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## POLYMORPHISM OF THE CSN2 GENE (VERSIONS A1/A2) IN DIFFERENT BREEDS OF DAIRY CATTLE IN THE CENTRAL BLACK SOIL REGION OF RUSSIA

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### ABSTRACT

USING the method PCR-RFLP, they performed genotyping of cattle for the CSN2 gene (A1/A2 polymorphism). The work studied the cows of six main dairy breeds: Holstein, Simmental, Ayrshire, Brown Swiss, Jersey, Red-and-White from nine different farms of the Central Black Earth Region of Russia. The gene for beta-casein (CSN2) is located in the chromosome 6 and may be an important marker of milk yield and milk quality. Purpose of the study: to assess the frequencies of the A2 variant and the A2A2 genotype in various breeds of dairy cattle at the breeding farms of the Central Chernozem region of Russia. Materials and Methods: Genomic DNA was isolated from whole blood. 106 b.p DNA fragment was amplified using the primers from the CSN2 gene containing the target mutation. The PCR product was hydrolyzed with the restriction enzyme BstDEI. Gel blocks were stained with ethidium bromide and visualized on UV transilluminator to detect DNA fragments. The samples with the genotype A1A1 had the DNA fragments of 106 b.p., A1A2 - the fragments of 106, 74, 32 b.p., and A2A2 - 74, 32 b.p. Result: The highest frequency of the variant A2 was found in the livestock of the Jersey (0.94) breed, and the lowest in the Simmental breed (0.38). At the same time, there is a significant scatter of A2 frequencies in different cattle of the same breed: in the Holstein breed - from 0.53 to 0.70, in the Ayrshire breed - from 0.48 to 0.91. In general, the frequencies of the A2 CSN2 variant in most of the studied breeds take values higher than the average as compared with the data on the same breeds from other regions and countries. In some herds, they revealed a significant deviation of genotype frequencies from the Hardy-Weinberg equilibrium, which indicates the breeding work related to milk quality.

**Key words:** Cattle, CSN2, polymorphism, PCR-ACRS.

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## BsmI AND HinfI POLYMORPHISMS OF THE FABP4 GENE OF DIFFERENT PIG BREEDS OF THE BELGOROD REGION IN RUSSIA

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### ABSTRACT

PIGS were genotyped for FABP3 BsmI (G5005A) and HinfI (C406T) gene. The study examined 188 boars of four breeds (Duroc, Large White, Landrace and Yorkshire) from the Belgorod region of Russia. Fatty acid binding proteins (FABP) are involved in the regulation of metabolism, bind and transport fatty acids, and play an important role in the regulation of fat accumulation and distribution. Purpose of the study: to assess the frequencies of alleles and genotypes for BsmI and HinfI polymorphisms in FABP4 gene among the Duroc, Large White, Landrace and Yorkshire breeds at the farms of the Belgorod region of Russia. Materials and methods: Genomic DNA was isolated from alcoholized earmarks. Pigs were genotyped by PCR-RFLP method. The PCR product in the amount of 10 µl was hydrolyzed with 4 units of the corresponding restriction enzyme BsmI or HinfI (SibEnzyme, Russia) for 16 hours. The hydrolysis products were separated by horizontal electrophoresis in 2% agarose gel (Mini-Sub Cell GT, BioRad, USA). Gel blocks were stained with ethidium bromide (0.5 µg/ml) and visualized on a UV transilluminator to detect DNA fragments. Result: For C406T mutation, the presence of the T mutant allele was noted only for the Duroc breed (0.467), and the C allele was fixed for the other breeds. According to the G5005A mutation, the highest frequency of the A allele associated with the content of intramuscular fat increase was noted among Duroc pigs (0.440), and the smallest - in the Yorkshire breed (0.213). According to the  $\chi^2$  test, all studied breeds, except for monomorphic groups, are in the state of genetic equilibrium according to Hardy-Weinberg. Conclusion: Our data on the variability of these markers generally corresponds to previously published materials for the pigs of Chinese, Canadian and Korean selection. At the same time, the increase of the A BsmI allele frequencies is associated with a high content of IMF for the pigs of the Belgorod region, which may be associated with the result of breeding work to improve the meat quality.

**Key words:** pigs, FABP3 gene, BsmI (G5005A) and HinfI (C406T) polymorphisms.

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## HINFI POLYMORPHISM OF THE H-FABP GENE OF DIFFERENT BREEDS OF PIGS IN THE BELGOROD REGION OF RUSSIA

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### ABSTRACT

USING the method PCR-RFLP, the pigs were genotyped for the H-FABP gene (T314C mutation). The study examined 188 boars of four breeds (Large White, Landrace, Duroc, and Yorkshire) from the Belgorod region of Russia. H-FABP3 plays a key role in fatty acid metabolism and fat deposition and is considered a candidate gene for the assessment of pig fatness and intramuscular fat (IMF) levels. Purpose of the study: Evaluation of alleles and genotype occurrence frequency for the HinfI polymorphism in the H-FABP gene among the Large White, Duroc, Landrace and Yorkshire breeds at the farms of the Belgorod region of Russia. Materials and methods: Genomic DNA was isolated from alcoholized earmarks using the DNA-Extran-2 reagent kit (SINTOL, Russia) according to the protocol. Pig genotyping for the T103C mutation was performed by PCR-RFLP method. The PCR product in the amount of 10 µl was hydrolyzed with 4 units of the HinfI restriction enzyme (SibEnzyme, Russia) for 16 hours. The hydrolysis products were separated by horizontal electrophoresis in 2% agarose gel (Mini-Sub Cell GT, BioRad, USA). Gel blocks were stained with ethidium bromide (0.5 µg/ml) and visualized by UV transilluminator to detect DNA fragments. Result: The highest frequency of the economically valuable allele H and the genotype HH was noted for the Yorkshire breed (0.968 and 0.94, respectively), and the smallest frequencies - for the Large White breed (0.628 and 0.36, respectively). Based on the  $\chi^2$  test, all examined breeds are in a status of genetic equilibrium based on Hardy-Weinberg. They revealed the dependence of H-FABP alleles and genotype frequencies on the T3469C polymorphism of the leptin gene (LEP). In particular, they revealed the tendency to C allele frequency increase for T3469C (LEP) with H allele frequency decrease of the H-FABP gene ( $r = -0.81 \pm 0.41$ ). Besides, a positive correlation was found between the frequencies of homozygous genotypes TT (LEP) and HH (H-FABP) ( $r = 0.87 \pm 0.35$ ), as well as heterozygous genotypes CT (LEP) and Hh (H-FABP) ( $r = 0.93 \pm 0.26$ ). Conclusion: The highest frequency of the economically valuable H allele and the HH genotype were noted for the Yorkshire breed, and the lowest - for the Large White breed. They determined the originality of the gene pool of the main breeds of pigs in the Belgorod region for the polymorphism H-FABP T314C, caused by the peculiarities of local selection and crossing systems thorough the producing meat products. They revealed the dependence of alleles and genotype frequencies in the FABP and LEP genes.

**Keywords:** pigs, H-FABP gene, T314C mutation

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## T3266G AND A1112G POLYMORPHISMS IN THE LEPTIN GENE OF VARIOUS PIG BREEDS OF THE BELGOROD REGION OF RUSSIA

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### ABSTRACT

USING the PCR-RFLP method, pigs were genotyped for the leptin gene (the mutations T3266G and A1112G). The boars of four breeds (Duroc, Large White, Landrace and Yorkshire) from the Belgorod region of Russia were studied in the work. The LEP gene of pigs is located in the 18-th chromosome and serves as a promising candidate gene for pig growth and obesity evaluation. Purpose of the study: Evaluation of allele frequencies for the polymorphisms A1112G and T3266G among the boars of the Duroc, Large White, Landrace and Yorkshire breeds at the farms of the Belgorod region (Russia). Materials and methods: Genomic DNA was isolated from alcoholized earmarks using the DNA-Extran-2 reagent kit (SINTOL, Russia) according to the protocol. Pigs were genotyped by PCR-RFLP. PCR products were hydrolyzed with 4 units of restriction enzyme FokI or TaqI (SibEnzyme, Russia) for 16 hours. Gel blocks were stained with ethidium bromide (0.5 µg/ml) and visualized on UV transilluminator to detect DNA fragments. The sequencing reaction was performed using the BigDye v. 3.1 Cycle Sequencing Kit. DNA sequencing was performed on the ABI 3500 genetic analyzer (Applied Biosystems, USA). Result: For both polymorphisms in all breeds of pigs they revealed a higher frequency of mutant alleles recorded in Yorkshires, as well as homozygous genotypes for them. At the same time, they showed a high degree of alleles and genotype frequencies dependence for two polymorphisms caused by linked inheritance. The allele frequencies of the studied breeds for the A1112G polymorphism correspond to the data of European selection. For T3266G polymorphism the Durocs revealed a significant deviation of genotype frequencies from the Hardy-Weinberg equilibrium, which may indicate an active breeding process in this breed. In addition to the studied polymorphisms relative to the reference sequence of the chromosome 18 (NC\_010460.4), we identified 11 other mutations, including 8 SNPs and 3 deletions. Of these, 10 mutations were previously described during sequencing of the leptin gene (the sequences U66254.1 and AJ865080.1). For the first time, we described the G3297A mutation in the leptin gene intron among Duroc pigs, which can potentially act as a genetic marker of lipid metabolism and fat content in pigs. Conclusion: Based on the results of the pig LEP gene fragment sequencing, they confirmed the presence of polymorphisms T3266G and A1112G relative to the reference sequence U66254.1. They revealed a higher frequency of mutant alleles recorded in Yorkshires, as well as homozygous genotypes for both polymorphisms in all breeds of pigs. At the same time, they showed a high degree of dependence of alleles and genotype frequencies for two polymorphisms caused by linked inheritance. According to the T3266G polymorphism, a significant deviation of the genotype frequencies from the Hardy-Weinberg equilibrium was revealed among the Durocs, which may be the consequence of an active selection process in this breed. The Duroc breed showed a new SNP in the leptin gene G3297A.

**Key words:** pigs, leptin gene, T3266G, A1112G polymorphisms, PCR-RFLP

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## ACID-BASE RELATIONSHIPS IN THE BODY OF FARM ANIMALS OF DIFFERENT AGES AND PRODUCTIVITY

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### ABSTRACT

They revealed the disorders of acid-base balance in the organism of animals within the age aspect and at different levels of productivity. Subcompensated acidosis was established during the first day of life among chickens, ducklings and calves. The balance of acid-base ratios differed in the blood, first of all, by a significant and negative shift of the buffer bases ( $-3.0 \pm 0.8$  mmol/L),  $p\text{CO}_2 = 5.5 \pm 0.9$  kPa,  $\text{pH}_{\text{ist}}$  of venous blood makes  $7.34 \pm 0.07$  on average. Acidic substances predominated in the urine of calves,  $\text{pH} = 6.23 \pm 0.18$ , ( $P < 0.05$ ). The chickens, ducklings and calves of older ages, the roosters of the parent livestock had no violations of the acid-base balance of blood. All parameters were within the physiological norm. The nature of acid-base relationships in the blood of laying hens largely depended on egg production. At an egg production of 95%, they found the acidosis of the metabolic nature of the subcompensated phase. The peculiarities of acid-base relation development in ontogenesis and at high productivity of cattle and poultry must be taken into account in zootechnical practice.

**Key words:** acid-base balance, cattle, poultry, age aspect, productivity.

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## ENERGY METABOLISM AND PRODUCTIVITY OF FARM ANIMALS DURING ADAPTATION TO HIGH AIR TEMPERATURE

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### ABSTRACT

The unproductive energy consumption of calf bulls and chickens of different breeds and ages increased by 27% on average. This affected the efficiency of production, which significantly decreased. Black-and-white calf bulls have adapted to the air temperature of 38 °C with a great stress of physiological mechanisms, and, therefore, with large energy costs. Heat production among red steppe calf bulls increased maximally by 2.7 MJ, black-and-white calf bulls - by 5.7 MJ, which amounted to 18% and 42%, respectively. The growth energy with the same demand for exchange energy decreased by 3.5 MJ on average. At an air temperature of 28-30 °C, the consumption of mixed feed by hens decreased, the gross feed energy decreased by 12% on average. A significant part of the metabolic energy (23.6 kJ per 100 g of live weight) was released by hens in the form of heat, i.e., unproductively. The exchange energy by head per day was 951 kJ and turned out to be 18% less than at normal air temperature. The energy of sediments by head per day decreased by 25%, the average daily gain in live weight made 3 g on average. Thus, the preservation of thermal homeostasis in the body of calf bulls and chickens during adaptation to high air temperatures was achieved due to unproductive energy consumption increase and productivity decrease. In order to preserve the health and high productivity of animals during their maintenance and energy nutrition rationing, it is necessary to take into account the increase in unproductive energy consumption for the restoration of homeostasis and adaptation to the ambient temperature.

**Key words:** chickens, calf bulls, high air temperature, energy metabolism, productivity.

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## INFLUENCE OF PRODUCTION TRANSPORT ON PHYSIOLOGICAL STATE OF BROILER CHICKENS

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### ABSTRACT

In present research work, the effect of transportation and related factors on broiler chicken organism of the Hubbard F15 cross studied. Based on the analysis of leukogram parameters, leukocyte index dynamics, it was established that the transportation of poultry is characterized by a complex effect, leading to developing a stress reaction. Significant alterations in leukogram parameters (eosinophils, lymphocyte content decrease, the number of segmented lymphocytes, and leukocyte index increase) indicating the stress state of chickens are detected already 10 hours after the onset of exposure to a complex of transport factors.

**Keywords:** transportation, stress, Adaptation, leukogram, vibration, leukocyte indices, broiler chickens.

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## INFLUENCE OF INCREASED NOISE LEVEL ON PHYSIOLOGICAL STATE OF BROILER CHICKENS IN INDUSTRIAL CONDITIONS

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### ABSTRACT

The effect of noise with the sound level of 100 dB on broiler chicken body of the Hubbard F15 cross was studied in present work. Based on the analysis of the leukogram parameters, leukocyte index dynamics, it was established that the stress anxiety stage manifests itself 24 hours after the factor exposure, because by this time there is the decrease of lymphocyte content with a simultaneous increase of segmented neutrophil (heterophiles) number, the ratio of heterophiles to lymphocytes (H/L), and blood leukocyte shift index (BLSI). The effect of the studied sound level caused significant changes in the hematological and biochemical parameters of the chicken blood, so there was a significant change in total protein content, transamination enzymes Alanine aminotransferase (AIAT), Aspartate aminotransferase (AST),  $\alpha$ -amylase, glucose, immunoglobulins, erythrocytes, hemoglobin, and leukocytes. These changes in the blood system can characterize the activation of the energetic and protective properties of the poultry body to prevent stress.

**Keywords:** Adaptation, biochemical parameters, hematological parameters, leukogram, leukocyte indices.

## ADAPTATION OF FARM ANIMALS TO HOUSING CONDITIONS

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### ABSTRACT

The features of calves and chicken adaptation to new housing conditions, and to technological operations were studied in present work. It was found that the most serious stress factor for animals is transportation under conditions of high summer air temperatures. It is accompanied by regrouping, landing compaction, disruption of social contacts and leads to the organism adaptive mechanism stress. After transportation of calves to an industrial feeding complex, they revealed body and individual areas of skin surface temperature increase, stimulation of the cardiovascular and respiratory system functions, the heart rate increase by 17-25%, respiratory rate increase by 38-47%, increase of total protein concentration in blood serum, the decrease in hematocrit, etc. Acid-base balance disorders of an acidotic nature appeared with a significant decrease in blood pH, low values of buffer base concentration in the blood of both calves and chickens. After loading, transportation, and transplantation under conditions of summer heat, hens showed metabolic acidosis of the decompensated phase with a significant decrease in pH ( $7.27 \pm 0.07$ ), a negative and very significant shift in buffer bases ( $-16.7 \pm 5.9$ ), and their deficiency ( $12.1 \pm 3.1$ ). After transportation of chickens in winter at the temperature of  $-6^{\circ}\text{C}$ , the violations of the acid-base balance in chicken blood appeared, but they were less pronounced. It was found that the adaptation of calves and chickens to transport stress is very slow, affects health and productivity, which must be taken into account in the practice of animal husbandry and poultry farming.

**Key words:** Stress, cattle, housing conditions, adaptation

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## ACID-BASE BALANCE OF FARM ANIMALS DURING ADAPTATION TO HIGH AIR TEMPERATURE

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### ABSTRACT

The state of acid-base balance among the calf bulls of red steppe, black-and-white breeds and the chickens of different ages under the conditions of high air temperature was studied. At a temperature of 38 °C, the bulls increased the respiratory rate significantly (by 2.1-2.3 times), the volume of pulmonary ventilation (by 40.6-50.4%), oxygen consumption by 14.4% on average, and the release of carbon dioxide (by 31-40%). Thus, pCO<sub>2</sub> decreased by 0.55-0.65 kPa (P<0.01), the active reaction of the blood (pH<sub>ist.</sub>) increased by 0.017-0.018 (P<0.05). An alkalous shift of the respiratory character of the subcompensated stage was recorded in blood, which was more pronounced among black-and-white bulls, as they were less adapted to the heat. By the end of the four-hour exposure, the alkalous shift in the acid-base state of the calf blood persisted, but it passed into the compensated stage. At 21 °C and the humidity of 70% the balance of acid-base relations was observed among chickens. All parameters characterizing it were within the physiological norm. At 23 °C, respiratory subcompensated alkalosis was found in the blood of three-month-old pullets. Whereas among the poultry of other ages, the acid-base balance was maintained with the decrease of pCO<sub>2</sub> at this temperature. At 25 °C the chickens of all ages, except for 12-day-old chickens, had compensated respiratory alkalosis in blood. 12-day-old chickens showed metabolic acidosis in a compensated phase at this temperature. The shift of the buffer bases was negative and amounted to - 2.8 ± 0.5 mmol/L, pCO<sub>2</sub> - 4.5 ± 0.3 kPa. Due to the inclusion of the respiratory component of the compensation, the pH<sub>ist.</sub> remained in the lower limit of the norm. These features can be explained by the fact that the temperature of 25 °C was below the required temperature regime recommended for chicken growing of this age (29 - 31 °C). At 27 °C - the chickens of all studied ages had subcompensated respiratory alkalosis. At 31, 33, and 34 °C, respiratory alkalosis passed into a decompensated phase, even among the chickens at the age of 20 days, with the accumulation of buffer bases above the norm. Knowledge of the nature of disorders and adaptive capabilities of the acid-base balance of blood among farm animals and poultry under heat stress in the practice of animal husbandry and poultry farming will allow to take timely measures to maintain health and high productivity.

**Key words:** acid-base balance, cattle, poultry, heat stress, productivity.

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## THE STATE AND PROSPECTS OF SMART SPECIALIZATION OF THE AGRO-INDUSTRIAL COMPLEX OF THE BELGOROD REGION

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### ABSTRACT

Rational specialization of agribusiness is an actual subject of research, both at the level of individual enterprises and at the regional level. The latter is particularly important in the light of the significant differences between the regions of the Russian Federation in the results achieved by the agricultural sector of the economy in absolute and relative terms. In this regard, it is interesting to study the experience and prospects of development of one of the most successful regional agro-industrial complex. In contrast to the rational industry structure of the enterprise, the smart specialization of the agro-industrial complex is not just a successful combination of industries. At the regional level, smart specialization is characterized by effective interaction of economic entities, sustainable development of agribusiness both in individual enterprises and in agro-industrial formations that comprise several links in the value chain. Smart specialization of the regional agro-industrial complex cannot be achieved by directive methods. It is achieved through the coordinated interaction of public authorities and business structures. The experience of the Belgorod region shows that the creation of a favorable investment climate, political and organizational support allow private businesses to realize their commercial interests simultaneously with the implementation of regional and national economic interests. As a result, the Belgorod Region currently occupies a leading position among the regions of the Russian Federation. Promising areas for improving the specialization of the agro-industrial complex of the Belgorod region are also based on the effective interaction of the public authorities of the region and business structures.

**Keywords:** region, agro-industrial complex, smart specialization, agro-industrial integration, value chain.

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## THE POLYMORPHISM IN THE DYSTROPHIN GENE (DMD) OF VARIOUS BREEDS OF THE PIGS IN THE BELGOROD REGION OF RUSSIA

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### ABSTRACT

The pigs were genotyped for the dystrophin gene (DMD) using the ACRS-PCR method. The study examined 215 boars of four breeds (Duroc, Large White, Landrace and Yorkshire) from the Belgorod region of Russia. The mutation in the DMD gene leads to dystrophin expression decrease in the muscles of the lumbar region, longissimus dorsi, and also in the diaphragm. The animals with the mutant allele T had the average daily gain decrease in live weight, the fat thickness of the muscle layer depth reduction. To assess the frequency of alleles and genotype occurrence for the DMD gene among the Landrace, Duroc, Large White and Yorkshire breeds that are bred on the farms of the Belgorod region of Russia. Genomic DNA was isolated from alcoholized earmarks. Pig genotyping was performed using ACRS-PCR (Amplification Created Restriction Sites). Using primers, 151 b.p. DNA fragment was amplified from the forty-first exon of the dystrophin gene containing the target mutation. Primers were designed using the Primer3 software. 10 µl of the PCR product were hydrolyzed with 4 units of the Mox20I restriction enzyme (SibEnzyme, Russia) for 16 hours. The hydrolysis products were separated by horizontal electrophoresis in 4% agarose gel. Then, based on the results obtained, the pigs were genotyped by TaqMan PCR. The mutant allele T is observed only in the Landrace livestock with the frequency of 0.405. This is significantly higher than in the livestock of this breed from other regions and can be caused by the peculiarities of local selection. Based on the verified genotypes, they developed the method of pig genotyping for the DMD gene using real-time PCR technology based on TaqMan fluorescent probes. The high efficiency and reliability of the results of the TaqMan PCR technique and the possibility of its use in routine practice have been shown. The methods of pig genotyping for DMD gene mutations have been developed using ACRS-PCR and real-time PCR using TaqMan technology. The analysis of allele frequencies and genotypes of boars from the Duroc, Large White, Landrace and Yorkshire breeds at the farms of the Belgorod region of Russia was carried out according to the polymorphism of the dystrophin gene. As compared to previously obtained data from other regions of Russia, higher frequencies of the mutant allele T were revealed for the Landrace breed, which may be associated with the peculiarities of local selection for productive traits.

**Key words:** pigs, dystrophin gene, ACRS-PCR, TaqMan PCR