Research News For U (RNFU) ISSN: 2250 –3668, Vol. 7, 2012
Available online http://www.doublehelixresearch.com/RNFU

DNA barcoding as a tool for algal species Identification and diversity studies

K. Sasikumar[1] and C. Anuradha[2]

[1]H.H. The Rajah's College, Pudukottai, Tamil Nadu, 622 001, India.

[2]National Research Centre for Banana, Trichy, Tamil Nadu, India.

In recent years there has been a drive to speed up the rate at which species on earth are identified and described in response to the diversity of life which is disappearing at an ever increasing rate. A major initiative that has been advocated to achieve this goal is DNA barcoding, i.e., sequencing a short, diagnostic segment to discriminate between species. Until now, the emphasis of DNA barcoding has been on animals, now, however, the algae and the higher plants are being considered for DNA barcoding and the search is on for molecular markers which can be successfully used across a wide spectrum of organisms for DNA barcoding. DNA barcoding is a valuable tool, especially when coupled with traditional taxonomic tools and is fundamental in revealing hidden diversity.

Although algae have a long history of study, they are still poorly understood taxonomically and phylogenetically. New species are discovered each year and molecular investigations have revealed many cryptic species and unexpected phylogenetic relationships. Identification of algae can be difficult because many species lack obvious structural features and some of the observable characteristics are variable within species. Often, diagnostic characteristics are only detectable using light or electron microscopy, taxonomy of algae is somewhat inaccessible to non-specialists and rapid identification of some species even by microscopy is impossible. DNA barcodes would allow non-specialists to identify algae consistently and rapidly regardless of life stage. They would also make it possible for researchers to identify suboptimal material and potentially recognize cryptic species or intraspecific variation correlated to geographic distribution.

Molecular barcoding methods have been applied to land plants and a number of algal groups. In practice, however, researchers are using 'barcodes' which includes at least one molecular marker and taxonomists now routinely use DNA markers to distinguish species of algae. The ideal DNA barcode would be sufficiently variable to distinguish among closely related species. The marker should be found in all taxa, amplify using universal primers and sequence cleanly. Finding an ideal molecular barcode for all algae is complicated by the fact that algae are an ancient and diverse lineage. Although little is known about the arrangement of their nuclear genomes it is now clear that organellar genomes in algae vary considerably in content and organization. Several loci have been proposed for algae and land plants. The 5' end of cytochrome oxidase I (COI) is widely used in animals and some algal lineages including red algae, brown algae and some diatoms. However, COI can be difficult to amplify from some taxa. In green algae, this gene can contain

several introns (e.g., five in *Chaetosphaeridium*) although the position and number of introns in the gene are not known for most species.

The internal transcribed spacer (ITS) of the nuclear ribosomal operon also has been proposed as a DNA barcode for algae and land plants and has been widely used in species-level phylogenetics of green algae and at deeper nodes. The locus lies between the large and small nuclear ribosomal subunit genes and includes the 5.8S rDNA gene. The length of the ITS varies from a few hundred to more than a thousand base pairs depending on the taxon. Interpretation of ITS phylogenies is not always straight forward due to within-species variation, difficulties with alignment and determination of orthology, although incorporation of information from secondary structure and compensatory base changes holds promise.

A third barcode has been recently proposed for plastid-containing organisms: a portion of the chloroplast ribosomal large subunit (23S), sometimes called the Universal Plastid Amplicon (UPA). This locus has not been broadly sampled and recent investigations suggested that it is less variable than other markers, including other chloroplast. The plastid encoded rbcL gene has been widely used for phylogenetic inference in green algae. The 3' end of *rbcL* may be sufficiently variable to identify species of marine green algae. Truly universal primers are not available for *rbcL*, although some primers can be used on a wide range of taxa. The plastid trnH-psbA is being proposed as possible markers for flowering plants which can also be used in the red algae.

Several additional molecular markers have been developed to address phylogenetic questions in various groups of green algae. One of these, *tufA*, is generally conserved and has been investigated in marine algae. Although tufA may be useful as a DNA barcode in chlorophytes, the gene was transferred from the plastid to the nuclear genome in some lineages of charophytic algae and in land plants and in a few cases a copy is maintained in each genetic compartment.

Genetic barcoding will thus not signal the end of taxonomy for phycologists, but will initiate a revolution of molecular assisted alpha taxonomy that will greatly change the number and distribution of species that are recognized in this lineage. It is not, however, a tool that should be used in isolation, particularly during the development stages when it will be desirable to accompany the molecular results with thorough anatomical observations, and in the case of closely related species where it will be prudent to assess the mitochondrial data with nuclear markers to search for introgression, hybridization and incomplete species boundaries.