

Species delimitation of the genus *Trentepohlia* (Ulvophyceae, Chlorophyta) in Central Europe using molecular and morphological approaches

Introduction

Trentepohliaceae is a group of subaerial green algae in otherwise predominantly marine or freshwater class Ulvophyceae. These algae often form conspicuous and brightly coloured layers on natural as well as artificial substrata. Some of the taxa are parasites of ornamentally or agriculturally important plants (Almeida, 1985), some are just epiphytes. The genus *Trentepohlia* is the richest one on species. Although the family Trentepohliaceae belongs to the Chlorophyta lineage, *Trentepohlia* possess some features typical for the lineage Streptophyta (MLS-like structures, phragmoplast present in cytokinesis; Chapman et al., 2001). It is also known for its production of large amounts of carotenoids and polyhydroxyalcohols (Boedeker et al., 2013) and it enters symbiotic relationships with large variety of fungi thus forming lichens (Nelsen, 2011).

This genus has been intensively studied for about two hundred years now, being already mentioned by Linnaeus (1753) for the first time. All these years, the main focus has been directed mainly towards the morphological criteria, such as the length and width of cells, shape of cells, shape of reproductive structures or substrata colonized etc. (Printz, 1964; Brand, 1902; Fischer, 1922). However, in the last two decades, first molecular data were obtained proving large inconsistencies in molecularly and morphologically delimited species (López-Bautista et al., 2006). Many of the traditional morphological criteria used for species delimitation rendered without phylogenetic significance and some of the species were marked as complexes of cryptic species rather than individuals (Rindi et al., 2009a). It is now clear, that it is no longer sufficient to focus separately on morphology or phylogeny. It is however advantageous to use both approaches and study as many aspects as possible.

In this Master's thesis I put emphasis on both molecular data and detailed morphological observation for I believe it is the best way to elucidate the current situation. To do that, I carry out a complex methodical approach. Firstly, I focus on the original descriptions of the European *Trentepohlia* species written mainly in the 18th and 19th century. Secondly, I study the morphology of fresh natural samples collected within Europe. I also maintain some cultures of these specimens, although they serve more as a demonstration of a common problem of many algae, changes of their morphology in the culture (Lakeman, 2009). For the fresh samples, I perform molecular analyses using chloroplast (*rbcL*) markers. In this study, the cloning method, inserting of the DNA sample into bacterial plasmids to separate contaminants from trentepohlialean DNA, will be used for the first time in the history of *Trentepohlia* research. It will help to answer some burning question, which still remain unclarified: Firstly, do the crusts and tufts formed by *Trentepohlia* consist of one or more species? This aspect has never been considered by the authors collecting *Trentepohlia* into cultures, though being of a great importance. Cloning revealed the existence of common mixture of species in what on the first sight appears to be a homogeneous crust. Secondly, Should all of the current European species according to Brand (1902), Fischer (1922) and Printz (1939) really be considered valid to date? After a careful examination of the oldest literature I concluded that some of the species cannot be labeled as valid anymore. The last but certainly not least goal of this thesis is to assign the most striking morphological features to particular phylogenetic lineages. The results will help to enlighten our current state of knowledge of the still enigmatic relationships among species of *Trentepohlia*.

