Abstract

MCPIP1 (*monocyte chemoattractant protein induced protein 1*), also known as Regnase 1, exerts anti-inflammatory activity attributed to its functions as RNase and deubiquitinase. The molecular targets of RNase activity of MCPIP1 are transcripts of e.g. inflammatory cytokines, while deubiquitinase activity has been described towards TRAF proteins (*tumor necrosis factor receptor associated factor*). The role of MCPIP1 role was broadly described in the inflammation induced by LPS (*lipopolysaccharide*), however, its role in tolerance to bacterial endotoxin has not been studied. Tolerance to LPS is a phenomenon characterized by reduced sensitivity of host cells to repeated exposure to bacterial endotoxin. Among others, this process plays a significant role in septic shock. After the initial phase of rapid inflammatory response, the phase of immunoparalysis occurs, manifested by insensitivity to LPS. During the research conducted in this study, it was shown that the level of MCPIP1 protein increases in LPS tolerance. This is a result of both MCPIP1 *de novo* synthesis induced by MCP-1 (*monocyte chemoattractant protein-1*) and increased stabilization of MCPIP1 caused by decreased activity of MALT-1 (*mucosa-associated lymphoid tissue 1*) paracaspase, which catalyze the proteolysis of MCPIP1. Decreased expression of MCPIP1 protein in macrophages inhibits the development of LPS tolerance *in vitro*. These results were confirmed using *in vivo* model of transgenic mice with selective downregulation of MCPIP1 expression in macrophages and neutrophils. This indicates the significant role of MCPIP1 in shaping the process of decreased sensitivity to LPS, while also confirming the dominant role of phagocytes in the phenomenon of LPS tolerance. The molecular mechanism of MCPIP1 role in the regulation of LPS tolerance is the effect of downregulation of MAP kinases (MAPK, *mitogen-activated protein kinases*) and in consequence inhibition of NF-κB activation (*nuclear factor kappa-light-chain-enhancer of activated B cells*). These results support the key role of the deubiquitinase activity of MCPIP1 towards TRAF proteins in the described phenomenon. Additionally, it was shown that MCPIP1 RNase activity is not crucial for the regulation of LPS tolerance. The presented results expand our knowledge of the biological significance of the MCPIP1 protein in phagocytes and provide significant insights into the molecular mechanisms of LPS tolerance.

MCPIP1 is described as a negative regulator for signaling pathways induced by Toll-like receptors (TLRs, *Toll like receptors*), but also for receptors recognizing host inflammatory mediators, including cytokines. Nevertheless, little is known about MCPIP1 contribution in JAK1/2-STAT1 signaling pathway, activated, among others, after recognition

of interferon type II. Using myeloid cells deprived of MCPIP1 it was demonstrated that this protein has a significant role in controlling JAK1/2-STAT1 signaling pathway. It was proven that under constitutive conditions, in the absence of IFN-γ, MCPIP1 promotes degradation of STAT1 (*signal transducer and activator of transcription 1*) transcript which accompanies the reduced expression of ISGs genes (*interferon stimulated genes*). Those data indicate for anti-inflammatory nature of MCPIP1 protein. However, in the presence of IFN-γ MCPIP1 promotes phosphorylation of STAT1 protein and induction of ISGs genes. This observation can be explained by the reduction of SOCS1 (*suppressor of cytokine signaling*), the protein which inhibits STAT1 phosphorylation. MCPIP1 controls the stability of SOCS1 transcript. Moreover, the level of SOCS1 is controlled indirectly by MCPIP1 via IL-6, a cytokine which stimulates SOCS1 expression. In summary, it has been demonstrated that MCPIP1 controls the IFN-γ signaling pathway giving an important contribution to further research on the systemic level. Moreover, it indicates that the control of the JAK1/2-STAT1 pathway by MCPIP1 is dependent on the level of IFN-γ in the microenvironment.

To sum up, the research presented in this thesis complements our knowledge of MCPIP1 protein biology and reveals its new role in controlling signal transduction pathways. Moreover, it demonstrates how significant function this protein plays in phagocytic cells, which is reflected in the systemic response of the organism. Furthermore, it was proven that MCPIP1 may differently regulate this response under homeostatic conditions and during inflammation associated with the accumulation of inflammatory mediators.