such a way that the level of concentration in the organs decreases: blood <wall of the small intestine <liver <wall of the abdominal cavity <spleen ≥ shell ≥ bone tissue> muscles. The cobalt content was as follows: coat> liver> bone> blood> abdominal wall ≥ intestinal wall ≥ kidney> muscle. The shrinking rows of manganese, copper, zinc and iodine were similar to each other with some slight signs (coat> bone tissue> blood> liver> kidney> renal wall> spleen> intestinal wall> muscle). Table 4 shows that in the organs and tissues of the studied Soviet Merino sheep there were a low level of Se, Co, I2 and other important microelements, which did not reach the normal level, it was in agreement with (Vorobiov *et al.*, 2017; Polkovnichenko *et al.*, 2019) who performed biogeochemical monitoring of the main components of terrestrial ecosystems of the Astrakhan region and recorded a low level of Se, Co an I2. Dietary resources of trace elements for animals include pasture, range plantation, forages and dietary supplements. Level of trace elements in livestock depends on interactions among nature of soil, plant types and species, maturity status, dry matter magnitude and environmental conditions (McDowell, 2003). Adequate levels of selenium are necessary for proper bone metabolism, immune, endocrine, iodine metabolism, and reproductive processes (Muegge *et al.*, 2016). Selenium is a key component of the active site of glutathione peroxidase (GSH-Px), which is essential to neutralize reactive oxygen that species produced during oxidation of unsaturated fatty acids (Rotruck *et al.*, 1973). Severe deficiency of Selenium in sheep results in white muscle disease (WMD) (Yildirim *et al.*, 2019). In ruminants, rumen microbes incorporate dietary cobalt into vitamin B_{12}, which is released in the abomasum and then absorbed by the small intestine. In the liver vitamin B_{12} forms an essential component of gluconeogenesis from propionate, a major source of energy in ruminants. Dietary deficiency of cobalt is known to be associated with 'pine', to which sheep are particularly sensitive (Hamer *et al.*, 2020). Low levels of the microelement iodine in the soil, water, plants, negatively influences the pituitary-thyroid system of the body and leads to the pathology of the thyroid gland in the result of violation of synthesis of thyroxine (T4) and triiodothyronine (T3) (Lomet *et al.*, 2018). Iodine deficiency has numerous negative effects on the development and formation of the body of animals and humans. The clinical manifestation of iodine deficiency in farm animals is characterized by small stature of adult animals and underdevelopment of young animals, alopecia (violation of wool growth), enophthalmos, the phenomenon of myxedema in the form of mucous edema of the intercellular region in cattle and sheep (Skripkin *et al.*, 2019).

CONCLUSION

In conclusion, the results of this study indicate that low level of some microelements in the ecosystem of Astrakhan region. Moreover, microelements level was low in organs and tissues of Soviet Merino sheep. Small changes in concentration of microelements due to soil and plant factors may create deficiency problems in grazing animals in this area and can predetermine oxidative stress and long-term development of the combined hypomicroelementosis.

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