

Advancing the science of algal taxonomy

“The primary aims of taxonomy are to name, circumscribe, describe and classify species. The first goal is convention but the remainder are science” (Seberg et al. 2003, p.63).

The science of algal taxonomy has come a long way, and represents a vast body of work that has accumulated over the past two and a half centuries. Important progress in algal taxonomy has often been accompanied by technological or analytical progress, including advances in light and electron microscopy, culture techniques, and digital access to collections (De Clerck et al. 2013).

Unquestionably, the main revolution in recent decades has come with the application of DNA sequence analysis. The use of molecular data has pointed phycologists toward more realistic species boundaries, and has resulted in the discovery of hundreds of new species. These data have profoundly reshaped our understanding of algal diversity and distributions, and exemplifies our inability to accurately assess species boundaries in algae based on morphological features alone (Leliaert et al. 2014).

Molecular approaches to species delimitation in algae are increasingly used, and have become the rule rather than the exception in assessments of algal species-level diversity (De Clerck et al. 2013). The use of molecular markers have been particularly valuable for assessing species boundaries in closely related taxa where morphological species delimitation is difficult or impossible. Molecular analyses have uncovered numerous cryptic species, and have revealed that widely distributed taxa may actually comprise different species with more restricted ranges, illustrating that an accurate understanding of species limits is essential for deducing fundamental processes in biogeography and evolution of algae (e.g., Vieira et al. 2017, Leliaert et al. 2018, Pinseel et al. 2020). The past decade has also witnessed a progress in methods and criteria used for species delimitation. Early studies frequently used some kind of subjective threshold, such as genetic distance. The conception that species are separately evolving metapopulation lineages (Wiley 1978, De Queiroz 2007) along with progress in phylogenetic and population genetic analyses has led to the development of an array of model-based methods aimed at making DNA-based species delimitation more objective and repeatable (Camargo and Sites 2013, Leliaert et al. 2014, Dellicour and Flot 2018). These methods have also found their entry in phycological research (e.g., Payo et al. 2013, Malavasi et al. 2016, Montecinos et al. 2016, Díaz-Tapia et al. 2020). It is clear, however, that uncertainties about species boundaries are inevitable, especially in recently diverged lineages, due to the very nature of speciation, which is a process and not a single event in time. These uncertainties in DNA-based species delimitation, along with conflicts between phylogenetic and traditional species definitions, and problems with linking DNA sequences to existing species names, has made phycologists hesitant about describing new species based solely on DNA sequence data. This is one of the reasons why taxonomic studies, in which new species are formally described, are often lagging behind DNA-based species diversity studies (De Clerck et al. 2013). Several researchers have advocated the importance of an integrative approach to taxonomy in which, in addition to molecular data, other lines of evidence, such as morphological, ecological, and breeding data, are required for delimiting and describing species (e.g., Darienko et al. 2015). Others have argued that DNA sequence data serve as a reliable source for testing species boundaries even in the absence of additional phenotypic evidence (Leliaert et al. 2014). In any case, phenotypic data are extremely valuable in providing additional biological evidence for defining species boundaries, and therefore remain essential in taxonomic studies. Finally, the formal description of new species is important. Molecular studies where newly discovered species are not described will result in lineages that remain unnamed, and species diversity that will not be recognized in the Linnaean taxonomic system in which we operate, and which underpins all biological research and associated applied fields (Godfray and Knapp 2004).

The study by Hoshino et al. (2021), published in this issue, is an excellent example of how phycology has embraced DNA-based species delimitation methods, combined with other lines of biological evidence, including morphological, biogeographical and breeding data, to arrive at new taxonomic conclusions. The paper focuses on *Scytosiphon lomentaria*, a common brown seaweed from intertidal pools and subtidal habitats with a wide geographical distribution in cold to warm temperate waters of both hemispheres (Guiry and Guiry 2021, GBIF 2021) (Fig. 1). The genus and species have a confounded taxonomic history with many species and infraspecific taxa that have been described based on subtle differences in thallus form or dimensions. Detailed field and culture observations have revealed that much of this morphological diversity can be attributed to environmentally induced plasticity, and as a result several of these species have been placed into synonymy with the morphologically variable *S. lomentaria* (Clayton 1978). More recently, molecular phylogenetic studies have shown that this species actually consist of distinct genetic lineages, suggestive of multiple cryptic species (Cho et al. 2007, Contreras et al. 2007, Kogame et al. 2015b, Kogame et al. 2015a, McDevit and Saunders 2017). A species complex was further supported by crossing experiments between isolates of different Japanese lineages, showing the existence of pre- or post-zygotic reproductive barriers (Hoshino et al. 2018). In Japan, five clades of *S. lomentaria* have been recognized, but associating these clades with species names remained tentative.

Hoshino et al. (2021) build on these studies by considerably increasing taxon sampling in Japanese waters. Two unlinked genes, *cox1* and *rbcl*, were analyzed from more than 200 specimens collected from various localities in Japan and the Russian Far East. Species boundaries were tested using Poisson Tree Processes (PTP), which delimits species by modelling speciation events in terms of number of substitutions on a phylogenetic tree (Zhang et al. 2013). In total six putative species were delimited, including the five previously detected species in Japan, in addition to a newly detected lineage from the subtropical Okinawa Island. Linking these six lineages to existing species names and deciding on whether or not lineages actually represented new species were one of the main challenges of the study, and required a thorough revision of the genus, including all previous heterotypic synonyms of *S. lomentaria*. A first important question to resolve was which of these six lineages corresponded to *S. lomentaria*, as several lineages have been reported from the type region in the north-eastern Atlantic (Kogame et al. 2015b). By combining available morphological, geographical, and phylogenetic data (McDevit and Saunders 2017), the authors were able to pinpoint the *bona fide* *S. lomentaria*, as well as that of the widely distributed *S. promiscuus*. Hoshino et al. (2021) further moved the field of algal taxonomy forward by describing the remaining four lineages as new species with molecularly defined types. Although some morphological features could be discerned between the six species in Japan, morphology was found to be a poor indicator of species boundaries due to extensive intraspecific variation, reinforcing the notion that DNA sequence data are essential for accurate identification of *Scytosiphon* species.

The paper also highlights once again that misconceptions about species boundaries impact on our understanding of distributions and evolution of seaweeds. Geographical ranges were found to differ markedly between species with three species (including *S. lomentaria*, *S. promiscuus*, and the newly described *S. shibazakiorum*) being widely distributed, while the others appear restricted to Japan. Except for the subtropical Okinawan species *S. subtropicus*, the temperate Japanese species have overlapping ranges. The former was found to easily form hybrid zygotes with the other species, although the zoospores from the hybrid sporophytes appeared to have low survival rate. Crossing experiments further showed that gametic incompatibility (pre-zygotic barrier) was less developed between the allopatric species (*S. subtropicus* and the five others) compared with the sympatric temperate species. Hoshino et al. suggested that these pre-zygotic barriers may have evolved among the geographically co-occurring species due to reinforcement, an evolutionary process in which natural selection strengthens pre-zygotic isolation as a response to unfit hybrid offspring between sympatric species. Reinforcement has only been studied in a few seaweed groups (Hoarau et al. 2015, Lipinska et al. 2016), but may be a common speciation mechanism in algae.

The paper of Hoshino et al. (2021) is certainly not an endpoint for the taxonomy of *Scytosiphon*. The genus is globally distributed and several regions have not been sampled molecularly. Given the discovery of multiple species within *S. "lomentaria"*, it can thus be expected that unexplored regions contain additional unrecognized diversity. In addition, analysis of multi-locus data will likely further refine our view on species diversity, especially in recently diverged lineages. For example, within *S. shibazakiorum*, a genetically highly variable and widespread species for which species delimitation was found to be uncertain, additional nuclear gene data may result in the recognition of less inclusive entities, which will further our understanding of the biogeography and evolution of species within this group.

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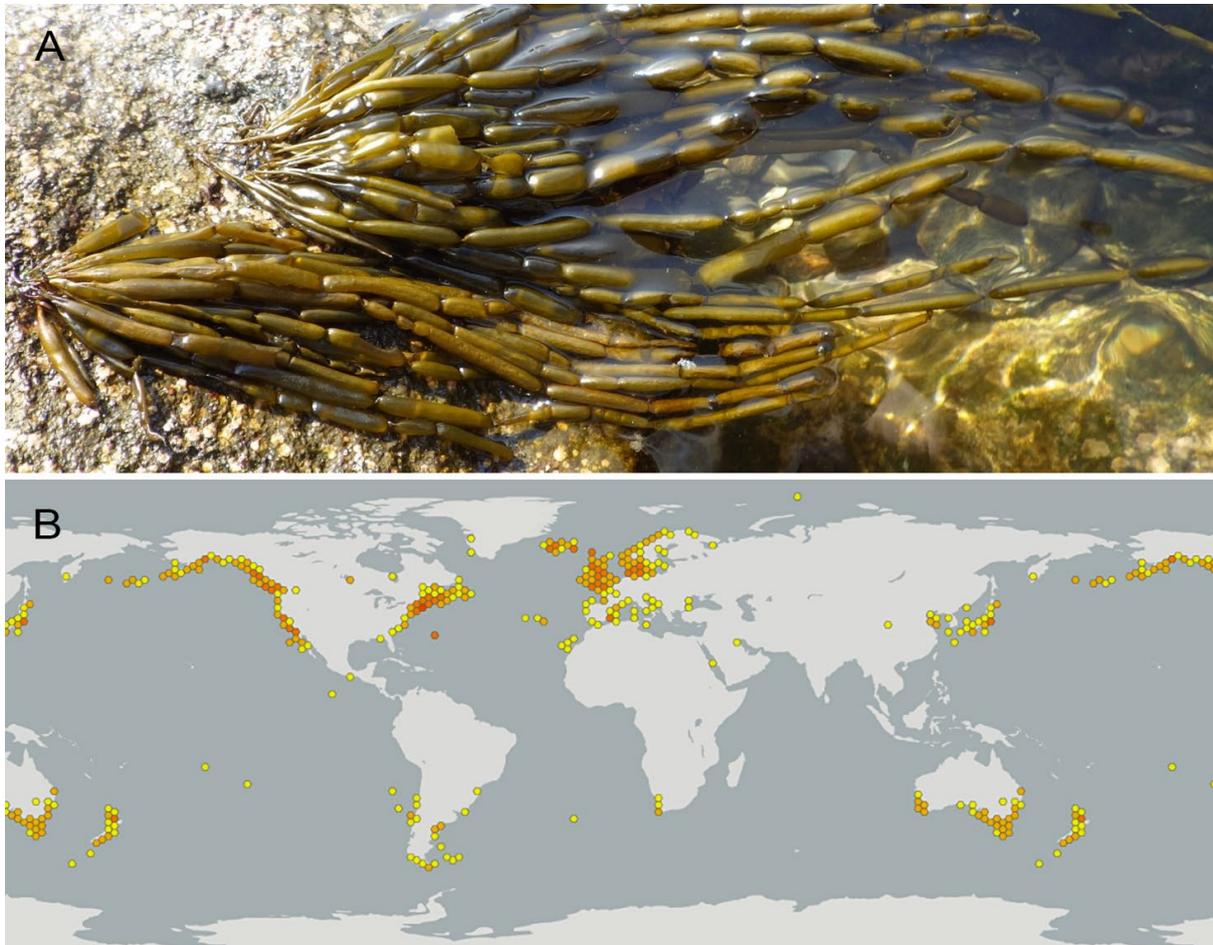


Fig. 1. A. *Scytosiphon lomentaria* growing in an intertidal rock pool in Enoshima, Kanagawa Pref., Japan. Photo by Masakazu Hoshino. B. Geographical distribution of *Scytosiphon* "lomentaria" in the broad sense based on preserved specimens, literature, and observation data (GBIF 2021).