Fertility disorders are observed in cattle breeding, since selection of animals for breeding traits significantly decreased fertility parameters. Also in human medicine infertility is a world-wide problem, which affects 8 to 12% of reproductive-aged couples. Although male infertility contributes to more than half of all cases of global infertility, in many communities this problem is mainly a woman's burden. Female fertility status is a complex trait combining the effect of numerous factors, such as nutrition, physiological status etc. All these parameters may influence not only the global metabolic processes within the female, but also affect oocyte and embryo quality via the maternal disorders of fatty acids or glucose metabolism. Studies show that metabolic disorders of fatty acids and glucose metabolism play crucial role in female health formation and furthermore have a strong impact on fertility. It is observed in human (obese or diabetic women) but also in other species e.g. cattle (negative energy balance of milking cows). The main product of lipid and glucose metabolism is energy, which is necessary to proceed the total metabolism of the cell. Within the oocyte the main origin of energy is glucose, however the literature describes a possible balance between these two pathways in order to keep the cell in a proper homeostasis. Therefore the main goal of the proposed project is to characterize the energy metabolism of bovine oocyte during in vitro maturation, with a special impact on glucose and fatty acids changes.

Within the proposed project it is planned to block selectively pathways of either glucose or fatty acid metabolism in order to demonstrate how these changes influence the quality of the oocyte and further embryo. There are planned control group and 4 experimental groups, which will differ with regard to the supplementation factor blocking a selective metabolic pathway [1 – control group, 2 – maturation under inhibition of glycolysis (a pathway of glucose metabolism), 3 – maturation under inhibition of pentose phosphate pathway (PPP, a pathway of glucose metabolism), 4 – both glycolysis and PPP pathways will be inhibited in order to block almost entire glucose metabolism 5 – maturation under inhibition of β-oxidation of fatty acids].

From each experimental group the following samples will be collected and the following analysis will be performed: (a) individual oocytes (analysis of selected products of glucose or fatty acids metabolic pathways e.g. ATP, NADPH, GSH, reactive oxygen species etc.), (b) cumulus cells originating from individual complexes of cumulus-oocyte (COCs; analysis of mRNA expression of selected genes involved in glucose and fatty acids metabolism; real time PCR) and (c) maturation medium. The metabolomic analysis of individual oocytes, cumulus cells originating from individual COCs as well as maturation media originating from individual wells will give data on e.g. glucose, pyruvate, lactate and fatty acids content (mass spectrometry). There is also planned in vitro production of bovine embryos (in vitro fertilization with the bull of high quality, embryo in vitro culture) after oocytes maturation in experimental groups described above. Time lapse system of on-line imagining of embryo development will be applied to visualize the impact of energy metabolism modifications during maturation on preimplantation embryo development. The final stage of embryo development will be blastocyst (obtained 9 days post fertilization), which will be analyzed with regard to blastocyst rate, morphology, fatty acids content and mRNA expression of genes involved in fatty acids and glucose metabolism.

Authors of the proposed project plan to answer the question, whether bovine oocyte during in vitro maturation to some extend may selectively modify the metabolism of fatty acids and glucose to adapt to environment changes. Within the proposed experiment, modifications of maturation environment take place in maturation medium, however it may be related to in vivo environment, when follicular fluid composition is influenced by the female health. Obtained data may show whether oocytes may adapt to unfavorable environment of female organism and in case of positive observation – how this process takes place.