Proceedings of the 7th International Symposium on Metal Elements in Enviroment, Medicine and Biology, Timosoara, 247 – 250

POSSIBILITY OF SPECTROPHOTOMETRIC DETERMINATION OF MOLYBDENUM IN FEED

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ABSTRACT

In the work the results of testing the method of spectrophotometric determination of molybdenum are presented. Corn, sunflower meal, fish meal and mixed feed for poultry were used as matrix. With "recovery" test mean value of chemical yield was determined and it was 95.9% with a variation factor of only 2.5%. The results show that the above mentioned method can be applied in determining molybdenum in feed, having in mind its specificity, accuracy and short time that is needed for its performance.

***Key words**: feed, molybdenum, spectrophotometry*

INTRODUCTION

Molybdenum is an essential microelement, necessary for functioning several metalloenzymes, including xanthine oxydase, aldehyde oxydase and sulphite oxydase (Hurley and Doane, 1989). This is a transitional element, and in the compounds it appears in different oxidation state: +2, +3, +4, +5, +6.

Quantity of molybdenum in feed of herbal and animal origin is mostly according to animal needs. In natural conditions of nutrition, both of humans and animals, no problems have been noticed regarding Mo deficiency. In practice it is not the question of deficiency, but the problem of its excess. The excess amounts of molybdenum originating from feed causes reduced concentration of some microelements, like for example iron, copper and zinc. Cattle are most sensitive on surplus of Mo in feed (Puls, 1990). Molybdenum toxicity depends on proportions of Cu and Mo in diet, but also on compounds where Mo may be found. The toxic effect of molybdenum is most probably related to reduced metabolism of copper (Hurley and Doane, 1989).

The content of molybdenum is usually determined by atomic absorption and technique of inductively coupled plasma. Giving the fact that the mentioned methods require expensive equipment, and knowing what is the importance of Mo monitoring in biologic systems, the aim of this paper was to test possibilities of spectrophotometric method as an alternative analytic method for determination the content of molybdenum in feed.

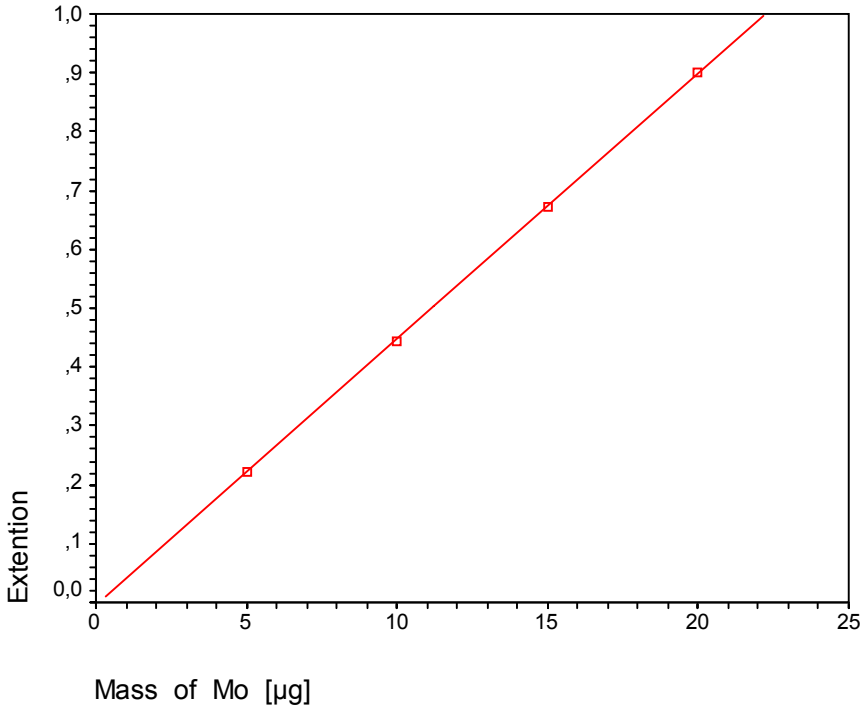
MATERIAL AND METHOD

Feed samples and full mix were taken in quantity of 1 kg, according to the standards for sampling. About 10 g of fine ground sample was mineralized, and the ashes were diluted in hydrochloric acid. The dilution was transferred into a funnel for separation with adding c.c. HCl, iron standard solution, $Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$ (100 μg/ml) and potassium thiocyanate solution (10%). After shaking for half a minute, stannous chloride solution (10%) was added, diluted with 25 ml of water and then again shaken. After that, 5 ml of isoamyl alcohol was added and shaken vigorously 1 min, phases were separated, drained and aqueous layer was discarded. It was extracted into alcohol without delay, since colored complex was somewhat unstable in aqueous solution. When alcohol layer was not clear, it was centrifuged 5 min at ca 2000 rpm. The developed color was read on a suitable spectrophotometer at λ=475 nm *(JUS, 1964; AOAC, 1998).*

RESULTS AND DISCUSSIONS

Calibration curve (Figure 1) was obtained with working standard solutions containing 5, 10, 15 and 20 μg molybdenum, respectively. Intensity of extended color was measured spectrophotometrically against reagent blank. Absorbance against corresponding mass of Mo [μg] was plotted.

Figure 1: Calibration curve of molybdenum solution



By the method of linear regression the following linear equation was obtained:

$$Ex = (0{,}0453 \times mass) - 0{,}006 ;$$

$$\text{Mass of Mo [µg]} = \frac{Ex + 0{,}006}{0{,}0453}$$

For determination of liquids, the "recovery" test was used, i.e. known quantity of standards was determined (1 mg/kg), than was added to different kinds of samples where molybdenum quantity had previously been measured. The obtained results are displayed in Table 1.

Table 1: The results of determination Mo content in feed samples with standard addition of Mo ("recovery" test)

Samples	**Mo [µg/kg]**	**Standard Mo addition [µg/kg]**	**Mo content in samples with standard addition [µg/kg]**	**Yield of method [%]**
Corn – grain	227 ±41	1 000	1 152 ±151	93.9
Sunflower meal	458 ±47	1 000	1 371 ±18	94.0
Mixed feed for poultry	1802 ±210	1 000	2 708 ±93	96.6
Fish meal	260 ±45	1 000	1 248 ±101	99.0

Accuracy of spectrophotometric detection of molybdenum for the samples without standard addition was determined by the mean value for standard deviation of all the samples and it was 14.3%. For serial measuring of the samples with standard addition of molybdenum, accuracy of the method was determined by the mean value of standard deviation for all the types of samples and it was only 6.5%. This shows that increase of Mo level in the examined material enhances method accuracy.

Table 2. Statistical analyses of chemical yield

Mean value of yield (%)	**Standard deviation (%)**	**Variation coefficient (%)**	**Standard error (%)**
95.9	2.4	2.5	1.2

Using "recovery" test (Table 2) the mean value of chemical yield was determined and it was 95.9% with variation coefficient of only 2.5%. The method was applied on chemically very different test-samples (matrixes). In all the samples there was a high yield (93.9 – 99.0%) with very low standard error (1.2%). This means that the method is highly specific, and interfering matters have minimal effect.

This points to the conclusion that the method is highly accurate and satisfactorily precise. Using this method we measured molybdenum in the following samples: wheat flour feed (1.11 mg/kg), soybean meal (1.28 mg/kg), mixed feed for cattle (0.85 mg/kg). The measured values of Mo content are according to the data in literature (NRC, 2001), as well as with the data on Mo content obtained by different analytic methods (Ćupić et all., 2006).

CONCLUSIONS

Based on the obtained results, it may be conclude that spectrophotometric determination of molybdenum content, given the fact of its simplicity and time of performance, accuracy and reproducibility, may be an alternative analytic method for determination of molybdenum in feed. The obtained results show that the level of molybdenum didn't exceed MDK 5 mg/kg (NRC, 1996), what refers to cattle as the most sensitive animal species.

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