

3.2.3. Microscopic Evaluation

Microscopic evaluation of a drug allows detailed examination so that the organised crude drugs (either in entire or powdered forms) can be identified by their known histological features. It is a qualitative evaluation type involving identification of organised drugs by cutting them in Transverse Section (T.S.), Longitudinal Section (L.S.), Radial Longitudinal Section (R.L.S.), or Tangential Longitudinal Section (T.L.S.).

Due to its magnification property, a microscope enlarges the minute structure being studied and also confirms the structural details of drugs obtained from plants. Effective results can be obtained by distinguishing the cellular structure using various reagents or stains. Microscopic evaluation also involves studying constituents by applying chemical

tests to small quantities of powdered drugs or to histological sections of drugs. Histological studies are conducted with very thin sections of drugs. Detailed study of the characteristics of cell wall, cell contents, trichomes, fibres, vessels, etc. can be obtained by microscopic evaluation.

Microscopic evaluation also involves microscopic linear measurement and quantitative microscopy.

3.2.3.1. Leaf Constants

Given below are some of the leaf constants:

- 1) **Palisade Ratio:** It is the average number of palisade cells present below each epidermal cell. It is determined using powdered drugs.
- 2) **Vein-Islet Number:** It is the number of vein-islets present in per sq. mm of the leaf surface midway between the midrib and margin.
- 3) **Vein-Termination Number:** It is the number of veinlet terminations present in per sq. mm of the leaf surface midway between the midrib and margin.
- 4) **Stomatal Number:** It is the average number of stomata present in per sq. mm of leaf epidermis.
- 5) **Stomatal Index:** It is the percentage which the number of stomata forms to the total number of epidermal cells; each stoma being counted as one cell. It is calculated by:

$$S.I. = \frac{S}{E + S} \times 100$$

Where,

S.I. = Stomatal Index

S = Number of stomata per unit area

E = Number of epidermal cells in the same unit area

The leaf constants are determined for microscopic evaluation of some leaf drugs, e.g., senna, datura, digitalis, buchu, coca, belladonna, etc.

3.2.3.2. Stomata

A stoma is a **minute epidermal opening** having a **central pore**, and two kidney-shaped **guard cells** containing chloroplasts; the guard cells are covered by variable number of subsidiary (epidermal) cells. All the plants do not necessarily have stomata; **for example**, the leaves of bryophytes and submerged leaves of aquatic parts lack stomata.

Stomata are basically present in leaves (green plant parts), most abundantly in dicot leaves. In some plants, they are present on the upper surface of leaves, in some on the lower surface of leaves (e.g., coca and cherry), while in others on both the surfaces (e.g., senna, belladonna, datura, etc.). The stomata distribution between upper and lower epidermis in dicot leaves presents great variation. Stomata may also be present in stems (ephedra), flowers (clove), and fruits (fennel). They are absent in roots.

Functions of Stomata: Its primary function is **gaseous exchange** and secondary function is **transpiration**.

Types of Stomata: Stomata are divided into **four types** based on the **type of guard cells** and **arrangement of subsidiary cells**. The fourth type of stomata shows diagnostic significance. The stomata types are:

- 1) Moss type,
- 2) Gymnospermous type,
- 3) Gramineous type, and
- 4) Dicotyledonous type.

Dicotyledonous Stomata: These stomata based on the form and arrangement of subsidiary cells, are divided into:

- 1) **Paracytic or Rubiaceae or Parallel-Celled Stomata:** These stomata have two guard cells covered with two subsidiary cells, whose long axes are arranged parallelly to that of stoma, e.g., coca and senna leaves.
- 2) **Diacytic or Caryophyllaceous or Cross-Celled Stomata:** These stomata have two guard cells covered with two subsidiary cells arranged at right angle to the stoma, e.g., peppermint, spearmint, and vasaka.
- 3) **Anisocytic or Cruciferous or Unequal-Celled Stomata:** These stomata have two guard cells covered with three subsidiary cells (one is smaller than the other two), e.g., belladonna, datura and stramonium.
- 4) **Anomocytic or Ranunculaceous or Irregular-Celled Stomata:** These stomata are surrounded by variable number of subsidiary cells (similar to other epidermal cells), e.g., buchu, digitalis, and lobelia.

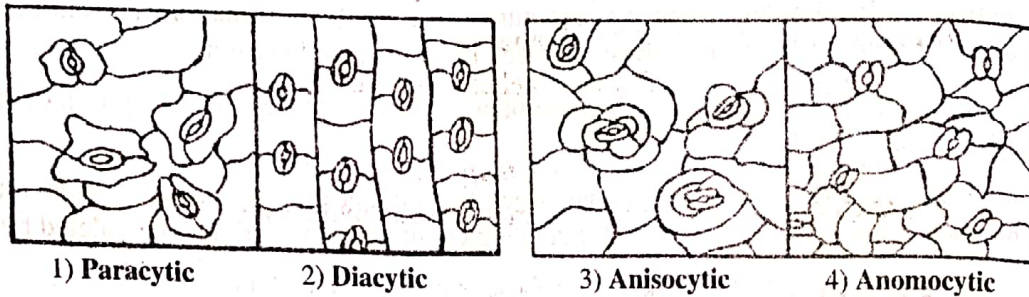


Figure 3.6: Types of Dicotyledonous Stomata

3.2.3.3. Trichomes

Trichomes are also important diagnostic feature used for identifying drugs and detecting adulterants. Trichomes (or **plant hair**) are outgrowths of the epidermal cell either **tubular**, or **elongated**, or **glandular** in shape.

The two parts of trichomes are **root** (inside the epidermis) and **body** (outside the epidermis). Although trichomes are functionless, sometimes they perform secretory functions; they excrete water and volatile oil (e.g., in peppermint). Trichomes are found in leaves (e.g., senna and digitalis), seeds (e.g., nux-vomica and strophanthus), fruits (e.g., *Helicteris isora* and lady finger), etc. Thus, they are present only in the aerial parts of plant, and not in roots. They are absent in coca, sevin, and hemlock; and sometimes present in buchu and henna.

Trichomes are classified as follows based on the structure and number of cells present in them (figure 3.7):

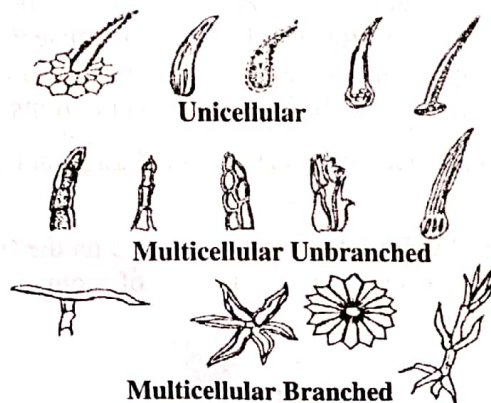


Figure 3.7: Types of Covering Trichomes

1) **Covering Trichomes**i) **Unicellular**

- Lignified trichomes (e.g., nux-vomica and strophanthus).
- Short, sharply pointed, curved trichomes (e.g., cannabis).
- Large, conical, strongly shrunken trichomes (e.g., lobelia).
- Short, conical, unicellular trichomes (e.g., tea and buchu).
- Strongly waved, thick walled trichomes (e.g., *Yerba santa*).

ii) **Multicellular – Unbranched Trichomes**a) **Uniseriate**

- Bi-cellular, conical trichomes (e.g., datura).
- Three-celled long trichomes (e.g., stramonium).
- Three- to four-celled long trichomes (e.g., digitalis).
- Four- to five-celled long trichomes (e.g., belladonna).

b) **Biseriate:** e.g., *Calendula officinalis*.c) **Multiseriate:** e.g., Male fern.iii) **Multicellular - Branched Trichomes**

- Stellate:** e.g., Hammamelis, helicteris-isora.
- Peltate:** e.g., Humulus.
- Candelabra:** e.g., *Verbascum thapsus*.
- T-shaped Trichomes:** e.g., *Artemisia* and pyrethrum.

2) **Glandular Trichomes:** At the top of these trichomes, glandular (spherical) cells are present. These trichomes are further classified into:

- Unicellular Glandular Trichomes:** In these trichomes, the stalk is absent, e.g., piper, betel, and vasaka.

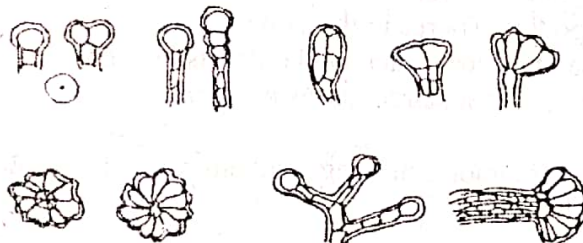


Figure 3.8: Types of Glandular Trichomes

ii) **Multicellular Glandular Trichomes:** These trichomes are further divided into:

- Trichomes with unicellular head and stalk, e.g., *Digitalis purpurea*.
- Trichomes with unicellular head and uniseriate multicellular stalk, e.g., *Digitalis thapsi*, belladonna, etc.
- Trichomes with multicellular head, and multicellular, biseriate stalk, e.g., Santonica and plants of Compositae, such as sunflower, etc.
- Trichomes with unicellular stalk and biseriate head, e.g., *Digitalis purpurea*.
- Trichomes with short stalk with secreting head formed of rosette or club-shaped cells, e.g., mentha species.
- Trichomes with multicellular, multiseriate, cylindrical stalk, and a rosette of secretory cells, e.g., *Cannabis sativa*.
- Trichomes with multicellular, multiseriate head and multicellular, uniseriate stalk, e.g., Indian hemp and tobacco.

3) **Hydathode:** These are water absorption or secretion organs developed in some particular plants, e.g., *Piper betel*, London pride, etc.