Chapter 21

General Discussion

Pharmacognosy

The present study provides innumerable characters to distinguish all the original drug plants and their substitutes/adulterants. These are discussed below.

1. Fumaria parviflora and its substitutes/adulterants

The presence of light brown contents in cortical parenchyma, fan shaped vascular bundle and pitted fiber tracheids in root was the characteristic features of Fumaria parviflora. All these characters are conspicuously absent in all substitutes/adulterants. It also showed the presence of reticulate and scalariform thickened vessels. The stomata present in the leaves of F. parviflora was of anomocytic type and mesophyll of leaf was not differentiated into palisade and spongy parenchyma. In stem of F. parviflora the vascular bundle was capped with sclerenchymatous sheath. In case of Justicia procumbens, the centre wood dominated by vessels and vessels had elongated parallel bordered pits while in the stem the presence of interfascicular wood prosenchyma and thick-walled unicellular as well as multicellular uniseriate trichomes with broad basal cell, blunt tip and warty walls and thin and wavy lateral walls of epidermal cells were the characteristics. The leaves of Oldenlandia corymbosa showed the presence of paracytic type of stomata, cells of upper epidermis about double the size of the lower epidermis and network of lobed spongy cells and association of libriform fibres with tracheids in stem was characteristic but beside this the presence of raphide bundles was the striking character. The leaf of Peristrophe bicalyculata showed the presence of glandular and thin walled unicellular as well as multicellular uniseriate trichomes with pointed tip while the stomata was of diacytic type and the cells of epidermis were wavy. Brown deposits in cortical cells and isolated acicular crystals in pith of stem were also characteristic. Polycarpea corymbosa showed the many diagnostic characters such as leaves showed anomocytic type of stomata outer walls of epidermis thick and papillose, parenchyma containing reddish brown contents, vascular bundle capped by thick walled sclerenchymatous sheets and rosette crystals.

Absence of rays in root and stem was also characteristics. In case of *Rungia repens*, the stem showed the thick walled and rosette crystals while its leaves showed the presence of diacytic type of stomata and thick-walled unicellular as well as multicellular uniseriate trichomes with broad basal cell, blunt tip and warty walls here the stem showed the typical pitted sclerenchyma in a pairs wherein the adjoining walls were straight.

Among these plants trichomes were found absent in both *F. parviflora* and *O. corymbosa* while it was present in all other substitutes/adulterants viz. *P. corymbosa, J. procumbens, R. repens* and *P. bicalyculata*. Among them, *P. corymbosa* differs in having branched trichomes while *R. repens* and *J. procumbens* differ in having multicellular uniseriate trichomes. *J. procumbens* differs in showing groups of sclereides and *P. bicalyculata* showing presence of aciaular crystals. *O. corymbosa* differs in having raphide bundles.

2. Bergenia ligulata and its substitutes/adulterants

The B. ligulata rhizome showed the distinct characters such as presence of various shaped starch grains typically having beak and rosette crystals in cork, phelloderm, cortex, ray and in pith, loosely arranged cortical parenchyma showing light brown deposits, 'V' shaped vascular bundles with cambium and spiral and simple pitted vessels. The important characters of Aerua lanata root were thin walled cork cells, narrow compactly arranged cortical parenchyma filled with rosette crystals and occasionally with rhomboidal crystals. The root also showed secondary anomalous secondary growth where in successive rings separated by thin walled parenchyma and separated from the central phloem by thick walled parenchyma contained rosette and rhomboidal crystals. The condition of central wood was triarch was also characteristics. The root of Ammannia baccifera showed the diagnostic characters such as thick walled light brown coloured cork cells, rosette crystals and presence of collenchyma between primary and secondary vascular bundles and thick walled fibres with pointed ends. The presence of thick walled brown colour cork cells, microspheroidal crystals and angular boarded pitted vessels were the characteristic of Celosia argentea. The root of Coleus amboinicus showed presence of cortical parenchyma with the reddish brown deposits, stone cells, thin and thick walled sclereids and scalariform vessel having oblique end walls while the diagnostic characters observed in root of Glossocardia linearifolia were cork with thick light yellow coloured wavy walled cork cells, cortical parenchyma with reddish-brown

deposition, stone cells, thick walled ray parenchyma with pits and blunt ends and broad lumen fibres. Among these plants the important diagnostic character such as presence of rosette crystals in B. ligulata, were also found in Aerua lanata and Ammannia baccifera while it was found absent in Coleus amboinicus, Celosia argentea and Glossocardia linearifolia. Another important character of B. ligulata was the presence of starch grains was also found in C. amboinicus but it was big, simple, various shaped and having beak and plenty in B. ligulata while in C. amboinicus it was small, simple and compound, spherical and few. The vascular bundles in B. ligulata was 'v' shaped, while Aerua lanata, Ammannia baccifera and Celosia argentea showed the anomalous secondary growth of vascular bundles. In case of Aerua lanata the vascular bundles were separated from the central phloem by thick walled parenchyma containing rosette and rhomboidal crystals while in case of Ammannia baccifera the primary and secondary vascular bundles separated by layers of collenchyma containing rosette crystals and Celosia argentea showed discrete secondary vascular bundles separated by 2 layered thick medullary rays of rectangular cells. None of the substitutes/adulterants of B. ligulata were showed the deposition of light brown contents in ray cells which was specific to the B. ligulata. The absence of pith cells in substitutes/adulterants of B. ligulata was also distinguishing features as the source of all substitutes/adulterants was of root. The presence of crystalline matters in *C. argentea*, rhomboidal crystals in *Aerua lanata*, thick cellulosic wall of parenchyma in Ammannia baccifera and stone cells in G. linearifolia was the distinguishing characters.

3. Glycyrrhiza glabra and its substitutes/adulterants

Microscopically Abrus precatorius, Alysicarpus longifolius, and Taverniera cuneifolia the substitutes/adulterants of Glycyrrhiza glabra were similar to Glycyrrhiza in characters like starch grains and prismatic crystals of calcium oxalate. Maerua arenaria was very different in the absence of such characters and in having parenchyma with oil droplets. The absence of tyloses and indistinct cambium in *T.cuneifolia* were the distinguishing characters and it was also differed in having characters like collapsed condition of phellogen, polygonal shaped cortical parenchyma, typical found in a pairs wherein the adjoining walls were straight while Alysicarpus longifolius and Abrus precatorius were differ in having ray parenchyma containing prismatic crystals and stone cells respectively and both had indistinct

phellogen. In *Alysicarpus longifolius* the vessel were characteristically associated with pitted parenchyma.

4. Polygala senega and its substitutes/adulterants

In case of substitutes/adulterants of *Polygala senega*, all substitutes/adulterants are look different. *Acalypha indica* differs in having rosette crystals, *Adhatoda vasica* in having pitted stone cells with distinct striations, *Polygala chinensis* showing deposition of pale yellow amorphous masses in some cortical and phloem parenchyma of shows parenchyma and *Xerornphis spinosa* differs in having starch grains.

5. Saraca indica and its substitutes/adulterants

The diagnostic characters observed in the bark of *Saraca indica* were brown thick walled cork cells, spherical starch grains, thick walled narrow lumened stone cells, sclereids, rhomboidal crystals, septet and aseptet fibers, crystal fibres. The bark of *Bauhinia variegate* showed the presence of distinct characters such as thin and thick walled cork cells, parenchyma containing reddish-orange deposits, starch grains, sclereids, thick walled narrow lumened fibers, rhomboidal and prismatic crystals. The presence of stone cells in the medullary rays and association of it with crystal fibers was also characteristics. *Bombax ceiba, Polyalthia longifolia, Shorea robusta,* and *Trema orntentails* closely resemble to *Saraca* in terms of microscopic characteristics like starch grains, crystals and stone cells. The differences observed are thin walled stone cells with simple pits in *Bauhinia variegata,* thick walled stone cells with distinct striations and pit canals in *Shorea robusta* and parenchyma with yellow content and elongated stone cells with branched lumen in *Trerna orientalis.*

Besides these characters the organoleptic characters like colour, surface taxture, fracture etc. of individual drugs mentioned in early Chapters which was characteristic to that drugs could be used for differentiating the genuine drugs from their substitutes/adulterants.

304

Phytochemistry

1. Fumaria parviflora and its substitutes/adulterants

In case of the substitutes/ adulterants of Fumaria parviflora, Oldenlandia corymbosa seems to close to that of Fumaria in having flavonol quercetin, 3'-OMe quercetin, phenolic acid vanillic acid and presence of alkaloids, steroids and tannins, while it differs in having the flavonol 3', 4'-di OMe quercetin, phenolic acids syringic, ferulic (cis-and trans- isomers), p-hydroxy benzoic, protocatechuic, melilotic acids, and iridoids. Rungia repens is similar in having quercetin, vanillic acid, alkaloids, steroids and tannins and differs in having 7'- OMe quercetin and kaempferol along with syringic, ferulic (cis- and trans-isomers), p-coumaric and p-hydroxybenzoic acids. Polycarpea corymbosa is similar in having vanillic acid, alkaloids, steroids and tannins and differs in having apigenin, acacetin, 3'- OMe luteolin, 7,3-di OMe quercetin. Justicia procumbens and Peristrophe bicalyculata are similar in having vanillic acid and steroids but Justicia differs in having 6-OH kaempferol and 7- OMe 6- OH kaempferol, and ferulic (cis-and trans- isomers) acid and Peristrophe in having ferulic (cis-and trans- isomers) acid and Peristrophe in having ferulic (cis-and trans- isomers) acid and Peristrophe in having ferulic (cis-and trans- isomers) acid and Peristrophe in having ferulic (cis-and trans- isomers) acid and Peristrophe in having ferulic (cis-and trans- isomers) acid and Peristrophe in having ferulic (cis-and trans- isomers) acid and Peristrophe in having ferulic (cis-and trans- isomers) acid and Peristrophe in having ferulic (cis-and trans- isomers) and p-hydroxy benzoic acids.

2. Bergenia ligulata and its substitutes/adulterants

In case of *Bergenia ligulata, Ammannia baccifera* is similar to *Bergenia* in having steroids, vanillic, syringic and gallic acids and differs in having alkaloid ephedrine and melilotic acid. *Coleus* similar in showing presence steroids, vanillic and syringic acids and differs in having melilotic acid. *Aerua lanata* similar in having vanillic, syringic acids and differ in having quinones and ferulic (*cis-and trans*-isomers) acid whereas *Glossocardia linearifolia* and *Celosia argentea* are similar in having flavonol acacetin and ferulic (*cis-* and *trans-* isomers) acid and ferulic (*cis-* and *trans-* isomers) acid and *ferulic* (*cis-* and *trans-* isomers) acid and ferulic (*cis-* and *trans-* isomers) acid and *ferulic* (*cis-* and *trans-* isomers) acid and *Celosia* differs in having flavonol acacetin and ferulic (*cis-* and *trans-* isomers) acid and *Celosia* differs in having phenolic acids such as *p*-coumaric and melilotic acids.

3. Glycyrrhiza glabra and its substitutes/adulterants

Among the substitutes/adulterants of *Glycyrrhiza*, *Abrus precatorius* and *Taverniera cuneifolia* seem to be more close to that of *Glycyrrhiza* in having saponin, coumarins, alkaloids, steroids and phenolic acids such as vanillic, syringic, ferulie

(cis- and trans-isomers) but, Abrus differs in having quinones and melilotic acid and Taverniera cuneifolia in having o-coumaric and p-hydroxybenzoic acids whereas Maerua arenaria and Alysicarpus longifolius are similar in having saponin, steroids, vanillic and syringic acids while differs in having melilotic acid.

4. Polygala senega and its substitutes/adulterants

In case of *Polygala senega*, all substitutes/adulterants are similar to that of *senega* in having saponins, vanillic and syringic acids but *Acalypha indica* differs in having alkaloids, *p*- coumaric and ferulic(*cis*- and *trans*-isomers) acids. *Adhatoda vasica* differs in having alkaloids, *p*- coumaric and *p*-hydroxy benzoic acids, *Polygala chinensis* in having coumaric acids and *Xeromphis spinosa* in having scopoletin.

5. Saraca indica and its substitutes/adulterants

Among the substitutes/adulterants of *Saraca asoca, Trema orientalis* seems to be more close to that of *Saraca asoca* in showing the presence of flavonoids, steroids, cyanidin, vanillic and syringic acids but differs in having ferulic (*cis-* and *trans*isomers)acids. *Bauhinia variegata* is similar in having steroids, vanillic and syringic acids but differs in having kaempferol and *o*-coumaric acid. *Bombax ceiba* is similar in having cyanidin, vanillic and syringic acids but differs in having ferulic (*cis-* and *trans-* isomers) acid, whereas *Polyalthia longifolia* and *Shorea robusta* are similar in having vanillic and syringic acids but *Polyalthia* differs in having ferulic (*cis-* and *trans-* isomers)acid and *Shorea* in having 3- OMe quercetin, pelargonidin derivatives and *p*- hydroxy benzoic acids.

The work on all these substitutes/adulterants provided a large amount of data useful in understanding the multifarious activities of these drugs and in quality control procedures. The phytochemical studies revealed that all the substitutes/adulterants, in spite of having more dissimilarity are rich in phytochemicals such as **flavonols**, **anthocyanidins**, **phenolic acids and mucilages**. Flavonols are the most common flavonoids. The flavonols located are kaempferol, quercetin, acacetin etc. and their various methoxylated its derivatives. Almost all these compounds are physiologically active bioflavonoids. Tannins especially proanthocyanins also are widely distributed in these plants. Alkaloids were rather rare, located in only few plants. The phenolics reported above are found to exert a multitude of pharmacological properties. Of late, Flavonoids which are found to be the important components of the medicinal plants screened here are found to have profound health benefits. **Quercetin**, the most common flavonol, appeared to have many beneficial effects on human health including cardiovascular protection, anti-cancer activity, antiulcer effects, antiallergic activity, cataract prevention and antiviral and anti-inflammatory effects (Miller, 1996) and also inhibits lipid peroxidation *in vitro*. **Kaempferol** had a stimulatory effect on alkaline phosphatase activity in MG-63 human osteoblasts through ERK and estrogen receptor pathway. It was also shown to inhibit proliferation and increase mediator content in human leukemic mast cells. The activities of phenolic acids, which are found to be omnipresent, as well as those of quinones also are well recognised.

Another important chemical component overlooked in almost all the studies are the simple phenolics such as phenols, phenolic acids, coumarins etc. They are the minor components, which are never attributed with any activity. It is in this context, Duke's (1997) observations are interesting and informative. Duke (1997) describes ferulic acid, gentisic acid, kaempferol glycosides and salicylic acid as pain relievers while ascorbic acid, cinnamic acid, coumarin and resveratrol are explained to be antiinflammatory. The present study revealed that all the plant parts screened contained a variety of phenolic acids, that too in good concentrations. These compounds may exert their own specific pharmacological actions or add to the activities of other compounds or act as efficient antioxidants. This variety of chemicals and their richness (concentration) in a medicinal herb is of great value in assessing its property. Duke's database states that both coriander and liquorice contain 20 chemicals with antibacterial action; oregano and rosemary have 19; ginger17; nutmeg15; cinnamon and cumin 11; Black pepper 19; Bay 10 and garlic 13. Quantity wise, liquorice contains up to 33%; bactericidal compounds (dry weight basis), thyme 21%, oregano 88%, rosemary 4-8%, coriander 22% and fennel 1.5%.

The role of **mucilages** in general in medicinal plants is never understood properly. It was not considered as a pharmacologically active component. But of late, the mucilages are found to exert a large number of pharmacological actions. They were known laxative agents, demulcents, emollients and anti-diarrhoeal agents. They also check fermentation, bacterial growth and adsorb toxins and wastes helping their elimination from the body. Even cholesterol is being lowered by these mechanisms, as also there are repots in which they cause a blood sugar lowering effects. Immunostimulating activities of polysaccharides have been brought out recently into light by studies on traditional Chinese and Japanese medicines (Chang, 2002). Antiulcerogenic activities and stimulating proliferation of bone marrow cells are the other activities attributed to polysaccharides (Pengelly, 2006). In this context the presence of mucilages in all the drugs screened, that to in good amounts, is highly significant. These mucilages are found to be differing in their monomer composition, and also vary from plant parts to plant parts in their chemistry. The activities of these mucilages is to be studied in detail to understand their role in all these substitutes/adulterants or other medicinal plants.

HPTLC fingerprinting

Though the methods of extraction and chromatographic conditions for all the substitutes/adulterants were kept identical, the HPTLC chromatograms obtained when the HPTLC plate was scanned at 254 nm and 366 nm showed immense variations.

HPTLC profile of *F. parviflora* and its substitutes/adulterants observed under UV 254 nm showed that *J. procumbens* found more close to *F. parviflora* as it is similar in 3 peaks next is *O. corymbosa* is similar in only 2 peaks whereas under UV 366 nm *J. procumbens* is similar in 4 peaks, *R. repens* in 3 peaks, *P. corymbosa* and *P. bicalyculata* similar in 2 peaks. while *O. corymbosa* was not show any peaks similar to that of *F. parviflora*. This indicates that *J. procumbens* has more common compunds and is the closest to *F. parviflora*.

HPTLC profile of *B.ligulata* and its substitutes/adulterants observed under UV 254 nm showed that presence of 5 similar peaks keep the drug more close to *B.ligulata* while *G. linearifolia* is not show any common peak to that of *B.ligulata* showing that *G. linearifolia* is total different.

HPTLC profile of *G. glabra* and its substitutes/adulterants observed under UV 254 nm showed that *T. cuneifolia* was similar in 3 peaks while *Abrus precatorius* and *Alysicarpus longifolius* were similar in 1 peak and *Maerua arenaria* was not show any peak similar to that of *G. glabra* showed that among all substitutes/adulterants, *T. cuneifolia* is found more close to *G. glabra*.

HPTLC profile of *Polygala senega* and its substitutes/adulterants showed that at UV 254 nm *Xeromphis spinosa* is similar in 2 and under UV 366 nm 4 peaks seemed that *Xeromphis spinosa* is the closest drug to *Polygala senega*.

HPTLC profile of *S. indica* and its substitutes/adulterants observed under UV 254 nm showed that *P. longifolia* was similar in 2 peaks and *T. orientalis*, *B. ceiba* and *S. robusta* are similar in 1 peak only but at UV 366nm *T. orientalis* is similar in 3

peaks and *P. longifolia* in one peak only showing that *T. orientalis* is more close to *S. indica* followed by *P. Longifolia*.

However, the HPTLC fingerprint for each substitutes/adulterants needs to developed separately as a quality control parameter for the plant.

Physico-chemical analysis

Total ash content

The total ash content of a crude drug is the inorganic residue remaining after incineration. It includes not only the inorganic salts, e.g. Calcium oxalate, occurring naturally in the drug; but also inorganic matter from external sources. The ash value may be useful in to ensure the absence of an undue proportion of extraneous mineral matter introduced accidently or by design due to the type of manufacturing process used, e.g. earth, sand, floor sweepings etc. and to detect adulteration of the drug. In this study the total ash content of all the drugs does not show significant variation along the material collected in different season. Total ash content of Fumaria parviflora and its substitutes/adulterants along the material collected in different season does not show significant variation and the closest value to the substitute/adulterant is Polycarpea corymbosa while other substitutes/adulterants have higher ash values i.e almost double the value of Fumaria parviflora. In case of Bergenia ligulata the closest value to the substitute/adulterant is of Aerua lanata and Coleus amboinicus but diffrence in values are of more than 3%. Among the substitutes/adulterants Glycyrrhiza glabra, the total ash value of Taverniera cuneifolia is almost equal to Glycyrrhiza glabra. The ash values of Adhatoda vasica and Polygala chinensis are found to be close to Polygala senega as compare to other the substitute/adulterant while in case of substitutes/adulterants of Saraca indica. Shorea robusta and Bombax ceiba are close to saraca indica.

The Total Ash content mainly is a measure of the presence of inorganic compounds. A larger value indicates that the plant material contains more of inorganic compounds.

Acid insoluble ash content

Crude drugs containing larger quantity of calcium oxalate, can give variable results depending upon the conditions of ignition. Treatment of the ash with Hydrochloric acid leaves virtually only silica. Hence acid insoluble ash forms a better test to detect and limit excess of soil present as an impurity in the drug, than does the total ash. In this study the acid insoluble ash content of all the drugs does not show significant variation along the material collected in different season. In case of acid insoluble ash content of Fumaria parviflora the closest value to the substitutes/adulterants is of Oldenlandia corymbosa. Amongst other substitutes/adulterants, the acid insoluble ash content of Coleus amboinicus, Celosia argentea and Aerua lanata and among substitutes/adulterants of Glycyrrhiza glabra, the Abrus precatorius, Taverniera cuneifolia, Alysicarpus longifolius are found tobe more close to that of genuine drug. The acid insoluble ash content of substitutes/adulterants of Polygala senega does not show much variatons, whereas the acid insoluble ash content value of aduterant Shorea robusta is same as to that of Saraca indica.

Alcohol soluble extractive

These are indicative of the approximate measure of chemical constituents of crude drug. The closest value of alcohol soluble extractive to their substitutes/adulterants of Fumaria parviflora, Bergenia ligulata, Glycyrrhiza glabra, Polygala senega and Saraca indica are of Rungia repens, Glossocardia linearifolia, Taverniera cuneifolia. Polygala chinensis Polvalthia and longifolia respectively. However extractive value of Celosia argentea showed the maximum and is higher than that of Bergenia ligulata.

Water soluble extractive

Amongst all substitutes/adulterants of Fumaria parviflora, Bergenia ligulata, Glycyrrhiza glabra, Polygala senega and Saraca indica show the closest value of water soluble extractive are shown by Justicia procumbens, Glossocardia linearifolia, Taverniera cuneifolia, Polygala chinensis and Trema orientalis respectively. However extractive value of substitutes/adulterants Rungia repens, Celosia argentea, Polygala chinensis and Polyalthia longifolia showed the maximum, higher than that of their genuine drugs.

Higher values of water and alcohol soluble extractive indicate the richness of the plant material in context to its phytochemicals.

310