Taxonomic Evidences:

The term taxonomy was first introduced to the plant science in 1813 by A. P. de Candolle, which meant the theory of plant classification. But later this term became more inclusive and at present it includes identification of plants, their nomenclature and classification.

Traditionally taxonomy was based largely on gross morphological features of a plant. However now virtually all other scientific information is used from branches like anatomy, genetics, cytology, chemistry, reproductive biology, ecology, physiology, molecular biology, etc.

Hence taxonomy is a very dynamic and synthetic science and is the basic to all other sciences. At the same time it is dependent upon them, i.e. the information used by taxonomists is gathered using information from other disciplines such as genetics, ecology, morphology, anatomy, physiology, etc. (Fig. 1.1)

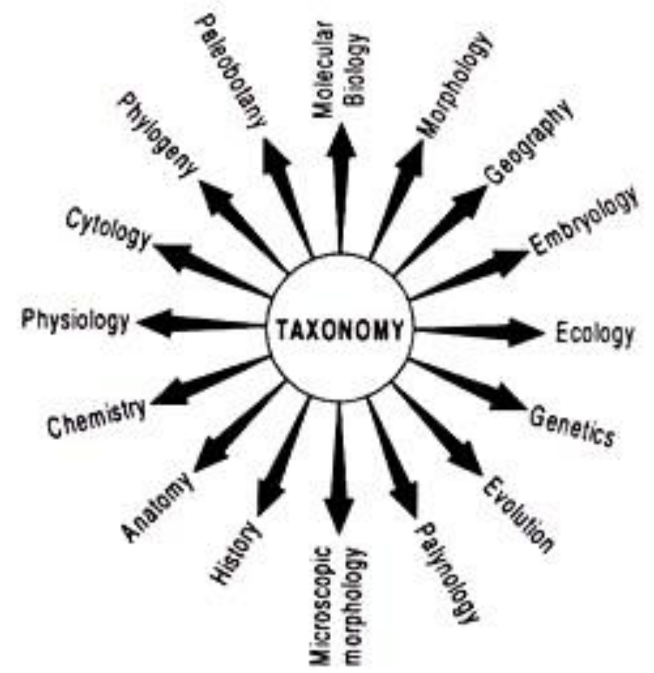

Fig. 1.1 : Taxogram showing relationships of various disciplines with plant taxonomy

1. Palynology in Relation to Taxonomy:

Palynology is the science, which deals with Pollen grains. Pollen grains are often easily disseminated by wind etc., Pollen grains are found everywhere . Some families, such as Asteraceae, show different types of pollen grains (**eurypalynous**), whereas several others have a single morphological pollen type (**stenopalynous**). Such stenopalynous groups are of considerable significance in systematic palynology. Pollen grains present a number of features of taxonomist interest. Fossil spores are found in peat and other sediments, in lignite, coal and shales. They are evident since Pre-Cambrian times hundreds of millions of years ago.

Pollen grains morphology plays an important role in classification. Pollen grains may be vesiculate (with air sacs); saccate or non-saccate, fenestrate or non-fenestrate, colpate (furrows or colpi present) or porate (apertures present at the poles). According to position of apertures six subdivisions are made e.g., ceta (down, inwards in a tetrad), ann (up; outwards in a tetrad), zone is the zonal position i.e., at the equator, and panto is uniform distribution all over the spore surface. The number of nuclei present at the time of shedding is also significant. Most primitive angiosperms are shed at 2-nucleate stage, whereas in more advanced groups pollen is shed at 3-nucleate stage. Angiosperms mostly have pollen grains of radial symmetry, bilateral symmetry being found in several gymnosperms. Most pollen grains are globose in shape, although boat- shaped, ellipsoidal and fusiforms are also met in different angiosperms.

Basic evidentiary characters:

(i) Pollen unit type, (ii) Pollen grain polarity, (iii) Pollen grain shape, (iv) Pollen grain symmetry, (v) Pollen grains nuclear state, (vi) Pollen wall architecture, (vii) Exine stratification, (viii) Exine structure, (ix) Exine sculpture, (x) Aperture type, (xi) Aperture number, (xii) Aperture position, (xiii) Aperture shape, and (xiv) Aperture structure.

Pollen aggregation

Microsporogenesis yields four microspores which mature into pollen grains. In large majority of angiosperms the pollen grains separate prior to release. Such single pollen grains are known as monads. In rare cases pollen grains are released fused in pairs, when they are known as dyads. In many angiosperms the four microspores do not separate and the pollen grains form a tetrad. Five different types of tetrads are differentiated:

1. **Tetrahedral tetrad**-four pollen grains form a tetrahedron: four grains compacted in a sphere. Such pollen grains are found in family **Ericaceae.**

2. **Linear tetrad**- four pollen grains arranged in a straight line as in genus ***Typha*.**

3. **Rhomboidal tetrad**- four pollen grains in one plane, with two separated from one another by close contact of the other two.

4. **Tetragonal tetrad-** four grains are in one plane and equally spaced as in ***Philydrum***.

5. **Decussate tetrad-** four grains in two pairs, arranged at right angles to one another, as in genus ***Lachnanthes***.

6. **Polyad:** A polyad generally consists of eight pollen grains. such as *Calliandra* of Mimosoideae, the pollen grains are connate in a group of more than four.

Pollen wall

The pollen grain wall is made of two principal layers, outer **exine** and inner **intine**. The exine is hard and impregnated with **sporopollenin**, a substance that makes it resistant to decay, and enables preservation in fossil record. Exine is further differentiated into two layers: outer **ektexine** and inner **endexine**. The ektexine is further distinguished into basal **foot layer,** radially elongate **columella** and roof like **tectum.** In some taxa the columella may be replaced by granular middle layer. Similarly in some primitive angiosperms tectum is lacking (**atectate** pollen grain), and the exine appears granular. Above layers of exine are clearly visible under an electron microscope, but when observed under a light microscope, the inner layer known as **nexine**, includes endexine plus foot layer of ektexine. The upper layers consisting of columella, tectum and the supratectal sculpturing constitute **sexine**.

Pollen wall sculpturing

Present on the outer surface of tectum are often certain supratectal projections, which provide a variety of sculpturing to exine wall. In some cases lacking tectum, the sculpturing is formed by columellae. The common types of sculpturing include: **baculate** (rod- shaped elements, each known as **baculum**), **clavate** (club-shaped elements), **echinate** (spine-like elements longer than 1 micron), **spinulose** (spine-like elements shorter than 1 micron; **scabrate**), **foveolate** (pitted sur- face with pores), **reticulate** (forming net- work, each element known as murus and space in between as lumen), **fossulate** (lon- gitudinal grooves), **verrucate** (short wart-like elements), **gemmate** (globose or ellipsoid elements), **psilate** (smooth surface), and **striate** (having thin striations on surface).

Pollen aperture

Pollen aperture is a specialized region of pollen wall through which the pollen tube comes out. The exine may be **inaperturate** (with- out an aperture) or aperturate. An aperturate pollen may have a single pore (**monoporate**), a single slit running at right angles to the equator (**monocolpate**), three slits (**tricolpate**), three pores (**triporate**) three slits each with a geminate pore in middle (**tricolporate**), with many pores (**multiporate**) accompanied by a variety of surface ornamentations. Pollen with one or more slits located at the polar end is accordingly termed, **monosulcate**, **disulcate** and **trisulcate**, depending on the number of slits. Pollen grain with slits joined at poles is termed **syncolpate**. Aperture having three branches is termed **tricho- tomosulcate**.

Monocolpate condition is widely spread in primitive dicots and a majority of monocots. The pollen of anemophilous plants is usually small, rounded, smooth, rather thin- walled and dry with shallow furrows. Anemo- philous pollen is found in *Populus*, Poaceae, Cyperaceae, Betulaceae and several other families. Insect- and bird-pollinated pollen, on the other hand, is large, sculptured and often coated with adhesive waxy or oily substance. The pollen of Asteraceae is generally highly elaborate but simplification towards loss of sculpturing has occurred in several genera with wind pollination. The vestigial scattered patches of adhesive layer on wind pollinated pollen have been considered as evidence of the derivation of anemophily from entomophily.

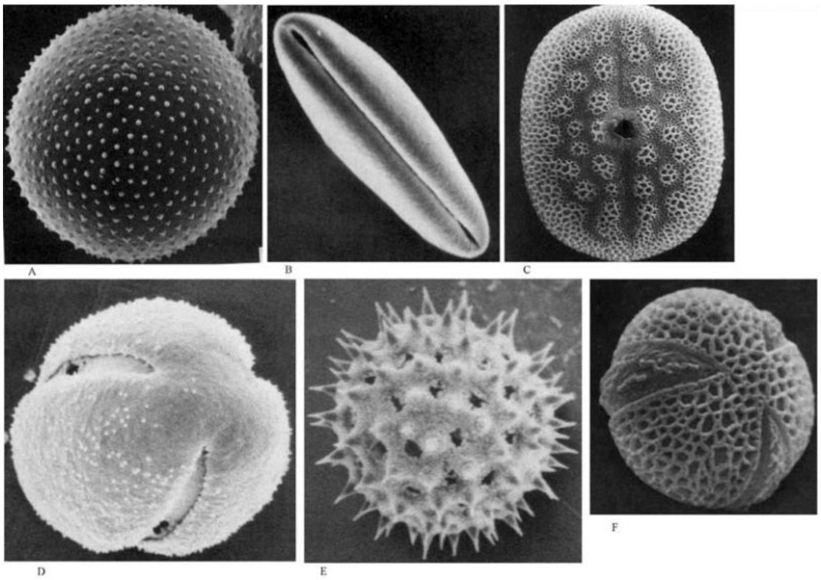

Fig. SEM of pollen grains. **A:** Nonaperturate pollen grain of *Persea americana*; **B:** Monosulcate pollen grain of *Magnolia grandiflora*; **C:** Monoporate pollen grain of *Siphonoglossa*; **D:** Tricolporate pollen grain of *Scaevola glabra*; **E:** Polyporate spinose pollen grain of *Ipomoaea wolcottiana*; **F:** Tricolpate pollen grain of *Disanthus cercidifolius*. (A, after Fahn, 1982; C, after Mauseth, 1998 courtesy R. A. Hilsenbebeck, Sul Ross State University; F, after Endress, 1977; rest, after Gifford and Foster, 1988). (Singh G.)

Among examples of the role of pollen grains in systematics is *Nelumbo* whose separation from Nymphaeaceae into a distinct family Nelumbonaceae is largely supported by the tricolpate pollen of *Nelumbo* as against the monosulcate condition in Nymphaeaceae.

Few examples related to palynology study:

In Magnoliidae the pollen is binucleate, In Caryophyllidae the pollen is trinucleate. In Ericaceae the pollen is in tetrads. In Asclepiadaceae pollen remain in Pollinia. Napenthaceae and Droseraceae (except Drosophyllum) have spinuliferous pollen tetrads. Such type of pollen tetrads are not found in any other plant.

Podophyllum is separated from Berberidaceae as it has united pollen grains. Some families are recognized on the basis of pollen sculpture e.g., Malvacae and Asteraceae has spinuous exine; Plumbaginacae has verrucate exine and Poaceae has smooth sulcate exine of pollen grain. On the basis of Palynological characters Fumariaceae is separated from Papaveraceae and Nelumbonaceae from Nymphaeaceae. Relationship between Polygalaceae and Ephedraceae are based on similarity between their pollen grains. Seven genera of Polygpnaceae i.e., Koenigia, Persicaria, Polygonum, Pleuropteropyrum, Bistoria, Tiniaria and Fagopyrum are different in their Pollen morphology.

2. **Phytochemistry in Relation to Taxonomy: (ChemoTaxonomy):**

The science of chemical taxonomy is based on classification of Plants on the basis of their chemical constituents related with the molecular characteristics.

Chemotaxonomy includes:
(i) Investigation of pattern of the compounds existing in plants,

(ii) Investigation pattern of the compounds in plant parts likes bark, wood, eaves, roots etc.

Basic characters as evidence come from:
(i) Flavonoids, (ii) Terpenoids, (iii) Carotenoids, (iv) Polysaccharides, (v) Alkaloids, (vi) Aminoacids, (vii) Fattyacids, (viii) Aromatic compounds, and (ix) C_3-C_4 photosynthesis etc.

Development of plant natural product chemistry revealed possibility of characterizing classifying, and establishing phyletic relationships of genera, in

(1699) it was first indicated correlating between chemical properties and morphologically character i.e., morphologically similar plants possess similar chemicals. A large variety of chemical compounds are found in plants and quite often the biosynthetic pathways producing these compounds differ in various plant groups.

Popularity of Phytochemistry is due to:

(a) Development of rapid analytical techniques.

(b) Belief that data from many sources should be employed for classification.

Classification on the basis of Mol. wt.
- → Micromolecules (mol. wt. less than 1000) (AA, Alk, Phenol, Terpenes)
- → Macromolecules (mol. wt. more than 1000) Protein, Nucleic acids etc.

Mentzar (1966) provided biogenetic classification on the basis of natural relationships between various constituents.

A. Micromolecules:

(a) Primary metabolites: These are compounds present in vital metabolic pathways. They are universal in distribution and of little taxonomic value. Eg. Organic acid, Amino acid, Sugar and Chlorophyll present in each plant. **Alanine** (in leaf extract), **proline** (in seed extract) and **asparagine** (in flower extract)

(b) Secondary metabolites: Secondary metabolites perform non-vital functions and are less widespread in plants and are the by-products of metabolism. They were earlier considered to be waste products, but they are important in chemical defense against predators, pathogens, allelopathic agents and also help in pollination and dispersal (Swain, 1977). Gershenzon and Mabry (1983) have provided a comprehensive review of the significance of secondary metabolites in higher classification of angiosperms. For eg. Alkaloids, Terpenoids, phenols, Specific Glucosids etc., present in plants.

B. Macromolecules:

Chemicals for various functions Semantides (DNA, RNA, Protein etc.) Non-semantides (Starch, cellulose etc.)

Lipids:

Members of Asteraceae lack unsaturated lipids. Lipids are heterogenous group present in storage organs. It depletes in dark.

(1) Linolenic rich seeds e.g., Rhamnaceae.

(2) Linoleic rich seeds e.g., Juglans, Liliac etc. Oleic and Palmitic rich e.g., Acanthaceae, Annonaceae, Malvaceae etc.

Pigments:

Chlorophyll and carotenoids are fat soluble Biloproteins and Anthocyanins are soluble in water. Anthocyanins and Betalins never coexist. Betalins are low molecular weight substances. Betacyanin gives purple colour and Betaxanthin gives yellow colour. It is mainly based on the supposition that related plants will have a similar chemistry e.g., in Pinus every species has different type of terpentine. In Lichen chemical methods are largely used for the identification of genera an species.

Betacyanin and Betaxanthin:

Chemistry of Betacyanins led to recognition of 10 families containing them (Centrospermae); they do not occur in plants containing anthocyanins. On this basis Caryophyllaceae and Illeceberaceae is separated from centrospermae as these two families lack betalin pigment. Takhtajan and Cronquist placed anthocyanins producing families in Caryophyllales. The Betacyanin containing families included in Centrospermae are Chenopodiaceae Portulaccaceae, Azoaceae, Cactaceae, Nyctaginaceae, Phytolaccaceae Stenospermaceae, Basellaceae, Amaranthaceae, and Didieraceae etc. These are taxonomic makers.

Glucosinolates (Mustard oil glucosides):

Glucosinolates are widely distributed in the families kept under Capparaceae (Capparidaceae) have specialized myrosin cells. Myrosin is enzyme involved in the formation of mustard oil.

Seed oil fatty acids:

Fatty acid in Juglandaceae and Juice sac fatty acid of Citrus are used as taxonomic significance. Four species of Nyssa and eleven species of *Cornus* and one species each of *Dauidua, Camptotheca* etc. are analysed and observed the percentage of Palmitic, Stearic, Linolic acid etc. and found that *Nyssa biflora* lies intermediate to *N. sylvatica* and *N. aquatica*. Petroselinic acid is found in Apiaceae.

Terpenes:

Embodem and Lewis (1967) studied the terpene composition in Salvia. β carotene is present in all green cells. Terpenoid, glycosides (cardiac glycosides) are present in Apocynaceae, Liliac, Moraceae etc. Monoterpenes are also known as essential oils. It depends on odors and essence etc. e.g., Lamiaceae, Apiaceae, Rutaceae etc. Sesquiterpene Lactones is a group of bitter tasting compounds.

Ieridoid Compounds:

These are bitter in taste e.g., monoterpenoids, Cyclopentanoids, Lactones etc. These are present in over 50 families of Sympetalae. Dahlgren kept all of them together e.g., Cornaceae, Scrophulariaceae, Gyrostemonaceae etc. Asperuloside is present in Rubiaceae and Buddlejaceae.

Lignin:

It is a highly branched polymer of three simple phenolic alcohols. Whereas gymnosperm lignin is composed of coniferyl alcohol subunits, the angiosperm lignin is a mixture of coniferyl and sinapyl alcohol subunits. The alcohols are oxidized to free radicals by peroxidase enzyme, and the freed radicals react to form lignin.

Phenolics:

These secondary pigments (Phenolics) are exclusive in nature (betalins). Phenolic compounds fall into a general class called Flavonoids. All contain characteristic flavonoid C_{15} nucleus. Most flavonoids are present in vacuole of plant cell. Phenolic compounds are inert end products of metabolism. These are widely distributed in the plant kingdom; common examples being catechol, hydroquinone, phlo- roglucinol and pyragallol. Coumarins, a group of natural phenolics, have a characteristic smell. The crushed leaves of *Anthoxanthum odoratum* can thus be identified by this characteristic odour.

Biflavonoids:

Biflavonoids contain 2 flavonoid and glycones linked by a carbon-carbon (C-C) bond. These are primitive and are found in most of the Gymnosperms. Four woody genera of angiosperms are known to contain biflavonoids e.g., *Viburnum* (Caprofoliaceae), *Garcinia* (Guttiferae), Heuea (Euphorbiaceae) and *Casuarina* (Casuarinaceae). The flavonoid pattern in monocots and dicots does not differ much except the presence or absence of ellagic acid.

Raphides:

Needle like crystals of Ca-oxalate arranged parallel to the bundles are found in families like Balsaminaceae, Onagraceae and Rubiaceae in Dicots and

Orchidaceae in Monocots. Trapa member of Onagraceae which does not contain raphides and since been separated into a new family Trapaceae.

Silica:

Silica is generally present in member of Arecaceae (Palmae) and Poaceae (Gramineae). Metcalfe (1960) reported 20 types of silica bodies in the epidermal cells of leaves and found them of taxonomic significance. Gypsum Crystals of gypsum are reported in the members of Tamaricaceae and some members of Capparidaceae and Asteraceae. It is not found in members containing Ca-oxalate crystals. Tanniferos plants are found in Sapindaceae. Mustard oils are typical of Brassicaceae. Alkaloids are found in Solanaceae (e.g., *Nicotiana*, *Datura*) Plants contain highly aromatic compounds in Lamiaceae.

Alkaloids:

Alkaloids are organic nitrogen-containing bases, They form one of the largest class of secondary metabolites, They are insoluble in water but soluble in organic solvents. Their distribution is restricted to some 20% of angiosperms. They are mostly present in storage tissues, seeds, fruits and roots. They act as chemical defence of plants against herbivory, and allelopathic reactions between plants. Tobacco alkaloid Nicotine (Nicotiana) is synthesized from nicotinic acid and caffeine (coffee beans and tea leaves) from purine. Isoquinolene alkaloids morphine, codeine and papaverine are found in opium poppy (Papaver somniferum). Their distribution is often specific and thus taxonomically significantConalium is the simplest known alkaloid found in Conium maculatum (Apiaceae). Some of them are of medicinal importance at low concentration, but toxic at high concentration. Some such as lycotonine (Delphinium), scopolamine (Datura), and atropine (Atropa) cause poison- ing of livestock.

3. **Cytology in Relation to Taxonomy:**

Cytology is the study of the morphology and physiology of cells. Normally anatomists deal with shapes, size, wall structure, pattern, etc. but cytologists deal with the internal organelles of the cell and detailed structure of cell wall.

Some evidential characters are:

(i) Chromosome number, structure, type, (ii) Chromosome meiotic behaviour, (iii) Ploidy level and type, and (iv) Chromosome aberration etc.

Cytological evidences is used for distinguishing taxa; to determine the origin of groups and to understand the evolutionary history of related taxa particularly those at the infraspecific and specific levels cytotaxonomy is a part of experimental taxonomy. Term **Karyotype** is used for phenotypic appearance of the somatic chromosome and the diagrammatic representation of karyotype is known as **Idiogram.** The study of homologies of the chromosome in the hybrids as determined in meiosis, is significant indicator in knowing the degree of genetic relationship.

The gametophytic chromosome number of diploid species is designated as base number (x). In diploids n = x, in polyploids n is multiple of x. e.g., in hexaploid sp 2n = 6x and n = 3x as 2n = 24 and n = 21. Angiosperm, the chromosome number varies greatly e.g., n = 2 in *Haplopappus gracilis* (Asteraceae) and highest is n = 132 in *Poa litloroa* (Poaceae). Homoploids (n=12) eg. Pinus, Quercus; Polyploids (n=9 or n=18 or n=27) eg. in species Asteraceae; Aneuploidy (n=6,7,8,9 0r 10) eg. in species of Brassicaceae. In Poaceae the subfamily Poideae has x = 7 and Bambusoideae has x = 12. Ploidy level also plays a significant role in taxonomy e.g., Triticum contains diploid (2n = 14), Triploid (2n = 21) and Hexaploid (2n = 42) etc., Senecio (Asteraceae) includes S. squalidus (2n = 20) a diploid, S. vulgaris (2n = 40) a tetraploid and S. combrensis (2n = 60) a hexaploid.

Chromosomes show variation in size, position of centromere and secondary construction etc. The structure of genome (chromosome set) in a species is called Karyotype and its diagrammatic representation as Idiogram. The chromosomes are commonly differentiated as **metacentric** (with centromere in middle), **submetacentric** (away from middle), **acrocentric** (near the end) or **telocentric** (at the end). The chromosomes are also characterized by their size. In addition the occurrence and position of **secondary constriction,** which demarcates a **satellite** is important in chromosomal identification and characterization. The karyotype study of members of Agavaceae confirms the shifting of Agave from Amaryllidaceae (inferior ovary) and Yucca from Liliaceae (superior ovary) into Agavaceae. The members of Agavaceae have two type of Karyotypes consisting of 5 large and 25 small chromosomes. Cyperaceae and Juncaceae are separated due to distinct floral structure. They have holocentric chromosomes and now considered closely related. Due to different karyotype of Butomus from that of Limnocharis, Hydrocharis, Tenagocharis, it is kept in Butomaceae while others are retained in Alismataceae. An interesting example of utilization of chromosomal information is family Agavaceae. The family contains about 16 genera such as Agave (and others formerly placed in Amaryllidaceae due to inferior ovary) and Yucca (and others formerly placed in Liliaceae due to superior ovary). These genera were shifted and brought into Agavaceae on the basis of great overall similarity. This was supported by the distinc- tive bimodal karyotype of Agavaceae con- sisting of 5 large chromosomes and 25 small ones.

Chromosome Behaviour at Meiosis refers to degree of sterility and occurrence of hybridization are determined by the behaviour of chromosomes during meiosis. Abnormalities in meiosis such as crossing over, non-pairing, unequal interchanges, bridge formation, translocations, lagging chromosomes etc. are utilized for taxonomic studies and comarisons. Meiotic behaviour of chromosomes is helpful in comparing the genomes to detect degree of homology e.g., *Triticum aestivum* is haxaploid (AABBDD) where 'A' is derived from *T. monococcum* (diploid) and 'B' from *Aegilops speltoides* and D is derived from *Aegilops squarrosa* (diploid). 2n = 26 is the characteristic of Amborellaceae; 2n= 16 of Trimeniaceae, Babcock (1947) separated the closely related genera on the basis of chromosomal number and morphology. Youngia is separated from Crepsis while Pterotheca was merged with Crepis. Rudall (1997) suggested transfer of Hosta (Hostaceae) Camassia and Chlorogatum (Liliaceae), to family Agavaceae on the basis of bimodal karyotype. Judd 2002 and Thorne (2003) also supported the statement.

Referred Links:

http://cdn.biologydiscussion.com/wp-content/uploads/2016/07/clip_image004-5.jpg

https://books.google.co.in/books/about/Plant_Systematics.html?id=In_Lv8iMt24C