Influence of (5'*R*) and (5'*S*) 5',8-cyclo-2'-deoxypurines on the repair process of clustered DNA lesions

Each cell in the human body is continuously exposed to various agents, such as ionisation radiation (UV, gamma, x-ray), reactive oxygen species (ROS) or products of endocellular processes. Their activity can cause *in a single cell* up to $1.5x10^5$ oxidative events per day. DNA is a storage house of genetic information which is described in nucleotide sequence. It is important to mention here that in evolution, prolonging longevity is irrelevant: the 'aims' of evolution are maintaining the stability of genetic information, and the survival of the species. Exposure of the genome to ionisation radiation and/or ROS activity can lead to various types of DNA damage, in both the sugar and base nucleotide subunits. Up to now around 80 different types of DNA damage have been identified. A single/isolated lesion (one lesion per one helix turn) like 2'deoxyuridine (dU), 8-oxo-2'-deoxyguanosine (^{oxo}dG), apurinic/apirimidinic site (AP-site), is repaired by the base excision repair (BER) system initiated by specific mono- or bifunctional glycosylases. On the other hand **clustered lesions** (two or more DNA lesions per one or two helix turns, induced by single ionisation tracks) can be removed from the genome by more complicated systems such as nucleotide excision repair (NER), homologus recombination (HR), and non-homologus end joining (NHEJ). As an example of clustered lesions the following DNA damage can be given: (**5'*R*) and (5'*S*) 8,5'-cyclo-2'-deoxypurine (cdPu)**, cis-platine adducts, cyclobutane pyrimidine dimers, pyrimidine-(6,4)-pyrimidone products, inter- or intrastrand crosslink, and double-strand brakes (DBS). A defect in NER activity can lead, in the context of cluster lesions and **cdPu**, to an accumulation of DNA lesions in the genome, which cause cancer, neurodegenerative disorders, *etc.* **8,5'-cyclo-2'-deoxypurines** are a unique kind of DNA damage, i.e. a . tandem lesion, where both the sugar and base moieties have been modified in the structures of the same nucleoside/nucleotide. Both diastereomers (**5'*R*)** and **(5'*S*)** of **cdPu** are removed from DNA by NER activity, perhaps due to the lack of specific glysosylases. Some data in literature indicates that **cdPu** can induce cancers and neurodegenerative disorders including Parkinson's, Alzhimer's, Huntington's, and*; while* defects in nucleotide excision repair systems in the cases of *Xerodermy Pigmentosum*, Cockayne syndrome and trichothiodystrophy can lead to **cdPu** accumulation in a patient's cells. **The aim of this study project is to assess the force of (5'*R*) and (5'*S*) 8,5'-cyclo-2'-deoxypurines' impact on the repair process of a clustered lesion in the context of cell lines with some defects in NER machinery.** Therefore, the following tasks will be undertaken: **A)** investigating the influence of **cdPu** on the efficiency of BER machinery in clustered DNA lesions, located in the same or both DNA strands, **B)** estimating the influence of **cdPu** on SspI and BsmAI restriction enzyme activity, **C)** influence of **cdPu** on the frequency of mutation formation in the BER repair region of the genome. Moreover, we will attempt to shed light on the question of whether **long patch BER is able to remove cdA from DNA as a part of clustered lesions**.

The obtained results during the project should: A) advance our knowledge of clustered lesion repair machinery and allow us to ascertain the role cdPu plays in it; B) prompt further research into new therapeutic strategies in the field of cancer, neurodegenerative disorders as well as disease entities with NER defects, C) indirectly increase the safety and effectiveness of radiotherapy and radiodiagnostics. It is important to mention here that even a low dose of radiation at the level of 1Gy can induce clustered lesions, and it can be expected that **cdPu** does so too.