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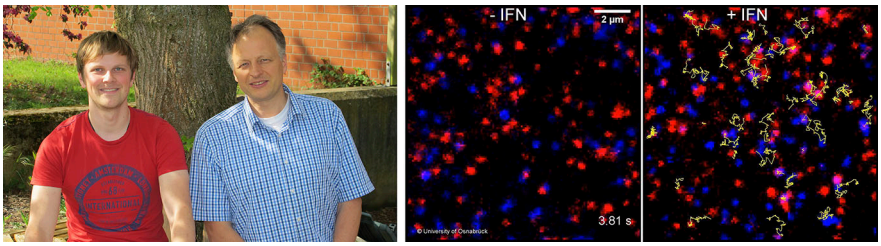
Dimerization dictates the message

Feedback regulation of receptor dimerization determines the cell's response to different interferons.

Focal Point



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Stephan Wilmes (left), Jacob Piehler (right), and colleagues investigate how different type I interferons can trigger different cellular responses, despite binding to the same dimeric cell surface receptor. In the absence of interferon (left), the receptor subunits IFNAR1 (red) and IFNAR2 (blue) remain separate from each other in the plasma membrane. Ligand addition (right) induces receptor dimerization, indicated by the colocomotion trajectories (yellow) of individual IFNAR1 and IFNAR2 molecules. The feedback regulator USP18 inhibits receptor dimerization such that only interferons with a relatively high affinity for IFNAR1 can recruit this subunit into the receptor complex and maintain signaling over longer time periods. These interferons can therefore induce different cellular responses than interferons with lower affinity for IFNAR1, which are quickly deactivated by USP18.

The different members of the human type I interferon (IFN) family can induce distinct cellular responses, even though they all bind to the same cell surface receptor. All IFNs evoke a potent antiviral response, for example, but IFNβ is far more effective than IFNα2 at inhibiting cell proliferation. Such "functional plasticity" is a common feature of cytokine signaling, but the mechanisms by which different ligands use the same receptor to induce different responses remain unknown. Wilmes et al. now demonstrate that the effects of IFNs depend on their ability to promote receptor dimerization (1).

The type I IFN receptor consists of two subunits, IFNAR1 and IFNAR2, which have low and high ligand binding affinities, respectively. Different IFNs, in turn, have different affinities for the receptor subunits, and IFNβ's relatively high affinity for IFNAR1 appears to be crucial for its specific functional effects (2). Cytokine receptors are often thought to "preassemble" in the plasma membrane before they bind their ligands, but Jacob Piehler, from the University of Osnabrück in Germany, previously found that, at least in vitro, IFNs first bind to the high-affinity IFNAR2 subunit, and then recruit the low-affinity IFNAR1 chain to form an active ternary complex (3). Moreover, due to its higher affinity for IFNAR1, IFNβ was better than IFNα2 at inducing receptor dimerization (2). "So it seemed like the recruitment of IFNAR1 might be critical for tuning the cellular response," Piehler explains. "But we needed to demonstrate this in cells." Piehler and colleagues, led by graduate student Stephan Wilmes, were also interested in how functional plasticity might be regulated by a protein called USP18, which, after being transcriptionally induced by IFN signaling, feeds back to inhibit the pathway (4, 5).

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Wilmes et al. first used single-molecule TIRF microscopy to monitor the binding of fluorescently labeled IFNα2 to the endogenous IFN receptor in vivo (1). By comparing the behavior of IFNα2 mutants with different affinities for the two receptor subunits, the researchers concluded that IFNs do, in fact, recruit IFNAR1 into a ternary complex with IFNAR2, and that this process is inhibited by USP18. "Cells expressing USP18 lose the ability to bind IFNα2 because receptor dimerization isn't as efficient," Piehler says.

To confirm this finding, Wilmes et al. directly visualized the assembly of individual receptors using fluorescently labeled receptor subunits expressed at physiological levels. In the absence of IFNs, the subunits were not preassembled or even organized in close proximity to each other. In the presence of ligand, however, the researchers could observe individual IFNAR1 and IFNAR2 chains come together to form a ternary complex. "The subunits float independently in the plasma membrane until the ligand is there, when they dimerize very efficiently," Piehler explains.

As in the ligand-binding assays, dimerization efficiency depended on the ligand's affinity for IFNAR1, and the process was inhibited by USP18. Previous reports have demonstrated that USP18 binds to the cytoplasmic tail of IFNAR2, potentially displacing the kinase Jak1 (4, 6). Wilmes et al. found that IFNAR2's Jak1-binding domain promoted dimerization, suggesting that the kinase somehow stabilizes the ternary ligand–receptor complex.

Thus, after its early induction by IFN signaling, USP18 might interfere with Jak1-mediated complex stabilization, thereby reducing the efficiency of receptor dimerization so that ligands with a relatively low affinity for IFNAR1, such as IFNα2, can no longer recruit this subunit to form an active signaling complex. IFNβ, with its relatively high affinity for IFNAR1, would still be able to induce receptor dimerization in the presence of USP18, allowing it to trigger additional cellular responses that require sustained IFN signaling.

The researchers now want to further investigate this mechanism of functional plasticity. "We want to pinpoint the role of Jak1 in receptor dimerization and to determine how USP18 interferes with the process," Piehler says.

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