I. Abstract

We studied how oligonucleotides' terminal ends and nature of present cations influence formation and multimerization of G-quadruplexes to long nanostructures, G-wires. Terminal GC ends in oligonucleotide might promote G-quadruplex multimerization via interlocking and thus formation of longer nanostructures. The effect of the presence of $^{15}NH_4^+$ and K^+ ions was studied on G-quadruplexes formed by oligonucleotides d($GCG_2AG_4AG_2$) and d($GCG_2AG_4AG_2CG$), named *GCn* and *GCnCG*. We showed that the presence of $^{15}NH_4^+$ or K^+ ions induces multimerization via stacking of 3'-terminal G-quartets in *GCn* G-quadruplex, which is precluded by 3'-GC ends in the case of *GCnCG* G-quadruplex. We observed five $^{15}NH_4^+$ ions bound in 3'-3' stacked *GCn* G-quadruplex multimer, with one located at 3'-3' stacking interface. $^{15}NH_4^+$ ions bound within 3'-3' stacked *GCn* G-quadruplex multimer exhibit slow exchange dynamics. Contrary, presence of 3'-GC ends accelerates exchange of bound $^{15}NH_4^+$ ions between binding sites in *GCnCG* G-quadruplex and with $^{15}NH_4^+$ ions in bulk solution. $^{15}NH_4^+$ ions within *GCnCG* G-quadruplex show unidirectional movement, which is characteristic for ion channels. We showed that in the presence of K^+ ions, d($G_2AG_4AG_2$) self-assembles into G-wires. By varying solution conditions and sample preparation procedure, we found five G-quadruplex structures, which are formed in d($G_2AG_4AG_2$) G-wire self-assembly. Using NMR spectroscopy we determined folding topologies of mentioned five G-quadruplex structures and thus obtained insight into mechanism of G-wire self-assembly on molecular level. Changing the nucleotides in loops enabled us to manipulate G-wires' properties. MD simulations provided rationale on how nucleotides in loops influence length of formed G-wires. We also studied the possibility of higher-order G-quadruplex structure formation in biological context on oligonucleotide from human telomere region, containing five G-tracts, d($TAG_3(T_2AG_3)_4$). We showed that the presence of additional G-tract leads to formation of parallel G-quadruplex with 3'-terminal T_2AG_3 overhang. Multimerization is more likely for parallel than hybrid G-quadruplexes, where lateral loops hinder stacking of terminal G-quartets.

Key words: G-quadruplexes, multimerization, higher-order structures, G-wires, self-assembly, NMR, DNA nanotechnology