ABSTRACT

Microtubules (MTs) play crucial roles in intracellular organization and transport, cell polarity, motility, signalling, division and differentiation. MTs form complex arrays, which are, due to their highly dynamic nature, capable of rapid reorganization in response to cellular requirements. Dynamics, stability and spatial organization of MTs are regulated by many factors including MT regulatory proteins. In the presented study we functionally characterized three selected MT regulatory proteins: Ca^{2+}-sensor STIM1, MT severing protein spastin and γ-tubulin that is essential for MT nucleation.

We found out that activation of bone marrow mast cells (BMMCs) leads to the formation of plasma membrane protrusions containing MTs. Formation of these MT protrusions is dependent on an influx of extracellular Ca^{2+} regulated by protein STIM1, located in endoplasmic reticulum. STIM1 associates with MTs and its depletion prevents formation of MT protrusions. This indicates that Ca^{2+} ions might be involved in MT regulation. Since STIM1 depletion also causes defects in chemotaxis, we propose that MT protrusions might be involved in sensing of external signals recognized by BMMCs.

Glioblastoma multiforme is the most common and most aggressive malignant primary brain tumor in humans. We demonstrated that MT severing protein spastin is overexpressed in glioma and glioblastoma cell lines and that its expression level increases with tumor malignancy. Glioblastoma cells depleted of spastin exhibit significantly lower motility and an increased proliferation rate. Modulation of these spastin functions in cell migration and proliferation has a potential to become a part of novel approaches to treatment of invasive gliomas.

We showed for the first time that γ-tubulin is present in the nucleoli of various cell types. We identified new γ-tubulin interacting protein C53 in the nucleus using mass spectrometry and found out that γ-tubulin can modulate C53 function in G2/M checkpoint activation after DNA damage. Furthermore, we showed that mammalian γ-tubulin 2 is able to nucleate MTs and substitute for γ-tubulin 1 in cultured cells and that these γ-tubulins are differentially expressed in mouse early embryogenesis and in adult tissues. Based on our results we propose that mammalian γ-tubulins are functionally equivalent with respect to their MT nucleation activity.