



PHENETIC STUDY OF SOME INDIAN SOLANACEAE BASED ON FOLIAR MICROMORPHOLOGY, PALYNOLOGY AND REPRODUCTIVE POTENTIALITY

Mridushree Basak, Aparna Saha Sonar and Monoranjan Chowdhury*

Taxonomy of Angiosperms & Biosystematics Laboratory, Department of Botany,
University of North Bengal, Siliguri (West Bengal), India.

Abstract

Solanaceae Juss. comprises of about 98 genera and more or less 2,700 species around the world. Many of the plant products of this family are consumed and hence economically important. In the present study, a phenogram was prepared to understand the interrelationships and phylogeny of 14 plant species based on phenetic characters from foliar micromorphology, palynology and reproductive potentiality.

Key words : Solanaceae, foliar micromorphology, seed, pollen, phenetics.

Introduction

Taxonomy is a craft of delimiting ordered unit, referred as taxon and is constructed fundamentally in light of assessment and elucidation of perceptions. For distinguishing proof and remaking of connection between plants, components of regenerative organs are utilized from the antiquated ages. Despite the fact that reproductive characters have set up extremely valuable tools for identification and translating their connections at the same time, some of the time these organs are not accessible for study. Since some tropical plants bloom rarely and sporadically (Watson and Dallwitz, 1992), now a days, we generally uses various sterile characters from lamina for identification of plants (Hickey, 1973).

Solanaceae comprises of around 98 genera and pretty much 2,700 species around the world (Olmstead *et al.*, 2007). About 15 genera and 90 species are reported from India (Hickey *et al.*, 1988), whereas, in West Bengal, eight genera and 30 species were reported (Prain, 1903).

Gupta (1961) examined the absolute vein-islet numbers of some Indian Solanaceous plants. Leaf architecture and venation design assumes essential part in distinguishing the species. Adjacent to venation, stomata and epidermal cells and indumentums likewise give systematically essential analytical characters. The

micromorphological characters of foliar indumentums assume a critical part in plant identification, particularly at species levels and below ranks (Hardin, 1979).

The term 'pollen' was first presented by Swedish botanist Linnaeus. The pollen characters are quite unique and the exine characters are hereditarily settled and stay unaltered with any natural changes. These characters are taxon-particular and might be change in species to species and in this manner have been perceived as a solid apparatus for handling taxonomic problems (Erdtman, 1952). Kumar *et al.* (2015) explored the pollen morphological characters of some species of *Solanum* growing in various localities of Southern parts of India.

Investigation of regenerative potentiality of plant species is quite vital because, it helps to understand the rate of successful establishment of specific plant species in respective agro-climatic environment. The regenerative limit, which is positively hereditarily controlled is exceptionally species particular and is of extensive intrigue. At this same time, seed morphology, weight and number add to the foundation of species for the era to come (Salisbury, 1942).

The family Solanaceae is regularly known as Potato Family. The name Solanaceae was embraced in light from the type genus *Solanum*, "The Nightshade Plant". The name may originate from the latin verb Solari, signifying

*Author for correspondence : E-mail: mono_malda@yahoo.co.in

“to alleviate”, probably referring to the calming pharmacological properties of some of the Psychoactive genera of the family.

Materials and Methods

Study area

Specimens were gathered from various zones of West Bengal including the Himalayas in the north, huge areas of plains and the Bay of Bengal in the south with an areas of 88,752 km² (Anonymous, 2012). The atmosphere changes from tropical savanna in the southern parts to subtropical humid in the north. The fundamental seasons are summer, the rainy season, a short fall and winter. While the late spring in the delta district is noted for extreme stickiness, the western highlands encounter a

dry summer, with the most noteworthy daytime temperature running from 38°C to 45°C (Anonymous, 2006). A cold and dry northern breeze blows in the winter, generously bringing down the moistness level. The Darjeeling Himalayan Hill district encounters a harsh winter, with periodic snowfall at high altitudinal ranges of Tiger hill and Sandakfu (Anonymous, 2017).

Materials

The present work has been carried out with the fresh specimens of *Solanum americanum* Mill., *Solanum villosum* Mill., *Solanum torvum* Sw., *Solanum viarum* Dunal, *Solanum sisymbriifolium* Lam., *Datura innoxia* Mill., *Datura metel* L., *Datura stramonium* L., *Brugmansia suaveolens* (Humb. & Bonpl. ex Willd.) Bercht. & J.Presl., *Nicotiana tabacum* L., *Nicotiana*

Table 1 : References to the collected and used taxa along with their known ranges of distribution.

S. no.	Taxa	Distribution in West Bengal	Indian Distribution	Global distribution
01.	<i>Solanum americanum</i>	Darjeeling, Jalpaiguri, Malda	Assam, Nepal, Bihar, Bhutan	South Africa, Greece, Turkey, Australia, Indonesia
02.	<i>Solanum villosum</i>	Darjeeling, jalpaiguri, Malda	Assam, Meghalaya, Bihar	South Africa, Greece, Turkey, Australia, Indonesia
03.	<i>Solanum torvum</i>	Darjeeling, Bakura, Howrah, Malda, Jalpaiguri	North-East, North West & South India	North & Central America, Eastern Brazil, Africa, Madagascar
04.	<i>Solanum viarum</i>	Throughout West Bengal	Assam, Himachal Pradesh, Nepal	Southern Japan, Mexico, Australia, North & Central America
05.	<i>Solanum sisymbriifolium</i>	Throughout West Bengal	North-East (Assam) to Uttarakhand (Garhwal)	West Indies, Eastern Brazil, Indonesia, Madagascar
06.	<i>Datura innoxia</i>	Darjeeling, Jalpaiguri, Malda	Throughout India	Australia, North & Central America
07.	<i>Datura metel</i>	Darjeeling, Jalpaiguri, Malda	Bhutan, Nepal, North West & South India	Africa, Malaysia, Tropical Asia
08.	<i>Datura stramonium</i>	Throughout West Bengal	Throughout India	North & Central America, Turkey, Eastern Brazil
09.	<i>Brugmansia suaveolens</i>	Darjeeling, Jalpaiguri	Throughout India	Australia, Mexico, Greece, Malaysia
10.	<i>Nicotiana tabacum</i>	Throughout West Bengal	North-East to Uttarakhand	West Indies, Japan, Bangladesh
11.	<i>Nicotiana plumbaginifolia</i>	Darjeeling, Jalpaiguri, Burdwan	Assam, Himachal Pradesh, Bhutan	Tropical Asia, Malaysia, Africa
12.	<i>Solanum pimpinellifolium</i>	Darjeeling, Jalpaiguri, Malda	Throughout India	Tropical Asia, North & Central America
13.	<i>Withania somnifera</i>	Darjeeling, Jalpaiguri	North-East region, Nepal, Bhutan	Australia, Japan, North & Central America
14.	<i>Physalis minima</i>	Darjeeling, Jalpaiguri, Malda	North-East (Assam) to Uttarakhand (Garhwal)	Tropical Asia, Africa, North & Central America

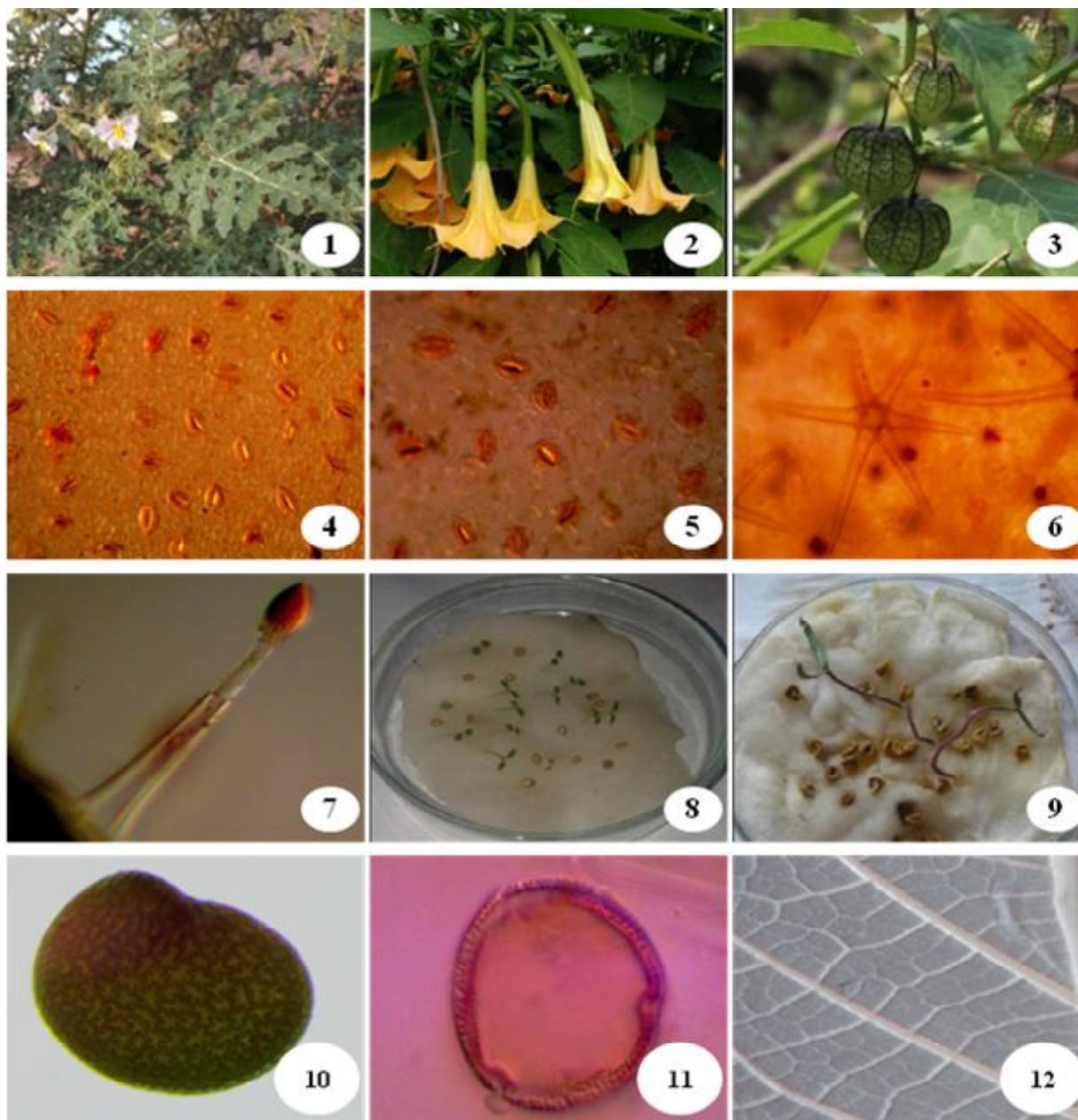


Plate 1 : *Solanum sisymbriifolium*, **2.** *Brugmansia suaveolens*, **3.** Fruits of *Physalis minima*, **4.** Stomata of *Solanum villosum*, **5.** Stomata of *Nicotiana tabacum*, **6.** Stellate hair of *Solanum torvum*, **7.** Glandular hair of *Solanum sisymbriifolium*, **8.** Seed germination of *Solanum viarum*, **9.** Seed germination of *Datura metel*, **10.** Seed coat ornamentation of *Withania somnifera*, **11.** Pollen of *Datura innoxia*, **12.** Venation pattern of *Datura stramonium*.

plumbaginifolia Viv., *Solanum pimpinellifolium* L., *Withania somnifera* (L.) Dunal and *Physalis minima* L.

Methods

Wide ranges of array of methodology were followed to accumulate various taxonomic data from the leaves, pollens and seeds for all species. For vein study some mature leaves were drenched in 4-5% solution of NaOH until cleared (Mishra *et al.*, 2011). A large portion of the samples were exceptionally delicate, thus needed 15-20

days to be fully transparent. The highest orders of veins were recognized upto 5⁰. The veins and indumentums were seen under 5X and 10X objective of a compound magnifying microscope (OLYMPUS – OIC 81789) utilizing a 10X eye piece.

Stomatal investigation was executed through peeling of lamina principally from abaxial surface (as stomatal thickness is substantially higher in down surface) with the assistance of forceps and mounted in glycerine for perception. At last photos were taken utilizing high power

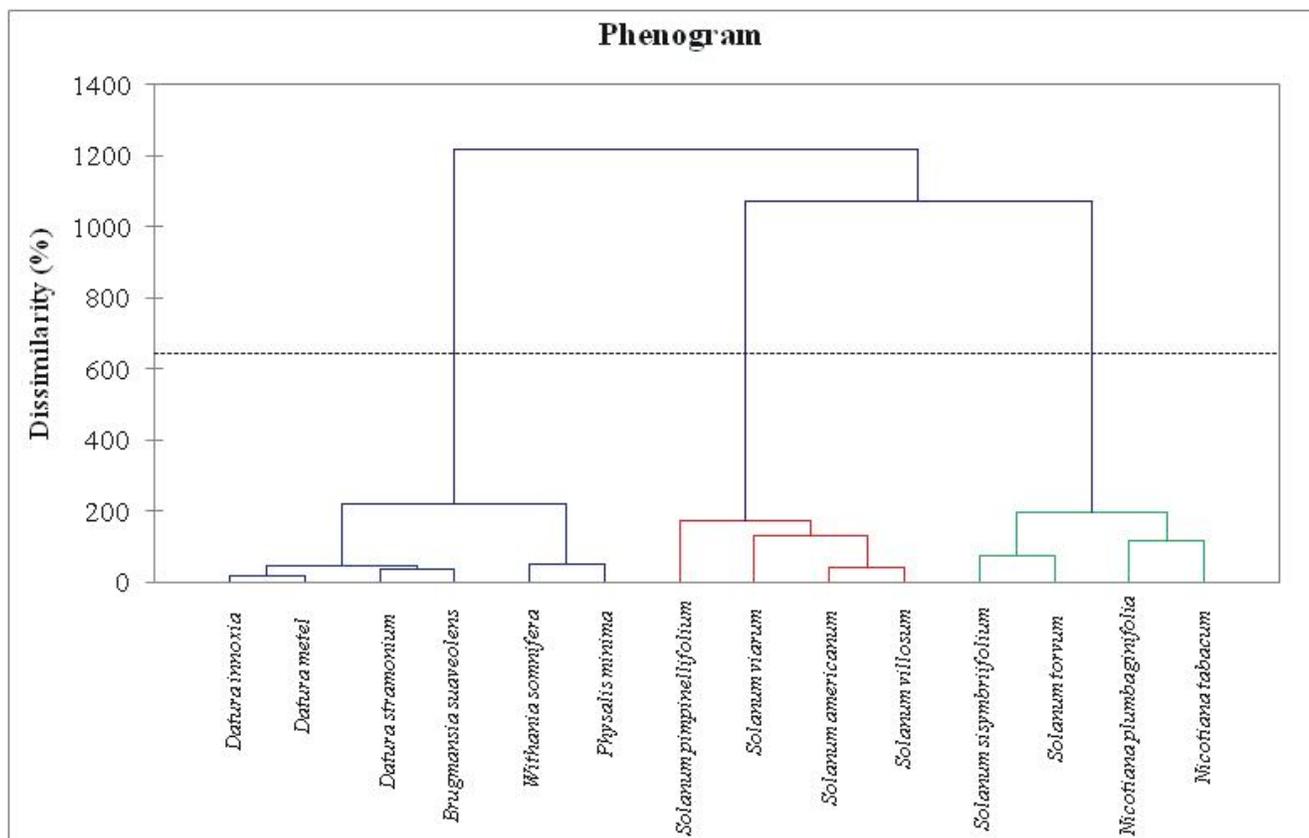


Fig. 1 : Phenogram showing the interrelationships of studied specimens of Solanaceae.

objective and stomatal frequency and stomatal index of considered specimens were determined as the technique suggested by Salisbury (1927).

Pollens were investigated under light microscope after preparing the permanent slides following acetolysis method as proposed by Erdtman (1952, 1969) after a few alterations recommended by Nair (1970) and Chanda (1966).

Following the technique for Hill *et al.* (1988), the regenerative capability of the considered plants was evaluated. Seeds were gathered and counted properly to calculate the total average seed output by individual plants. 50 seeds of one species with three replicas were germinated in Petri dishes on wet whatman blotting paper and regular monitoring and watering were done. The numbers of germinated seeds was recorded after every 20 days interval. Seed size index, seed shape index, viability percentage, non-viability percentage and germination percentage were calculated following method of Hill *et al.* (1988). Reproductive capacity and seed output was determined by standard method (Salisbury, 1942). Finally, a phenogram was prepared using XLSTAT software considering all the accumulating data from the studied sources.

Results and Discussion

Micro and macro-morphological examination of 14 species have demonstrated different imperative characters. Lamina size ratio is recorded highest in *Solanum sisymbriifolium* and least in *Solanum pimpinellifolium* and *Withania somnifera*. Except *Solanum torvum* and *Solanum viarum*, all the species show marginal petiolar attachment and decurrent leaf base. They also show enormous type of variation in their venation pattern such as *Nicotiana tabacum* and *Nicotiana plumbaginifolia* show Festooned brochodromous type of 2^0 vein category, whereas others possess semicraspedodromous type. Principally 4 sorts of stomatas (*viz.* anisocytic, actinocytic, anomocytic, paracytic) were recorded during the investigation. The range of stomata frequency is 22.22-104 per mm^2 and the range of stomatal index is 34.89-47.48%. *Solanum viarum* shows highest stomatal frequency (104/ mm^2) and *Solanum pimpinellifolium* shows lowest stomatal frequency, whereas highest stomatal index found in *Brugmansia suaveolens* (47.48%) and lowest in *Solanum torvum* (34.89%). In the present perceptions, these contemplated species have multicellular, uniseriate, branched, glandular and non-glandular kind of

Table 2 : Characterization of Pollen grains of studied species.

Taxa	Shape	Size(μm)	Aperture	Unit	Exine
<i>Solanum americanum</i>	Prolate-spheroidal	26-27	Tricolporate	Monad	Psilate
<i>Solanum villosum</i>	Prolate-spheroidal	14-18	Tricolporate	Monad	Psilate
<i>Solanum torvum</i>	Prolate-spheroidal	20-22	Trizonocolporate	Monad	Psilate
<i>Solanum viarum</i>	Prolate-spheroidal	88-99	Trizonocolporate	Monad	Psilate
<i>Solanum sisymbriifolium</i>	Oblate-spheroidal	75-79	Trizonocolporate	Monad	Reticulate
<i>Datura innoxia</i>	Oblate-spheroidal	88-93	Tricolporate	Monad	Striate with distinct proliferaton
<i>Datura metel</i>	Oblate-spheroidal	69-86	Tricolporate	Monad	Striate with distinct proliferaton
<i>Datura stramonium</i>	Oblate-spheroidal	77-90	Tricolporate	Monad	Striate with distinct proliferaton
<i>Brugmansia suaveolens</i>	Prolate-spheroidal	80-95	Tricolporate	Monad	Reticulate
<i>Nicotiana tabacum</i>	Prolate-spheroidal	24-25	Tricolporate	Monad	Reticulate-foveolate
<i>Nicotiana plumbaginifolia</i>	Prolate-spheroidal	17-20	Tricolporate	Monad	Reticulate-foveolate
<i>Solanum pimpinellifolium</i>	Oblate-spheroidal	73-77	Tetrazonocolporate	Monad	Granulate
<i>Physalis minima</i>	Prolate-spheroidal	25-27	Tricolporate	Monad	Psilate

indumentums. Stellate kind of indumentums was seen in just *Solanum torvum* and it likewise has most noteworthy length of indumentums (44.66 μm).

From seed study, it has been presumed that, seed output is a highly variable character. Maximum seed output was found in *Solanum americanum* (Ave. 4884.80) and minimum in *Solanum pimpinellifolium* (640). From the study of germination test, it has been recorded that *Solanum americanum* shows highest germination capacity and *Datura metel* has the lowest value.

From previously available literary works, the local, Indian and World distributions of the studied specimens were tabulated (table 1).

The pollen morphology of the family is quite distinctive (Erdtman, 1952). Pollen grains are usually radially symmetrical, isopolar, prolate-spheroidal to sub-prolate or prolate, rarely oblate-spheroidal. The pollen morphological study is shown in table 2.

The result of the survey and the data produced through the application of different formulae related to seed size, seeds shape, viable percentage, germination percentage, reproductive capacity and seed output of the studied specimens except *Datura stramonium* and *Brugmansia suaveolens* were presented in table 3.

A phenogram was prepared using XLSTAT software based on the overall various data from the characters such as Stomatal type, Mean no. of stomata (S), Mean

no. of epidermal cells (E), Stomatal frequency (No. of stomata/ mm^2), Stomatal index ($S \times 100 / E+S$) (%), Mean length and breadth of stomata (μm), Mean length and breadth of stomata+guard cell (μm), Mean length and breadth of guard cell (μm), Mean length and breadth of stomata+subsidiary cell (μm), Mean length and breadth of subsidiary cell (μm), 2^o, 3^o, 4^o and 5^o vein category, Origin of indumentums, Presence or absence of glandular indumentums, Length and breadth of indumentum (μm). The obtained phenogram (fig. 1) is quite significant where, the X-axis shows the plants species and Y-axis shows the percentage of dissimilarity among the species. The phenogram shows the six species of *Solanum* and two species of *Nicotiana* are close relatives. *Withania* is very closely related to *Physalis*, whereas, *Brugmansia* and *Datura* are close relatives.

Conclusion

Most of the species show aniso- or actinocytic stomata while, only *Solanum sisymbriifolium* possess anomocytic type. Stomatal frequency and index were calculated from the apical, middle and basal portions of leaves in each case, which showed a large variation. 2^o vein arrangements in all the cases are semicraspedodromous, except species of *Nicotiana*, which contain festooned brochidodromous arrangement. There are variations in 3^o, 4^o and 5^o vein categories also. The indumentums are in some cases, short, stout and glandular, while in other cases, elongated and

Table 3 : Seed morphological and reproductive capability study of the specimens.

Taxa	Seed length (µm)	Seed width (µm)	Size index (µm)	Shape index (µm)	Weight/ 100 seeds (gm)	Fruit/ Plant	Seeds/ Plant	Seed output	Germi- nation %	Viable %	Non- viable %	Reprod- uctive capacity
<i>Solanum americanum</i>	4.18	2.01	8.40	2.07	0.28	56.80	86	4884.80	28	28	72	1,367.74
<i>Solanum villosum</i>	4.24	2.88	12.21	1.47	0.24	34.40	37.33	1284.15	16	16	84	205.46
<i>Solanum torvum</i>	5.70	3.29	18.75	1.73	1.67	65	88	5720	20	20	80	1144
<i>Solanum viarum</i>	5.13	3.82	19.59	1.34	0.54	7	113	791	44	44	56	348.04
<i>Solanum sisymbriifolium</i>	6.22	5.20	31.87	1.17	0.42	29.6	37.33	1104.96	23	23	77	254.14
<i>Daturainnoxia</i>	9.85	8.13	80.08	1.21	0.96	4	160	640	36	36	64	409.6
<i>Daturametel</i>	8.77	6.73	55.86	1.37	0.86	8.66	124	1073.84	8	8	92	85.90
<i>Nicotianatabacum</i>	6.21	4.30	26.70	1.44	0.52	25	91	2275	92	92	8	2093
<i>Nicotiana plumbaginifolia</i>	1.02	0.98	0.99	1.04	0.10	13.4	52	696.80	65	65	35	45,292
<i>Solanumpimpinellifolium</i>	5.80	3.58	20.76	1.62	0.36	50	67	3350	8	8	92	268
<i>Withaniasomnifera</i>	5.26	3.24	17.04	1.62	0.12	35.2	27	950.40	8	8	92	76.03
<i>Physalismsinima</i>	3.13	2.42	7.57	1.291	0.14	24.2	85.66	2072.97	38	38	62	787.72

nonglandular. The pollen aperture type, though mostly tricolporate and trizonocolporate, *Solanum pimpinellifolium* has tetrazonocolporate aperture. There is significant difference in the seed germination percentage too.

Comparative phenetic study on 14 species of Solanaceae reveals significant interrelationship that is clearly reflected in the phenogram. The species of *Solanum* are closely located along with *Nicotiana*. The species of *Datura*, *Brugmansia* and *Withania* and *Physalis* located on separate sister clade.

References

- Anonymous (2012). "Area, population, decennial growth rate and density for 2001 and 2011 at a glance for West Bengal and the districts: provisional population totals paper 1 of 2011: West Bengal". Registrar General & Census Commissioner, India.
- Anonymous (2006). "Climate". West Bengal: Land. Suni System (P) Ltd. Anonymous. "Under 7-inch snow, Sandakphu a hot favourite among tourists now". *Times of India*. 12 March 2017. Retrieved 2 July 2017.
- Chanda, S. (1966). On the pollen morphology of Centrolepidaceae, Restionaceae and Flagellariaceae with special reference to taxonomy. *Grana Palynol.*, **6(3)** : 365-415.
- Erdtman, G. (1952). *Pollen morphology and plant taxonomy of angiosperms*. Stockholm and Waltham, Mass: USA.
- Erdtman, G. (1969). *Hand book of palynology*. Munksgaard, Copenhagen.
- Gupta, R. (1961). Correlation of tissues in leaves. I. Absolute veinlet numbers and absolute veinlet termination numbers. *Ann. Bot.*, **25** : 65-70.
- Hardin, J. W. (1979). Patterns of variation in foliar trichomes of eastern north American *Quercus*. *American J. Bot.*, **6**: 576-585.
- Hickey, M. and C. King (1988). *Hundred families of flowering plants*. Cambridge University Press, Cambridge.
- Hickey, L. J. (1973). Classification of the architecture of dicotyledonous leaves. *Amer. J. Bot.*, **60**: 17-33.
- Hill, H. J., A. G. Taylor and X. L. Huang (1988). Seed Viability Determinations in Cabbage Utilizing Sinapine Leakage and Electrical Conductivity Measurements. *J. Exp. Bot.*, **39(207)** : 1439-1447.
- Kumar, A. V. S., M. C. Nair and K. Murugan (2015). Pollen morphology of selected taxa of the genus *Solanum* from Southern Western Ghats, Kerala, India. *Rheedea*, **25(2)**: 128-145.
- Mishra, M. K., P. Dandamudi, S. P. Nayani, S. S. Munikoti, S. S. Chelukunda and Jayarama (2011). Variability in stomatal features and leaf venation pattern in Indian coffee (*Coffea arabica* L.) cultivars and their functional significance. *Bot.*

- Sarb.*, **35(2)** : 111-119.
- Nair, P. K. K. (1970). Pollen morphology of angiosperms: A historical and phylogenetic study. *Vikas publishing house*, Lucknow.
- Olmstead, R. G. and L. Bohs (2007). A Summary of molecular systematic research in Solanaceae : 1982-2006. *Acta Hort.*, **745** : 255-68.
- Prain, D. (1903). *Bengal plants*. Volume II. Bishen Singh Mahendra Pal Singh, Dehradun.
- Salisbury, E. J. (1927). On the cause and ecological significance of stomatal frequency with special reference to the woodland flora. *Philosoph. Trans. Royal Soc. B.*, **216** : 848-852.
- Salisbury, E. J. (1942). *The reproductive capacity of Plants*. G. Bell and Sons.
- Sharma, E. and K. Tsering (2009). *Climate Change in the Himalayas: the vulnerability of biodiversity*. Biodiversity and climate change in the Himalayas; Sustainable Mountain Development No.55. Spring 2009, International Centre for Integrated Mountain Development (ICIMOD), Nepal.
- Watson, L. and M. J. Dallwitz (1992). *The grass genera of the world*. CAB International, Cambridge.