

P051 Development of a kinase assay for high-throughput screening of DNA-PK inhibitors

Ihor Yakymovych, Birgitta Mörk, Dali Zong, Petra Hååg, Rolf Lewensohn and Kristina Viktorsson

Department of Oncology/Pathology, Karolinska Biomics Center, Karolinska Institutet, Stockholm, Sweden.

Inhibition of DNA double strand break repair can greatly enhance the efficiency of radio- and chemotherapies. We have previously shown that increased DNA-dependent protein kinase (DNA-PK) expression correlates with an increased DNA repair capacity in non-small cell lung carcinoma cells and influence their resistance to DNA double strand break-inducing treatment. It suggests that DNA-PK inhibitors may reverse the resistance of tumour cells to the DNA damaging treatment. Existing DNA-PK inhibitors are not effective *in vivo* because of their toxicity and/or low selectivity. The aim of our study is to search for new substances with potential to inhibit DNA-PK. Here we report about the development of an non-radioactive *in vitro* Kinase Assay for DNA-PK and its successful application for screening of the inhibitors. As readout system for the evaluation of kinase activity we used an ATP-monitoring system based on firefly luciferase (easyLite-Kinase™, PerkinElmer). The assay relies on the consumption of ATP during a kinase reaction without the need for radioactive-labelled substrates. The results have shown that our assay is compatible with previously described assays which use $[\gamma\text{-}^{32}\text{P}]\text{ATP}$ and can be used in highthroughput screening for DNA-PK inhibitors.