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Interaction between short oligonucleotides and recombinant signaling proteins and their receptors

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Background/Aim. Oligonucleotides are short fragments of DNA or RNA that play a significant role in many fundamental biological processes and have pharmacological activity [1]. The aim of this study was to investigate the interaction of synthetic purine ribo- and deoxyoligonucleotides (OLNs) with recombinant signaling proteins, in particular interferon α2-β and insulin, their receptors, and somatropin. **Methods.** Oligonucleotides were synthesized using the solid-phase phosphoamidite method with purification by solid-phase extraction. Fluorescence titration with interaction analysis by the Stern-Volmer equation in its general and modified forms was used to analyze interactions [2]; docking in the PyRx program [3]. **Results.** Fluorometric titration showed the binding of OLNs to proteins in the middle affinity zone of Kd in the range of 10^{-5}–10^{-7} M, forming non-fluorescent complexes, the most active interaction being with shorter oligonucleotides. Negative cooperative two-ligand binding of INS to rG5, INSR receptor to rA5 oligonucleotide and INFR receptor to rG5 and rA5 preparations, and positive cooperative two-ligand binding of A5 to INFR were found. At the same time, only single-ligand binding was observed for G5 ligand. Oligoribonucleotides bound more strongly to insulin, interferon and its receptor, and oligdeoxynucleotides to the insulin receptor and somatotropin (difference up to an order of magnitude), with A5 binding 1.3 times stronger than G5 and rG5 binding 1.5 times stronger than rA5, respectively. These data correlated with the data on docking with ΔG_{bind} values ranging from –6.9 to –10.5 kkal/mol, the binding to the insulin receptor being stronger than to insulin, and interferon being stronger than to its receptor, which was confirmed experimentally by fluorescence quenching studies. **Conclusions.** The results of our experiments indicate that OLNs interact with recombinant signaling proteins and their receptors by different mechanisms (single-, double-ligand positive and negative cooperative) depending on the protein and the ligand itself, which, in turn, may be important for their various therapeutic applications. **Grants/Fundings.** Simons Support Grant 1290589**.**

K e y w o r d s: interferon, receptor, insulin, somatotropin, protein-ligand interaction, spectroscopy, oligonucleotide, docking.

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