Pavel Jůda - Abstract

The cell nucleus is a complex cellular organelle. The nucleus and nuclear processes are organized into functionally and morphologically separated nuclear subcompartments. This thesis is particularly concerned with the three following nuclear subcompartments: sites of DNA replication, Polycomb bodies and nuclear inclusions constituted of inosine monophosphate dehydrogenase 2 (IMPDH2).

First, we examined the relationship between MCM proteins and DNA replication. Using immunofluorescent labeling of cells extracted prior fixation and applying cross-correlation function analysis, we showed that MCM proteins are present at the sites of active DNA synthesis. Our results contributed to the solving of the first part of so-called MCM paradox.

Second, we studied the structural basis of the Polycomb bodies. Based on fluorescence microscopy studies, Polycomb bodies have been considered to be the nuclear subcompartments formed by the accumulation of Polycomb proteins in the interchromatin compartment. In our work, using correlative light electron microscopy and experimental changes in macromolecular crowding, we clearly showed that a Polycomb body is a chromosomal domain formed by an accumulation of heterochromatin structures, rather than a typical nucleoplasmic body.

Third, we were interested in the inclusions composed of inhibited IMPDH2 protein. Using microscopic methods, we showed the presence of IMPDH2 in the cell nucleus and its ability to form nuclear inclusions. We described in details the ultrastructure of these inclusions as well as the IMPDH2 inclusions in the cytoplasm.

Our results significantly contribute to the knowledge about the organization of the cell nucleus and in many aspects they change the current view on the structural basis and composition of some nuclear subcompartments.