# Determination of Some Chemical Elements in Animal Feed from Three Regions in Republic of Macedonia

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-----ABSTRACT-----

The objective of this investigation was to determine the content of heavy metals (arsenic, cadmium, cobalt, chromium, copper, iron, manganese, nickel, lead, vanadium and zinc) in animal feed (concentrate feed and silages of alfalfa and straw) from the regions of Kumanovo, Tetovo and Gostivar, Republic of Macedonia, and to evaluate the potential health risks of these metals to humans via milk consumption. The analysis was performed by using of atomic emission spectrometry with inductively coupled plasma (ICP-AES). It was found that the content of As, Cd, Co, Cr, Ni and Pb was bellow 1 mg/kg in the all analysed samples. It was established that the content of the other heavy metals (Cu, Fe, Mn, V and Zn) was lower in the animal feed samples from Kumanovo region that those originated from Tetovo and Gostivar regions. Target hazard quotient values of less than 1 indicate a relative absence of health risks associated with the consumption of milk and dairy products, as well as of meat and meat products.

Keywords: Animal feed, atomic emission spectrometry, different regions, heavy metals

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## I. INTRODUCTION

Heavy metal contaminants in animal feed pose a hazard to animal health and human food safety. Heavy metals including copper (Cu), cadmium (Cd), chromium (Cr), and lead (Pb) are potential bio-accumulative toxicants that may cause severe health problems even at low concentrations [1, 2]. Heavy metals, belonging in the group of undesirable substances are a definite human health hazard because of their biocummulativity [3]. The extensive contamination of various foods and beverages with heavy metals as well as their constant and continuous use represents a serious risk to human health, the most dangerous being lead, cadmium and mercury [4, 5]. Heavy metals are widely used in all fields of life (batteries, dyes, alloys, chemical compounds, pharmaceutical and cosmetic products) [6, 7, 8], thus suggesting that the risk of pollution is very high and therefore a strengthened control along the entire food chain is necessary.

The goal of this study was to determine the content of heavy metals (arsenic, cadmium, cobalt, chromium, copper, iron, manganese, nickel, lead, vanadium and zinc) in animal feed (concentrate feed and silages of alfalfa and straw) from the regions of Kumanovo, Tetovo and Gostivar in Republic of Macedonia, and to evaluate the potential health risks of these metals to humans *via* milk consumption.

## 2.1 Material and Methods

# **II. EXPERIMENTAL WORK**

For digestion the samples (0.5000 g) were placed in Teflon digestion vessels and 7 ml of trace pure HNO<sub>3</sub> (Merck, Germany), as well as 2 ml  $H_2O_2$  p.a. (Alkloid, Macedonia) were added. The vessels were capped closed,



tightened and placed in the rotor of Mars microwave digestion apparatus (CEM, USA). The plant samples were digested at 180 °C. After cooling the digested samples were quantitatively transferred into 25 ml calibrated flasks.

Stock solutions (11355 – ICP Multi Elements Standard IV, Merck) of 20 elements, with a concentration of 1000 mg  $l^{-1}$ , served for further dilutions. All chemical reagents used were of analytical grade. All vessels used were pre-cleaned by leaching for 24 h each in proportions of 1 part of HNO<sub>3</sub> and 3 parts of HCl, followed by rinsing with double distilled water.

The analyses were performed at the Institute of Chemistry, Faculty of Natural Sciences and Mathematics in Skopje. All analysed elements were determined by the application of ICP-AES (Varian, 715ES) applying ultrasonic nebulizer CETAC (ICP/U-5000 AT) for better sensitivity. The optimal instrumental parameters for these techniques are already described [9].

## **III. RESULTS AND DISCUSSION**

The contents of 21 elements deremined in the samples of animal feed from Kumanovo region are presented in Table 1, while the contents of the same elements in the feeds originated from the regions of Tetovo and Gostivar are given in Table 2. As it is shown in Tables 1 and 2, the concentrations of As in all samples is < 1 mg/kg. The content of arsenic in all feed samples was <1 mg/kg, showing that in comparison with the reference standards established by NRC [10] and EU [11], was within the permissible limits of 30 mg/kg and 2 mg/kg, respectively. The toxicity of As is dependent on the chemical valence and form. Inorganic As is usually more toxic than the organic one [12]. The EU maximum allowable level of As in general feed materials is 2 mg/kg, and for feed material containing fish, other aquatic animals and products derived is 25 mg/kg [13]. The maximum level of As in premixes is 12 mg/kg. The NRC maximum tolerable level is 30 mg/kg for cattle and 5 mg/kg for fish.

The content of the heavy metals such as: Cd, Co, Cr, Li, Ni, Pb, is also lower than 1 mg/kg. Cadmium is not an essential nutrient for animals or humans. Accumulation of Cd was seen in human kidney and liver, which can lead to kidney disease including proteinuria and kidney stone formation [14]. Recently, cadmium was classified as a carcinogen, showing with experimental animals after oral exposure and has been also shown to pose hazards to fish and aquatic invertebrates [15, 10]. Exposure to high concentrations of Pb can lead to blood disorders in mammals [16]. In animals and humans, cardiovascular, hematological and neurological signs occur at the lowest levels of Pb exposure whereas renal, gastrointestinal, hepatic and immunologic signs occur with higher doses or longer exposure times [10]. The maximum tolerable level in animal feed is 10 mg/kg for poultry, swine, fish and horse; and 100 mg/kg for cattle and sheep. Chromium concentration was found to be <1 mg/kg, which was much lower than the permissible limit set by NRC (500 ppm), [10]. There is no permissible limit for chromium in feeding stuff given by the EU. Although  $Cr^{3+}$  has been proven to be essential for biological pathways such as glucose metabolism, studies also showed that high concentrations of this ion can reduce weight gain for chicks and rats [10], while hexavalent  $Cr^{6+}$  is the most toxic, as well as carcinogen, allergen and acute irritant in humans.

The content of copper ranged from 4.04 mg/kg in the silage of straw to 21.3 mg/kg in the sample 2 of feed concentrate with an average value of 16.95 mg/kg. The content of copper in the silage of lucerne (alfalfa) was higher (8.51 mg/kg) compared with the silage of straw (4.04 mg/kg). The values of copper in the samples from Tetovo and Gostivar regions are presented in Table 2. It could be seen that the content of Cu in the animal feed from these regions is higher than that from the region of Kumanovo. The content of Al in the animal feed was variable, with the highest determined content in the alfalfa silage sample (2428 mg/kg) from Tetovo region, compared with the sample from Kumanovo region (220 mg/kg) and Gostivar region (136.5 mg/kg). The content of K in the animal feed was also variable, with the highest one observed in the alfalfa silage sample collected from the Kumanovo region (166369 mg/kg), while in the silage of straw it was 12599 mg/kg. The content of K in the silage to 44.3 mg/kg in the feed concentrates. As it was shown, the concentration of Zn was sixfold higer in the feed concentrates, compared to that in the silage. The content of phosphorous ranged from 627 mg/kg to 5196 mg/kg in the feed concentrates, compared to that of P in the silage (627 mg/kg).

From the results obtained it was established that the content of the other heavy metals (Cu, Fe, Mn, V and Zn) was lower in the animal feed samples from Kumanovo region that in those from Tetovo and Gostivar regions. In general, target hazard quotient value is less than 1 indicating a relative absence of health risks associated with the consumption of milk and dairy products, including meat and meat products.

Table 1. Content of chemical elements in animal feed and silage from Kumanovo region (mg/kg)

| Element | Feed         | Feed         | Alfalfa silage | Straw silage |
|---------|--------------|--------------|----------------|--------------|
|         | Concentrate, | Concentrate, | (lucerne)      |              |

|                | Sample No. 1 | Sample No. 2 |       |       |
|----------------|--------------|--------------|-------|-------|
| Aluminium, Al  | 110          | 10.8         | 220   | 15.5  |
| Arsenic, As    | < 1          | <1[          | <1    | <1    |
| Boron, B       | 3.45         | 2.15         | 80.5  | 2.35  |
| Barium, Ba     | 1.75         | 10.7         | 49.7  | 27.1  |
| Calcium, Ca    | 153          | 562          | 5802  | 1096  |
| Cadmium, Cd    | <1           | <1           | <1    | <1    |
| Cobalt, Co     | <1           | <1           | <1    | <1    |
| Chromium, Cr   | <1           | <1           | <1    | <1    |
| Copper, Cu     | 12.6         | 21.3         | 8.51  | 4.04  |
| Iron, Fe       | 104          | 66.5         | 184   | 26.8  |
| Potassium, K   | 2619         | 6832         | 16639 | 12599 |
| Lithium, Li    | <1           | <1           | <1    | <1    |
| Magnesium, Mg  | 869          | 2095         | 1257  | 513   |
| Manganese, Mn  | 6.64         | 97.5         | 70.9  | 19.7  |
| Sodium, Na     | 28.7         | 46.2         | 123   | 60.8  |
| Nickel, Ni     | <1           | <1           | <1    | <1    |
| Phosphorous, P | 1990         | 5196         | 627   | 627   |
| Lead, Pb       | <1           | <1           | <1    | <1    |
| Strontium, Sr  | 0.77         | 3.53         | 7.37  | 7.37  |
| Vanadium, V    | <1           | <1           | <1    | <1    |
| Zinc, Zn       | 10.4         | 44.3         | 7.6   | 7.6   |

**Table 2.** Content of chemical elements in animal feed and silage from the regions of Tetovo and Gostivar (mg/kg)

|                | Tetovo       | region      |                | Gostivar   | region         |
|----------------|--------------|-------------|----------------|------------|----------------|
| Element        | Feed         | Feed        | Alfalfa silage | Feed       | Alfalfa silage |
|                | concentrate, | concentrat, | (lucerne)      | concentrae | (lucerne)      |
|                | sample No.1  | sample No.2 | × ,            |            | × ,            |
| Aluminium, Al  | 99.8         | 46.6        | 2428           | 53.4       | 136.5          |
| Arsenic, As    | <1           | <1          | <1             | <1         | <1             |
| Boron, B       | 12.6         | 6.76        | 15.7           | 3.56       | 9.51           |
| Barium, Ba     | 5.29         | 5.64        | 50.9           | 16.8       | 10.5           |
| Calcium, Ca    | 15283        | 7461        | <1             | 743        | 4093           |
| Cadmium, Cd    | <1           | <1          | <1             | <1         | <1             |
| Cobalt, Co     | <1           | <1          | <1             | <1         | <1             |
| Chromium, Cr   | <1           | <1          | <1             | <1         | <1             |
| Copper, Cu     | 34.8         | 24.9        | 13.7           | 29.9       | 10.7           |
| Iron, Fe       | 171          | 99.6        | 2243           | 130        | 114            |
| Potassium, K   | 8702         | 4913        | 17205          | 8419       | 14768          |
| Lithium, Li    | <1           | <1          | <1             | <1         | <1             |
| Magnesium, Mg  | 3072         | 1862        | 1928           | 2944       | 1142           |
| Manganese, Mn  | 133          | 89.3        | 96.4           | 94.6       | 11.4           |
| Sodium, Na     | 2944         | 6387        | 625            | 69.0       | 528            |
| Nickel, Ni     | <1           | <1          | <1             | <1         | <1             |
| Phosphorous, P | 7537         | 3217        | <11904         | 6785       | 1748           |
| Lead, Pb       | <1           | <1          | <1             | <1         | <1             |
| Strontium, Sr  | 13.3         | 8.58        | 22.5           | 5.32       | 16.2           |
| Vanadium, V    | <1           | <1          | <1             | <1         | <1             |
| Zinc, Zn       | 299          | 81.8        | 20.9           | 64.3       | 10.7           |

## **IV. CONCLUSION**

The content of some chemical elements (arsenic, cadmium, cobalt, chromium, copper, iron, manganese, nickel, lead, vanadium and zinc) in the animal feed (feed concentrate and silages of alfalfa and straw) from the regions of Kumanovo, Tetovo and Gostivar in Republic of Macedonia was determined. It was found that the content of heavy metal elements As, Cd, Co, Cr, Ni and Pb was bellow 1 mg/kg in the all analysed samples. It was established that the content of the other elements (Cu, Fe, Mn, V and Zn) was lower in the animal feed samples from Kumanovo region that those from Tetovo and Gostivar regions. According to the obtained results it can be concluded that the all samples analysed were bellow the no permissible limit for content of heavy metals in

feeding stuff setted by the EU. The target hazard quotient values of less than 1 indicates a relative absence of health risks associated with the consumption of milk and dairy products, as well as of meat and meat products.

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