



**Katedry biochémie a genetiky**  
Prírodovedeckej fakulty Univerzity Komenského

Vás pozývajú na **37.** prednášku v rámci Kuželových seminárov:

**Dr. Július Lukeš**  
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České Budějovice

**Functional analysis of proteins involved  
in RNA editing in *Trypanosoma brucei*  
using RNA interference**

ktorá sa uskutoční

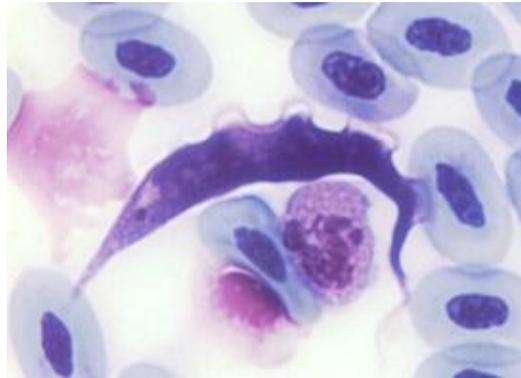
**24.6. 2003** (utorok)  
o **10:00** v miestnosti **CH1-224** PriF UK

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## Dr. Július Lukeš.

1991-present	Research Scientist,
1992-present	Member of the Scientific Board of the Institute of Parasitology
1993	Postdoctoral Fellow, E.C.Slater Institute, University of Amsterdam, The Netherlands
1994-1997	Assistant, Faculty of Biology, University of South Bohemia, České Budějovice
1997-1998	Postdoctoral Fellow, Department of Biology, University of California, Riverside
1998-1999	Visiting Scientist, Molecular Biology Institute, University of California, Los Angeles

In the mitochondrion of kinetoplastid flagellates the insertion/deletion RNA editing occurs via a “cut-and-paste” mechanism carried out by a complex machinery consisting of numerous proteins. In procyclic form *T. brucei*, we generated both single and double tetracycline-regulated RNAi knock-down cell lines for two guide (g) RNA binding proteins, gBP21 and gBP25, which have been shown to promote annealing of gRNA and pre-mRNA. Both proteins co-migrate in glycerol gradients and elimination of gBP21 protein results in a marked decrease of gBP25 and *vice versa*, suggesting some type of interdependence between the two proteins. The amount of some other proteins involved in editing (TbREL1, RBP16, REAP) is not affected by the disruption of the gBPs. We have shown that a marked growth phenotype is associated with a strong effect on editing *in vivo* upon repression of the gBPs. As revealed by primer extension, Northern analysis and quantitative PCR, the levels of several edited (RPS12, cox3, cytB,) and also some never-edited (cox1, ND4) transcripts were reduced in all three knock-downs. Interestingly, the levels of never-edited 12S rRNA and unedited and edited cox2 mRNAs (the only mRNA with gRNA in cis), as well as those of the GAPDH control were unaffected, while editing of ND7 seems to be up-regulated. *In vitro* analysis of editing in the gBP knock-downs is under way.



### **Selected most recent publications:**

- Gažiová I. & Lukeš J. (2003). Mitochondrial and nuclear localization of topoisomerase II in the flagellate *Bodo saltans* (Kinetoplastida), a species with non-catenated kinetoplast DNA. *J. Biol. Chem.* **278**: 10900-10907.
- Dyková I., Fiala I., Lom J. & Lukeš J. (2003). Perkinsiella amoebae-like endosymbionts of Neoparamoeba spp., relatives of the kinetoplastid *Ichthyobodo*. *Eur. J. Protistol.* **39**: 37-52.
- Maslov D.A., Zíková A., Kyselová I. & Lukeš J. (2002). A novel nuclear-encoded putative subunit of the cytochrome c oxidase complex in trypanosomatids. *Mol. Biochem. Parasitol.* **125**: 113-125.
- Lukeš J., Guilbride D.L., Votýpka J., Zíková A., Benne R. & Englund P.T. (2002). The kinetoplast DNA network: Evolution of an improbable structure. *Eukaryot. Cell* **1**: 495-502.
- Horváth A., Neboháčová M., Lukeš J. & Maslov D.A. (2002). Unusual polypeptide synthesis in the kinetoplast-mitochondria from *Leishmania tarentolae*. Identification of individual *de novo* translation products. *J. Biol. Chem.* **277**: 7222-7230.
- Oborník M., Van de Peer Y., Hypša V., Frickey T., Šlapeta J.R., Meyer A. & Lukeš J. (2002) Phylogenetic analyses suggest a possible lateral gene transfer from the mitochondrion to the apicoplast. *Gene* **285**: 109-118.
- Lukeš J., Hines J.C., Evans C.J., Avliyakov N.K., Prabhu V.P., Chen J. & Ray D.S. (2001). Disruption of the *Crithidia fasciculata* KAP1 gene results in extensive structural rearrangement of the kinetoplast disc. *Mol. Biochem. Parasitol.* **117**: 179-186.
- Šlapeta J. R., Modrý D., Votýpka J., Jirků M., Oborník M., Lukeš J. & Koudela B. (2001) *Eimeria telekii* n. sp. (Apicomplexa: Coccidia) from *Lemniscomys striatus* (Rodentia: Muridae): morphology, pathology and phylogeny. *Parasitology* **122**: 133-145.