Original scientific paper
Izvirni znanstveni prispevek

SULPHUR AND CHLORINE CONTENT IN FORAGES FROM DEFINED REGION

Marija RAJČEVIČ [a)], Anton ILC [b)], Peter KUMP [c)] and Marijan NEČEMER [c)]

[a)] Poslovni sistem Mercator, d.d., Dunajska 107, SI-1113 Ljubljana, Slovenia, Ph.D., M.Sc., senior scientist.
[b)] M-Kmetijsko gospodarstvo Kočevje, Kolodvorska 25, SI-1330 Kočevje, Slovenia.
[c)] "Jožef Stefan" Institute, Jamova 25, SI-1000 Ljubljana, Slovenia, Ph.D.

Received April 06, 2000, accepted October 16, 2000.
Delo je prispelo 06. aprila 2000, sprejeto 16. oktobra 2000.

ABSTRACT

In 1999 5 samples of pasture grass, 13 samples of grass silage, 6 samples of hay and 7 samples of maize silage were taken at random. All samples were from the defined region, and the forage was produced in similar soil and weather conditions. The grass botanical composition was comparable as well. The pasture and grasslands were manured by pig and bovine slurry. The content of sulphur and chlorine was determined by roentgen fluorescent analysis. Grass dry matter contained 2.56 ± 0.70 g of sulphur (CV = 27.29%) and 9.15 ± 4.18 g of chlorine (CV = 45.49%), grass silage 2.38 ± 0.49 g of sulphur (CV = 20.67%) and 7.19 ± 1.89 of chlorine (CV = 26.34%), hay 1.93 ± 0.48 g of sulphur (CV = 24.84%) and 5.72 ± 1.98 of chlorine (CV = 34.68%), and maize silage contained 0.88 ± 0.25 g of sulphur (CV = 28.14%) and 1.98 ± 0.86 g of chlorine (CV = 43.39%). Some results for sulphur as well as for chlorine deviated a great deal from the mean value and median. More attention should be paid to collection and preparation of samples for the analysis and determination of ash insoluble in hydrochloric acid.

Key words: forage / composition / sulphur / chlorine / content

VSEBNOST ŽVEPLA IN KLORA V VOLUMINOZNI KRMI Z DOLOČENEGA OBMOČJA

IZVLEČEK

V letu 1999 smo naključno odvzeli 5 vzorcev pašne trave, 13 vzorcev travne silaže, 6 vzorcev mrve in 7 vzorcev koruzne silaže. Vsi vzorci so bili z določenega območja, krma pa je bila pridelana v podobnih talnih in klimatskih razmerah. Tudi botanična sestava trav je bila podobna. Pašniki in travniki so bili gnojeni s prašičjo in govejo gnojevko. Vsebnost žvepla in klora smo določili z rentgensko fluorescenčno analizo. V suhi snovi trave je bilo 2,56 ± 0,70 g žvepla (KV = 27,29%) in 9,15 ± 4,18 g klora (KV = 45,49%) ,v travni silaži 2,38 ± 0,49 g žvepla (KV = 20,67%) in 7,19 ± 1,89 g klora (KV = 26,34%), v mrvi 1,93 ± 0,48 g žvepla (KV = 24,84%) in 5,72 ± 1,98 g klora (KV = 34,68%), v koruzni silaži pa 0,88 ± 0,25 g žvepla (KV = 28,14%) in 1,98 ± 0,86 g klora (KV = 43,39%). Nekatere določitve tako za žveplo kot za klor so precej odstopale od srednje vrednosti in mediane, vendar gre za manjše število analiziranih vzorcev. Priporočamo več pozornosti odvzemu in pripravi vzorcev za analizo ter določitev pepela, netopnega v klorvodikovi kislini.

Ključne besede: voluminozna krma / sestava / žveplo/ klor / vsebnost

INTRODUCTION

In the developed countries as well as in Slovenia it has been found out that actual milk production per cow does not match milk production that cows can achieve regarding their inherited ability. This difference varies from country to country.

Animal-friendly breeding technology, good control and suitable and balanced nutrition diminish those differences, with nutrition being the most important factor. The more we know the composition and traits of fed forage the better composition of rations can be achieved. Besides proper calculation of rations, it is important that supply of nutrients, minerals and vitamins enables good consumption, digestion and metabolism. It is especially important that rations contain a lot of fresh forage.

Calcium, phosphorus, magnesium, potassium and sodium are those macro elements that are most frequently studied in rations while sulphur and chlorine are not. Sulphur and chlorine are usually added as sulphates and chlorides in mineral and vitamin mixtures. We know their traits and role in metabolism but we do not know the contents of both minerals in forages produced in Slovenia. There are not many data on the content of sulphur and chlorine in fresh forages in literature.

On the agricultural estate Mercator in Kočevje the production of forages responds the needs of 1100 dairy cows and adequate number of heifers for reproduction of herd. The produced forages have regularly been chemically analysed.

MATERIAL AND METHODS

In 1999 five samples of pasture grass, thirteen samples of grass silage, six samples of hay and seven samples of maize silage were taken at random. Besides determination of usual mineral elements the contents of sulphur and chlorine were determined as well. Pastures are intensively supplied. The grass and legume mixture consists of 40% *Lolium perenne*, 20% of *Festuca rubra*, 20% of *Poa pratensis* and 20% of *Trifolium repens*. Pastures and grasslands are manured by pig and bovine slurry besides mineral fertilizers. The analysed voluminous forage was produced in similar soil, weather and agro-technical conditions. The botanical composition of grass is comparable. Samples of airing dried hay in the drying house were taken for analysis. The contents of sulphur and chlorine were determined by roentgen fluorescent analysis (Charalambous, 1984) at Jožef Stefan Institute in Ljubljana.

Preparation and analysis of sample by RFA

The content of S, Cl, K and Ca was determined by roentgen fluorescent spectrometry (RFS). The physics base of this technique, which has been used for nearly hundred years, as well as quantitative analyses have been well described in literature (Jenkins *et al.*, 1981, Kump *et al.*, 1996). Radioactive source ^{55}Fe was used for stimulation, and energy dispersion spectrometer with semi-conductive Si (Li) detector for measuring the roentgen spectra.

The procedure of simple preparation of sample starts with drying at 105° C following by grinding and homogenisation (mixing) of sample and preparation of pressed pill from achieved powdered material. Quantification is based on intensity of measured characteristic lines of each element that are hulled from spectrum by special computer program. The relation between the intensity of measured roentgen rays and the concentration of element, which is not linear in general, is obtained by measuring the analysing system and calculation of matrix corrections on the base of additional absorption measure in the sample (Kump *et al.*, 1996). The errors estimated in all phases of mentioned quantification procedure are summed and presented as uncertainty at the end. This parameter corresponds to parameter of standard deviation in statistical analysis of repeated set of measurements (Guide …, 1993). Results obtained by roentgen fluorescent analysis are more precise than estimated uncertainty, being 2 to 3% in general depending on numeric statistics in the measured spectrum while the uncertainty of analysis results ranges between 5 and 10%. The accuracy of obtained results is estimated by comparison of results obtained by other techniques (Kump *et al.*, 1996) or by analysing the suitable standard reference materials. In our case the SRM NIST 1570 analysis (spinach leaves) shows the accuracy of our results from 5 to 10%, which is within the estimated uncertainty of obtained results also presented in the table 1.

Table 1. Results for some elements in the SRM NIST Certified Reference Material No 1570 Spinach Levels, n = 4

	Content (% of DM)	
Elements	Obtained results	Certified value
Phosphorus	0.54 ± 0.05	0.55 ± 0.03
Potasium	3.48 ± 0.15	3.56 ± 0.03
Calcium	1.21 ± 0.05	1.35 ± 0.03

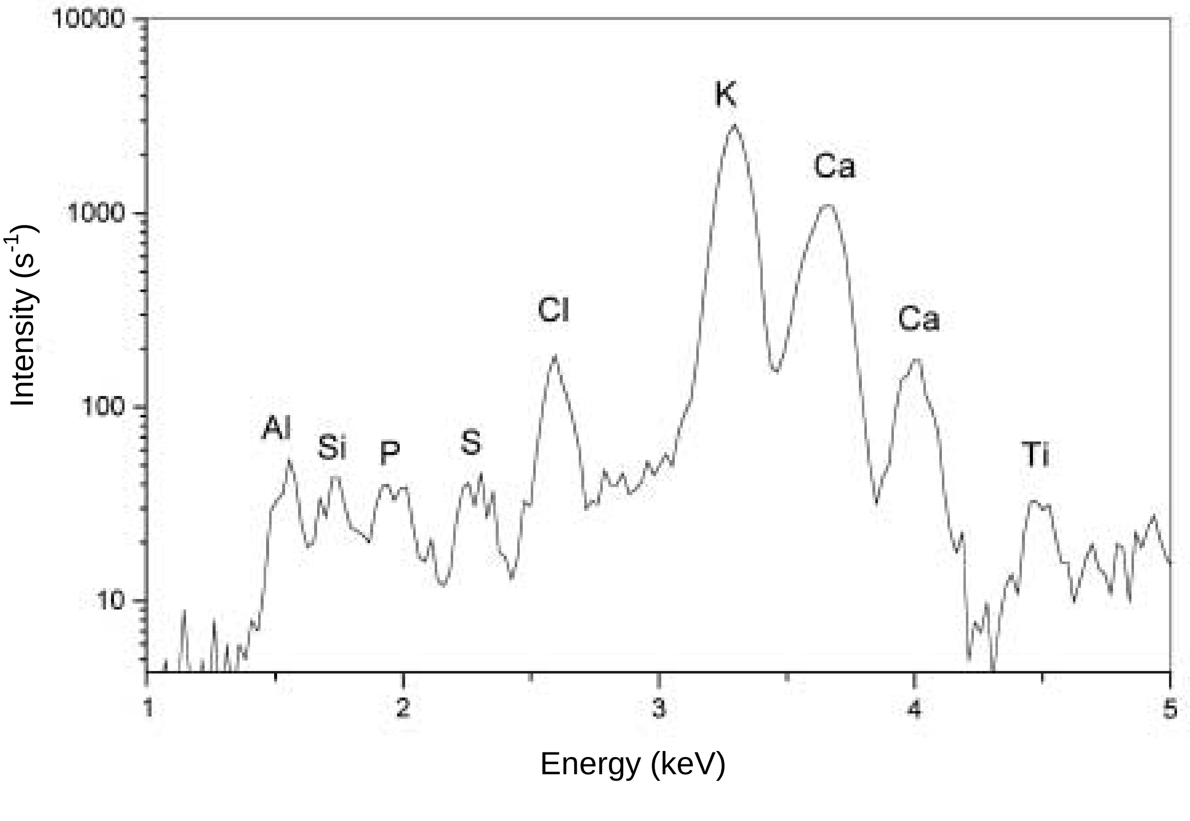

Figure 1. Part of roentgen spectrum for grass silage sample.

RESULTS AND DISCUSSION

The content of sulphur and chlorine in the studied samples of forage is shown in Tables 2 to 5.

Table 2. The sulphur and chlorine content in pasture, g kg^{-1} DM

Sample	Dry matter, g kg^{-1}	Crude ash	Sulphur	Chlorine
1	162.20	94.30	2.52	12.97
2	137.80	104.50	3.33	4.00
3	121.40	115.30	3.21	5.79
4	139.50	107.50	1.87	9.70
5	142.80	112.70	1.88	13.30
Mean	140.74	106.86	2.56	9.15
Median	139.50	107.50	2.52	9.70
Standard deviation (±)	14.57	8.20	0.70	4.18
Coefficient of variability (%)	10.35	7.68	27.29	45.69

Table 2 displays that values for sulphur are within the normal distribution (mean value ± 3 SD) in the five samples of pasture grass. Despite that, the determinations deviate from the mean value and median. Only sample 1 approximates them, while determinations for samples 2 and 3 deviate and reach the upper value and samples 4 and 5 the lower one. The range among determinations is significant, the coefficient of variability being 27.29%.

There were no analytical data on the content of sulphur and chlorine in forage produced in Slovenia, and few data were found in the foreign literature. Underwood (1981) reported that the content of sulphur in the pasture grass ranged between 0.1 and 0.5% of dry matter and NRC tables (1988) quote 0.33% of sulphur in dry matter in clovers. The NRC tables (1988) and McDowell (1992) quoted data on mineral elements in grasses and legumes. The most frequently reported three of four plants have the following contents of sulphur in dry matter: *Lolium perenne* 0.30% (McDowell, 1992), *Poa pretensis* 0.15 to 0.17% (NRC, 1988) and 0.29% (McDowell, 1992) respectively, and *Trifolium repens* 0. 21% (NRC, 1988). McDowell reported that *Dactylis glomerata* contained 0.21% of sulphur, and *Phleum pratense* 0.13% in dry matter. According to the NRC tables (1988) *Trifolium incarnatum* contains 0.28% of sulphur in dry matter, while *Trifolium pratense* contains 0.17% as quoted in the NRC tables (1988) and by McDowell (1992).

Underwood (1981) supposed that the shortage of sulphur in pasture is hardly believable. McDowell (1985) reported that ruminants on pasture needed between 0.10 and 0.32% of sulphur in dry matter. Naylor and Ralston (1991) stated that with great amounts of wheat and grass silage and little dry legumes in rations cows consumed less sulphur. More hay from legumes in the ration satisfies to all needs of sulphur. McDonald *et al.* (1995) concluded that animals in intensive breeding needed extra sulphur whenever manuring did not restore its content in plants.

Above mentioned refers to the content of chlorine in pasture grass too. The mean value is 9.15 g kg^{-1} in dry matter. The difference between determinations is significant with the coefficient of variability 45.69% and a low number of analysed samples. Determinations in samples are within the normal distribution but the value for sample 4 equals to median (9.70 g kg^{-1} dry matter), determinations for samples 1 and 5 reach the highest values (12.97 and 13.5 g kg^{-1} dry matter) while samples 2 and 3 are within the limits of lower value (4.0 and 5.79 g kg^{-1} dry matter).

According to the literature the content of chlorine in pasture grass varies a lot. McDonald *et al.* (1995) reported on wide range of chlorine content in pasture grass, i.e. between 3 and 25 g kg^{-1} dry matter. Several factors affect the contents. Underwood (1981) informed that the content of chlorine in the pasture grass was 0.5% of dry matter and 0.4% in legumes. As indicated by Kellner and Becker (1971), the content of chlorine in grass from intensively supplied grasslands varied between 15.5 g kg^{-1} dry matter in young grass (16% dry matter) and 5 g kg^{-1} dry matter in grass of worse quality (25% dry matter). McDowell (1992) stated that *Poa pratensis* contained 0.4% of chlorine, but NRC tables (1988) quote 0.3% of chlorine in *Trifolium repens*. NRC tables (1988) quote 0.32% of chlorine in dry matter for *Trifolium pratense* and McDowell (1992) 0.72%. The contents of chlorine in the studied samples were within the range stated by Kellner and Becker (1971) and McDonald *et al.* (1995).

Table 3. The sulphur and chlorine content in grass silage, g kg^{-1} DM

Sample	Dry matter, g kg^{-1}	Crude ash	Sulphur	Chlorine
1	289.00	143.59	2.39	7.85
2	439.40	82.61	2.06	4.42
3	338.70	98.61	2.38	6.02
4	419.30	130.21	2.13	5.52
5	512.50	**213.26**	2.51	**11.03**
6	260.20	105.68	**1.74**	4.78
7	248.60	146.00	**3.42**	9.86
8	256.20	93.67	2.03	7.87
9	261.00	98.08	1.84	6.13
10	246.00	98.25	2.20	7.09
11	430.20	157.37	2.74	7.45
12	327.00	**246.48**	3.18	8.36
13	336.50	134.35	2.29	7.05
Mean	335.74	134.47	2.38	7.19
Median	327.00	130.21	2.29	7.09
Standard deviation (±)	88.24	48.83	0.49	1.89
Coefficient of variability (%)	26.28	36.32	20.67	26.34

The mean value for crude ash is 134.35 g kg^{-1} dry matter and is high. Sample 5 (213.26 g kg^{-1} dry matter) and sample 12 (246.48 g kg^{-1} dry matter) especially deviate. If we exclude them, the content of ash will be 117.31 g kg^{-1} dry matter. The high content of ash in some samples is the consequence of unfavourable weather conditions during the ensiling. Therefore, ash insoluble in hydrochloric acid should be determined as well.

The mean value for thirteen determinations of sulphur in the grass silage is 2.38 g kg^{-1} dry matter. Median is a bit lower (2.29 g kg^{-1} dry matter). The values of 13 determinations are within the limits of normal distribution. Nevertheless, some determinations deviate from the main value and median (samples 6 and 9, 7 and 12). Sample 6 contains the lowest amount of sulphur and sample 7 the highest. The coefficient of variability is 20.67%.

The mean determined value for chlorine in grass silage was 7.19 g kg^{-1} dry matter, with a bit lower median (7.09 g kg^{-1} dry matter). The determinations ranged between 4.42 g (sample 2) and

11.03 g kg^{-1} in dry matter (sample 5); the coefficient of variability being 26.34%. The values for all thirteen determinations were within the limits of normal distribution. The determination in sample 5 deviated the most. Kellner and Becker (1971) reported that silage from young grass contained 17 g of chlorine in a kg of dry matter.

Table 4. The sulphur and chlorine content in hay, g kg^{-1} DM

Sample	Dry matter, g kg^{-1}	Crude ash	Sulphur	Chlorine
1	883.00	71.85	1.94	**2.40**
2	871.10	71.80	**2.52**	5.04
3	890.30	68.34	**1.46**	5.90
4	843.70	82.49	1.85	7.24
5	830.30	69.60	**1.37**	**8.13**
6	920.00	73.00	**2.44**	5.59
Mean	873.07	71.85	1.93	5.72
Mediane	877.05	71.82	1.89	5.74
Standard deviation (±)	32.54	6.48	0.48	1.98
Coefficient variability (%)	3.73	9.02	24.84	34.68

Table 4 shows that the content of sulphur in all six samples of hay is within the normal distribution. Nevertheless, only two determinations approach the mean value (1.93 g kg^{-1} dry matter) and median (1.89 g kg^{-1} dry matter) respectively, while the determinations of samples 2 and 6 deviate upwards and for samples 3 and 5 downwards. The coefficient of variability is 24.84%.

NRC tables (1988) quote 0.16% of sulphur in dry matter in *Poa pratensis*, 0.17% in *Phleum pratense*, 0.21 in *Trifolium repens*, 0.17% in *Trifolium pratense* and 0.28% in *Trifolium incarnatum*, all in dry matter. The reported data concern the sun-dried hay.

Chlorine determinations in hay range between 2.40 and 8.13 g kg^{-1} dry matter, the coefficient of variability being 34.68%. The number of samples was low. The difference between the mean value (5.72 g kg^{-1} dry matter) and median (5.74 g kg^{-1} dry matter) is slight but not any determination approaches it. Determinations 1 and 5 deviate the most although within the limits of normal distribution.

Kellner and Becker (1971) reported that the hay of best quality contained 11.29 g kg^{-1} chlorine in dry matter, while the hay of worse quality 4 g. The first cut hay of good quality contained between 3.5 and 16 g of chlorine, while good quality second cut hay contained between 7.8 and 16 g and hay of mid quality between 8.82 and 12.9 g of chlorine in a kg of dry matter. NRC tables (1988) quotes 0.3% of chlorine in dry matter in the hay of *Trifolium repens*, 0.28% in the hay of *Trifolium incarnatum* and 0.32% in the hay of *Trifolium pratense*. The same value for *Trifolium pratense* was determined by McDowell (1992), while he quoted 0.53% of chlorine in dry matter in the hay of *Poa pratensis* and 0.51% in the hay of *Phleum pratense*.

Table 5 indicates that the content of sulphur in sample 7 significantly deviates but chemical analyses show that its value is within the limits of normal distribution. If we omit this sample from calculations, the mean value for sulphur is 0.80 g kg^{-1} dry matter. Determination 2 deviates too while the values of other five determinations quite match. Since the value of sample 7 deviates, the range among determination is wide, between 0.55 and 1.37 g kg^{-1} dry matter with the coefficient of variability 28.14%. Sample 7 contains a lot of crude ash. If we omit this sample, the mean value for ash is 34.38 g kg^{-1} dry matter.

Table 5. The sulphur and chlorine content in maize silage, g kg^{-1} DM

Vzorec	Dry matter, g kg^{-1}	Crude ash	Sulphur	Chlorine
1	354.00	32.78	0.82	1.85
2	328.00	32.12	**0.55**	1.57
3	321.30	34.86	0.80	1.74
4	327.90	33.25	0.92	1.87
5	332.80	33.20	0.87	1.22
6	294.60	40.05	0.81	1.77
7	352.50	**74.33**	**1.37**	**3.87**
Mean	330.16	40.08	0.88	1.98
Median	328.00	33.25	0.82	1.77
Standard deviation (±)	20.11	15.34	0.25	0.86
Coefficient of variability (%)	6.09	38.26	28.14	43.39

NRC tables (1988) quote that maize silage contains between 0.08 and 0.15% of sulphur in dry matter. McDowell (1992) reported 0.13%. Mean value of determinations quoted in Table 5 (0.88 g kg^{-1} dry matter) is within the lower limit of NRC quotations.

Sample 7 is an exception in chlorine determination even if the value 3.87 g kg^{-1} dry matter is still within the limits of normal distribution. If we omit this determination, the mean value for chlorine will be 1.67 g kg^{-1} dry matter. Nevertheless, mean value as well as median approach the quoted values in literature, i.e. 0.18% of chlorine in dry matter of maize silage (McDowell, 1992).

CONCLUSION

The content of sulphur and chlorine in the studied forages significantly varied although the samples were taken in the defined region and in similar production conditions. The contents of sulphur are within the quotations in literature as well as the content of chlorine in the maize silage. Forage produced on grassland contains as much chlorine as Kellner and Becker (1971) reported and is within the limits of wide range reported by McDonald *et al.* (1995), i.e. between 3 and 25 g in a kg of dry matter. We recommend more attention to collection and preparation of samples for analysis. Data for ash insoluble in hydrochloric acid are recommended as well.

POVZETEK

Od makroelementov običajno v obrokih izračunavamo kalcij, fosfor, magnezij, kalij in natrij, medtem ko žveplo in klor le redko. Slednja dva najpogosteje dodajamo živalim v obliki različnih virov (sulfati, kloridi) v mineralno-vitaminskih mešanicah. Njune lastnosti in vloga v presnovi so nam poznane, manj pa poznamo vsebnost teh dveh mineralnih elementov v krmi, pridelani v Sloveniji.

Na Mercatorjevem Kmetijskem gospodarstvu v Kočevju letno pridelamo krmo za 1100 krav molznic in potrebno število podmladka za obnovo črede. Krmo redno kemično analiziramo. V letu 1999 smo v 5 vzorcih pašne trave, 13 vzorcih travne silaže, 6 vzorcih sena in 7 vzorcih koruzne silaže analizirali vsebnost žvepla in klora. Uporabili smo rentgensko fluorescentno analizo (Charalambous, 1984).

Določili smo naslednje povprečne vrednosti (g kg^{-1} SS):

- pašna trava: 2,56 ± 0,699 S in 9,15 ± 4,18 Cl,
- travna silaža: 2,38 ± 0,49 S in 7,19 ± 1,89 Cl,
- mrva: 1,93 ± 0,479 S in 5,72 ± 1,98 Cl,
- koruzna silaža: 0,877 ± 0,247 S in 1,98 ± 0,86 Cl.

Rezultati kažejo veliko variabilnost, čeprav so vzorci vzeti na definiranem področju, krma pa je pridelana v podobnih talnih in klimatskih razmerah. Priporočamo še več pozornosti pri pripravi vzorcev za analizo ter določitev pepela, netopnega v klorvodikovi kislini.

REFERENCES

Charalambous, G. Analysis of Foods and Beverages: Modern Techniques. St.Louis, Missouri, Academic Press, Inc., 1984, 398 p.

Guide to the expression of uncertainty in measurement. Geneva, International Organization for Standardization, 1993, 101 p.

Jenkins, R./ Gould, R.W./ Gedcke, D. Quantitative X- ray Spectrometry. New York, Marcel Dekker, 1981, 586 p.

Kellner, O./ Becker, M. Grundzüge der Fütterungslehre. Hamburg, Berlin, Verlag Paul Parey, 1971, 333-348.

Kump, P./ Nečemer, M./ Smodiš, B./ Jačimovič, R. Multielement analysis of rubber samples by X- ray fluorescence. Appl. Spectrosc., 50(1996), 1373-1377.

McDonald, P./ Edwards, R.A./ Greenhalgh, J.F.D./ Morgan, C.A. Animal nutrition. New York, Longman Scientific and Technical, 1995, 97-127.

McDowell, L.R. Nutrition of grazing ruminants in warm climates. New York, Acad. Press, 1985, 329 p.

McDowell, L.R. Minerals in animal and human nutrition. New York, Academic Press, Inc., 1992, 523 p.

Naylor, M.J./ Ralston, S.L. Large animal clinical nutrition. St. Louis, Mosby Year Book, Inc. 1991, 576 p.

NRC. Nutrient requirements of dairy cattle. Washington, National Academy Press, 1988, 157 p.

Underwood, E.J. The mineral nutrition of livestock. 2nd Ed. Wallingford, CAB International, 1981, 177 p.