Abstract Submission Form

Title (Mr./Mrs/Dr./Prof.)	Prof.
Presenting author	Peter Dovc
Institute	Institute/company: University of Ljubljana, Biotechnical Faculty
	Adress: Jamnikarjeva 101
	ZIP/Postal code: 1000 City: Ljubljana
	Country: Slovenia
Insert all authors and institutions Zorc M. (1), Dovc P. (1) (1) UL, Biotechnical faculty, Slovenia	
Preferred presentation	Oral
Preferred session	Session 9: WG DNA – Genomic's impact on Livestock Sustainability
Email of corresponding author	peter.dovc@bf.uni-lj.si
Title of your paper	Milk cell transcriptome opens a new dimension in the mammary gland biology research

Insert ABSTRACT text

The mammary gland, a relatively recent acquisition of mammalian evolution, is essential for successful reproduction, as it provides nourishment and immune protection for the neonate. The regenerative potential of the mammary gland is crucial for its cyclical nature, which is regulated by hormones and growth factors. A consequence of the complex function of the mammary gland and the intensive milk secretion is also the presence of somatic cells in the milk. The most important fractions of somatic cells in milk are epithelial cells, lymphocytes, polymorphonuclear neutrophils (PMN) and macrophages. While the somatic cell count (SCC), which is often used as a marker for udder health, only indicates the cumulative number of somatic cells in the milk, the differential somatic cell count (DSCC) allows differentiation between two cell groups: PMN and lymphocytes and macrophages. Therefore, DSCC is an important step in understanding the dynamics of the somatic cell population in the mammary gland during lactation and infections. In cattle and sheep, the epithelial cell fraction accounts for only a relatively small proportion of somatic cells in milk, whereas in pig milk, similar to human milk, epithelial cells are the predominant cell type in milk. Mammary stem cells can self-renew and differentiate into different cell types during the development and renewal of the mammary gland.

In order to gain insight into the molecular processes in the lactating mammary gland at the cellular level, access to appropriate biological material is required. Taking biopsies from the mammary gland is one



option, but due to their invasive nature, researchers were looking for alternatives to biopsies. Comparison of different RNA sources, including biopsies, laser microdissected mammary epithelial cells, somatic mammary cells, milk fat globules and antibody-captured mammary epithelial cells, showed that isolation of total RNA directly from somatic mammary cells released into milk during lactation is an effective alternative to mammary gland tissue biopsies. The sequencing of total RNA from bovine mammary cells at different stages of lactation revealed the expression of more than 17 thousand genes. Regardless of the lactation stage, about 9000 genes showed ubiquitous expression. Genes coding for caseins, whey proteins and enzymes of the lactose synthesis pathway showed higher expression in early lactation, and the majority of genes of the fat metabolism pathway also had high expression in transition and peak lactation.

Here we report the application of scRNA-seq to elucidate the cell type repertoire in bovine milk based on transcriptomic differences between different cell clusters. We identified 21 cell clusters and classified them as T cells (CD8+, CD4+), neutrophils, progenitor cells, monocytes, mast cells, macrophages, B cells, NK cells, dendritic cells, monocytes, luminal cells and luminal progenitor cells. The cells responsible for milk production were identified in three clusters, and clusters expressing epithelial markers were identified as progenitor cells The results of our study will contribute to a better understanding of the complex processes in the mammary gland, including tissue remodeling and involution. Single-cell RNA sequencing opens a new horizon for the documentation of cell type-specific expression profiles in the mammary gland and even the possibility to determine different cell types based on cell type-specific transcriptomic profiles.

Enter keywords

milk cell, single cell RNA sequencing, immune response, progenitor cells, tissue renewal