

**University of South Bohemia**

**Faculty of Science**

Department of Molecular Biology

***Paratrypanosoma* is a novel early-branching trypanosomatid**

RNDr. Thesis

**Mgr. Tomáš Skalický**

Supervisor: Prof. RNDr. Julius Lukeš Csc.

České Budějovice, 2014

Flegontov P, Votýpka J, Skalický T, Logacheva MD, Penin AA, Tanifuji G, Onodera NT, Kondrashov AS, Volf P, Archibald JM, Lukeš J. Paratrypanosoma is a novel early-branching trypanosomatid. *Curr Biol.* 2013 Sep 23; 23(18):1787-93. doi: 10.1016/j.cub.2013.07.045. Epub 2013 Sep 5. PubMed PMID: 24012313.

### **Annotation**

The kinetoplastids are a widespread and important group of single-celled eukaryotes, many of which are devastating parasites of animals, including humans. We have discovered a new insect trypanosomatid in the gut of *Culex pipiens* mosquitoes. Glyceraldehyde-3-phosphate dehydrogenase- and SSU rRNA-based phylogenetic analyses show this parasite to constitute a distinct branch between the free-living *Bodo saltans* and the obligatory parasitic clades represented by the genus *Trypanosoma* and other trypanosomatids. From draft genome sequence data, we identified 114 protein genes shared among the new flagellate, 15 trypanosomatid species, *B. saltans*, and the heterolobosean *Naegleria gruberi*, as well as 129 protein genes shared with the basal kinetoplastid *Perkinsela* sp. Individual protein phylogenies together with analyses of concatenated alignments show that the new species, here named *Paratrypanosoma confusum* n. gen., n. sp., branches with very high support at the base of the family Trypanosomatidae. *P. confusum* thus represents a long-sought-after missing link between the ancestral free-living bodonids and the derived parasitic trypanosomatids. Further analysis of the *P. confusum* genome should provide insight into the emergence of parasitism in the medically important trypanosomatids.

## **Declaration in Czech**

Prohlašuji, že svoji rigorózní práci jsem vypracoval samostatně, pouze s použitím pramenů a literatury uvedených v seznamu citované literatury.

Prohlašuji, že v souladu s § 47b zákona č. 111/1998 Sb. v platném znění souhlasím se zveřejněním své rigorózní práce, a to v úpravě vzniklé vypuštěním vyznačených částí archivovaných Přírodovědeckou fakultou elektronickou cestou ve veřejně přístupné části databáze STAG provozované Jihočeskou univerzitou v Českých Budějovicích na jejích internetových stránkách, a to se zachováním mého autorského práva k odevzdanému textu této kvalifikační práce. Souhlasím dále s tím, aby toutéž elektronickou cestou byly v souladu s uvedeným ustanovením zákona č. 111/1998 Sb. zveřejněny posudky školitele a oponentů práce i záznam o průběhu a výsledku obhajoby kvalifikační práce. Rovněž souhlasím s porovnáním textu mé kvalifikační práce s databází kvalifikačních prací Theses.cz provozovanou Národním registrem vysokoškolských kvalifikačních prací a systémem na odhalování plagiátů.

České Budějovice 24. 4. 2014

.....  
Tomáš Skalický

### **Declaration of corresponding author**

I hereby declare, that Tomáš Skalický had a major contribution to the following paper. He has cultivated *P. confusum*, isolated its genomic DNA and assembled genome. He has also performed single gene and multigene phylogenetic analyses, cross-validation analyses, created figures for publication, uploaded sequencing data to public database and contributed to text of the manuscript.

České Budějovice 24. 4. 2014

.....  
Prof. RNDr. Julius Lukeš CSc.

### **Financial support**

This work was supported by the Czech Grant Agency (P305/11/2179, 206/09/H026) and a Praemium Academiae award to Julius Lukeš, who is also a Fellow of the Canadian Institute for Advanced Research (CIFAR). Research in the Archibald Lab was supported by an operating grant from the Canadian Institutes of Health Research (CIHR). John M. Archibald holds a CIHR New Investigator Award and is a CIFAR Fellow. Maria D. Logacheva, Aleksey A. Penin., Alexey S. Kondrashov were supported by Russian Ministry of Education and Science (G34.31.0008).

# REPORT

## *Paratrypanosoma* is a novel early-branching trypanosomatid

**Pavel Flegontov<sup>1,\*</sup>, Jan Votýpka<sup>1,2,\*</sup>, Tomáš Skalický<sup>1,3,\*</sup>, Maria D. Logacheva<sup>4,5</sup>, Aleksey A. Penin<sup>4,6</sup>, Goro Tanifuji<sup>7</sup>, Naoko T. Onodera<sup>7</sup>, Alexey S. Kondrashov<sup>4,8</sup>, Petr Volf<sup>2</sup>, John M. Archibald<sup>7</sup>, Julius Lukeš<sup>1,3,#</sup>**

<sup>1</sup> Institute of Parasitology, Biology Centre, 37005 České Budějovice (Budweis), Czech Republic

<sup>2</sup> Department of Parasitology, Faculty of Science, Charles University, 12844 Prague, Czech Republic

<sup>3</sup> Faculty of Science, University of South Bohemia, 37005 České Budějovice (Budweis), Czech Republic

<sup>4</sup> Faculty of Bioengineering and Bioinformatics, Lomonosov Moscow State University, Moscow, 119991, Russia

<sup>5</sup> A.N. Belozersky Institute of Physico-Chemical Biology, Lomonosov Moscow State University, Moscow, 119991, Russia

<sup>6</sup> Kharkevich Institute for Information Transmission Problems, Russian Academy of Sciences, Moscow, 127994, Russia

<sup>7</sup> Department of Biochemistry and Molecular Biology, Dalhousie University, Halifax, Nova Scotia, B3H 4R2, Canada

<sup>8</sup> Life Sciences Institute and Department of Ecology and Evolutionary Biology, University of Michigan, Ann Arbor, MI 48109, USA

\* These authors contributed equally

# Correspondence: jula@paru.cas.cz

### Summary

The kinetoplastids are a widespread and important group of single-celled eukaryotes, many of which are devastating parasites of animals, including humans. We have discovered a new insect trypanosomatid in the gut of *Culex pipiens* mosquitoes. Glyceraldehyde-3-phosphate dehydrogenase- and SSU rRNA-based phylogenetic analyses show this parasite to constitute a distinct branch between the free-living *Bodo saltans* and the obligatory parasitic clades represented by the genus *Trypanosoma* and other trypanosomatids. From draft genome sequence data we identified 114 protein genes shared among the new flagellate, 15 trypanosomatid species, *B. saltans*, and the heterolobosean *Naegleria gruberi*, as well as 129 protein genes shared with the basal kinetoplastid *Perkinsela* sp. Individual protein phylogenies together with analyses of concatenated alignments show that the new species, here named *Paratrypanosoma confusum* n. gen., n. sp., branches with very high support at the base of the family Trypanosomatidae. *P. confusum* thus represents a long-sought after missing link between the ancestral free-living bodonids and the derived parasitic trypanosomatids. Further analysis of the *P. confusum* genome should provide insight into the emergence of parasitism in the medically important trypanosomatids.