RESEARCH PAPER

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Morphological and biochemical investigation of five *Solanum* species

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Solanaceae is one of the families with a number of important agricultural plants as well as many toxic plants. In the present investigation a comparative and comprehensive leaf, branch, fruit and seed have been analyzed and complemented with leaf proteins for identifications. As expected taxonomical characters within *Solanum* species revealed great morphological differences. Some *Solanum* species are phenotypically close to each other, but have different taxonomic status. These morphological similarities lead to difficulty in identification of these species. Hence, a study was undertaken to understand the similarities and dissimilarities at morphological and protein level. Molecular weight of the protein of the five species varied from 199.53kD to 1.78kD. The result showed *S. surattense* and *S. trilobatum* were closer morphologically whereas, *S. surattense* and *S. melongena* were closer at protein level.

Key words : Solanaceae, SDS-PAGE, Paired affinity, Similarity, Phylogeny

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INTRODUCTION

The Solanaceae is a large family comprised of over 3000 species. It is the third economically important plant family and ranks the first in terms of vegetable crops and number of important medicinal plants. Plant morphology deals with the external characteristics of plants. It deals with the structure, development and modification of the various plant parts like roots, stem, leaf, inflorescence, flower, fruit and seed. Morphology forms are the fundamental basis for the study of plant taxonomy. Taxonomic classification of plant species basically depends on the morphological and anatomical characters, these features are changeable and sometimes difficult to observe, so it is necessary to be supported by molecular techniques, in which molecular markers are used to detect the genetic variability. Molecular markers are biochemical constituents (*e.g.* secondary metabolites in plants) and macromolecules, *viz.,* proteins and deoxyribonucleic acids (DNA) that play a very important role in taxonomy, physiology, embryology, plant breeding, ecology, genetic engineering etc. SDS-PAGE (Sodium dodecyl sulphate) has been used successfully to resolve taxonomic and evolutionary problems of several plants (Khan, 1992; Rabbani *et al.,* 2001). In this technique protein is separated according their molecular weights. Resolution of this technique is very high and more than ten bands can be detected (Bartke *et al.,* 1966). Therefore, it could be a reliable tool for taxonomic purposes. *Solanum* species were selected to study the morphological and protein profile.

RESEARCH METHODOLOGY

The following *Solanum* species, *Solanum nigrum* L., *Solanum torvum* Sw., *Solanum trilobatum* L., *Solanum surattense* Burm. and *Solanum melongena* L. were collected from Tirunelveli region. The collected materials were stored in deep freezer (-70^{0}C) for SDS-PAGE analysis.

Morphological studies:

The taxonomic characters of the selected *Solanum* species were studied by using dissection microscope. Morphological characters were divided into two main categories.

– Vegetative characters
– Reproductive characters

Vegetative characters:

Habit	Herb, shrub or tree
Stem	Texture, presence of pubaceous.
Leaves	Sessile or petiole, glabrous, hairy, glaucous, shape of lamina, margin, apex, base, absence/presence of prickles, texture of lamina

Reproductive characters:

Inflorescence	Type of inflorescence, presence or absence of peduncle, presence or absence of prickles, presence or absence of pubescence.
Calyx	Colour, shape, number of sepals, presence or absence of prickles.
Corolla	Colour, shape, number of petals, presence or absence of prickles
Androecium	Number, filament shape, fixation of anther, length, presence / absence of prickles, anthers shape.
Gynoecium	Number, length of style, stigma shape, placentation, number of ovules.
Fruit and seed	Size of the fruit, colour of the fruit, texture, seed shape, color, texture.

Paired Affinity Index (Ellison *et al.,* 1962)

Paired Affinity Index (PA) was calculated by using the following formula:

$$\text{Paired affinity index (PA)} = \frac{\text{No. of common characters for species A and B}}{\text{Total no. of characters in A and B}} \times 100$$

SDS – PAGE analysis:

SDS –PAGE of leaf protein was carried out in vertical slab gel discontinuous buffer system following the method of Laemmli (1970) using 10% acrylamide gel concentration. A total volume of 12µl protein extract solution was loaded into each well and electrophoresis was carried out at 100V until the bromophenol blue dye reaches the bottom of the gel.

Data analysis:

Protein bands were scored for similarity index analysis based on the presence and absence of bands. The dendrogram was constructed based on this similarity index table using NTSYS- PC, version 2.01 (Rohlf, 1999). The molecular weights of the dissociated polypeptides were determined by using the standard curve.

RESULTS AND ANALYSIS

The findings of the present study as well as relevant discussion have been presented under following heads:

Morphological description of *Solanum* species:

According to the morphological characters of the selected species *S. nigrum* and *S. surattense* are closely related to each other based on the habit, habitat, root and medicinal values of berries. The shape and texture of leaf surface of *S. torvum* and *S. melongena* are closely related. The berries of these two species are used as vegetables (Table 1).

Paired affinity index:

Paired affinity (PA) indices can best be illustrated in the form of polygonal graphs (Hutchinson 1936). PA indices are expressed from 0 (at the centre of circle) to 100 (at the periphery of circle) along the appropriate radii, one value for each species will always equal 100, since one radius represents the species under consideration (Fig. 1).

The paired affinity index values of selected five *Solanum* species were below 50% (Table 2). The results indicate that all the species were distantly related to each other. Paired affinity index value was high (38.88%) between *S. surattense* and *S. trilobatum*. *S. torvum* and *S. melongena* showed 20% similarity in paired affinity index, it is very low compared to other three species of *Solanum*. Commonly all the species have below 40% similarities between them. Morphological similarities of characters contributed a lot toward the difficulty in identification of these species. There are very few morphological markers for distinguishing them. *Solanum americanum* can be distinguished from *S. nigrum* by its smaller seeds, umbellate inflorescence rather than the raciform as in *S. nigrum*, smaller anthers and shiny fruit (Edward 1999).

SDS-PAGE Analysis:

The protein profile (Fig.2) obtained by electrophoresis has been successfully used to resolve the taxonomic problems of *Solanum* species. Molecular weight of these five species varied from 199.53kD to 1.78kD. Based on the presence and absence of the bands, the similarity between the species was studied and the UPGMA dendrogram was constructed using NTSYS pc software. The similarity index table represents the similarity indices between the species (Table 3). In dendrogram five different species of *Solanum* species were separated into

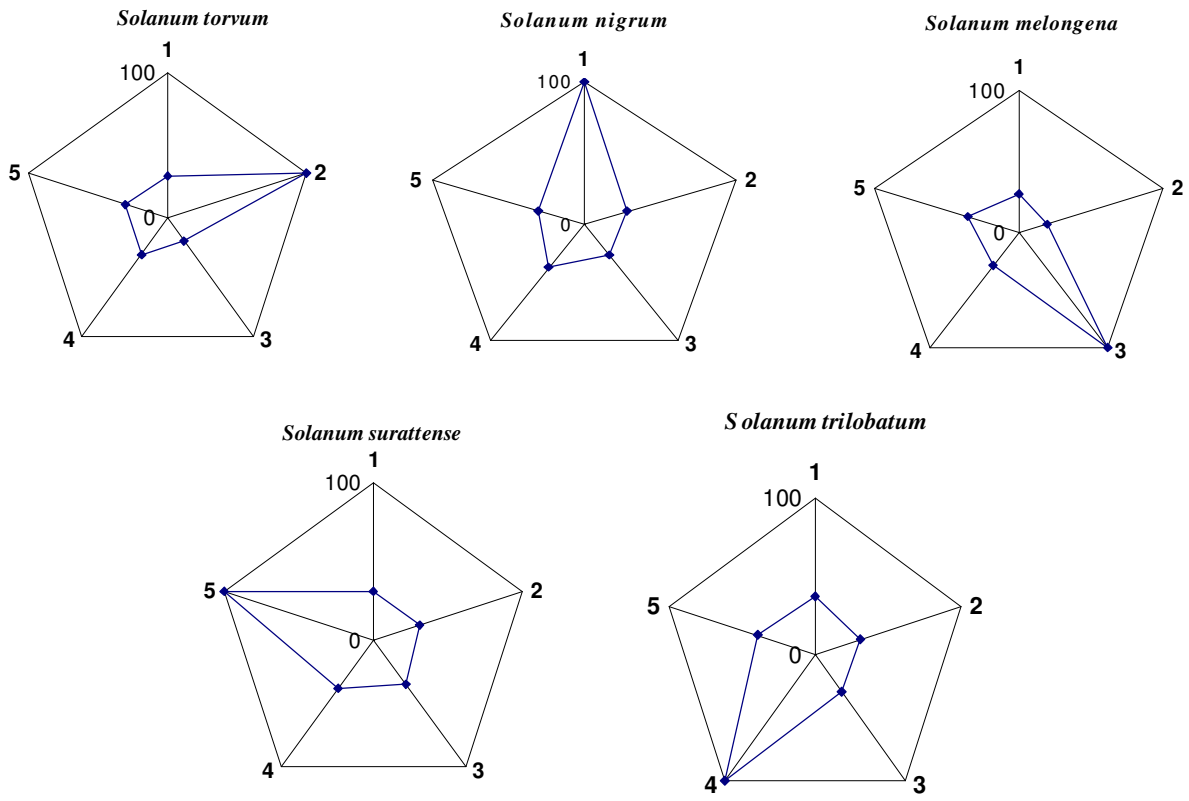

Fig. 1: Paired affinity index of five *Solannum* species

three main clusters (Fig. 3). A very close association was found between the species of *S. surattense* and *S.melongena* in the first cluster. *S.trilobatum* and *Solanum nigrum* showing more than 75% of similarity in the second cluster. Third cluster contains only one *Solanum* species *S.torvum* showing more than 50% similarities with each other. The status of critical taxa can be justified by using protein profile (Khan, 1992). *Solanum surattense* is another medicinally important species of the family Solanaceae; it has more resemblance with *Solanum nigrum*. Khan (1992) used SDS-PAGE to identify *Brachypodium* species, which overlap in most of their morphological characters, and to elucidate the relationship of critical taxa. Based on protein profile,

Table 2: Paired affinity index of *Solanum* species

Species	*S. nigrum*	*S. torvum*	*S. melongena*	*S. trilobatum*	*S. surattense*
S. nigrum	100.00	28.57	26.66	37.5	31.25
S. torvum		100.00	20.00	31.25	31.25
S. melongena			100.00	29.41	35.29
S. trilobatum				100.00	38.88
S. surattense					100.00

Table 3: Similarity index for *Solanum* species

Species	*S. surattense*	*S. melongena*	*S. trilobatum*	*S. nigrum*	*S. torvum*
S. surattense	1.00				
S. melongena	0.48	1.00			
S. trilobatum	0.64	0.56	1.00		
S. nigrum	0.78	0.54	0.78	1.00	
S. torvum	0.63	0.45	0.74	0.70	1.00

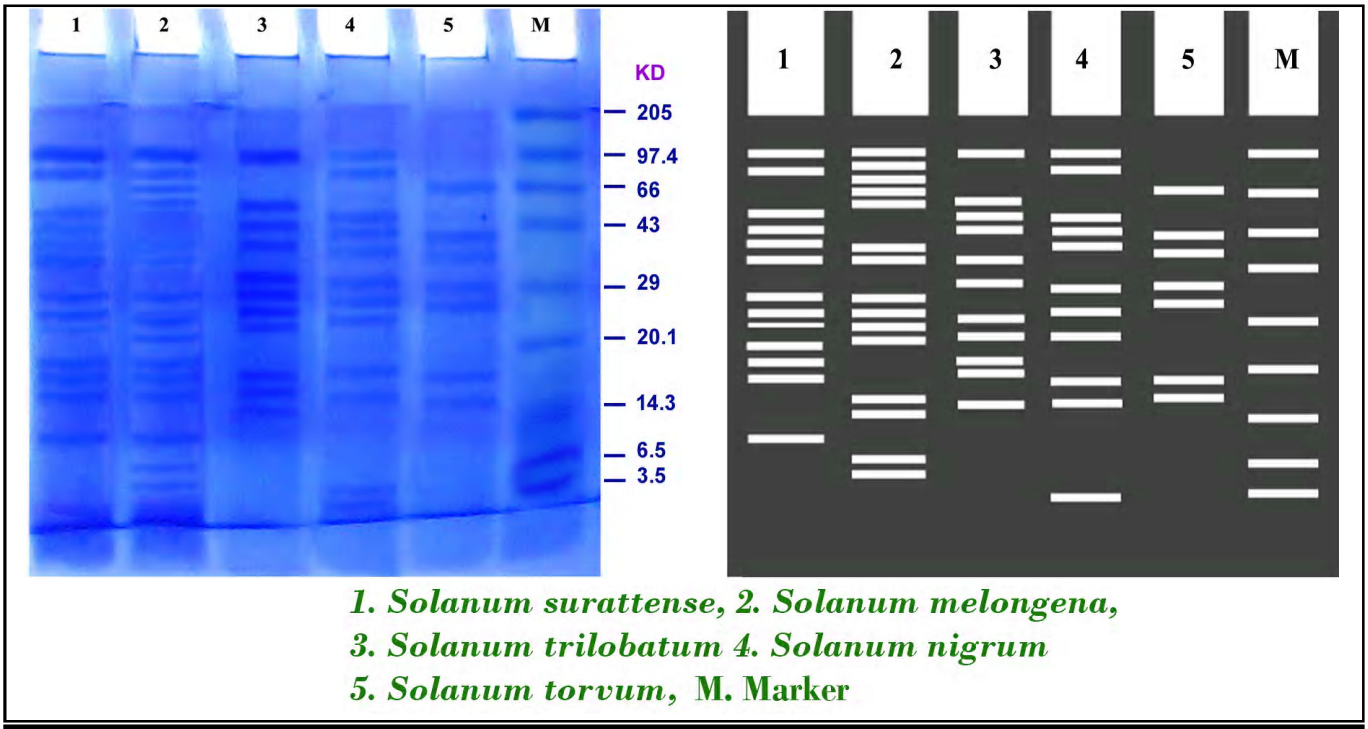

Fig. 2: Protein profiles of *Solanum* species

phylogenetic relationship among the various species of legumes was studied by Valizadeh (2001).

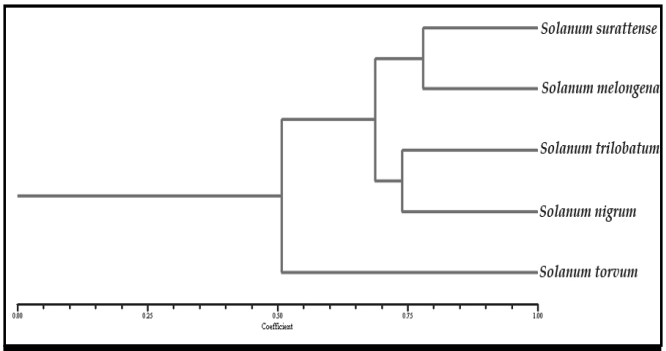

Fig. 3: UPGMA dendrogram of *Solanum* species by SDS-PAGE

Conclusion:

In conclusion, taxonomic classification of plant species basically depends on the morphological and anatomic characters, these features are changeable and sometimes difficult to observe, so it is necessary to be supported by molecular techniques. Studies on morphology and SDS-PAGE have provided an insight into the interrelationships among the five species of *Solanum* and have thereby helped to understand the relationship between these species.

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