In-depth structural analysis of the 'conformational change' of DNA aptamers

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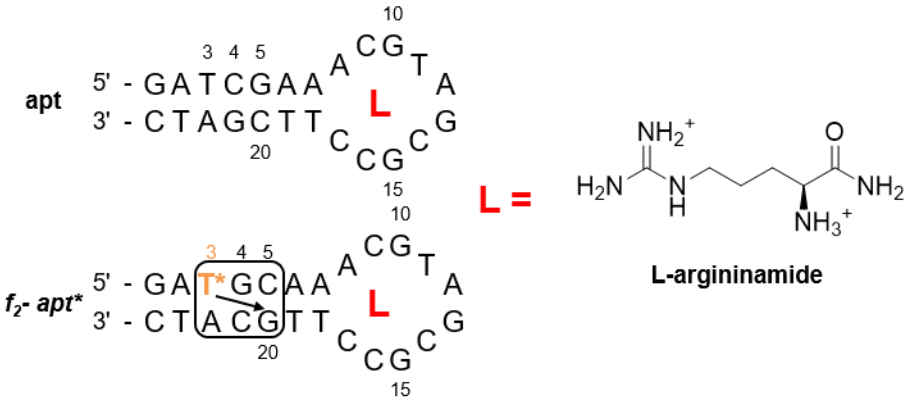
Aptamers are the RNA and/or DNA based analogues of antibodies, capable of binding both small molecules or larger targets. An aptamer against a particular target can be obtained through SELEX (Systematic Evolution of Ligands by Exponential enrichment). Compared to antibody based binding, their benefits include a larger range of targets, a longer shelf-life and more straightforward production conditions. Numerous examples can be found in literature where these singly stranded oligonucleotides bind their target in a very specific manner with limits of detection down to the femto molar range. Rather surprisingly however, very few aptamers have made it to the market so far.

In general, little information is available with respect to the recognition event itself, making it hard to optimize the selectivity and affinity towards the desired range. Additionally, the conditions under which proof-of-principle for aptamer function is delivered is often not similar to actual application conditions.

To address these issues we use liquid state NMR to investigate the recognition mechanism, involved in binding the ligand, on a molecular level. Eventually, we hope this will help to guide the design of tailor-made aptamers ready to be used in several fields of application. Two examples of how NMR can make a difference in aptamer research will be illustrated.

First, we show how the lack of response of the cocaine aptamer when coated onto an electrode surface can be explained by its supposed interaction with a common adulterant levamisole.

In another, well researched model system, we show how our previously discovered duplex stabilizing pKa motif, based on an imidazole-modified thymine (T*), can be used to increase the stability of the L-argininamide binding aptamer. Thereby demonstrating its ability to assist in pre-organizing the aptamer while making it also more robust against thermal denaturation.



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