

Tree Seed Technology

Practical Manual

For

M. Sc. (Forestry) Silviculture and

Agroforestry



Dr. Swati Shedage



Rani Lakshmi Bai Central Agricultural University, Jhansi
(UP) - 284003, India

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Experiment No: 1

Identification of forest tree seeds

Introduction: Seed is defined as matured ovule consisting of embryonic together with store of food surrounded by protective coat. Seed can be defined as a fertilized ovule consisting of intact embryo, stored food and seed coat which is viable and has got the capacity to germinate. Botanically seed may be defined as a fertilized mature ovule that possesses an embryonic plant, stored food material covered by a protective coat or coats which is viable and has got capacity to germinate under favorable environmental conditions. It is sometime difficult to identify seed of tree species, especially seeds of different species but same genera.

Objectives: 1. To become familiar with forest tree seeds. 3. To begin to identify seeds and its morphological characteristics

Materials: Different tree seeds, Polythene and Plastic bottle.

Parts of a typical dicotyledonous seed:

- Seed coats: seed coats consist of two layers of integument, united or free, the outer being called tests and the inner is called tegmen. The seed coats are provided with hilum which represents the point of attachment with the stalk, micropyle, minute pore above the hilum and raphe (a ridge formed by the funicle or stalk in many seeds).
- Embryo: Embryos lying within the seeds consist of an axis and two cotyledons. The pointed end of the axis is the radicle and the feathery leaf end is called plumule. As the seed germinates the radicle gives rise to the root and plumule to the shoot.
- Endosperm: Endosperm is the fleshy food storage tissue. In some seeds endosperm is present until maturity. Such seeds are called endospermic or albuminous seeds. In some seeds it is consumed in the young stage by the developing cotyledons and such seeds do not possess endosperm at maturity. Such seeds are called non endospermic or exalbuminous seeds.

Parts of a typical monocotyledonous seed:

- Seed coat: Seed coat is the brownish membranous layer adherent to the grain. This layer is made up of the seed coat and the wall of the fruit fused together.
- Endosperm: It forms the main bulk of the grain and is the food storage tissue of it, being laden with reserve food material, particularly starch. In a longitudinal section of a grain, it is seen to be distinctly separated from the embryo by a definite layer known as the epithelium.
- Embryo: It is very small and lies in a groove at one end of the endosperm. It consists of only (a) one shield shaped cotyledon known as scutellum (b) a short axis with the plumule and the radicle. The radicle is protected by a root cap. The plumule as a whole (growing point and foliage leaves) is surrounded by a protective sheath called coleoptile; similarly the radicle is surrounded and protected by a sheath called coleorrhizae. The

surface layer of the scutellum lying in contact with the endosperm is the epithelium, its function is to digest and absorb the food material stored in the endosperm.

Seeds of some important forest trees



Tectona grandis



Ailanthus excelsa



Terminalia arjuna



Acacia auriculiformis



Prosopis juliflora



Cassia fistula



Diospyrus melanoxylum



Anacardium Semicarpus



Thespesia populnea



Terminalia chebula



Melia dubia



Holoptelia integrigolia

Activities: Collect seeds of any 10 forest tree species and describe its morphological characters

Worksheet

Experiment No: 2

Study of Seed sampling

Introduction: Sampling is the process of taking a small part or quantity of something for testing or analysis; it is the first step in seed testing. In sampling, it is essential: (1) to obtain a sample of proper size, and (2) to obtain a sample representative of the main seed lot. The results of the laboratory tests can only show the quality and characteristics of the sample submitted for the analysis; therefore, the validity of test results for a large seed lot is determined by the success of obtaining a representative sample. Sampling seed lots for quality evaluation must be done systematically, using appropriate techniques, tools, and procedures, to ensure that the seed sample represents the entire lot.

Objectives: Quantify a seed lot according to accepted standards. Determine sampling intensity according to size and characteristics of the seed lot. Learn about appropriate sampling instruments and techniques according to recognized standards.

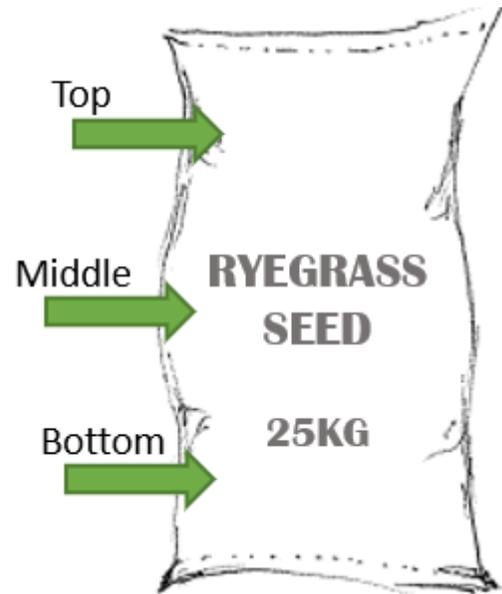
Materials required: Nobbe Type Trier: Used for sampling bags or sacks, Long Nobbe Trier: can be used for sampling bulk bags and bulk bins where it is possible to sample through the liner wall. Sleeve Trier / Spiral Sleeve Trier: Used for sampling seed in bulk bags or bulk bins. Pelican Sampler: Used to take samples from free-flowing seed streams. Sample container: Sample containers used to collect primary samples, composite samples and during mixing and dividing must be clean and static free. Riffle Divider: Used to prepare the submitted sample for testing by reducing the composite sample. Balances (weighing scales): Used for ensuring samples for submission meet minimum sample weights. Automatic Sampler: Used to mechanically sample through a cross section of the seed stream during processing. An Automatic Sampler will be approved provided installation and operation meet requirements. Seed sample envelope, calico bag, or approved moisture sample bag: Used to send samples to the Official Seed Testing Laboratory. Seals: Paper seal for envelopes or sequentially numbered metal seal (silver) for calico bags, attaching extra labels or re-labelling.

Procedure:

Sampling with the Nobbe Trier

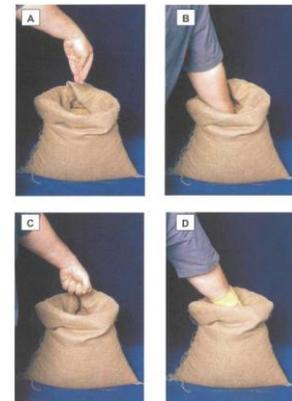
- Select the correct seed trier. (Check the trier has a Unique Number, either engraved, stamped, or with an identification sticker and is listed on the Seed Sampling Equipment Register.
- Check the trier is clean and fit for use.
- Insert the point of the trier gently into the side of the bag / bin with the slot facing down to prevent seed entering the trier.
- Angle the trier upwards at approximately 30° and push the trier up to the handle (this should place the slot in the centre of the bag / bin).
- Hold the sample container over the end of the trier then turn the trier half a turn (180°) so the slot is now facing upwards.

- Withdraw trier immediately at decreasing speed from the centre to the side of the bag ensuring that seed is running freely.
- Gently agitate the trier while it is being withdrawn to maintain an even flow of seed.
- Run the point of the trier over the hole a few times to pull the sack threads back together. If the bag fibres break or there are holes in paper bags, use sticky tape to repair the bags. Check that the sample collection container is clean and anti-static.
- Take primary samples (sub-samples) alternately from top, middle and bottom of bag, and from different random positions in bulk bins. Start sampling the lower bags on pallets and work upwards to prevent contamination spilling into the sample container.
- Regularly check the primary samples to ensure homogeneity.
- If the sample does not appear uniform, stop sampling and notify the Seed Store Manager.
- Repeat until sufficient primary samples are obtained
- Weigh the composite sample and check against Standard Minimum Weights of Submitted Samples



Sampling Using the Hand Method

- Primary samples are taken by removing handfuls of seed from random positions and depths within the open bag. This position should be varied between bags.
- Prior to sampling ensure sleeve is rolled up and hands are clean.
- Insert the open hand into the bag of seed to the required position then close and withdraw the hand, taking care to keep fingers tightly closed about the seeds so none escape
- Empty hand into the seed collection container.
- Brush hand down thoroughly into the sample collection container between primary samples.
- To ensure that lower layers of bags or bins are effectively sampled, it may be necessary to request that some bags be partially emptied, then refilled.
- Repeat until sufficient primary samples are obtained



Activities:

1. What methods are adopted in the laboratory to obtain a working sample?
2. Explain the differences between the various kinds of equipment used in mechanical sample reduction. In your opinion, which is the most suitable method? Why?

Worksheet

Experiment No: 3

Study of storage methods

Objectives: To study the methods of storing seeds of economic plants to preserve planting stocks from one season until the next

Materials Required: Seeds, air dryer, refrigerator, deep freezer, gunny bags, polybags, muslin clothbags

Procedure

1. Determine whether the seeds can be stored

Not all seeds can be stored. If you are uncertain, try looking in the Royal Botanic Garden Kew Seed Information Data Base (SID). First, find the scientific name of the plant. Find the SID storage classification for your seeds. If the seeds are “orthodox”, you should be able to store the seeds for years by drying it as described below, then storing them refrigerated or frozen in airtight containers. If the seeds are “intermediate”, storage is trickier. If the seeds are “recalcitrant”, it is not practical to store them for more than a short time. It is best to germinate them soon after collection. If you cannot find the seeds in the SID, you may be able to guess whether the seeds can be stored. Small seeds that dry on the parent plant are usually “orthodox”. Large seeds that are moist when ripe (like mangos) are almost always “recalcitrant.”

2. Collect ripe, healthy seeds

Collect healthy, ripe seeds. Unripe seeds will not store well. Seeds are usually ripe when fruits are fully ripe or starting to dry. Ripe orthodox seeds are typically firm and dry, and are often darker than unripe seeds. (Some seeds may require additional ripening after collection.) Remember, viable seeds are alive. After collection, treat them like fresh produce. Keep them in a cool, ventilated place (if you keep them in plastic bags, leave the top unsealed so that the seeds/fruits can breathe). To prevent mold, they should not be wet. Try to process them as soon as practical after collection.

3. Separate and clean the seeds

If you process the seeds wet, air-dry them. Do not use heat to speed drying, as this will shorten storage life. Use moving air instead.

4a. Dry the seeds to the correct moisture level for storage

Under warm, humid conditions most stored seeds deteriorate rapidly. Air drying will not dry them enough for good storage life.

4b. Meanwhile, germinate some of the seeds

This is to make sure that the seeds are really viable. You may need to use special treatments to break dormancy. Seeds of some plants need a period of after harvest ripening before they will germinate. Ask other growers for advice.

5. Pack the seeds for storage

After you dry the seeds, you need to pack them in airtight containers. Otherwise, the seeds will absorb moisture from the air and undo the results of step 4a. Many containers are not as airtight as you might think. For airtight storage, we recommend using containers with rubber gaskets. If you have many small containers of seeds, you can put the small containers into larger gasketed containers. (Exception: If you use a self defrosting refrigerator for both drying and storing the seeds, airtightness does not matter so much. If you are going to store the seeds for more than a few years, consider getting archival labels, or enclosing a piece of paper with the label information written in pencil inside the storage container. Ordinary stationery store labels may become brittle and fall off after a few years.

6. Store the seeds

Seeds of most garden and crop plants store best when dried and kept at low temperatures. In most national seed banks, the preferred method for long-term storage is freezing to near 0° F, which is the temperature in a home freezer or in the freezing compartment of a home refrigerator. For year to year storage, many store the seeds near 39° F, the temperature inside a home refrigerator. In developing countries, dried seeds are often stored at room temperature inside airtight containers, but cold storage is better if available. If the freezer or refrigerator fails, it is not a disaster. Changing temperatures during storage will not harm the seeds, but when they are warm, they will age much faster than when they are cold. When the temperature drops, they will go back to aging slowly. Our recommendations for storage conditions, once the seeds have been dried: First choice: Freezing. Second choice: Refrigeration. (Seeds of some wild plants store better refrigerated than frozen, but this is uncommon for cultivated plants.) Third choice: Room temperature after proper drying, in airtight containers.

7. Germinate a sample from time to time

If you store the seeds for more than a few years, you may want to take out some to test from time to time. Even under good storage conditions, seeds will eventually deteriorate. When the first signs of reduced germination appear, it is time to replenish the seed supply.

8. Prepare the seeds for sowing

Seeds that have been dried for storage may become damaged if they absorb water too fast. This will lead to poor germination or unhealthy seedlings. To prevent this, allow the stored seeds to pick up moisture from the air slowly before sowing them. You can do this by putting them in a sealed plastic container with a damp paper towel for a day. (Do not let the damp towel contact the seeds.) Note: If you have stored the seeds in a freezer or refrigerator, allow them to come to room temperature before opening the storage container. Otherwise, moisture will condense on the seeds. If you do not use all of the seeds, reseal the container immediately. You do not have to

redry the remaining seeds unless the volume of air in the container is much greater ($>100x$) the volume of seeds.

9. Sow the seeds: You can now sow the seeds normally.

Activities: What are storage methods used to different forest tree seeds

Worksheet

Experiment No: 4

Study Seed quality testing-purity

Introduction: What is quality seed? Quality seed is genetically pure, characterized by a high germination percentage and appropriate moisture content; it is free from diseases, and has a high content of pure seeds and no weed seeds. Quality seed is important in both the formal and the informal sector.

The formal sector encompasses specific activities to make available new varieties and maintain their purity; and to certify seed and distribute it to farmers through recognized seed channels. Quality seed is produced under supervised conditions, which may vary depending on the specific seed class or category.

The informal sector – also known as the traditional or farmer seed system – lacks public sector regulation. Seed is exchanged and bartered among farmers or sold on the local market. According to Cromwell, Friis-Hansen, and Turner (1992), five key features distinguish the informal system: it is based on tradition, is semi-structured, operates at individual community level, uses a wide range of exchange mechanisms and usually deals with small quantities of seeds as widely demanded by farmers. This traditional system has maintained local varieties and landraces for hundreds of years.

Objectives: To determine the extent to which a given seed lot meets the standards set for certain attributes like genetic, physical, physiological and health that determining the quality status of seeds.

Materials required: Purity board, hand lances , seed blower, sieve, Analytical balance, forceps and fine needles

Procedure:

- **Pure seed** – the species stated by the applicant, or that found to predominate in the test, and including all botanical varieties and cultivars of that species. The pure seed fraction comprises also:- Mature undamaged seeds of the species; and - Pieces of broken seeds that are more than half the original size.
- **Other seeds** – seed units of any plant species other than that of pure seed.
- **Inert matter** – seed units and all other matter and structures not defined by ISTA as pure seed or other seed, for example:- Broken pieces of pure seed and crop seed species that are half or less their original size; - Soil particles, sand, stones, chaff, stems, leaves, flowers; and - Smut balls, ergots and nematode galls.

Identify the seed.

- Determine the correct weight of the working sample ($\geq 2\ 500$ seeds with a maximum weight of 1 000 g). The ISTA Rules stipulate that the analysis can be done on one working sample of this weight or on two subsamples of at least half this weight, each independently drawn.
- Weigh the working sample (or each subsample) in grams to the minimum number of decimal places necessary to calculate the percentage of its component parts
- Divide the working sample on the working board into three components (pure seed, other seed and inert matter).
- Weigh the individual fractions independently using an analytical balance.

For example:

- Pure seed = X (g)
- Other seeds = Y (g)
- Inert matter = Z (g)

Example 1.

Components	Weight (g)	Percentage	Example Example (rice)	
			Weight (g)	%
Pure seed	Pure seed	$[X \times 100] \div W$	X = 68.88	98.4
Other seeds	Other seeds	$(Y \times 100) \div W$	Y = 0.14	0.2
Inert matter	Inert matter	$(Z \times 100) \div W$	Z = 0.98	1.4
Total	Total	100	W = 70	100.0

- Pure seed = 98.4%
- Other seeds = 0.2%
- Inert matter = 1.4%

Activities: Estimate purity percentage of given seed sample

Experiment No: 5

Date:

Determination of seed viability by using tetrazolium

Introduction: Viability is the capability of the seed to germinate and produce a normal seedling. It indicates that a seed contains the structures and substances required to germinate under favorable conditions in the absence of dormancy. External physical appearance alone cannot determine whether a seed is alive or dead. Seed viability testing is therefore carried out to determine the percentage of viable seed in a given lot. The test is valid for all species for which a method is described in the ISTA Rules.

Objectives: To Make a rapid assessment of seed viability and seed vigour

Materials: Petri dishes, filter paper, magnifying glass, dropper and bottle, solution of tetrazolium, dissecting needles, forceps

Procedure:

- Draw four replicates of 100 pure seeds at random, either from the pure seed fraction of a purity test carried out or from a representative fraction of the submitted sample.
- Mix the pure seed fraction thoroughly taking care to not select seeds causing biased results.
- Soak seeds in water overnight to soften the embryo and endosperm and activate the enzyme system.
- Make a cut or completely remove the seed-coat (depending on the species) – to expose the embryo and facilitate contact with the tetrazolium solution.
- Immerse the prepared seeds or embryos in tetrazolium salt solution. Avoid exposure to direct light, as it would cause a reduction of the tetrazolium salt. Refer to the ISTA Rules for optimum temperatures and staining times.
- Wash seeds repeatedly with distilled water.
- Examine seeds under a magnifying glass.

Activity: Determine the viability of given seed sample

Observations and result:

Experiment No: 6

Determination of seed viability by using Hydrogen peroxide

Introduction: The primary advantages of the H_2O_2 method are that it is objective and technically simple. It is performed by clipping the seed coat and incubating the seeds in a dilute H_2O_2 solution for one week, at which time seeds with radicles longer than 1 mm are counted and the test is terminated. Because it is the only method that actually measures growth, the H_2O_2 test is often chosen in preference to the tetrazolium, X-ray, and excised embryo methods

Objectives: To Make a rapid assessment of seed viability and seed vigour

Materials: Petri dishes, filter paper, magnifying glass, dropper and bottle, solution of tetrazolium, dissecting needles, forceps

Procedure:

1. Soaking

Withdraw 300 seeds using random sampling techniques, and soak the seeds in 100 ml 1.0% H_2O_2 at room temperature overnight. Only four replications of 50 seeds each are needed for the test but extra seeds should be included to replace ones damaged during cutting.

2. Cutting

Using a single edge razor blade, cut the tip of the radicle end of the seed deep enough to expose, but not damage, the radicle tip. The cut should be made at an angle not straight across in order to reduce damage to the radicle. Place the seeds in water after cutting to prevent desiccation.

*Some radicles may curl during elongation. The total length of the radicle should be taken in to account for purposes of classification.

* Embryos may separate from the seed coats during soaking, especially if too much of the coat was removed during cutting. If this occurs, every effort should be made to match the free embryos with empty seeds in order to avoid errors in classification. For assessment purposes, very long embryos should be classified as "evident", but small embryos must be removed from the tally. If a substantial number of embryos cannot be classified, repeat the test.

3. Incubation

Pour 150 mL 1.0% H_2O_2 (for large e seeds, 200 mL) in to each of four beakers. Add 50 cut seeds per beaker and incubate in the dark at 20-25° Co to allow embryo elongation to occur.

4. Assessment

After 3 days incubation, remove and count any seeds with "evident" radicles. Return remaining seeds to a fresh H₂O₂ solution. On day 7 remove and assess all seeds and terminate test. Data is generally expressed as the proportion of "evident" germinant to total seeds used in the test.

Activity: Determine the viability of given seed sample

Experiment No: 7

Study of Germination Testing

Introduction: Germination is the emergence and development of the seedling to a stage at which the appearance of its essential structures indicates whether it can develop further into a satisfactory plant under favorable conditions in the field.

Objectives: Determine the germination potential of a seed lot, which is vital to compare the quality of different lots and estimate the field planting value.

Materials required: Petri dishes, forceps, covering net, water, blotting paper, sand, Germination incubator and room germinator

Procedure:

- Take a sample of 400 seeds at random from well-mixed pure seed. It is important to not select seeds, as this would give biased results.
- Use four replicates of 100 seeds to ensure adequate spacing. Split replicates of 50 seeds (or even 25, particularly where there are seed-borne pathogens or saprophytes present) to minimize the effect of adjacent seeds on seedling development.
- Place seeds uniformly and sufficiently apart on the moist substrate on the Petri dish. If seeds grown on paper substrates are heavily infected, at an intermediate count, transfer remaining seeds and seedlings to fresh media.
- Place Petri dishes in the germination apparatus; record the number of seeds set and the date.
- Make two counts of seedlings. Schedule the first and final countings according to the ISTA Rules.
- Keep the seed moist throughout the test period.

Activities: Determine the germination percentage of given seed sample

Observations result:

Experiment No: 8

Study of collection of seeds

Introduction: Seed collection for plant propagation is an opportunity to reverse trends of genetic degradation and species loss. Nurseries have a key role in conserving the gene pool of native plants. After selecting and marking good mother trees, several seed collection methods can be used.

Objectives: To study scientific collection of seed

Material: Rake, Sieve, Seed container, large canvas, cloth or plastic sheet

Procedure for collecting seeds

- Effective native seed collection involves a number of steps to ensure quality seeds are collected at the right stage. Proper seed collection requires the following practices:
- Locate populations of desired species before or during flowering.
- Investigate the viability of seeds after dispersal or maturation on a species-by-species basis. Monitor potential sites directly after flowering when fruits are becoming visible.
- Record the dates of flower, fruit, and cone formation. Cones are often a 2-year crop, so you can assess cone crop the year before collection.
- Observe carefully the weather patterns during pollination, fruit formation, and maturation.
- Visit the site frequently to monitor the development and quality of the seed crop.
- Use collection dates from previous years to predict target collection dates and other information.
- Use a cutting test of a few sample seeds to determine maturity before collection.
- Collect seeds during dry weather, if possible

Collecting from natural seed fall

- Clear the ground beneath the tree of leaves, branches, and weeds before seeds begin to fall. This will make seed collection easier. Or, spread plastic sheets, cloth or canvas under the mother trees so that the seeds will fall onto them.
- Use a rake to gather the seeds and collect them daily. Or, fold sheets to collect seeds daily. Chances of insect attack and fungal infection which could occur if seeds are left on the ground too long will be minimized.
- Extract seeds from the litter by sieving

Shaking the tree

- If natural seed fall is spread over a long period of time, manual shaking of the tree is a useful method to get seeds to fall to the ground at the same time. This makes their collection easier. In some cases, however, fruits or pods are strongly attached to the branches and will not drop off easily, even when the tree is shaken.
- Clean the ground, or lay down a plastic or canvas sheet.
- Shake the trunks of trees or low branches by hand. (Higher branches may be shaken using a stick, long pole, hook on rope).
- Separate seed from the dry pods

Pruning off seed bearing branches

- When the seed is out of reach for hand picking various pole implements may be used for pruning branches.
- Select branches with a heavy load of good looking pods.
- Carefully locate the ground sheets so that pods and seeds will fall onto them from pruned branches.
- If necessary, prune out “windows” so that seed bearing branches are able to fall to the ground and not get entangled in the tree as they fall.
- Cut the branches.
- Collect the pods.
- Remove the seeds.

Throwing a rope with weighted end to break off a seed bearing branch

- As the last possibility this destructive method may be used to reach high seed bearing branches from the ground, without having to climb the tree. Branches up to 12 metres from the ground can be reached. Skill is required to throw the rope over the selected branch and in the correct position for ease of breakage.

Climbing trees to collect seed

- To use this method, you must have skill in climbing trees and using some specialized equipment. This is the method normally used to collect from standing dry zone trees as they are of open form and relatively small. Several methods can be used when collecting seed from standing trees. The roof of a car may serve as a platform. Or, climb into the crown of the tree and use a saw, large knife or similar implement to cut down seed bearing branches.

Collecting seed from felled trees

- If a tree is to be felled, try to wait until its seed is ripe. Never fell trees just for seed collection

Activities: Collect the seed of any wild species, write the procedure you followed for collection of seeds

Experiment No: 9

Study of processing of seeds

Introduction: The way in which seeds and fruits are handled during collection, temporary storage, postharvest handling, and cleaning can directly affect seed quality, viability, and storage life. Proper processing of fruits and seeds begins the moment the fruit or seed is removed from the parent plant. Proper processing includes short-term handling from the field back to the nursery, temporary storage at the nursery, and prompt and proper seed extraction if necessary

Objectives: To study scientific processing of seed

Processing for dry seeds:

- After they arrive at the nursery, small quantities of dry fruits and cones can be dried in paper bags or envelopes as long as the contents are loose.
- Large quantities must be dried immediately by spreading the material evenly on a tarp or drying rack.
- The materials will need to be turned several times per day to prevent it from becoming too hot, drying unevenly, or becoming moldy.
- Dry, dehiscent fruits, should also be covered with a fine mesh cloth as well to prevent the loss of seeds after fruits open
- Good air movement, low relative humidity, and temperatures between 65 to 80 °F (18 to 27 °C) promote even drying and eliminate moisture buildup that can cause mold and damaging temperature.
- Separating seeds from dry, dehiscent fruits is usually easy because the fruits split open at maturity. Shaking the fruit inside paper bags so that the seeds fall out will readily separate small lots of seeds and the woody capsules can then be removed from the bag.
- Screening is the easiest way to separate extracted seeds from debris such as dry leaves, wings, and small pieces of dried fruits. Screens can be constructed of hardware cloth and wooden frames.
- If you are cleaning large, tough, leathery pods, you may need pliers, hammers, vices, or screwdrivers. Seeds that are contained in tough woody capsules can be extracted by heating the capsules in ovens or exposing them to fire in a portable barbecue grill. The heat treatment causes the pods to become brittle, which aids in the extraction of the seeds using hand tools. When using fire or heat and hand tools, be certain to not damage the seeds. By simply immersing them in water to separate fine chaff and other impurities, species with hard seed coats can be cleaned.
- The final step is fanning or winnowing, which separates detached wings, hollow seeds, and seed-sized impurities from good seeds

Processing for Fleshy Fruits:

- Fleshy fruits and cones are very susceptible to fermentation, mummification, excessive heating, or microbial infestation, all of which can damage seeds.
- On the other hand, it is important not to let the fruits dry out because it can make cleaning them much more difficult. The best procedure is to temporarily store fleshy fruits in white plastic bags in a cool place or refrigerator until the seeds can be processed.
- Seeds in fleshy fruits need to be processed shortly after collection.

- The first step in cleaning is to soak fleshy fruits in water to soften the pulp.
- The soak may need to last a few hours to a few days, depending on the species, and the water needs to be changed every few hours.
- Depulping After the pulp is soft, flesh can be removed by hand squeezing or mashing using a wooden block, rolling pin, or other device. The flesh can also be removed by wet screening, which involves hand rubbing the fruits against screens using a steady stream of water to eliminate the pulp.
- Cleaning-After a course of tumbling, the contents is dumped into a sieve and the pulp or debris is washed off, leaving clean seeds.

Activities: Collect the seed of any wild species and process it, write the procedure you followed for processing the seeds

Experiment No.10

Seed health testing primarily to the presence or absence of disease by ocular method

Introduction: It is important to understand the distinction between seed pathology and seed health. Seed pathology – the study of seed-borne diseases, including the infection mechanism; seed transmission; role of seed-borne inoculum in disease development; techniques for detection of seed-borne pathogens; seed certification standards; deterioration due to storage fungi, mycotoxins and mycotoxicoses; and control of seed-borne inoculum.

Objectives: To determine the presence or absence of disease-causing organisms (e.g. fungi, bacteria and viruses) and animal pests (e.g. nematodes and insects).

Materials required: Purity box, stereo-binocular microscope, compound microscope, Petri dishes, watch glass, balance, sodium hydroxide (NaOH).

Procedure:

There are various ways of detecting the presence or absence of seed-borne organisms: 1. Inspection of dry seed (Ocular method) 2. Whole embryo count method 3. Blotter method 4. Agar plate method 5. Water agar plate method 6. Freezing method 7. Seedling symptom method, 8. Serological test, 9. Growing on test, 10. Enzyme-linked immunosorbent assay (ELISA), 11. Molecular biology methods (PCR), 12. Field trials, 13. Inspection of seed crops

1. Inspection of dry seed

- The seed sample is examined with the naked eye or hand lens stereo-microscope; the presence of fungi, if any, is observed and recorded. Fungi affects the physical appearance of the seeds:
- Fruiting structures of fungi are visible as *acervuli* or *pycnidia*; seeds are partly or completely smutted or bunted.
- Whole or broken ergot sclerotia are mixed with the seed.
- Spores or spore masses of fungi are visible on the seed surface (rusts, smuts, downy mildews, spores of other fungi – e.g. *Drechslera*, *Alternaria*, *Nigrospora*, *Curvularia*).
- Nematode galls are present. Inspection also reveals physical abnormalities (e.g. shrivelling of seed-coat, reduced or increased seed size, discoloration of or spots on seeds)

Procedure:

- Take a sample – the same size as that used for a purity test.
- Put seeds on the purity box and separate into pure seeds, inert matter and seeds of other crops.
- Weigh the three components and record details in the seed health report.
- Examine the pure seeds with the naked eye and then under a stereobinocular microscope.

- Pick out isolated spores using a thin brush or wet needle. Alternatively, dip the seed on the slide in a drop of water causing some spores to be released. This can be done under a stereo-binocular microscope.
- Record observations in the seed health report.

Observations and result:

Experiment No: 11

Seed health testing to the presence or absence of disease by wash test

Introduction: It is important to understand the distinction between seed pathology and seed health. Seed pathology – the study of seed-borne diseases, including the infection mechanism; seed transmission; role of seed-borne inoculum in disease development; techniques for detection of seed-borne pathogens; seed certification standards; deterioration due to storage fungi, mycotoxins and mycotoxicoses; and control of seed-borne inoculum.

Objectives: To determine the presence or absence of disease-causing organisms (e.g. fungi, bacteria and viruses) and animal pests (e.g. nematodes and insects).

Materials required: Compound microscope, balance, shaker, centrifuge, centrifuge tubes, capillary tubes, conical flasks, beakers, measuring cylinder, glass slides and cover slips, test tube racks, cheesecloth, Tween 20, mounting solution, haemocytometer.

Washing test

Seeds are washed and the suspension is examined. This test is used principally to detect fungi whose inoculum is present on the seed surface – e.g. teliospores of bunts and smuts, oospores of downy mildew, chlamydospores of *Protomyces macrosporus*, rust of sugar beet (*Uromyces betae*) and safflower (*Puccinia calcitrapae*).

Procedure:

- Take a working sample.
- Transfer seeds into a flask and add water until the seeds are submerged.
- Add 1–2 drops of Tween 20 (Polysorbate-type non-ionic surfactant).
- Place seeds on shaker for 5-10 minutes.
- Filter contents into a beaker through muslin or cheesecloth.
- Transfer contents into a centrifuge tube; put water in another centrifuge tube for control.
- Centrifuge contents at 1 500–3 000 for 2–10 minutes.
- Decant upper liquid and add 1 ml of water or mounting solution to the centrifuge tube.
- Mix contents with a needle.
- Insert a capillary tube; suck out a few drops and place on slides.
- Place cover slip over drops.
- Examine slide under a compound microscope.
- Pour contents into a Petri dish directly and examine the washings directly under a stereo-binocular microscope at 50X.
- Pick spores using capillary tubes and place on slide for compound microscope examination. This is useful when only a small number of seeds are available for testing (e.g. germplasm samples in quarantine clearance).
- Record results in the seed health report.

Observations and result:

Experiment No: 12

Seed health testing primarily to the presence or absence of disease by blotter method

Introduction: This is an incubation method (seeds are placed on moist blotters) with seeds incubated for 7 days at 22 °C under alternating cycles of light and dark. After incubation, the fungi developed are observed under a stereo-binocular microscope and identified based on the habit characteristics and morphology of the spores. Fruiting bodies are observed under a compound microscope. Normally 400 seeds are tested in the blotter method. It is recommended that one analyst examines 200 seeds and another analyst examines the other 200.

Objectives: To determine the presence or absence of disease-causing organisms (e.g. fungi, bacteria and viruses) and animal pests (e.g. nematodes and insects).

Materials required: Incubation room/incubator with racks equipped with blacklight tubes (for near ultraviolet light) fitted with an automatic timer for 12-h light/ dark and a deep-freezer (-20 °C). Eye protection glasses (for NUV lights), plastic disposable gloves and masks. Compound and stereo-binocular microscopes. Petri dishes, filter paper discs, trays (30 × 60 cm) for holding plates, trays and spoons of different sizes for sampling, container for water, glass slides and cover slips. Sodium hypochlorite, distilled water, measuring cylinders (25 ml, 25 ml), refrigerator, toolbox and cheesecloth.

Procedure:

- Prepare required number of Petri dishes.
- Disinfect surface of the plates.
- Place 5–50 seeds per plate on three wet blotter papers in circular pattern.
- Incubate dishes at the selected temperature (20–25 °C) – e.g. 22 °C for 7 days in alternating 12 h cycles of light and darkness.
- Examine fungi on seed under stereo-microscope.
- Observe growth characters and identify fungi.
- Count various fungi in each Petri dish and calculate the percentage of infected seed.

Observations and result:

Experiment No: 13

Visit to seed processing units

Q. 1.	Where did you visit a seed processing units?
Q. 2.	Which crop(s) was processed in this seed processing unit?
Q. 3.	Which are the important steps followed by the seed processing unit that you had visited?
Q. 4.	How seed were dried in the dryer?
Q. 5.	Explain the method of seed cleaning in details.
Q. 6.	How seed packaging and storage was done in the processing unit?
Q. 7.	What are the factors affecting seed longevity in seed storage?
Q. 8.	List out the equipments/apparatus used in the seed processing unit

Experiment No: 14

Visit to seed testing laboratory

Q. 1.	Which seed testing laboratory you have visited?
Q. 2.	Which types of tests were performed in seed testing laboratory?
Q. 3.	What are the key objectives of seed testing laboratory?
Q. 4.	How working samples were drawn in the testing laboratory for various tests?
Q. 5.	List out the equipments/apparatus and chemical used in the seed testing laboratory?
Q. 6.	How seed germination and seed moisture tests were conducted in the seed testing laboratory?
Q. 7.	How seed vigour and seed viability tests were performed in the seed testing laboratory?
Q. 8.	How many seed testing laboratories are working in the state? Name them along with their location.

Experiment No: 15

Visit to seed production area or seed orchard

Q. 1.	Where did you visit seed production area or seed orchard?
Q. 2.	Name of the species
Q. 3.	Seed Source and date of planting/sowing in seed orchards?
Q. 4.	Size of seed orchard?
Q. 5.	What are the different operations and management methods followed in seed orchard?

GLOSSARY

Abnormal seedlings- Seedlings which in a germination test show damages on critical structures of the embryo with likelihood that the capacity for continued development into a normal plants may not materialize. The critical structure(s) may be damaged, deformed, decayed, or show other defects

Air screen cleaner- The basic piece of equipment for cleaning seed, utilizing air flow and perforated screens for sieving action in the separation of the seed from inert materials, weed seed and other crop seed (using differences in the size, shape and weight of seed and that of the contaminants) resulting in cleaner seed of more uniform size.

Analytical purity - The percentage of the seed that is of the same crop species but not necessarily the same crop variety. The balance can include inert matter, weed seed, damaged seed. While regular seed testing procedures may not, in all cases, distinguish between different varieties of the same species, the seeds of different crop (species) can be identified in the seed laboratory by close examination of the seed.

Certified seed - Seed of a prescribed standard of quality produced under a controlled multiplication scheme either from basic seed or from a previous generation of certified seed. It is intended either for the production of a further generation of certified seed or for sowing to produce food, forage, etc.

Clone – A group of individuals (plants) of common ancestry which have been propagated vegetatively, usually by cuttings or by multiplication of bulbs or tubers.

Commercial seed – Seed which is intended for crop production, but has not been produced under a recognized certification scheme. Composite sample – A sample that is made by mixing together the primary samples drawn from containers of the seed lot for testing purposes.

Cultivar – synonymous with the term ‘variety’

Dormancy - The condition in which a seed with a viable embryo fails to germinate in conditions conducive to plant growth.

Embryo - The generative part of the seed that will develop into a plant.

Endosperm – The nutritive tissue within a seed but external to the embryo on which the developing seedling can draw nutrients until it is able to photosynthesize on exposure to light. F1 – The first generation arising from a cross between two genetically different parents, usually in-bred lines.

Foundation seed – The progeny of breeder seed; used as planting stock for registered and certified seed

Genetic purity – trueness to type or variety usually referring the specified crop variety as represented by seed.

Germination – Initiation of active growth of all essential embryonic parts required for a successful seedling establishment. In a seed test it is regarded as the emergence and development

from the seed of those essential structures which indicate the ability of the embryo to develop into a normal plant under favourable field conditions.

Germination capacity – The percentage of pure seed which germinate in a standard test to give normal seedlings as defined in the Rules for Seed Testing.

Hybrid vigour – The increase in vigour of hybrids over the their parental inbred lines; also known as heterosis

Inert matter – One of the four components of a purity test; it includes non seed material, straw, stones, and seed material which is classified as inert according to the Rules of Seed Testing.

Inbred – self-fertilized over several generations

ISTA – The International Seed Testing Association that with it member laboratories establishes the international standards and procedures for seed testing.

Isolation – The separation of the field of seed crop from the field of other crops in order to prevent mechanical or genetic contamination of the seed to be harvested. Isolation could be in form of distance, time and physical barrier such as plant species like Sesbania.

Moisture content – The weight of available water in a seed sample expressed as a percentage of the total weight of the seed at the time of determination.

Normal seedlings – Seedlings which in a germination test show the capacity for continued growth and development into normal plant.

Noxious weeds – A weed species that is defined by law as being noxious; usually highly objectionable when found in crop seed lots. Technically, it is a weed seed that is difficult to control by any known cultural means.

Off-type – A plant in a seed crop which deviates from the typical description for the cultivar.

Open pollinated variety – A heterogeneous variety of a cross pollinated crop which is allowed to inter-pollinate freely during seed production. In contrast to hybrid seed production representing controlled cross pollination.

Phyto sanitary certificate – A certificate issued by a legally constituted authority of federal or state government stating that a seed lot has been inspected and found to be free of quarantine disease infestation. These certificates are frequently used in international seed trade agreements to prevent the spread of seed borne diseases among countries.

Pollination – The transfer of pollen grains from an anther of a flower to a stigma of the same or another flower followed by fertilization of the ovule.

Primary sample – A small portion of seed taken from one point in a seed lot during the sampling process.

Progeny – offspring

Pure seed – Refers to the species stated on the label or found to predominate in the test and shall include all botanical varieties and cultivars of that species including both whole seed, immature seed, diseased seed, and seed larger than one-half their original size or as defined by ISTA rules for seed testing

Registered seed – A class of seed in a certified seed scheme which is produced from foundation seed and planted to produce certified seed.

Relative humidity – The ratio, expressed as a percentage of the quantity of water vapour actually present in the air to the greatest amount it could contain at that temperature.

Respiration – The metabolic process by which a plant oxidizes its food and provides energy for assimilation of breakdown products.

Rogue – A contaminant (cultivar, other species or weed) in a seed crop. Roguing is the process of removal rogues from the crop.

Sampling – The method by which a representative sample is taken from a seed lot to be sent to a laboratory for analysis.

Seed – The ripened ovule consisting of an embryonic plant together with a store of food or other structure including the ovule used by farmers as planting material.

Seedling – A young plant as it emerges from the seed until it is established physically and physiologically as a completely independent plant.

Seed lot – A quantity of seed of one cultivar, of known origin and history, and controlled under one reference number.

Seed equilibrium moisture content- the percentage of moisture in a seed at a particular temperature and relative humidity. Stamens – The parts of the flower which contain the anthers (represents the male part)

Seed vigour – Is the sum of the properties that determine the activity and performance of the seed lots of acceptable germination in a wide range of environmental conditions. A vigorous seed lot is one that is potentially able to perform well even under environmental conditions which are not optimal for the species.

Stigma – The surface to which pollen grains are transferred for fertilization of ovules (represents female part)

Submitted sample – Is a sample submitted to the testing laboratory. It must be of at least the size specified by ISTA regulations and may comprise either the whole or a subsample of the composite sample

Sub-sample – Is the portion of a sample obtained by reducing the sample using one of the sampling methods prescribed in ISTA regulations.

Variety – synonymous with the term ‘cultivar’ as defined in the International Code of Nomenclature for Cultivated Plants, 1980, Article 10: ‘The international term cultivar denotes an

assemblage of cultivated plants which is clearly distinguishable by a group of characters(morphological, physiological, cytological, chemical or other) and which when reproduced (sexually or asexually) retains its distinguishing characters’.

Weed - An unwanted plant appearing in a cultivated crop.

Working sample – The sample taken in a laboratory from a submitted sample and actually used in a test.