

Effect of glucose and its metabolism on the physiological state, reproductive potential and lifespan of the yeast *Saccharomyces cerevisiae*

Summary

Glucose is not only the primary source of energy for the cells of most organisms, but also provides carbon skeleton for biosynthesis of many cellular macromolecules. Therefore, glucose plays an important role in the metabolism and maintenance of the proper physiological state of the cell, which in turn determine their proliferative efficiency. This is particularly pronounced in the case of unicellular organisms, including yeast *Saccharomyces cerevisiae*, in which the changes in the availability of glucose determine the way of obtaining energy. This causes that in the case of yeast the influence of glucose on the physiological state of the cell is more direct than in the case of multicellular organisms, which have developed mechanisms that allow to regulate glucose concentration. Currently, glucose is one of the most intensively studied nutrients which influence on the lifespan of a number of organisms. The studies conducted in this area are mainly focused on the reduction of glucose and carbohydrate supply, i.e. calorie restriction. The positive effect of calorie restriction is observed in a wide range of species, including yeast *S. cerevisiae*. Significantly less data can be found in regard to the calorie excess and its effect at the cellular level. Moreover, this problem is practically not performed in studies using the yeast *S. cerevisiae* cells as a research model. However, the analysis of influence of calorie excess and high glucose concentrations on cell physiology seems to be particularly important due to the problems associated with disorders in glucose metabolism.

The aim of the doctoral dissertation was to make a comprehensive analysis of the impact of calorie excess and calorie restriction, obtained by using different concentration of glucose in the culture medium, on the physiological state, reproductive potential and lifespan of the yeast cells. The studies also tried to explain the relationships between different intracellular pathways of glucose utilisation, with particular emphasis on the pathways responsible for the cellular biosynthetic capabilities.

The research were conducted using yeast cells cultured on media containing different glucose concentrations. The wild-type BY4741 yeast strain and three mutant strains isogenic to BY4741, with deletion of selected genes related to glucose metabolism, i.e. $\Delta gpa2$, $\Delta gpr1$ and $\Delta hxk2$, were used for the analysis. This experimental approach and the analysis of the rate

of glucose uptake allowed to test the role of both extracellular and intracellular glucose concentration in the regulation of metabolic changes. The physiological state of the yeast cells was assessed *inter alia* by determination of: growth rate of cell population, cell viability, cell size and cell biomass, functional status of mitochondria, generation of reactive oxygen species, intracellular level of ATP, NADP(H) and enzymatic activity of pentose phosphate pathway enzymes. The reproductive potential and total lifespan of the yeast cells were determined using the micromanipulation method.

It was found that there was a significant relationship between glucose concentration, biosynthetic efficiency, cell size, reproductive potential and total lifespan of the yeast cells. The results indicate that cells possess a specific metabolic trade-off between different pathways of glucose utilisation, which additionally can be actively modulated, among others by the different glucose concentration. However, it is worth emphasizing that intracellular glucose level is more important for cell functionality than its extracellular concentration. This is confirmed by the analysis of the rate of glucose uptake by the yeast cells of strains lacking selected genes related to glucose metabolism. Moreover, it was found that an increase in the glucose concentration in the medium leads to an increase in the level of reactive oxygen species in the yeast cells. However, the obtained results suggest that in this case reactive oxygen species have an extra-mitochondrial source. In addition, high glucose levels corresponding to calorie excess conditions increase the cellular biosynthetic capabilities. On the one hand, this leads to an increase in cell size and cell biomass, but on the other hand, it reduces the reproductive potential and shortens the total lifespan of the yeast cells. The obtained results draw attention to negative impact of calorie excess on the physiological state of the cell, which due to the complexity of intracellular metabolic pathways, need further investigation.