

Analysis of Cisplatinated DNA Using Magnetic Immunoseparation

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Introduction

Magnetic separation employs spherical nanometric or micrometric structures with paramagnetic or superparamagnetic properties for isolation of various interesting compounds¹, including the cisplatinated (cisPt) DNA. This method can be employed as an easy way of detecting guanine-rich regions in DNA² or to monitor the pharmacokinetics of cisplatin-DNA interactions³.

Materials/methods

DNA fragment of phage λ *xis* gene was mixed with cisPt of various concentrations and incubated to form adducts. Dynabeads Protein G particles were used for immunoseparation. The anti-dsDNA antibodies were bound on the MPs' surface according to the manufacturer's instructions. CisPt-DNA sample was added and incubated to form complex. Anti-cisPt-modified DNA antibody was added to the complex and CdTe QDs modified with HWR peptide were bound to this antibody. The fluorescence of the QDs was measured after purification with the excitation wavelength of 360 nm.

Results and conclusion

The reactivity of both used antibodies was tested using dot blot. Anti-dsDNA antibody was able to detect 0.2 nM concentration of DNA with a linear dependence on concentration. This reactivity was not lost after cisplatin of DNA. The reactivity of anti-cisPt-DNA was linearly increased with increasing incorporated platinum in DNA. The capacity of the magnetic particles was 10 µg of antibodies per mg of the particles. The sandwich immunoassay caused 6.6-fold increased fluorescence of QDs labelled anti-cisPt-DNA antibodies compared with isolation of DNA without cisPt. Using this approach, 86% of applied DNA can be detected on the particles.

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- 2 Siddik, Z. H. Cisplatin: mode of cytotoxic action and molecular basis of resistance. *Oncogene* **22**, 7265-7279, doi:10.1038/sj.onc.1206933 (2003).
- 3 Chen, H. Y., Chen, H. F., Kao, C. L., Yang, P. Y. & Hsu, S. C. N. Interaction of electrons with cisplatin and the subsequent effect on DNA damage: a density functional theory study. *Phys. Chem. Chem. Phys.* **16**, 19290-19297, doi:10.1039/c4cp02306d (2014).