Abstract

Yeast, although non-motile and unicellular organisms can create organized structures, colonies, in which cells communicate and cooperate and which in some ways resemble multicellular organisms. Our previous studies on yeast colony development revealed that colonies periodically change pH of their surroundings. Alkalization of an agar medium is accompanied by production of volatile ammonia that acts as the long-range signal. Microarray analysis of the expression changes in Saccharomyces cerevisiae colonies during their transition from acid to alkali phase revealed significant changes in yeast transcriptome. Among others, strong induction of expression of three homologous genes ATO1 (YNR010c, ADY2), ATO2 (YNR002c, FUN34) and ATO3 (YDR384c) at the beginning of the alkali phase was found. These genes encode membrane proteins that may function as ammonium/H⁺ antiporters. This work contributes to better understanding of both the ammonia signaling and the role of putative ammonium exporters - Ato proteins. It was revealed, that other volatile compounds - methylamine and propylamine - are (in addition to ammonia) able to induce entry into the alkali phase of yeast colony development. Moreover, the significant impact of the transport of monocarboxylic acids on ammonia production and yeast colony development was shown. Limited transport of weak organic acids into the cells of selected deletion strains apparently prevented the proper ammonia signaling and subsequent developmental changes. In addition to the established model of giant yeast colonies, another two types of yeast populations were introduced - static and shaken (aerobically cultivated) liquid cultures of S.cerevisiae. Among others, we found that during growth of both types of cultures, pH of the media changes and pH increase is associated with ammonia and Ato proteins production. Thus, ammonia can probably act as a general warning signal of the approaching shortage of nutrients in the environment. Data concerning Ato proteins suggest that these proteins are necessary for growth in ambient neutral to alkaline pH. Moreover, some of Ato proteins (Ato1p and Ato2p, Ato1p and Ato1p and Ato3p and Ato3p) are able to interact. The results were achieved using new approaches suited to measure interactions between yeast membrane proteins, which are based on monitoring changes in fluorescence lifetime due to FRET (Fluorescence Resonance Energy Transfer).

Keywords: *Saccharomyces cerevisiae*, yeast colony, ammonia, ammonia signaling, differentiation, Ato1p, Ato2p, Ato3p, pH, Sok2p, Jen1p, monocarboxylic acids, FLIM-FRET