Abstrakt

Respiration State of *Saccharomyces Cerevisiae* Yeast Colonies with Deletions in Genes *CCR4*, *CAF40* and *RIM15*.

The topic of my graduation theses has been the yeast colonies of *Saccharomyces cerevisiae* genus with deletions in genes *CCR4*, *CAF40* and *RIM15*. Our laboratory began interested in study of these genes because of their assumed influence on yeast colony morphology. The Ccr4 and Caf40 proteins form part of Ccr4-NOT complex, which is involved in genetic expression regulation, participates on RNA degradation, a stress response and a cell cycle regulation. The Rim15p influences entering into the stationary phase and adaptation to respiratory growth; *RIM15* gene deletions cause the thermo tolerance loss, sporulation failures, defect of trehalose accumulation and glycogen synthesis.

I studied the effect of *RIM15* gene deletions on colony morphology. Comparing it to the original parental strain I found that rather than the *RIM15* deletions it is the conditions under which the colony is growing that influence the morphology (temperature 28°C or 37°C, media glucose content 0,5%, 1% or 2%, carbon source 1% glucose or 3% glycerol). I found out the adverse effect of the deletion on stressful conditions adaptation, demonstrating itself by delayed or nonexistent ammonia production on media containing glycerol compared to parental strain.

Further, I detected the respiratory metabolism in colonies of parental strain and deletants in CCR4, CAF40 and RIM15 genes using TTC (2, 3, 5 – triphenyltetrazolium chloride). With functional respiratory chain, TTC is reduced by dehydrogenases to red formazane and enables the detection of functionality of the respiratory chain. I found that the powerful respiration can be detected in giant colonies not only on GM media, but also on glucose containing media (though somewhat later), and further that the respiration efficiency decreases in old colonies and also that both the giant colonies and mono-colonies $ccr4\Delta$ a $caf40\Delta$ create sectors with decreased or non-existent respiration.

In the final part of my graduation thesis I identified respiratory deficient (rd) mutants, which exhibit the inability to make use of unfermentable carbon sources and in the media containing limited amount of glucose they grow in the form of small (petit) colonies. Studied strains revealed high percentage of spontaneous rd mutants

getting formed in colonies, with the larger portion of them in less TTC colored areas of colonies.

Key words: yeast, *S. cerevisiae*, *RIM15*, colony morphology, TTC, respiratory deficient mutants, respiratory metabolism, alkalinization.

Klíčová slova: kvasinka, *S. cerevisiae*, *RIM15*, morfologie kolonií, TTC, respirační metabolizmus, respiračně deficientní mutanti, alkalizace.