

PREFACE

In a bid to standardize higher education in the country, the University Grants Commission (UGC) has introduced Choice Based Credit System (CBCS) based on five types of courses viz. *core, generic, discipline specific elective, ability and skill enhancement* for graduate students of all programmes at Honours level. This brings in the semester pattern which finds efficacy in sync with credit system, credit transfer, comprehensive continuous assessments and a graded pattern of evaluation. The objective is to offer learners ample flexibility to choose from a wide gamut of courses, as also to provide them lateral mobility between various educational institutions in the country where they can carry their acquired credits. I am happy to note that the university has been recently accredited by National Assessment and Accreditation Council of India (NAAC) with grade “A”.

UGC (Open and Distance Learning Programmes and Online Programmes) Regulations, 2020 have mandated compliance with CBCS for U.G. programmes for all the HEIs in this mode. Welcoming this paradigm shift in higher education, Netaji Subhas Open University (NSOU) has resolved to adopt CBCS from the academic session 2021-22 at the Under Graduate Degree Programme level. The present syllabus, framed in the spirit of syllabi recommended by UGC, lays due stress on all aspects envisaged in the curricular framework of the apex body on higher education. It will be imparted to learners over the six semesters of the Programme.

Self Learning Material (SLMs) are the mainstay of Student Support Services (SSS) of an Open University. From a logistic point of view, NSOU has embarked upon CBCS presently with SLMs in English / Bengali. Eventually, the English version SLMs will be translated into Bengali too, for the benefit of learners. As always, all of our teaching faculties contributed in this process. In addition to this we have also requisitioned the services of best academics in each domain in preparation of the new SLMs. I am sure they will be of commendable academic support. We look forward to proactive feedback from all stakeholders who will participate in the teaching-learning based on these study materials. It has been a very challenging task well executed, and I congratulate all concerned in the preparation of these SLMs.

I wish the venture a grand success.

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Vice-Chancellor

Netaji Subhas Open University
Under Graduate Degree Programme
Choice Based Credit System (CBCS)
Subject : UG Botany (HBT)

Course : Economic Botany and Plant Biotechnology
Course Code: GE-BT-41

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**Netaji Subhas
Open University**

**UG : Botany
(HBT)**

**Course : Economic Botany and Plant Biotechnology
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MODULE 1
ECONOMIC BOTANY

Unit 1 □ Origin of Cultivated Plants

Structure

- 1.0 Objectives**
- 1.1 Introduction**
- 1.2 Centres of origin and domestication of crop plants**
- 1.3 Limitations of Vavilov's views**
- 1.4 Summary**
- 1.5 Questions & Answers**

1.0 Objectives

This unit will help to know the basic knowledge about centres of origin and domestication of crop plants.

1.1 Introduction

The origin of crop plants is now basic to economic botany in order to locate wild relatives, related species, and new genes (especially dominant genes, sources of disease resistance). Knowledge of the origins of crop plants is vitally important in order to avoid genetic erosion, the loss of germplasm due to the loss of ecotypes and landraces, loss of habitat (such as rainforests), and increased urbanization. Germplasm preservation is accomplished through gene banks (largely seed collections but now frozen stem sections) and preservation of natural habitats (especially in centers of origin).

1.2 Centres of origin and domestication of crop plants

The Russian scientist Nikolai Ivanovich Vavilov and his colleagues visited several countries and collected a large number of crop plants and their wild relatives. They used this collection in Russian breeding programme of developing improved varieties. His deductions were based on evidences from morphology, anatomy, cytology, genetics, plant geography and distribution.

He considered that great centres of origin were always located in lower mountains and hills of tropical, sub-tropical regions. He also recognizes some secondary centres of origin where two or more species crossed together. Secondary centres of origin are the places where natural and artificial selection occurred one after another. He stated that plants were not domesticated at random but it was a continuous process. He published his results in 1926 and developed a theory on the centres of origin of cultivated plants. He proposed 8 centres of origin of crop plants

Vavilov considered that “as a rule the primary foci of crop origins were in mountainous regions, characterized by the presence of dominant alleles.” In his work entitled *The Phytogeographical Basis for Plant Breeding* (Vavilov 1935) he summarizes and pulls together all his previous work on centers of origin and diversity. In this he recognizes eight primary centers, as follows.

- I. **The Chinese Center** - in which he recognizes 138 distinct species of which probably the earlier and most important were cereals, buckwheats and legumes.
- II. **The Indian Center** (including the entire subcontinent) - based originally on rice, millets and legumes, with a total of 117 species.
- IIa. **The Indo-Malayan Center** (including Indonesia, Philippines, etc.) - with root crops (*Dioscorea* spp., *Tacca*, etc.) preponderant, also with fruit crops, sugarcane, spices, etc., some 55 species.
- III. **The Inner Asiatic Center** (Tadjikistan, Uzbekistan, etc.) - with wheats, rye and many herbaceous legumes, as well as seed-sown root crops and fruits, some 42 species.
- IV. **Asia Minor** (including Transcaucasia, Iran and Turkmenistan) - with more wheats, rye, oats, seed and forage legumes, fruits, etc., some 83 species.
- V. **The Mediterranean Center** - of more limited importance than the others to the east, but including wheats, barleys, forage plants, vegetables and fruits- especially also spices and ethereal oil plants, some 84 species.
- VI. **The Abyssinian (now Ethiopian) Center** - of lesser importance, mostly a refuge of crops from other regions, especially wheats and barleys, local grains, spices, etc., some 38 species.
- VII. **The South Mexican and Central American Center** - important for maize, *Phaseolus* and Cucurbitaceous species, with spices, fruits and fibre plants, some 49 species.

- VIII. South America Andes Region** (Bolivia, Peru, Ecuador) - important for potatoes, other root crops, grain crops of the Andes, vegetables, spices and fruits, as well as drugs (cocaine, quinine, tobacco, etc.), some 45 species. This region is further sub-divided into following two groups.
- VIIIa. The Chilean Center** - only four species - outside the main area of crop domestication, and one of these (*Solanum tuberosum*) derived from the Andean center in any case. This could hardly be compared with the eight main centers.
- VIIIb. Brazilian-Paraguayan Center** - again outside the main centers with only 13 species, though *Manihot* (cassava) and *Arachis* (peanut) are of considerable importance; others such as pineapple, *Hevea* rubber, *Theobroma cacao* were probably domesticated much later.

1.3 Limitations of Vavilov's views

The expansion of our understanding on cultivated plants pointed certain limitations on Vavilov's views. These views require some modifications,

1. Vavilov considered the region with greatest genetic diversity of a species as the centre of origin of that species. But now, many such species are known whose centres of origin and genetic diversity are different. For example, Maize and Tomato
2. The centres of origin of cultivated plants as per Vavilov are limited to the mountains and small hills in tropical and sub-tropical regions. But recent evidences also suggest plains as the centres of origin of many cultivated plants.
3. Today several crops are known whose centres of origin are different from the ones suggested by Vavilov. Moreover there is more than one centre of origin. Also, the origin of many of the species cannot be traced due to lack of sufficient evidence.

According to Vavilov primary centre is marked by high frequency of dominant alleles towards the centre and recessive towards the periphery. But this view is not acceptable as per the latest knowledge.

1.4 Summary

Knowledge of the origins of crop plants is now basic to economic botany and vitally

important to locate wild relatives, related species and new genes. Under Russian breeding programme, scientist Nikolai Ivanovich Vavilov and his colleagues visited several countries and collected a large number of crop plants and their wild relatives. His deductions were based on evidences from morphology, anatomy, cytology, genetics, plant geography and distribution. He proposed 8 primary centers of origin of crop plants viz. The Chinese Center, The Indian Center, The Indo-Malayan Center, The Inner Asiatic Center, Asia Minor, The Mediterranean Center, The Abyssinian (now Ethiopian) Center, The South Mexican and Central American Center and South America Andes Region. South America Andes Region is subdivided into two centre. Latest knowledge on cultivated plants pointed certain limitations on Vavilov's views.

1.5 Questions & Answers

- i. What are the limitations of Vavilov's views ?

Ans. See Para. 1.3

- ii. According to Vavilov how many primary centers are exist ?

Ans. See Para 1.2

- iii. How many species are recognized from The Chinese center ?

Ans. See Para 1.2

- iv. Indonesia belongs to which primary center ?

Ans. See Para 1.2

- v. Cucurbitaceous species originated in which center ?

Ans. See Para 1.2

Unit 2 □ Cereals

Structure

- 2.0 Objectives**
- 2.1 Introduction**
- 2.2 Wheat**
 - 2.2.1 Origin**
 - 2.2.2 Variety**
 - 2.2.3 Morphology**
 - 2.2.4 Climate Requirement**
 - 2.2.5 Soil**
 - 2.2.6 Fertilizer Management**
 - 2.2.7 Cultivation**
 - 2.2.8 Uses**
- 2.3 Summary**
- 2.4 Questions & Answers**

2.0 Objectives

- This unit will help you to know the origin and variety of the major cereals i.e. wheat.
- You will be able to know the morphology of Wheat plant.
- You will be able to discuss the climate requirement, soil type and fertilizer management for wheat cultivation.
- You will know the cultivation process and different uses of wheat.

2.1 Introduction

The cereals are the most important sources of plant food for mankind and also for some animals since the earliest time. Certain plants of the family Gramineae (Poaceae)

whose grains are used as food are called cereals. 'Cereal' name has been given in honour of the Roman goddess 'Ceres', whom Romans once worshiped as the giver of grain. Cereals are cultivated for their grains which form the foremost staple food of India and other Asian countries.

Wheat, rice, maize are the most important among cereals; others are barley, oats and rye. Regarding food value, cereals contain a high percentage of carbohydrates (68-79%), a considerable amount of proteins (7- 16%) and some fats; vitamins are also present. Cereals belong to the family Gramineae (Poaceae), which is characterized by caryopsis fruits, flowers, grouped in spikelets, half-alternate leaves with ligules and open sheathing leaf bases, delicate and rounded stems and annual or perennial herbs mainly.

The world production of cereals (excluding Sorghum and Millets) in 2021 amounted to 2.1 million tonnes. In India, the area under cultivation of cereals is 98 million (2018) hectares and the corresponding production of grains is 282 million tonnes (2021).

2.2 Wheat

2.2.1 Origin:

Cultivation of wheat started after 8000 BC. Jared Diamond traces the spread of cultivated emmer wheat starting in the Fertile Crescent about 8500 BC. Archaeological analysis of wild emmer indicates that it was first cultivated in the southern Levant with Iran as far back as 9600 BC. Genetic analysis of wild einkorn wheat suggests that it was first grown in the Karacadeg Mountain in South Eastern Turkey.

Remains of harvested emmer from several sites near the Karacadag Range have been dated between 8600 (at Cayonu) and 8400 BC (Abu Hureyra). That is in the Neolithic period. Iraq-ed-Dubb is the exception where the earliest Carbon dated remains of domesticated emmer wheat were found is the earliest levels of Tell Aswad, in the Damascus basin near Mount Hermon in Syria.

The cultivation of emmer reached Greece, Cyprus and India by 6500 BC, Egypt after 6000 BC and Germany and Spain by 5000 BC. The early Egyptians were developers of bread or the use of over and developed baking into one of the first large-scale food production industries.

By 3000 BC, wheat had reached England and Scandinavia A millennium later it reached China. The first identifiable bread wheat (*Triticum aestivum*) with sufficient gluten

for yeasted breads has been identified using DNA analysis in samples from a grainary dating to approximately 1350 BC at Assiros in Greek Macedonia.

Wheat (*Triticum* sp.) is a cereal grain originally from the Levant region of the near East and Ethiopian Highlands. It is now cultivated worldwide. Wheat was already an important crop when history was first recorded and so accurate information on the exact time and place of its origin is not available. The distribution of the wild wheats and grasses, believed to be the progenitors of the cultivated wheats, supports the belief that wheat originated in Southeastern Asia. Some species were cultivated in Greece, Persia, Turkey, and Egypt in prehistoric times while the cultivation of other species may be of more recent origin. In India, evidences from Mohen-Jo-Daro excavations, indicate that wheat was cultivated there more than 5000 years ago.

The modern origin of wheat is of interest for it is a classical example of how closely related species may be combined in nature into a polyploid series. The species of *Triticum* genus to which the cultivated wheat belongs and their close relatives may be divided into diploid, tetraploid and hexaploid groups, with chromosome number $2n=14$, 28 and 42 respectively. The tetraploid group has originated from two diploid species as indicated by the combination of genomic formulae. The hexaploid species originate by the addition of third genome to a tetraploid specie. The 21 chromosomes of the hexaploid wheat have been assigned into seven homeologous groups, each homeologous group containing a partially homologous chromosome from each of the A, B, and D genomes.

Archeological data suggests that wheat was cultivated first around 9600 BCE. Syria, Jordan, Israel are lands of the large area in the Eastern Mediterranean region of Western Asia that is named the Levant in historical terms. The first cultivation of wheat is related to the Southern Levant. Genetic analysis of wild wheat depicts that wheat was grown early in the Karacadag Mountains in Turkey. Thus, wheat originated in the Southwest regions of Asia.

Wheat is the main cereal crop in India and the most important staple food for Indian people. The cultivation of wheat dates back to more than 5000 years back during the era of Indus valley civilization where the original species was *Triticum sphaerococcum* popularly known as Indian wheat has now disappeared and replaced by present day species- *Triticum aestivum* or the common Bread Wheat, *Triticum durum* or the Macaroni wheat and the *Triticum dicoccum* or the Emmer Wheat. Wheat cultivation in India traditionally has been dominated by the northern region of India. The northern states of Punjab and Haryana Plains in India have been prolific wheat producers.

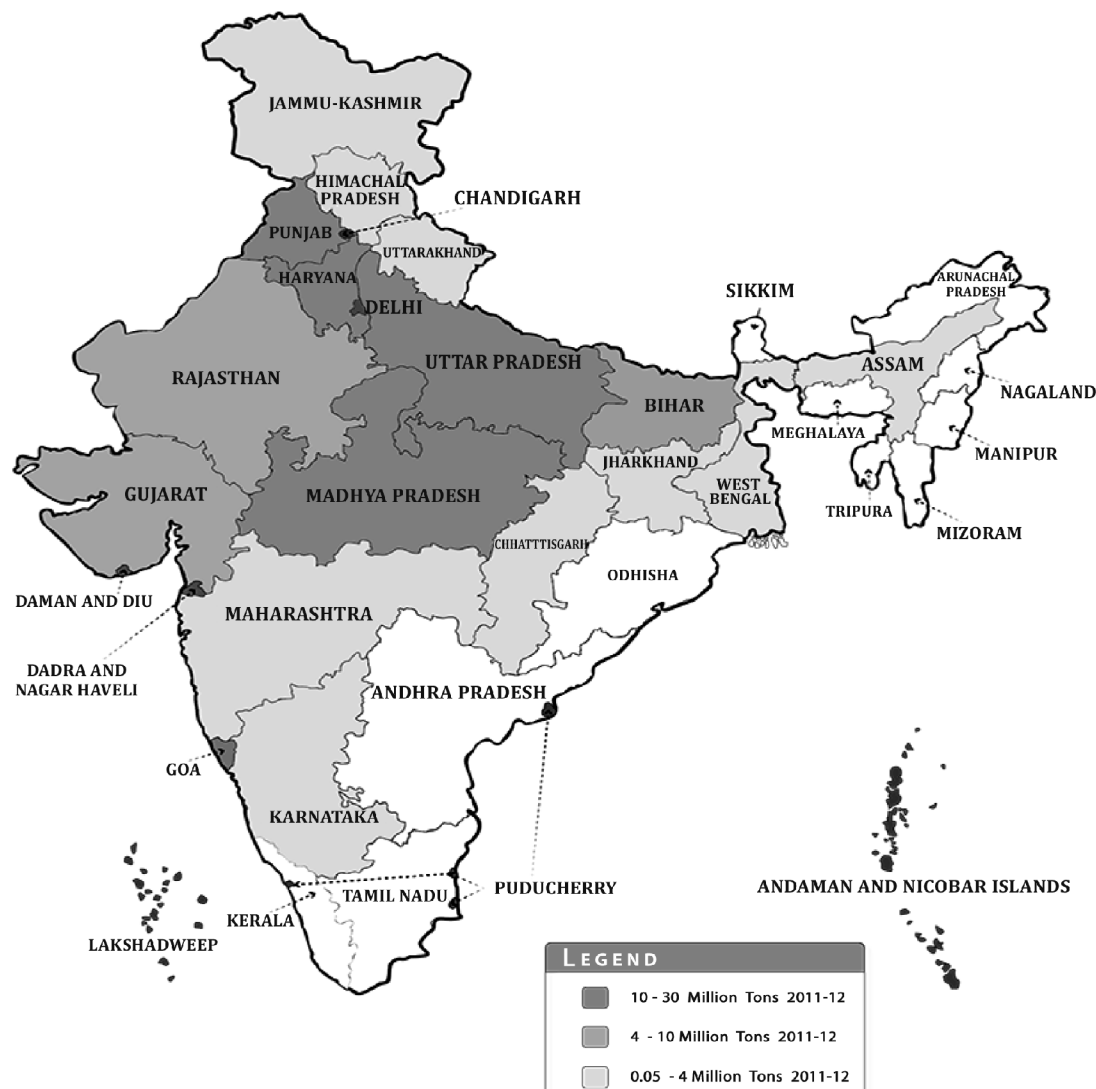


Fig 2.1: Major Wheat Producing States (2018)

The total area under the crop is about 30.60 million hectares (2017) in the country. This crop plant is widely cultivated in UP, Punjab, Madhya Pradesh, Haryana and Rajasthan. Global production of wheat in 2021 was 773.7 million metric tonnes. The major increase in the productivity of wheat has been observed in the states of Haryana, Punjab and Uttar Pradesh. Higher area coverage is reported from MP in recent years. In West Bengal 509 thousand metric tons of wheat was produced during 2020.

Currently, India is the second largest producer of Wheat in the world after China with about 12% share in total world Wheat production.

Triticum aestivum L. is the botanical name of the bread wheat plant which is cultivated extensively in major parts of India (Fig 2.1). Other species are :-*Triticum durum* (Macaroni wheat)-cultivated on the black soils of Central and Peninsular India; *Triticum dicoccum* (Emmer wheat) is grown in Karnataka, Andhra Pradesh. Nilgiri hills of South India and Maharashtra.

Triticum aestivum (wheat) belongs to the family Gramineae (Poaceae). Plant is an annual herb. Leaf blades are linear, flat, auricled. Inflorescence-erect spikes: spikelets solitary, sessile, laterally compressed, 2-several flowered. Glumes ovate, sub-equal, persistent, often unequal sided, obtuse or shortly awned.

Indian Wheat Growing Zones

The entire wheat growing areas of our country has been categorized into 6 major zones as follows-

Zones	States/Regions Covered	Approx Area (million ha)
Northern Hill Zone(NHZ)	Hilly areas of J&K (except Jammu, Kathua and Samba districts), Himachal Pradesh (except Una & Paonta valley), Uttarakhand (excluding Tarai region) & Sikkim.	0.8
North Western Plains Zone (NWPZ)	Punjab, Haryana, Western UP (except Jhansi Div), Rajasthan (excluding Kota & Udaipur div), Delhi, Tarai region of Uttarakhand, Una & Paonta valley of HP, Jammu, Samba & Kathua districts of J&K and Chandigarh.	11.55
North Eastern Plains Zone (NEPZ)	Eastern UP(28 dist), Bihar, Jharkhand, West Bengal, Assam, Odisha and other NE states (except Sikkim).	10.5
Central Zone	MP, Gujarat, Chattisgarh, Kota & Udaipur Div of Rajasthan & Jhansi Div of UP.	5.2
Peninsular Zone	Maharashtra, Tamil Nadu (except Nilgiris & Palani Hills), Karnataka & Andhra Pradesh.	1.6
Southern Hill Zone (SHZ)	Nilgiris & Palani Hills of Tamil Nadu.	0.1

Wheat namely, (i) *T. aestivum*, (ii) *T. durum* and (iii) *T. dicoccum* are being cultivated in the country, as per details given as under :

Species	% share of Production	Major Growing Areas
<i>T. aestivum</i>	95 %	Uttar Pradesh, Punjab, Haryana, Rajasthan, Bihar, West Bengal, Assam, Parts of Madhya Pradesh, Himachal Pradesh, Jammu & Kashmir.
<i>T. durum</i>	4 %	Madhya Pradesh, Maharashtra, Gujarat, Southern Rajasthan and few locations in Punjab.
<i>T. dicoccum</i>	1 %	Karnataka, Maharashtra & Tamil Nadu.

2.2.2 Variety :

Among the high yielding wheat varieties recommended for cultivation mention may be made of ‘Sonalika’, ‘Kalyansona’, ‘Arjun’, ‘Janak’, ‘UP 301’, ‘Pratap’, ‘UP 262’, ‘PV 18’, ‘WG 357’, ‘WG 377’, ‘Girija’, ‘Shailaja’, ‘Sharbati sonora’, ‘Choti Lerma’ etc. In West Bengal ‘Sonalika’ and ‘UP 262’ are recommended for the plains and laterite tract; for terrai region only ‘Sonalika’ is suitable. Varieties released by IIWBR (Indian Institute of Wheat and Barley Research) during last 10 years are-

- Karan Narendra (DBW222)
- Karan Vandana (DBW187) – NWPZ
- Karan Vandana (DBW187)-NEPZ
- Karan Shriya (DBW252)
- DDW 47- Durum

The genus *Triticum* embraces 22 species. Only two of them, *Triticum aestivum* (soft wheat) *Triticum durum* (hard or durum wheat, macaroni wheat) are cultivated. Of all other wheat species only *Triticum dicoccum* (Emmer wheat - Tetraploid 2n=28) is occasionally grown in wheat fields. The grain of durum wheat contains more protein up to 18 - 20 % but its gluten is not porous and not elastic, that is why bread made out of durum wheat is of low porosity. It is extremely good for manufacturing macaroni and confectionaries.

2.2.3 Morphology :

Triticum aestivum (wheat) belongs to the family Gramineae (Poaceae).

- **Habit** : Erect, annual which grows from 30 to 120 cm.
- **Roots** : Mostly adventitious and fibrous.
- **Stem** : Cylindrical with distinct nodes and internodes. Nodes are swollen and internodes are hollow. Usually there are six internodes and the sixth is the spike bearing one (Fig. 2.2A).
- **Leaves** : Simple, alternate in distichous alternate leaves arranged to right and left side of the stem on one plane arrangement, long, linear lamina with leaf sheath covering the internodes, ligule membranous, auricles prominent, claw shaped and clasp the stem. The two halves of the lamina are unequal and show a tendency to twist (Fig. 2.2A).
- **Inflorescence** : It is called as head, spikes or ear. Often 15 - 20 spikelets are borne on rachis in a zigzag shape. The spikelets have two glumes, which may end as an awn (lemma) (Fig. 2.2B).
- **Structure of spikelet** : The wheat inflorescence is a terminal distichous spike (ear), spikelets are sessile and borne singly at the nodes on alternate sides of the zigzag rachis. Each spikelet consists of two to five florets attached alternately on opposite sides of a short central axis called rachilla and is covered by two sterile or empty glumes. Lemma broad with an acute tip or awn, palea thin. The lower lemmas are fertile while the top one or two are sterile. Lodicules two, stamens three with thin filaments and large anthers superior ovary, styles two, single ovule, bifid feathery stigma (Fig. 2.2C-E).
- **Structure of the grain** : The fused pericarp and testa surrounds both the endosperm and the embryo with the scutellum in direct contact with the surface of the endosperm. The axis consists of the primary root, which is enclosed by the coleorhiza, and the plumule with the protecting sheath of the coleoptile enclosing the primordia of two or three foliage leaves and the shoot apex. The part of the axis between the point of attachment of the scutellum and the plumule is called the mesocotyl which is the internode between the scutellum representing the cotyledon and the coleoptile representing the next leaf (Fig. 2.3).

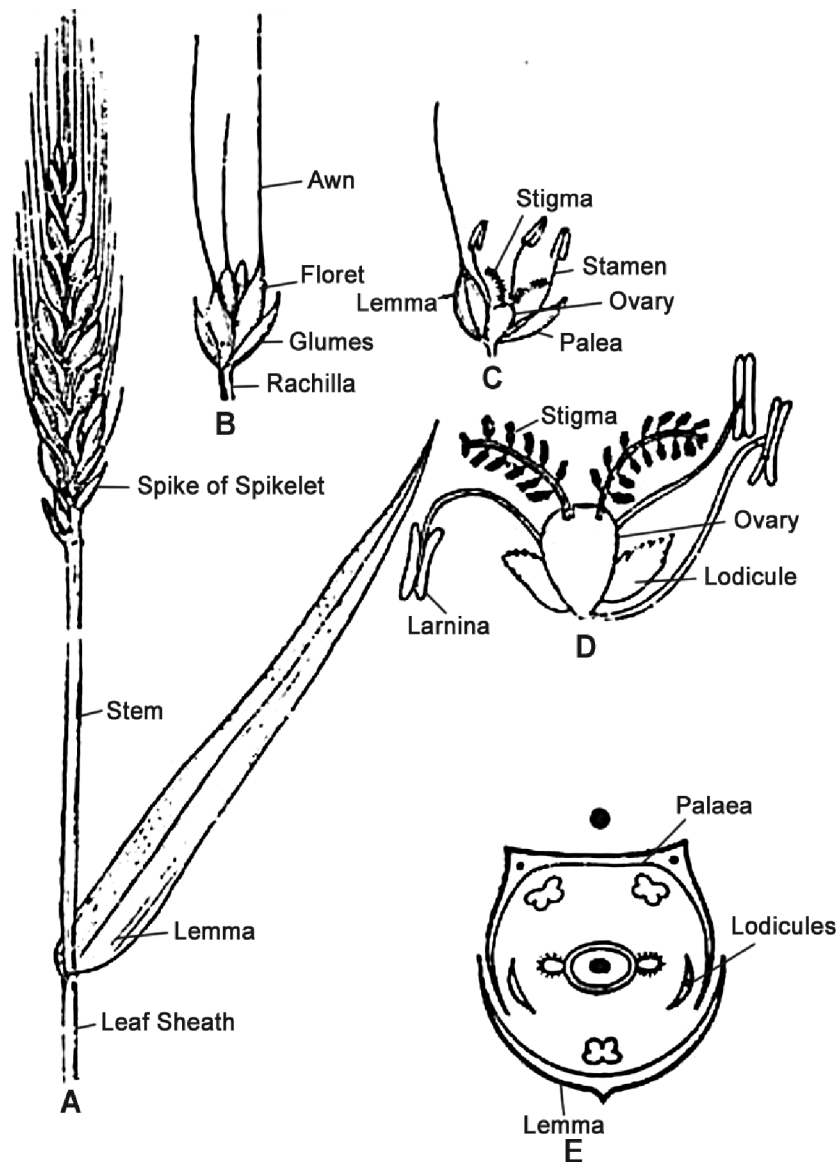


Fig 2.2: Different parts of Wheat plant (A-flowering twig, B-Spikelet, C-A floret opened out, D-floret which lemma and Palea removed, E-floral diagram).

- **Fruit (grain) :** Dry one seeded indehiscent known as caryopsis having more or less oval shape. The apex of the grain has tufts of hairs called bush. The other side has a longitudinal groove or a furrow often called the crease. The either sides of the furrow are called cheeks. The embryo is situated at the base on the dorsal side of the grains (Fig. 2.3).

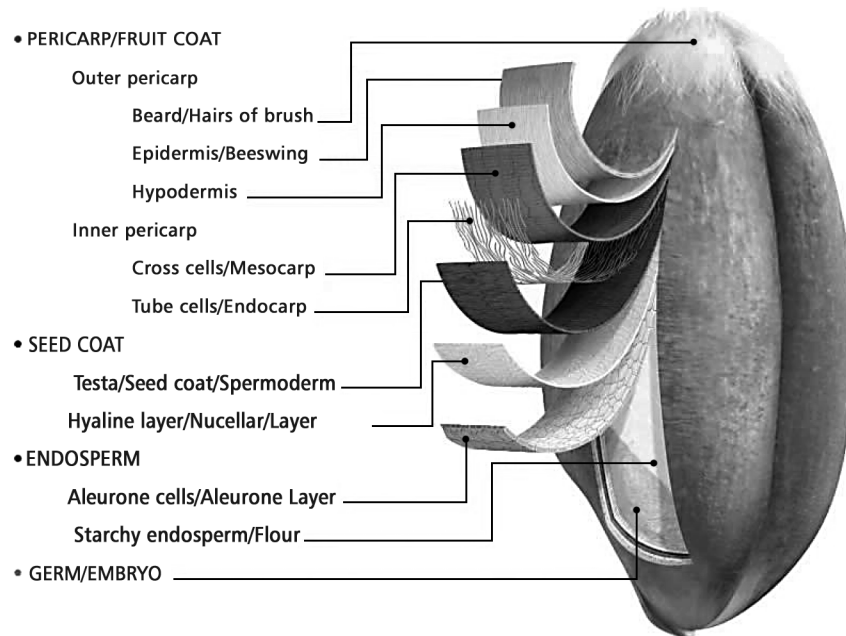


Fig 2.3: Wheat Grain showing different structure.

2.2.4 Climate Requirement :

Wheat is mainly a rabi (winter) season crop in India. Wheat crop has wide adaptability. It can be grown not only in the tropical and sub-tropical zones, but also in the temperate zone and the cold tracts of the far north, beyond even the 60 degree north altitude. Wheat can tolerate severe cold and snow and resume growth with the setting in of warm weather in spring. It can be cultivated from sea level to as high as 3300 meters.

The best wheat is produced in Indo-Gangetic Plain areas favoured with cool, moist weather during the major portion of the growing period followed by dry, warm weather to enable the grain to ripen properly. The optimum temperature range for ideal germination of wheat seed is 20-25 °C though the seeds can germinate in the temperature range 3.5 to 35 °C. Rains just after sowing hamper germination and encourage seedling blight. Areas with a warm and damp climate are not suited for wheat growing. Within an annual rainfall range of 130-1,000 mm, wheat is grown effectively.

2.2.5 Soil :

Wheat is grown in a variety of soils of India. Soils with a clay loam or loam texture, good structure and moderate water holding capacity are ideal for wheat cultivation. Care

should be taken to avoid very porous and excessively drained soils. Soil should be neutral in its reaction. Heavy soil with good drainage are suitable for wheat cultivation under dry conditions. These soils absorb and retain rain water well. Heavy soils with poor structure and poor drainage are not suitable as wheat is sensitive to water logging. Wheat can be successfully grown on lighter soils provided their water and nutrient holding capacity are improved.

2.2.6 Fertilizer Management :

It was demonstrated that 120 kg nitrogen, 60 kg phosphorus and 30 kg potash per hectare were required for optimum productivity. Recently, the new wheat varieties have responded up to 180 kg N/ha with optimum dose around 150 kg/ha. In the Indo-Gangetic plains, application of zinc @ 25kg/ha in rice-wheat system was found to increase the yield substantially. Recently, the use of sulphur has been found beneficial for enhancing the productivity as well as the grain protein content of wheat. Response to Mn (pockets in the Indo-Gangetic plains) and boron (eastern and far eastern region) has also been realized.

2.2.7 Cultivation :

For uniform and healthy germination, the seed bed should be well-pulverized and compact. Several ploughings, repeated harrowing in the rainy season followed by 3 or 4 cultivations and planking immediately before sowing produce a good seed bed for unirrigated crop on alluvial soils. For irrigated crop, the land is irrigated before sowing and the number of ploughings is reduced. Seed, only highly viable and free from weed-seeds and seed borne diseases, is sown by broadcasting, drilling or dibbling at the rate of 100-125 kg per hectare for bold seeded, shy-tillering varieties like "Sonalika", and 75-100 kg per hectare for medium grained varieties. For late sowing the rate is increased to 125 kg / hectare. For rainfed condition the seed rate is 100 kg / ha. The optimum sowing time is first fortnight of November for medium long duration varieties and second fortnight of November for short duration ones. Line sowing (20-22 cm apart) with drills at about 5 cm depth deposits the seed in moist soil which results in good germination and plant stand. In rough, dry and light soils, deeper sowing is the general rule while in moist or heavy lands, comparatively shallower sowing (5 cm) is desirable. For irrigated wheat crop, first irrigation should be given 20-25 days after sowing. Irrigations (3 - 4) at tillering, heading and grain filling stages are essential. Weeding is also equally important which may be done by spraying 2, 4-D (0.4 kg a.e./hectare in 750 litre water-4-6 weeks after sowing) though hand weeding is generally practiced. The dry (non-irrigated) wheat crop

is not normally manured although application of well-rotted farmyard manure or compost or oil-cake at the rate of 20-30 quintal per hectare about six weeks before sowing is desirable. For the irrigated wheat crop, though actual dose should be based on soil fertility status, 80-120 kg/ha N, 40 - 60 kg / ha P_2O_5 and 40-60 kg / ha K_2O is recommended where fertilizer supply is assured; and 60-80 kg/ha N , 30-40 kg/ha P_2O_5 and 30-40 kg/ha K_2O with limited supply. In acid soils of North Bengal and for the red lateritic soils of West Bengal applications of 2 t/ha of rock phosphate one month before sowing is recommended.

Harvesting and Threshing-Harvest begins in mid February and continues till April, depending on variety, when the grain is deadripe and the straw is golden yellow and brittle. Plants are cut by sickle. Harvested grains are then threshed by treading under the feet of cattle on a threshing floor or by pedal and power threshers. Next winnowing is done with winnowing baskets.

Storage- Thoroughly dried grains should be stored in moisture-proof and fumigated store rooms.

Yield-The yield of unirrigated or rainfed crop is generally low; about 300-400 kg/ha in Peninsular or Central India. Under irrigated condition, with high yielding varieties and following the correct production technology yield upto 8 tonnes/hectare has been reported from National Demonstrations. The present all India average wheat yield is **3.5 thousand kilograms per hectare (2021)**.

Diseases and Insect Pests

Common wheat diseases-

- (a) Hill bunt caused by *Tilletia foetida* and *T. caries*. it can be controlled by treating seeds with Ceresan or Agrosan GN@ 2.5 gms per kg and growing resistant varieties.
- (b) Flag smut caused by *Urocystis tritici*-can be controlled by growing resistant varieties, following crop rotation and seed dressing with Ceresan or Agrosan GN@ 1 : 500.
- (c) Karnal bunt caused by *Neovossia indica*-can be controlled by growing resistant varieties.
- (d) Leaf rust (Brown rust) caused by *Puccinia recondita* control like that of (c) and also spraying Zineb and Dithane M 45 @ 3 kg / ha

- (e) Stem rust (Black rust) caused by *Puccinia graminis tritici*, can be controlled by growing resistant varieties, and by seed treatment with Oxycarboxin.
- (f) Stripe rust (Yellow rust) caused by *Puccinia striiformis*, can be controlled by growing resistant varieties and seed treatment with Oxycarboxin.
- (g) Loose smut caused by *Ustilago nuda* can be controlled by growing resistant varieties, solar heat or hot water treatment of seed, rouging of smutted plants, dry seed treatment with Carboxin (0.25%).
- (h) Leaf blight caused by *Alternaria triticina*: Control measures are-growing resistant varieties, hot water treatment of seed, spraying with Mancozeb (2g/ litre) or Dimethyl-dithiocarbamate (3ml / litre) .

2.2.8 Uses :

- **Food:** Wheat is milled to produce flour which is used to make a variety of products around the world including bread across the world. Wheat contains a protein called gluten which is necessary for the basic structure in forming the dough system for bread, rolls and other baked goods. Many of the foods we consume on a daily basis such as bread, cookies, cakes, pies, pastries, cereals, crackers, pasta, flour tortillas and noodles are all made from wheat flour.
- **Straw:** Wheat straw chaff is fed to cattle, poultry and other livestock feed. In many countries, rural houses still use wheat straw to thatch their houses as well as mattresses and pillows. It is also used in the manufacture of paper.
- **Drinks:** Wheat also forms the base for three extremely popular alcoholic drinks - whiskey, vodka, and beer.
- **Wheatgrass:** Young wheatgrass is becoming increasingly popular as a nutritional supplement offering vitamin A, B-complex, C, E, I and K. It is also extremely rich in protein, and contains 17 amino acids, which are the building blocks of protein.
- **Bioethanol:** Since 2010, wheat has been substituted for corn and used to produce bioethanol in the United Kingdom and United States.
- **Biosorbent:** Wheat bran serves as a biosorbent for a variety of inorganic and organic toxic compounds that are not often degradable (such as heavy metal ions, eg. Chromium and Lead).

- **Fiber:** Some strains of wheat are grown to produce starch in South Asia used in textile manufacturing.
- **Others:** Wheat is also being used for plastics manufacturing and aquaculture feed purposes for both fish and shrimp.

2.3 Summary

Certain plants of the family Gramineae (Poaceae) whose grain are used as food are called cereals. Wheat, rice, maize are the most important among cereals. Cultivation of wheat started after 8000 BC. The modern origin of wheat is of interest for it is a classical example of how closely related species may be combined in nature into a polyploid series. The species of *Triticum* genus to which the cultivated wheat belongs and their close relatives may be divided into diploid, tetraploid and hexaploid groups, with chromosome number $2n=14$, 28 and 42 respectively. The total area under the crop is about 30.60 million hectares (2017) in India. Currently, India is the second largest producer of Wheat in the world after China. Wheat namely, *T. aestivum*, *T. durum* and *T. dicoccum* are being cultivated in India. Wheat is mainly a rabi (winter) season crop and is produced in Indo-Gangetic Plain areas favoured with cool, moist weather during the major portion of the growing period followed by dry, warm weather to enable the grain to ripen properly. Soils with a clay loam or loam texture, good structure and moderate water holding capacity are ideal for wheat cultivation, 120 kg nitrogen, 60 kg phosphorus and 30 kg potash per hectare were required for optimum productivity. Common wheat diseases are Hill bunt, Flag smut, Karnal bunt, Leaf rust, Stem rust, Stripe rust, Loose smut and Leaf blight. Wheat is used as food. It is used in alcoholic drinks and bioethanol production, textile manufacturing, plastics manufacturing and aquaculture. Wheat bran serves as a biosorbent for a variety of inorganic toxic compounds. Young wheatgrass is becoming increasingly popular as a nutritional supplement offering vitamin A, B-complex, C, E, I and K.

2.4 Questions & Answers

- i. What are the uses of Wheat ?

Ans. See Para 2.2.8

- ii. Which is the casual organism of Hill bunt disease of wheat ?

Ans. *Tilletia foetida* and *T. caries*.

iii. Describe the process of wheat cultivation ?

Ans. See Para 2.2.7

iv. Describe the soil requirement of wheat cultivation ?

Ans. See Para 2.2.5

v. In India best what is produced in which areas ?

Ans. Indo-Gangetic Plain areas.

vi. How much annual rainfall is needed for wheat cultivation ?

Ans. 130-1000 mm.

Unit 3 □ Legumes

Structure

- 3.0 Objectives**
- 3.1 Introduction**
- 3.2 Gram or Chick pea (Chola, Chana)**
 - 3.2.1 Morphology**
 - 3.2.2 Uses**
 - 3.2.3 Varieties**
- 3.3 Soyabean (Geri kalai, Bhat, Ramkurthi)**
 - 3.3.1 Morphology**
 - 3.3.2 Uses**
 - 3.3.3 Varieties**
- 3.4 Summary**
- 3.5 Questions & Answers**

3.0 Objectives

This unit will help you to acquire knowledge about morphology, uses and varieties of two legumes, i.e. Chickpea and Soyabean.

3.1 Introduction

Next to cereals, legumes are the most important sources of human food. Legumes contain high amount of proteins more than other vegetables, hence they are nearer to animal protein in food value. Highly nutritious legumes are easily grown and mature rapidly. They have high energy content. In addition to protein, carbohydrates and fats are also present in legumes. They are also rich in minerals and vitamin B. Majority of legumes possess root nodules that contain nitrogen fixing bacteria-these bacteria can fix free nitrogen from the air and enhance the soil fertility. All legumes belong to the sub-family Papilionatae of the family Leguminosae which is characterized by having legume type of fruit (developing

from monocarpellary gynoecium and dehiscing along two sutures), one carpel, marginal placentation, racemose type of inflorescence and mostly compound leaves with pulvinus leaf base. In India, the total annual area under the cultivation of legumes/pulse crops is about 283.4 lakh hectares (2019-2020) and the corresponding average production of pulses is about 23.2 million tonnes (2019-2020). In 2021, Higher area is reported from the States of Gujarat (1.07 lakh ha), Rajasthan (0.44 lakh ha), Telangana (0.37 lakh ha), Jharkhand (0.29 lakh ha), Uttar Pradesh (0.25 lakh ha), Maharashtra (0.24 lakh ha), Madhya Pradesh (0.21 lakh ha), Karnataka (0.16 lakh ha), Jammu & Kashmir (0.08 lakh ha), Haryana (0.01 lakh ha), Uttarakhand (0.01 lakh ha) and Punjab (0.01 lakh ha). Less area is reported from the States of Odisha (0.61 lakh ha), Bihar (0.57 lakh ha), Tamil Nadu (0.53 lakh ha), Chhattisgarh (0.45 lakh ha), Andhra Pradesh (0.23 lakh ha), West Bengal (0.18 lakh ha), Nagaland (0.07 lakh ha) and Assam (0.02 lakh ha).

3.2 Gram or Chick pea (Chola, Chana)

- Scientific Name: *Cicer arietinum* L.
- Family: Fabaceae

Cicer arietinum, commonly known as the gram, chickpea, chick pea, or garbanzo bean, is a high protein legume. Gram is the oldest known crop grown in India. It is considered to have originated in Western Asia and spread at a very early date to India and Europe. Gram or chick pea is a native of southern Europe, now successfully grown in North Indian States.

Gram (whole seed) contains 61.2 percent carbohydrates, 5-3 percent fat and 17.1 percent protein among other constituents. In India average annual area for cultivation and production is about 81.43 lakh hectares and 6-7 million tones of crop respectively.

3.2.1 Morphology:

Plant is small bushy annual herb. Leaves even-pinnate compound, rigid, stipulate; leaflets and foliaceous stipules strongly veined and deeply toothed, the rachis ending in a tendril or bristle or with a leaflet. Flowers axillar solitary, bracts small. Sepals 5, connate in an oblique tube. Petals exerted; standard broad and narrowed to a wide claw; wings obliquely obovate, free; keel incurved. Stamens 10, diadelphous, anthers uniform. Ovary sessile, one chambered, 2-many ovuled; style filiform, incurved, beardless; stigma terminal, capitate. Fruit an oblong sessile turgid pod, narrowed into the persistent style. Seeds sub-globose or irregularly obovoid; hilum small, funicle slender (Fig. 3.1).



Fig. 3.1. : 1-Flowering twig of Gram plant. 2- Leaflet. 3- Pod. 4- Flag. 5- Wing. 6 – Keel, 7 – Anther. 8 – Pistil and 9 – Seed.

3.2.2 Uses :

- The seeds of the plant are edible, cotyledons of seeds are used in the form of ‘dal’, ‘besan’ etc.
- The vegetative parts are used as salad and fodder.

- For the treatment of scurvy disease germinated seed is very useful.
- Green leaves contain malic and oxalic acids and are prescribed for intestinal disorders.

3.2.3 Varieties :

About 40 improved varieties of gram are recommended in different states. In West Bengal Mahamaya-1 and Mahamaya-2 are the recommended varieties.

3.3 Soyabean (Geri kalai, Bhat, Ramkurthi)

- Scientific Name: *Glycine hispida* (Syn *Glycine max* (L) Merr.)
- Family: Leguminosae
- Sub family: Papilionatae

Glycine hispida, commonly known as soyabean is a native of South-East Asia and its first cultivation was recorded in China, in 2838 BC. In India, soyabean is now cultivated in the Madhya Pradesh, Punjab, Himachal Pradesh, West Bengal, the Khasia and Naga hill, hilly tracts of Assam up to an altitude of 1,822 m, Manipur and Bihar. In some countries of East Asia, soyabean is grown mainly as a food crop.

3.3.1 Morphology :

Glycine hispida is an annual sub-erect or twining herb covered with hairs, reaching up to a height of 1.8 m. Stem erect or climbing. Leaves trifoliate, ovate-lanceolate, long petioled. Flowers small, white or purple to reddish purple on short axillary racemes. Sepals 5, connate in a campanulate tube, lobes equally distinct. Petals little exerted, standard sub-orbicular; wings narrow, keels obtuse, Stamens 10, monadelphous or at length diadelphous. Ovary sub-sessile, style short, incurved, beardless; stigma terminal, capitate. Pods 3.5-5.0 cm long in clusters of 3-5, densely hairy. Seeds elliptical, compressed, yellow or chocolate or black in colour (Fig. 3.2).

3.3.2 Uses :

- Now-a-days soyabean has become highly essential and vital crop. Soyabean, with about 40% high quality protein is the richest source of vegetable protein. Soyabean protein contains all the essential amino acids in large quantities.

- In addition to proteins, soyabean contains considerable amount of vitamins and minerals, specially calcium in rich quantity.
- The seeds of soyabean are used as pulse. Green seeds are used as vegetables; roasted and salted seeds are used in çakes and candies.

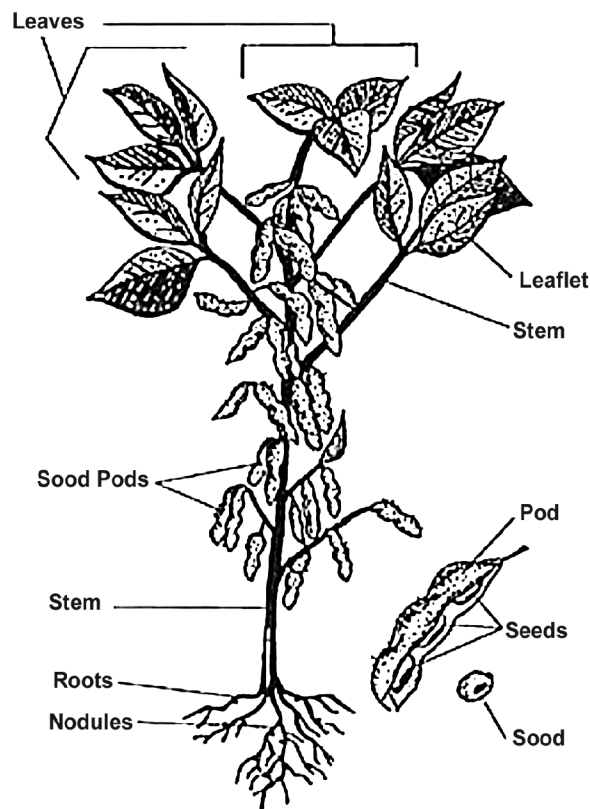


Fig. 3.2. : Whole plant of Saya bean. (*G. hispida*).

- The flour, obtained from the seeds, contains low carbohydrate and high protein which is an excellent food for diabetics.
- Seeds are processed to give milk-like products, curd or cheese. Soyabean milk extracted from the seed, is used as substitute of caesin in the preparation of sweets also.
- At present artificial “meat”, made from soyabeans, has been developed by a Japanese food manufacturer-this meat will be used for mixing into processed meat such as saussages, meat balls and meat paste.

- **Animal Feed Production-** The high-protein fiber that remains after soyabean oil is removed is toasted and prepared into animal feed for poultry, pork, cattle, fish, and pets. Over half of the soyabeans processed for livestock feed are fed to poultry. Soya protein is increasingly found in fish food, both for home aquariums and for farm-raised aqua protein. Most marine species were fed fish meal at one time, but wild caught fish feed has become scarcer, making soyfeed a more sustainable and affordable choice.
- **Biodiesel-**Fuel for biodiesel engines can be produced from soyabeans through a process called transesterification. This process simply removes the glycerin from the oil. Soya biodiesel is desirable because it burns cleaner than petroleum-based diesel. Advantages include reduction of particle emissions, non-toxicity, renewability, and environmentally-friendliness.
- **Soy Crayons-**Soy crayons made by the Dixon Ticonderoga Company replace the petroleum found in regular crayons with soy oil making them non-toxic and safer for children.
- **Biocomposites-**These are building materials made from recycled newspaper and soyabeans. They replace other products traditionally made from wood such as furniture, countertops, and flooring.
- **Candles-**Candles made with soyabean oil burn longer, but with less smoke and soot.
- **Carpets-** Soy products are used in many home and commercial carpets, as well as automobile interior upholstery.
- **Cleaners-**Soy oil produces environmentally friendly solvent that safely and rapidly removes oil from streams, creeks, and shorelines without harming people, animals or the environment.
- **Soy Tires-** Goodyear company has recently launched a line of tires made from soyabean oil that rival traditional tires for performance, and are more environmentally friendly than petroleum-based tires.
- **Soy Ink-** Soy ink is superior to petroleum ink because it is non-toxic and easy to clean up.
- **Soy Foams-** Soy-based foams can be used in refrigerators, coolers, automotive interiors, and footwear.

- **Concrete Protector-** Poreshield brand have created non-toxic concrete sealants made from soyabean oil that extend the life of paved surfaces.

3.3.3 Varieties :

Ankur, Alankar, Bragg. Clark 63, Improved Pelicon, Loo, Punjab-1, Shilajeet, JS-2 UPSM 19, R 184, etc.

3.4 Summary

Highly nutritious legumes contain high amount of protein, minerals and Vitamin B. All legumes belong to the family leguminosae, characterized by legume type of fruit. High protein legume *Cicer arietinum*, commonly known as the gram, chickpea, chick pea, or garbanzo bean and is the oldest known crop grown in India. It is considered to have originated in Westem Asia and spread at a very early date to Inda and Europe. Plant is small bushy annual herb. Seeds of plant are edible and green leaves are prescribed for intestinal disorder. Mahamaya 1 and Mahamaya 2 are recommended varieties in West Bengal.

Glycine hispida, commonly known as soyabean is a native of South-East Asia and its frist cultivation was recorded in China, in 2838 BC. In India, soyabean is now cultivated in the Madhya Pradesh, Punjab, Himachal Pradesh. West Bengal, the Khasia and Naga hill, hilly tracts of Assam up to an altitude of 1,822 m, Manipur and Bihar. It is an annual sub-erect or twining herb covered with hairs, reaching up to a height of 1.8 m. Now-a-days soyabean has become highly essential and vital crop due to the richest source of vegetable protein. Soyabean protein contains all the essential amino acids in large quantities. In addition to proteins, soyabean contains considerable amount of vitamins and minerals, specially calcium in rich quantity. The seeds are used as pulse, Green seeds are used as vegetables, roasted and salted seeds are used in cakes and candies. Artificial “meat”, made from soyabeans, will be used for mixing into processed meat such as saussages, meat balls and meat paste. The flour, obtained from the seeds, is an excellent food for diabetics. Soyabean milk extracted from the seed, is used as substitute of caesin in the preparation of sweets also. The high-protein fiber is used in animal Feed Production. Soya protein is increasingly found in fish food, both for home aquariums and for farm-raised aqua protein. Soya biodiesel is desirable than petroleum based diesel because it is renewable and clean burning fuel. Soy crayons replace the petroleum found in regular crayons with soy oil making them non-toxic and safer for children. Soya oil, Soy foam

and Soy ink is used in several industries. Ankur, Alankar, Shilajeet, UPSM 19, R 184 etc. are some Varieties of this crop.

3.5 Questions & Answers

- i. What is the scientific name of Chick pea ?

Answer : *Cicer arietinum L.*

- ii. What are the uses of Chick pea ?

Answer : See Para 3.2.2

- iii. Name two varieties of Chick pea.

Answer : See Para 3.2.3

- iv. What are the uses of *Glycine hispida* ?

Answer : See para 3.3.2

- v. Name two varieties of *Glycine hispida*.

Answer : See para 3.3.3

Unit 4 □ Spices

Structure

- 4.0 Objectives**
- 4.1 Introduction**
- 4.2 Clove**
 - 4.2.1 Morphology**
 - 4.2.2 Uses**
 - 4.2.3 Varieties**
- 4.3 Black Pepper**
 - 4.3.1 Morphology**
 - 4.3.2 Uses**
 - 4.3.3 Varieties**
- 4.4 Summary**
- 4.5 Questions & Answers**

4.0 Objectives

This unit will help you to acquire knowledge about morphology, uses and varieties of two spices, i.e. Clove and Black Pepper.

4.1 Introduction

Spices may be bark, buds, sprouts, natural items, leaves, rhizomes, roots, seeds, characteristics of disgrace and styles or the entire plant tops. The history and culture of Indian spices is likely as old as human advancement itself. The Vedas, the Bible and the Quran are generally stuffed with references - quick or circumlocutory - to Indian spices. The most timely unique record in India on spices is the Rig Veda (around 6000 BC), and the other three Vedas - Yajur, Sama and Atharva. In India total spice production was estimated to be around 10.4 million metric tons (2021). The state of Madhya Pradesh was the largest producer of spices across India in 2021. Rajasthan and Gujarat were the other major producers that year.

4.2 Clove

- Scientific Name: *Syzygium aromaticum* (L.) Merr. & L.M.Perry
- Family: Myrtaceae
- Chemical Compound: **Eugenol** comprises 72–90% of the essential oil extracted from cloves, and is the main cause of clove aroma.

Syzygium aromaticum, also known as clove, is a dried flower bud belonging to the Myrtaceae family that is indigenous to the Maluku islands in Indonesia but has recently been farmed in different places worldwide. The clove tree is composed of leaves and buds (the commercial part of the tree) and the flowering bud production begins four years after plantation. Afterward, they are collected either by hand or using a natural phytohormone in the pre-flowering stage. Interestingly, they are commercially used for many medicinal purposes and in the perfume industry, and clove is considered one of the spices that can be potentially used as preservatives in many foods, especially in meat processing, to replace chemical preservatives due to their antioxidant and antimicrobial properties. Several reports have documented the antibacterial, antiviral, anticarcinogenic, and antifungal activities of some aromatic herbs including cinnamon, oregano, clove, thyme, and mint. However, clove has gained much attention among other spices due to its potent antimicrobial and antioxidant activities.

Clove is rich in many phytochemicals viz. sesquiterpenes, monoterpenes, hydrocarbon, and phenolic compounds. Eugenyl acetate, eugenol, and β -caryophyllene are the most significant phytochemicals in clove oil.

The islands of Zanzibar, Pemba (now part of Tanzania) and Indonesia are the major producers of clove in the world. In India, clove is mostly grown in the hilly tracts of Tamil Nadu, Kerala and Karnataka.

4.2.1 Morphology :

The clove tree is an evergreen that grows up to 8–12 metres (26–39 ft) tall, with large leaves and crimson flowers grouped in terminal clusters. The flower buds initially have a pale hue, gradually turn green, then transition to a bright red when ready for harvest. Cloves are harvested at 1.5–2 centimetres (0.59–0.79 in) long, and consist of a long calyx that terminates in four spreading sepals, and four unopened petals that form a small central ball (Fig 4.1, 4.2).



Fig. 4.1: Twig of Clove plant with fruit.

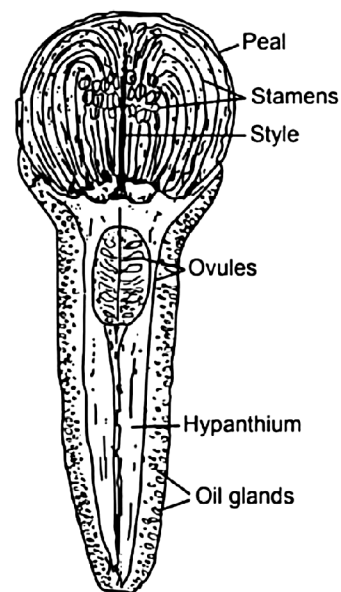


Fig. 4.2: Longitudinal section of a Clove bud.

4.2.2 Uses :

- **Cuisine-** Cloves are used in lending flavor to meats, curries, and marinades, as well as fruit (such as apples, pears, and rhubarb). It may be used to give aromatic and flavor qualities to hot beverages, often combined with other ingredients such as lemon and sugar.
- **Aromatherapy-** The essential oil is used in aromatherapy.
- **Traditional medicine-** Cloves are used in traditional medicine as the essential oil, which is used as an analgesic mainly for dental emergencies and other disorders. Cloves are also helpful in nasal and respiratory infections. Aids in digestion and treating stomach ulcers.
- **Cigar-** Since 2009, clove cigarettes have been classified as cigars in the US. This spice is used in a type of cigarette called *kretek* in Indonesia.
- **Ant repellent-** Bioactive chemicals of clove may be used as an ant repellent.

4.2.3 Varieties:

Penang, Zanzibar and Amboyan varieties are popular in India. Some other famous varieties include Madagascar, Posi Posi, White, Afo, Kotok, and Siputi cloves.

4.3 Black Pepper

- Scientific Name: *Piper nigrum* L.
- Family: Piperaceae
- Chemical Compound: **Piperine**

Piper nigrum, commonly known as Black Pepper is a flowering vine in the family Piperaceae, cultivated for its fruit, known as a peppercorn. The fruit is usually dried and used as a spice and seasoning. Black pepper is the world's most traded spice. Known as the 'king of spices', it forms a significant part of a large number of cuisines all over the world, used in cooking as well as garnishing. Its spiciness is due to the chemical compound piperine, which is a different kind of spicy from the capsaicin characteristic of chili peppers.

In 2019, Ethiopia was the world's largest producer and exporter of black peppercorns, producing 374,413 tonnes or 34% of the world total. Other major producers were Vietnam, Brazil, Indonesia, India, China, and Malaysia.

4.3.1 Morphology :

Black Pepper is a perennial woody vine growing plant up to 4 m (13 ft) in height on supporting trees, poles, or trellises. It is a spreading vine, rooting readily where trailing stems touch the ground. The leaves are alternate, entire, 5 to 10 cm (2.0 to 3.9 in) long and 3 to 6 cm (1.2 to 2.4 in) across. The flowers are small, produced on pendulous spikes 4 to 8 cm (1.6 to 3.1 in) long at the leaf nodes, the spikes lengthening up to 7 to 15 cm (2.8 to 5.9 in) as the fruit matures. A single stem bears 20 to 30 fruiting spikes (Fig. 4.3).

4.3.2 Uses :

- Culinary Uses-

Black pepper is used in cooking as well as garnishing. It is used as an ingredient in recipes to add flavor and spice to meats, fish, vegetables, salad dressings, soups, stir-fries, pasta, and more.

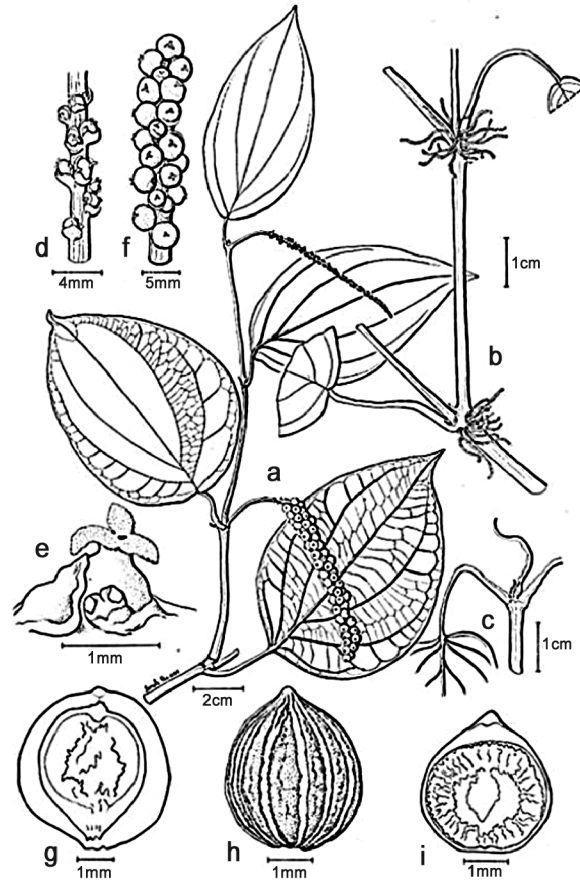


Fig. 4.3: a- Twig of black pepper, b-Terminal stem node, c- Shoot tip, d-Inflorescence, e- Single flower, f- Mature stage of fruits, g & i- Section of fruit, h- Whole fruit.

- Non-Culinary Uses-

- ❖ Black **pepper** is a major component of Ayurvedic medicines that have been used for as long as a cure for the most common respiratory problems that affect all of us. It has a stimulatory effect on the human brain. It especially helps patients with neurodegenerative diseases. It improves memory and cognitive function in an individual by stimulating the chemical pathways in the brain.
- ❖ It is useful in controlling the sugar level in patients with type 2 diabetes. Black pepper's antioxidant properties help to stabilize blood sugar, while also boosting the digestive tract's health. It helps to fight obesity, which is also one of the major contributing factors of diabetes.

- ❖ Piperine in black pepper helps reduce cholesterol uptake, reduces the levels of 'bad' cholesterol (LDL- Low-density lipoprotein) while simultaneously increasing the levels of high-density lipoprotein 'good' cholesterol (HDL- High-density lipoprotein).
- ❖ Eating freshly ground black **pepper** can help you to keep cancer at bay, due to its anti-inflammatory, antibacterial and antioxidant effects. It helps to increase the absorption of essential nutrients like calcium and selenium etc.
- ❖ It helps prevent vitiligo and maintains the original colour of the skin, also help to clear acne by doing away with the intoxicants in it.
- ❖ Quitting smoking-Black pepper oil could help to quit smoking. A small 2013 study measured the intensity of people's smoking cravings before and after smelling black pepper oil for two minutes. The results showed that the oil could effectively reduce nicotine cravings. Some of the participants even quit smoking altogether.
- ❖ Intravenous catheter insertion- Black Pepper oil aids in intravenous catheter insertion. The *Journal of Alternative and Complementary Medicine* published a study about one experimental use of the oil. Nurses added black pepper oil to a gel and rubbed it on the skin of patients where they had difficulty placing intravenous catheters. The research showed that the oil significantly helped with catheter insertion.
- ❖ Swallowing problems-Black pepper could cut down on swallowing problems for the elderly. Swallowing difficulties — especially for the elderly — often lead to a lot of other serious conditions, including pneumonia. A study found that smelling black pepper oil was a risk-free way of stimulating the reflexes in the body that cause you to swallow. Because it's so easy to use, elderly people who have trouble swallowing, like those who have recently had a stroke, may benefit from smelling the essential oil.

4.3.3 Varieties :

In India over 75 varieties or cultivars of pepper are grown. **Karimunda** is the most popular among them. Other important varieties are Billimalligesara, Karimalligesara, Doddigya, Mottakare and Uddagare in Karnataka State and Kottanadan, Narayakodi, Aimpiriyan, Neelamundi, Kuthiravally, Balancotta, and Kalluvally in Kerala State.

The first hybrid, **Panniyur 1** (Fig below), was produced at Panniyur Pepper Research Station in Kerala over three decades ago. This hybrid variety is very popular among pepper farmers in India. At present 12 varieties have been released including Panniyur 1 for cultivation in India by different research stations located at Kozhikode, Panniyur and Palode, in Kerala. **Panchami** is the highest yield variety among them. Piperine content is highest in **Panniyur 2** variety.

4.4 Summary

Syzygium aromaticum or clove, is a dried flower bud belonging to the Myrtaceae family. It is indigenous to the Maluku islands in Indonesia but has recently been farmed in different places worldwide. The clove tree is an evergreen that grows up to 8-12 metres (26-39 ft) tall, with large leaves and crimson flowers grouped in terminal clusters. The main cause of clove aroma is Eugenol. The islands of Zanzibar, Pemba (now part of Tanzania) and Indonesia are the major producers of clove. In India, clove is mostly grown in the hilly tracts of Tamil Nadu, Kerala and Karnataka. Cloves is used in Cuisine, Aromatherapy and traditional medicine. It is also used as cigar and ant repellent. Penang, Zanzibar and Amboyan are popular varieties in India for cultivation.

King of Spices or Black Pepper (*Piper nigrum*) is a flowering view in the family Piperaceae, cultivated for its fruit, known as a peppercorn. Black Pepper is a perennial woody vine growing plant up to 4 m (13 ft) in height on supporting trees, poles, or trellises. It is the world's most traded spice. Ethiopia was the world's largest producer and exporter of it in 2019. It is used in cooking as well as garnishing. It is a major component of Ayurvedic medicines that have been used for as long as a cure for the most common respiratory problems. Piperine in black pepper helps reduce cholesterol uptake, reduces the levels of 'bad' cholesterol while simultaneously increasing the levels of 'good' cholesterol. Black pepper can help to keep cancer at bay, due to its anti-inflammatory, antibacterial and antioxidant effects. Study reports showed that Black pepper oil could help to quit smoking and used in intravenous catheter insertion.

4.5 Questions & Answers

- i. What is the scientific name of Clove ?

Answer : *Syzygium aromaticum*

ii. Which spice is called as King of Spices ?

Answer : Black Pepper or *Piper nigrum*.

iii. What are the non-culinary uses of Black Pepper ?

Answer : See Para 4.3.2

iv. King of Spices belongs to which family ?

Answer : Piperaceae

v. Mention two Indian varieties of Clove ?

Answer : See Para 4.2.3

Unit 5 □ Beverages

Structure

- 5.0 Objectives**
- 5.1 Introduction**
- 5.2 Tea**
 - 5.2.1 Genesis of Tea**
 - 5.2.2 Morphology**
 - 5.2.3 Processing of Tea**
 - 5.2.4 Uses**
- 5.3 Summary**
- 5.4 Questions & Answers**

5.0 Objectives

After reading this unit you will acquire knowledge about genesis, morphology, processing and uses of Tea.

5.1 Introduction

Beverage is a liquor meant for drinking. Various type of beverages also form some sort of important human food because of their liquid content. Since very early times man was in the habit of finding out palatable drinks or beverages, most of them are plant based products and have become of commercial importance. Two types of beverages may be recognised viz., non alcoholic and alcoholic.

Non-alcoholic beverages include tea, coffee, cocoa, cola etc. These beverages contain caffeine (an alkaloid) and are used world-wide for their stimulating and refreshing qualities. Of the various beverages, tea which originated in China, is important and used by one-half of the population of the world; next in importance comes coffee, it also originated in regions adjacent to South-western Asia and is now in 2020/2021, around 166.63 million 60 kilogram bags of coffee were consumed worldwide. Cocoa, a native of tropical America, is also used today as both food and drink globally.

Alcoholic beverages contain alcohol-a poison, which when taken in excess, produces deleterious effects on the human system. Alcoholic beverages are manufactured by the fermentation of sugars present in various fruit juices or sugars produced by the transformation of starch.

5.2 Tea

One of the most important and popular caffeine containing beverage is Tea. It is obtained from the leaves of the tea plant, the tender leaves are processed into cured products which, when added in boiled water, yield the beverage of the same name. It contains 13 - 1 % tannin, 2-5% theine-an alkaloid identical with caffeine and a volatile oil. Volatile oil and alkaloids are readily dissolved in hot water. Tea has served as principal beverage since the early part of the 5th century, before which it was valued only for its medicinal properties.

The word 'tea' came from the Chinese Amoy word **t'e**, pronounced **tay**. The Dutch, who were the first to import tea into Europe from the port of Amoy in Fujian Province, called it **thee**, which became '**tea**' in English. The Mandarin word for tea is **cha**, became **ch'a** (pronounced **tcha**) in Cantonese, in Indian it is **chai**.

The actual centre of origin of tea is not clearly known. Some scholars believed that the centre of origin might be the Tibetan Plateau including Sze Chuan, Yunan, Sain, North East India or China. The idea of tea as beverage travelled gradually from the East to the West by the sixteenth century. India is the largest producer of Black tea while China is the largest producer of Green tea in the world.

- Scientific Name: *Camellia sinensis* (L) Kuntze.
- Family: Theaceae.

5.2.1 Genesis of Tea :

- In 2737 B.C tea plant was discovered in China by the Chinese King Shen Nung.
- In 350 A.D for the first time, one of the Chinese dictionaries mentioned about tea.
- In between 400-600, the demand of tea was raised in China. In that time, tea was consumed as medical beverage. The cultivation process was developed. During this period, the concept of tea was travelled to Japan.

- In between 648-749, Gyoki, the Japanese monk planted tea in some 50 temple gardens. In Japan, tea was rare and expensive, only the aristocrats and priests would enjoy it.
- In 780, for the first time tax was imposed on tea in China.
- In 1211, Abbot Eisai, a Japanese Buddhist wrote a book on tea titled 'Kitcha-Yojoki'.
- In 1589, the Europeans learned about tea.
- In 1597, the word 'tea' got translated in English for the first time.
- In 1610, tea was bought by the East India Company. They marketed it as a foreign medical drink. According to them, tea was expensive and common people could not afford it.
- In 1661, there was a debate on the benefits versus harmful aspects of tea. Dutch doctors were in favor of health benefits of tea and on the other hand, German and French doctors were of its detrimental aspects.
- In 1723, the import tax was reduced on tea by Robert Walpole, the British Prime Minister.
- In 1765, tea gained popularity in America.
- In 1778, Joseph Bank, the British Naturalist suggested to cultivate tea in India.
- In 1823, indigenous tea was originated in India by Robert Bruce, a Scottish Adventurer.
- In 1835, tea cultivation was started in Assam by the East India Company.
- **In 1856, tea plantation was started in Darjeeling, India.**
- During 1865- 1867 a situation of tea disaster was hit to the tea planters in India.
- In 1904, the concept of iced tea was generated by Richard Blechynden.
- In 1909, Thomas Lipton started packaging and blending of tea.
- In 1953, first instant tea was introduced.

5.2.2 Morphology :

The tea plant in wild condition is an evergreen tree attaining a height of 15 - 20m, but under cultivation the plant is maintained as a much branched shrubby bush about 0.6-1.5 m high. Leaves are simple, alternate exstipulate with lanceolate and leathery lamina having serrated margin and numerous oil glands. Flowers are actinomorphic, hypogynous, bisexual, white or pinkish and fragrant solitary, axillary at the axil of leaves. Sepals and petals 5, free and imbricate; stamens many, free; carpels 3, united, ovary 3-celled and each cell 2-ovuled. Fruit capsular (Fig. 5.1).

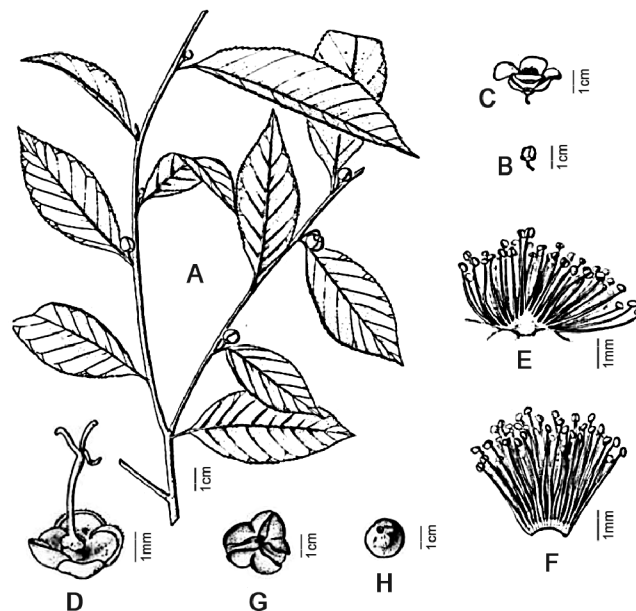


Fig. 5.1: A- Twig of Tea plant, B- Flower bud, C- Single flower, D- Gynoecium, E & F- Androecium, G & H- Fruit.

There are two main varieties of *Camellia sinensis*

1. ***Camellia sinensis* var *bohea*** : It is Chinese tea. Chinese variety is a tall plant with soft leaves, it can grow up to an altitude of 1, 520 meters.
2. ***Camellia sinensis* var *assamica*** : It is the Indian i.e.. Assam tea. Assam variety is a dwarf bush with leathery leaves and can be grown up to an altitude of 2,432 meters.

Tea is mainly a crop of tropical and hot temperate regions. It is a native of India (Assam) or of China. In India, tea is cultivated largely in Assam, West Bengal (Darjeeling and Jalpaiguri districts), Kerala and Tamil Nadu and to a small extent in Tripura, Karnataka

and Dehra Dun of UP. Area under tea cultivation in West Bengal 96,000 hectare and production 16.25 crore kg in 1990.

5.2.3 Processing of Tea :

We will discuss different processing techniques of four major commercial varieties of tea.

- (a) Black Tea,
- (b) Brick Tea
- (c) Green Tea,
- (d) Oolong Tea.
- (e) CTC Tea.

5.2.3.1 Black Tea

For the manufacture of this type, following four major operations are necessary.

- Withering,
 - Rolling,
 - Fermenting and
 - Firing
- **Withering**–The plucked leaves are taken to withering houses where they are spread out at the rate of 0.423 kg leaf to 1 square yard either on slopping wire netting racks or on horizontal hessian cloth ‘chung’ for 18-24 hours. The main object of withering is to eliminate about half of the water content of the leaves so that they can stand the strain of rolling without breaking up.
 - **Rolling**-Withered leaves are then taken to rolling rooms where they are subjected to three rolls by machine, each of 30 minutes duration, with 10 minutes between each for unloading, Kutcha sifting and refiling. This process curls the leaves and burises the cells of the leaves so that their sap is exposed to the action of the oxygen to the air. A low temperature of the room (24-26°C) should be maintained.
 - **Fermenting**-After rolling and shifting the leaves need to be fermented for a period which depends on weather, temperature and amount of rolling they have undergone. Rolled leaves are spread on a cement floor or some other clean surface of fermenting bed having 1.2 cm to 2.5 cm thick surface layer for a

period varying between 2.30 and 4.30 hours. Optimum temperature for this process should be low i.e., 24°C or 26.5°C. During this process, tannins present in the leaves are acted on by enzymes-as a result colour of leaves changes from light red to brown.

- **Firing**-Fermented leaves are taken from fermenting bed and dried in a current of hot air by 'pressure' type drier machine as quickly as possible. The temperature of the drier should be kept between 60-65.5°C. A sufficiently high inlet temperature (87.5°C - 92.5°C) should be maintained to keep the temperature at the top of the machine steady. Time required for firing process varies from 30 to 40 minutes.

The main object of this process is to arrest fermentation and to desiccate leaves slowly to extract the moisture without scorching the tea but at the sametime preserving quality and other characters of tea. Then the dried tea leaves are passed through sieves of different meshes. thus sorting out 'leaf, 'broken' and 'dust' tea. These are again graded into different grades like BOP ("broken orange pekoe"), OP ("orange pekoe"), BP ("broken pekoe"), etc. Different grades are then packed carefully in separate plywood tea chests lined with aluminium foil and paper, and thus ready for marketing.

(b) Brick Tea-This tea is consumed in Tibet and to some extent in former U.S.S.R. Brick tea is manufactured in the following ways:

- **Panning**-Coarse plucked leaves together with twigs are subjected to heat treatment at a temperature of 71 - 94°C in a large cast iron vessel built into a brick stove for about 10 minutes. In this process the leaves become a little soft and they turn olive green in colour. No withering is necessary.
- **Rolling**- Leaves are then passed to rolling machine in which they are rolled for about half an hour; in this process the fibre of the leaf being well lacerated and all broken. There is no curling, as the leaves are too coarse.
- **Fermentation**- Coarse leaves are placed in a heap upon a mat or upon a cement floor, about 15.2 cm deep (or 0.6-0.9 m deep), covered with a sheet or tarpaulin and left for about five days of very dry leaves days. The heap generates a considerable amount of heat and after a few days a black fungus begins to grow amongst it-this fungus plays an important role in the fermentation process, typical black colour and flavour of brick tea.

- **Drying-** After fermentation is complete, the leaves are dried either in sun or on small brick furnace fired by charcoal. The dried leaves can be kept for long time before moulding into bricks.
- – The moulds used in Tibet and China are 1.2 m long and 3.0 cm × 11.3 cm internally with strips in corners. The bulk of leaf, immediately after drying, is rammed as tight as possible into the moulds. Each mould holds material for four bricks of about 1.8 kg each. After about three days, the bricks have settled to allow of the moulds opening. The bricks are then packed in paper and put in the sun.

(c) Green Tea-This type of tea is mainly produced in China and Japan. The main feature in the manufacture of green tea, as distinguished from black tea, is that the green tea does not require artificial withering and fermenting processes. Instead natural withering or wilting is done which retain the greenness of the leaves. Following operations are followed in the manufacture of green tea:

- **Panning-**This process is done in the same way as described for brick tea.
- **Steaming and Rolling-**In this process, leaves are first heat treated at about 120 °C either by pan-firing or by steaming. As soon as the leaves have become soft enough, they are rolled by hand on a bamboo mat.
- **Drying-**Next, rolled leaves are taken to brick furnace where they are dried upon trays over charcoal fires until they are perfectly crisp.

(d) Oolong Tea-This is an intermediate type between black and green tea. Oolong tea is partially fermented and has the colour of the black tea and the flavour of the green tea. Oolong tea is mainly produced in Formosa (former name for the island of Taiwan).

(e) CTC Tea- At present, methods of tea processing are very much reduced e.g., (a) crushing, (b) tearing and (c) curling. This process is known as CTC method. In India, about 85% tea is processed in this method widely consumed than green tea. India is the largest producer of CTC tea. The process involves additional steps of aeration, withering leaves for several hours which oxidises the flavonoids as well as darkening the leaves' colour.

CTC tea manufacturing is the contribution of Sir William Ner, Superintendent of Amgoorie Tea Estate. He invented CTC machine in 1930. Method of manufacturing CTC tea started gaining importance during 1950s. CTC tea manufacturing technology involves disruption of the cellular integrity of tea shoots; in doing so, the mixing of substrates,

polyphenols and the enzymes, polyphenols oxidase is facilitated. This results in the initiation of a series of biochemical and chemical reactions with the uptake of atmospheric oxygen and formation of pigmented hot water soluble polyphenolic compounds, characteristic of CTC or Black tea.

The withered leaves are passed through CTC machine, causing severe rupturing of leaf cells. Machine consists of two steel engraved rollers rotating at different speeds in opposite direction (70 and 700 rpm). The leaves are allowed to consecutively pass through 2-3 such machines to achieve rupturing of cells and desired size.

The CTC machine has 3 sets of rollers: (a) first cut (coarse), (b) second cut (fine) and (c) third cut (super fine), after which the leaves are completely rolled. The capacity of CTC machine ranges from 750 to 1000 kg/h.

5.2.3 Uses :

Tea is mainly used as a drink, making an excellent drink that has attractive color, fragrance and taste. However, In addition to being used to drink, tea has many other special uses that could be described as having practical value in daily life. Tea has been part of life for thousands of years and over the years, naturally, many special uses have been developed. Some notable uses of tea are introduced below:

- **Plant Fertilizer** : Used tea leaves still contain organic salts, carbohydrates and other nutrients and if buried in the garden or in a plant pot they will help plants grow and reproduce.
- **Eliminating bad breath** : By gargling with strong tea infusion can eliminate bad breath.
- **Making tea eggs** : To make the eggs, first boil eggs until they are done, then break the shell gently and add tea leaves and others ingredient to the water and continue to simmer, this way the tea flavor will be better absorbed by the eggs.
- **Tea pillow** : Don't throw away used tea leaves, dry them on a wooden board and accumulate over a period of time and they can be used as the filling for a pillow. Tea pillows are an ancient Chinese tradition. It is believed that through the tea's natural aroma and its beneficent qualities, the tea pillow gifts the sleeper with a grounding of one's spirit for a restful sleep, an aid to boosting the immune system, opening of the sinuses, relieving overall tension and prevents cold and/ or flu.

5.3 Summary

Beverage is a liquor meant for drinking. Two types of beverages may be recognised viz., non alcoholic and alcoholic. *Camellia sinensis* or Tea is most important and popular caffeine containing non alcoholic beverage. it contains 13 - 18% tannin, 2-5% theine-an alkaloid identical with caffeine and a volatile oil. It belongs to Theaceae family. The tea plant in wild condition is an evergreen tree attaining a height of 15 - 20m, but under cultivation the plant is maintained as a much branched shrubby bush about 0.6-1.5 m high. In 1856, tea plantation was started in Darjeeling, India. *Camellia sinensis* has two varieties, *bohea* and *assamica*, Depending upon processing techniques tea can be categorized into four major commercial varieties i.e. (a) Black Tea (b) Brick Tea (c) Green Tea (d) Oolong Tea and (e) CTC Tea. In addition to being used to drink, tea has many other special uses i.e. as fertilizer, tea pillow, tea eggs etc.

5.4 Questions & Answers

- i. What is Beverage ?
Answer : See Para 5.1
- ii. Tea belongs to which type of beverage ?
Answer : See Para 5.1
- iii. What is the scientific name of Tea ?
Answer : See Para 5.2
- iv. Describe the processing of green tea ?
Answer : See Para 5.2.3
- v. What is the full form of CTC tea ?
Answer : See Para 5.2.3

Unit 6 □ Oils and Fats

Structure

- 6.0 Objectives**
- 6.1 Introduction**
- 6.2 Groundnut Oil**
 - 6.2.1 Source**
 - 6.2.2 Origin**
 - 6.2.3 Production**
 - 6.2.4 Plant Description**
 - 6.2.5 Classification of Groundnut**
 - 6.2.6 Climatic Requirement**
 - 6.2.7 Soil**
 - 6.2.8 Rotation**
 - 6.2.9 Cultivation**
 - 6.2.10 Harvesting and Yield**
 - 6.2.11 Uses**
 - 6.2.12 Varieties**
 - 6.2.13 Diseases and Insect Pests**
- 6.3 Summary**
- 6.4 Questions & Answers**

6.0 Objectives

- This unit will help you to gain knowledge about the centre of origin, morphology, varieties and uses of Ground nut plant.
- You will able to discuss about climate and soil requirement for ground nut production.

- You will know about some common disease and insect pest of pea nut / ground nut plant with their control measures.

6.1 Introduction

The word 'lipid' is the scientific name given to a wide range of natural compounds based on fatty acids. Oils and fats are an important subsection of lipids differing from one another in whether they are liquid or solid at ambient temperature. Oils are chemical compounds of carbon, hydrogen and oxygen (but the ratio of hydrogen to oxygen is not which remain liquid at ambient temperature (10°C- 20°C). The solid state of oils is termed as fats. Oils obtained from the various parts of plants fall under two main categories viz.(1) Essential or Volatile oils and (2) Fatty or Fixed oils. Essential oils are highly aromatic substances, mostly benzene or terpene derivatives or straight-chain hydrocarbon compounds of intermediate molecular length; they evaporate or volatilize in contact with air and are extracted by distillation or by solvents mainly. Fatty or Fixed oils do not evaporate or volatilize and they can not be extracted by simple distillation method. Fatty oils are composed of glycerine together with fatty acid, these are stored up in many plant parts like seeds, fruits and other organs. Fatty oils are produced by a large number of both tropical and temperate plants.

Fatty oils are of four types viz., (a) Drying oils which form thin elastic film on exposure to air e.g., linseed oil, niger-seed oil, soyabean oil etc; (b) semi drying oils which form a soft film on exposure to air e.g., cotton-seed oil; (c) non-drying oils, this never form a film but remain liquid at ordinary temperature e.g., mustard oil, castor oil, groundnut oil, olive oil etc and (d) fats or 1//OW /s that remain solid or semi-solid at ordinary temperature e.g.. coconut oil, mohua oil etc.

6.2 Groundnut Oil

6.2.1 Source :

Groundnut or Peanut oil is obtained from the seeds of the plant *Arachis hypogea* L belonging to the sub-family Faboideae of the family Fabaceae or Leguminosae.

Arachis hypogea is derived from two Greek words "Arachis" meaning to legume and "hypogea" meaning below ground, referring to the formation of pods in the soil (Fig 6.1).

6.2.2 Origin :

Recent botanical survey has indicated that Brazil in South America is the most likely center of origin of this plant. In India, the Jesuit Father (Missionaries) introduced it in the first half of the 16th century. It was introduced in Gujarat by Shri Padmabhai Patel of Pipaliya village, taluka dhoraji(Rajkot) in 1910 from Tamil Nadu. It is also known as peanut, earthnut, manilla nut, monkey nut, goober, pinda and kingpin of oilseeds, unpredictable legume and energy capsule.

6.2.3 Production :

World production of groundnut reached a record of about 21 million tonnes. The most important groundnut producing countries in the world are India, China, USA, West Africa, Sudan, and Nigeria etc. India ranks first in the world in area (8.5 million hectares contributes about 40 % of the total world's area) and production (8.4 million tonnes contributes about 33 % of the total world's production). The average productivity of India is only about 988 kg/ha as against 2995 kg/ha in USA, 2688 kg/ha in China, 1379 kg/ha in Brazil, 1360 kg/ha in Indonesia and 1145 kg/ha in Nigeria.

6.2.4 Plant Description :

Arachis hypogea is a prostrate much branched annual herb. Leaves are pinnate compound, stipulate. Roots are of both tap and adventitious types. Flowers in dense axillary spikes, sessile or shortly pediceled-generally two types of flowers e.g., sterile and fertile types are developed. Former is with yellow-coloured papilionaceous corolla and latter is without corolla. Stamens 10. Carpel one, ovary sessile, 2-3 ovuled, style long and filiform. Fruit a thick, oblong, reticulate lomentum, burying itself to ripen underground. Seeds 1-3, irregularly ovoid; cotyledons thick, fleshy.

6.2.5 Classification of Groundnut :

Groundnut is classified as follows:

1) **According to Growth habits.**

- a) The erect or bunch type: include *Arachis hypogea* sub species *fastigiata* short duration (95-105), early maturing, and high yielding and almost free from dormancy, high germination percentage (90-95).
- b) The spreading or trailing type: include *Arachis hypogea* sub species *procumbens*. Lal. Long duration (110-120 days), late maturity, high yielding

ability and have dormancy (60-75%), low germination per cent (85-90).

2) **Commercial classification of groundnut**

- a) **Coromandal type:** It bears smaller pods, with thin husk, Oval shape and smaller seeds.
- b) **Big Japan or Bombay bold type:** The pods and seeds of this type are bigger than the coromandal type.

6.2.6 Climatic Requirement :

Groundnut is a tropical plant that requires a long and warm growing season. It may be cultivated both in tropical and sub-tropical climates and up to an elevation of about 1,060 metre. It grows well in areas receiving 50 to 125 cm of well-distributed rainfall during growing season, abundance of sunshine and relatively warm temperature. Soil temperature is an important factor and its effects are critical on seed germination, emergence of seedlings, early plant growth, rate of flowering, and pod development. When soil temperature goes below 19 °C, emergence of seedlings is low. The optimum temperature for vegetative growth of groundnut is ranging in between 26 to 30 °C depending on the cultivar.

6.2.7 Soil :

The crop grows best in loam, sandy loam and well-drained black soils. Groundnut is sensitive to soil salinity. It gives good yields in the soil having the pH between 6.0 to 7.5, well supplies with calcium and a moderate amount of organic matter.

6.2.8 Rotation :

Groundnut may be grown alone or in rotation with wheat, jowar, bajra, paddy, cotton and also with garden crops. It is also cultivated as an inter-crop with cotton, castor, millets etc.

6.2.9 Cultivation :

In India, groundnut is generally sown in four seasons

- a) **Kharif:** About 85 % of the total groundnut in India is sown in the kharif season under rainfed conditions.
- b) **Rabi:** Groundnut is grown in rabi season in a limited area where winter is not severe and night temperature does not go below 15 °C. This crop is usually

cultivated in rice fallow field and utilizes the residual moisture after harvest of rice or with minimal irrigation (required 5-9 approx). Rabi groundnut is raised on coastal regions, river deltas of Krishna, Godavari and Kaveri and other irrigated areas in part of Tamil Nadu, Andhra Pradesh and Karnataka.

- c) **Summer:** Summer cultivation of groundnut is mainly taken in the states of Tamil Nadu, Andhra Pradesh, Karnataka, Gujarat and Maharashtra states (required 9-12 irrigations).
- d) **Spring:** Spring cultivation of groundnut is taken in the states of Uttar Pradesh, Punjab and West Bengal etc. during March to May.

6.2.10 Harvesting and Yield :

The prominent symptoms of maturity are yellowing of foliage, spotting of leaves and dropping of old leaves. When the basal leaves of the plant turn yellow and start shedding of leaves, the crop is harvested. The bunch variety is harvested by pulling out the entire plant by hand while the spreading variety is harvested by digging the soil. Then pods are immediately stripped from the harvested plants, dried in the sun and finally stored as unshelled nuts they are then despatched to the market.

In India, it is grown in an area of about 85 lakh hectares with the total production of 84 lakh tonnes. Its cultivation in India is mainly confined to the States of Tamil Nadu, Gujarat, Andhra Pradesh, Karnataka, Maharashtra, Madhya Pradesh, Uttar Pradesh, Rajasthan, Punjab and Orissa. About 80 % of the total area and 84 % of the total production in the country are confined to first five States. The highest productivity of groundnut (1604 kg/ha) is in the State of Tamil Nadu.

The average yield of unshelled pods of spreading variety is 543.5-640 kg per acre while that of bunch variety 362.4-453 kg per acre. The average yield of seeds i.e., kernel by pods varies from 70-75% by weight. The oil content of the seed varies from 44-50%.

6.2.11 Uses :

Groundnut oil is extensively used for cooking purposes and also in the manufacture of hydrogenated vanaspati, margarine, soap and toilet products. The seeds are also used for roasting or salting and for the preparation of peanut butter. Seeds are rich in vitamin A, B and proteins the calorific value is 349 per 100 gms. The oil-cake is used as a fertilizer and as a cattle feed.

6.2.12 Varieties :

Followings are the important and recommended varieties for cultivation in different states of India–

West Bengal- AH 25, B-30, B-31, J-11, Pollachi 1.

Andhra Pradesh-TMV 2, TMV 3, Spanish peanut 5 Madhya Pradesh-AK10, AK 12-24, Improved Small Japan, Improved Spanish.

Bihar-Big Japan, Spanish peanut, AK 12-24, TMV 1.

Karnataka-HG 1, HG 7, HG 9, Pondichery 8.

Maharashtra-Spanish improved, Kopergaon 1, Kopergaon 3, Spanish peanut 5, AK 10 etc.

Rajasthan-RS 1

Tamil Nadu-TMV 1,2,3,4

Uttar Pradesh-Type 25, Type 28, Type 100.

6.2.13 Diseases and Insect Pests :

Common diseases are:-

- **Tikka disease** caused by *Mycosphaerella personata*-it can be controlled by spraying the crop with 1% Bordeaux mixture or dusting with sulphur.
- **Rosette and mosaic diseases** due to virus-control measure is not yet known.
- **Collar rot** caused by *Pellicularia rolfsii* can be controlled by disinfecting soil with chloropicrin and growing resistant varieties.
- **Irregular leaf spot** caused by *Mycosphaerella arachidicola* control by crop rotation, growing resistant varieties and by spraying with 1% Bordeaux mixture.

Common insect pests are-

- Aphids (*Aphis laburni*) and Red hairy caterpillar (*Amsacta albistriga*)-they can be controlled by dusting with 10% BHC or Endrin at 0.012% concentration in the early stage. Dusting of Toxaphene and spraying Folidol or Endrin also give good result.
- Stem borer (*Sphenoptera perroteti*) is another important insect pest, it can be controlled by removing and destroying infested plants.

6.3 Summary

Groundnut oil is non-drying oil derived from *Arachis hypogea* L. (Groundnut). It is a prostrate much branched annual herb. Recent botanical survey has indicated that Brazil in South America is the most likely center of origin of this plant. India ranks first in the world in case of cultivation area and production (33% of the total world's production). Groundnut is classified as growth habit or commercial type. Groundnut is a tropical plant and grows best in loam, sandy loam and well-drained blank soils (pH range 6.0-7.5). About 85% of the total groundnut in India is sown in the kharif season under rainfed conditions. The prominent symptoms of maturity are yellowing of foliage, spotting of leaves and dropping of old leaves. Groundnut oil is extensively used for cooking purposes. Seeds are rich in vitamin A, B and proteins and used for roasting or salting and for the preparation of peanut butter. The oil-cake is used as a fertilizer and as a cattle feed. Tikka disease, Rosette and mosaic diseases, Collar rot are some common diseases found in ground nut. Common insect pests of this plant are aphids and stem borer.

6.4 Questions & Answers

- i. What is the scientific name and family of ground nut ?
Answer : *Arachis hypogea* L. family- Fabaceae or Leguminosae.
- ii. Which country ranks first in the World for ground nut production ?
Answer : India.
- iii. Classify the ground nut as per commercial classification.
Answer : See Para 6.2.5
- iv. What is the range of soil pH for good yields of ground nut ?
Answer : See Para 6.2.7
- v. Mention two varieties of ground nut cultivated in West Bengal.
Answer : See Para 6.2.12
- vi. What is the causal organism of Tikka disease ? How it can be controlled ?
Answer : See Para 6.2.13
- vii. What are the types of fatty oils ?
Answer : See Para 6.1

Unit 7 □ Fibre Yielding Plants

Structure

- 7.0 Objectives
- 7.1 Introduction
- 7.2 Cotton (Karpas, Kapas)
 - 7.2.1 Varieties or Kinds of Cotton:
 - 7.2.2 Morphology
 - 7.2.3 Uses of Cotton
- 7.3 Summary
- 7.4 Questions & Answers

7.0 Objectives

- After reading this unit you will acquire knowledge about morphology, varieties and uses of fibre-yielding plant cotton.

7.1 Introduction

Fibre-yielding plants stand second to food plants in their usefulness to human beings. Next to food and shelter, man needed some form of clothings made from the fibres of different parts like stems, leaves, fruits and roots of many plants. From the inception of civilization, plant fibres have had a more extensive use than animal fibres (eg. silk, wool and fur). Use of vegetable fibres for various purposes has also been increased gradually. There are numerous species of fibre-yielding plants, of which the greater number comprising native species used for various purposes by local people in all parts of the world. As per the origin of fibres, fibres may be bast fibres, wood fibres, sclerenchyma cells associated with vascular strands in leaves, surface fibres developing as hair-like outgrowths on the seeds etc. There are lots of fibre-yielding plants belonging to different families; of these more important families are Agavaceae, Bombacaceae, Bromeliaceae, Liliaceae, Leguminosae, Linaceae, Malvaceae, Moraceae, Musaceae, Palmae (Arecaceae) Tiliaceae, Urticaceae, etc.

Hill (1952) has classified fibres into the following six groups, on the basis of their utilization :

- (a) Textile fibres: Used in textile industry in the manufacture of fabrics, netting and cordage. Textile fibre is classified into 3 sub-classes:
 - i. Surface fibre- **Example:** *Gossypium arboretum*, *G. herbaceum*, *G. hirsutum*, *G. barbadense* (Malvaceae).
 - ii. Soft fibre- **Example:** *Linum usitatissimum* (Linaceae). *Corchorus capsularis*, *C. olitorius* (Tiliaceae).
 - iii. Hard fibre- **Example:** *Agave sisalina* (Agavaceae).
- (b) Brush fibres- These are tough and stiff fibres, used in the manufacture of brushes and brooms. **Example:** *Cocos nucifera* (Arecaceae).
- (c) Planting and Rough weaving fibres- These are flat, pliable fibrous strands which are used in making baskets, chair seats, straw hats, sandals etc. **Example:** *Bambusa tulda* (Poaceae).
- (d) Filling fibres- These fibres are used for stuffing cushions, mattresses. **Example:** *Bambusa ceiba* (Bombacaceae).
- (e) Natural fabrics- These are bast fibres of plants which are used sometimes as substitutes for cloth or lace. **Example:** *Antiaris toxicaria* (Moraceae).
- (f) Paper making fibres- Generally these are wood fibres and textile fibres used in either the raw or manufactured state. **Example:** *Cyperus papyrus* (Cyperaceae), *Bambusa sp.* (Poaceae).

Fibres are mostly long, thick-walled sclerenchyma cells with pointed ends, differ in texture, strength and chemical composition. Cell walls contain cellulose and lignin.

7.2 Cotton (Karpas, Kapas)

The word 'cotton' has been derived from the Arabic term 'qutn'. It is the oldest and greatest industrial crop of the world. Since 1800 BC cotton has been used in India. Cotton was introduced in Europe from India by Arabians. Main cotton growing countries are India, USA, USSR, China, Pakistan, Brazil, Egypt, Turkey, Mexico and Sudan. In India it is grown in over 8 million hectares producing 7.7-7.9 million bales of lint. Large scale cultivation of cotton is practiced in Maharashtra, Gujarat, Karnataka, Madhya Pradesh, Punjab, Rajasthan, Uttar Pradesh, Haryana and Tamil Nadu.

7.2.1 Varieties or Kinds of Cotton:

- Scientific Name: *Gossypium* sp.
- Family: *Malvaceae*.

Cotton is the surface fibre obtained from four major species of *Gossypium*. Fine fibrous hairs occur on the surface of the seeds. These fibrous hairs constitute the raw materials of the industry. Hairs are flattened, twisted and tubular which compose the staple, lint or floss-the length and qualities of which vary in different species. Four species of cultivated *Gossypium* are:

1. ***Gossypium arboreum* L:** This species is the desi cotton of India (Diploid, $2n=26$ chromosomes). It is also grown in Arabia and Africa. The staple is coarse, very short (about 9.5mm - 19.05 mm in length) but strong. This cotton was probably the first to be used commercially.
2. ***Gossypium herbaceum* L:** It is the chief cotton of Asia and also forms desi cotton (diploid, $2n=26$ chromosomes). In India, this species has been growing since time immemorial. It is utilized for low quality fabrics, carpets, blankets etc and for blending with wool; cottons obtained from this species are generally coarse and short to medium stapled.
3. ***Gossypium herbaceum* L:** A tetraploid ($2n=52$ chromosomes) and new world species, native of America and constitutes American and Cambodia cotton and is commonly called upland cotton. Upland cotton constitutes the greater part of the cultivated cotton of the world.
4. ***Gossypium arboreum* L var *nadam* (Watt) Prokh (Syn *G barbadense* L):** It is also a new world tetraploid ($2n=52$ chromosomes) species. The native home is probably South America. There are two distinct types of cotton, under this species.
 - a. **Sea-island Cotton**—Purely cultivated form. Sea-island cotton was introduced in USA from West Indies in 1785. In India, another variety under this, known as **Andrews variety** having extra long staple can be cultivated successfully in the coastal areas of Kerala and Karnataka.
 - b. **Egyptian Cotton**—It is grown mainly in Nile basin of Egypt, and was introduced from Central America.

7.2.2 Morphology :

- ***Gossypium arboreum* L:** Plant is a perennial tree. Leaves palmately lobed, lobes mucronate. Flowers axillary, large, solitary on jointed peduncles; bracteoles 3, large, leafy, cordate. Calyx cupular, truncate or slightly 5-toothed. Petals 5, connate at base. Stamens numerous, monadelphous forming a staminal column, anther one celled. Ovary 5-chambered, style clavate, 5-grooved at the apex, stigmas 5; ovules many in each chamber. Fruit capsule, oblong, pointed. Seeds free, covered with white wool overlying a dense green down (Fig. 7.1).

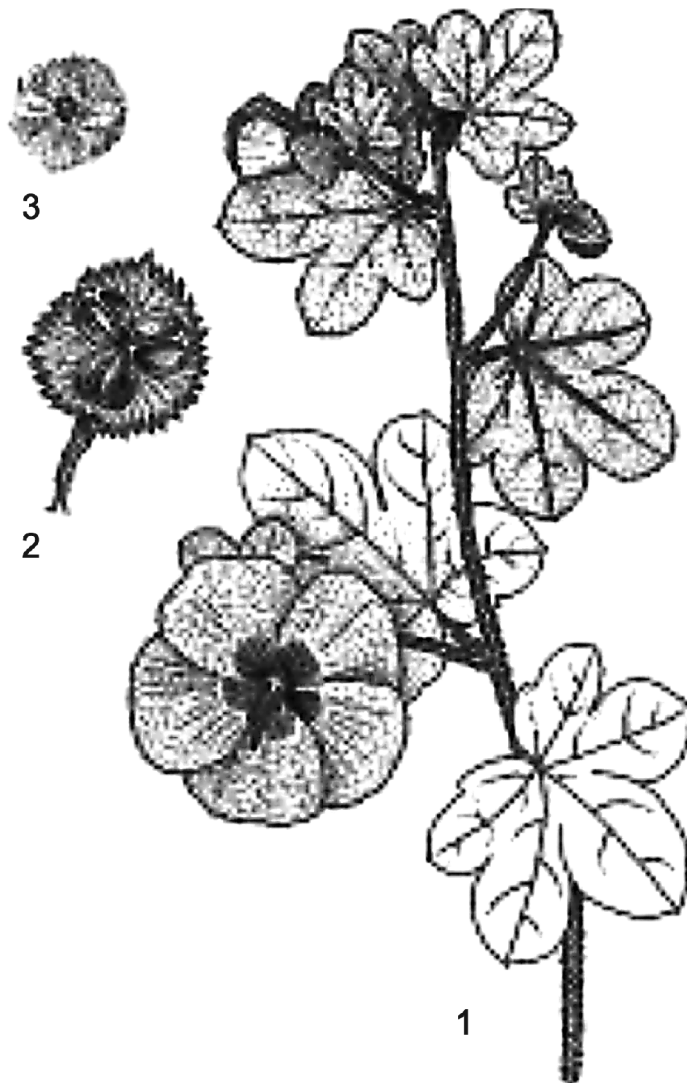


Fig. 7.1: 1- Flowering twig of *Gossypium* sp. 2- Dehiscent fruit. 3- Seed.

- ***Gossypium herbaceum* L:** Plant is an annual shrub, sub-glabrous or hairy. Leaves cordate, 3-5 or 7-lobed, lobes broadly acuminate. Flowers yellow with purple centre, rarely yellow or white or purple, petals spreading, calyx truncate, obovate or cuneate; bracteoles equalling the capsule, not divided below the middle. Capsule ovate, globose, mucronate, 3-5 valved. Seeds 5-7 in each cell, ovoid. Cotton white, rarely brownish, overlying a greenish or grayish brown.
- ***Gossypium hirsutum* L:** The fibres are fine and white, staple length ranges from 6.2 mm to 25.7 mm. This species is characterized by the following characters. Flowers are white or light yellow and unspotted. Fruits or bolls as these are called, are 4 or 5 valved. Seeds are fuzzy all over. Other characters are like those of *G. arboreum*.
- ***Gossypium arboreum* L var *nadam* (Watt) Prokh (Syn *G. barbadense* L.)**
 -
 - (a) **Sea-island Cotton-** Fibres are fine, light cream coloured, strong, regular in the number and uniformity of twist and silkier in appearance. Staples range in length from 38.0 mm to 50.8 mm or more. This variety is susceptible to a large number of pests and diseases and very sensitive to environmental conditions.
 - (b) **Egyptian Cotton-** In appearance, this plant is very similar to sea-island cotton and is probably of hybrid origin. Staples are brown in colour and somewhat shorter than sea-island cotton, measuring from 25.7 mm to 26.1 mm. Plants are perennial shrubs or tall herbs, branches purplish. Leaves nearly glabrous, cordate, 3-5 lobed; lobes oblong acuminate, bracteoles very large, deeply gashed; stipules linear-lanceolate. Flowers are bright yellow with purple spots. Petals convolute, Ovary ovoid, pitted, 3-5 celled. Fruits or bolls are oval acuminate, 3-valved. Seeds are fuzzy only at the ends, black, free or coherent. This cotton is used for hosiery, tyre fabrics and fine dress materials.

7.2.3 Uses of Cotton :

Cotton has played an important role in human history ever since it was first used, which anthropologists date back to prehistoric times. Cotton is extremely versatile, soft,

and sturdy and the most widely used fiber in the world.

- **Fabrics**

Many of the fabrics commonly seen on clothing labels originated from cotton fibers. Cotton can be woven or knitted into denim, velvet, corduroy, jersey, flannel, velour, and chambray. Cotton and related fabrics are used to make just about everything in the apparel industry – from leisure wear to underwear to socks and t-shirts. They are also used to make bed linens, comforters, and blankets.

- **Baby Care & Feminine Care (Nonwovens)**

For many years, nonwoven products in baby care and feminine care featured man-made fibers almost exclusively. But as time passed, consumers demanded next-to-skin topsheets made from natural fibers that were softer and hypoallergenic. Now, an increasing number of many high-quality diapers and baby wipes are made from soft, breathable cotton, the perfect match for a baby's delicate skin.

Cotton is also a common ingredient in extra-gentle lotions, body washes, and shampoos designed for babies. In feminine care, too, cotton is often seen in pure, natural products. High-quality hygiene pads, panty liners, and tampons may be made from 100% cotton, both conventional and organic, or in some cases a blend of cotton with synthetic materials.

- **Other Consumer Products**

Cotton can be easily processed into a number of products that we use on a daily basis, like coffee filters, book binding, paper, and bandages. Cottonseed oil, which is made from crushed seeds of cotton plants, is used in a multitude of products including soap, cosmetics, and margarine.

Cotton is also an important component of many products used for travel and recreation, including tents and tarpaulins, fishing nets, cords, and ropes.

- **Agriculture and Industrial Applications**

Cotton is a food and fiber crop. Cotton seed is often fed to cattle and horses as a healthy source of protein. Humans can't digest the cellulose found in cotton,

but animals have a special enzyme that breaks it down.

Even stalks and leaves from the cotton plant can be made useful; stalks are ploughed underground to enrich soil, and fiber extracted from them is used to make pressed paper and cardboard.

Additionally, cotton serves a function in just about every industry, from pharmaceuticals to rubber and plastics. Cotton linters (the short fibers that remain on the plant after it is ginned) are used in x-rays, swabs, and cotton buds in the medical field, as well as in cleanroom suits and supplies. Cotton linters are also used in mattresses, furniture, automobile cushions, and even flat screen televisions.

7.3 Summary

Plant fibres have had a more extensive use than animal fibres from the inception of civilization. As per the origin, fibres may be bast fibres, wood fibres, sclerenchyma cells associated with vascular strands in leaves, surface fibres developing as hair-like outgrowths on the seeds etc. On the basis of utilization, Hill (1952) has classified fibres into six groups i.e. Textile fibres, Brush fibres, Planting and Rough weaving fibres, Filling fibres, Natural fabrics and Paper making fibres. Cotton is the oldest and greatest industrial crop of the world and used as textile fibres. Since 1800 BC it has been used in India. It was introduced in Europe from India by Arabians. Cotton is extremely versatile, soft, and sturdy surface fibre obtained from four major species of *Gossypium* (family-Malvaceae). It is most widely used fiber in the world.

7.4 Questions & Answers

- i. What is the scientific name of Cotton plant ?

Answer : *Gossypium sp.*

- ii. Briefly describe different varieties of Cotton.

Answer : See para 7.2.1

- iii. Write a short note on Egyptian cotton.

Answer : See para 7.2.2

iv. What are the uses of cotton ?

Answer : See para 7.2.3

v. Classify the fibres as per Hill (1952).

Answer : See para 7.1

MODULE 2
PLANT BIOTECHNOLOGY

Unit 8 □ Introduction to Biotechnology

Structure

- 8.0 Objectives**
- 8.1 Introduction**
- 8.2 Objectives of Biotechnology**
- 8.3 Biotechnology : Definition, Branches**
 - 8.3.1 Branches of Biotechnology**
 - 8.3.1.1. Medical Biotechnology (Red)**
 - 8.3.1.2. Agricultural Biotechnology (Green)**
 - 8.3.1.3. Industrial Biotechnology (White)**
 - 8.3.1.4. Food Biotechnology (Yellow)**
 - 8.3.1.5. Environmental Biotechnology (Gray)**
 - 8.3.1.6. Marine Biotechnology (Blue)**
 - 8.3.2 Application**
- 8.4 Summary**
- 8.5 Questions & Answers**

8.0 Objectives

- This unit will help you to know about the definition, different branches & applications of biotechnology.

8.1 Introduction

“Biotechnology”, as the word, is combination of biology and technology. Biotechnology is diverse field which involves either working with living cells or using molecules derived from them for applications oriented towards human welfare using different types of tools and technology. It has also impacted the environment which is loaded by toxic compounds due to human industrialization and urbanization.

People were using biotechnology techniques thousand years but they did not name their working field as biotechnology. The name 'Biotechnology' was given by Hungarian engineer Karoly Ereky in 1919. In early days, selectively bred and microorganism were used to make beer, wine, cheese, and bread. However, the field gradually evolved, and presently it is the use or manipulation of living organisms to produce beneficiary substances which may have medical, agricultural and/or industrial utilization. Modern biotechnology deals with the technique that makes use of cellular molecules like DNA, monoclonal antibodies, biologics, etc.

8.2 Objectives of Biotechnology

The main objectives of biotechnology :

- 1) To develop industrial processes for production of antibiotics, enzymes etc.
- 2) To develop gene surgery and gene therapy to cure genetic disease.
- 3) To create improved varieties of plants and animals through genetic engineering and plant breeding.
- 4) To develop techniques for tissue culture, cell culture and organ transplantation.
- 5) To develop bioenergy.
- 6) To develop biological processes of waste treatment to reduce the impact of pollution.
- 7) To develop biological process of plant disease control.

8.3 Biotechnology : Definition, Branches

Biotechnology is defined as :

- 1) "Biotechnology is the application of biological organisms, system or process to manufacturing and service industries." (British or processes to manufacturing and Biotechnologist)
- 2) "Biotechnology is the controlled use of biological agent, such as microorganisms or cellular components." (US National Science Foundation)
- 3) "Biotechnology is the integrated use of biochemistry, micro-biology and

engineering sciences in order to achieve technological (industrial) application of the capabilities of microorganism cultured tissue cells.” (European Federation of Biotechnology)

8.3.1. Branches of Biotechnology :

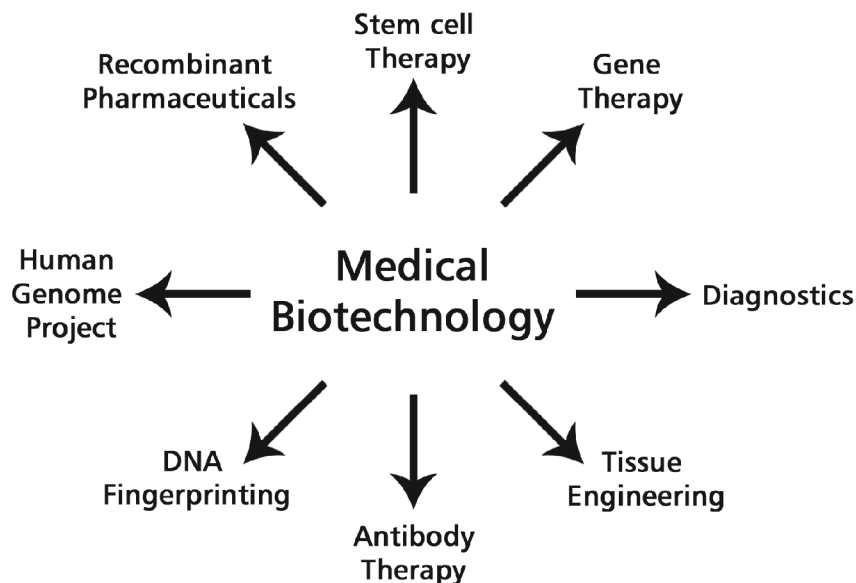
The main branches of biotechnology are –

- Medical (Red) Biotechnology
- Agricultural (Green) Biotechnology
- Industrial (White) Biotechnology
- Food (Yellow) Biotechnology
- Environmental (Gray) Biotechnology
- Marine (Blue) Biotechnology

In this chapter we come to know about the potential applications of biotechnology in several fields like production of medicines; diagnostics; therapeutics; agricultural biotechnology; pollution control; industrial and marine biotechnology.

8.3.1.1. Medical Biotechnology : (Red)

This field of biotechnology has many applications and is involved in production of recombinant pharmaceuticals, tissue engineering products, regenerative medicine such as



stem cell and gene therapy, and many more biotechnology products for human life. Genetic testing allows the genetic diagnosis of vulnerabilities to inherited diseases, and can also be used to determine a child's parentage or in general a person's ancestry.

Many bio-therapeutic agents in clinical use are biotech pharmaceuticals. Insulin was among the earliest recombinant drugs. Canadian physiologists Frederick Banting and Charles Best discovered and isolated insulin in 1921. The first "Bioengineered" drug, a recombinant form of human insulin, was approved by the US Food and Drug Administration (FDA) in 1982. The first recombinant vaccine, approved in 1986, was produced by cloning a gene fragment from the hepatitis B virus into yeast (Merck's Recombivax HB). This avoided the need to extract the antigen from the serum of people infected with hepatitis B. Stem cell research is very promising and potential to treat neurodegenerative disorders, spinal cord injuries, and other conditions related to organs or tissue loss. Biotechnological techniques also improves diagnosis of diseases and therapeutic capabilities.

8.3.1.2. Agricultural Biotechnology : (Green)

The man has made tremendous progress in crop improvement in terms of yield; still the ultimate production of crop is less than their full genetic potentials. There are many reasons like environmental stress, disease, pests, and many other factors which reduce the ultimate desired yield affecting crop productivity. Agricultural biotechnology aims to introduce sustainable agricultural practice to with best yield potential and minimal adverse effects on environment.¹

¹* For example, to combat pests the gene from bacterium, the BT gene, that functions as insect-resistance gene when inserted into crop plants like cotton, corn, etc. helps prevent the invasion of pathogen. This management is helpful in reducing usage of potentially dangerous pesticides on the crop. Resistance to infectious agents through genetic engineering—

- 1) Bt crops
- 2) Protection against nematodes
 - Agricultural biotechnology has the potential to produce –
 - ❖ High crop yields
 - ❖ Crops are engineered to have desirable nutrients and better taste.
 - ❖ Insect and disease resistant plant.

- ❖ Genetic modification avoids nonselective changes.
- ❖ Longer self life of fruits and vegetables.

8.3.1.3. Industrial Biotechnology : (White)

Industrial biotechnology is the application of biotechnology for industrial purpose, including industrial fermentation. It includes the practice of using cells such as micropropagation, or components of cells like enzymes to generate industrially useful products such as chemicals, foods, detergents, paper and pulp, textiles and biofuels.

The aim of Industrial biotechnology is to develop newer industrial manufacturing processes and products, which are economical and better than preexisting ones with minimal environmental impact. In industrial biotechnology,

- 1) microorganisms are being explored for producing material goods like fermentation products as cheese;
- 2) biorefineries where oils, sugars, and biomass may be converted into biofuels, bioplastics, and biopolymers;
- 3) and value - added chemicals from biomass.

8.3.1.4. Food Biotechnology : (Yellow)

Food biotechnology is an emerging field, which can increase the production of food, improving its nutritional content and improving the taste of the food. The food is safe and beneficial as it needs fewer pesticides and insecticides. The technology aims to produce foods which have more flavour, contain more vitamins and minerals and absorb less fat when cooked.

8.3.1.5. Environmental Biotechnology : (Gray)

Environmental biotechnology focused on waste treatment, monitoring of environmental changes, and pollution prevention. Bioremediation is the main application of environmental biotechnology. Microorganisms are widely used to reduce environmental pollution by acting on sewage wastage. The production of biofuel like biogas from waste can resolve the fuel crisis. Microbs may be engineered to produce enzymes required for conversion of plant and vegetable materials into building blocks for biodegradable plastics. Beside these helpful approaches biotechnology has some negative effect on environment. It can disturb the ecological balance and natural interaction of organisms. It may threatens biodiversity on a wider level.

8.3.1.6. Marine Biotechnology : (Blue)

Marine biotechnology is also referred as “blue biotechnology”, deals with exploring and utilizing the marine resources of the world. This field aims to –

- Fulfill the increasing food supply needs.
- Identify and isolate important compounds which may benefit health of humans.
- Manipulate the existing traits in sea animals for their improvement.
- Protects marine ecosystem and gain knowledge about the geochemical processes occurring in ocean.

Major applications :-

- 1) Aquaculture
- 2) Biotechnology discoveries and products like –
 - a) Transgenic species of salmon with growth hormone gene has accelerated growth of salmon.
 - b) MIH (Molt-inhibiting) from blue crab.
 - c) Antifreeze protein (AFP) was identified and isolated from Northern cod.
 - d) Green fluorescent protein obtain from jelly fish *Acquorea victoria*.
 - e) Salmon calcitonin for osteoporosis.
 - f) Hydroxyapatite (HA) obtained from coral reefs, an important component of bone and cartilage matrix.
 - g) Byssal fibers, protein-rich super adhesive obtained from *Mytilus edulis*
 - h) Many anti-inflammatory, analgesic, anticancerous compounds have be identified from sea organisms.

8.3.2 Application :

The biotechnological advancement have resulted in–

- 1) Many biopharmaceuticals and vaccines,
- 2) New and specific ways to diagnose,
- 3) Increasing the productivity and introduction of quality traits in agricultural crops,
- 4) The ways to tackle the pollutants efficiently for sustainable environmental practices,

- 5) Helped the forensic experts by DNA finger printing and profiling,
- 6) Fermentation technology for production of industrially important products,
- 7) Food industries. Production of SCP, spirulina, enzymes.

8.4 Summary

- Biotechnology is the production of interaction between biological science and modern technology.
- During last two decades biotechnology has achieved greatest advancements in the fields of recombinant DNA technology, PCR, gene cloning, DNA finger-printing etc.
- The technology holds promise through stem cell research and gene therapy and holds applications in forensic medicine.
- It is also helpful in maintaining environment by bioremediation and other treatment.
- Biotechnology is enabling industries to make new or better products, often with greater speed, efficiency and flexibility.

8.5 Questions & Answers

1) What is Biotechnology ?

Ans : Cellular and Biomolecular processes to develop technologies and products that help improve our lives and the health of our planet.

2) Who coined the term 'Biotechnology' ?

Ans : The term 'Biotechnology' was coined by a Hungarian engineer Karl Ereky in 1919.

3) What are the branches of Biotechnology ?

Ans : View 8.3.1

4) What are applications of Biotechnology in maintaining Environment?

Ans : View 8.3.1.5

5) Give some applications of Biotechnology in Agriculture.

Ans : View 8.3..1.2

6) What is meant by Bioremediation ?

Ans : Bioremediation is a branch of biotechnology that employs the use of living organisms, like microbes and bacteria, in the removal of contamination, pollutants, and toxins from soil, water and other environments.

7) What are Bt-crops ?

Ans : Bt-crops are transgenic crops that produce the same as the bacterium *Bacillus thuringiensis* in the plant cell, thereby protecting the crops from pests. The bacterium secretes specific proteins known as cry proteins that are toxic to insects. Eg. - Bt cotton, Bt brinjal.

8) Write two application of Medical Biotechnology.

Ans : a) Enabling mass production of safe and more effective therapeutic drugs.
b) Gene therapy to treat genetic disorders.

9) Write two application of Biotechnology in Industry.

Ans : a) Improvement in Fermentation Production.
b) Microbial production of synthetic fuels.

10) What is Marine Biotechnology ? Write its importance ?

Ans : View 8.3.1.6

Unit 9 □ Plant Tissue Culture

Structure

- 9.0 Objectives**
- 9.1 Introduction**
- 9.2 Objective of Plant Tissue Culture**
- 9.3 Micropropagation**
 - 9.3.1 Stages of Micropropagation**
 - 9.3.2 Advantages of Micropropagation**
- 9.4 Haploid production through androgenesis and gynogenesis**
 - 9.4.1 Androgenesis**
 - 9.4.1.1. Comparison between anther and pollen culture**
 - 9.4.1.2. Factors affecting androgenesis**
 - 9.4.2 Gynogenesis**
 - 9.4.2.1. Limitations of Gynogenesis**
- 9.5 Embryo and Endosperm culture and their applications**
 - 9.5.1 Protocol of Embryo culture**
 - 9.5.2 Application of Embryo culture**
 - 9.5.3 Steps of Endosperm culture**
 - 9.5.4 Application of endosperm culture**
- 9.6 Application of Tissue Culture**
- 9.7 Summary**
- 9.8 Questions & Answers**

9.0 Objectives

- This unit will help you to acquire knowledge about micropropagation, androgenesis and gynogenesis.

- After reading this unit you will be able to discuss the protocol & application of embryo & endosperm culture. You will also know the different application of tissue culture.

9.1 Introduction

A mature plant cell, unlike an animal cell, retains the ability to differentiate into a whole plant, if it has retained an intact membrane system and a viable nucleus. This capacity of a cell (or a group of cells) to give rise to an entire organism is known as totipotency.

In 1902, the German botanist G. Haberlandt develop the concept of in vitro cell culture and its importance. Unfortunately Haberlandt was unsuccessful to culture cells. The term totipotency coined by Steward in 1968.

The first commercial use of plant tissue culture on artificial media was in the germination and growth of orchid plants, in the 1920's. In the 1950's and 60's there was a great deal of research, but it was only after the development of a reliable artificial medium (Murashige & Skoog, 1962) that plant tissue culture really "took off" commercially. Plant tissue culture refers to growing and multiplication of cells, tissues and organs of plants on solid or liquid media with nutrients under aseptic and controlled environment. The plant tissue or organ excised and used for in vitro culture is known as explant. The plants so grown would be exactly similar to the mother plant in all aspect.

9.2 Objective of Plant Tissue Culture

- 1) A single explant can be multiplied into several thousand plants in less than a year.
- 2) Once established, it can give a continuous supply of young plants throughout the year.
- 3) Disease free plants can be formed by tissue culture techniques.
- 4) This technique is rapid, continuous and efficient.
- 5) Breeding cycle is reduced.
- 6) Germplasm prevention possible.
- 7) This technique is useful in hard to propagate plant.

9.3 Micropropagation

Micropropagation is the rapid, vegetative propagation of plants under *in vitro* conditions of high light intensity, controlled temperature and a defined nutrient medium. Use of tissue culture for micropropagation was started by G. Morel (1960) who used this as a commercially viable approach for orchid propagation.

Here, a large number of miniatures of vegetative shoots are produced from a clone within a short time and space. The products of these rapid vegetative propagation can also be regarded as clones when it is established that the cells are genetically identical.

9.3.1 Stages of Micropropagation –

The process of micropropagation can be subdivided into *four* prominent stages :-

- (i) **Stage 0** – It is the initial step of micropropagation in which stock plants are grown under controlled condition before being used for culture initiation.
- (ii) **Stage 1** – This stage includes *preparation of explant* from stock plants through : *explant isolation ; surface sterilization ; washing ; establishment of explant on appropriate culture medium.*
- (iii) **Stage 2** – This stage involves *multiplication of shoots or rapid somatic embryo formation* using a defined culture medium.

Multiplication may be :

- By Meristematic tissue of axillary and apical shoots
 - By adventitious shoots
 - By adventitious embryo formation
 - Through callus culture
- iv) **Stage 3** – Shoots obtained from stage II are transferred to the next rooting (soil) or storage medium. These shoots are directly established in soil as micro-cuttings to develop roots.
 - v) **Stage 4** – Plantlets are *transferred to sterilized soil* for hardening under greenhouse environment i.e. under controlled condition of light, temperature and humidity. For marketing, sometimes these plantlets are established in an artificial growing medium. It takes 4-16 weeks for marketing of the finished products.

9.3.2 Advantages of Micropropagation –

- Micropropagation is an alternative approach to conventional methods of vegetative propagation.
- Millions of shoot tips can be obtained from a small, microscopic piece of plant tissue within a short period of time and space.
- This method is more helpful in case of bulb or corn-producing plants.
- The propagules can be maintained in soilfree environment which facilitates their storage on a large scale.
- Micropropagation helps in maintenance of germplasm for many years.
- This method of propagation requires minimum growing space e-g. thousands of plantlets can be maintained within culture vials.
- Micropropagation method will help to maintain genetic uniformity in the propagules which is not always possible through seed production.

9.4 Haploid production through androgenesis and gynogenesis

In plant biotechnology programmes, haploid production is achieved by 2 methods :-

- a) **Androgenesis** – This involves haploid production through *anther* or *pollen culture*, and they are referred to as *androgenic haploids*.
- b) **Gynogenesis** – This involves *gynogenic haploid* production through *ovary* or *ovule culture*.

9.4.1 Androgenesis

The principle behind this technique is to arrest the development of male gametophyte (pollen or microspore) into a gamete and force it to form a haploid plant.

Androgenesis may be through *anther culture* or *pollen culture*.

(i) **Anther culture : The Steps include –**

- Selected flower buds of young plants are surface sterilized.
- Anthers are removed alongwith their filaments.

- Anthers are cut under aseptic conditions and crushed in 2% aceto-cormine to test the stage of meiosis.
- Anthers in the proper stage are inoculated on proper media.
- Within few days or weeks (3-8 weeks) the microspore within the anther divides to form either *callus* or *embryo*
- The anther wall ruptures and the embryo or plant tetras are subcultured in proper media to get the whole plant in rooted condition.

(ii) Pollen (microspore) culture :

Microspore or immature pollen can be used as explant to produce haploid plant. The steps include :

- Flower buds are collected, surface sterilized and the anther lobes are dissected out from the flower buds.
- Pollens are extracted by squeezing the anthers.
- Pollen suspension is filtered to remove the anther tissue debris.
- Viable and large pollen are washed and collected.
- Pollen are cultured on a solid or liquid medium.
- The callus / embryo formed is transferred to a suitable medium to produce a haploid plant.

9.4.1.1 Comparison between anther and pollen culture

- Anther culture is easy, quick and practicable.
- They are efficient for haploid production.
- The major limitation is that the plants originate not only from pollen, but also from other parts of anther.
- So the population of plants are at different ploidy levels (e.g. diploids, aneuploids)
- Disadvantages of anther culture can be overcome by pollen culture.

9.4.1.2 Factors affecting androgenesis :

- 1) Genotype of donor plants
- 2) Stage of microspore or pollen

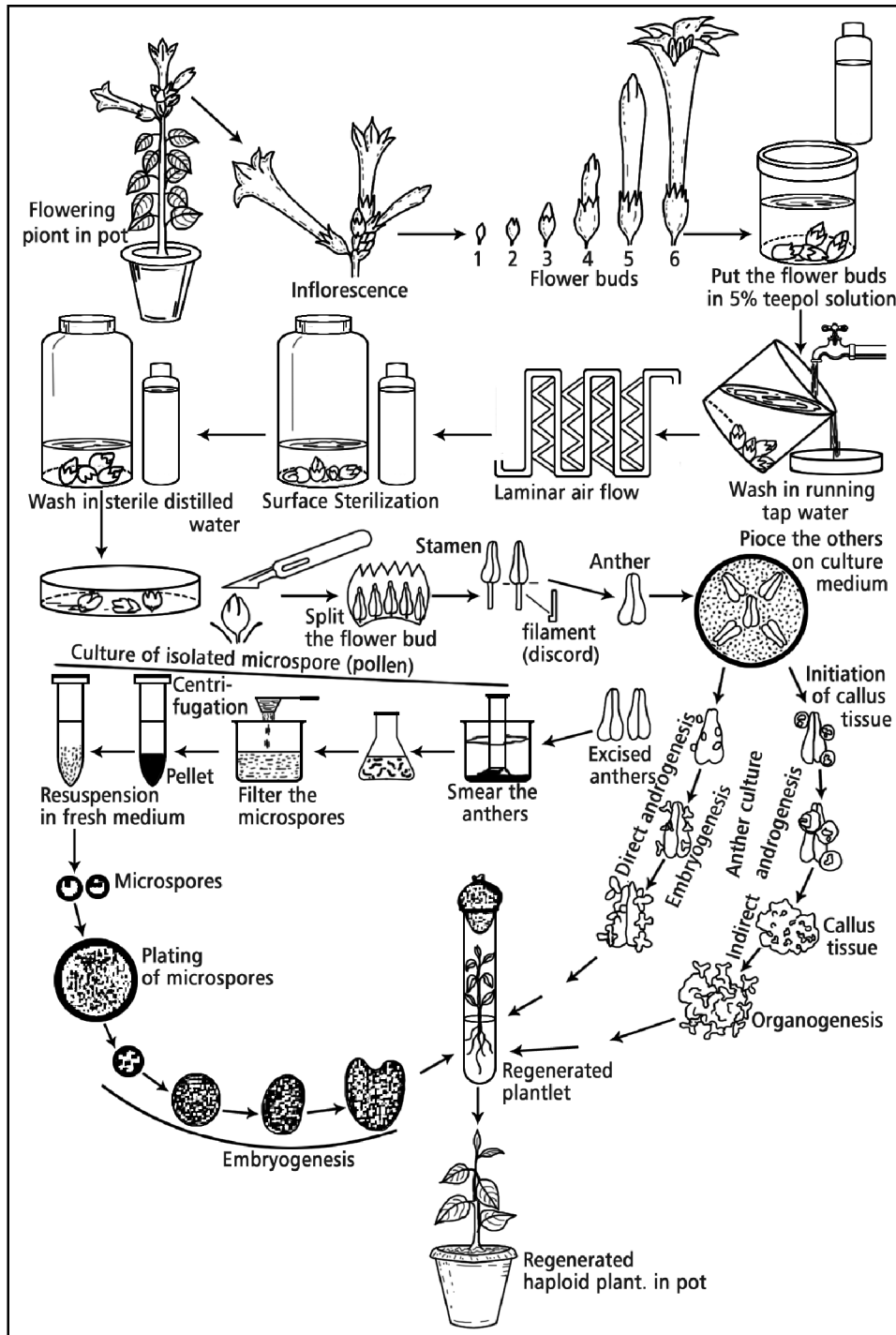


Fig. 9.1 : Schematic representation of the culture of excised anther and isolated microspores and the development of haploid plant directly by embryo formation or through haploid callus.

- 3) Physiological status of the donor plant
- 4) Pretreatment of anthers.

9.4.2 Gynogenesis

It is the process of development of haploid plants from ovary or ovule cultures. The plants so produced are referred to as *gynogenic haploids*. They were first developed by San Noem (1976) from the ovary cultures of *Hordeum Vulgare*. The technique was later applied for raising haploid plants of rice, wheat, maize, sunflower, sugar-beet and tobacco. The flower buds are excised 24-48 hrs prior to anthesis from un-pollinated ovaries. After removal of calyx, corolla and stamens, the ovaries are subjected to surface sterilization. The *ovary is inserted in the solid culture medium*. Whenever a liquid medium is used, the ovaries are placed on a filter paper or allowed to float over the medium with inserted through filter paper. The commonly used media are MS, Whites, N6 and Nitsch, supplemented with growth factors.

9.4.2.1 Limitations of Gynogenesis :

The major limitations of gynogenesis are :

- The dissection of unfertilized ovaries and ovules is rather difficult.
- The presence of only one ovary per flower is another disadvantage.

9.5 Embryo and Endosperm culture and their applications

Embryo Culture is isolation and growth of an immature or mature embryo *in vitro*, with the aim of obtaining a viable plant. The *first attempt* to grow the embryos of angiosperms was made by Hannig (1904) from two crucifers *Cochleria* and *Raphanus* .

Types of embryo culture include :

- *Mature embryo culture*
- *Immature Embryo culture/Embryo rescue.*

Mature embryo Culture :

It is the culture of mature embryos derived from ripe seeds. This type of culture is done when embryos do not survive *in vivo* or become dormant for long periods of time. This culture is done to eliminate the inhibition of seed germination. The medium required is a basal salt medium with a carbon energy source.

Immature embryo culture :

It is the culture of immature embryos to rescue the embryos of wide crosses. This type of culture is mainly used to avoid embryo abortion to produce a viable plant. The medium used requires different vitamins, amino acids and growth regulators.

Embryo rescue – In immature embryo culture, wide hybrids often suffer from early spontaneous abortion due to several barriers which act at pre and post-fertilization levels.

Pre-fertilization barriers include all factors that stop effective fertilization (due to inhibition of pollen tube growth)

Post-fertilization barriers effect the development of the zygote after fertilization and normal development of the seed. (due to failure of endosperm).

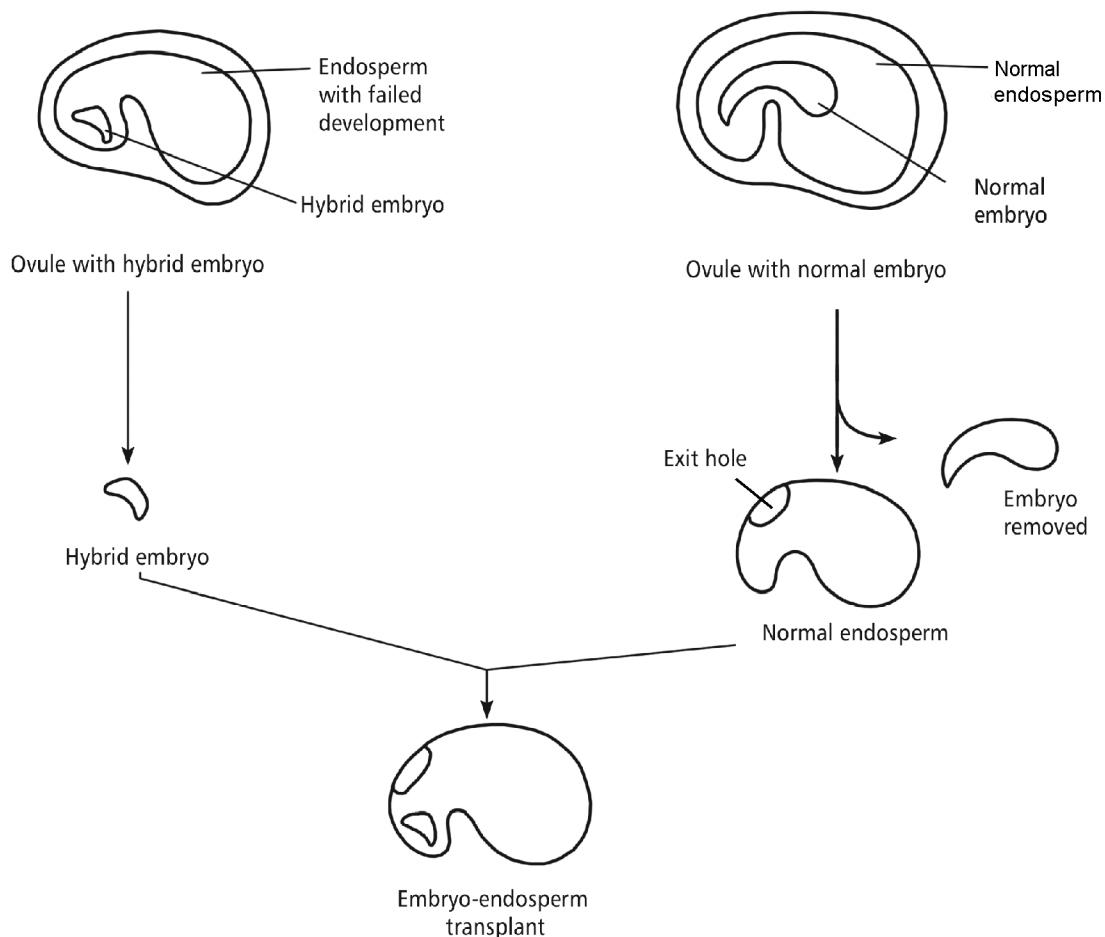


Fig. 9.2 : Embryo-endosperm transplant technique used in embryo rescue.

This is known as *embryo-endosperm incompatibility*. Where the endosperm produces toxins which kill the embryo.

9.5.1 Protocol for Embryo culture

The following protocol is based on the method used for *Capsella bursapastoris*. It is a basic protocol for embryo culture, in general.

Steps :

1. Capsules in the desired stages of development are surface sterilized for 5-10mins. in 0.1% HgCl₂ either in a preirously UV-illuminated closed room or in Laminar air flow.

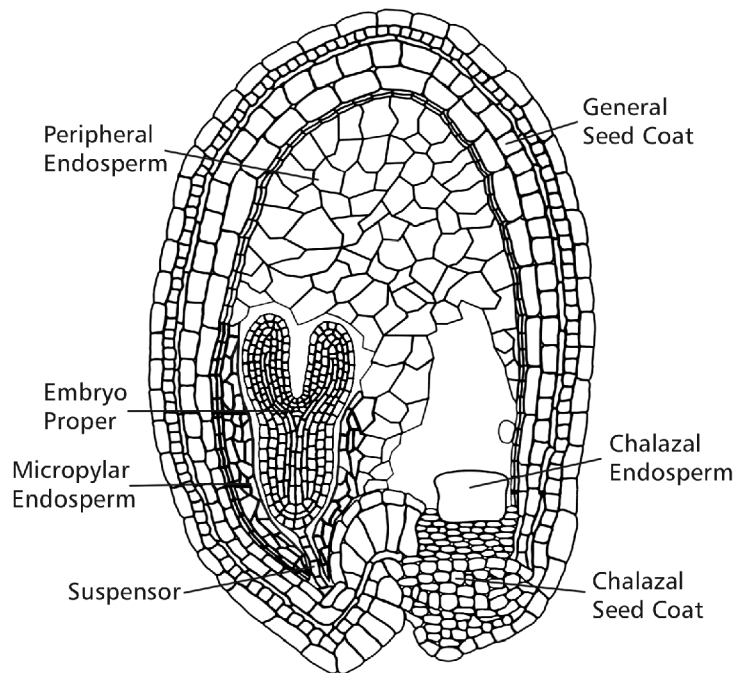


Fig. 9.3 : Structure of Ovule

2. Repeatedly washed in sterile water.
3. The outer wall of the capsule is removed under a dissecting microscope at 90X, and cut in the region of placenta. The ovules are exposed.
4. The embryos are extracted from the ovule.

5. The excised embryos (6-8) are transferred to petridishes containing solid standard medium.
6. Cultures are kept in a culture room at $\sim 25^{\circ}\text{C}$ with 16 hrs illumination.
7. Subcultures into fresh medium are made at approximately 4 weeks interval.

9.5.2 Applications of Embryo culture

- Production of haploids
- Prevention of embryo abortion in wide crosses
- Overcoming seed dormancy
- Shortening of breeding cycle
- Prevention of embryo abortion in stone fruits.

Endosperm Culture :

Endosperm is an unique tissue, because of its function of providing nutrition to the developing embryo and also due to its triploid ($3n$) nature. The endosperm of angiosperms is $3n$, due to the double fertilization (forming $2n$ zygote and $3n$ endosperm). Endosperm lack many organogenic potential and vascular differentiation.

Zampe and Mills (1933) first reported the proliferation of immature endosperm of may grown on medium containing extract of potato.

Nakano et al (1975) successfully cultured immature endosperm of rice and also achieved *organogenesis*.

Thus, *endosperm culture* is the *in vitro* development of isolated mature or immature endosperm from seed at proper stage on a suitable culture medium to obtain triploid plantlet.

Types –

- (1) *Mature endosperm culture* is the *in vitro* development of isolated, mature endosperm from ripen endospermic seed to obtain triploid plantlet.
- (2) *Immature endosperm culture* is the *in vitro* development of isolated, immature endosperm isolated at precise stage from immature seed to obtain triploid plantlet.

9.5.3 Steps of Endosperm culture

It consists mainly of 3 steps –

- The immature or mature seeds are dissected under aseptic conditions and endosperms, along with embryos are excised.
- Excised endosperms are cultured on a suitable medium and embryos are removed after initial stage.
- Callus formation, followed by embryogenesis or shoot bud differentiation.
- Complete plant formation.

9.5.4 Applications of Endosperm culture –

- Help in production of triploid plants which are sterile and *usually seedless*. This increases the edibility of fruits and is a desirable character in banana, apple, grapes, watermelon etc.
- In timber and fuel-yielding plants, triploids show better performance compared to their corresponding diploids or tetraploids. Seed-sterility does not impose a problem as they can be multiplied by vegetative means.

9.6 Application of Tissue Culture

- 1) Clonal Propagation
- 2) Somaclonal Variation
- 3) Production of Virus free plants
- 4) Production of Synthetic Seeds
- 5) Somatic Hybridization
- 6) In vitro plant Germplasm conservation
- 7) Mutation Breeding
- 8) Molecular Farming
- 9) Genetic Engineering
- 10) Production of Secondary Metabolites

9.7 Summary

Tissue culture is one of the most important part of applied biotechnology. In the coming decades the world's population will increase more and accommodation space agricultural lands will decrease significantly. Plant produced via tissue culture are uniform. Plant production is reliable and consistent Multiplication rates are also high. But there are some limitations such as all the plants are genetically similar, there is reduction is genetic diversity. If the precautions are not taken the whole stock, may be contaminated or infected. Tissue culture is of great significance in biological studies due to its wide range of applications. Because of the complexities that may be involved in some of the steps, this may not be an experiment for everyone.

9.8 Questions & Answers

1) What is Plant Tissue Culture ?

Ans. Plant tissue Culture is *in vitro* cultivation of plant cell or tissue under aseptic and controlled environmental conditons, in liquid or on semisolid well defined nutrient medium for the production of primary and secondary metabolites or to regenerate plant.

2) What is totipotency ?

Ans. The capacity of a cell or tissue to give rise to an entire organism is known as totipotency.

3) What is micropropagation ?

Ans. Micropropagation refers to the *in vitro* multiplication and/or regeneration of plant material under aseptic and controlled environmental conditions to produce thousands or millions of plants for transfer to the field.

4) What is embryo genesis ?

Ans. Organogenesis means the development of adventitious organs or prionordia from undifferentiated cell mass in tissue culture by the process of differentiation.

5) What is embryogenesis ?

Ans. The developmental pathway of numerous well-organised small embryoids resembling the zygotic embryos from the embryogenic potential somatic plant cell of the callus tissue or cell of suspension culture is known as embryo-genesis.

6) What is an artificial seed?

Ans. Artificial seeds are artificially encapsulated somatic embryos that were able to survive and would not desiccate even after planting into soil.

7) What is endosperm culture ?

Ans. Endosperm culture is the *in vitro* development of isolated mature or immature endosperm from seed at proper stage on suitable culture medium to obtain triploid plantlet.

8) What is embryo rescue technique ?

Ans. In immature embryo culture, wide hybrids often suffer from early spontaneous abortion due to several barriers which act at pre and post-fertilization levels.

9) Write the different stages of micropropagation.

Ans. View 9.3.1

10) What is androgenesis ? What is gynogenesis ?

Ans. Androgenesis involves haploid production through anther or pollen culture. Gynogenesis involves gynogenic haploid production through ovary or ovule culture.

Unit 10 □ Recombinant DNA Technologies

Structure

- 10.0 Objective**
- 10.1 Introduction**
- 10.2 Objective of Recombinant DNA Technologies**
- 10.3 Blotting techniques**
 - 10.3.1 Northern Blotting**
 - 10.3.2 Significance**
 - 10.3.3 Western Blotting**
 - 10.3.4 Application**
 - 10.3.5 Southern Blotting**
 - 10.3.6 Application**
- 10.4 DNA Finger printing**
- 10.5 Molecular DNA markers**
 - 10.5.1 Types of genetic markers**
 - 10.5.1.1 RAPD**
 - 10.5.1.2 RFLP**
 - 10.5.1.3 SNPs**
- 10.6 Nucleic Acid Sequencing**
 - 10.6.1 Sauger method / Chain termination method**
 - 10.6.2 Flow Chart of DNA sequencing**
- 10.7 Polymerase Chain Reaction (PCR)**
- 10.8 RT-PCR**
- 10.9 Application of rDNA Technology**
- 10.10 Summary**
- 10.11 Questions & Answers**

10.0 Objectives

After reading this unit,

- You will be able to learn about different rDNA technologies & their application.
- You will know about different types of DNA markers
- You will be able to discuss about nucleic acid sequencing, PCR & RT-PCR.

10.1 Introduction

Recombinant DNA Technology is a new born discipline of science which aims to alter the heredity apparatus of a living cell. *Recombinant DNA* is a form of artificial DNA that is made through the combination or insertion of one or more DNA strands, therefore combining DNA sequences as per our requirements within different species i.e. DNA sequences that would not normally occur together. Recombinant DNA technology is also popularly known as genetic engineering which is performed under highly controllable laboratory conditions so that the cell can perform completely new functions.

Principles of rDNA technology :

- Generation of DNA fragments & selection of the desired piece of DNA.
- Insertion of the selected DNA into a cloning vector to create rDNA or chimeric DNA.
- Introduction of the recombinant vectors into host cells.
- Multiplication & selection of clones containing the recombinant molecules.
- Expression of the gene to produce the desired product.

Recombinant DNA technology is one of the recent advances in biotechnology, which was developed by two scientists named Boyer and Cohen in 1973. They constructed the first recombinant DNA using bacterial DNA and plasmids. Some molecular methods used in rDNA technology is discussed in this chapter.

10.2 Objective of Recombinant DNA Technologies

- 1) Artificially synthesize new genes.

- 2) Altering the genome of an organism.
- 3) Bring about new gene combinations not found in nature.
- 4) Understanding the hereditary diseases and their care.
- 5) Improving human genome.

10.3 Blotting techniques

It is a tool used in the Identification of biomolecules e.g. DNA, mRNA & Protein.

10.3.1 Northern Blotting

Northern blotting is a technique used to *analyze RNA*. In northern blot, *RNA* is transferred *from agarose gel to nitrocellulose* paper for hybridization analysis. Total cellular RNA or poly (A) RNA is separated by size on agarose gel containing formaldehyde or dimethylsulfoxide. The formaldehyde is used to alter secondary structure of RNA molecules. Nitrocellulose filter paper binds strongly to denatured RNA, but not with RNA

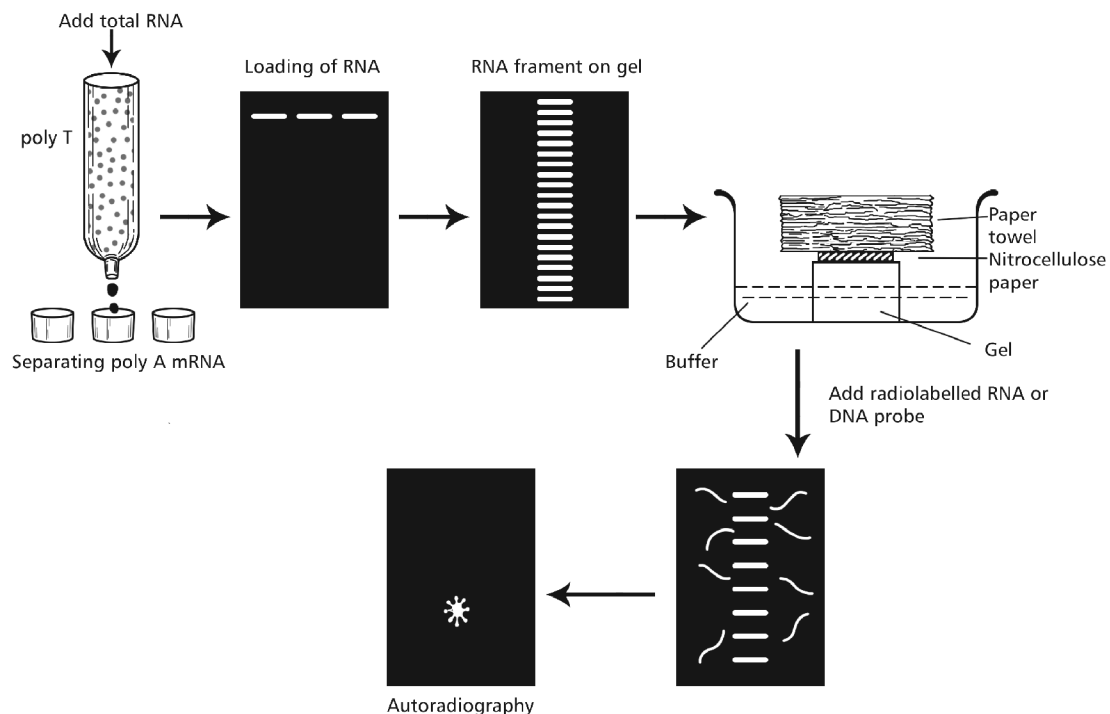


Fig. 10.1 : Northern hybridization

having secondary structure. The NC paper becomes reactive after treating with aminobenzyl oxymethy.

After blotting RNA to chemically reactive paper, they are *hybridized* to radiolabeled DNA probe. Autoradiography is then carried out to locate RNA bands that are complementary to the probe.

10.3.2 Significance

- It is useful in the identification of a particular gene expression in a tissue or a cell-tube.
- It is useful in cDNA cloning because the size of a specific mRNA can be compared with the size of cloned DNA

10.3.3 Western Blotting

Western blotting technique is used for identification of particular protein from the mixture of proteins. In this method labelled antibody against particular protein is used to identify the desired protein. So it is a specific test, and also known as immunoblotting.

Cellysate is the most common sample for western blotting. *Protein* is extracted from cell by mechanical or chemical lysis of cell. The concentration of protein is determined by spectroscopy. Protein is then diluted in loading buffer containing glycerol Tracking dye (bromothymol blue) is also added in sample to monitor the movement of protein.

The sample is then loaded in well of SDS-PAGE. The proteins separate on the basis, of electric charge, isoelectric point, molecular weight. The small size protein moves faster than large size protein. Proteins are negatively charged so they move towards positive (anode) pole, when electricity is applied.

The *Nitrocellulose membrane* is placed on the gel, so that the separated protein pass the gel, get transferred to the paper by capillary action. This type of blotting may take 1-2 days.

The membrane with transferred protein is then probed with a primary antibody, washed and reacted with a secondary antibody labeled with an enzyme. e.g. horseradish peroxidase (HRP). The bound enzyme activity is used to detect the target protein and visualized by a chemiluminiscent or chromogenic method.

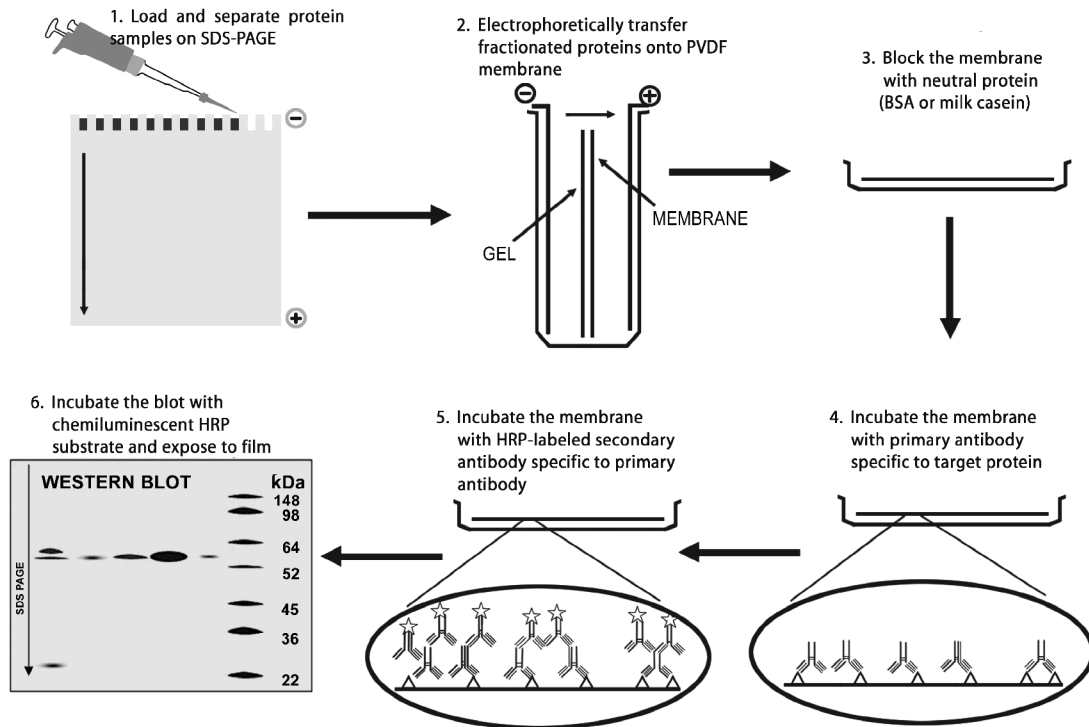


Fig. 10.2 : Western Blotting Procedure

10.3.4 Application

1. To determine the size and amount of protein in given sample.
2. Detects antibody against virus or bacteria in serum.
3. Detects anti HIV antibody in patients serum.
4. Useful to detect defective proteins.

10.3.5 Southern Blotting

The technique of southern blotting developed by E. M. Southern, is one of the most important methods used in molecular biology.

Here, DNA is transferred from a gel to a membrane for hybridization analysis. DNA is cut with suitable restriction enzymes and run on a gel. Treatment with NaOH denatures the DNA to form single strand. The transfer of DNA from agarose gel to the membrane is performed by capillary action. The gel is placed above the buffer-saturated filter paper. The nitrocellulose membrane is placed above the gel and covered by 2-3 layers of dry

filter paper towel. A flow of buffer occurs through the gel and membrane to the top papers. The flow carries the DNA fragment with it. However, DNA cannot pass through the membrane and is fixed firmly to the paper. The membrane with DNA is then exposed to a solution containing the radio-labelled cDNA, overnight.

The binding of probe to its complementary sequence is then detected by autoradiography.

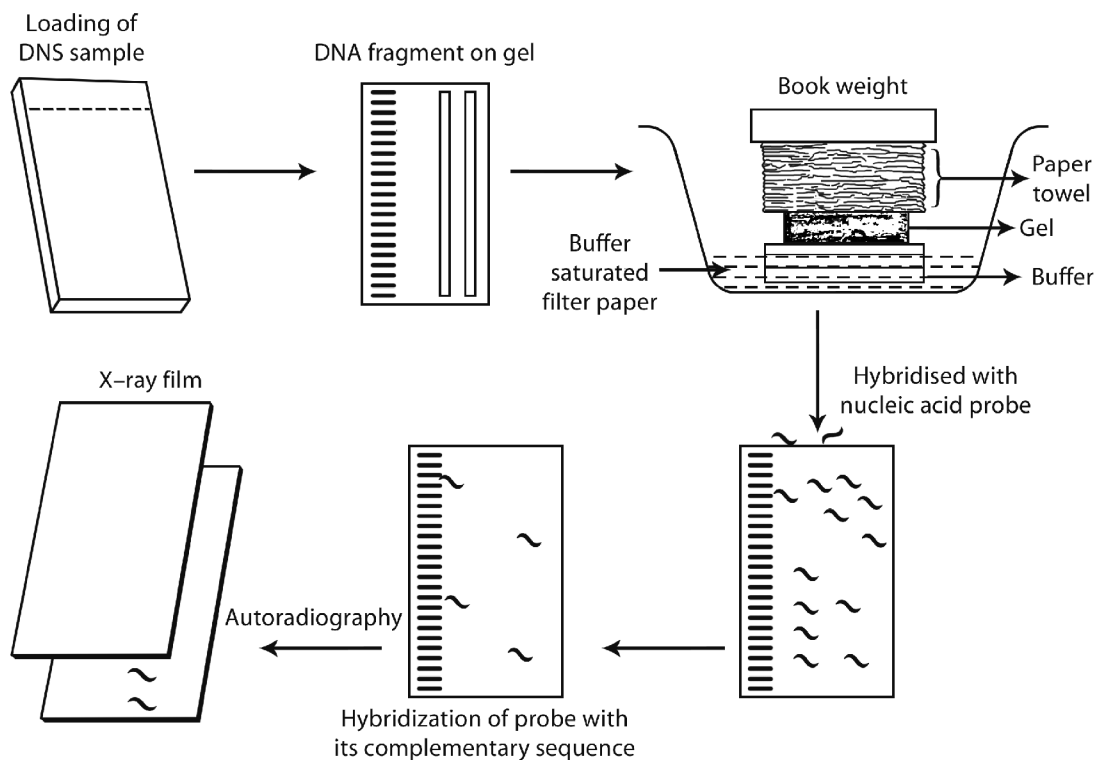


Fig. 10.3 : Southern blotting technique

10.3.6 Applications

Southern blotting is useful for detecting major gene arrangements. This technique plays important role in DNA fingerprint, identification of novel gene, identification of structurally related genes in the species etc.

10.4 DNA Finger printing

DNA finger printing (also called DNA typing or DNA profiling) *is a technique of*

determining nucleotide sequences of certain areas of DNA which are unique to each individual. Each person has a unique DNA finger print which is the same for every cell, tissue and organ of a person.

This method of DNA profiling uses *repetitive sequences* that are highly variable, called *variable number tandem repeats* (VNTRs), in particular *short tandem repeats* (STRs), also known as *microsatellites* and *minisatellites*. VNTR loci are similar between closely related individuals, but are so variable that unrelated individuals are unlikely have the same VNTRs.

The *Steps of DNA finger printing* include–

- *Isolating the desired DNA* chemically, mechanically or enzymatically
- *Cutting the DNA* into several pieces by restriction endonuclease.
- Sorting the DNA pieces by *gel electrophoresis*, by size.
- *Denaturing the DNA* pieces.
- *Blotting DNA* by applying the gel to a sheet of nitrocellulose paper.
- *Probing for VNTRs* is done using labelled DNA probes.
- *Hybridized fragment* can be detected by autoradiography.

10.5 Molecular DNA markers

Molecular marker is a DNA or gene sequence within a recognized location on a chromosome which is used as identification tool.

In the pool of unknown DNA or in a whole chromosome, these molecular markers help in identification of particular sequence of DNA at particular location.

10.5.1 Types of genetic markers

10.5.1.1. Random Amplified Polymorphic DNA (RAPD)

RAPD was developed by Welsh and McClelland along with Williams in 1990. *It is based on PCR assay* and it does not need any prior sequencing of DNA. This process uses *short arbitrary primers of 8-12 bp* that randomly amplifies the region of DNA.

It proceeds when a *single primer anneals* to the genomic DNA at 2 distinct sites on the complementary strand of DNA template. The *amplification* of the segment of DNA depends on the positions complementary to the primer's sequence.

The fragments obtained from RAPD are between 0.2 to 5.0 kb and can be viewed by using agarose gel electrophoresis. If any mutation occurs in primer-binding region, then no PCR product will be produced.

Application–

- Distinct pattern of amplification is seen in different samples. So RAPD can be used for studying polymorphism.
- It is applicable for the mapping of genome analyzing linkage & specific genotyping.

10.5.1.2 Restriction Fragment Length Polymorphism (RFLP)

It was one of the oldest methods used for the analysis of DNA in fields of forensic science.

It is a *hybridization based technique*, invented by Alec Jeffreys in 1984.

The method uses particular *restriction endonuclease* that cuts at its specific site yielding fragments of various lengths, along with fragment of interest.

The restriction fragments are then examined using *gel electrophoresis*.

The gel is now treated with luminiscent dyes for the visibility of DNA bands.

The gel is placed in sodium hydroxide solu for *denaturation*, so that single stranded DNA are formed.

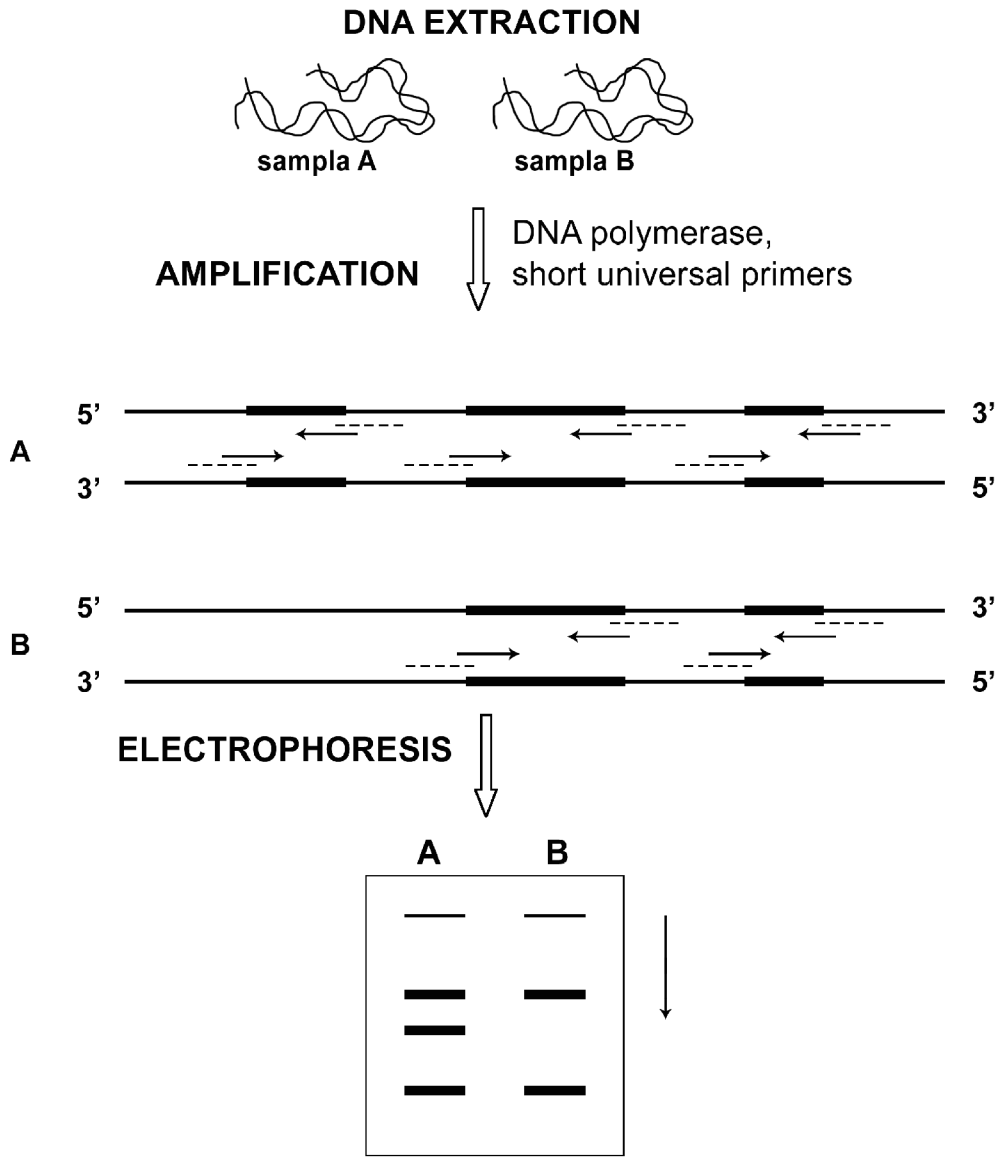
These ssDNA are transferred *into nitrocellulose paper* by the process of *capillary blotting*.

The labelled RFLP probe is hybridized with DNA on the nitrocellulose paper.

The probes form colour bands, seen by autoradiography.

Application of RFLP test

- Genome mapping
- Genetic disease analysis



----- primer
 ——— amplified fragment
 → direction of amplification

- To detect mutated gene
- DNA finger printing (forensic test).

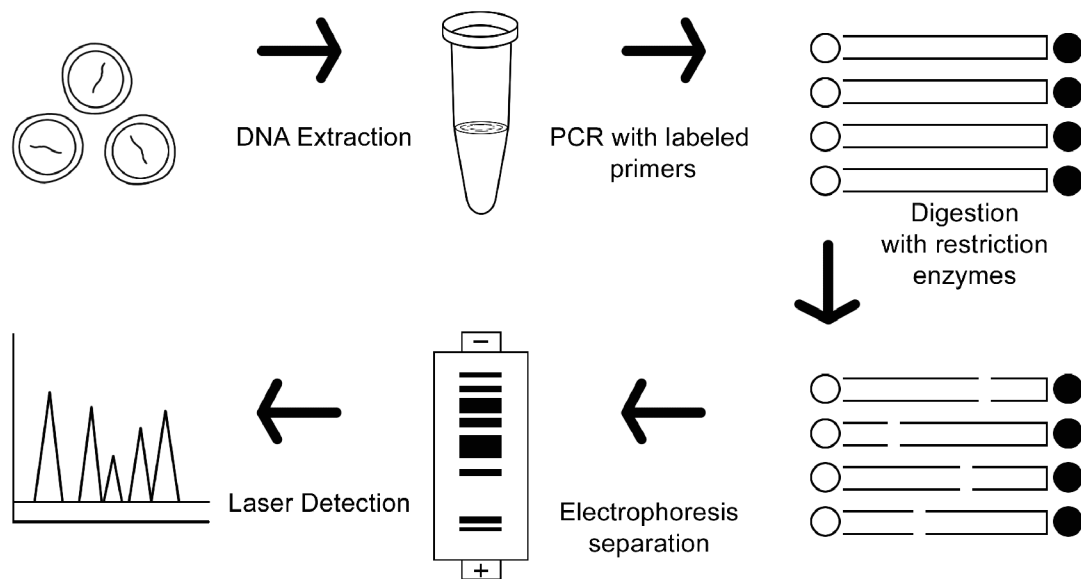
10.5.1.3. Single Nucleotide Polymorphisms (SNPs)

SNPs are the most common type of genetic variation among people. Each SNP represents a difference in a single DNA building block, called a *nucleotide*. SNPs occur normally throughout a person's DNA. They occur almost once in every 1000 nucleotides, on average ie there are approximately 4-5 million SNPs in a person's genome.

These SNPs belong to the last-generation molecular markers which help scientists to locate genes associated with diseases.

An example of an SNP is the substitution of a *C* for a *G* in the nucleotide sequence AACGAT, thereby producing the sequence AACCAT. SNP detection technologies are used to scan for new polymorphisms and to determine the alleles of a known polymorphism in target sequences. Researchers have found SNPs that may help to predict an individual's response to certain drugs, susceptibility to environmental factors such as toxins, and risk of developing particular diseases.

STEPS INVOLVED RFLP



10.6 Nucleic Acid Sequencing

Nucleic acid sequencing encompasses biochemical methods for determining the order of the nucleotide bases (adenine, guanine, cytosine, thymine) in a DNA molecule.

Some of the methodology for DNA sequencing are –

- Chain termination method / Sanger method
- Chemical degradation method / Maxam – Gilbert method.
- Pyrosequencing method

10.6.1 Chain termination method / Sanger method

The enzymatic chain termination method was developed by Frederick Sanger and coworkers in the year 1977. The method is based on the DNA polymerase – dependent synthesis of a complementary DNA strand in the presence of natural deoxynucleotides (dNTPs) and dideoxynucleotides (ddNTPs) that serve as terminators.

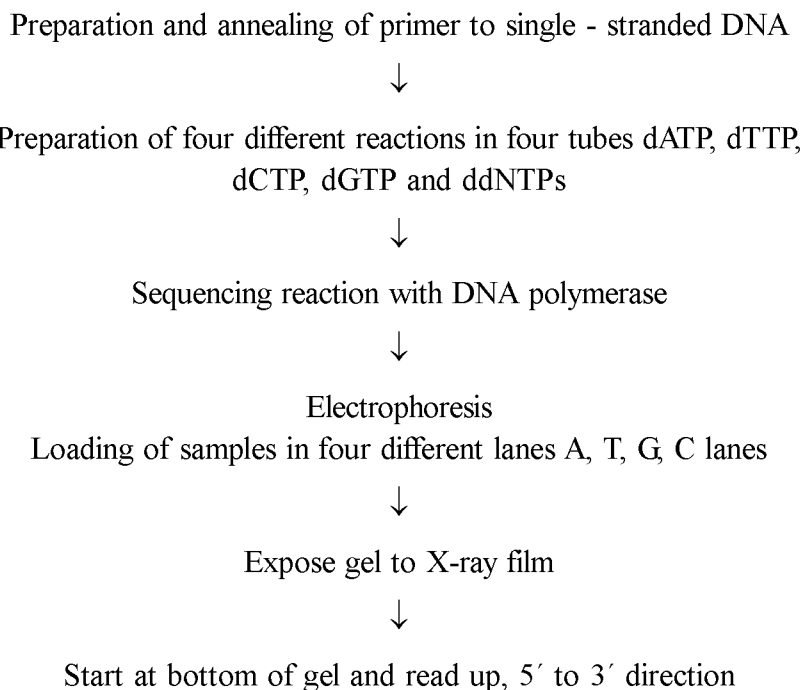
The enzymatic synthesis of DNA takes place by the sequential formation of a phosphodiester bond between the free 5' phosphate group of an incoming nucleotide and the 3' OH group of the growing chain. This process takes place throughout the length of the DNA molecule. The strand synthesis reaction, which is catalyzed by a DNA polymerase enzyme and required the four deoxyribonucleotide triphosphates (dNTPs – dATPs, dGTPs, dCTPs, dTTPs) as substrates. In the presence of a dideoxynucleotide, the synthesis of DNA stalls because the phosphodiester bond cannot be formed as dideoxynucleotide lack 3' OH group instead have a 3' H group. The chain terminates at that point and the last base in the 3' end of the chain is a dideoxy terminator. In Sanger's sequencing technique each dideoxynucleotides is labeled with a different fluorescent marker.

After primer hybridization, the sample is equally distributed into four different tubes, each containing a different dideoxy terminator nucleotide (ddATP, ddCTP, ddGTP, ddTTP) along with all the other reagents required for the synthesis of DNA.

A small amount of each of the tubes then loaded onto a denaturing poly acrylamide gel and electrophoresed for 2-6 hours to separate DNA molecules.* The DNA molecule resulting from the synthesis reaction will be sorted according to size. by electrophoresis. Larger molecules located near the sample well and the smallest molecules moving further down the gel. An autoradiograph is performed after electrophoresis and the band pattern is read.

* As the synthesised strands are double-stranded in nature they need to be denatured during the gel electrophoresis because of the presence of urea and high temperatures.

10.6.2 Flow Chart of DNA sequencing (Fig 10.4)



10.7 Polymerase Chain Reaction (PCR)

Polymerase chain reaction (PCR) is a rapid and versatile in vitro method. With the PCR, any particular stretch of genetic material can be pinpointed and amplified numerous times by selecting a pair of primers that flank the desire stretch of DNA.

PCR is carried out in a single tube simply by mixing DNA with a set of reagents and placing the tube in a thermal cycler, a piece of equipment that enables the mixture to be incubated at a series of tempartures that are varied in a preprogrammed manner. The basic steps in PCR experiment are (Fig 10.5) :

1. **Denaturation** : The mixture is heated to 94°C, at this temperature the hydrogen bonds of the double-stranded DNA molecule are broken. The two strands of the target DNA molecule are separated (denatured).
2. **Annealing of primers** : The mixture is then cooled down to 50-60°C. At this temperature the two strands of DNA molecules could join back together but do not because the mixture contain oligonucleotides or primer (short DNA molecules) which anneal to the target DNA molecules at specific positions.

• Sanger method

- synthesize complementary DNA strand in vitro
- in each tube:
 - “normal” N-bases
 - dideoxy N-bases
 - ddA, ddC, ddG, ddT
 - DNA polymerase
 - primer
 - buffers & salt

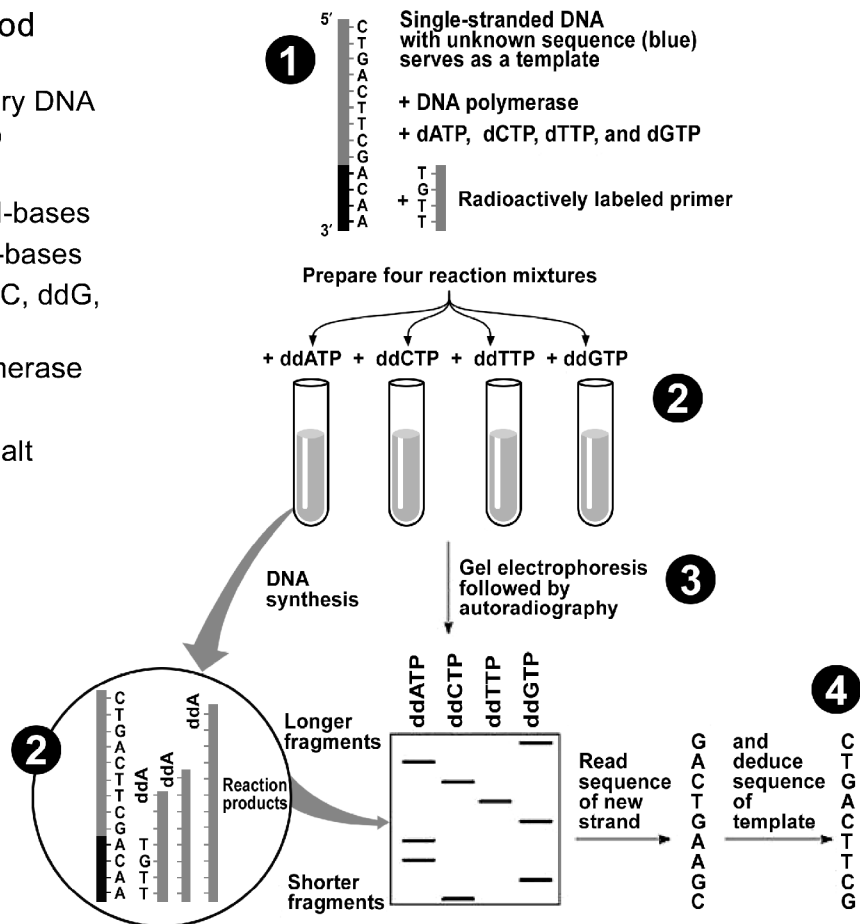


Fig. 10.4 : DNA sequencing

3. **DNA polymerization** : After annealing of primers the temperature is raised to 74°C. At this stage the Taq DNA polymerase attaches to one of the ends of each primer and synthesizes new strands of DNA, complementary to the template DNA molecules.
4. **Repetition** : The temperature is increased back to 94°C. This begins a new cycle of denaturation-annealing-synthesis. These steps are repeated 20-50 times to obtain large quantities of the desired DNA fragments.

The PCR products are called amplicons. The number of molecules is obtained by applying the following formula :

$$M_f = M_i \times 2^n$$

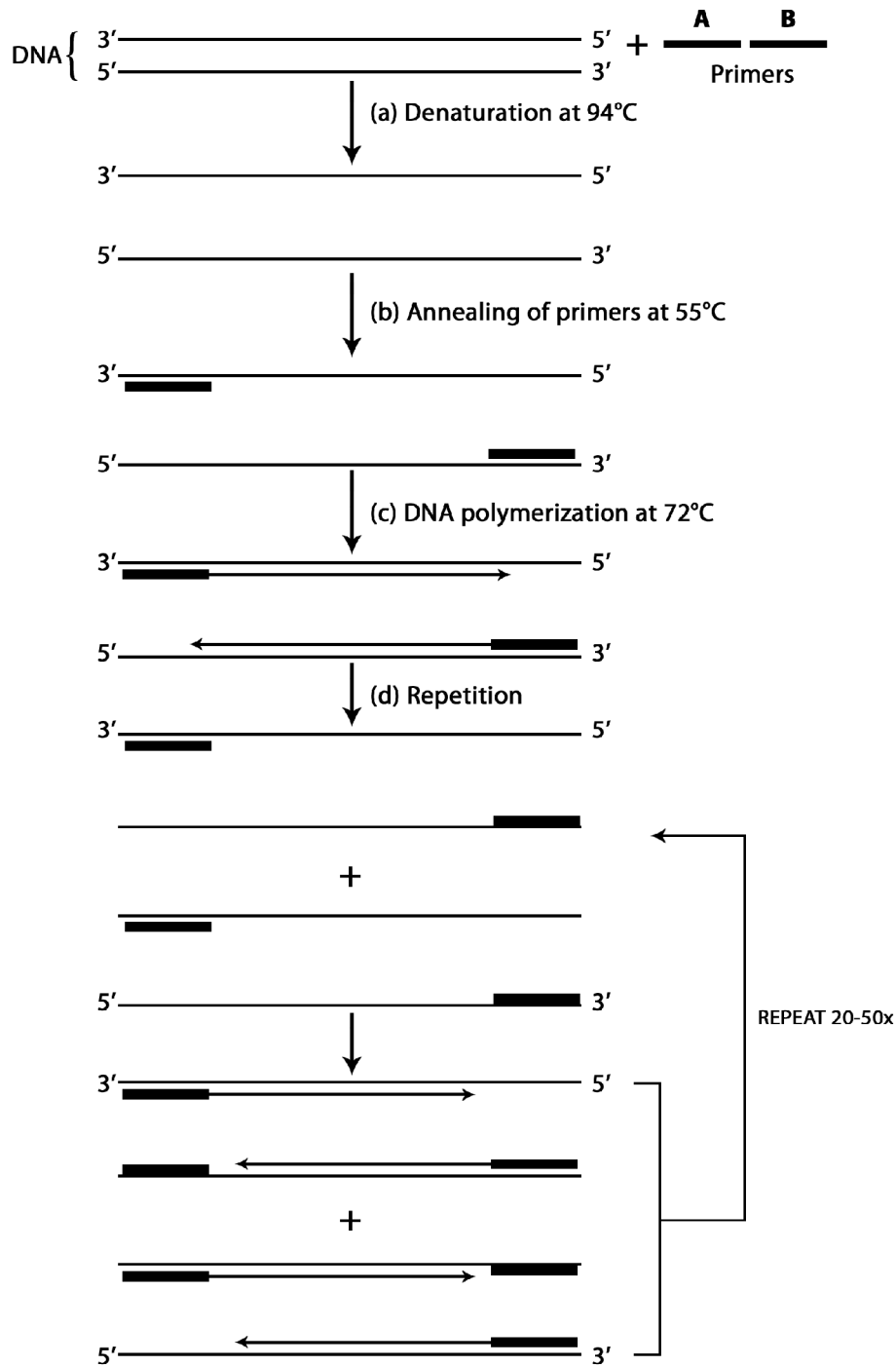


Fig. 10.5 : Schematic representation of a typical PCR assay.

where, M_1 = initiate number of molecules (templete)

n = number of cycles performed.

M_f = final number of DNA segment produced by the PCR.

10.8 RT-PCR can also amplify target RNA

The thermostable polymerases used in the basic PCR requires a DNA templete and hence is limited to the amplification of DNA samples. There are numerous instances in which the amplification of RNA would be preferred. RT-PCR involves the use of the enzyme reverse transcriptase to make DNA copies of RNA molecules, which are then amplified using standard PCR protocol. In this assay, RNA extracted from the cell or virus is converted to cDNA (complementary DNA) as a first step. Once this preliminary step has been carried out, the PCR primers and Tag polymerase are added and the experiment proceeds as the standard protocol.

10.9 Applications of rDNA Technology

- Analysis of Gene Structure and Expression
- Production of Pharmaceutical Products like Drugs, Vaccines
- Genetically modified organisms (GMO)
- Application in medicine
- Prenatal diagnosis
- Industrial application
- Forensic medicine etc.

10.10 Summary

Recombinant DNA technology is not only an important tool in scientific research, but has also resulted in enormous progress in the diagnosis and treatment of certain diseases and genetic disorders in many areas of medicine.

In spite of having great future aspects it has certain limitations. Destruction of native species in the environment by the genetically modified species are introduced. Recombinant

organisms contaminating the natural environment. Many people worry about the safety of modifying food and medicines using recombinant DNA technology. Such a system might lead to people having their genetic information stolen and used without permission.

10.12 Questions & Answers

1) Which technique involves in DNA fingerprinting ?

Ans. Southern blotting.

2) Name the enzyme which is used in PCR technology.

Ans. Taq polymerase

3) What is DNA fingerprinting ?

Ans. DNA fingerprinting (also called DNA profiling) is a technique of determining nucleotide sequences of certain areas of DNA which are unique to each individual.

4) Who develop DNA fingerprinting technique ?

Ans. It is developed by Alec Jeffrey.

5) Who develop the PCR technique ?

Ans. Kary Mullis

6) At what temperature does denaturation of DNA double helix takes place ?

Ans. 94°C

7) What is the main enzyme component of Sanger sequencing ?

Ans. The chain-termination or dideoxy method of DNA sequencing capitalizes on two unique properties of *DNA polymerase* enzyme.

8) Which is act as chain terminator ?

Ans. Dideoxynucleotide

9) What is a primer ?

Ans. A primer is a chemically synthesized oligonucleotide.

10) How many types of deoxynucleoside triphosphates are used in Sanger sequencing ?

Ans. 4 different types of deoxynucleoside triphosphate are used in Sanger sequencing.

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Note
