

Poznań, 30.08.2022

dr hab. Marcin Pszczoła

Assistant Professor

Katedra Genetyki i Podstaw Hodowli Zwierząt

Wydział Medycyny Weterynaryjnej i Nauk o Zwierzętach

Uniwersytet Przyrodniczy w Poznaniu

**Review of the doctoral thesis of MSc Eng. Barbara Elżbieta Kosińska-Selbli entitled:  
'The analysis of genetic structure of selected dairy cattle breeds'.**

The doctoral thesis presented for the review concerns the genetic analyses of different cattle breeds. It is a compilation of three scientific papers published in well-known journals. One paper is a review article published in *Animal* in 2020 and has already received eight citations. Two other papers are original research papers published in *Frontiers in Genetics and Livestock Science*. The research was financed with funds from two projects, one from local funds through National Science Centre and the second from EU funds within ERA-NET-SusAn project. The reviewed thesis was supervised by prof. Joanna Szyda and co-supervised by Dr. Morten Kargo.

The dissertation is written in English and has a classical set-up. It comprises 72 pages, of which 16 contain literature containing 101 citations. First, the candidate introduces the general introduction, after which the main information from the papers is provided, after which a general summary of the thesis is given. The papers included in the thesis were subject to the peer review process. Therefore, the methodological aspects of the study were already checked by the author, co-authors, reviewers, and editors of the scientific journals. Now they were also read by myself while reviewing the doctoral thesis. I have found no major flaws in the methods applied in the analyses presented in the dissertation. Nonetheless, I will give my personal opinion on the presented papers and will raise some discussion points.

The thesis was entitled "The analysis of the genetic structure of selected dairy cattle breeds" At first, the candidate introduces a reader to the topic by presenting a short description of cattle, including the domestication process and history of genome analyses, and stresses

the importance of genetic diversity in cattle breeds, followed by a short description of cattle genome assembly and QTL database and genomic variability described by CNVs. Afterward, the author presents information on the purpose of breeding goals and economic values. At the end of the general introduction, the candidate presents the scientific goal of the doctoral thesis as an exploration of genomic variation observed in various cattle breeds. The goal was achieved by reviewing the literature on lncRNA, analyzing genomic data in combination with phenotypic information on hoof and leg health, and estimating economic values for functional traits of Polish Holstein-Friesian and Red Polish cattle.

The goal of the first research paper was to describe genome variability based on long non-coding fragments of RNA (lncRNA). This review paper aimed to understand better the function of long non-coding RNA in cattle based on 15 studies published between 2017 and 2019. The performed review revealed that lncRNA plays an essential role in regulating gene expression related to milk and beef production and other characteristics, some of which may have economic importance. It is worth noting that the review summarized in this chapter is only a more extensive article in which information on poultry and pigs was also included. The chapter ends with the statement that obtaining more knowledge on the topic is essential. I would appreciate a comment on how such information can be used in breeding practice.

The following paper presents research on the exploration of genetic heterogeneity in the incidence of the hoof disorders recorded in Austrian Fleckvieh and Braunvieh cattle. In this study, the authors perform a genome-wide association study for a total number of leg disorder incidences recorded until 100 days in milk. The GWAS study, performed separately for each breed, was followed by analyzing allelic heterogeneity using a window of 50 neighboring SNPs. The authors conclude that the analyzed dataset did not allow to confirm or exclude the hypothesis that genetic heterogeneity in the incidence of hoof disorders exists and gave a possible explanation for this fact. I would like to explore this issue further. Hoof health is a complex phenotype. It is collected by veterinarians, hoof trimmers, farmers, or even with automated detectors. I presume this can be a source of considerable variation in the obtained phenotypes. This was reflected in the model used to pre-correct phenotypes. I wondered what was the reason for selecting such a complicated and specific trait to work with when the dissertation title suggests more broad interest of the study. I can imagine that more standardized traits could serve a similar purpose of assessing the heterogeneity between breeds and possibly would allow for gathering a more extensive data set for analysis.

For calculating genomic relationship matrices, a method used by Yang et al. 2011 who derived an equation equivalent to vanRaden 2008 was used. In these calculations, allele frequencies estimated from data or assumed allele frequencies could be used. Breeds can differ concerning allele frequencies due to differences in breeding strategies and history. It is not clearly stated in the dissertation nor the full paper of Kosińska-Selbi et al. 2020b what approach was used in this study. This information provided a justification for the selected approach and could provide a better understanding of the obtained results.

The authors decided to use a window of 50 SNPs in the analysis. They then indicated that: *"it was not possible to differentiate between breeds based on SNP genotypes corresponding to the 50-SNP windows"* Have the authors considered different window sizes, and what would be a possible implication of such an approach on the outcomes of the analyzes?

The authors discuss no overlap of the GWAS outcomes between breeds and state that this is common finding in other studies. Were the cited studies analyzing similar phenotypes? Is this the general rule for different hoof health traits? The analyzed trait is a complex trait collecting number of different hoof problems of which each can have a different genetic background. The full paper describes, in short, the differences in frequency of the number of incidences of hoof disorders between breeds. However, it does not compare the contribution of particular disorders in the total count. This information could enable a better understanding of the lack of overlap between GWAS results for the two analyzed breeds.

The last contribution describes using a bio-economic model to simulate herd structure and economic profit of two breeds. The paper itself is an interesting contribution. However, I do not see a clear connection between this paper and the dissertation's general topic. Defining a breeding goal could indeed lead to more or less genetic diversity. However, in such cases, genetic diversity is usually included in the breeding program with a mating strategy that minimizes the loss of genetic diversity due to selection (e.g., optimal contribution). This topic is not included in the paper.

I appreciate the efforts to construct economic indices for selected breeds and pointing out some unique characteristics of local Polish breeds that could be used while breeding for more resilient animals. I wondered what justified using the unknown parameters on the health of Nordic cattle. I understand that the reason could be the availability of the information. However, because of this fact and selection for health traits in the Nordic countries, our Polish parameters can be substantially different. Also, parameters taken from German Holsteins could lead to results

further away from the Polish situation. Have the authors assessed the possible uncertainties in the obtained results? Also, for several years, the Polish Federation of Cattle Breeders and Dairy Farmers has published results of an economic index for dairy cows, have authors compared the obtained results to the published official economic index?

Another minor issue is breed definition. It is unclear to me which two breeds were used in the study. Polish Federation for Cattle Breeders and Dairy Farmers indicates the Polish Holstein-Friesian breed with two color varieties, the Red Polish breed, which is a one-colored breed, and Polish Red-White breed. The information on the breed used could be more specific.

All the issues mentioned above are more discussion points that would enable a better understanding of the presented dissertation and do not diminish the value of the work included in the doctoral thesis. The doctoral dissertation by Ms. Barbara Elżbieta Kosińska-Selbi proves good knowledge of techniques used to analyze genomic variability measured with different tools. It contributes to the current state of knowledge in the Discipline of Animal Science and Fisheries with valuable information that could potentially be utilized in breeding practices in Poland and other countries. The candidate demonstrated her ability to contribute to the scientific process by producing several scientific papers published in well-respected journals. During their Ph.D., the candidate, next to the research included in the thesis, contributed to other research articles, which shows her potential for collaborative work, which is an essential ability in the field of animal sciences. I acknowledge that two out of three papers included international co-authors. Given the above, I state that the reviewed dissertation complies with Act on academic degrees and academic title and degrees and title in art (art. 13 Ustawy z dnia 14.03.2003 r. o stopniach naukowych i tytule naukowym oraz o stopniach i tytule w zakresie sztuki (Dz. U. 2017 poz. 1789) w związku z art. 179 ust. 2 oraz art. 179 ust. 3 pkt 2b Ustawy z dnia 3 lipca 2018 r. Przepisy wprowadzające ustawę – Prawo o szkolnictwie wyższym i nauce (Dz. U. 2018 poz. 1669 ze zm.), and therefore, I recommend admitting Ms. Barbara Elżbieta Kosińska-Selbi to public defense of their doctoral thesis.

Marcin Tszepiła