Supramolecular forces guide the assembly of carbon nanotube and oligonucleotide vectors: implications for gene delivery

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Establishing a comprehensive picture of the chemical identity of ammonium-functionalized carbon nanotube (f-CNT)-based nucleic acid constructs is critical to design and implementation to mediate the delivery of siRNA. Therefore, a quantitative investigation of the several different non-covalent supramolecular bonding interactions between oligonucleotides and f-CNT was necessary to maximizing their potential efficiency as drug delivery vehicles. This work aimed to explore the thermodynamic, kinetic and stoichiometric properties of single- and double-strand oligonucleotide sequences binding to f-CNT. The binding affinity of f-CNT and short oligonucleotide sequences was approximately 10 nM; the kinetics of complexation were very rapid (< 0.5 min.) in aqueous solution at 37^{o}C; and from one to several single- or double-strand sequences were bound per f-CNT. Fluorescence-quenching spectrophotometry and ^{31}P-NMR were the techniques employed to interrogate this system in aqueous solution. A tactical combination of components, methods, and conditions revealed that primarily ionic and hydrogen-bonding interactions, and to a lesser extent, π-stacking forces, all contributed to the supramolecular assembly of oligonucleotide and functionalized carbon nanotubes constructs in aqueous solution. Key evidence for the mechanism by which the bound oligonucleotide cargo could be off-loaded from the f-CNT platform was acquired from these data, and justified further evaluation in biological systems. The key advantage of quantifying these parameters will be realized in the design optimization of these constructs in order to accomplish specific and efficient systemic delivery of gene-vectors to cellular targets in vivo using f-CNT. These data have now been applied to build constructs that were able to silence the expression of protein more effectively and with lower cytotoxicity than a conventional transfection agent.