

European Research Priorities in Farm Animal Genomics

Peter DOVC¹ (✉)
Gregor GORJANC¹
Jernej OGOREVC¹
Sonja PRPAR¹
Mojca SIMCIC¹
Tanja KUNEJ¹
Mario POLI²

Summary

Production of safe and healthy high quality food for human population, preservation of stable rural social economic infrastructure and sustainable use of agricultural land and animal resources are important goals of European livestock sector. Research and innovation in the livestock sector will play an important role in development of more efficient and sustainable strategies for supply of healthy high quality food, reduction of environmental impact of animal production and development of broader genetic base for our livestock species. Research, especially high throughput -omics technologies are therefore important component of the research agenda within the new EU framework programme, Horizon 2020. In the present article we review the most important developments in animal genetics research during the last decade and evaluate some new strategies which should contribute to establishment of excellent science in the European livestock sector.

Key words

genomic selection, animal genetic resources, RNA interference, animal health, integratomics

¹ University of Ljubljana, Biotechnical Faculty, Department of Animal Science, Groblje 3, 1230 Domzale, Slovenia

✉ e-mail: peter.dovc@bf.uni-lj.si

² INTA, Buenos Aires, Argentina

Received: June 14, 2013 | Accepted: July 15, 2013

ACKNOWLEDGEMENTS

The authors acknowledge the financial support of Slovenian Research Agency (ARRS) through the Research programme P4-0220 and Slovene-Argentinian bilateral grant.

Aim

The livestock sector contributes 130 billion € annually to Europe's economy (Eurostat regional yearbook, 2012) and represents more than 44% of the total agricultural economic activity (Eurostat, 2012). The research and innovation in the livestock sector contributed significantly to the development of efficient, competitive, and environmentally acceptable production systems which offer employment to almost 30 million people all over the continent (Eurostat, 2012). Production of safe and healthy high quality food for human population, preservation of stable rural social economic infrastructure and sustainable use of agricultural land are important goals of European livestock sector. Social development in the last decades introduced in addition to these primary goals of the animal production several new aspects which will play an important role in successful integration of the livestock sector into the new, knowledge based bio economy in Europe. Research and innovation in the livestock sector will play an important role in development of more efficient and sustainable strategies for supply of healthy high quality food, reduction of environmental impact of animal production and development of broad genetic base for our livestock species as proposed in the position paper issued by the Animal Task Force (ATF), the leading body of expertise linking European Technology Platforms and research providers for developing innovation in the livestock sector (Animal Task Force, 2013). The most important pillars represent sustainable and efficient use of animal resources, permanent care for animal health and welfare, new solutions for environmental challenges and contribution to human health. This includes preservation of genetic resources as a reservoir of genetic variants already adapted to different environmental conditions, exploitation of genetic variation in resistance or susceptibility for infectious diseases, selection of robust, healthy animals which will produce healthy food and development of animal models for biomedical applications. In this paper we will focus on strategic goals which can be achieved through basic and translational research in livestock genomics.

Research topics relevant for animal science

Animal genomics revolutionized strategies for genetic improvement of farm animal species. Since the consumers worldwide, but especially in Europe, are not willing to accept genetically modified animals in the food chain, the development of genomic tools in livestock species is focused to diagnostic procedures and identification of genetic mechanisms playing important role in shaping of complex phenotypes. In order to meet the society needs and to support sustainable, smart and inclusive livestock economy the ATF proposed following issues to be addressed in the frame of the new EU framework programme, Horizon 2020:

- Efficient use of resources
- Development of responsible livestock farming systems
- Improvement of human and animal health
- Support of knowledge exchange towards innovation, and
- Identification of opportunities and needs in Excellent Science (Animal Task Force, 2013).

New technologies enabling high throughput genotyping with large number of markers, mainly represented by high density

SNP micro arrays, allow identification of very complex genotypes for a reasonable price, which offer sufficient information about genetic background of complex phenotypes. This information can be used for computation of haplotypes which can be associated with complex traits. For more comprehensive information, technology for deciphering individual genomes is already available (Oti *et al.*, 2008). In addition to genetic blueprint, encrypted in the DNA sequence, more and more attention is focused on epigenetic modifications of the genome, mainly represented by methylation of certain DNA regions and posttranslational modification of histone proteins, which importantly contribute to structural changes in the chromatin and so directly affect gene expression in farm animals (Vanselow *et al.*, 2006) but also in humans (Marquez *et al.*, 2008). Another mechanism of epigenetic regulation of gene expression, relevant to animal production represents RNA interference (Kunej *et al.*, 2010), which plays an important role in the regulation of expression of about one third of eukaryotic genes. Rapid accumulation of different types of genomic information requires more and more sophisticated bioinformatics tools which allow their interpretation and exploitation (Ogorevc *et al.*, 2011).

Animal welfare and environmental impact of animal production represent two important topics related to the development of responsible livestock farming systems. Concerns of European citizens regarding animal welfare have been documented in a number of studies in different European countries (Verbeke and Viaene, 2000; Schloderer, 2003; Bennett and Yee, 2004, European Commission 2005, 2007). As a response to these concerns several policy instruments and animal welfare assurance schemes have been developed (European Commission, 2007; FAO, 2008; FAWC, 2009). However, in spite of the large number of instruments available, there is still a gap in the agri- and food policy literature regarding the decision criteria which instrument should be applied under which conditions. It became clear that "one size fits all" cannot be the effective solution for the plethora of different situations which exist in the EU livestock sector (Ingenbleek *et al.*, 2012). Regarding the environmental impact of the animal production the greenhouse gas (GHG) emissions play an important role. It has been estimated that production of beef and dairy predominantly on grassland, as opposed to intensive grain fed production, together with reduction of production and food waste could contribute up to 60% to the reduction of GHG emissions of all livestock products which was estimated to be between 630 and 863 Mt in EU27 in 2007 (Bellarby *et al.*, 2013). The measurements which could be effective for mitigation of GHG emissions from livestock sector could also contribute to better public health.

In the following sections we will present several examples how these new technologies can contribute to better understanding of genetic architecture of complex traits in animal production.

Genomic selection

Genomic selection represents an advanced form of marker-assisted selection (MAS), where a large number of genetic markers covering the entire genome are exploited in a way that each QTL is in linkage disequilibrium with at least one marker (Meuwissen *et al.*, 2001; Goddard and Hayes, 2007). Sequencing projects in the last years revealed sufficient number of poly-

morphic single nucleotide markers (SNP) which enable complete coverage of the genome and development of high density SNP micro arrays allowed high throughput genotyping of large number of individuals. Genome wide association studies in animals but also in humans revealed that the effects of single QTL on complex traits are small and therefore a large number of QTL would be needed to explain large portion of variation observed in those traits (Hayes and Goddard, 2010). Utilization of the high number of densely spaced genetic markers allows estimation of the breeding value as a sum of the effects of these markers across the entire genome. An example of successful application of this approach in combination with novel bioinformatics tools allowed to explain about 35% of deregressed estimated breeding value for backfat thickness in Canchim beef cattle using a set of only five SNP markers (Barichello Mokry *et al.*, 2013). One of the main advantages of the genomic estimated breeding values (GEBV) obtained using this technology is that they can be obtained early in life, the fact that is extremely important in selection schemes for traits where phenotypic information is available only after several years, e.g. lactation traits. However, in order to precisely estimate the marker effects a relative large reference population with reliable phenotypic records is required (Hayes and Goddard, 2010). In general is assumed that prediction of genome-wide breeding values in genomic selection is achieved exploiting linkage disequilibrium between markers and QTL taking into account genetic relationships. It has been shown that genome-wide prediction of breeding values based only on identity-by-descent genomic relationships within the known pedigree is equally or even more reliable GEBV based on identity-by-state, which implicitly also accounts for genomic relationships that occurred before the known pedigree (Luan *et al.*, 2012). In addition, inclusion of different numbers of generations from the pedigree showed that most of the information in genome-wide breeding value prediction comes from animals with known common ancestors less than four generations back in the pedigree. In spite of the fact that genomic selection based on identity-by-state does not require pedigree data, it does use the available pedigree structure. This may explain the fact that prediction equations derived for one breed may not predict accurate genome-wide breeding values when applied to other breeds, due to different family structures in different breeds (Luan *et al.*, 2012). However, genomic selection based on large number of markers and reliable phenotypic data for the reference population offers significant improvement in reliable prediction of breeding values which can significantly speed up the selection process.

In order to increase statistical power through the larger number of individuals included in the analysis and to allow international comparison of estimated breeding values (EBV) for production traits in cattle, the International Bull Evaluation Service, Interbull, was established in 1983. Currently, the international breeding values of dairy bulls are estimated three times a year and enable international comparison of genetic merit (Tyrisavä *et al.*, 2011). Recently, the new high throughput genotyping strategies required for genomic selection have been adopted successfully. Using this technology in the frame of the international genomic evaluation program, coordinated by Intebull, the genome wide association study for production traits and stature in the Brown Swiss cattle breed has been performed (Guo *et al.*, 2012). In this study, the major signals for milk production traits, stature and

body depth were identified on BTA 25, whereas the strongest signal for somatic cell score was found on BTA 25. In addition to the permanent growing amount of genomic data included in the evaluation, the special attention has to be focused on definition of phenotypic traits and statistical evaluation of the data set. In spite of the fact that the same traits are recorded in different countries, there are still differences in trait definition which cause changes in across-country genetic correlations. An example for that are across-country genetic correlations for overall conformation score (OCS) in Holstein bulls which are affected by changes in trait definition in different countries (Battagin *et al.*, 2013). In order to facilitate the estimation process for multiple-trait across-country evaluation (MACE), the use of principal component (PC) and factor analysis (FA) approaches were proposed to prevent over-parameterization of the model as well as unproportional increase of the sampling variance (Tyrisavä *et al.*, 2011). The availability of genomic information enabled the national preselection of young bulls, which is widely used in dairy breeding schemes. However, this approach causes bias in estimations of national breeding values and has to be accounted for in genetic evaluation models (Patry *et al.*, 2013).

RNA interference (RNAi)

The discovery of RNA interference (RNAi) in *C. elegans* (Fire *et al.*, 1998) opened a new avenue in understanding the complexity of mechanisms involved in the regulation of animal gene expression. Even if the RNAi was first considered rather as an exception than a common mechanism, a number of studies proved in a relatively short period of time, that RNAi is a wide spread mechanism involved in fine tuning of expression of a considerable number of eukaryotic genes. Among different forms of RNAi found in animals, the microRNA (miRNA) plays a central role. In different animal genomes several hundred or even more than thousand regions coding for miRNAs were identified. The variable expression of miRNAs and polymorphisms within the miRNA target sites, which are mainly situated at 3'-end of mRNA transcripts, represent a regulatory system where specific miRNAs have a number of targets leading to pleiotropic effect of the was majority of miRNAs. The research in the last few years showed that miRNAs and miRNA targets play an important role in the regulation of genes which affect important traits in livestock species (Zorc *et al.*, 2012; Jevšinek Skok *et al.*, in press). The most frequently studied mode of action for RNAi is translational inhibition and post-transcriptional silencing of mRNA via RISC complex. A nice example for phenotypic effect of mutated miRNA target site on muscle development is myostatin expression and consequently muscle development in Texel sheep (Clop *et al.*, 2006) which are known for exceptional meatiness. A whole genome scan for regions being associated with this trait in a Romanov x Texel F2 population has been performed and a QTL with major effect on muscle mass was detected at OAR 2 in the region where myostatin gene (GDF8) is located. A Texel allele, characterized by a G to A transition in the 3'-UTR region within the target site for two miRNAs, miR-1 and miR 206, both expressed in skeletal muscle causes translational inhibition of the GDF8 and muscular hypertrophy in Texel sheep. This finding clearly demonstrates the relevance of RNAi for shaping phenotypic traits. Although the miRNAs found in different mammalian species differ in number and their location in the genome,

the comparative analysis can help to identify regulatory pathways in which miRNAs are involved. Especially intragenic miRNA genes seem to be evolutionary conserved and our recent study revealed that approximately half of currently annotated miRNA genes resided within host genes in human, mouse and chicken (Godnic *et al.*, 2013). This miRNAs are involved in either synergistic or antagonistic mode of host gene regulation. We found 27 miRNA-host gene pairs with conserved co-localization, co-expression and potential co-regulation. This is the first step in functional annotation of intragenic miRNAs involved in health and disease (Godnic *et al.*, 2013).

Comparative genomics and animal health

Breeding of healthy animals which produce healthy food is one of the central goals in European animal production which indirectly includes also important animal welfare issues. One of the most important problems in animal health is mastitis which causes about 160 EUR additional costs per cow per year (Ogorevc, 2011). In addition to preventive measures as hygiene and proper milking technology, the improvement of genetic base of animal populations seems to be the most effective strategy for prevention of mastitis. Implementation of different genomic tools for study of host – pathogen interactions during mastitis was supported by EU Network of excellence – EADGENE, promoting new research resources and collaboration between European research centres (<http://www.eadgene.info/>). The experimental approach includes search for favourable alleles at candidate loci, development of cell models and comparative genomic approach for better understanding of innate immunity of the mammary gland. Transcriptomic studies *in vivo* revealed important response mechanisms which regulate udder response to acute infection. One of the most prominent changes in the infected mammary gland is abrupt shut-down of α S1-casein synthesis during acute mastitis, which is due to DNA-remethylation around a STAT5-binding enhancer in the α S1-casein promoter (Vanselow *et al.*, 2006). The experiments *in vitro* on the primary culture of the mammary gland epithelial cells revealed strong activation of chemokines, interleukins, beta-defensins, serum amyloid A and haptoglobin (Günther *et al.*, 2009). The complexity of the host response was also studied in goat primary mammary epithelial cells (Prpar *et al.*, 2012) triggering differential expression of 1562 genes 24 hours after infection. The use of mammary gland cells of different species gives us the opportunity to compare the immune response in most important dairy species and to identify common pathways of immune response in the mammary gland. Using this information, new breeding strategies towards better udder health could be designed.

Bioinformatics – integratomics

The common characteristics of all –omics approaches is an enormous flood of data, which are often fragmented and difficult to compare. To overcome this problem, different bioinformatics approaches have been developed which enable integration of data produced using different approaches. Some complex phenotypes like cryptorchidism were extensively studied using classical approaches but there were no clear candidate genes responsible for this congenital disorder affecting male fertility in man and many other mammalian species. Recently, we demonstrated the power of integratomics approach combining comparative inter

species approach with integration of different types of data. We collected literature data from seven mammalian species, organized it in different gene networks and used them finally for gene map based integration (Cannistraci *et al.*, 2013), resulting in the cryptorchidism gene atlas (<http://www.integratomics-time.com/cryptorchidism>). The analysis revealed 43 novel network-predicted candidate genes. Interestingly, joint pathway analysis revealed the central role of the muscle-contraction pathway in cryptorchidism, but also genomic associations with cardiomyopathy pathways in RASopathies, giving an example for the explanation for co-presence of diverse and apparently unrelated clinical syndromes in some syndromes (Cannistraci *et al.*, 2013).

High throughput phenotyping

A serious limitation for the use of vast amount of genomic data for selection purposes is lack of phenotypic counterpart in the complex datasets. Traditionally, the conventional phenotypic data, such as milk yield, protein and fat content, daily weight gain and feed conversion are used for identification of associations with genomic markers. However, dissecting traditional complex traits into better defined traits with clearer physiological background requires a new type of phenotypic data, which may be difficult and expensive to obtain. There are several examples in plant breeding where researchers try to introduce high throughput phenotyping techniques based on near-infrared spectroscopy (NIRS) and plant spectral reflectance as a high-throughput field phenotyping methods for complex traits (Cabrera-Bosquet *et al.*, 2012). In animals, the most promising development goes in the direction of integrated biosensors for collection of a large number of physiological data on line. This would enable generation of large data sets which would describe complex phenotypic differences among animals and allow search for correlations with genetic architecture of complex traits. In addition to physiological parameters, the quality of animal products can give valuable information about the genetic merit of livestock species. High throughput automated procedures for evaluation of animal products can contribute important information for complex description of the phenotype. Determination of beef quality parameters using time-domain nuclear magnetic resonance is an example for this (Verbi Pereira *et al.*, 2013). Automatisations of working routines in animal production also offers a number of opportunities to collect data about physiological parameters (milking speed) and composition of animal products (protein, fat content in milk) as well as health status (somatic cell count) of the animal (Innocente and Biasutti, 2012).

Preservation of local genetic resources

A large number of local livestock breeds are endangered or even disappearing worldwide and also in Europe there are numerous local breeds which are close to extinction. Therefore, in different strategies dealing with preservation of animal genetic resources, some prioritization is needed. Due to unreliable records about some breeds and incomplete pedigree data, it is difficult to judge about the genetic uniqueness of some breeds and their genetic potential. Breeds, adapted to the very extreme environmental conditions, are certainly good candidates to be preserved, but breeds adapted to more moderate conditions might not be so attractive. In such situations the understanding of breed history and genetic set up of the breed may be important

criteria in decision making process. A very valuable contribution in the field of description of animal genetic resources has been made at international level with high density genotyping of a large collection of European cattle breeds which allowed establishment of relationships among breeds and revealed historical interconnections among them (Feliús *et al.*, 2011). An example where genetic analysis nicely completed relative scarce data about the breed is Slovenian autochthonous cattle breed Cika. Using molecular marker approach and STRUCTURE analysis we could confirm a unique genetic identity of Cika in spite of Pinzgauer admixture in the Cika population (Simcic *et al.*, 2013). Such studies can help in decision making and support preservation of valuable, unique genetic resources which may be of great importance for further selection work. In this context it is important to have an overview about phenotypic variation among breeds for certain traits, as already shown for body size and carcass characteristics, in order to identify valuable genetic resources (Alberti *et al.*, 2008). Similar as for cattle breeds application of genetic markers for establishing genetic relationships among pig breeds was performed on the global scale using X chromosome single nucleotide polymorphisms (Burgos-Paz *et al.*, 2012). In relation to product quality, considerable contribution has been made through the elucidation of single gene effects on carcass quality and fat deposition in pigs (Fontanesi *et al.*, 2010; Fontanesi & Russo, 2013) and in cattle (Jevšinek Skok *et al.*, 2011). Certainly, for protection of valuable genetic resources it is inevitable to recognize the uniqueness of their products and to introduce analytical methods which will prevent marketing frauds, as recently demonstrated with the water buffalo mozzarella and other buffalo dairy products (Sakaridis *et al.*, 2013).

Conclusion

A novel technologies, including different -omics approaches as well as bioinformatics and powerful statistical tools allow for the first time that animal genomics can be considered as a “big science”, mainly due to the enormous amount of data available but also due to structured animal populations and powerful tools which are available for data generation and interpretation. Therefore, the stakeholder in the field have to make sure that reasonable financial support will be available at national as well as at international level in order to allow European livestock sector to remain competitive player on the global market and to integrate more efficiently research excellence which already exists in Europe into development strategies and technological progress.

References

- Alberti P., Panea B., Sañudo C., Olleta J.L., Ripoli G., Ertbjerg, P., Christensen M., Gigli S., Failla S., Concetti S., Hocquette J.F., Jailler R., Rudel S., Renand G., Nute G.R., Richardson R.I., Williams J.L. (2008). Live weight, body size and carcass characteristics of young bulls of fifteen European breeds. *Livestock Science* 114:19-30
- Animal Task Force (2013): www.animaltaskforce.eu
- Baricello Mokry F., Higa R.H., de Alvarenga Mudau M., Oliveira de Lima A., Conceição Meirelles S.L., Barbosa da Silva M.V.G., Cardoso F. F., de Oliveira M.M., Urbinati I., Meo Niciura S.C., Tullio R.R., de Alencar M.M., de Almeida Regitano L.C. (2013). Genome-wide association study for backfat thickness in Canchim beef cattle using Random Forest approach. *BMC Genetics* 14:47-57
- Battagin M., Forabosco F., Penasa M., Cassandro M. (2013). Cluster analysis on across-country genetic correlations for overall type traits and body condition score of Holstein bulls. *Livestock Science* 151:108-114
- Belarby J., Torado R., Leip A., Weiss F., Lesschen J.P., Smith P. (2013). Livestock greenhouse gas emissions and mitigation potential in Europe. *Global Change Biology* 19:3-18
- Bennett R.M., Yee W. (2004). UK review of Literature on Consumer Food Animal Welfare Concerns and Requirements for Animal Welfare and Welfare Friendly Products. Welfare Quality WP1.1, Consumers Literature Review Country Report UK: <http://www.welfarequality.net>
- Burgos-Paz W., Souza C.A., Castello A., Mercade A., Okumura N., Sheremet'eva I.N., Huang L.S., Cho I.C., Paiva S.R., Ramos-Onsins S., Perez-Enciso M. (2012). Worldwide genetic relationships of pigs as inferred from X chromosome SNPs. *Animal Genetics* 44:130-138
- Cabrera-Bosquet L., Crossa J., von Zitzewitz J., Serret M.D., Araus J.L. (2012). High-throughput phenotyping and genomic selection: the frontiers of crop breeding converge. *J Integr Plant Biol* 54(5):312-20
- Cannistraci C.V., Ogorevc J., Zorc M., Ravasi T., Dovc P., Kunej T. (2013). Pivotal role of the muscle-contraction pathway in cryptorchidism with cardiomyopathy pathways in RASopathies. *BMC Medical Genomics* 6:5
- Clop A., Marcq F., Takeda H., Pirotton D., Tordoir X., Bibé B., Bouix J., Caumont F., Elsen J.-M., Eychenne F., Larzul C., Laville E., Meish F., Milenkovic D., Tobin J., Charlier C., Georges M. (2006). A mutation creating a potential illegitimate microRNA target site in the myostatin gene affects muscularity in sheep. *Nature Genetics* 38, 813 – 818
- EADGENE - <http://www.eadgene.info/>
- European Commission (2005). Attitudes of EU Citizens towards Farmed Animals. Special Eurobarometer 229, Brussels, Belgium
- European Commission (2007). Attitudes of EU Citizens towards Animal Welfare. Special Eurobarometer 270/Wave 66.1, Brussels, Belgium
- Eurostat (2012):http://epp.eurostat.ec.europa.eu/portal/page/portal/agriculture/data/main_tables
- Eurostat regional yearbook (2012). electronic version:
- Eurostat regional yearbook, (2012). http://epp.eurostat.ec.europa.eu/cache/ITY_OFFPUB/KS-HA-12-001-09/EN/KS-HA-12-001-09-EN.PDF
- FAO (2008). Capacity Building to Implement Good Animal Welfare Practices. Report of the FAO Expert Meeting, FAO, Rome, Italy
- FAWC (2009). Opinion on Policy Instruments for Protecting and Improving Farm Animal Welfare. Farm Animal Welfare Council, London, UK
- Fire A., Xu S., Montgomery M.K., Kostas S.A., Driver S.E., Mello C.C. (1998). Potent and specific genetic interference by double-stranded RNA in *Caenorhabditis elegans*. *Nature* 391:806-811
- Fontanesi L., Scotti E., Buttazzoni L., Dall'Olio S., Bagnato A., Lo Fiego D.P., Davoli R., Russo V. (2010). Confirmed association between a single nucleotide polymorphism in the FTO gene and obesity-related traits in heavy pigs. *Mol Biol Rep* 37, 461-466
- Fontanesi L., Russo V. (2013). Nucleotide variability and haplotype heterogeneity at the porcine fat mass and obesity-associated (FTO) gene. *Anim Genet* 44, 96-100
- Goddard M.E., Hayes B.J. (2007). Genomic selection. *J. Anim Breed Genet* 124(6):323-30
- Godnic I., Zorc M., Jevšinek Skok D., Calin G.A., Horvat S., Dovc P., Kovac M., Kunej T. (2013). Genome-wide and species-wide in silico screening for intragenic microRNAs in human, mouse and chicken. *PLoS one* 8(6):1-14, e-65165

- Gunther J., Koczan D., Yang W., Nurnberg G., Reipsilber D., Schuberth H.J., Park Z., Maqbool N., Molenaar A., Seyfert H-M. (2009). Assessment of the immune capacity of mammary epithelial cells: comparison with mammary tissue after challenge with *Escherichia coli*. *Veterinary Research* 40(4) 31
- Guo J., Jorjani H., Carlborg Ö. (2012). A genome-wide association study using international breeding-evaluation data identifies major loci affecting production traits and stature in the Brown Swiss cattle breed. *BMC Genetics* 13: 82-91
- Hayes B.J., Goddard M.E. (2010). Genome-wide association and genomic selection in animal breeding. *Genome* 53(11):876-83
- Ingenbleek P.T.M., Immink V.M., Spoolder H.A.M., Bokma M.H., Keeling L.J. (2012). EU animal welfare policy: Developing a comprehensive policy framework. *Food Policy* 37:690-699
- Innocente N., Biasutti M. (2013). Automatic milking systems in the Protected Designation of Origin Montasio cheese production chain: Effects on milk and cheese quality. *J. Dairy Sci.* 96:740-751
- Jevšinek Skok D., Godnič I., Zorc M., Horvat S., Dovc P., Kovac M., Kunej T. (2013). Genome-wide in silico screening for microRNA genetic variability in livestock species. *Animal Genetics* in press
- Jevšinek Skok D., Kunej T., Rencelj A., Razpet A., Dovc P., Žgur S., Petric N., Planinc M., Malovrh Š., Kovac M., Horvat S. (2011). Use of SNP markers within the fat mass and obesity-associated (FTO) gene to verify pedigrees and determine haplotypes in paternal half-sib families in Slovenian Simmental cattle. *Agriculturae Conspectus Scientificus* 76(3, 4):333-336
- Kunej T., Jevšinek Skok D., Horvat S., Dovc P., Jiang Z. (2010). The glypican 3-hosted murine Mir717 gene : sequence conservation, seed region polymorphisms and putative targets. *International journal of biological sciences* 6 (7):769-772
- Felius M, Koolmees P.A., Theunissen B., European Cattle Genetic Diversity Consortium, Johannes A. Lenstra (2011): On the breeds of cattle - historic and current classifications. *Diversity* 3(4):660-692
- Luan T., Wooliams J.A., Ødegård J., Dolezal M., Roman-Ponce S.I., Bagnato A., Meuwissen T.H.E. (2012). The importance of identity-by-state information for the accuracy of genomic selection. *Genet Sel Evol* 44(1): 28-35
- Marques C. J.; Costa P.; Vaz B.; Carvalho F.; Fernandes S.; Barros A.; Sousa M. 2008. Abnormal methylation of imprinted genes in human sperm is associated with oligozoospermia. *Molecular Human Reproduction*, 14:67-73
- Meuwissen T.H.E., Hayes B.J., Goddard M.E. (2001). Prediction of total genetic value using genome-wide dense marker maps. *Genetics* 157:1819-1829
- Ogorevc J. (2011): Identification of candidate genes for mastitis resistance using bioinformatics and transcriptomics. PhD thesis, University of Ljubljana, pp. 147.
- Ogorevc J., Kunej T., Razpet A., Dovc P. (2009). Database of cattle candidate genes and genetic markers for milk production and mastitis. *Anim. genet.* 40:832-851
- Oti M., Huynen M.A., Brunner H.G. (2008). Phenome connections. *Trends in genetics* 24(3):103-106
- Patry C., Jorjani H., Ducrocq V. (2013). Effects of a national genomic preselection on the international genetic evaluations. *J. Dairy Sci.* 96:3272-3284
- Prpar S., Martignani E., Dovc P., Barata M. (2012). Identification of goat mammary stem/progenitor cells. *Biol. reprod.*, 86(4): 1-7
- Sakaridis I., Ganopoulos I., Argiriou A., Tsafaris A. (2013). High resolution melting analysis for quantitative detection of bovine milk in pure water buffalo mozzarella and other buffalo dairy products. *International Dairy Journal* 28:32-35
- Schloderer J. (2003). Quality of free range pork: What consumers want. In: MAPP Conference, 8 October, 2003
- Simcic M., Lenstra J.A., Baumung R., Dovc P., Cepon M., Kompan D. (2013). On the origin of the Slovenian Cika cattle. *J. Anim. Breed. Genet.* 1-9
- Tyrisevā A-M., Meyer K., Fikse W.F., Ducrocq V., Jakobsen J., Lidauer M.H., Mäntysaari E.A. (2011). Principal component and factor analytic models in international sire evaluation. *Genetics Selection Evolution* 43: 33-42
- Vanselow J., Yang W., Herrmann J., Zerbe H., Schuberth H.J., Petzl W., Tomek W., Seyfert H-M. (2006). DNA-methylation around a STAT5-binding enhancer in the αS1-casein promoter is associated with abrupt shut-down of αS1-casein synthesis during acute mastitis. *J. Mol. Endocrinol.* 37:463-477
- Verbeke W.A.J., Viaene J. (2000). Ethical challenges for livestock production: meeting consumer concerns about meat safety and animal welfare. *J. Agric. Environ. Ethics* 12:141-151
- Verbi Pereira F.M., Bertelli Pflanzler S., Gomig T., Lugnani Gomes C., de Felicio P.E., Colnago L.A. (2013). Fast determination of beef quality parameters with time-domain nuclear magnetic resonance spectroscopy and chemometrics. *Talanta* 108:88-91
- Zorc M., Jevšinek Skok D., Godnič I., Calin G.A., Horvat S., Jiang Z., Dovc P., Kunej T. (2012). Catalog of MicroRNA seed polymorphisms in vertebrates. *PLoS one* 7(1):1-8, e30737