Laboratory of Viral and Cellular Genetics

Receptors for retroviruses, retroviral vectors, endogenous retroviruses, silencing of retroviruses, epigenetics

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Retrovirus replication cycle starts by specific binding of retroviral envelope proteins to host cell receptors. After entering host cells, retroviruses integrate into the host chromosomes, and use the cell transcription and proteosynthesis machineries to express retroviral proteins and propagate their own progeny. At multiple levels, cellular restriction factors regulate retroviral replication. Retroviruses can broaden their host range by mutations of the env gene, and vice versa, host cells develop resistance to retroviruses by mutations of genes encoding the specific receptors. Avian leukosis virus subgroup J (ALV-J), an important pathogen of domestic poultry, infects chickens and turkeys, whereas other galliform species are resistant thanks to a single amino-acid substitution in cell surface Na+/H+ exchanger (NHE1), the receptor for the virus. We now screen the NHE1 receptor in domestic chickens and wild ducks in order to find genetic sources for the resistance and predict the spread of ALV-J in natural reservoirs. Another defence mechanism used by the host cells is the inactivation of integrated invaders at the level of transcription via DNA methylation and modifications of adjacent histones. This might be an obstacle in the case of retroviral vectors used for the gene transfer in gene therapy trials. We have demonstrated that vectors derived from avian sarcoma and leukosis viruses are efficiently silenced through DNA methylation and we developed strategies of protection from DNA methyltransferases by simple modifications of retroviral regulatory sequences. Now, our main effort is focused on the epigenomics of retroviral integration sites. The epigenomic approach to retrovirus integration reveals that transcriptionally active proviruses are preferentially localized close to the transcription starts of targeted genes or in enhancer regions. The protective region around the transcription start site is marked by enrichment in H3K4 trimethylation, demethylation, and H3K9 acetylation. Retrovirus restriction by the host cell is particularly effective in cross-species transmission. In a classic model of rodent cells transformed with Rous sarcoma virus, we studied the molecular mechanisms of virus rescue. Fusion with chicken cells permissive for Rous sarcoma virus provides the rodent cells with factors required for proper splicing and transport of viral RNA. Last but not least, our laboratory deals with endogenous retroviruses. We studied the epigenetic control and the possible role of hydroxymethyl cytosine in expression of syncytin-1, a fusogenic envelope glycoprotein encoded by human endogenous retrovirus ERVWE1. We are interested in the process of endogenization displayed in a model of polymorphic and infectious endogenous retrovirus in the mule deer genome. Using a new two-step computational screen, we discovered a new endogenous retrovirus in the genome of Malayan colugo. This is the first endogenous lentivirus identified in the Euarchonta lineage, which includes primates, and represents the oldest member of the lentivirus genus.

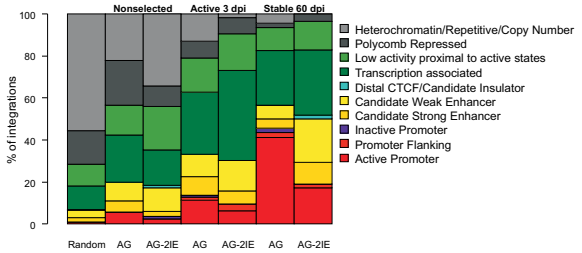

Fig. 1. An example of retrovirus integration analysis. In contrast to random distribution, retroviruses are overrepresented in certain chromatin segments. Selection for transcriptional activity favours retroviruses integrated into active promoter and enhancer regions.

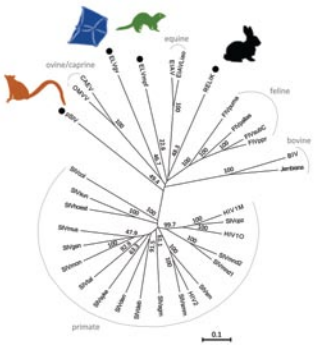

Fig. 2. Phylogenetic relationship of new endogenous retrovirus ELVgv to other lentiviruses. Endogenous lentiviruses described so far in rabbit, ferret, grey mouse lemur, and colugo are denoted.

- GACR, GAP304/12/1736 – Development of CpG methylation of HIV-1 promoter and its stabilizing effect on the latent provirus reservoir: longitudinal study in patients, 2012-2014, K. Trejbalová
- MA, QJ1210041 – New type of vaccine against chicken viral diseases, 2012-2016, J. Hejnar
- MEYS, LK11215 LK-NÁVRAT – Regulation of active endogenous retroviruses in the mammalian genome, 2012-2016, D. Elleder
- MH, NT14601 – Syncytins as markers of germ line tumours, 2013-2015, J. Hejnar
- GACR, GA13-37600S – Expression of fusogenic human endogenous retroviruses in germ line tumours, 2013-2015, J. Hejnar
- GACR, GA13-30983S – Chicken polymorphisms in retroviral receptors and their potential for cross-species transmission, 2013-2015, J. Plachý
- GACR, 14-34873S – Epigenomics of retroviral integration, 2014-2016, J. Hejnar
- GACR, 14-32547S – Sensing of HIV-1 by plasmacytoid dendritic cells: dichotomy of immunoreceptor signalling, 2014-2017, I. Hirsch
- GACR, GAP502/11/2207 – Avian sarcoma and leukosis virus-based vectors and their potential for transgenesis in chicken, 2011-2013, J. Hejnar

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From the left: Markéta Reinišová, MSc / Research Assistant, Martina Benešová / PhD Student, Kateřina Trejbalová, PhD / Research Associate, Jan Vlasatý / Bachelor Student, Ivan Hirsch, PhD / Research Fellow, Kryštof Štafl / Diploma Student, Josef Geryk, PhD / Research Fellow, Helena Fábryová, MSc / PhD Student, Jiří Plachý, PhD / Research Fellow, Magda Matoušková, PhD / Postdoctoral Fellow, Dalibor Miklík, MSc / PhD Student, Tomáš Hron, MSc / PhD Student, Volodymyr Stepanets, PhD / Postdoctoral Fellow, Dana Kučerová, MSc / Research Assistant, Jiří Hejnar, PhD / Head of Laboratory, Albert Font Haro, MSc / PhD student, Anna Lounková, MSc / PhD Student, Martina Slavková / Diploma Student, Daniel Elleder, PhD / Research Associate, Denisa Kovářová, MSc / PhD Student.

Not in the picture: Prof Jan Svoboda, DSc / Research Fellow, Filip Šenigl, PhD / Research Associate, Lenka Mikušová / Technician.