25. Purity analysis and germination test for seeds

Purity analysis

The purity test is the first test to be made. Seed samples can contain impurities such as weed seeds, seeds of other crop species, detached seed structures, leaf particles and other material. The object of purity analysis is to determine the composition of the sample being tested by weight. To do this, a purity test is conducted, in which the working sample is separated into the following component parts:

i) **Pure seed** refers to the species under consideration. In addition to mature, undamaged seed, it includes, undersized, shriveled, immature and germinated seeds, provided they can be definitely identified as the species under consideration, more over it includes pieces resulting from breakage that are more than one half their original size Pure seeds includes the following:

a. Intact seeds

b. Achenes and similar fruits like caryopsis, schizocarpand mericarp with or without pedicel, perianth and whether they contain true seed unless it is apparent and when difficult to identify.

c. Pieces of seeds, achenes, mericarp and caryopsis resulting from breakage that is more than half the original size (Half seed rule). However, seeds of Leguminosae, Cruciferae and Coniferae are considered as inert matter if their seed coat is removed.

d. Clusters of Beta or pieces of such clusters with or without seeds that are retained by 200 x 300 mm sieve.

e. Florets and caryopsis of Grammae.

Florets and one flowered spikelet's with an obvious caryopsis containing endosperm provided, also that, the caryopsis of particular genera and species have attained minimum sizes.

f. Free caryopsis

All florets and caryopsis (except broken florets and caryopsis half or less than half the original size and in the case of *Dactylis glomerata* excluding one-fifth of the weight of multiple floret in which the sterile floret extends to or beyond the tip of the fertile floret) remaining in heavy protein after blowing at an uniform blowing speed.

With reference to specific species

Allium sp., ***Capsicum*** sp., ***Cucumis*** sp., and ***Lycopersicon*** sp., seed with or without seed except pieces of seed more than 1/2 the original size with or without seed coat.

ii) **Other seeds** shall include seeds and seed like structures of any plant species other than that of pure seed.

iii) **Inert matter** includes seed units and all other matters and structures not defined as pure seed or other seed. It includes, seeds and seed like structures eg., achenes, caryopsis, mericarp and seeds of leguminaceae less than ½ the original size with no seed coat.

To perform purity analysis, the working sample is kept over the purity work board at the base end. A small quantity of sample is brought to the middle of the board and split into two basic components as pure seed and inert matter. The inert matter is further divided as pieces of seeds less than 1/2 the original size, stones, pieces of leaves, weed seeds, other crop seed etc. The pure seed is further divided into pure seed and other distinct variety (ODV) etc. The pure seed and inert matter are weighed upto three decimals and percentage worked out. The weed seed, OCS, ODV are counted and reported as number per kg.

Instruments

1. Seed blower

It is used to remove the light weighted inert matter from the seeds. Working sample is kept at the lower portion of the tube and the required uniform upward flow of air is regulated upto prescribed period of time. Lighter matter is separated from the sample by air flow and settle down in the partition provided in the tube of the blower. The tube is removed and inert matter is collected.

2. Diapanascope

The purity work board is provided with light source in the background which facilitates easy separation of different component. It also helps better distinguishing of red pericarp from white pericarp and short bold grains long slender grains from medium types.

The percentage by weight of each of the component parts shall be calculated to one decimal place. Percentage must be based on the sum of the weight of the components not on the original weight of the working sample, but the sum of the weights of the components must be compared with the original weight as a check against loss of material or other error. The result shall be reported to one decimal place and the percentage of all components must total 100. Components of less than 0.05% shall be reported as Trace. If the purity is less than the standard retirement the certification department will reject the seed.

Seed germination test

Principle

Germination tests shall be conducted with a pure seed fraction. A minimum of 400 seeds are required in four replicates of 100 seeds each or 8 replicates of 50 seeds each or 16 replicates of 25 seeds each depending on the size of seed and size of containers of substrate. The test is conducted under favourable conditions of moisture, temperature, suitable substratum and light if necessary. No pretreatment to the seed is given except for those recommended by ISTA.

Materials required

A. Substratum

The substratum serves as moisture reservoir and provides a surface or medium for which the seeds can germinate and the seedlings grow. The commonly used substrata are sand, paper and soil.

I. Sand

a. Size of sand particle

Sand particles should not be too large or too small. The sand particles should pass thorough 0.80 mm sieve and retained by 0.05 mm sieve.

b. Toxicity

Sand should not have any toxic material or any pathogen. If there is presence of any pathogen, found, then the sand should be sterilized in an autoclave.

c. Germination Tray

When we use the sand, germination trays are used to carry out the test. The normal size of the tray is 22.5 x 22.5 x 4 cm. They tray may either zinc or stainless steel.

B. Method of seed placement

1. Seeds in sand(s)

Seeds are planted in a uniform layer of moist sand and then covered to a depth of 1 cm to 2 cm with sand.

2. Top of sand (TS)

Seeds are pressed into the surface of the sand

C. Spacing

We must give equal spacing on all sides to facilitate normal growth of seedling and to avoid entangling of seed and spread of disease. Spacing should be 1-5 times the width or diameter of the seed.

D. Water

The amount of water to be added to the sand will depend on size of the seed. For cereals, except maize, the sand can be moistened to 50% of its water holding capacity. For large seeded legumes and maize sand is moistened to 60% water holding capacity.

II. Paper

Most widely used paper substrates are filter paper, blotter or towel (kraft paper). It should be have capillary movement of water, at vertical direction (30 mm rise / min.). It should be free from toxic substances and free from fungi or bacteria. It should \ hold sufficient moisture during the period of test. The texture should be such that the roots of germinating seedlings will grow on and not into the paper.

A. Methods

a. Top of Paper (TP)

Seeds are placed on one or more layers of moist filter paper or blotter paper in petridishes. These petridishes are covered with lid and placed inside the germination cabinet. This is suitable of those seeds which require light.

a. **Between paper (BP)**

The seeds are placed between two layers of paper

b. Roll towel method

The seeds are placed between two layers of paper and rolled in towels. The rolled towels are placed in a water source and kept in germinator or germination room.

c. Inclined plate method

Germination on glass plate with germination paper and kept at an angle of 45^0C.

III. SOIL

Should be non-caking, free from large particles. It must be free from weed seeds, bacteria, fungi, nematode and other toxic substances. Soil is not recommended for reuse.

B. TEMPERATURE

Normally most of the seeds germinate between 20-30^0 C.

C. LIGHT

Light requirement seeds should provided with light eg. Lettuce

Germination requirements for different crops

Crop	Substratum	Temp ^{0} C	First count (Days)	Final count (days)	Pre - treatment
Brinjal	TP,BP	20-30	7	14	EthreI (25 ppm) 48 hrs.
Tomato	TP,BP	20-30	5	14	
Chillies	TP,BP	20-30	7	14	(Hot water 85° C 1 min)
Bhendi	BP,S	20-30	4	21	
Onion	TP,BP	15-20	6	21	KN03
Carrot	TP,BP	20-30	7	14	KN03
Radish	TP,BP	20-30	4	10	Pre chill
Cabbage					Pre chill
Cauliflower	TP	20-30	5	10	Pre chill, KN03
Ash gourd	S	30-35	5	14	light
Biter gourd	BP,S	20-30	4	14	
Bottle gourd	BP,S '	20-30	4	14	-

TP- Top paper, BP-Between paper, S-Sand method

Germination apparatus

1. Germination Cabinet / Germinator

This is called chamber where in temperature and relative humidity is controlled. We can maintain the required temperature

2. Room germinator

It works with same principle of germinator. This is a modified chamber of larger one and the worker can enter into it and evaluate the seedlings. Provisions are made to maintain the temperature and relative humidity. This is used widely in practice.

3. Counting Board

This is used for accurate counting and spacing of seeds. This consists of 2 plates. The basal one is stationary and top one is movable. Both top and basal plates are having uniform number of holes viz., 50/100, when the plates are in different position. After taking the sample, the top plate is pulled in such a way that the holes are in one line so that the fixed number of seeds falls on the substratum.

4. Vacuum Counter

Consists of a head, pipe and wall. There are plates of 50 or 100 holes which can be fitted to the head. When vacuum is created the plate absorbs seeds and once the vacuum is released the seeds fall on the substrate.

5. Impression Board

Made of plastic / wood with 50 or 100 holes/pins. Here the knobs are arranged in equal length and space. By giving impression on the sand it makes uniform depth and spacing for seed.

D. Seedling Evaluation

ISTA classified the seedlings into different categories based on the development of essential structures

CATEGORIES OF SEEDLIGS

1. Normal seedlings
2. Abnormal seedlings
3. Hard seeds
4. Fresh ungerminated seeds
5. Dead seeds

1. ormal seedlings

Seedlings which show the capacity for continued development into normal plant when grown in favorable conditions of soil, water and temperature.

Characters of normal seedling

1. A well developed root system with primary root except in certain species of graminae which normally producing seminal root or secondary root
2. A well developed shoot axis consists of elongated hypocotyls in seedlings of epigeal germination.
3. A well developed epicotyls in seedlings of hypogeal germination.
4. One cotyledons in monocots and two in dicots
5. A well developed coleoptile in graminae containing a green leaf
6. A well developed plumule in dicots
7. Seedlings with following slight defects are also taken as normal seedlings. Primary root with limited damage but well developed seminal root system in leguminaceae (Pisum), graminae (maize), cucurbitaceae (cucumis) and malvaceae (cotton)
8. Seedlings with limited damage or decay to essential structures but no damage to conducting tissue
9. Seedlings which are decayed by pathogen but it is clearly evident that the parent seed is not the source of infection.

II. Abnormal Seedlings

Seedlings which do not show the capacity for continued development into normal plant when grown in favorable conditions of soil, water and temperature.

Types of abnormal seedling

A. Damaged seedlings