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Method of Sampling Beef Cattle Hair for Assessment of Elemental Profile

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Abstract: The development of research methods of sampling of beef cattle hair are given in the article. Data on filthiness, growth and elemental composition of different components of hair were taken into account. Hair from mature beef cattle (Bos Taurus) was used. Animals were of Hereford and Angus breeds. Hair sampling of one and the same animals was performed in winter and summer period. Hair for study was cut from six areas: back of head, the first tail vertebra, projection of the 12th rib median line, dewlap, withers, switch. Elemental composition of hair and its components was tested by 25 chemical elements. It was observed that guard hair from withers is the fastest growing (0.38±0.033 mm/d). Hair from withers (8.14±1.02% in winter period and 4.80±0.83 in summer period) and dewlap were with less dirt. No correlation between area of samples and its elemental composition was observed. Down hair contained more cobalt, manganese and nickel compared to guard hair of the animals. It was found that average sample of hair from withers is more preferable for research of elemental composition. It is recommended to use the developed method in science and agricultural industry for assessment of elemental composition of beef cattle hair.

Key words: Bos Taurus (beef cattle), hair, elemental profile

INTRODUCTION

Diagnostics, prevention and treatment of elementosis may be conducted taking into account data of the hair elemental composition. Medicine accumulated experience proves it.

Over the last years the method of diagnostics and treatment of human elementosis based on the analysis of hair elemental composition (method of Dr. Skalny) was used for more than 220 thousand patient's treatment and prevention of diseases (www. microelements.ru). The base of this method is a technic considering individual variations of elemental composition of human hair from the physiological range of elemental composition (Skalnaya *et al*., 2003).

Lately, hair has become a fundamental biological specimen, alternative to the usual blood and urine samples, as well as biopsy material, in clinical toxicology and chemistry (Rebacz-Maron *et al*., 2013; Pasha *et al*., 2010). Elemental analysis is used for diagnostics of cancer diseases (Czerny *et al*., 2014); pathological diseases caused by heavy metal intoxication (Grabeklis *et al*., 2011); metabolic syndromes (Park *et al*., 2006; Farkhutdinova *et al*., 2009); thyroid diseases (Momcilovic *et al*., 2012).

At the moment there are no analogical noninvasive methods in the animal breeding. There are no databases of multielement composition of animal hair and other animal bio substrates. There is a lack of information about elemental composition of different hair parts (guard hair, down hair). Different structural elements of hair may have different multielement composition (Cygan-Szczegielniak *et al*., 2014).

The noninvasive research method of animal elemental profile may be wide applicable in dairy cattle breeding for individual assessment and metabolism correction of high-producing animals. This may increase the longevity and livestock efficiency indexes and improve reproductive capacity.

The objective of this research was to develop the hair sampling technic for sampling of beef cattle hair taking into account hair filthiness, elemental composition of different parts of hair etc.

MATERIALS AND METHODS

Animals: Hair from mature Bos Taurus (beef cattle) of Hereford and Angus breeds was researched. The animals were born and stayed in Orenburg region (Russia). The total number of beef cattle used in the research was 120 animals. The live weight was 450-600 kg.

Sampling: Hair sampling of one and the same animals was performed in winter and summer period. Samples of hair were taken in winter and summer period from the same animals. Hair was cut from six areas: back of head, the first tail vertebra, projection of the 12th rib median line, dewlap, withers, switch. Hair sampling was made from 4-5 places of the area determined for

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sampling. Hair was cut at 1 cm from the root and in length no more than 5 cm. Samples were divided into guard hair, down hair and awn hair.

Hair analysis: Hair filthiness was measured by weighing of hair samples before and after the cleaning procedure. Cleaning included the following: steeping for three hours in the distilled water (t = 40-60°C); washing for two hours in each conditions - in the ethyl alcohol solution (40%) and in the distilled water together with ultrasonic treatment (frequency of 35 kHz, power-300 (450) W, vibration amplitude -10 mm) (Kovalenok, 2012). Then samples were dried to constant mass at the temperature of 65°C.

Speed of hair growth was measured on certain places of animal's body surface. Areas 2.5 x 2.5 cm of body surface were shaved. The weight and length of the grown back hair was measured each 60 days.

Elemental composition of hair and its components was tested by 25 elements (Al, As, Be, Cd, Hg, Li, Ni, Pb, Sn, Ti, V, I, Ca, Co, Cr, Cu, Fe, K, Mg, Mn, Na, P, Se, Si, Zn). The atomic emission and mass spectrometry methods were applied using the following equipment: Elan 9000 (Perkin Elmer, USA) and Optima 2000 V (Perkin Elmer, USA). Samples were tested in the laboratory of ANO the Center for Biotic Medicine (Registration Certificate of ISO 9001: 2000, Number 4017-5.04.06) with consideration of recommendations (Skalny *et al*., 2009).

Statistical analysis: Statistical analysis was performed by means of statistical software package Statistica 10 Ru. Statistical comparison of the results was made using the Mann-Whitney U and Student's tests. The parameter $p<0.05$ was the significance level. Elemental composition of hair samples taken from six separate areas was compared with the average value of elemental composition of samples taken from six body surface areas.

RESULTS AND DISCUSSION

Hair filthiness: Assessment of hair taken from different areas of animal body surface showed that hair from withers and dewlap are the less dirty (Table 1).

Content of dirt in hair from withers and dewlap was more than that in hair from the back of head in winter period by 39.3% ($p\leq0.05$) and 25.9% ($p\leq0.05$), in summer period by 64% ($p\leq0.05$) and 45.2% ($p\leq0.05$).

Most of contaminations were found in hair from switch 843.2 mg/g in winter period and 287.8 mg/g in summer period. Weight of dirt in hair from switch in winter period was 10.4 times more than in hair form withers ($p\leq0.001$), in summer period -6.0 times more ($p\leq0.001$).

Hair growth rate: It was found that guard hair on withers has the highest growth rate 0.375±0.033 mm/d. It was by 79 % ($p<0.001$) more than these figures for dewlap and by 29% ($p<0.001$) for guard hair from first tail vertebra projection area. Ground hair on the surface of first tail vertebra grows faster (0.22±0.02 mm/d; $p<0.01$) (Table 2).

Table 1: Hair filthiness, mg/g

	Period	
Parameter	winter	Summer
Back of head	113.4±9.2*	78.7±6.8*
Tail head	121.4±12.1*	87.8±8.2*
Last rib median line	141.7±8.3***	67.1±6.4
Dewlap	90.1±7.3	54.2±7.1
Withers	81.4±10.2	48.0±8.3
Switch	843.2±86.5***	287.8±32.1***

*$p\leq0.05$; ***$p\leq0.001$ compared with withers

Table 2: Hair growth rate on different body areas of animals, mm/d

	Type of hair	
Parameter	Down hair	Guard hair
Back of head	-	0.31±0.03
Tail head	0.22±0.02	0.29±0.02*
Last rib median line	0.19±0.03	0.24±0.01***
Dewlap	0.16±0.02*	0.21±0.02***
Withers	-	0.37±0.03
Switch	-	0.33±0.04

*$p\leq0.05$; ***$p\leq0.001$

Hair elemental composition: Samples of hair taken from different body surface areas had different elemental composition that varies from the average value of chemical elemental concentration in the hair from six places of sampling (Table 3). Composition of hair samples from withers was the nearest to the average elemental concentration. The only exemption was nickel which concentration was 10.0 % ($p\leq0.01$) less than the average.

Elemental composition of guard and down hair: Researches showed difference between elemental composition of guard and down hair (Table 4). Concentration of cobalt in down hair was by 49.5% ($p\leq0.001$), of manganese by 56.8% ($p\leq0.001$), of nickel by 39.0% ($p\leq0.01$) more.

Medicine experience of assessment and correction of human elemental profile through the research of hair elemental composition let us consider this method for use in cattle breeding. Researches of elemental composition of animal hair appeared to be promising for diagnostics, prevention and treatment of high-producing cow's diseases (Zhao *et al*., 2015).

Therefore the objective was to develop method of sampling of beef cattle hair. The research was necessary due to the absence of information about elemental composition and growth rate of hair and its components on different body areas.

Difference in the elemental composition of hair from different body surface areas of animals was revealed by

Table 3: Concentration of chemical elements in hair samples of beef cattle, microgram/g

	Parameter						
Element	Back of head	Tail head	Last rib median line	Dewlap	Withers	Switch	Average
K	4663±176.0**	5137±202.7*	4878±95.9*	4189±157.7**	4644±88.5	4319±170.8	4638±47.3
Ca	2212±48.5**	1917±53.1**	2482±59.0***	1758±32.5***	2099±28.2	1927±39.3**	2065±17.7
Mg	865±34.8	896±39.5**	975±23.1***	367±16.4***	802±19.5	836±20.1**	770±12.6
Na	1218±58.2	1140±92.3	1023±86.0	1407±67.0	1174±57.5	1017±89.1	1163±24.2
P	327±7.8	311±11.6	337±7.9	251±4.2***	304±4.2	275±6.5***	301±2.9
Zn	103±0.8	106±1.0***	102±0.8	99±1.3	102±0.6	99±0.6***	102±0.3
Fe	69.2±2.9**	54.3±1.1	67.7±2.4***	48.8±1.8***	58.1±1.9	49.7±1.9	57.9±1.2
Cu	7.2±0.3	6.55±0.18**	5.96±0.12***	8.17±0.13***	7.52±0.12	7.96±0.25**	7.23±0.11
Mn	14.4±1.4	16.2±0.9	23.0±1.4***	14.8±0.8	17.7±0.8	15.9±1.3	17.0±0.8
I	0.50±0.03	0.58±0.028*	0.63±0.021***	0.39±0.017***	0.52±0.016	0.47±0.019*	0.52±0.01
Se	0.46±0.014	0.53±0.017***	0.55±0.045*	0.28±0.009***	0.45±0.008	0.42±0.012*	0.45±0.01
Cr	0.20±0.010*	0.19±0.012	0.17±0.006	0.15±0.007	0.17±0.009	0.13±0.006***	0.17±0.01
Co	0.063±0.003*	0.05±0.003	0.07±0.003***	0.037±0.001***	0.053±0.002	0.054±0.002	0.055±0.001
Si	34.4±1.22	35.1±1.15*	27.3±0.97	26.8±1.67	31.5±1.02	32.9±1.73	31.4±1.08
B	1.64±0.063	1.65±0.069	1.85±0.050***	0.92±0.024***	1.62±0.038	1.61±0.059	1.55±0.02
Li	1.46±0.055	1.21±0.037**	1.29±0.061	1.17±0.041***	1.34±0.039	1.55±0.048***	1.34±0.03
Ni	0.78±0.054***	0.56±0.020	0.63±0.010*	0.34±0.010***	0.54±0.015**	0.72±0.064	0.60±0.01
V	0.26±0.014*	0.22±0.012	0.33±0.014***	0.13±0.005***	0.24±0.006	0.22±0.007	0.23±0.01
As	0.18±0.014	0.14±0.006***	0.13±0.005***	0.21±0.013	0.19±0.006	0.25±0.019***	0.18±0.01
Al	45.5±1.94**	37.0±0.72*	45.7±1.92***	31.5±1.32***	41.7±0.94	36.7±1.84	39.7±0.83
Sr	12.7±0.58	12.7±0.61	15.3±0.50***	9.5±0.14***	11.7±0.24	10.4±0.32***	12.0±0.17
Pb	0.22±0.02	0.17±0.006***	0.16±0.004***	0.13±0.004***	0.18±0.004	0.33±0.054*	0.19±0.004
Sn	0.047±0.0039	0.054±0.0060**	0.027±0.0016***	0.028±0.0021**	0.036±0.0019	0.032±0.0033	0.037±0.001
Cd	0.0096±0.0006	0.012±0.0006***	0.013±0.0006***	0.0067±0.0004***	0.0094±0.0005	0.0058±0.0005***	0.0094±0.003
Hg	0.0068±0.0003	0.0065±0.0003	0.0064±0.0004	0.0062±0.0002	0.0066±0.0003	0.0070±0.0004	0.0066±0.0002

p≤0.05; **p≤0.01, ***p≤0.001

Table 4: Elemental composition of animal guard and down hair, microgram/g

Element	Average guard hair	Average down hair	Values of t-distribution	p	Variation guard hair	Variation down hair
Al	30.3	35.7	-0.78	0.435	24.6	28.0
As	0.07	0.06	0.22	0.825	0.03	0.0
B	6.1	5.7	0.27	0.785	4.5	4.1
Ca	2515	2878.5	-1.60	0.113	901.4	688.1
Cd	0.03	0.03	-0.26	0.796	0.02	0.01
Co	0.09	0.14	-2.15	0.034	0.05	0.1
Cr	0.11	0.12	-0.83	0.407	0.07	0.08
Cu	4.9	4.4	1.66	0.099	1.2	0.9
Fe	32.4	39.2	-1.18	0.240	20.5	24.4
Hg	0.01	0.008	0.45	0.650	0.02	0.007
I	0.6	0.8	-1.52	0.132	0.4	0.4
K	606.5	594.0	0.17	0.863	272.4	272.4
Li	0.5	0.5	-0.08	0.932	0.4	0.3
Mg	561.0	629.6	-0.97	0.332	277.4	218.9
Mn	24.1	37.8	-3.41	0.001	12.5	21.02
Na	390.2	388.4	0.04	0.961	153.4	123.3
Ni	0.6	0.8	-2.64	0.010	0.3	0.4
P	185.8	167.6	1.04	0.299	69.3	51.4
Pb	0.2	0.2	-0.45	0.648	0.07	0.07
Se	0.6	0.5	0.30	0.763	0.3	0.2
Si	25.2	19.4	0.95	0.343	24.4	18.0
Sn	0.03	0.03	-0.33	0.742	0.02	0.02
Sr	16.6	20.5	-1.91	0.060	8.02	6.7
V	0.2	0.2	-1.02	0.307	0.2	0.2
Zn	93.7	91.0	0.58	0.560	17.7	16.6

our study. Hair elemental composition is closely related to the blood elemental composition and reflects characteristics of chemical elements metabolism in the organism during certain life periods (Christodoulopoulos *et al*., 2003; Smith *et al*., 2010; Cygan-Szczegielniak *et al*., 2014). Thus hair composition variance might be conditioned by the different growth rate on certain body areas. It was found that growth rate variance between sampling areas was up to 79%. So the changes of separate chemical elements metabolism did not equally affect hair composition along the hair.

Hair elemental composition is determined by the elemental composition of guard and down hair. Concentration differences of cobalt, manganese and nickel in guard and down hair were revealed by our study. Therefore withers is more preferable for sampling of average hair. Hair from withers includes minimal

Table 5: Value of Student's t-distribution calculated by concentration of chemical elements in the composition of hair from withers and tail head of beef cattle

Value of Student's t-distribution by group of elements							
Macro elements		Vital essential micro elements		Conditionally essential elements		Toxical microelements	
K	0.222	Zn	0.0124	Si	0.2211	Al	0.0015
Ca	0.012	Fe	0.0003	B	0.4722	Sr	0.0008
Mg	0.014	Cu	0.0001	Li	0.0571	Pb	0.2315
Na	0.285	Mn	0.0224	Ni	0.2852	Sn	0.2010
P	0.214	I	0.0034	V	0.0001	Cd	0.1333
		Se	0.3420	As	0.1573	Hg	0.1859
		Cr	0.0067				
		Co	0.0101				

k = 50, α = 0.05, t-table = 2.009. Where k-degree of freedom for two sets of sampling data; α = 0.05-significance level; t-table-table value of Student's t-distribution determined based on the degree of freedom and set significance level

quantity of down hair that makes the research of elemental composition more objective. This body area has the maximum hair growth rate (0.38±0.033 mm/d) so it allows fairly assess animal's elemental profile.

However the elemental composition of beef cattle hair on different body areas is formed by the same principles. This conclusion is confirmed by the hypothesis that average values of two sampling (elemental composition from two areas) belong to the same population.

Absolute values of Student's t-distribution were determined to assess deviation of average values of hair elemental composition of beef cattle. Student's t-distribution values were calculated based on the concentration of chemical elements in the composition of hair from withers and tail head of beef cattle with degree of freedom k = 50 and significance level α = 0.05. Comparison of value of Student's t-distribution calculated for each element (Table 5) with the table value (t-table = 2.009) showed that average values of two sets of sampling data refers to the same population, e.g. both sets of sampling data (data on elements concentration in hair samples from withers and tail head) belongs to the same population and have no significant deviations.

Similar calculations of values of Student's t-distribution were made for the rest sampling areas (samples from withers and back of head; withers and last rib median line; withers and brisket; withers and switch). It was found that average values of tested samples had no significant differences by each element. Thus the information of concentration of certain chemical element in hair may be used regardless of the sampling area.

Based on the above the selection of average hair samples from withers is more preferable for the elemental composition test. All that is proved by the following:

1: Samples form withers contained less dirt: 8.14±1.02% in winter period and 4.80±0.83 in summer period. Filthiness of other four areas was definitely greater with the maximum values for switch 84.3±8.65% in housing season and 28.8±3.21% in grazing season
2: Guard hair on withers has the highest growth rate 0.38±0.033 mm/d. It is by 79% ($p<0.001$) more than these figures for dewlap and by 29% ($p<0.001$) more for guard hair from first tail vertebra projection area
3: The average elemental composition of hair from animal's body surface corresponds to the samples from withers by 24 of 25 chemical elements

Moreover when the animal is fixed in a cattle chute the withers area is convenient for hair sampling.

Conclusion: Taking into account hair filthiness, elemental composition of different components of hair ant etc. hair sampling of beef cattle shall be conducted from withers. Hair growth rate approximately 0.38±0.033 mm/d may be considered for taking hair samples formed in the certain life period of the animal. Considering medicine experience accumulation of objective data on hair elemental composition of the animal in connection with its health and productivity will allow developing new method of diagnostics, preventive measures and treatment of beef cattle elementosis.

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REFERENCES

Christodoulopoulos, G., N. Roubies, H. Karatzias and A. Papasteriadis, 2003. Selenium concentration in blood and hair of holstein dairy cows. Biol. Trace Elem. Res., 91: 145-150.

Cygan-Szczegielniak, D., M. Stanek, E. Giernatowska and B. Janicki, 2014. Impact of breeding region and season on the content of some trace elements and heavy metals in the hair of cows. Folia Biol. (Krakow), 62: 163-169.

Czerny, B., K. Krupka, M. Ozarowski and A. Seremak-Mrozikiewicz, 2014. Screening of trace elements in hair of the female population with different types of cancers in Wielkopolska region of Poland. Sci. World J., 2014:953181. doi: 10.1155/2014/953181.

Farkhutdinova, L.M., V.V. Speranskii and A.Zh. Gil'manov, 2009. Hair tissue mineral analysis and metabolic syndrome. Biol. Trace Elem. Res., 130: 218-228.

Grabeklis, A.R., A.V. Skalny, S.P. Nechiporenko and E.V. Lakarova, 2011. Indicator ability of biosubstances in monitoring the moderate occupational exposure to toxic metals. J. Trace Elem. Med. Biol., 1: S41-44.

Invention patent of the Russian Federation No. Preparation method of beef cattle hair samples for analysis of macro and microelemental composition /Y.K. Kovalenok: published on 27.05.2012.

Momcilovic, B., J. Prejac, V. Visnjevic, M.G. Skalnaya, N. Mimica, S. Drmic and A.V. Skalny, 2012. Hair iodine for human iodine status assessment. Thyroid, 24: 1018-1026.

Park, S.B., S.W. Choi and A.Y. Nam, 2006. Hair trace elements in patients with goiter Klin Lab Diagn, 19-21.

Pasha, Q., S.A. Malik, N. Shaheen and M.H. Shah, 2010. Comparison of trace elements in the scalp hair of malignant and benign breast lesions versus healthy women. Biolog. Trace Elem. Res., 134: 160-173.

Rebacz-Maron, E., I. Baranowska-Bosiacka, I. Gutowska, N. Krzywania and D. Chlubek, 2013. The content of fluoride, calcium and magnesium in the hair of young men of the bantu language group from tanzania versus social conditioning. Biolog. Trace Element Res., 156: 91-95.

Skalnaya, M.G., V.A. Demidov and A.V. Skalny, 2003. Limits of physiological (normal) concentration of Ca, Mg, Fe, Zn and Cu in human hair. Microelements in medicine. O., 2: 5-10.

Skalny, A.V., V.V. Kuznetsov, E.V. Lakarova and M.G. Skalnaya, 2009. Analytical methods in bioelementology. St-Petersburg, Sci., 576.

Smith, K.M., M.P. Dagleish and P.W. Abrahams, 2010. The intake of lead and associated metals by sheep grazing mining-contaminated floodplain pastures in mid-Wales, UK: II. Metal concentrations in blood and wool. Sci. Total Environ., 408: 1035-1042.

Zhao, X.J., X.Y. Wang, J.H. Wang, Z.Y. Wang, L. Wang and Z.H. Wang, 2015. Oxidative stress and imbalance of mineral metabolism contribute to lameness in dairy cows. Biol. Trace Elem. Res., 164: 43-49.