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## UNIT – III

### 4. Cultivation, Collection, Processing & Storage of crude drugs:

- (A) Factors influencing cultivation of medicinal plants, Type of Soils & fertilizers of common use.
  - (B) Pest & Pest Management, natural pest control agents.
  - (C) Plant hormones and their applications.
  - (D) Polyploidy, Mutation & Hybridization with reference to medicinal plants
  - (E) Poly Houses/Green Houses for cultivation
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## CULTIVATION

Though several countries in the world have a rich heritage of herbal drugs, very few can put claim for their procurement only from cultivated species. It is recently that some of these drugs have been subjected to systematic cultivation based on modern scientific information. Our reliance on wild sources of crude drugs and the lack of information of sound cultivation technology have resulted in gradual depletion of raw material from wild sources. Eventhough, cultivation of medicinal plants offers wide spectrum of advantages over their wild sources, it may be an uneconomical proposition for certain types of crude drugs which occur abundantly in their natural habitat e.g. nux-vomica, acacia, myrobalan, etc. On the other hand, crude drugs like cardamom, clove, Indian hemp, poppy latex, tea, cinchona, ginger, linseed, isabgol, ceylon, cinnamon, saffron, peppermint, fennel, etc. are obtained from cultivated plants. The cultivation of vegetable drugs involves convergence of various factors from agricultural and pharmaceutical sphere, such as soil, climate, rainfall, irrigation, altitude, temperature, use of fertilizers and pesticides, genetic manipulation and biochemical aspects of natural drugs. When all such factors are precisely applied, the new approach to scientific cultivation technology emerges out.

The advantages of cultivation may be briefly summarized as follows:

1. It ensures quality and purity of medicinal plants. Crude drugs derive their utility from chemical contents in them. If uniformity is maintained in all operations during the process of cultivation, drugs of highest quality can be obtained. Cultivation of rhizomes demands an adequate quantity of fertilizers and proper irrigation. Systematic cultivation results in raising a crop with maximum content of volatile oil and other constituents. The examples of ginger, turmeric and

liquorice can be cited to illustrate this point. If the cultivated plants are kept free of weeds, the contamination of crude drugs can be conveniently avoided.

2. Collection of crude drugs from cultivated plants gives a better yield and therapeutic quality. However, it is a skilled operation and requires some professional excellence. If the collection of crude drugs for market is done from cultivated plants by skilled and well-experienced personnel, the high yield and therapeutic quality of drugs can be maintained. For example, collection of latex from poppy capsules and oleo-resins from *Pinus* species, If done by experiences persons, can result in better yield of crude drugs. Preservation of green colour of senna leaves and minimizing the deterioration of cardiac glycosides in freshly collected leaves of digitalis can be achieved only by highly skilled labour.
3. Cultivation ensures regular supply of a crude drug. In other words, cultivation is a method of crop-planning. Planning a crop-cultivation regularizes its supply and as a result the industries depending upon crude drugs, do not face problem of shortage of raw material.
4. The cultivation of medicinal and aromatic plants also leads to industrialisation to a greater extent. The cultivation of coffee and cocoa in Kerala has given rise to several cottage and small scale industries. The cultivation of cinchona in West Bengal has led to the establishment of the cinchona-alkaloid factory near Darjeeling. The government owned opium factory at Ghaziabad is an eloquent testimony to the significance of well planned cultivation of poppy.
5. Cultivation permits application of modern technological aspects such as mutation, polyploidy and hybridization.

The high cost of cultivated drugs as compared to wild sources and the losses due to ecological imbalances such as storms, earthquakes, floods, droughts, etc. are major disadvantages of cultivation.

## **METHODS OF PROPAGATION**

Medical plants can be propagated by two usual methods as applicable to non-medicinal plants or crops. These methods are referred as Sexual method and Asexual method. Each of these methods have certain advantages, and also, disadvantages.

## **1. Sexual method (Seed propagation)**

The sexual method of propagation enjoys following advantages.

1. Seedlings are long-lived (in case of perennial drugs).
2. Seedling is comparatively cheaper and easy to raise.
3. Propagation from seed has been responsible for production of some chance-seedlings of highly superior merits which may be of great importance to specific products, such as orange, papaya, etc.
4. In case of plants where other vegetative methods cannot be utilised, propagation from seeds is the only method of choice.

Sexual method suffers from following limitations.

1. Generally, seedling-trees are not uniform in their growth and yielding capacity, as compared to grafted trees.
2. They require more time to bear, as compared to grafted plants.
3. The cost of harvesting, spraying of pesticides, etc. is more as compared to grafted trees.
4. it is not possible to avail of modifying influence of root stocks on scion, as in case of vegetatively propagated trees.

For propagation purpose, the seeds must be of good quality. They should be capable of high germination rate, free from diseases and insects and also free from other seeds, used seeds and extraneous material. The germination capacity of seeds is tested by rolled towel test, excised embryo test, etc. The seeds are preconditioned with the help of scarification to make them permeable to water and gases. If the seeds are not to be germinated in near future, they should be stored in cool and dry place to maintain their germinating power. Long storage of seeds should be avoided.

Before germination, sometimes a chemical treatment is given with stimulants like gibberellins, cytokinins, ethylene, thiourea, potassium nitrate or sodium hypochlorite. Gibberellic acid ( $GA_3$ ) promotes germination of some type of dormant seeds and stimulates the seedling growth. Many freshly harvested dormant seeds germinate better after soaking in potassium nitrate solution. Thiourea is used for those seeds which do not germinate in dark or at high temperatures.

To enhance germination, many times, special treatments to seeds are given, such as soaking the seeds in water for a day. e.g. castor-seeds, and other slow-germinating seeds. Sometimes, seeds are soaked in sulphuric acid. e.g. henbane seeds. Alternatively, testa is partially removed by grind-stone or by pounding seeds with coarse sand. e.g. Indian senna.

## 2. Asexual method

In case of asexual method of vegetative propagation, the vegetative part of a plant, such as stem or root, is placed in such an environment that it develops into a new plant. Asexual propagation enjoys following advantages.

1. There is no variation between the plant grown and plant from which it is grown. As such, the plants are uniform in growth and yielding capacity. In case of fruit trees, uniformity in fruit quality makes harvesting and marketing easy.
2. Seedless varieties of fruits can only be propagated vegetatively e.g. grapes, pomegranates and lemon.
3. Plants start bearing earlier as compared to seedling trees.
4. Budding or grafting encourages disease-resistant varieties of plants.
5. Modifying influence of root-stocks on scion can be availed of.
6. Inferior or unsuitable varieties can be over-looked.

It suffers from following disadvantages:

1. In comparison to seedling trees, these are not vigorous in growth and are not long-lived.
2. No new varieties can be evolved by this method.

**(a) Vegetative propagation:** It is done by sowing various parts of the plants in well prepared soil. The following are the examples of vegetative propagation.

- |             |      |  |
|-------------|------|--|
| 1. Bulls    | .... | Squill, garlic                         |
| 2. Corms    | .... | Colchicum, saffron                     |
| 3. Tubers   | ...  | Jalap, aconite, potato                 |
| 4. Rhizomes | ...  | Ginger, turmeric                       |
| 5. Runners  | ...  | Peppermint                             |
| 6. Suckers  | ...  | Mint, pineapple, chrysanthemum, banana |
| 7. Offsets  | ...  | Aloe, Valerian                         |
| 8. Stolons  | ...  | Arrow-root, liquorice                  |

**(b) Aseptic methods of micropropagation :** It is novel method for propagation of medicinal plants. In micropropagation, the plants are developed in an artificial medium under aseptic conditions from fine pieces of plants like single cells, callus, seeds, embryos, root tips, shoot tips, pollen grains, etc.

### **FACTORS AFFECTING CULTIVATION**

Cultivation of medicinal and aromatic plants takes cognizance of plant habitats and climatic requirements for their favorable growth.

The factors which are given special attention for cultivation are listed below:

1. Altitude, temperature and humidity
2. Rainfall or irrigation
3. Soil and soil fertility
4. Fertilizers
5. Pests and pest control

#### **1. Altitude, temperature and humidity**

Altitude is a very important factor in cultivation of medicinal plants. Tea, cinchona and eucalyptus are cultivated favourably at an altitude of 1000-2000 metres. Cinnamon and cardamom are grown at a height of 500-1000 metres, while senna can be cultivated at sea level. The following are the examples of medicinal and aromatic plants indicating the altitude for their successful cultivation (Table)

**Altitude for Drug cultivation**

<b>Plant</b>	<b>Altitude (metres)</b>
Tea	1000-1500
Cinchona	1000-2000
Camphor	1500-2000
Cinnamon	250-1000
Coffee	1000-2000
Clove	upto 900
Saffron	upto 1250
Cardamom	600-1600

**Temperature** is another factor affecting the growth of a plant. Excessive temperature, as well as, frost also affect quality of medicinal plants adversely. The following

are few examples of ranges of temperature necessary for luxuriant growth of certain medicinal plants. (Table)

<b>Altitude for Drug cultivation</b>	
<b>Plant</b>	<b>Optimum Temperature (°F)</b>
Cinchona	60-75
Coffee	55-70
Tea	70-90
Cardamom	50-100

Comphor and coffee cannot withstand frost, whereas saffron needs only cold climate and pyrethrum requires dry weather for cultivation.

## **2. Rainfall or irrigation**

Except the xerophytic plants like aloe, acacia and few others, most of the plants need either proper arrangements for irrigation or sufficient rainfall for their favourable development. In few cases, well distributed rainfall throughout the year is desired.

## **3. Soils and soil fertility**

**Soil** is the most important natural resource as it supports growth of all plants. Soil provides mechanical anchorage, as well as, water and essential plant food elements for plant growth. The capacity of soil to supply plant nutrients in quantities and proportions required and to provide a suitable medium for plant growth is known as soil fertility. Soil makes chemical make up and nutrients available to plants. Plant growth depends upon physical arrangement and nature of soil particles, organic matter content of soil and its living organisms.

The commonly known soil is the shallow upper layer and is the friable material in which plants find foot-hold and nourishment.

Clay is one of the highly weathered portions of the soil, consisting of finest particles. This provides the soil adhesive and cohesive properties and also holds plants nutrients with the result that nutrients are not lost through leaching.

Soil consists of mineral matter, air, water and organic matter. It is the mineral matter which makes a lot of difference in various forms of soil. Mineral matter may be coarse gravel, coarse sand or in the form of finest particles of clay and silt. Air and water give rise to pores, while purified and decayed plant and animal parts constitute the organic matter.

Depending upon the size of mineral matter, following names are given to the soil.

<b>Particle size (diameter)</b>	<b>Types of Soil</b>
Less than 0.002 mm	... Fine clay
0.002 to 0.02 mm	... Coarse clay or silt
0.02 to 0.2 mm	... Fine sand
0.2 to 2.00 mm	... Coarse sand

Depending upon the percentage covered by clay, soils are classified as given below.

<b>Types of soil</b>	<b>Percentage covered</b>
1. Clay	... More than 50% of clay
2. Loamy	... 30 to 50% of clay
3. Silt loam	... 20 to 30% to clay
4. Silt loam	... 10 to 20 % of clay
5. Sandy soil	... More than 70% sandy soil
6. Calcareous soil	... More than 20% of lime

Any type of soil containing less than 0.5% of organic matter is described as poor. If more than 1.5 to 5% of organic matter is present, it is described as rich soil. The soil with 0.5 to 1.5% of humus is termed as intermediate soil.

A soil good for plant growth should have half of the pore spaces filled with water and the rest with air, since good aeration is essential for root development.

The pH of soil decides favourable growth of plants and presence of micro-organisms. The maximum availability of plant nutrients is in between the pH range of 6.5 to 7.5 To bring the pH to neutral, acidic soils can be limed or alkaline soils can be reclaimed by application of gypsum.

Acidic soils are not suitable for leguminous plants due to poor development of nodular bacteria. Therefore, ground nut, sunflower seeds, cotton and rice grow better in

alkaline soils only. Acidic pH is disadvantageous as it solubilizes more iron. Some of the plants like, tobacco, cinchona, tea, and potato grow well only in acidic soils. In alkaline soils, phosphorus is converted to insoluble forms of calcium phosphate and so it cannot be made available to plants.

### **Soil fertility**

It is the capacity of soil to provide nutrients in adequate amounts and in balanced proportion to plants. If cropping is done without fortification of soil with plant nutrients, soil fertility gets lost. It is also diminished through leaching and erosion. Soil fertility can be maintained by addition of animal manures, nitrogen-fixing bacteria or by application of chemical fertilizers. The latter is time-saving and surest of all above techniques.

### **4. Fertilizers**

For the vegetative growth, plants need carbon dioxide, sunlight, water and mineral matter. Plants are also in need of 16 nutrient elements for synthesizing various compounds. Some of them are known as primary nutrients like nitrogen, phosphorus and potassium. Magnesium, calcium and sulphur are required in small quantities and hence they are known as secondary nutrients. Trace elements like copper, manganese, iron, boron, molybdenum and zinc are also necessary for plant growth. Carbon, oxygen, hydrogen and chlorine are provided to plants from water and air.

Every element has to perform some specific function in growth and development of plants. Its deficiency is also characterized by certain symptoms. Various parts of plants are used in pharmaceutical industry for their active constituents, for example, leaves, rhizomes, roots, etc. Some of the nutrients are responsible for growth of particular parts of the plant body and hence, their usefulness and deficiency need to be described systematically.

### **TYPES OF FERTILIZER**

The fertilizers are two types:

1. Biological origin fertilizer.
2. Chemical fertilizer



1. **Biological origin fertilizer** : Soil is generally poor in organic matter and nitrogen. The substances of biological origin used as fertilizer are thus selected if these could provide the elements required. These are two type :

(i) **Manures** : Manure is material, which are mixed with soil. These supply almost all the nutrients required by the crop plants. This results in the increase in crop productivity. Manures are three types:

- **Farmyard manure** : This is a mixture of cattle dung and remaining unused parts of straw and plants stalks fed to cattle.
- **Composited manure** : This consists of a mixture of rotted or decomposed and useless parts of plants and animals.
- **Green manure** : It is a herbaceous crop ploughed under and mixed with the soil while still green to enrich the soil. The plants used as green manure are often quick growing. These add both organic as well as nitrogen to the soil. It is also forms a protective soil cover that checks soil erosion and leaching. Thus, the crop yield increases by 30-50%.

(ii) **Biofertilizer** : It can be defined as biologically active products or bacteria, algae and fungi, which useful in bringing about soil nutrient enrichment. These mostly include nitrogen fixing microorganisms Some of the Biofertilizer are as follows :

- Legume – *Rhizobium symbiosis*.
- Azola – *Anabaena symbiosis*
- Free – living bacteria.
- Loose association of nitrogen fixing bacteria.
- Cyanobacteria (blue green algae)
- Mycorrhiza
  1. *Ectomycorrhizae* : Increase the interface surface between plant root and soil. Mycorrhizae absorb and store nitrogen phosphorous, potassium and calcium.
  2. *Endomycorrhizae*

## 2. Chemical fertilizers :

1. Macronutrients :

- Nitrogen
- Phosphorous

- Potassium
- Calcium
- Magnesium
- Sulphur.

2. Micronutrients :

- Iron
- Manganese
- Zinc
- Boron
- Copper
- Molybdenum

Carbon, oxygen, hydrogen and chlorine are provided from water and air.

**Examples :** Urea, Potash.

## 5. Pests and pest control

Population increases in an arithmetic progression, whereas the agricultural produce is enhanced in geometric progression. What man has reaped must be protected. At the same time, future yields must be improved. But, unfortunately this is not so because, between the time a medicinal crop is harvested and consumed by man, considerable quantity of crude drug is wasted or destroyed by pest. Also, loss in quality occurs when these pests are allowed to grow on produce. The overall losses due to pest thus, assume primary importance in the context of cultivation of medicinal and aromatic plants.

Pest is an undesired animal or plant species and pesticides are chemicals derived from synthetic and natural sources effective in small concentrations against pest.

### Types of pests

The different types of pests infesting medicinal plants are fungi, viruses, weeds, insects and non-insect pests including rodents.

**(i) Fungi and viruses:** Different types of fungi are known to occur on medicinal plants.

*Ascochyta atropae* causes the formation of greyish-white irregular spots which further cause necrosis of leaves. The disease is called leaf necrosis. *Cercospora atropae* causes round to angular brown spots with chestnut coloured margins on both sides of leaves. It is called as leaf-spot. *Phytophthora nicotianae* is a dreadful disease occurring on belladonna

and other plants in which dropping of young leaves and branches, yellowing of older leaves and drying of whole apical portion occurs. This disease is called as phytophthora rootrot. The association of *Fusarium solanii* and *Pythium butleri* causes damping off in young seedlings. The disease occurs on isolated branches of the roots of older plants. *Phytophthora erythrosceptica* causes damping off in young seedlings and wilt in matured plants. It causes black coloration of root. The disease is called phytophthora rot disease.

Several other pathogenic fungi infest the medicinal and aromatic plants and cause diseases, like *Pythium spinosum* (pythium rhizome rot); *Currularia prasadii* (leaf blight); and *Collectotrichum fuscum* (anthracnose) on digitalis; *Septoria digitalis* and *Phyllosticta digitalis* (leaf spot); *Ascochyta kashmiriana* (leaf spot), *Peronospora hyoscyami* (mildew) and *Thielavia basicola* (root rot) occurring on hyoscyamus; *Fusarium oxysporum* (wilt), *Levellula taurica* (powdery mildew), *Cercospora rauwolfiae* (leaf spot) and *Alternaria tenuis* (leaf blight and bud rot) occurring on rauwolfia; *Sclerotium rolfsii* (wilt), *Corticium solanii* (root and root rot) and *Alternaria tenuissima* (leaf spot) occurring on *Datura* species; *Fusarium solanii* (root rot); *Sclerotinia minor* (damping off), and *Ramularia bellunensis* (bud disease) occurring on pyrethrum; *Puccinia dioscorea* (rust) and *Cercospora dioscorea* (leaf spot) on *Dioscorea*. *Phytophthora* species (blight) on vinca; *Erysiphe cinchoracearum* (powdery mildew), *Verticillium albotarum* (wilt), *Puccinia menthae* (rust), *Alternaria* sp. (leaf blight), *Curularia lunatus* (leaf spot) and *Sclerotium rolfsii* (sclerotium rot) on *Mentha* species; *Cercospora canescens* (leaf spot) on *Ocimum* species; *Cylindrocladium scoparium* on *Eucalyptus*; *Cercospora jasminicola* (leaf blight) and *Uromyces hobsonii* (rust) on *Jasminum* species and *Collectotrichum graminicola* on lemongrass.

Many different viruses are also the cause of some diseases occurring on plants. They are mosaic causing necrosis of leaves, petioles and stem on different solanaceous plants. Tobacco mosaic virus, cucumber mosaic virus and tobacco ring spot virus are observed on digitalis and a strain of cucumber mosaic virus is detected on hyoscyamus. The viruses show disease symptoms on rauwolfia, tobacco, datura, vinca and eucalyptus. Other viruses reported on medicinal plants are yellow vein mosaic, graft transmissible virus, distortion mosaic, rugose leaf curl, and *Rugose leaf curl virus*.

**(iii) Insects:** It is found that the total number of insect species in the world is larger than the total number of species of all other forms of life put together. Throughout the world,

about one million species of insects have been reported. These small creatures cause a drastic problem from which the plants must be properly protected.

Various insect pests which attack medicinal plants that can be enumerated here are *Agrotis* species; *Heliothis armigera*, and *Odontotermes obesus*. Flea beetle. *Empoasca pteridis*, *Laphygma exigna* and *Odontotermes obesus* and *Phytomyza atricornis* are the insect pests occurring on *Mentha* species. *Rauwolfia* is attacked by *Diaphania nilgirica*, *Indomia cretaceus*, *Plantia viridicolis* and various beetles. Dill is affected by *Papilio machon* and *Hyadaphis coriandri*. *Belladonna* loses the leaves due to *Gonocephalum* species and *Agrotis flammata*.

The other insect pests known to cause damage are caterpillar *Lepidopterus* larvae, cutworms, termites, weevil, Hessian fly, aphids, phylla, grass-hoppers, locusts, spiders, ticks, mites, etc.

All the insect pests belong to phylum *Arthropoda* of animal kingdom they are further placed under two morphological groups, viz. (1) biting and chewing, (2) piercing and sucking insects.

In most cases, the adult form in the metamorphosis of these insects is damaging in nature, but in certain instances the earlier stage like larvae is also destructive to plants.

**(iii) Weeds :** A weed is undesired plant. Weeds are considered as dreadful pests because losses due to them are estimated to be more than those occurring due to other pests and diseases combined together. If the problem of control of weeds is not handled properly, it leads to loss of nutrients, water, light and space, increase in cost of labour and equipment, low product quality and problems in marketability, enhanced chances for attacks of bacteria, fungi, viruses and insects. But among all these, losses of nutrients and moisture due to weeds are the problems of major concern to all plants yielding foods, medicines, fibres and other economic raw materials.

Some weeds cause allergies e.g. hay fever caused by ragweed, *Medican tea*, yellow dock, *parthenium*, etc. While the others, like corn cockle contains cyanophore glycoside and the seeds of this plant may cause fatal effects. Dermatitis is caused by poison ivy, western poison oak, varnish tree, poison sumac, etc. Some plants growing as weeds may be poisonous like *Datura* and *Menispermus* species, etc.

**(iv) Non-insect pests:** They are grouped into two categories :

1. Vertebrates like rats, monkeys, birds, rabbits and hares, squirrels, antelopes, deer, pigs, etc.
2. Invertebrates like nematodes, crabs, snails, mites, and symphylids.

The rodents have sharp and gnawing incisor teeth with which they cause considerable spoilage to stored crude drugs and also the faecal matter of such animals causes serious contamination of crude drugs.

## **METHODS OF PEST CONTROL**

Different techniques followed for an integral approach to pest control are briefly summarized as under.

**(1) MECHANICAL METHOD :** It employs manual labour alongwith different devices for collection and destruction of pest.

The simple techniques used are hand-picking, pruning, burning and trapping of pests. A proper approach is made for collection and destruction of eggs, larvae, pupae and adults of insects.

The better way for protection from rodents like rats is construction of concrete warehouses. The warehouses should have metal reinforcement corners on window frames. The rat and mouse traps are also used.

The method adopted for trapping flying insects is flavoured attractants placed in funnel shaped container, which are formulated with rose oil, anise oil, etc. mixed with saw dust. The insects can easily get an entry in the trap. but find it very hard to come out.

**(2) AGRICULTURAL METHOD:** It covers advanced plant breeding techniques capable of inducing genetic manipulations resulting in production of pest-resistant species. It has achieved much success in producing hybrid varieties, which are resistant to fungal and bacterial attack, as compared to limited success with insects. The systemic insecticides have been developed which are absorbed through the roots and reach to leaves by which all the foliage portion becomes distasteful for insects.

Another aspect in agricultural control is ploughing which should be sufficiently deep so as to eradicate weeds, as well as, early stages of insects. If a plant is found out to be favoured by insects as major source of food, the land under cultivation of such plant should

be subjected to crop rotation. Another method for checking supply of food to insects is by changing the environment which in many cases, may lead to obstruction in their life cycle. Proper drainage serves this purpose to a great extent.

**(3) BIOLOGICAL CONTROL :** This method is practiced by combating the pests, mostly the insects, with other living organisms. The latter is frequently the parasite form. If this method is properly designed, it may emerge as an effective, safe and economic method of pest control.

The chemical substances produced and released by some female insects are capable of eliciting a sexual response from the opposite sex, which could be properly exploited for biological control of pests. Such substances are called as sex pheromones, e.g. 7,8 – epoxy – 2 methyloctadecane from gypsy-moth.

**(4) CHEMICAL CONTROL :** The control of pests is brought about with the use of chemical pesticides, which include insecticides, fungicides, herbicides and rodenticides.

Because of toxic effects of all such chemicals used as pesticides, their use is regulated by the Insecticides Act in India; Federal Insecticide, Fungicide and Rodenticide Act in United States and the Agriculture (Poisonous substances) Regulations in U.K.

The chemical pesticides are further classified as rodenticides, insecticides, acaricides, fungicides, herbicides, predacides, ovicides, bactericides, arboricides, etc.

**(i) Rodenticides :** Warfarin, strychnine, arsenic trioxide, thallium sulphate, red squill, etc.

**(ii) Insecticides (Table 3.3) :** D.D.T., gammaxine, methoxychlor, parathion, malathion, sodium arsenate, pyrethroids, rotenoids, carbamates, etc.

**(iii) Acaricides (miticides) :** Tetradifon, chlorobenzolate.

**(iv) Fungicides :** Bordeaux mixture, chlorophenols, antibiotics, quaternary ammonium compounds, etc.

**(v) Herbicides :** 2, 4- dichlorophenoxy acetic acid, calcium arsenate, sulphuric acid.

The insecticides are applied to vegetative parts for protective or eradicator activity in the form of spray, aerosol, solution, suspensions and fine dust. The stomach poisons are protective in action, while contact poisons act as eradicants. The fumigation keeps away the

insect from the area under the influence of insect repellents. An ideal insecticide is required to satisfy following parameters.

- (i) It should be non-toxic and non-injurious to medicinal plants and human beings.
- (ii) It should be selective in action and highly toxic to insects in small concentrations.
- (iii) The pesticides should be stable under ordinary conditions of storage, non-inflammable, non-corrosive and free from obnoxious odours.

**Types of Insecticides used in Cultivation and Post-Harvest Technology Chlorinated hydrocarbons Carbamates**

Aldrin Carbaryl [Carbaril (Sevin)]

Benzene hexachloride (BHC, Gammaxine)

DDT

Dieldrin

**Inorganic Compounds**

Endrin

Calcium arsenate

Heptachlor

Lead arsenate

Methoxychlor

Paris green

Chlordane

Sodium fluosilicate

DDE

Zinc phosphide

**Organo-phosphorus Compounds**

**Fumigants**

Carbophenothion

Aluminium phosphide

Demeton

Hydrogen cyanide

Dichlorvos

Ethylene dichloride

Diaznon

Methyl bromide

Dimethoate(Rogor)

Phosphine

Malathion

Methyl parathion

Phorate

**Natural products**

Phosphamidon(Dimecron)

Tobacco

Schraden

Pyrethrum

Trichlorophon	Derris
Tetra ethyl pyro phosphate (TEPP)	Neem
Thiometom	Sabadilla

The mode of action of different insecticides is given below in Table-

### Mode of Action of some Pesticides

Chemical Compound	Biological Effect
<b>Effect on Animals</b>	
1. Organophosphorus compounds and carbamates	Inhibition of acetyl cholinesterase
2. Chlorinated hydrocarbons and Pyrethroids	Neurotoxication
3. Nicotinoids	Inhibition of neuromuscular junction
<b>Effect on Plants</b>	
4. Carbamates, substituted ureas, triazines	Inhibition of photosynthesis
5. Carbamates	Inhibition of oxidative phosphorylation
6. 2,4-D; 2,4,5-T	Hormone analogs
7. Metals, sulphur	Unknown causes
8. 3- amino-1, 2, 4-triazole	Inhibition of chlorophyll synthesis
9. Chlorinated aliphatic hydrocarbons	Inhibition of pantothenate synthesis

### PLANT GROWTH REGULATORS (PLANT HORMONES)

Plant growth regulators are the organic compounds, other than nutrients which affect the morphological structure and/or physiological processes of plants in low concentrations. Phytohormones or plant hormones are naturally occurring growth regulators which in low concentrations control physiological processes in plants. More commonly, the term plant growth regulators is used, because it includes both the native (endogenous) and the synthetic (exogenous) substances; which modify the plant growth.



As the native plant growth regulators, five major kinds of substances are reported, viz. auxins, gibberellins, cytokinins, abscisic acid and ethylene. All of them, except ethylene and abscisic acid, are multiple forms of endogenous plant growth regulators. In general, the plant growth regulators or substances serve role in regulating cell enlargement, cell division, cell differentiation, organogenesis, senescence and dormancy. They are employed in seed treatment to achieve earlier growth and root development, quality improvement like protein level and amino acid balance, etc. Plant growth regulators have given a real boost to plant tissue culture techniques by which now it is possible to culture almost any part of the plant in-vitro.

### **Auxins**

Auxin is a general term used to indicate substances that promote elongation of coleoptile tissues. Indole acetic acid (IAA) is an auxin that occurs naturally in plants. They are either natural auxins which are produced by plants themselves or synthetic auxins, which have the same action as natural auxins. IAA is the principle auxin and other natural auxins are indole-3-acetonitrile(IAN), 4-chloroindole 3-acetic acid and phenyl acetic acid. The synthetic auxins are indole-3-butyric acid(IBA), 2-naphthyloxyacetic acid(NOA),  $\alpha$ -naphthyl acetic acid (NAA), 1-naphthyl acetamide (NAD)2, 4-dichlorophenoxy acetic acid (2,4-D), 2, 4, 5 trichlorophenoxy acetic acid and 5-carboxymethyl-N, N-dimethyl dithiocarbamate.

Auxins are involved in different growth processes in plants like internodes elongation, leaf growth, initiation of vascular tissues, cambial activity, fruit setting in absence of pollination, fruit growth, apical dominance, inhibition of root growth, influencing physical and chemical properties in leaf abscission and inhibition of lateral buds.

The proposed mechanism of action of IAA is its interaction with one or more components of biochemical systems involved in the synthesis of proteins. The other hypothesis suggested is the role of IAA to alter the osmotically active contents of cell vacuole during cell expansion or cell wall extension.

IBA and NAA in combination are used in rooting of cuttings. NOA is used as a fruit setting spray 2, 4-D and 2, 4, 5-T are used both as plant growth regulators and in higher concentrations, as selective weed killers, especially for dicot plants. IBA has shown promising results to induce rooting in cuttings for Cinchona, Pinus, Papaya and Coffee. The addition of different auxins like. IAA, NAA and 2, 4-D in tissue cultures of ergot has led to

increase in indole alkaloids. Treatment of derivatives of NAA given to seedlings and young plants of *Mentha piperita* has shown about 40 per cent increase in volatile oil content.

## **GIBBERELLINS**

They are a class of endogenous plant growth regulators and at present over 50 gibberellins are known. About 40 of them occur in green plants, while others are present in some fungi. They are present in different organs and tissues like roots, shoots, buds, leaves, floral apices, root nodules, fruits and callus tissues. The commercial formulations of gibberellins are used currently for promoting vegetative and fruit growth, breaking dormancy, flower initiation and induction of parthenocarpy. Kurosawa, a Japanese physiologist, is credited for initiating the discovery of gibberellin from fungus *Gibberella fujikuroi* (previously known as *Fusarium heterospermum*) grown on rice. According to Paleg, gibberellins are compounds having gibbane skeleton and biological activity in stimulating cell division or cell elongation, or both.

The thorough research on gibberellins carried out in Japan, United States of America and Great Britain has shown that gibberellin A (isolated in 1938) is actually a mixture of at least 6 gibberellins referred to as GA<sub>1</sub>, GA<sub>2</sub>, GA<sub>3</sub>, GA<sub>4</sub>, GA<sub>7</sub>, GA<sub>9</sub>. GA<sub>3</sub> is termed as gibberellic acid.

All of them are the derivatives of gibbane ring skeleton. In addition to free gibberellins, they are also present in conjugated forms. GA has not yet been synthesized, but can be produced by large scale fermentation on commercial scale. The angiosperms, gymnosperms, ferns, algae, fungi and bacteria contain several forms of gibberellins, but no single plant contains all of them together. Many activities attributed to gibberellins are promotion of rapid expansion of plant cells, stimulation of seed germination, breaking dormancy of overwintering plants, induction of flowering under non-inductive conditions, marked increase in stem elongation, increase in the size of leaves and induction of parthenocarpic fruit leading to seedless fruit sets.

The effect of gibberellins in cell division is an increase in cell size similar to the effect of auxins. It is observed that gibberellins are more effective in intact plants, While major auxin effects are on excised organs.

The applications of gibberellins are extended to various medicinal plants. The use of gibberellins in lower dose has shown increased yield of digitalis glycosides per shoot. The hormone tried with leaf and root culture of digitalis, showed higher production of digoxin. In

case of Tinnevelly senna, GA shows a little positive effect on dry weight of shoot, but reduction in sennoside content of leaves. It is observed that GA treatment can cause an increase in height of castor plant upto five times, but does not show any change in fixed oil content. The treatment significantly causes reduction in alkaloid content of vinca, datura, hyoscyamus, etc.

The mechanism of action of gibberellic acid appears mainly to induce activity of gluconeogenic enzymes during early stages of seed germination and this specificity ensures a rapid conversion of lipid to sucrose, which is further used in supporting growth and development of the embryonic axis to a competent root and shoot system. It is also found that gibberellins induce the synthesis of  $\alpha$ -amylase and other hydrolytic enzymes during germination of monocot seeds. They are also involved in mobilizing seed storage reserves during germination and seedling emergence.

## **CYTOKININS**

These are either natural (zeatin) or synthetic (kinetin) compounds with significant growth regulating activity. Zeatin has effect on cell division and leaf senescence and synthetic cytokinins are useful in promoting lateral bud development and inhibition of senescence.

Cytokinins influence a broad spectrum of physiological processes in plants like promotion of cell division. The other activities exerted are participation in orderly development of embryos during seed development, influencing the expansion of cells in leaf discs and cotyledons, delaying breakdown of chlorophyll and degradation of proteins in ageing leaves.

Miller isolated the crystalline substance from autoclaved herring sperm DNA capable of inducing cell division in tobacco cultures and named it as kinetin, which was found to be 6-furfuryl adenine. Further, some other adenine derivatives were also found having similar biological activity and were called 'kinins' collectively known as cytokinins. These substances are found in young and actively dividing tissues like embryos, seedlings and apical meristems.

The naturally occurring cytokinins are zeatin, N<sup>6</sup> dimethyl amino purine, and N<sup>6</sup>- $\Lambda^2$ -isopentenyl aminopurine. The synthetic cytokinins are kinetin, adenine, 6-benzyl adenine benzimidazole and N. N<sup>1</sup>-diphenyl urea.

Cytokinins are reported to increase marginally sennoside content in Tinnevelly senna leaves and also enhance the dry weight of shoots. In opium, they cause formation of elongated capsule and reduce alkaloid content. In *Duboisia* hybrids, the cytokinin activity present in extract of a seaweed, shows marked increase in both leaf content and also hyosine content.

Kinetins are reported to play the role in nucleic acid metabolism and protein synthesis. In plant metabolism, it is proposed that some t-RNA contain cytokinin like activity. They have an action on some enzymes responsible for formation of certain amino acids.

### **ETHYLENE**

It is a simple organic molecule present in the form of volatile gas and shows profound physiological effects. It is present in ripening fruits, flowers, stems, roots, tubers and seeds, It is present in very less quantity in plant, normally about 0.1 ppm. Possibly, its quantity is increased in local areas during the time of growth and development.

Ethylene is produced by incomplete burning of carbon rich substances like natural gas, coal and petroleum. Denny (1924) showed the yellowing of lemons due to stove gas. Also, the plant damage was noticed after the introduction of illuminating gas. Gane (1935) found that a gas evolved from ripe apples can also effect the ripening of green apples showed that it was ethylene gas which has a role in ripening of other fruits.

Ethylene shows a broad array of growth responses in plants, which include fruit ripening, leaf abscission, stem swelling, leaf bending, flower petal discoloration, and inhibition of stem and root growth. It is commercially used for promotion of flowering and fruit ripening, induction of fruit abscission, breaking dormancy and stimulation of latex flow in rubber trees.

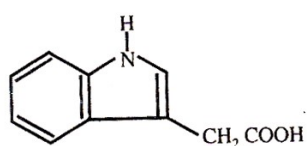
### **ABSCISIC ACID (ABA)**

The physiological activities in plants like retaining or shedding of different organs such as leaves, stems, flowers and fruits have led to finding of natural growth inhibitor. A diffusible abscission-accelerating substance was found by Osborne (1955) in senescent leaves. Carns et al. isolated several abscission accelerating substances from cotton plants and named them as abscisin I and abscisin II.

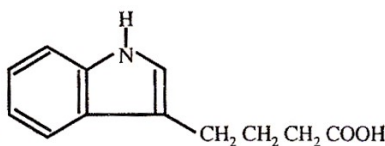
In an inhibitory way, ABA interacts with other plant growth substances. It inhibits the GA-induced synthesis of  $\alpha$ -amylase and other hydrolytic enzymes. During maturation, ABA accumulates in many seeds and helps in seed dormancy. ABA concentrations are found to be enhanced in stress conditions, like mineral deficiency, injury, drought and flooding. ABA serves an important role as potential antitranspirant by closing the stomata, when applied to leaves.

A number of other synthetic growth inhibitors and retardants reported are maleic hydrazide, daminozide, glyphosine, chromequat chloride, S, S, S-tributyl phosphorotrithioate, ancymidol, chlorophonium chloride, piproctanyl bromide, etc. However, commercial use of these compounds is yet to be reported. A group of synthetic substances called morphactins is potent inhibitor of auxin transport causing tropic responses, reduction of apical dominance and promoting lateral growth. Morphactins include chloroflurecol methyl, flurecol-butyl and TIBA (2, 3, 5-tri-iodobenzoic acid).

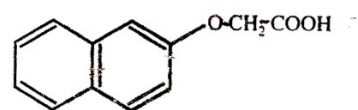
### EXAMPLES OF CHEMICAL STRUCTURE OF SOME PLANT GROWTH REGULATORS



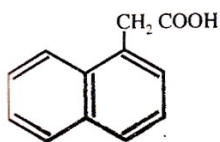
IAA



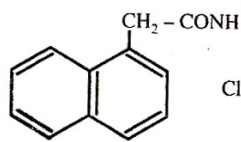
IBA



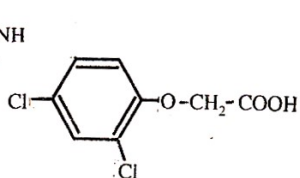
NOA



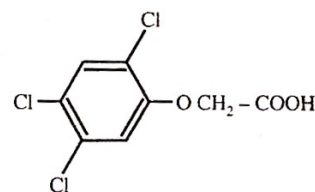
NAA



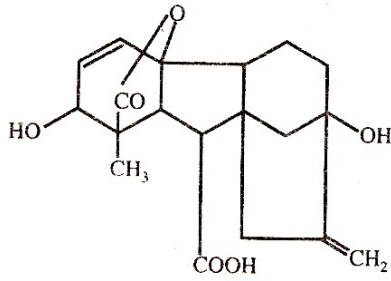
NAD



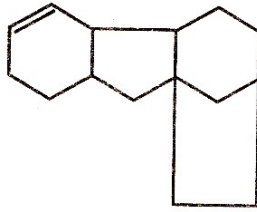
2,4-D



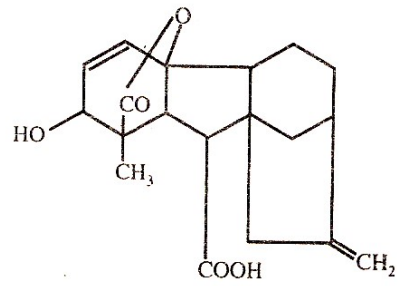
2,4,5 T



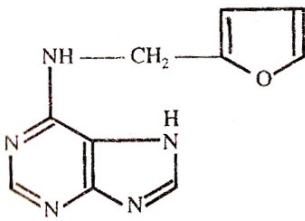
(Gibberellic acid)



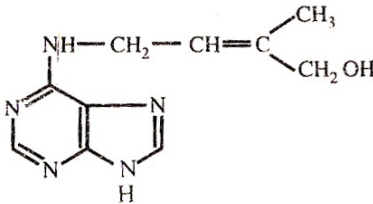
Gibbane skeleton



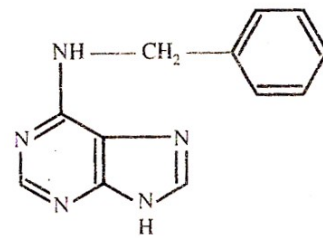
GA<sub>7</sub>



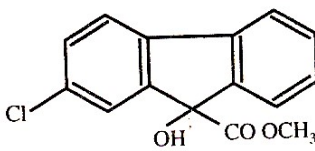
Kinetin



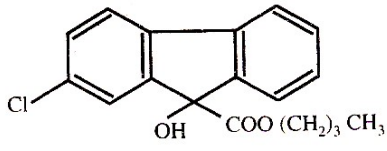
Zeatin



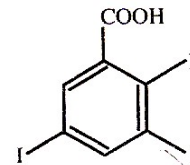
(6 - Benzyl adenine)  
(6 - Benzyl aminopurine)



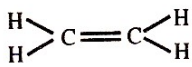
Chloroflurecol methyl



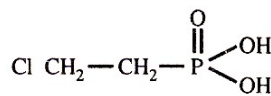
Flurecol-butyl



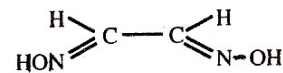
(2, 3, 5-tri-iodobenzoic acid) TIBA



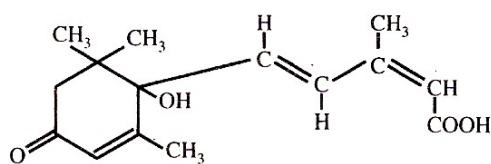
Ethylene



Ethephon



Glyoxime



**Abscisic acid (Abscisin II)**

## COLLECTION AND PROCESSING OF CRUDE DRUGS

After collection of the crude drugs, they are required to be processed prior to marketing. The reasons for preparation of drugs are to stabilize them in transport and storage and to ensure the absence of foreign organic matter and substitutes. Market preparation of crude drugs also takes care of pharmaceuticals elegance. While preparing drug for commerce, several methods are adopted to meet the standard pharmacopoeial requirements. Generally, these methods include proper methods of collection, harvesting, drying and garbling. Sometimes, coating and bleaching are also necessary for converting the drug into suitable form for the market. While doing so, it could be observed that neither the action of the drug is lowered down nor it is changed, due to additives used in the process.

## COLLECTION

Irrespective of the type of crude drug and area of collection, there cannot be two opinions that the drugs are collected suitably when they contain maximum concentration of active constituents. The advantage of existing environmental conditions is also taken into consideration while collecting the crude drugs. The drugs which constitute leaf and flowering tops of plants are collected just before they reach their flowering stage(maturity); e.g., senna, digitalis, vinca, belladonna, etc, while the leaves of aloe are collected when they are sufficiently thick. Flowers need to be collected just before pollination or many a times, before their full expansion. e.g. saffron, clove buds, chamomile, arnica, etc, They are collected in dry weather and preferably during morning hours. Barks are generally collected in spring or early summer when cambium is active, as it is easy to detach them from the stem. Sometimes, they are collected in autumn (wild cherry) or in rainy season (cinnamon). Three different methods for collecting barks are (i) felling, (ii) uprooting and (iii) coppicing. In felling method, the tree is cut at base and bark is peeled out. In uprooting technique, the

roots are dug out and bark is stripped off from roots and branches. In coppicing method, the plant is allowed to grow for a definite period and then it is cut off at specific distance from soil. The stumps which remain in ground are allowed to send shoots, which develop further independently yielding aerial parts. These new parts are cut off and bark is collected from shoots. As compared to other methods of collection of bark, this technique is more economical and less time-consuming. It is, therefore, the method of choice for collecting barks commercially. Cascara and Cinnamon are collected by this method.

The fruits are collected depending upon the part of the fruits used. They are collected either ripe or half ripe, but fully grown. For example, cardamom fruits are collected just before their dehiscence; bael and tamarind, after their, full maturity, while caraway, fennel and coriander are collected, when they are fully ripe. The roots are collected in spring, before the vegetative process stops. Usually, the roots are sliced transversely or longitudinally to facilitate drying. Rhizomes are collected, when they store ample of reserve food material and also contain maximum content of chemical constituents.

The unorganized drugs such as resins, gums, latices are collected, as soon as, they ooze out of the plant. Acacia gum is collected 2-3 weeks after making incisions on the bark of the tree and when it is sufficiently hard. Opium and papaya latices are collected after coagulation of latex. Turpentine oleoresin and balsam of peru are collected when the plant is about 8-10 years old.

## **HARVESTING**

Harvesting is an important operation in cultivation technology, as it reflects upon economic aspects of the crude drugs. An important point which needs attention over here is the type of drug to be harvested and the pharmacopoeial standards which it needs to achieve. Harvesting can be done efficiently in every respect by the skilled workers. Selectivity is of advantage in that the drugs other than genuine, but similar in appearance can be rejected at the site of collection. It is, however, a laborious job and may not be economical. In certain cases, it cannot be replaced by any mechanical means, e.g. digitalis, tea, vinca and senna leaves. The underground drugs like roots, rhizomes, tubers, etc. are harvested by mechanical devices, such as diggers or lifters. The tubers or roots are thoroughly washed in water to get rid of earthy-matter. Drugs which constitute all aerial parts are harvested by binders for economic reasons. Many a times, flowers, seeds and small fruits are harvested by a special device known as seed stripper. The technique of beating plant with bamboos is used in case



of cloves. The cochineal insects are collected from branches of cacti by brushing. The seaweeds producing agar are harvested by long handled forks. Peppermint and spearmint are harvested by normal method with mowers, whereas fennel, coriander and caraway plants are uprooted and dried. After drying, either they are thrashed or beaten and the fruits are separated by winnowing. Sometimes, reaping machines are also used for their harvesting.

## **DRYING**

Before marking a crude drug, it is necessary to process it properly, so as to preserve it for a longer time and also to acquire better pharmaceutical elegance. This processing includes several operation or treatments, depending upon the source of the crude drug (animal or plant) and its chemical nature. Drying consists of removal of sufficient moisture content of crude drug, so as to improve its quality and make it resistant to the growth of microorganisms. Drying inhibits partially enzymatic reactions. Drying also facilitates pulverizing or grinding of a crude drug. In certain drugs, some special methods are required to be followed to attain specific standards. e.g. fermentation in case of *Cinnamomum zeylanicum* bark and gentian roots. The slicing and cutting into smaller pieces is done to enhance drying, as in case of glycyrrhiza, squill and calumba. The flowers are dried in shade so as to retain their colour and volatile oil content. Depending upon the type of chemical constituents, a method of drying can be used for a crude drug. Drying can be of two types-(1) **natural (sun drying); (2) artificial.**

### **1. Natural Drying (Sun-drying)**

In case of natural drying, it may be either sun-drying or in the shed. If the natural colour of the drug (*digitalis*, clove, *senna*) and the volatile principles of the drug (*peppermint*) are to be retained, drying in shed is preferred. If the contents of the drugs are quite stable to the temperature and sunlight, the drugs can be dried directly in sunshine(*gum acacia*, seeds and fruits).

### **2. Artificial Drying**

Drying by artificial means includes drying the drugs in (a) an oven; i.e. tray-dryers; (b) vacuum dryers, and (c) spray dryers.

**(a) Tray dryers :** The drugs which do not contain volatile oils and are quite stable to heat or which need deactivation of enzymes are dried in tray dryers. In this process, hot air of the desired temperature is circulated through the dryers and this facilitates the removal of water content of the drugs (belladonna roots, cinchona bark, tea and raspberry leaves and gums are dried by this method).

**(b) Vacuum dryers :** The drugs which are sensitive to higher temperature are dried by this process e.g. tannic acid and digitalis leaves.

**(b) Spray dryers :** Few drugs which are highly sensitive to atmospheric conditions and also to temperature of vacuum-drying are dried by spray-drying method. The technique is followed for quick drying of economically important plant or animal constituents, rather than the crude drugs. Examples of spray drying are papaya latex, pectin, tannins, etc.

## **PACKING**

The morphological and chemical nature of drug, its ultimate use and effects of climatic conditions during transportation and storage should be taken into consideration while packing the drugs. Aloe is packed in goat skin. Colophony and balsam of tolu are packed in kerosene tins, while asafoetida is stored in well closed containers to prevent loss of volatile oil. Cod liver oil, being sensitive to sunlight, should be stored in such containers, which will not have effect of sunlight, whereas, the leaf drugs like senna, vinca and others are pressed and baled. The drugs which are very sensitive to moisture and also costly at the same time need special attention. e.g. digitalis, ergot and squill. Squill becomes flexible, ergot becomes susceptible to the microbial growth, while digitalis loses its potency due to decomposition of glycosides, if brought in contact with excess of moisture during storage. Hence, the chemicals which absorb excessive moisture (desiccating agents) from the drug are incorporated in the containers. Colophony needs to be packed in big masses to control auto-oxidation. Cinnamon bark, which is available in the form of quills, is packed one inside the other quill, so as to facilitate transport and to prevent volatilization of oil from the drug. The crude drugs like roots seeds and others do not need special attention and are packed in

gunny bags, while in some cases bags are coated with polythene internally. The weight of certain drugs in lots is also kept constant. e.g. Indian opium.

## **STORAGE OF CRUDE DRUGS**

Preservation of crude drugs needs sound knowledge of their physical and chemical properties. A good quality of the drugs can be maintained, if they are preserved properly. The following illustrations shall help in understanding the factors to be considered while preserving the drug. All the drugs should be preserved in well closed and, possibly, in the filled containers. They should be stored in the premises which are water-proof, fire-proof and rodent-proof. A number of drugs absorb moisture during their storage and become susceptible to the microbial growth. Some drugs absorb moisture to the extent of 25% of their weight. The moisture, not only increases the bulk of the drug, but also causes impairment in the quality of crude drug. The excessive moisture facilitates enzymatic reactions resulting in decomposition of active constituents. e.g. digitalis leaves and wild cherry bark. Gentian and ergot receive mould infestation due to excessive moisture. Radiation due to direct sun- light also causes destruction of active chemical constituents. e.g. ergot, cod liver oil and digitalis. Form or shape of the drug also plays very important role in preserving the crude drugs. Colophony in the entire form (big masses) is preserved nicely, but if stored in powdered form, it gets oxidized or loses its solubility in petroleum ether. Squill, when stored in powdered form becomes hygroscopic and forms rubbery mass on prolonged exposure to air. The fixed oil in the powdered ergot becomes rancid on storage. In order to maintain a good quality of powdered ergot; it is required that the drug should be defatted with lipid solvent prior to storage. Lard, the purified internal fat of the abdomen of the hog, is to be preserved against rancidity by adding siam benzoin. Atmospheric oxygen is also destructive to several drugs and hence, they are filled completely in well closed containers, or the air in the container is replaced by an inert gas like nitrogen, e.g. shark liver oil, papain, etc.

Apart from protection against adverse physical and chemical changes, the preservation against insect or mould attacks is also important. Different types of insects, nematode, worms, moulds and mites infest the crude drugs during storage. Some of the more important pests found in drugs are Coleoptera (*Stegobium paniceum* and *Calandrum granarium*), Lepidoptera (*Ephestia kuehniella* and *Tinea pellionella*), and Archnida or mites

(*Tyroglyphus farinae* and *Glyphogus domesticus*). They can be prevented by drying the drug thoroughly before storage and also by giving treatment of fumigants. The common fumigants used for storage of crude drugs are methyl bromide, carbon disulphide and hydrocyanic acid. At times, drugs are given special treatment, such as liming of the ginger and coating of nutmeg. Temperature is also very important factor in preservation of the drugs, as it accelerates several chemical reactions leading to decomposition of the quality. Hence, most of the drugs need to be preserved at a very low temperature. The costly phytopharmaceuticals are required to be preserved at refrigerated temperature in well closed containers. Small quantities of crude drugs could be readily stored in air-tight, moisture proof and light proof containers such as tin, cans, covered metal tins, or amber glass containers. Wooden boxes and paper bags should not be used for storage of crude drugs.

### **POLYPLOIDY**

Plants whose cells contain two sets of chromosomes, derived at fertilization from the union of one set from the pollen and one set from the egg cells, are described as diploids and denoted by “2n”. The term polyploidy is applied to plants with more than two sets of chromosomes in the cells; when four sets are present the plants are described as tetraploids and denoted by “4n”. Tetraploidy is induced by treatment with colchicine. Which inhibits spindle formation during cell division, so that the divided chromosomes are unable to separate and pass to the daughter cells. The two sets of chromosomes remain in one cells and this develops to give tetraploids plant. Treatment with colchicines may be applied in various ways but all depend on the effects produced in the meristem. The seeds may be soaked in a dilute solution of colchicines, or the seedlings, the soil around the seedling or the young shoot treated with colchicine solution.

Fertile seed and robust, healthy tetraploid plants were obtained, the tetraploid condition being indicated by the increased size of the pollen grains and stomata; chromosomes counts in root-tip preparation confirm the tetraploid condition. The average increase in alkaloids content compared with diploid plants of *Datura stromonium* and *Datura tatula* was 68%, with a maximum increase of 211.6%. This diploid of *Acorus calamus* is 2.1% volatile oil content but they are converted into tetraploid, they produce 6.8% of volatile oil contents.

### **MUTATION**

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**Definition:** Sudden heritable change in the structure of a gene or chromosome or change in the chromosome number.

**TYPE OF MUTATIONS:**

1. Spontaneous and induced mutations.
2. Recessive and dominant mutation
3. Somatic and germinal mutations
4. Forward, back and suppressor mutation
5. Chromosomal, genomic and point mutations.

Mutations can be artificially produced by certain agents called mutagens or mutagenic agent. They are two types :

**(a) Physical mutagens :**

- Ionizing radiations : X-rays, gamma radiation and cosmic rays.
- Non-ionizing radiation : U.V. radiation.

**(b) Chemical mutagens :**

- Alkylating and hydroxylating agents\_: Nitrogen and sulphur mustard; methyl and ethylsulphonate, ethylethane sulphonates.
- Nitrous acid
- Acridines : Acridines and proflavins.

Ionizing radiation cause breaks in the chromosome. These cells then show abnormal cell divisions. If these include gametes, they may be abnormal and even die prematurely.

Non-ionizing radiation like Ultra Violet rays are easily absorbed by purine and pyrimidines. The changed bases are known as photoproducts. U.V. rays cause two changes in pyrimidine to produce pyrimidine hydrate and pyrimidine dimers. Thymine dimer is a major mutagenic effect of U.V. rays that disturbs DNA double helix and thus DNA replication.

**Example :** Penicillin, as an antibiotic was first obtained from *Penicillium*. However, the yield was very poor and the preparation was commercially expensive. Since then mutants with higher yield of penicillin have been selected and produced. *Penicillium chrysogenum* used in the production of penicillin yielded about 100 units of penicillin per ml of culture medium. By single – spore isolation, strains were obtained which yielded upto to 250 units per ml of medium. X-ray treatment of this strain gave mutants which produce 500 units per ml and ultraviolet mutants of latter gave strain which produced about 1000 unit per ml. Similarly improvements have been obtained with other antibiotic – producing organism.

Mutant strains of *Capsicum annum* with increasing yields (20-60%) of capsaicin have been isolated from M<sub>3</sub> and M<sub>4</sub> generations originating from seed treated with sodium azide and ethyl methane sulphonate.

### HYBRIDIZATION

It is mating or crossing of two genetically dissimilar plants having desired genes or genotypes and bringing them together into one individual called hybrid. The process through which hybrids are produced is called hybridization. Hybridization particularly between homozygous strains, which have been inbred for a number of generations, introduces a degree of heterozygosity with resultant hybrid vigour often manifest in the dimensions and other characteristic of the plants. A hybrid is a organism which results from crossing of two species or varieties differing at least in one set of characters.

The following steps are involved in hybridization of plant :

1. **Choice of parents** : *The two parents to be selected, at least one should be as well adopted and proven variety in the area. The other variety should have the characters that are absent in the first chosen variety.*
2. **Emasculation** : Removal of stamens or anthers or killing the pollen grains of a flower without affecting the female reproductive organs is known as emasculation. Emasculation is essential in bisexual flowers.
3. **Bagging** : Immediately after emasculation, the flowers or inflorescences are enclosed in bags of suitable sizes to prevent random cross-pollination.
4. **Pollination** : In pollination, mature, fertile and viable pollens are placed on a receptive stigma. The procedure consists of collecting pollens from freshly dehisced anthers and dusting them on the stigmas of emasculated flowers.
5. **Raising F<sub>1</sub> plants** : Pollination is naturally followed by fertilization. It results in the formation of seeds. Mature seeds of F<sub>1</sub> generation are harvested dried and stored. These seeds are grown to produce F<sub>1</sub> hybrid.

Hybrids of cinchona yield more amount of quinine. A hybrid developed by crossing *Cinchona succirubra* with *Cinchona ledgeriana* yields a bark, which contains 11.3% of alkaloids. The parent species produced 3.4% and 5.1% of alkaloids, respectively.

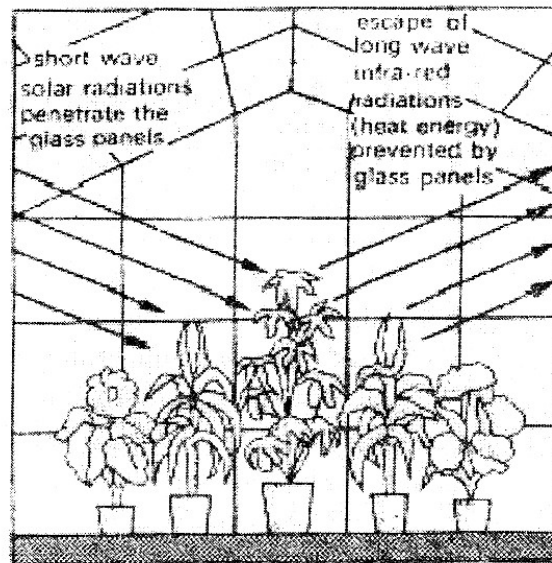
Pyrethrum hybrids have been used for pyrethrin production; these hybrids are produced either by crossing two clones assumed to be self-sterile or planting a number of

desirable clones together and bulking the seed. The hybridization of plant to increase the Pyrethrin contents.

### GREEN HOUSE EFFECT

Normal conditions sun rays reach the earth and heat is radiated back into space. However, when carbon dioxide concentration increases in the atmosphere, it forms a thick cover and prevents the from being re-radiated. Consequently, the atmosphere gets heated and the temperature increases. This is called green house effect. In recent past, amount of carbon dioxide has increased from 290 ppm o 330 ppm due to cutting of forests and excessive burning of fossil fuels. The rate at which the amount of carbon dioxide in the atmosphere is increasing, it is expected to cause rise in global temperature.

The global warming by two or three degrees would cause polar ice caps to melt, floods in coastal areas, change in hydrologic cycle and islands would get submerged. The following gases produce green house effect like carbon dioxide, sulphur dioxide, oxides of nitrogen, chlorofluorocarbons, etc.



Condition in green house; Glass panel prevent escape heat

Condition on earth; Thick CO<sub>2</sub> mantle like glass panel

