

Filamentous fungi such as *Trichoderma* are very interesting for biotechnology. These fungi more efficiently produce and secrete proteins compared to yeast *S. cerevisiae*. Fungal proteins such as hydrolytic enzymes are produced and utilized on an industrial scale. It is not known why filamentous fungi are so good protein producers. To clarify this we intend to look for differences in the activities of processes related to the synthesis of proteins in the cell and differences in the construction of cellular structures associated with the synthesis and maturation of proteins and their transport outside the cell. We know the subsequent steps which the protein passes during synthesis, maturation and secretion. In these processes cell membranes play an important role. Cell membranes are the place where enzymes modifying proteins are localized. These enzymes attach sugars to proteins protecting them against lysis and such proteins could leave the cell. Proteins are transported inside membrane vesicles from one structure to the other where they are modified and at the end to the cell membrane. Membrane vesicles integrate with the cell membrane and their content is liberated outside.

Differences in the cell membrane composition may result in the looser or more compact structure and that way affect membrane permeability and conformation of membrane-bound enzymes. The environment of membrane-bound proteins affects their biological activity. In addition, enzymes modifying proteins by attachment of sugar residues require dolichol an carrier of sugar residues. Such modifications precede secretion of many proteins outside the cell.

Our preliminary studies have shown that dolichols from filamentous fungi have a different structure than dolichols commonly found in both yeast and other organisms including humans. Structure of dolichols from filamentous fungi will be studied in this project. Dolichols have two different functions: they are components of cell membranes so their structure is important for properties of the cell membrane, and, furthermore, dolichols transfer sugar residues thus influence activity of dolichol-dependent protein-modifying enzymes. It is believed, that structure of dolichols is determined by enzyme catalyzing their synthesis. We will examine whether filamentous fungi, in contrast to the yeast additionally modify dolichols giving them the characteristic structure. We will also study whether dolichol-dependent enzymes from yeast and fungi prefer their own dolichols to reach maximum activity or can be similarly active in the presence of foreign dolichols.

In addition, we suspect that in fungi, as well as in yeast, enzyme synthesizing dolichols forms complex with another protein. We intend to characterize this protein. We will also examine how yeast and fungal dolichols influence properties of the cell membranes such as permeability and fluidity.

Our research will be conducted using genetic engineering, and biochemical and biophysical methods. We will be able to determine how the different cellular environment affects protein synthesis, modification and secretion. We will know structures of dolichols from fungi and yeast in detail and the impact of these structures on the function of cell membranes and dolichol-dependent enzymes.

The results will help answer the question why filamentous fungi as efficiently synthesize, modify and secrete proteins. Is that an advantage of the environment in which these processes occur?