Molecular systematics*

Our understanding of the relationships among organisms at various levels of the Tree of Life has been advanced greatly in the last decade with the aid of DNA molecular systematics techniques and phylogenetic theory. Molecular systematics encompasses a series of approaches in which phylogenetic relationships are inferred using information from macro-molecules of the organisms under study. The type of molecular data acquired includes those from DNA sequences, DNA restriction sites, microsatellites, RAPDs and AFLPs.

Molecular systematic studies have primarily been done in the developed countries. With the rich biological resources and available floristic expertise in many developing countries, it will greatly advance the field of systematics and related sub-disciplines if more scientists and students in developing countries are involved in molecular systematic studies along with other integrative systematics investigations. India is one of the richest countries of the world as far as biodiversity is concerned. However, not many molecular studies have been carried out due to lack of proper training in the field of molecular systematics. The present workshop was organized with a view to provide basic training on molecular techniques to taxonomists. This was the first molecular systematics workshop on flowering plants organized in India.

The workshop comprised a total of 30 participants covering 13 States of India and Nepal. It was organized under the auspices of the Indian Association for Angiosperm Taxonomy (IAAT).

In the inaugural session, Arun K. Pandey (Course Coordinator) introduced the theme of the workshop. He pointed out that application of molecular data in systematics and its implications in plant classification and evolution are now well established. With the development of PCR, cloning, automated sequencing machines, and other techniques and instrumentation, it has now become possible to realize the dream of establishing the relationship of different plant groups at DNA level. Molecular systematists have unlocked the treasure chest of information on evolutionary history of organisms. Using conserved molecular sequences, it is possible to define and identify most species. DNA data also allow investigation of the evolutionary patterns of diversity.

The workshop was inaugurated by K. K. Mandal (Pro-Vice-Chancellor, TM Bhagalpur University (TMBU), Bhagalpur). He spoke about the importance of DNA taxonomy and its role in genome mapping and conservation of biodiversity.

The keynote address was delivered by Rosabelle Samuel (Department of Systematic and Evolutionary Botany, University of Vienna) on application of molecular markers at population and systematic levels. She gave an overview of molecular systematics and outlined its importance in evolutionary studies and conservation of biodiversity. While recognizing the rich phytodiversity in India, Rosabelle Samuel advocated the need for molecular systematic studies on Indian taxa. She shared her experiences on working with tropical plants. She said that although molecular systematics is an extremely expensive enterprise, scientists in the developing countries can do better in collaboration with those from the developed countries. Rosabelle Samuel further pointed out that molecular systematics can be useful not only in plant identification and genealogical history, but also in understanding its phytogeography which can ultimately help in conservation of biodiversity. She also released the souvenir published by the Plant Systematics Research Centre, TMBU.

After the inaugural session, Arvid K. Misra (North Eastern Hill University, Shillong) delivered a lecture on DNA-based molecular markers. He gave an illustrated account of the basic tools that are extensively used for developing such markers. He discussed various techniques and strategies deployed for designing DNA-based molecular probes.

Deepthi Yakandawala (University of Peradeniya, Sri Lanka) delivered a lecture on the molecular systematic study of the genus Asparagus, inferred from both morphological and molecular data. She emphasized the use of combined analysis with both morphological and DNA data. H. S. Kathiriaarchchi (University of Colombo, Sri Lanka) explained the significance of molecular studies in the family Euphorbiaceae sensu lato. She highlighted the role of nuclear and plastid sequences in recognizing a separate family, Phyllantheaceae.

On the second day, Rosabelle Samuel delivered a lecture-cum-demonstration on DNA extraction. She gave a detailed account of different protocols used in genomic DNA extraction. Her lecture was followed by an interactive discussion and laboratory exercises. Participants prepared extraction buffers and extracted genomic DNA from fresh and silica-dried leaves using CTAB method and extraction kits.

Pandey discussed how to initiate DNA molecular systematic studies in developing countries. He said that contributions from colleagues in developing countries can offer many new opportunities to investigate questions on evolutionary diversification and conservation at the global scale and in many more plant groups.

In another lecture, Misra emphasized that modern tools of DNA analysis can detect differences at single-nucleotide level and sequences with such differences are then said to have single nucleotide polymorphism (SNP). Classical evolutionary studies have been concentrating on phenotypic differences. SNP studies can go beyond this level and can therefore differentiate between two apparently similar organisms.

On the third day, Kathiriaarchchi delivered a lecture on PCR. She explained the mechanism of PCR and provided a detailed account of primers, Taq polymerase, dNTPs, buffers and DNA template. She also discussed the mechanism of denaturation, annealing and extension process during PCR reactions. This was followed by laboratory exercises on PCR reactions and verification of PCR products. Kathiriaarchchi emphasized the role of primer design in effective amplification of genomic DNA.
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Rosabelle Samuel delivered a lecture on Amplified Fragment Length Polymorphism (AFLP). While explaining the importance of AFLP studies, she talked about taxa and sampling methodology, protocols involved in AFLP and data analyses. Unlike RAPDs, AFLP data are highly reproducible. The AFLP technique produces large sets of polymorphic markers that may be used to analyse closely related taxa. She highlighted the importance of AFLP data in the study of polyploidy evolution. She also demonstrated protocols for cycle sequencing.

D. Yakandawala introduced the methodology of GenBank search. Her lecture was followed by a panel discussion which was jointly conducted by Rosabelle Samuel, Deepthi Yakandawala, Harshendra and Pandey. Sequence alignment using different software was demonstrated to the participants using computers.

On the fourth day, Deepthi Yakandawala delivered a talk on PAUP (Phylogenetic Analysis Using Parsimony). She gave a detailed account of cladistic analysis using both morphological and molecular data. She presented an overview of maximum parsimony, maximum likelihood, Bayesian inference and other methods of analysis and tree-building. This was followed by laboratory exercises on computers.

On the final day of the workshop, Rosabelle Samuel presented two talks, viz. ‘Genus Leonotodon (Asteraceae) is biphyletic’ and ‘Molecular phylogeny of Ebaeaceae’. The lectures were followed by interactive sessions. Participants were also given training on writing papers on molecular phylogenetics and data analysis. All the participants were provided with handouts related to extraction procedure, amplification, sequencing, sequence alignment and phylogenetic analysis. The workshop was interactive and deliberations were active and lively. The workshop was productive and provided several occasions for interaction between participants and resource persons.

In her valedictory lecture, Prema Jha (Vice-Chancellor, TMBU, Bhuagulpur) emphasized the need for molecular systematic studies in developing countries and development of DNA banks of plants in India. She emphasized that the sequence data of Indian plants, when deposited in GenBank, can be retrieved when needed and can solve the problems of patents of Indian plants and products. In the valedictory session, the participants and resource persons shared their experiences.

The major outcome of the workshop was the practical training given to the participants and first-hand information on various aspects of molecular systematics. The workshop also provided an opportunity to the young and senior taxonomists to learn various techniques of extraction, amplification, sequencing, sequence alignment and phylogenetic analysis. Participants were of the view that several such workshops are needed to train young Indian taxonomists in cutting-edge technology.

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MEETING REPORT

Semiochemicals in crop protection*

Inaugurating the Annual meeting, M. S. Swaminathan (MSSRF, Chennai) highlighted the need to improve the economic viability of farming, protection and improvement of land, water, biodiversity and climate resources for sustainable advances in productivity, profitability and stability of major farming systems. Emphasizing the need to strengthen the areas of conservation and green agriculture, he visualized the need to strengthen strategic research with a focus on biotic and abiotic stresses, ensure environmental sustainability, biotechnology, biosafety and biotechnology research, monitoring and strengthening research in the areas of semiochemical technology, nanotechnology, space and information technology, renewable energy technologies, besides strengthening research on technology-delivery systems to accelerate progress in bridging the growing gap between scientific as well as field-level know-how.

Strengthening the ecological foundation for sustainable agriculture, extension and anticipatory research is an immediate necessity, with the establishment of a few national centres of excellence in agriculture.

Briefly outlining the theme of the meeting, T. N. Ananthakrishnan (Chennai) highlighted that semiochemicals are increasingly being integrated with a range of methods producing new schemes, with the application of semiochemicals being increasingly utilized for pest monitoring, mass trapping, mating disruption and natural enemy attraction. A bewildering array of secondary plant substances has been identified through diverse biosynthetic pathways, so that increased interest is being evinced on the impact of chemical communication between crop plants and insects. Compared to what has come to be known as the ‘supermarket utopia of monoculture’, prone to insect outbreaks, the significance of polyculture has come to be appreciated, with newer technologies involving genetic modification and molecular marker associated selection.

Stressing the need for better appreciation of the third component to host plant resistance, apart from vertical and horizontal ones, the need for better understanding of the third trophic level involving parasite/predator attracting semiochemicals, besides diverse blends of volatiles emitted by insect-damaged plants attracting natural enemies, has been highlighted. A better understanding of odour receptors, odour-binding proteins and

*A report on the Seventh Annual Discussion Meeting on ‘Semiochemicals in Crop Protection: Ongoing Technologies’ convened by T. N. Ananthakrishnan on 2 December 2006 at Centre for Cooperation in Science and Technology Among Developing Societies, Chennai.

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