

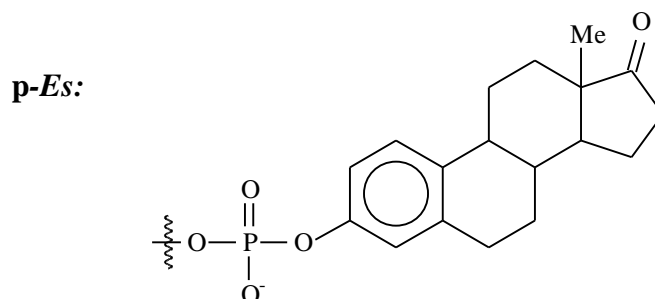
Solution Structure of Tandem Oligonucleotide Duplex with Two Spatially Adjacent Steroid Residues

Aleksej Yu. Denisov^{1*}, Tatyana V. Maltseva², Anders Sandstrom², Dmitrii V. Pyshnyi¹, Evgeniya M. Ivanova¹, Valentina F. Zarytova¹ and Jyoti Chattopadhyaya²

¹Novosibirsk Institute of Bioorganic Chemistry of SD RAS,
630090 Novosibirsk, Russia

²Department of Bioorganic Chemistry, Uppsala University,
75123 Uppsala, Sweden, Fax: +7-3832-333677 E-mail: denisov@niboch.nsc.ru

Covalent linking of steroids to oligonucleotide terminal phosphate group promotes stabilization of DNA complementary complexes. For detailed understanding of this effect in stereochemical terms, the spatial structure of estrone (*Es*) tethered $[pd(^5CAGC^3')p-Es] + [Es-pd(^5TCCA^3')] : pd(^5TGGAGCTG^3')$ tandem duplex with two spatially adjacent estrone residues was investigated by NMR, and compared with the solution structure of the same DNA duplex without steroids (1). The full assignment of all estrone and oligonucleotide proton signals in two-dimensional NMR spectra (NOESY, DQF-COSY, ROESY, *H,C*-HSQC, *etc.*) as well as MARDIGRAS relaxation matrix analysis and AMBER molecular dynamics refinements were used for this structure modeling. The NMR structures were based on 24 experimental constraints per residue.



It has been found that the general structure of both tandem duplexes corresponds to the B-type DNA. The aromatic protons of 3'-estrone residue have strong NOE-peaks with the sugar protons of cytidine bearing this 3'-steroid, and H2' of cytidine sugar ring is shifted to high field by 0.7 ppm due to the influence of 3'-estrone aromatic ring. The aromatics of the second 5'-estrone residue has strong NOE-peaks with aromatic and methyl group protons of the base of thymidine bearing this 5'-steroid. These typical NMR features suggested to us that both steroids are located into the major groove of DNA duplex. Also the methyl groups of both 3'- and 5'-estrones have NOE-contacts with the same H2 proton of adenosine base in octanucleotide that proves the spatial proximity of these estrone residues. The increase of thermal stability for such kind of tandem duplexes can be explained by hydrophobic interactions between two adjacent steroid residues.

1. A. Yu. Denisov *et al.*, *J. Biomol. Struct. Dyn.*, 15, 499-516 (1997).