Hanna Nowicka

Temat w języku polskim:

„Rola fosforylacji komponentów kompleksu ISGF3 w regulacji ekspresji ISG i ochrony przeciwwirusowej”.

Temat w języku angielskim:

“The role of phosphorylation of ISGF3 components in the regulation of ISG expression and viral protection”.

Summary

Interferons belong to the family of cytokines that play crucial role in triggering of antiviral response and are divided into three subfamilies: Type-I, -II and -III. IFNα-dependent response, in the most classical form, is based on Signal Transducer and Activator of Transcription (STAT) family members STAT1 and STAT2 that become phosphorylated and together with interferon regulatory factor 9 (IRF9) form ISGF3. This complex recognizes the specific ISRE sequence in the regulatory regions of ISGs, thus mediating their expression and providing ability to combat viral infections.

Over the years evidence has accumulated that this system is not as simple. It becomes clear that besides canonical ISGF3-dependent signalling, also other ISGF3-like complexes function and may cooperate to provide effective viral protection. The examples are U-ISGF3 (that is composed of unphosphorylated forms of STAT1 and STAT2 together with IRF9) and complexes of STAT2 and IRF9 both, in phosphorylated (STAT2/IRF9) or unphosphorylated form (U-STAT2/IRF9). The role of all mentioned complexes was tested in this thesis.

In this dissertation, we focused on the role of phosphorylation in both, basal as well as IFN- and -time dependent transcriptional responses under wild-type and STAT1-deficiency conditions, we as well considered the dependence of viral protection on the amount of the native STAT1, STAT2 and IRF9.

Using different molecular techniques, among them whole-genome approaches, such as RNA-Seq and ChIP-Seq, in combination with JAK Inhibitor I (that prevents STAT phosphorylation) we provide further evidence that in WT cells an IFNα-inducible transcriptional mechanism exists, that relies on the ISGF3 components STAT1, STAT2 and IRF9 in a phosphorylation- and time-dependent manner. It also points to an important role of classical ISGF3 in the regulation of prolonged ISG expressions and viral protection. However, the contribution of U-ISGF3 under these conditions cannot be ruled out.

Likewise, in cells lacking STAT1, an IFNα-inducible and STAT1-independent transcriptional mechanism exists, that depends on the STAT2/IRF9 components STAT2 and IRF9 in a phosphorylation- and time-dependent manner. It also provides further prove for the previous observation that STAT2/IRF9 can take over the role of ISGF3 in the absence of STAT1, to regulate expression of a common group of ISRE-containing genes and provide protection against viral infection.

Finally, comparative experiments in STAT1-KO cells overexpressing all ISGF3 components, revealed a potential role of U-ISFG3, and possibly U-STAT2/IRF9, in the regulation of constitutive and long-term IFNα-treated ISG expression and viral protection. This strongly suggests that a certain threshold of STAT1, STAT2 and IRF9 expression and levels of U-ISGF3 (and/or U-STAT2/IRF9) has to be reached to be able to trigger ISG transcription. As a consequence, together with classical ISGF3, U-ISGF3 and U-STAT2/IRF9, could be instrumental in IFN-dependent and independent ISG transcription and in combatting viral infection.