User-friendly image analysis pipeline for analysing granulosa cell line KGN

Bachelor thesis

Student: Eva Katarina Tambets
Student code: 213040LAAB
Supervisors: Agne Velthut-Meikas,
Department of Chemistry and Biotechnology, Associate professor
Simona Bartkova,
Department of Chemistry and Biotechnology, Researcher
Study program: Applied chemistry and gene technology

Tallinn 2024

User-friendly image analysis pipeline for analysing granulosa cell line KGN

Eva Katarina Tambets

The ovary is a complex structure that contains germ cells surrounded by somatic cells and its primary function is to support various stages of female reproductive life including pregnancy. Disruptions in the ovary can lead to many clinical syndromes including polycystic ovarian syndrome and premature ovarian failure, which can result in infertility. Infertility is a health issue affecting a large number of people globally and the prevalence of reproductive health problems is on the rise. Ovarian cells have an essential role in female fertility and by single-cell analysis, it has been determined that the somatic cell types in the ovary are more heterogeneous than previously anticipated.

The aim of this bachelor thesis was to develop a method for studying cell heterogeneity with a reduced cost using image analysis software. First it was shown that KGN cells express the protein markers chosen for this study. Markers used in this study were COL3A1, HSD17B1, FSHR and IGFBP5. Then a software CellProfiler was used to construct a pipeline for assessing protein gene expression in KGN cells with the aim to automatically evaluate cell population protein marker dependence on time. It was established that the proportion of positively stained cells changes between time points, however, only the decrease in FSHR expression was statistically significant. An additional pipeline was constructed in cooperation of softwares Ilastik and CellProfiler to automatically evaluate whether cell area changes in time. While the change of cell area over time was observed by eye, image analysis with the pipeline for automatic evaluation of cell area did not determine a statistically significant difference.

Obtained results suggest that image analysis softwares CellProfiler and Ilastik are suitable for image analysis, specifically for cells. The developed pipeline for automatic analysis of cell population protein marker dependence on time can be potentially used in further studies by being applied on the images of primary cell culture samples to study their heterogeneity. Additionally, it was established that markers chosen for this study have the potential to be incorporated in detecting somatic cell types by an immunocytochemistry-based workflow.