

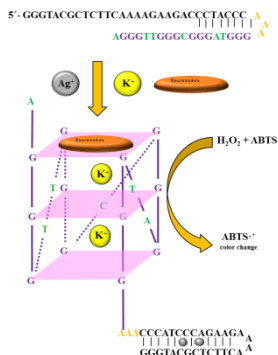
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Spektrofotometrická analýza nukleových kyselin tvořících kvadruplexy

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Abstrakt

G-quadruplexes form a complex with hemin called DNAzymes. These complexes exhibit peroxidase-like activity and effectively catalyse the H_2O_2 -mediated oxidation of 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)diammonium salt (ABTS), which can be determined spectrophotometrically. Firstly, effect of Ag^+ ions on DNAzyme activity was observed using spectrophotometric methods. Different times of DNA (rich in guanine) incubation with potassium, hemin and silver ions were studied. The absorption maximum of DNA with hemin and potassium ions at the wavelength 422 nm has been found. Ag^+ ions further increased absorbance values. Calibration curve was linear in the range of the Ag^+ ions concentrations 50-200 nM with the detection limit of 25 nM. Electrochemical behavior of oligonucleotide that is suitable for the G-quadruplex formation was determined by measuring electrochemical response of guanine, adenine and cytosine. So called G peak reflects anodic oxidation of a guanine reduction product. Cathodic CA peak reflects reduction of adenine and cytosine residues. Monitoring of statistically significant changes of both peaks during the G-quadruplex preparation was the main point of the experiment. Changes of the G peak were associated directly with the G-quadruplex structure formation while changes in the CA peaks were related to the binding and stabilizing of oligonucleotide strands by the silver ions.



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