
# Modern biotechnology used in animal breeding, elements of genetic engineering and advances in breeding

**Module no. 2: Conservation and Sustainable Use of Animal Genetic Resources**

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Modern biotechnology optimally exploits DNA recombination techniques and is associated with the development of microbiology, biochemistry,  molecular and cellular biology, chemistry, computer science, and physics.

BIOTECHNOLOGICAL RESEARCH IS INTERDISCIPLINARY RESEARCH

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Biotechnological research goals are:

- optimization of gene expression levels
- modification of nucleotide sequences encoding a protein or regulating gene expression
- sequencing of the genome and genes of a selected organism and comparison with previously known sequences
- construction of transgenic organisms with new traits
- somatic and reproductive gene therapy
- genetic diagnostics
- animal cloning

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Attaining biotechnological goals involves not only achievements, but also concerns and risks:

- Correct diagnoses and improved methods for preventing and treating genetic and infectious diseases.
- New beneficial varieties of plants and animals
- New strains of microorganisms with beneficial productivity traits
- New substances obtained through research and implementation
- New methods of environmental protection
- Potential damage to other organisms or the environment caused by transgenic strains and varieties
- A reduction in biological genetic diversity
- Interference with human and animal genotypes
- Violations of the boundaries of personal information
- Further privileging of wealthy people and countries due to the availability of expensive and patented technologies

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For about a decade, biotechnology has been divided into three main fields:

- Red biotechnology, leading to new means of preventing, treating and curing diseases, especially previously incurable ones, using new diagnostic and treatment methods
- Green biotechnology, i.e. genetic manipulation in plants and animals leading to improved yields and new agricultural products, as well as new traits in plants and animals
- White biotechnology, i.e. applications in industry

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Green biotechnology - – the production of genetically modified plants and animals. The effects of green biotechnology can be seen in the following areas:

- in vitro production of starting material for crops
- the introduction of genes determining desired traits in plants and animals – transgenesis
- **breeding using genetic markers**

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In modern animal breeding, which is focused on maximum genetic progress, the most important role is ascribed to genetic markers and their use for the assessment of genetic progress and the breeding value of animals.

**So what is a genetic marker?**

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A **genetic marker** can be a gene or DNA sequence of known location in the animal genome, linked to a gene or chromosome fragment determining a specific trait (usually an important quantitative trait in breeding of a given species).

A marker's usefulness depends on whether it is **polymorphic**, i.e. at least two alleles occur at the locus of the marker in the population.

Polymorphism of markers is tested by analytical methods, among which serological methods using test sera were once dominant, and biochemical methods, using techniques based on protein electrophoresis and techniques using DNA sequence analysis.

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Genetic markers are divided into two classes.

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**Class I** comprises classical markers, i.e. genes. Polymorphism of these markers was previously detected by analysing gene products (serological methods and protein electrophoresis), but it can also be tested by analysing the sequences of these genes, using methods such as DNA sequencing or **RFLP** (restriction fragment length polymorphism) or **SSCP** (simple sequence length polymorphism) tests.

**Class II** – polymorphic non-coding sequences, among which tandemly repeated **microsatellite** sequences, and to a lesser degree, **minisatellite** sequences, are considered the most important. Polymorphism of this class of markers is analysed exclusively by DNA sequence analysis.

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Apart from DNA markers, we can distinguish another group of markers associated with chromosomal polymorphism, which is detected using chromosome banding techniques. Chromosomal polymorphism mainly affects the size of constitutive heterochromatin blocks (CBG staining technique) and nucleolar organizer regions (Ag-NOR technique).

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Genetic progress in animals is undoubtedly influenced by properly conducted selection work.

Selection of animals on the basis of breeding value assessments estimated using genetic markers is known as marker-assisted selection (**MAS**). The usefulness of genetic markers in MAS programmes depends on how much of the genetic variance of a given trait can be explained by information available from genetic markers.

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The use of genetic markers in breeding programmes increases the accuracy of breeding value assessment and thereby increases genetic progress. The additional information about the breeding value of animals provided by genetic markers is especially useful in the case of traits with low heritability, traits whose measurements are available in the later period of the animal's life or only after it is slaughtered, and traits associated with the animal's sex. Another benefit of the use of genetic markers is shortening of the generation interval. The ability to quickly obtain information about an individual (made possible by genetic markers) can significantly shorten the generation interval and accelerate breeding progress.

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The usefulness of genetic markers in assessment of the breeding value of animals depends on how strongly they are linked to the quantitative trait locus (QTL). The strength of the linkage of the marker to the QTL is described by linkage disequilibrium (LD), i.e. the non-random association of alleles from two or more loci. If the genetic markers are linked to a given locus in a completely random way, they are in linkage equilibrium (LE). According to the strength of the link, genetic markers are classified as LD or LE. The most useful markers in animal breeding are those in strong linkage disequilibrium with the quantitative trait loci, which means that the linkage with the QTL is rarely broken by recombinations.

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The use of information provided by genetic markers to assess breeding value by the BLUP method involves including the QTL effect in a mixed model as an additional random effect. QTL effects can be estimated by analysing the linkages of genetic markers to qualitative trait genes. Based on this analysis, chromosome regions in which the sought-for QTL is most likely to be located are selected. In the case of breeding value assessment based on the model with the additional random QTL effect, MAS consists in combining the use of phenotypic information (as in classical phenotype-based methods of breeding value assessment) with the additional information provided by genetic markers.



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In recent years, research on the use of single nucleotide polymorphisms (SNPs) as a source of information about the breeding value of animals has become increasingly popular.

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In contrast to estimation of breeding value with the effect of the QTL in a mixed model, where identification of the QTL location requires a significance test, MAS using SNPs involves simultaneous estimation of the effects of all SNPs (or SNP haplotypes) without the need to test their significance. In these analyses it is assumed that the SNP markers will be the only source of information about the breeding value of animals (it will become unnecessary to gather phenotypic information), which will make it possible to obtain a precise breeding value assessment for very young individuals (immediately after birth). This will enable a significant reduction in the generation interval, faster genetic progress, and a reduction in the costs of breeding value assessment.

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The technology of SNP microarrays enables rapid genotyping of hundreds of thousands of SNP loci. From the vast number of SNPs identified, the most informative (*tag*SNPs) are selected. Then the most likely frequencies of SNP haplotypes are established, followed by a search for associations between SNP haplotypes and the traits whose breeding values are to be estimated

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After estimating the effects of haplotypes (which are assumed not to be specific to the individual but the same for the entire population), the genome-wide estimated breeding value (**GEBV**) is estimated as the sum of the effects of SNP haplotypes characteristic of a given genotype (individual). It is assumed that the effects of SNP haplotypes add up, and that epistatic interactions between SNP loci do not take place. GEBV can be estimated by the BLUP method, with the random effect of haplotypes included in the linear model.

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Polymorphism of genetic markers is exploited in research on the resistance/susceptibility of animals to various diseases and genetic disorders, owing to which carriers of deleterious alleles can be detected before the onset of disease symptoms. Particularly important research is conducted on the relationship between the antigens/genes of the major histocompatibility complex (MHC) and the occurrence of diseases, especially infectious ones.

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Genetic markers are also extremely useful for estimating the degree of inbreeding (homozygosity) in individual populations (lines), analysing the similarities and differences between populations, and determining the genetic distance between them. Estimation of genetic distance and the selection of breeds with high genetic variation can be used to preserve biological diversity in farm animals. Assessment of genetic variation between populations (lines) is helpful in choosing the optimal crossbreeding variant and obtaining the maximum heterosis effect, manifested mainly as improvement in the performance traits of animals. Characterization of selected genetic markers makes it possible to track the impact of selection work on the genetic structure of a given population and how specific genes are eliminated together with elimination of traits that are deleterious in terms of breeding.

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