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TAXONOMY AS RELATED TO GENETIC DIVERSITY IN PLANTS

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The main needs of classification are two-fold: (1) to categorise organisms so as to be able to identify individuals and groups for future references and (ii) to identify and subsequently predict inter relationships between putative groups and to suggest, if possible, the mode of evolution. Earlier classifications, even upto the 1950's, involved the weighting of characters. Various characters or clusters of characters were taken into account.

Of the different concepts proposed, *phenetics* takes into account the overall similarity. Similar organisms are grouped together and consequently, the origin of a group may be polyphyletic. *Cladistics*, however recognises only the genealogical relationships and thus only monophyletic group and sister group relationships are maintained. The *traditional or evolutionary* systematists follow the same principle as the pheneticists in grouping similar organisms together, thus recognising paraphyletic or polyphyletic groups. For general purpose classification, the commonality of characters used by pheneticists is preferred since these are verifiable. However, in the absence of sequenced fossil data, evolutionary homologies are not recognised by some taxonomists.

The post-Darwinian synthetic theory of evolution arose through the integration of cytology, genetics, population genetics, statistics, field experimentation and observational systematics. The biological species concept regarded the species as evolutionary units held together by gene flow and separated by genetical barriers from such other units. The recommendations by the United Nations Conference on Human Environment held in 1972 for the survey, collection and conservation of plant genetic resources gave a new impetus to this aspect of taxonomy.

The initial stress on comparative morphology and floristics was supplemented by data from anatomy, embryology and fossil records. Experimental approaches to classification used the observational investigative methods of cytology, the hybridisation and progeny

testing of genetics and then the ecological aspects. Later aids to taxonomy included relatively minor morphological characters, such as seed coat and pollen grain, together with the analysis of ultrastructure, protein and amino acid patterns, chemical components (chemotaxonomy) etc. The methods for cytology - genetics - ecological studies formed the basis of biosystematics. The emphasis shifted from the consideration of characteristics of an individual to those of a population and from the concept of a static situation to dynamic ecological changes.

The initial criteria for the detection of genetic diversity included hybridisation, meiotic behaviour, progeny analysis, intercompatibility, reconstruction and hybridisation of polyploid species. The study of chromosome number was replaced by karyotype analysis, followed by identification of individual chromosome segments through banding pattern analysis, - the G, Q, R, C, O, N and CT bands. These advances in methodology led to the golden era of cytotaxonomy.

Karyotype studies were principally based on the idea that symmetrical karyotypes were more primitive than asymmetrical ones; longer chromosomes than shorter ones; median centromeres with chromosome arms of equal length were more primitive than chromosomes with arms of unequal length; low basic numbers had given rise to higher ones. These criteria were based on the comparison of karyotypes of known relative antiquity, as determined through classical taxonomy. These methods were utilised extensively. Some instances of their use were in relation (i) to the different theories of the putative origin of the monocots; (ii) to the creation of Agavales as a separate group from Liliales and Amaryllidales and in the delimitation of the family Commelinaceae. All the work was however within the framework of the existing systems of classification. Despite the very large number of publications on cytotaxonomy, particularly between 1950 to 1970, following the advent of the squash

technique, there was no dramatic alteration to the principal systems of classification. The cytotaxonomical studies have only added to the existing knowledge and aided in clearing up controversial issues. I would like to cite some examples from the work carried out on monocotyledons by our workers during the past 40 years and more.

(i) In the evolution of monocotyledonous groups, the main controversies deal with the ancestral status and the homogeneity of the putative ancestors. According to Hutchinson, monocots have possibly been derived from the Helobiales - considered to be a homogenous group with aquatic habitat. In Engler's system, on the other hand, Pandanales, consisting of *Pandanus*, *Typha* and *Sparganium*, was regarded as primitive and a possible ancestor to the monocots. Hutchinson considered this group to be advanced and further divided it into Typhaceae and Pandanaceae on the basis of habitat. Chromosome studies have indicated that it is possible for the monocots to have been derived from the progenitors of Alismatales, thus supporting Hutchinson. However the Helobiales, as a whole, is a heterogenous group, as seen from karyotype analysis, and only *Alisma* has the so-called primitive relatively symmetrical karyotype. The Typhales, on the other hand, are definitely more advanced, as shown by the presence of very small acrocentric chromosomes in the karyotype with high number ($n=15$) in all the three genera. However, the karyotypes of these genera are so similar that there is no justification for separating them on the basis of their different habitat. These two examples show the weakness of relying on habitat as the principal criterion in classification.

(ii) The group Agavales was created by Hutchinson, taking genera from Amaryllidaceae and Liliaceae, on the basis of habit and morphological characters like long-stalked inflorescence, rhizomatous stock and arborescent nature. The karyotype of Agavales, as shown by *Agave*, is very distinctive - bimodal, with $x=30$, of which 5 chromosomes are long and 25 short. A comparison with the karyotypes of the other members of Amaryllidaceae and Liliaceae shows that this group is homogenous. It has been suggested that some genera, like *Funkia*, which show *Agave* type karyotype should be transferred to Agavales.

(iii) At the level of family studies, karyotype analysis in the Commelinaceae has shown the role of

chromosomal alterations in the evolution of two separate lines, one with $x=6$ and long chromosomes and the other with higher numbers and shorter chromosomes, within the family. *Callisia fragrans*, having both long and short chromosomes, may be a possible intermediate type.

After the initial enthusiasm, some limitations were observed in considering karyotype studies as confirmatory parameters in classification :

- (i) Chromosomal polymorphisms were observed where different chromosome numbers were recorded between two populations of the same species; between two individuals of the same population and even between two cells of the same individual, a phenomenon known as polysomaty.
- (ii) The trends from symmetry to asymmetry were reversed in some cases - fission of chromosome arms was followed by fusion, so that metacentric chromosomes were derived from telocentric ones; high base numbers were reduced by chromosomal alteration, following two successive reciprocal translocations and subsequent loss of short chromosome, as in *Crepis*.
- (iii) Polysomaty was maintained in vegetatively propagated plants, occasionally leading to the formation of new forms by entering into the growing apex of a daughter shoot. Examples of chromosomal polymorphisms have been recorded, among others, between (i) populations of *Chlorophytum variegatum* with $2n=28$ and 42 chromosomes; (ii) individuals of *Helichrysum bracteatum*, where two individuals, both with $2n=24$ chromosomes, have karyotypes with structural differences and (iii) in the same root-tip of *Zephyranthes mesochloa* where two cells have different chromosome numbers.

Certain other phenomena were also recorded which affected the role of karyotype analysis in solving major problems of taxonomy. These included : (i) occurrence of the same chromosome number in different species of the same genus. For example, in *Pogonia*, the two distinct species *P. gamutiana* and *P. juliana* both have $2n=68$ chromosomes; (ii) The same number has also been

observed in different genera as well. For example, *Erythronium dans canis* has the same number of $2n=24$ chromosomes as three species of *Lilium*, *L. henryi*, *L. giganteum* and *L. japonicum*. In such cases, the karyotypes of the different taxa differ only in structural details of the chromosomes. Species of *Arisaema*, collected from different altitudes mostly have the same chromosome number of $2n=28$ but differ from each other in the chromosome structure and the presence of B chromosomes.

In such cases, the investigations may be extended to more intensive studies to identify genetical parameters at the molecular level. The various criteria used for the detection of genetic diversity include the inheritance of clusters of characteristics involving one or more of the individual cellular components. Allozyme and protein patterns are used as well as the relationship between DNA sequences as determined through DNA/DNA hybridization and restriction fragment length polymorphisms. Protein sequencing and ultrastructure analysis may further be utilised for nucleosome and replication patterns. DNA content may be determined *in situ* or after extraction from the nucleus. The different parameters showing diversity are : (i) amount of total nuclear DNA; (ii) amount of the repeats and (iii) nature of the repeats. These parameters often uncover light cryptic differences: between taxa and can be used as additional evidences.

The amount of total nuclear DNA may, however vary between different species of the same genus, viz.

Picea albertina (85pg) and *P. sitchensis* (38 pg) while unrelated genera like *Larix decidua* and *Pinus sylvestris* may have the same amount (30 pg). Some monocots have relatively high amount of total nuclear DNA. In Liliaceae, the value ranges between 85 to 94 pg while in *Tradescantia*, it is 62 pg.

In taxa with similar nuclear DNA content, variations occur between species or strains in the relative quantities of repeats. The repeats may be separated into fast, moderate and minor fractions. The relative amounts differ at the interstrain level as seen in *Lathyrus* and *Phaseolus* but the total content remains the same. The very fast fraction is usually highly conserved. Such analysis of the genetic diversity at the molecular level forms a new line of approach to the problem of taxonomic relationships. It may be related to other parameters like growth. The amount of DNA is lower in the faster growing species of *Acacia*.

In spite of the rapid advancements, even now genetic studies can only assist in solving problems of taxonomic dispute within the taxonomic hierarchy. However, future refinements may unravel the patterns of evolution of the structure, behaviour and expression of genes. These improvements could lead to an understanding of the evolutionary and predictive aspects in systems of classification since ultimately any character, or clusters of characters, on which any classification is based, is the expression of gene action as related to environment.