

## POSTER 19

### HIV-2 LEADER RNA: STRUCTURE PROBING AND COMPLEX FORMATION WITH RECOMBINANT HIV-2 NUCLEOCAPSID PROTEIN NCP8

Katarzyna J. Purzycka<sup>1,2</sup>, Katarzyna Pachulska-Wieczorek<sup>2</sup>, Stuart F.J. Le Grice<sup>1</sup>, and R.W. Adamiak<sup>2</sup>

<sup>1</sup>RT Biochemistry Section, HIV Drug Resistance Program, NCI-Frederick, Frederick, MD 21702; <sup>2</sup>Laboratory of Structural Chemistry of Nucleic Acids, Institute of Bioorganic Chemistry, Polish Academy of Sciences, Noskowskiego 12/14, 61-704 Poznań, Poland

Retroviral genomes are assembled from two (+) sense RNAs by interactions at their 5' ends. The HIV-2 RNA DIS domain (DIS-2) mediates dimerization of the genome via a 'kissing hairpin' interaction. Both, Mg<sup>2+</sup> ions and nucleocapsid protein (NC) are important mediators of dimerization. HIV-2 NC (NCp8), a highly active chaperone, contains two zinc fingers of the type C-X2-C-X4-H-X4-C.

Previous studies on HIV-2 RNA/NC nucleoprotein complexes were performed with the *gag* polyprotein or chemically synthesised peptide. The NCp8 protein, expressed from the engineered pGEX-4T3-NC2 vector as a fusion protein with GST, cleaved-off with thrombin, migrates on SDS gels with the same mobility as the synthetically-derived polypeptide. Access to the pure recombinant nucleocapsid protein has allowed us to perform a detailed analysis of RNA/NCp8 complex formation.

As a part of the structural studies concerning conformational rearrangements of the HIV-2 leader RNA and its interactions with viral and cellular proteins [1], we will present results of structural probing of the 5'-UTR in the presence of Mg<sup>2+</sup> ions and NCp8. In addition the influence of antisense oligoribonucleotides designed to inhibit dimerization of the HIV-2 leader will be shown. Results we have obtained have allowed us to propose an *in vitro* model for HIV-2 leader RNA dimerization initiation.

- [1] K. Pachulska-Wieczorek, K. J. Purzycka and R. W. Adamiak, *New, extended hairpin form of the TAR-2 RNA domain points to the structural polymorphism at the 5' end of the HIV-2 leader RNA*, Nucleic Acids Res., 34, 2984-2997 (2006).