MEETING REPORT

Molecular systematics*

The use of molecular data in phylogenetic systematics and its implications for plant classification and evolution has been well established since the 1980s. The development of polymerase chain reaction (PCR), cloning, automated sequencing technologies, New Generation Sequencing (NGS) and sophisticated instrumentation, as well as diverse computational evolutionary approaches have provided the exciting possibility of establishing inter-relationships of all plant groups.

Molecular systematics encompasses a series of approaches in which phylogenetic relationships are inferred using information from macromolecules of the organisms under study. Sequences of the chloroplast, nuclear and mitochondrial genomes are analysed independently and/or in combination for inferring phylogenetic relationships at different hierarchical levels of angiosperm groups.

Molecular studies on plant systematics have been extensively done in developed countries. India is one of the richest countries of the world as far as biodiversity is concerned. However, not many studies on molecular systematics have been done, partly due to lack of training. Hence a workshop was held recently to provide basic training on molecular techniques and phylogenetic analyses to practising taxonomists. A total of 32 participants from 10 states of India participated in the workshop.

The inaugural session introduced the need and relevance of a workshop on molecular systematics. It was pointed out that diverse molecular approaches are now available to plant taxonomists for phylogenetic inference, and that the development of techniques in molecular biology has opened new avenues for the study of phylogenetic relationships of different plant groups at the molecular level. These approaches have unlocked the treasure chest of information on the evolutionary history of organisms. It was advocated that such workshops need to be organized in different parts of the country to train taxonomists. The need for integrating data from various sources in systematic studies was discussed. It was pointed out that some remarkable results have been obtained by the use of techniques that provide data on DNA sequences, and methods to analyse the complex phylogeny.

The inaugural session was followed by a lecture on concepts, methods and applications of molecular data in systematics. Several examples were cited to illustrate the use of a combination of nuclear and chloroplast DNA sequences to ascertain the patterns of evolution within a genus. The establishment of DNA banks of Indian plants was encouraged.

This was followed by a lecture on DNA-based molecular markers comprising an illustrated account of the basic tools that are extensively used for developing such markers, and discussion of various techniques and strategies deployed for designing DNA-based molecular probes.

The second half of the second day was devoted to practical aspects of molecular phylogenetic studies. An introduction to DNA extraction and detailed account of different protocols used in genomic DNA extraction was followed by a video to demonstrate DNA extraction methods.

This presentation was followed by a lecture on PCR with explanations of the mechanism of PCR, including denaturation, annealing and extension processes along with detailed descriptions of the ingredients – primers, *Taq* polymerase, dnTPs, buffers and DNA template.

This was followed by explanations of the use of methods of reading and editing chromatograms, as well as introduction to GenBank searches and sequence alignment. Sequence alignment using different software was demonstrated to the participants using computers, followed by practical exercises using different software.

The second day was devoted to the phylogenetic analyses of data. The first lecture was on cladistics, in which the terms used therein were explained and several examples were cited where the cladistic methodology has been used for phylogenetic inferences.

This was followed by a lecture on morphology and molecular markers and techniques for their analysis. The presentation emphasized the use of combined analysis of both morphological and molecular data. This lecture was followed by an interactive discussion.

The issue of how to initiate molecular systematics studies in India was discussed. Although excellent floristic studies have been done by taxonomists in India, not many of them are trained in molecular techniques to integrate the DNA sequence data. The advantages and disadvantages of plastid, nuclear and mitochondrial genomes in phylogeny reconstruction were also discussed.

On the third day, the lecture on phylogenetic methods included an overview of maximum parsimony, maximum likelihood, Bayesian inference, and other methods of analysis and tree-building. This was followed by a lecture (live virtual presentation) on understanding and reading phylogenetic trees.

This was followed by laboratory exercises, viz. retrieving sequences from GenBank, sequence alignment, construction of phylogenetic trees and character reconstruction on computers. The workshop provided several occasions for interaction between participants and resource persons.

In the valedictory session, emphasis was on the importance of traditional and molecular taxonomy, and the need for training of students and college teachers in the molecular techniques for use in the systematics of India taxa. The participants and resource persons shared their experiences.

The major outcome of the workshop was the hands-on training given to the participants, especially on using different software for phylogenetic analyses.

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