UDC 575:619:618.19-002:636.22/.28

GENETICS OF RESISTANCE TO CLINICAL MASTITIS IN COWS: A REVIEW

Fedota O. M.[1,2], Ruban S. Yu.[3], Bolotin V. I.[2], Klochko I. O.[2]

[1] V. N. Karazin Kharkov National University, Kharkiv, Ukraine, e-mail: afedota@mail.ru

[2] National Scientific Center 'Institute of Experimental and Clinical Veterinary Medicine', Kharkiv, Ukraine

[3] M. V. Zubets Institute of Animal Breeding and Genetics of NAAS, Chubynske, Ukraine

Summary. The article provides an overview of data about genetics of cattle and susceptibility to infectious diseases by the example of clinical mastitis. The problems of the genetic markers associated with mastitis susceptibility of cattle in studies of scientists from different countries are analyzed.

Clinical mastitis (CM) is an inflammation of the mammary gland associated with elevated somatic cell count (SCC) and is one of the biggest problems affecting commercial milk production. In the world literature the problems of mastitis etiology were discussed and it was noted that many factors promote the development of mastitis, including the genetic background and feeding environment. The unfavorable genetic correlations between milk production and clinical mastitis are well known. The complexity of mastitis infection suggests a polygenic and multi-factorial immune response comprising many different proteins. Most studies focused on polygenic variation of the trait and genetic correlation among phenotypic traits related to mastitis such as somatic cell counts and clinical cases.

At the present time, the role of Major Histocompatibility Complex (MHC genes) in the susceptibility or resistance to intrammamary infection is well studied. Researchers have shown that investigation of genetic markers associated with mastitis susceptibility, provide initial evidence for a phenotypic association between a single nucleotide polymorphisms of CXCR2, CD18, MBL1, TLR1, TLR2, CARD15, HMGB1, ATP1A1, BoLADQA1 genes and somatic cell counts and clinical manifestations in dairy cows, as well as potential insight into specific mechanisms affected in cows more susceptible to mastitis.

According researchers currently identified differences between breeds. Dairy breeds originating from eastern France (Montbéliarde, Abondance) or central Europe (Simmental, Brown Swiss) have lower SCC and clinical mastitis frequency than Holstein. Within breed, genetic variability is also quite large. Because the disease susceptibility may be genetically dependent, in which case disease resistance could be improved by animal selection through breeding programmes.

The most economical method of reducing SCC in Ukraine, based on the experience of other countries, including in our opinion, is testing monthly milk samples from each cow after the selection of genes for mastitis resistance.

Keywords: clinical mastitis, somatic cell count, genetic markers, mastitis susceptibility, mastitis resistance, dairy breeds

Clinical mastitis (CM) is an inflammation of the mammary gland associated with elevated somatic cell count (SCC) caused by microorganisms and is one of the most frequent infectious diseases in dairy cattle with important economic losses (Heringstad et al., 2006). At the present time, mastitis is one of the biggest problems affecting commercial milk production (Hinrichs et al., 2011).

In accordance with the data presented by Looper (2012), in the United States, federal law allows milk to be sold only if the bulk tank has an SCC of fewer than 750,000 cells/cm^3. The primary reason for dairy producers to reduce SCC is because it relates to milk losses due to mastitis. The author notes that lowering somatic cell count from 600,000 to 300,000 increases milk sales by $50 per cow per year, which is a small fraction of the total benefit. If they can reduce their SCC from 600,000 to 200,000 cells/cm^3, they can decrease milk production losses by 600 pounds per cow per year. In a 100-cow herd, these losses amount to $7,500/year if milk is valued at $12.50/cwt.

The requirements in Norway and England determine 150 cells/cm^3 of SCC as the upper limit, in Denmark — 200 cells/cm^3, in most European countries — 400 cells/cm^3 (Kantsevich, Rusko and Baksheiev, 2014).

Ukrainian companies in the dairy industry must meet the high requirements for the European market. Ukraine produces more milk than eastern European countries — Romania, Czech Republic, Hungary, Bulgaria, but the proportion of raw milk that goes to industrial processing, less 41–46% in the Ukraine compare to Poland, the Czech Republic and Hungary (74–87%). The reason is the low quality of raw milk produced in the households of the population in Ukraine (> 75% of raw milk in Ukraine).

In 2011–2015 in the eastern Ukrainian farms clinical serous form of mastitis had detected in 18.3% of the cows, subclinical form — at 81.7%. In 35.7% of clinically healthy cows, SSC in milk samples is higher than 400,000 cells/ml (DSTU 3662–97) and is within 404,000–7,178,000 cells/cm^3 (Levchenko,

2015; DSU, 1997). The most numerous breed in this region is a Ukrainian Dairy Black and White breed, created on the basis of Holstein (Ruban and Fedota, 2013).

Over the last 10 years the study of mastitis and prevention are held in many countries for cattle of different breeds, for example, in Spanish Holstein (Pérez-Cabal et al., 2009) and Czech Holstein cows (Wolf, Wolfová and Štípková, 2010), in Austrian Fleckvieh cows (Koeck et al., 2010), in Dutch Holstein (Bloemhof, Dejong and Dehaas, 2009) and Norwegian Red cattle (Opsal et al., 2008), in US Holsteins (Vazquez et al., 2009) and Holstein cattle from Argentina (Baltian et al., 2012), in Holstein × Zebu (Duangjinda et al., 2008).

It was currently identified differences between breeds. According to Rupp and Boichard (2003), these genetic differences could be estimated in herds with different breeds. Dairy breeds originating from eastern France (Montbéliarde, Abondance) or central Europe (Simmental, Brown Swiss) have lower SCC and clinical mastitis frequency than Holstein. Genetic variability is also quite large within a breed. The genetic standard deviation of clinical mastitis frequency reaches about 5%. This means that in an environment with 20% average frequency of clinical mastitis, the frequencies observed for extreme genotypes range from 10% to 30%.

The authors note that many factors promote the development of mastitis, including the genetic background and feeding environment (Takeshima et al., 2008). It is known (Park et al., 2004) that such factors as environment, pathogen, and host affect susceptibility or resistance of an individual cow to bovine mastitis. According to Sæbø and Frigessi (2004), the pathogens causing mastitis are various species of bacteria, but a cow's susceptibility to the disease also depends on many other factors. It is also important to maintain the animal welfare such as sanitation, climate change and the value of the stock, among other things, as an environmental factor.

Due to the fact that the disease susceptibility may be genetically dependent, it could be improved by animal selection through breeding programs (Sæbø and Frigessi, 2004).

Breeding for mastitis resistance is becoming increasingly important because of its effect on farm economy and animal welfare. At the present time, the unfavorable genetic correlations between milk production and clinical mastitis (CM) are well known (Heringstad, Klemetsdal and Ruane, 2000; Oltenacu and Broom, 2010). Most of the countries that perform genetic evaluations for mastitis resistance lack records of CM because disease recording systems are not well developed. Thus, most commonly SCC is used as an indirect measure (Koeck et al., 2010). Wherein mastitis data recording should be carried out in early lactation because these mastitis cases are more related to the genetics of the cow than cases in late lactation, which are more affected by the environment (Hinrichs et al., 2011).

According to Baltian et al. (2012), mastitis is a complex disease that involves three major environment factors: the microorganisms as the causative agent, the cow as host, and the environment which can influence both the cow and the microorganisms. Among the major microorganisms responsible for the development of mastitis can be mentioned *Streptococcus agalactiae*, *Staphylococcus aureus* and *Streptococcus dysgalactiae* (Baltian et al., 2012). According to Ukrainian authors (Levchenko, 2015), cows with clinical and subclinical mastitis serous isolated culture *Staphylococcus aureus* (60% and 33%) *Streptococcus agalactiae* (26.6% and 20.1%), *Escherichia coli* (13.4% and 13.3%). These bacteria produce toxins that injure the milk-secreting tissues and various ducts throughout the mammary gland (Takeshima et al., 2008).

Because high SCC in milk is a response to a presence of microbes in the mammary gland, SCC can be used both as an indicator of mastitis and as a measure of response to infection. In many countries, SCC (treated homogeneously) is used as an indirect selection criterion for improving mastitis resistance and as characteristic of genotypes for the genes of resistance to mastitis (Detilleux, 2009; Heringstad et al., 2006).

Resistance to mastitis is a complex function involving various biological pathways, molecules and cells. Therefore numerous functional candidate genes could be involved in the determinism of the function as reported by Detilleux (2009). Genetic selection for increasing antibody responsiveness seems to be possible, but it should be pointed out that trends in association with clinical mastitis occurrence were not straight forward and those further investigations are needed to address genetics of immunology in mastitis (Rupp and Boichard, 2003).

Several chromosomal regions have large effects on mastitis resistance. The multitude of results reflects, to some extent, the fact that mastitis resistance is a complex function that involves many molecules and pathways that can be regulated by many different genes (Rupp and Boichard, 2003).

The complexity of mastitis infection suggests a polygenic and multifactorial immune response comprising many different proteins (Russel et al., 2012). Effective elimination of bacterial infections, such as mastitis in dairy cattle, requires four basic steps: bacterial recognition, inflammatory mediator release, leukocyte

recruitment from the bloodstream, and bacteria removal (Riollet, Rainard and Poutrel, 2000). Researchers have shown (Rambeaud and Pighetti, 2005) that investigation of genetic markers associated with mastitis susceptibility, provide initial evidence for a phenotypic association between a single nucleotide polymorphism of CXCR2 gene and neutrophil function in dairy cows, as well as potential insight into specific mechanisms affected in cows more susceptible to mastitis. Cows with the CC or GC genotype at CXCR2 +777 showed significantly lower neutrophil migration to recombinant human interleukin-8 (rhIL-8) than cows with the GG genotype. Cows with the CC genotype at CXCR2 +777 also showed decreased neutrophil migrations to zymosan-activated serum compared to these same cows. According to the authors, polymorphisms in bovine CXCR2 may potentially be used to select cows that are more resistant to disease. Of the sixteen polymorphisms in seven immune genes genotyped, presented by Beecher et al. (2010) just CXCR1–777 tended to associate with SCS, albeit only in the on-farm study (Beecher et al., 2010).

Several authors (Rupp and Boichard, 2003) focused on a mutation in the CD18 gene (BTA1) associated with bovine leukocyte adhesion deficiency (BLAD) in Holstein cattle. Cattle that are homozygous for the deleterious allele exhibit impaired diapedesis of leucocytes, extreme sensitivity in any infection and premature death. However, no difference in susceptibility to intramammary infections has been found in heterozygous carriers at the CD18 gene.

Wang et al. (2011) described three novel single-nucleotide polymorphisms of MBL1 gene in Chinese native cattle and their associations with milk performance traits**.** Among the Chinese Holstein cattle, eight different haplotypes and 19 genotype combinations were detected. Statistical analyses revealed no correlation between either g.855 G>A or g.2686 T>C and somatic cell score (SCS) however, a significant association was found between g.2651 G>A and SCS, suggesting a possible role of this SNP in the host response against mastitis. The combined genotypes of GGC/AAC with the lowest SCS, AAT/AAT with the highest protein content and AGC/AGC with the highest 305-days milk yield were favorable combinations for mastitis resistance and milk production traits. According to researchers, GGC/AAC, AAT/AAT and AGC/AGC can be used as possible candidates for marker-assisted selection in the dairy cattle breeding program.

Toll-like receptors (TLR) are important cell-surface molecules mediating immune response (Opsal et al., 2008). Investigations of whether SNPs within the bovine TLR1 gene (bo*TLR1*) are associated with clinical mastitis (CM) were presented by Russell et al. (2012). Selected bo*TLR1* SNPs were analyzed within a Holstein Friesian herd. Significant associations were found for the tagging SNP –79 T>G and the 3'UTR SNP +2463 C>T. Animals with the GG genotype (from the tag SNP -79 T>G) had significantly lower bo*TLR1* expression in milk somatic cells when compared with TT or TG animals. In addition, stimulation of leucocytes from GG animals with the TLR1-ligand Pam3 csk4 resulted in significantly lower levels of CXCL8 mRNA and protein. The authors concluded, that rapid immune response, conferred by the favorable bo*TLR1* SNP -79 TT variant, could reduce detrimental clinical manifestations of diseases via an efficient yet controlled influx of inflammatory cells to neutralise pathogens and control infection.

The results of association research of toll-like receptor 2 gene (TLR2) polymorphisms with somatic cell score in Xinjiang Brown cattle showed (Bai et al., 2012) that the SCS of AB genotype was lower than AA, the SCS of cattle with Hap5 was lower than Hap3. This suggests that Hap5 might play an important role in sub-mastitis resistance in Xinjiang Brown cattle.

Results of the study associations between CARD15 and TLR2 gene polymorphisms and milk somatic cell score in Canadian Holsteins were introduced (Pant et al., 2008). According to the authors, Toll-like receptor-2 (TLR2) and caspase recruitment domain 15 (CARD15) are important pattern recognition receptors that play a role in the initiation of the inflammatory and subsequent immune response. The hap21 (TA) in CARD15 was significantly associated with increased SCS EBV (estimated breeding values) and SNP c.3020 A>T was associated with SCS EBVs and might play a role in the host response against mastitis. In a study of Opsal et al. (2008) presented, that dense linkage maps comprising single nucleotide polymorphisms (SNPs) have been constructed for the chromosomal regions harboring TLR2 and TLR4 on bovine chromosome 17 and 8.

The most likely marker orders for both regions were compared with the corresponding human map positions and used to reorder bovine scaffolds available from the bovine genome sequence assembly (Heringstad et al., 2006). Haplotype analysis of TLR4 gene and its effects on milk somatic cell score in Chinese commercial cattle had been carried out in Chinese commercial dairy cattle including Chinese Holstein, Sanhe cattle and Chinese Simmental breeds by Wang et al. (2014). Results showed that Hap1 (30.5%) and Hap2 (30.4%) were the most common haplotypes. Hap2, Hap4 and Hap12 might negatively effect on milk SCS whereas Hap13 might positively effect on milk SCS.

Li et al. (2012) mentioned that one SNP in the 3'-UTR of HMGB1 gene affects the binding of target bta-miR-223 and is involved in mastitis in dairy cattle. According to the authors, high-mobility group box protein 1 (HMGB1) gene has a universal sentinel

function for nucleic-acid-mediated innate immune responses and acts as a pathogenic mediator in the inflammatory disease. They showed that the relative expression of HMGB1 mRNA in cows with the genotype GG is significantly higher than those in cows with the genotype AA. One novel SNP (g. +2776 A>G) in the HMGB1 3'-UTR, altering the binding of HMGB1 and bta-miR-223, was found to be associated with somatic count scores in cows. The g. +2776 A>G-GG was an advantageous genotype which can be used as a candidate functional marker for mastitis resistance breeding program.

Due to the fact that mastitis affects the concentrations of potassium and sodium in milk, presumed that polymorphism of the *ATP1 A1* gene, which encodes the bovine Na_+, K_+-ATPase α1 subunit could be associated with mastitis (Liu et al., 2012). The authors showed that the cows with genotype *CC* in *ATP1 A1* had significantly lower somatic cell scores and 305-day milk yields than those with genotype *CA*. The Na_+, K_+-ATPase activity was significantly higher in dairy cows with genotype *CC* compared to the other two genotypes, and the Na_+, K_+-ATPase activity of the resistant group was significantly higher than that of the susceptible group in dairy cows. In this regard, researchers conclude that this polymorphism has potential as a marker for mastitis resistance in dairy cattle.

Sugimoto, Uchiza and Kuniyuki (2013) have reported that the forebrain embryonic zinc finger-like (FEZL) gene promotes immune responses that are associated with mastitis resistance. According to Takeshima et al. (2008), it is preferable to genotype all known candidate genes simultaneously for estimating the susceptibility or resistance of animals to multifactorial diseases like mastitis. However, it remains difficult to genotype MHC alleles, although most candidate genes, once identified, can be readily genotyped using a method for detecting single nucleotide polymorphisms.

It is now known, that several host genes have been suggested to promote resistance or susceptibility to clinical mastitis, particularly major histocompatibility complex (MHC) genes (Takeshima et al., 2008).

According to Baltian et al. (2012), in bovine MHC (named bovine lymphocyte antigen, BoLA) some polymorphisms were associated with resistance to infectious diseases such as mastitis, enzootic bovine leukemia and others diseases. The general structure of BoLA is similar to the MHC of others mammalian species and is made up of three regions: Class I, Class II and Class III, which fulfill different roles. The class II genes encode proteins that presented processed peptides derived from extracellular antigens to CD4 cells, like DRA, DRB, DQA and DQB. In the bovine DR subregion, there are at least three BoLA-DRB loci but only the BoLA-DBR3 gene in functional. This gene is the highly polymorphic since more than 103 alleles have been reported thus far.

Many reports have documented the association of BoLADRB3 alleles with the progression of mastitis. They have also detected disease-associated BoLADRB3 alleles. These results showed that while the existence of particular BoLADRB3 alleles is important for the progression of mastitis (Baltian et al., 2012; Duangjinda et al., 2008; Sharif, Mallard and Sargeant, 2000; Takeshima et al., 2008). In the works of Takeshima et al. (2008) it was shown that heterozygosity of the BoLADQA1 gene is associated with resistance to mastitis progression.

Park et al. (2004) have presented that susceptibility to mastitis was associated with major histocompatibility complex (MHC) haplotypes that have only a single set of DQ genes. The study also revealed that susceptible cows had CD4:CD8 ratios of less than one in both their mammary gland secretions and peripheral blood. These results raise the possibility that the number of DQ genes that a cow has and/or a cow's CD4:CD8 ratio could be used as indicators of susceptibility to bovine mastitis.

According to Takeshima et al. (2008), the MHC genes are a classic example of where heterozygote advantage may apply because heterozygosity at MHC loci may enhance resistance to infectious diseases as it increases the diversity of the antigens presented to T cells, thereby generating a more diverse T-cell repertoire. Such heterozygote-favoring selection is one of the mechanisms that maintain genetic polymorphism.

Potential relationships between amino acid motifs in the antigen binding groove of various alleles of the bovine major histocompatibility complex DR (BoLA-DR) molecule and occurrence of clinical mastitis caused by *Staphylococcus* species (non-*Staphylococcus aureus*) were presented by Sharif, Mallard and Sargeant (2000). A significant association was detected between the presence of glutamic acid at position beta 74 in BoLA-DRB3.2*22, *23 and *24 alleles and occurrence of mastitis caused by *Staphylococcus* spp. with a relative risk of 11. These positions (beta 13, beta 71 and beta 74) form pocket 4 of the antigen binding groove, which plays an instrumental role in antigen binding and recognition by T-lymphocytes. The authors concluded, pocket 4 of the BoLA-DR molecule is involved in conferring susceptibility to clinical mastitis caused by *Staphylococcus* spp.

The association of the polymorphism of bovine leukocyte antigen (BoLA-DRB3) genes with resistance and susceptibility to mastitis caused by *Streptococci*, coagulase-negative *Staphylococci*, *Escherichia coli* and *Staphylococcus aureus* was investigated by Yoshida et al. (2012). The researchers concluded that in the case

of *Escherichia coli* mastitis, amino acid substitutions at the 9, 11, 13, and 30 positions had little effect, but rather substitutions at the 47, 67 positions of pocket 7, and at the 71, 74 positions of pocket 4, Tyr (47), Ile (67), Ala (71), and Ala (74), were associated with resistance and this motif was present in DRB3*1201. In 2012 the authors provided data (33), that DRB3.2*8 (DRB3*1201) and DRB3.2*16(DRB3*1501) alleles were found to be associated with susceptibility, while DRB3.2*22(DRB3*1101), DRB3.2*23(DRB3*2703), and DRB3.2*24(DRB3*0101) alleles were found to be associated with resistance.

According to de Haas et al. (2008), genetic selection on lowering the log-transformed lactation-average SCC may, therefore, be more effective in decreasing mastitis incidence than genetic selection on lowering the lactation-average of log-transformed SCC.

Looper (2012) suggests that there are several methods of reducing SCC. The first method, culling cows, is a short-term solution, which can quickly reduce SCC in the bulk tank. The second method, controlling mastitis, is a long-term solution, which should be the basis of a sound management program. The most economical method, including in our opinion, and in Ukraine, to determine SCC is testing monthly milk samples from each cow after the selection of genes for mastitis resistance.

It needs to provide further in-depth studies among Ukrainian herds with genetical analysis. We believe that due the increase in the consumption of dairy products in the world and low current exports from Ukraine. It will give an opportunity to significantly increase participation in world trade and milk producers should improve the quality of dairy products.

References

Bai, J., Lin, J., Li, W. and Liu, M. (2012) 'Association of toll-like receptor 2 polymorphisms with somatic cell score in Xinjiang brown cattle', *Animal Science Journal*, 83(1), pp. 23–30. doi: 10.1111/j.1740-0929.2011.00909.x.

Baltian, L. R.,Ripoli, M. V.,Sanfilippo, S.,Takeshima, S. N., Aida, Y., Giovambattista, G., Sanfilippo, S., Aida, Y. and Giovambattista, G. (2012) 'Association between BoLA-DRB3 and somatic cell count in Holstein cattle from Argentina', *Molecular Biology Reports*, 39(7), pp. 7215–7220. doi: 10.1007/s11033-012-1526-y.

Beecher, C., Daly, M., Childs, S., Berry, D. P., Magee, D. A., McCarthy, T. V. and Giblin, L. (2010) 'Polymorphisms in bovine immune genes and their associations with somatic cell count and milk production in dairy cattle', [Online] *BMC Genetics*, 11(99), p. 1–9. doi: 10.1186/1471-2156-11-99.

Bloemhof, S., Dejong, G. and Dehaas, Y. (2009) 'Genetic parameters for clinical mastitis in the first three lactations of Dutch Holstein cattle', *Veterinary Microbiology*, 134(1–2), pp. 165–171. doi: 10.1016/j.vetmic.2008.09.024.

DSU [State Committee of Ukraine for Standardization, Metrology and Certification] (1997) *DSTU 3662–97. Whole cow milk. Requirements for purchasing [Moloko koroviache nezbyrane. Vymohy pry zakupivli]*. Kyiv: Derzhstandart Ukrainy. [in Ukrainian].

Detilleux, J. (2009) 'Genetic factors affecting susceptibility to udder pathogens', *Veterinary Microbiology*, 134(1–2), pp. 157–164. doi: 10.1016/j.vetmic.2008.09.023.

Duangjinda, M., Buayai, D., Pattarajinda, V., Phasuk, Y., Katawatin, S., Vongpralub, T. and Chaiyotvittayakul, A. (2008) 'Detection of bovine leukocyte antigen DRB3 alleles as candidate markers for clinical mastitis resistance in Holstein × Zebu', *Journal of Animal Science*, 87(2), pp. 469–476. doi: 10.2527/jas.2007-0789.

Haas, Y. de, Ouweltjes, W., Napel, J. ten, Windig, J. J. and de Jong, G. (2008) 'Alternative somatic cell count traits as mastitis indicators for genetic selection', *Journal of Dairy Science*, 91(6), pp. 2501–2511. doi: 10.3168/jds.2007-0459.

Heringstad, B., Gianola, D., Chang, Y. M., Ødegård, J. and Klemetsdal, G. (2006) 'Genetic associations between clinical mastitis and somatic cell score in early first-lactation cows', *Journal of Dairy Science*, 89(6), pp. 2236–2244. doi: 10.3168/jds.s0022-0302(06)72295-0.

Heringstad, B., Klemetsdal, G. and Ruane, J. (2000) 'Selection for mastitis resistance in dairy cattle: A review with focus on the situation in the Nordic countries', *Livestock Production Science*, 64(2–3), pp. 95–106. doi: 10.1016/s0301-6226(99)00128-1.

Hinrichs, D., Bennewitz, J., Stamer, E., Junge, W., Kalm, E. and Thaller, G. (2011) 'Genetic analysis of mastitis data with different models', *Journal of Dairy Science*, 94(1), pp. 471–478. doi: 10.3168/jds.2010-3374.

Kantsevich, S. I., Rusko, N. P. and Baksheiev, M. M. (2014) 'Estimation of milk quality impact on economic efficiency of milk production', *The Economy of Agro-Industrial Complex*, 4, pp. 24–27. Available at: http://eapk.org.ua/sites/default/files/translate/1404kantsevich_rusko_baksheiev.pdf.

Koeck, A., Heringstad, B., Egger-Danner, C., Fuerst, C., Winter, P. and Fuerst-Waltl, B. (2010) 'Genetic analysis of clinical mastitis and somatic cell count traits in Austrian Fleckvieh cows', *Journal of Dairy Science*, 93(12), pp. 5987–5995. doi: 10.3168/jds.2010-3451.

Levchenko, A. G. (2015) *The characteristics of the manifestation of mastitis in cows in farms with various technologies and the development of a comprehensive preventive therapeutic measures [Osoblyvosti proiavu mastytu u koriv u hospodarstvakh z riznymy tekhnolohiiamy ta rozrobka kompleksnykh profilaktychno likuvalnykh zakhodiv]*. The thesis for the scientific degree of the candidate of veterinary sciences, specialty 16.00.03 — veterinary microbiology, epizootology, infectious diseases and immunology. Kyiv: State Scientific Control Institute of Biotechnology and Strains of Microorganisms. [in Ukrainian].

Li, L., Huang, J., Zhang, X., Ju, Z., Qi, C., Zhang, Y., Li, Q., Wang, C., Miao, W., Zhong, J., Hou, M. and Hang, S. (2012) 'One SNP in the 3′-UTR of HMGB1 gene affects the binding of target bta-miR-223 and is involved in mastitis in dairy cattle', *Immunogenetics*, 64(11), pp. 817–824. doi: 10.1007/s00251-012-0641-1.

Liu, Y. X., Xu, C. H., Gao, T. Y. and Sun, Y. (2012) 'Polymorphisms of the ATP1A1 gene associated with mastitis in dairy cattle', *Genetics and Molecular Research*, 11(1), pp. 651–660. doi: 10.4238/2012.march.16.3.

Looper, M. (2012) *Reducing somatic cell count in dairy cattle*. Division of Agriculture Research and Extension, University of Arkansas System, FSA4002. Available at: http://www.uaex.edu/publications/pdf/fsa-4002.pdf.

Oltenacu, P. A. and Broom, D. M. (2010) 'The impact of genetic selection for increased milk yield on the welfare of dairy cows', *Animal Welfare*, 19(S1), pp. 39–49. Available at: http://www.fao.org/fileadmin/user_upload/animalwelfare/dairy.pdf.

Opsal, M. A., Lien, S., Brenna-Hansen, S., Olsen, H. G. and Våge, D. I. (2008) 'Association analysis of the constructed linkage maps covering TLR2 and TLR4 with clinical mastitis in Norwegian red cattle', *Journal of Animal Breeding and Genetics*, 125(2), pp. 110–118. doi: 10.1111/j.1439-0388.2007.00704.x.

Pant, S. D., Schenkel, F. S., Leyva-Baca, I., Sharma, B. S. and Karrow, N. A. (2008) 'Identification of polymorphisms in bovine *TLR2* and *CARD15*, associations between *CARD15* polymorphisms and milk somatic cell score in Canadian Holsteins, and functional relevance of SNP c.3020A>T', *Developments in biologicals (Basel)*, 132, pp. 247–253. doi: 10.1159/000317167.

Park, Y. H, Joo, Y. S., Park. J. Y., Moon, J. S., Kim, S. H., Kwon, N. H., Ahn, J. S., Davis, W. C. and Davies, C. J. (2004) 'Characterization of lymphocyte subpopulations and major histocompatibility complex haplotypes of mastitis-resistant and susceptible cows', *Journal of Veterinary Science*, 5(1), pp. 29–39. Available at: http://www.vetsci.org/journal/download_pdf.php?spage=29&volume=5&number=1.

Pérez-Cabal, M. A., de los Campos, G., Vazquez, A. I., Gianola, D., Rosa, G. J. M., Weigel, K. A. and Alenda, R. (2009) 'Genetic evaluation of susceptibility to clinical mastitis in Spanish Holstein cows', *Journal of Dairy Science*, 92(7), pp. 3472–3480. doi: 10.3168/jds.2008-1978.

Rambeaud, M. and Pighetti, G. M. (2005) 'Impaired neutrophil migration associated with specific bovine CXCR2 Genotypes', *Infection and Immunity*, 73(8), pp. 4955–4959. doi: 10.1128/iai.73.8.4955-4959.2005.

Riollet, C., Rainard, P. and Poutrel, B. (2000) 'Differential induction of complement fragment C5a and inflammatory cytokines during intramammary infections with *Escherichia coli* and *Staphylococcus aureus*', *Clinical and Vaccine Immunology*, 7(2), pp. 161–167. doi: 10.1128/cdli.7.2.161-167.2000.

Ruban, S. Yu. and Fedota, O. M. (2013) 'The directions of selection organization in the dairy and beef cattle breeding of Ukraine' [Napriamy orhanizatsii selektsiinoi roboty v molochnomu ta miasnomu skotarstvi Ukrainy], *Animal Breeding and Genetics [Rozvedennia i henetyka tvaryn]*, 47, pp. 5–13. Available at: http://nbuv.gov.ua/UJRN/rgt_2013_47_3. [in Ukrainian].

Rupp, R. and Boichard, D. (2003) 'Genetics of resistance to mastitis in dairy cattle', *Veterinary Research*, 34(5), pp. 671–688. doi: 10.1051/vetres:2003020.

Russell, C. D., Widdison, S., Leigh, J. A. and Coffey, T. J. (2012) 'Identification of single nucleotide polymorphisms in the bovine toll-like receptor 1 gene and association with health traits in cattle', [Online] *Veterinary Research*, 43(17), pp. 1–12. doi: 10.1186/1297-9716-43-17.

Sæbø, S. and Frigessi, A. (2004) 'A genetic and spatial Bayesian analysis of mastitis resistance', *Genetics Selection Evolution*, 36(5), p. 527–542. doi: 10.1186/1297-9686-36-5-527.

Sharif, S., Mallard, B. A. and Sargeant, J. M. (2000) 'Presence of glutamine at position 74 of pocket 4 in the BoLA-DR antigen binding groove is associated with occurrence of clinical mastitis caused by *Staphylococcus* species', *Veterinary Immunology and Immunopathology*, 76(3–4), pp. 231–238. doi: 10.1016/s0165-2427(00)00216-6.

Sugimoto, M., Uchiza, M. and Kuniyuki, M. (2013) 'Effects of a Forebrain embryonic zinc finger-like p.Gly105(12_13) polymorphism on mastitis resistance: An embryo-transfer study', [Online] *Molecular Biology and Genetic Engineering*, 1(1), pp. 1–3. doi: 10.7243/2053-5767-1-1.

Takeshima, S., Matsumoto, Y., Chen, J., Yoshida, T., Mukoyama, H. and Aida, Y. (2008) 'Evidence for cattle major histocompatibility complex (BoLA) class II DQA1 gene heterozygote advantage against clinical mastitis caused by *Streptococci* and *Escherichia* species', *Tissue Antigens*, 72(6), pp. 525–531. doi: 10.1111/j.1399-0039.2008.01140.x.

Vazquez, A. I., Weigel, K. A., Gianola, D., Bates, D. M., Perez-Cabal, M. A., Rosa, G. J. M. and Chang, Y. M. (2009) 'Poisson versus threshold models for genetic analysis of clinical mastitis in US Holsteins', *Journal of Dairy Science*, 92(10), pp. 5239–5247. doi: 10.3168/jds.2009-2085.

Wang, C., Liu, M., Li, Q., Ju, Z., Huang, J., Li, J., Wang, H. and Zhong, J. (2011) 'Three novel single-nucleotide polymorphisms of MBL1 gene in Chinese native cattle and their associations with milk performance traits', *Veterinary Immunology and Immunopathology*, 139(2–4), pp. 229–236. doi: 10.1016/j.vetimm.2010.10.023.

Wang, X. P., Luoreng, Z. M., Gao, S. X., Guo, D. S., Li, J. Y., Gao, X., Xu, S. Z., Li, F., Chen, G. and Wang, J. R. (2014) 'Haplotype analysis of TLR4 gene and its effects on milk somatic cell score in Chinese commercial cattle', *Molecular Biology Reports*, 41(4), pp. 2345–2351. doi: 10.1007/s11033-014-3088-7.

Wolf, J., Wolfová, M. and Štípková, M. (2010) 'A model for the genetic evaluation of number of clinical mastitis cases per lactation in Czech Holstein cows', *Journal of Dairy Science*, 93(3), pp. 1193–1204. doi: 10.3168/jds.2009-2443.

Yoshida, T., Furuta, H., Kondo, Y. and Mukoyama, H. (2011) 'Association of BoLA-DRB3 alleles with mastitis resistance and susceptibility in Japanese Holstein cows', *Animal Science Journal*, 83(5), pp. 359–366. doi: 10.1111/j.1740-0929.2011.00972.x.