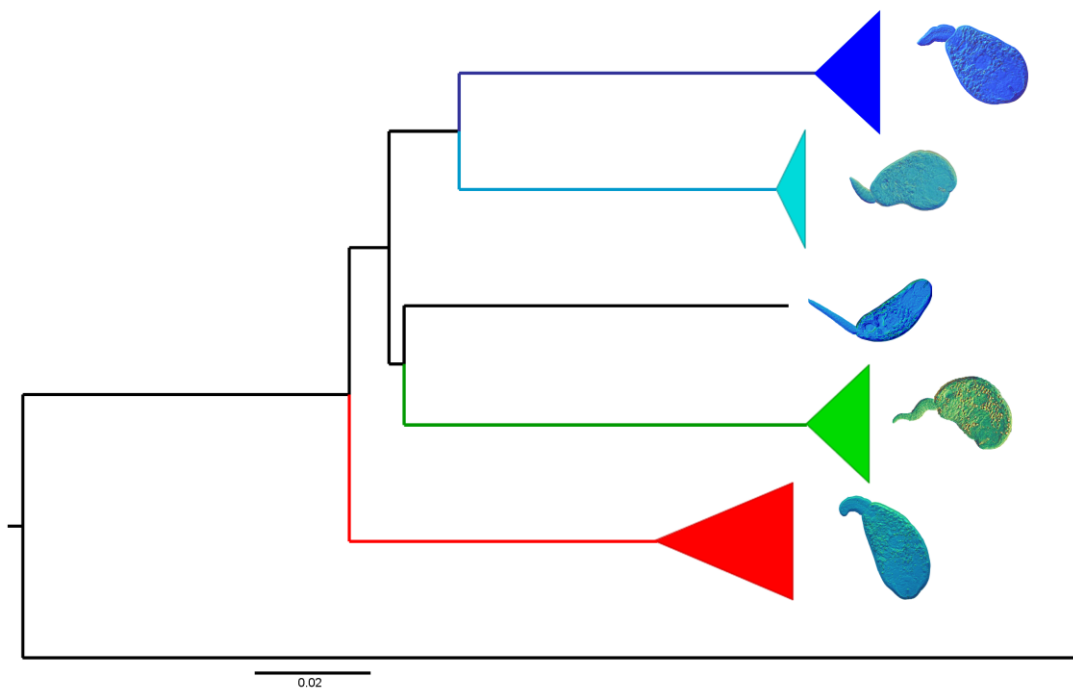


University of South Bohemia in České Budějovice
Faculty of Science

**SPECIES DIVERSITY OF *PLAGIORCHIS* LÜHE, 1899 (DIGENEA:
PLAGIORCHIIDAE) IN LYMNAEID SNAILS FROM FRESHWATER
ECOSYSTEMS IN CENTRAL EUROPE REVEALED BY MOLECULES AND
MORPHOLOGY**



RNDr. Thesis

Bc. Jana Zikmundová

České Budějovice
2014

Zikmundová, J., 2014: Species diversity of *Plagiorchis* Lühe, 1899 (Digenea: Plagiorchiidae) in lymnaeid snails from freshwater ecosystems in central Europe revealed by molecules and morphology. RNDr. Thesis, in English – 18 p., Faculty of Science, University of South Bohemia, České Budějovice, Czech Republic.

ANNOTATION

This study applies molecular and morphological approaches addressing the identification of morphologically similar larval stages (cercariae) of *Plagiorchis* spp. (Digenea: Plagiorchiidae) parasitising lymnaeid snail populations in the freshwater ecosystems of central Europe. Five morphologically homogeneous and genetically distinct lineages of *Plagiorchis* spp. were identified *via* matching molecular data for the mitochondrial *cox1* gene with detailed morphometric data. Phylogenetic and comparative sequence analyses using partial 28S rDNA and ITS1-5.8S-ITS2 sequences allowed molecular identification of three species (*P. elegans*, *P. maculosus* and *P. koreanus*) *via* matching sequences from larval and adult digenean stages. A key for the identification of the cercariae of *Plagiorchis* spp. parasitising lymnaeid populations in central Europe is provided.

DECLARATION

Prohlašuji, že svoji rigorózní práci jsem vypracovala 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.

THIS THESIS IS BASED ON THE FOLLOWING PUBLICATION:

Zikmundová, J., Georgieva, S., Faltýnková, A., Soldánová, M. & Kostadinova, A. (2014) Species diversity of *Plagiorchis* Lühe, 1899 (Digenea: Plagiorchiidae) in lymnaeid snails from freshwater ecosystems in central Europe revealed by molecules and morphology. *Systematic Parasitology*, 88 (1), 37–54. (IF = 1.260)

AUTHOR'S CONTRIBUTION

The co-authors listed below fully acknowledge that Jana Zikmundová contributed substantially to the above publication. She participated in the field sampling and the preparation of isolates for morphological and molecular analyses; obtained the morphometric data and performed statistical analyses; carried out molecular characterisation (DNA isolation, amplification, sequencing, assembling) for a substantial part of the isolates; participated in phylogenetic analyses and data interpretation and drafted the manuscript.

Agreement of the co-authors

Anna Faltýnková Aneta Kostadinova Simona Georgieva Miroslava Soldánová

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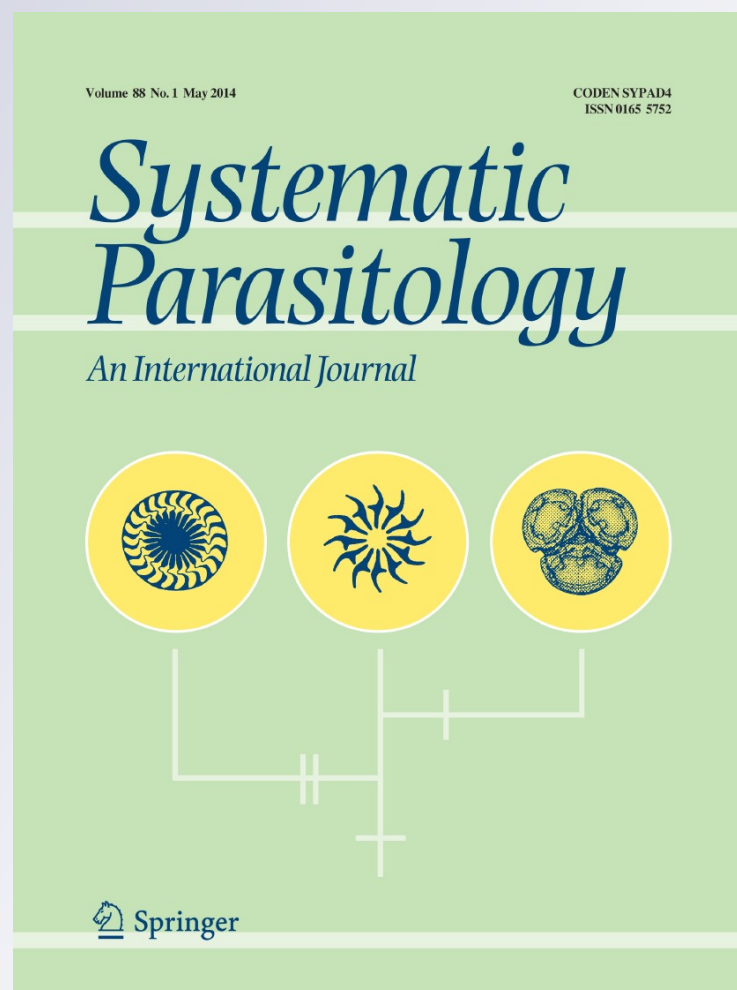
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Species diversity of *Plagiorchis* Lühe, 1899 (Digenea: Plagiorchiidae) in lymnaeid snails from freshwater ecosystems in central Europe revealed by molecules and morphology

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Abstract Larval stages of *Plagiorchis* spp. are both ubiquitous and ecologically important parasites in snail populations of freshwater ecosystems in Europe. However, difficulties in distinguishing the morphologically similar cercariae used for species identification, may lead to underestimation of species diversity. In this study, 38 isolates of *Plagiorchis* spp. infecting two lymnaeid snails, *Lymnaea stagnalis* (L.) and *Radix auricularia* (L.), in five central European freshwater ecosystems were subjected to morphological and molecular assessment. Five morphologically homogeneous and genetically distinct lineages of *Plagiorchis* spp. were identified *via* matching molecular data for the mitochondrial cytochrome *c* oxidase subunit I (*cox1*) gene with detailed morphological and morphometric data of the cercariae. Comparative sequence analysis using partial 28S rDNA and ITS1-5.8S-ITS2 sequences revealed that three distinct *cox1* lineages are conspecific with *Plagiorchis elegans* (Rudolphi, 1802), *P. maculosus* (Rudolphi, 1802) and

P. koreanus Ogata, 1938, respectively, whereas the lineage identified based on cercarial morphology as *P. neomidis* Brendow, 1970 plus a single isolate that could not be assigned to a described species, did not match any of the available sequences for *Plagiorchis* spp. A key to the cercariae of *Plagiorchis* spp. parasitising lymnaeid populations in central Europe is provided to facilitate identification.

Introduction

Plagiorchis Lühe, 1899 is the type- and perhaps the most speciose genus of the family Plagiorchiidae Lühe, 1901. Species of *Plagiorchis* utilise a three-host life-cycle using lymnaeid snails as first intermediate hosts, aquatic insects and freshwater crustaceans as second intermediate hosts, and birds and mammals, accidentally amphibians and reptiles, as definitive hosts.

Larval stages of *Plagiorchis* spp. are both ubiquitous and ecologically important parasites in snail populations of freshwater ecosystems in Europe (e.g. Faltýnková et al., 2007; Soldánová et al., 2011). Notably, a single species, *Plagiorchis elegans* (Rudolphi, 1802), is among the most frequently recorded in the inventories of larval trematodes of *Lymnaea stagnalis* (L.) in Europe (Väyrynen et al., 2000; Faltýnková, 2005; Faltýnková & Haas, 2006; Žbikowska et al., 2006; Žbikowska, 2007; Faltýnková et al., 2007). This species has been recognised in these

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