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CLINICAL AND BIOCHEMICAL ASPECTS OF DIAGNOSTICS OF OSTEODYSTROPHY OF GOATS

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Summary. The purpose of this study was to carry out the comparative analysis of data of the clinical examination of goats and biochemical testing of bone tissue metabolism of the metabolites level in blood serum and urine for determination of criteria at diagnostics of osteodystrophy. The study was conducted on 20 adult goats. Diagnosis for osteodystrophy was made according to physical examination of goats and analysis of biochemical tests of bone tissue metabolism using blood serum and urine. Activity of aminotransferase (alanine aminotransferase, aspartate aminotransferase) and alkaline phosphatase and also concentration of glycoproteins (GP), chondroitin sulphate and fractional content of glucosaminoglicans content of total and ionized calcium were determined in blood serum. The content of hydroxyproline and total uronic acid were defined in urine. Comparative analysis of total clinical status of goats and level of biochemical components in blood serum and urine that characterize the connective tissue status at diagnostics of osteodystrophy was conducted. The intergroup significant differences of some biochemical parameters (aspartate aminotransferase, alkaline phisphatase, chondroitin sulphate in blood serum and fraction II mucopolysaccharides, sum of fractions glucosaminoglicans in urine) were observed. Further testing must be conducted to determine the biochemical parameters in goat with subclinical signs of osteodystrophy.

Keywords: osteodystrophy, biochemical parameters, goats, blood serum, urine

Introduction. The problem of diagnostics of different stages of osteodystrophy is the relevant issue of modern veterinary science. One of the main directions of research in this field is working out and implementation of non-complicated methods of diagnostics, for including to laboratory practice (Kondrakhin, 1989).

Conducting of biochemical analysis of blood serum is limited by facilities of regional veterinary laboratories in the farms, taking into account that for diagnostics of osteodystrophy they mainly determine only content of total calcium and inorganic phosphorus in the blood serum. These indexes characterize in a certain measure the state of mineral homeostasis that changes substantially only in the perspectives of abnormalities metabolism and considerable impairment of structure of the bone tissue. It is not always possible to define such pathology as osteodystrophy, especially at the early stage. Methods for determination of content and resulting metabolites of organic matrix of connective tissue in blood serum and urine which reflect the state of bone tissue are more informative for diagnostics of early stage of osteodystrophy (Liesegang, Risteli and Wanner, 2007; Cruz, Lima and Peleteiro, 2002; Carstanjen et al., 2004). Information about the status of organic content of bone tissue of goats in the normal range and with osteodystrophy is quite limited (Timoshenko and Maslak, 2008).

Analysis of the level of biochemical indicators in blood serum of goats is a relevant problem these days.

The purpose of the present study was to conduct comparative analysis of data that have been obtained after the clinical examination of goats and analysis of bone tissue metabolism markers level in blood serum and urine for determination of criteria at diagnostics of osteodystrophy.

Materials and methods. Twenty 2–3-year-old goats were examined in this. They were bred in 'Study and Scientific Center of Plant Growing and Animal Husbandry of Kharkiv State Zooveterinary academy'. Samples of blood serum and urine were collected for biochemical parameter analysis.

Activity of aminotransferases (alanine aminotransferase, aspartate aminotransferase) and alkaline phosphatase (Kamyshnikov, 2000) and also concentration of glycoproteins (GP), chondroitin sulphate and fractional content of glucosaminoglicans (Shteynberg, and Dotsenko, 1962; Shtern et al., 1982) were determined in goat blood serum. Content of total and ionized calcium in blood serum were defined using the analyzer of electrolytes (AEK-01). The content of hydroxyproline and total uronic acid in urine were determined using the techniques by Krel' and Furtseva (1968) and Di Ferrante and Rich (1956) respectively.

The results of the research were processed with biometric statistics techniques using Microsoft Excel.

Results and discussion. It was established that the average temperature was 39.2±0.1 °C (Lim 38.5–40.0) and pulse rate was 89.0±6.0 (Lim 70.0–110.0) on the basis of clinical examination. Fourteen animals (70%) had dullness of heart tones. The number of respiratory movements per minute was 15.3±0.9 (Lim 12.0–20.0).

Three of the examined goats (15%) had pain of liver on palpation. So biochemical investigations of blood serum and urine were conducted for objective assessment of clinical data (Table 1).

More detailed information about the state of bone-articular system was collected by the determination of metabolites that characterize the state of components of organic matrix of connective tissue (Table 2).

Table 1 — Biochemical indicators of blood serum of goats with clinical signs of osteodystrophy

Indicator		Clinically healthy	Osteodystrophy
Alanine aminotransferase, U/L	M±m	28.50±1.04	33.80±2.71
	Confidence interval	25.7–31.3	27.8–39.8
Aspartate aminotransferase, U/L	M±m	26.00±1.08	38.90±1.56 ***
	Confidence interval	23.1–28.9	34.8–41.7
Alkaline phosphatase, m/L	M±m	3.78±0.63	8.31±1.09 *
	Confidence interval	2.08–5.48	5.85–10.77
Total calcium, m/L	M±m	2.67±0.08	2.78±0.05
	Confidence interval	2.45–2.89	2.68–2.88
Ionized calcium, m/L	M±m	1.28±0.04	1.32±0.04
	Confidence interval	1.18–1.38	1.24–1.40
Phosphorus, m/L	M±m	1.24±0.02	1.24±0.02
	Confidence interval	1.19–1.29	1.20–1.28
Chondroitin sulphate, g/L	M±m	0.11±0.01	0.24±0.04 *
	Confidence interval	0.09–0.13	0.16–0.32
Glycoprotein, m/L	M±m	0.60±0.01	0.67±0.05
	Confidence interval	0.58–0.62	0.44–0.70

Notes: * — $p<0.05$, *** — $p<0.001$

Table 2 — Biochemical indicators of the state of connective tissue at osteodystrophy of goats (according to the analysis of blood serum)

Indicator		Clinically healthy	Osteodystrophy
Fraction I mucopolysaccharides, U/L	M±m	5.9±0.55	8.1± 0.52
	Confidence interval	4.4–7.4	6.9–9.27
Fraction II mucopolysaccharides, U/L	M±m	2.63±0.08	3.42±0.17 *
	Confidence interval	2.41–2.85	3.04–3.8
Fraction III glucosaminoglicans, U/L	M± m	2.0±0.17	2.6±0.31
	Confidence interval	1.53–2.47	1.92–3.28
Sum of fractions glucosaminoglicans, U/L	M± m	10.2±0.62	14.2±0.84 *
	Confidence interval	8.5–11.9	12.4–16.0

Notes: * — $p<0.05$

Other clinical signs of osteodystrophy were the following: sixteen goats (80%) had thinning and tuberosity of ribs, the incisor teeth of fourteen animals (70%) were loose, four goats had partial lysis of the last pair of ribs. After clinical examination twenty goats were divided into two groups (four animals — clinically healthy, sixteen animals — with clinical signs of osteodystrophy).

In terms of obtained data we can conclude that the affected goats had clinical signs of osteodystrophy. It is known that acquired clinical signs could remain for the whole period of animals' life of the animal (tuberosity of ribs) and clinical signs not always reflect the state of bone tissue at the moment of examination.

Indicators of the mineral metabolism (total and ionized calcium, inorganic phosphorus) of affected goats were almost the same with another group of control that confirms low level of informational content. At the same time affected goats demonstrated enlarged level of chondroitin sulphate (by 54%) and increased activity of alkaline phosphatase that can designate the pathology of support-locomotion system. 50% of animals had increased activity of alanine amino transferase in blood serum that made up 41.2±1.9 U/L. Increased activity of aspartate aminotransferase of goats with clinical signs of osteodystrophy 38.90±1.08 U/L on comparison with 26.00±1.08 U/L in the normal range might be the result of abnormity of myocardium and liver as it is known that functions of these organs decay at the postprimary osteodystrophy. In particular, it might be abnormality of cardiac muscle that coincides with such clinical sign as dullness of heart tones.

Analysis of Table 2 shows that goats with osteodystrophy had larger concentration of fraction II glucosaminoglicans, (by 23%) that led to increase of total glucosaminoglicans, in blood serum. Increase of level of fraction II that contains mostly chondroitin-4-sulphate is evidence of pathology of bone system in particular, as fraction chondroitin-4-sulphate dominates quantitatively in bones (Borovkov, 2006). If indicators that were analyzed before cannot be considered as specific only for diagnostics of osteodystrophy of animals, determination of excretion of hydroxyproline with urine is undoubtedly a marker of bone resorption. It reflects its metabolism that with definition of level of excretion of glucosaminoglicans, and calcium with urine can give detailed information about direction of metabolism of bone tissue (Liesegang, Risteli and Wanner, 2007).

In terms of Table 3 it can be concluded that animals with osteodystrophy had larger excretion of hydroxyproline (by 33%), uronic acid (by 39%) and calcium (by 78%). From our point of view it is caused by different correlation of osteosynthesis and resorption of bone tissue during examination.

Table 3 — Biochemical indicators of connective tissue at osteodystrophy of goats (according to the analysis of urine)

Indicator		Clinically healthy	Osteodystrophy
Hydroxyproline, mg/l	M±m	50.6±4.04	75.1±4.82 *
	Confidence interval	39.5–61.7	64.1–86.1
Uronic acid, mg/l	M±m	3.17±0.34	8.08±1.35 *
	Confidence interval	2.09–4.25	5.09–11.07
Calcium, mg/l	M±m	92.5±12.4	420.6±60.43 **
	Confidence interval	59.0–126.0	284.1–557.1
Phosphorus, g/l	M±m	0.17±0.04	0.13±0.02
	Confidence interval	0.06–0.28	0.09–0.17

Notes: * — $p<0.05$, ** — $p<0.01$

Conclusions. On the basis of clinical examination of goats and taking into account data from biochemical analysis of blood serum and urine it was established that level of clinical appearance of osteodystrophy in goats coincide with changes in some of biochemical indicators in blood serum and urine.

Among biochemical components of blood serum the most informative for diagnostics of osteodystrophy is determination of content of chondroitin sulphate, fraction II glucosaminoglicans, and activity of alkaline phosphatase, and in urine — level of excretion of hydroxyproline, uronic acid and calcium.

References

Borovkov, S. B. (2006) *Clinical-biochemical indexes of the state of connecting tissue in diagnostics and treatment of osteodystrophy in cows [Kliniko-biokhimichni pokaznyky stanu spoluchnoi tkanyny v diahnostytsi ta likuvanni osteodystrofii koriv]*. The dissertation thesis for the scientific degree of the candidate of veterinary sciences. Bila Tserkva: Bila Tserkva State Agrarian University. Available at: http://www.irbis-nbuv.gov.ua/cgi-bin/irbis_nbuv/cgiirbis_64.exe?C21COM=2&I21DBN=ARD&P21DBN=ARD&Z21ID=&IMAGE_FILE_DOWNLOAD=1&Image_file_name=DOC/2006/06bsblok.zip. [in Ukrainian].

Carstanjen B, Hoyle, N. R., Gabriel, A., Hars, O., Sandersen, C., Amory, H. and Remy, B. (2004) 'Evaluation of plasma carboxy-terminal cross-linking telopeptide of type I collagen concentration in horses', *American Journal of Veterinary Research*, 65(1), pp. 104–109. doi: 10.2460/ajvr.2004.65.104.

Cruz, L. A., Lima, M. S. and Peleteiro, M. C. (2002) 'Osteodystrophia fibrosa in milking goats: report of a clinical case' [Osteodistrofia fibrosa em cabras de leite: descrição de um caso clínico], *Revista Portuguesa de Ciências Veterinárias*, 97(543), pp. 147–150. Available at: http://www.fmv.ulisboa.pt/spcv/PDF/pdf9_2002/08_rpcv543.pdf. [in Portuguese].

Di Ferrante, N. and Rich, C. (1956) 'The determination of acid aminopolysaccharides in urine', *The Journal of Laboratory and Clinical Medicine*, 48(3), pp. 491–494. PMID: 13367587.

Kamyshnikov, V. S. (2000) *Handbook of clinical and biochemical laboratory diagnostics. Volume 2 [Spravochnik po kliniko-biokhimicheskoy laboratornoy diagnostike. Tom 2]*. Minsk: Belarus'. ISBN: 9850104449.

Kondrakhin, I. P. (1989) *Alimentary and endocrine diseases of animals [Alimentarnye i endokrinnye bolezni zhivotnykh]*. Moscow: Agropromizdat. ISBN: 5100006471.

Krel', A. A. and Furtseva, L. N. (1968) 'Methods of determining hydroxyproline in biological fluids and their use in clinical practice' [Metody opredeleniia oksprolina v biologicheskikh zhidkostiakh i ikh primenenie v klinicheskoĭ praktike], *Problems of Medical Chemistry [Voprosy meditsinskoy khimii]*, 14(6), pp. 635–640. Available at: http://pbmc.ibmc.msk.ru/index.php/ru/article/PBMC-1968-14-6-635-ru. [in Russian].

Liesegang, A., Risteli, J. and Wanner, M. (2007) 'Bone metabolism of milk goats and sheep during second pregnancy and lactation in comparison to first lactation', *Journal of Animal Physiology and Animal Nutrition*, 91(5–6), pp. 217–225. doi:10.1111/j.1439-0396.2007.00695.x.

Shtern, M. R., Timoshenko, O. P., Leont'eva, F. S. and Klyueva, G. F. (1982) *Method for determination of hexosaminoglycansulphates in blood serum [Sposob opredeleniya geksozaminoglikansul'fatov v syvorotke krovi]*. Author's Certificate SU 960626. Available at: http://patents.su/3-960626-sposob-opredeleniya-glikozaminglikansulfatov-v-syvorotke-krovi.html.

Shteynberg, S. Ya. and Dotsenko, Ya. N. (1962) 'New method for determination of glycoproteins in serum and blood plasma' [Novyy metod opredeleniya glikoproteidov v syvorotke i plazme krovi], *Medical Practice [Vrachebnoe delo]*, 12, pp. 43–45.

Timoshenko, O. P. and Maslak, Yu. V. (2008) 'Biochemical indicators in blood serum in healthy goats' [Biokhimichni pokaznyky u syrovattsi krovi zdorovykh kiz], *Collection of Scientific Works of the Luhansk National Agrarian University. Series 'Veterinary Sciences' [Zbirnyk naukovykh prats Luhanskoho natsionalnoho ahrarnoho universytetu]*, 92, pp. 223–226.