BIOSTATISTICS AND SEED**TECHNOLOGY**

SUBJECT CODE: 18MBO24E

UNIT - 5

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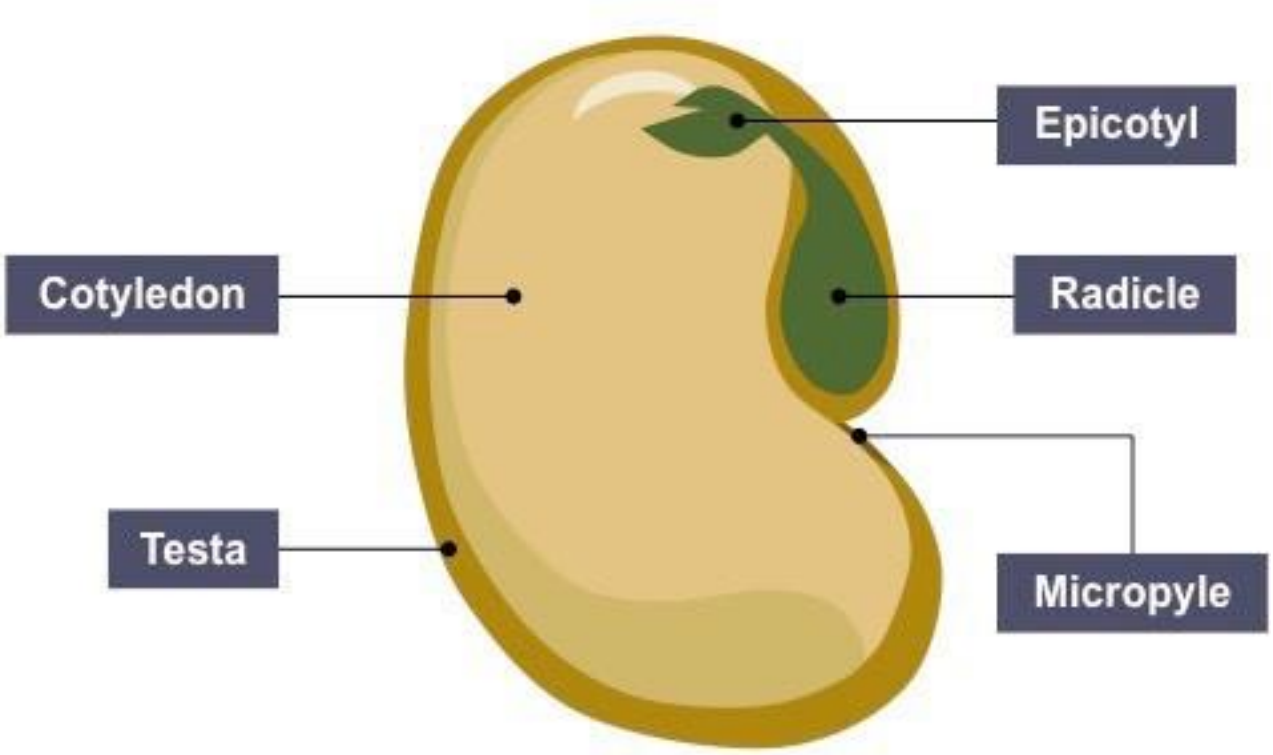
Structure of a Seed

Seeds of different plants may vary in many ways, but the basic anatomy remains the same. A typical seed consistsof the following parts:



Tesla: It is the outer coat of the seed thatprotects the embryonic plant.

Micropyle: It is a tiny pore in the testa thatlies on the opposite of the tip of the radicle. It permits water to enter the embryo before active germination.

Hilum: Is a scar left by the stalk whichattached the ovule to the ovary wall before it became a seed.

Cotyledon: In some plants, this containshigh quantities of starch and will providea source of food for the developing embryo prior to germination, in other

plants this role is performed by
an endosperm. In monocotyledons, thereis just one cotyledon whereas
in dicotyledons there are two. Dependingon the type of germination (epigeous or hypogeous) the cotyledons may remain below ground or be pulled above ground.

Radicle: This is the embryonic root which will develop into the primary root of the plant. It is usually the first part of the embryo to push its way out of the seed during germination.

Plumule: This is the embryonic shoot. It appears as a bud which will give rise to the shoot and the remaining structures inthe plant.

Endosperm: In many plants, a separate part for storage of starch develops and this is called the endosperm. It is seen inmaize and wheat.

Seed dispersal

Seed dispersal is the movement, spread or transport of seeds away from the parent plant. Plants have limited mobility and rely upon a variety of dispersal vectors to transport their propagules, including both abiotic vectors such as thewind and living (biotic) vectors like birds.

Seeds can be dispersed away from the parent plant individually or collectively, aswell as dispersed in both space and time. The patterns of seed dispersal are determined in large part by the dispersal mechanism and this has important implications for the demographic and genetic structure of plant populations, as well as migration patterns and species interactions. There are five main modes ofseed dispersal: gravity, wind, ballistic, water, and by animals. Some plants
are serotinous and only disperse their seeds in response to an environmental

stimulus. Dispersal involves the letting goor detachment of a diaspore from the main parent plant.

Animal dispersal (zoochory)

Many animal-dispersed fruits are dispersed by vertebrates—especially certain mammals and birds, although fishand reptiles can also act as dispersal agents—or ants. Vertebrate-dispersed fruits and seeds may be fleshy, or may have fleshy coverings; ant-dispersed seeds often have nutrient-rich appendages.

Wind dispersal (anemochory)

Fruits and seeds that are wind-dispersed frequently have modifications that help slow their descent to the ground and increase the chances that they will be blown laterally by air currents, so that theydo not land directly beneath or next to their parent plant. Seed modifications for wind dispersal can include small size and/or light weight, wings, hairs, and/or inflation.

One of the most obvious modificationsfor wind-dispersal is the wing. Winged fruits are common in the fossil record

beginning in the Paleogene. Winged fruitsor seeds often have a single wing, in which case the wing may be asymmetrical, or offset to one side of the fruit or seed. If they have more than one wing, the wings many be regularly arranged around the fruit or seed. Often, the structure of the wing or wings will cause a seed or fruit to spin or rotate as itfalls (known as autorotation, i.e., self-rotation). Maples (Acer) produce familiar wind-dispersed fruits that spin as they fall. If you live in a neighborhood withmaple trees, you can observe this yourself; watch the mature fruits as they fall from a tree on a windy day, or pick up the fallen mericarps (fruit halves) and drop them to watch them spin as they fall.

Water dispersal (hydrochory)

Plants that live in wetland environments or near the ocean may have buoyant, or floating, fruits or seeds. Cranberries (*Vaccinium* species) are low-growing plants found in boggy environments. Their bright red berries arenot particularly sweet, and thus probablynot terribly attractive to animals. Cranberries do, however, float, which aidsin their dispersal in wetland habitats. It has been hypothesized that cranberries

evolved from ancestors that had more palatable, animal-dispersed fruits. Humans take advantage of the berries' buoyancy during commercial production,as cranberry bogs can be flooded so thatthe floating berries can be more easily collected.

Some plants with floating fruits or seeds can disperse long distances over the ocean. The most obvious example of this is the coconut palm (*Cocos nucifera*), which has large, fibrous fruits that canfloat to and colonize oceanic islands. Similarly, legumes in the genus *Entada* produce large, buoyant seeds; each seed harbors an air pocket, which enhances it ability to float.

Explosive dispersal (autochory or bolochory)

Amongst the more novel and exciting ways in which seeds are dispersed is through ballistic or explosive dispersal. Inthis method of dispersal, the fruit forciblyejects the seed(s), scattering them for a short distance. The common garden plant (also known as balsam, touch-me-not, and jewelweed, amongst other names) is one such plant. It produces capsules. When ripe, an animal

brushing by the plant can cause the capsule to open instantly, scattering theseeds. Another plant with dramatic explosive seed dispersal is the squirting cucumber (*Ecballium elaterium*), which ejects its seeds as the fruit detaches fromits stalk.

Seed Germination Types

The three main types are:
(1) Hypogeal Germination (2) Epigeal Germination and (3) Vivipary (Viviparous Germination).

Type # 1. Hypogeal Germination:

In this kind of germination, the cotyledonsdo not come out of the soil surface. In such seeds the epicotyl (i.e., part of embryonic axis between plumule and cotyledons) elongates pushing the plumule out of the soil. All monocotyledons show hypogeal germination . Among dicotyledons, gram, pea , groundnut are some common examples of hypogeal germination.

In monocotyledons (e.g., wheat, maize, rice, coconut) radicle and plumule come out by piercing the coleorrhiza and coleoptile respectively. The plumule growsupward and the first leaf comes out of thecoleoptile. The radicle forms the primary root which is soon replaced by many fibrous roots.

Type # 2. Epigeal Germination:

In seeds with epigeal germination, the cotyledons are brought above the soil dueto elongation of the hypocotyl. In castor, cotton, papay, onion (Figs. 4.7., 4.8), flat green leaf like cotyledons can be seen in the young seedlings. Here the cotyledons,besides food storage, also perform photosynthesis till the seedling becomes independent. In some other plants like bean, the cotyledons being thick, do not become leaf-like; they shrival and fall off after their food reserves are consumed by the seedling.

Type # 3. Vivipary (Viviparous Germination):

Vivipary is the phenomenon of giving birthto young ones in advanced stage of development. It occurs in mammals (among animals) and mangrove plants. Inmangrove plants (e.g., Rhizophora, Sonneratia, Heritiera) the seeds cannot germinate on the ground because of the excessive salt content and lack of oxygenin marshy habitat. In such plants seed dormancy is absent.

The embryo of the seed (present inside the fruit) continues growth while the latteris attached to the parent plant. Hypocotyl elongates and pushes the radicle out of the seed and the fruit. Growth continues till the hypocotyl and radicle become several centimetres long (more than 70 cm in Rhizophora). The seedling becomesheavy.

As a result it breaks its connection with the fruit and falls down in the salt rich muddy water in such a position that the plumule remains outside the saltish waterwhile the tip of the radicle gets fixed in themud. This protects the plumule. The radicle quickly forms new roots and establishes the seeding as a new plant.

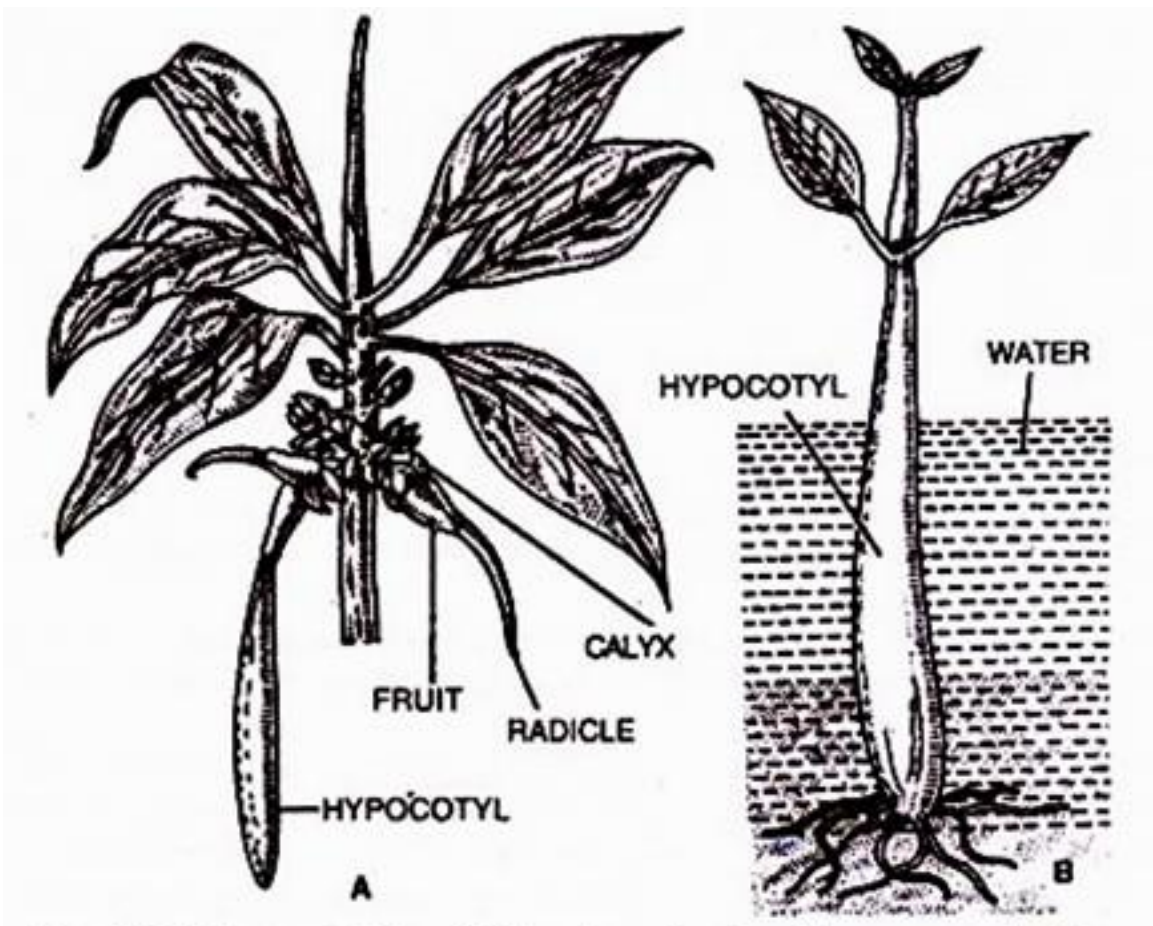


Fig. 4.9. Vivipary. A. twig of *Rhizophora* showing viviparous germination, **B.** A seedling has become established on tidal soil.

15. SEED VIGOUR TESTING

P.C. GUPTA

Seed vigour is an important quality parameter which needs to be assessed to supplement germination and viability tests to gain insight into the performance of a seed lot in the field or in storage. Several definitions have been offered to explain seed vigour. Looking into the complexity of the situation, the ISTA congress in 1977 adopted the definition of seed vigour as " the sum total of those properties of the seed which determine the level of activity and performance of the seed or seed lot during germination and seedling emergence". Although differences in physiological attributes of seed lots can be demonstrated in the laboratory, it was recommended that the term should be used to describe the performance of seeds when sown in the field (Perry, 1984a).

As the germination test is conducted in an optimum condition specific to different species, it is not always possible to get an idea of the performance of a seed lot in the field on the basis of germination test in the laboratory. It is mainly because of the reason that field conditions are seldom optimum and the emerging seeding suffers from one or the other kind of stress. In many cases seed lots having similar laboratory germinations may give widely differing field emergence values. Similarly, two seed lots having the same germination percentage in the laboratory may age differently when stored under ambient condition. These two situations indicate the incompletences of germination test in assessing the performance of a seed lot in the field or storage. This offers scope and possibility to determine vigour of a seed lot so that its field and storage performance can be assessed.

Seed vigour is still a concept rather than a specific property of a seed or seed lot. Several factors like; genetic constitution, environment and nutrition of mother plant, maturity at harvest, seed weight and size, mechanical integrity, deterioration and ageing and pathogens are known to influence seed vigour (Perry, 1984a). Therefore, care has to be exercised in selecting a seed vigour test to do the job. Two criteria have been employed by the ISTA seed vigour committee to evaluate the performance of seed vigour test methods for different crops :

(i) Reproducibility of vigour method

(ii) The relationship between vigour test results and seedling emergence in field soil.

There is no universally accepted vigour test for all kinds of seeds. The determination of following vigour tests will be useful in gaining additional information on seed quality.

1. Growth Tests

Principles : Growth tests are based on the principle that vigorous seeds grow at a faster rate than poor vigour seeds even under favourable environments. Vigorous seeds rapidly germinate, metabolize and establish in the field. Therefore, any method used to determine the rapidity of growth of the seedling will give an indication of seed vigour level.

Apparatus and equipment : All the equipments and materials needed to conduct a germination test are required. Additionally, a top loading balance and an air oven are also required.

Procedure

(a) First count : The test is done along with the regular germination test. The number of normal seedlings, germinated on the first count day, as specified in the germination test for each species, are counted. The number of normal seedlings gives an idea of the level of seed vigour in the sample. Higher the number of normal seedlings greater is the seed vigour.

(b) Seedling growth rate and dry weight : The seedlings are grown either in laboratory, green house or field. In laboratory, in between rolled towel paper method should be followed. Ten seeds are planted in the centre of the moist towel papers in such a way that the micropyles are oriented towards bottom to avoid root twisting. The rolled towel papers are kept in the germinator maintained at a temperature recommended for crop in reference. After a specified period of time (5-10 days) towel papers are removed and five seedlings are selected, their length is measured and mean seedling length is calculated. Seed lots producing the taller seedlings are considered more vigorous than the seed lots producing shorter seedlings. For dry weight determination, the seedlings are removed and dried in an air oven at 100°C temperature for 24 hours. The seedling dry weight provides additional information for assessing seed vigour.

(c) Speed of germination : One hundred seeds each in four replications are planted in recommended substratum for germination. The substratum is kept in a germinator maintained at recommended temperature for the crop in reference (Table 5.1). Number of seedlings emerging daily are counted from day of planting the seeds in the medium till the time germination is complete. Thereafter a germination index (G.I.) is computed by using the following formula :

$$G.I. = \frac{n}{d}$$

where, n = number of seedlings emerging on day 'd'

d = day after planting

The seed lot having greater germination index is considered to be more vigorous.

Example

Seed lot A — No. of seedlings= 0,0,0,40,30,12,7, counted

Day of counting = 1,2,3,4,5,6,7

Seed lot B — No. of seedlings =0,0,0,0,30,42,21 counted

G.I. of Seed lot A

$$\frac{0}{1}+\frac{0}{2}+\frac{0}{3}+\frac{40}{4}+\frac{30}{5}+\frac{12}{6}+\frac{7}{7}$$

$$= 10 + 6 + 2 + 1$$

$$= 19$$

G.I. of Seed lot B

$$\frac{0}{1}+\frac{0}{2}+\frac{0}{3}+\frac{0}{4}+\frac{30}{5}+\frac{42}{6}+\frac{21}{7}$$

$$= 6 + 7 + 3$$

$$= 16$$

In this example seed lot A has greater G.I. (19) than seed lot B (16), so seed lot A is more vigorous than seed lot B.

(d) Seed vigour index (S.V.I.) : This is calculated by determining the germination percentage and seedling length of the same seed lot. Fifty seeds each in four replications are germinated in towel papers as prescribed for the crop species in germination test. While evaluating the number of normal seedlings at the time of final count, the seedling length of 5 randomly selected seedlings are also measured. Seed vigour index is calculated by multiplying germination (%) and seedling length (mm). The seed lot showing the higher seed vigour index is considered to be more vigorous (Abdul-Baki and Anderson, 1973).

Example

Seed lot	% germination	Seedling length, mm	Vigour index
A	96	85	8160
B	95	76	7220
C	94	71	6674

In this example seed lot A is the most vigorous and seed lot C the least vigorous as they have the highest and the lowest values of seed vigour index, respectively.

2. Conductivity Test

Principle : Weakening of cell membrane in poor vigour seeds causes leakage of water soluble compounds like sugars, amino acids, electrolytes etc. when immersed in water. On the other hand, fresh seeds having intact membrane leach less quantity of these chemicals. The measurement of electrical conductivity (EC) of the leachate by a good and sensitive conductivity meter gives an accurate estimation of membrane permeability. The EC has been positively correlated with the emergence percentage of peas and broad beans (Mathews and Bradnock, 1968). The value of this test appears to be restricted to the large seed species of the Leguminoceae (Perry, 1984b).

Apparatus and equipment : Conductivity meter, beaker, 0.1% mercuric chloride, distilled water, seed sample, wash bottle an tissue paper.

Procedure : A seed sample of 2-5 gram is weighed and surface sterilized with 0.1% $HgCl_2$ for 5-10 minutes. The sample is washed thoroughly in distilled water. The clean seeds are immersed in 100 ml of water at 25 ± 1°C temperature for 10-12 hours. After this the seeds are removed with a clean forcep. The steep water left is decanted and is termed as leachate.

The conductivity meter is warmed for about 30 minutes before testing. First the conductance of distilled water is measured in a beaker. The electrode is then cleaned with a tissue paper and conductance of the leachate is read. The electrode is thoroughly washed using a wash bottle and wiped with a clean tissue paper before reusing. While recording the conductance, the lower bulb of the electrode should be fully emerged in the leachate. To get the EC of leachate the reading of distilled water is substracted from the sample reading. The value is then corrected for the temperature and multiplied by the cell constant factor. The reading is expressed as mu mhos/cm/g of seed. Lower the value of EC greater is the seed vigour.

3. Hiltner Test (Brick gravel test)

Principle : The test was developed by Hiltner in Germany in 1917. He observed that the seeds of cereal crops affected by Fusarium disease were able to germinate in regular test but were not able to emerge from brick gravels of 2-3 mm size. Compared to this, healthy seeds were able to emerge from the brick gravel (Robersts, 1972). The principle is that the weak seedlings are not able to generate enough force to overcome the pressure of brick gravels, so this method can be used to differentiate vigour levels in cereal seeds. Perry (1984b) found this method reproducible and associated with field emergence in case of wheat.

Apparatus and equipment : Germination box, aluminium tray, sand, sand marker brick gravel of 2-3 mm size, germinator, seed sample.

Procedure : The sand is seived, moistured and filled in the germination box leaving about 3 cm empty at the top. One hundred seeds are placed in each box in the impressions made by a sand marker. After this 2-2.5 cm of porous brick gravel is spread over the seeds. The box is kept in the germinator at appropriate temperature. After the period required for germination, the box is removed and the seedlings which have emerged through the brick gravel layer are counted. The percentage of emerged seedlings are used to compare seed vigour of different lots. The test should be repeated 3-4 times to get authentic value.

4. Paper Piercing Test

Principle : The principle of paper piercing test is similar to that of brick gravel test. High vigour seed lots are expected to produce strong seedlings which can pierce a particular type of paper while seedlings of poor vigour lots may not be able to pierce the paper. Therefore, the seedlings which emerge by piercing the paper

are more vigorous than those which are not able to emerge through the paper.

Apparatus and equipment : All the material required for conducting germination test in sand boxes or trays plus the special paper which should have the following characteristics :

(a) Basic weight = 90 g/m^2

(b) Thickness = 0.4 mm

(c) Bulk = 4

(d) Dry bursting strength = 0.3 kg/cm^2

(e) Breaking length = 1000-5000 mm

(f) Filtering speed = 500 ml/minute

(g) Wet bursting strength = 150 mm

(h) Ash content = 0.1%

(i) Fibre composition = Chemical wood pulp with high alpha percentage

Procedure : The cereal seeds are placed on 1.5 cm moist sand in a tray or sand box. The seeds are covered with specially selected dry filter paper, which is then covered with 2 cm of moist sand. After this, the sand boxes/trays are kept in a germinator maintained at 20°C temperature for 8 days. After 8 days sand boxes/trays are taken out and seedlings emerging above the paper are counted. A seed lot having maximum number of seedlings coming out of paper is considered to be most vigorous. *The test is highly dependant on the quality of paper and should be used when such papers are available.*

5. Cold Test

Principle : The cold test has been developed in USA to evaluate the seed vigour of maize (corn). In USA when the corn is planted in late spring, the soil is humid and cold. The weak seeds do not germinate and establish. Therefore, to simulate the actual field conditions witnessed at the time of corn planting, cold test

has been developed. The test aims to differentiate between weak and vigorous seed lots by subjecting them to low temperature prior to germination at optimum temperature. The test has been criticized for using field soil which greatly varies from place to place.

Apparatus and equipment : Aluminium tray, field soil, sand marker, germinator, seed sample.

Procedure : After grinding and properly sieving the soil is filled in tray upto 2 cm depth. Fifty seeds are placed over the sand and covered with another 2 cm thick layer of soil. The soil is compacted and enough water is added to make the soil about 70% of its water holding capacity. The temperature of the water should be 10°C. After watering the trays are covered with polythene bags and placed in the refrigerator maintained at 10°C temperature for one week. After one week the trays are removed and placed in the germinator at 25°C temperature. The seedlings emerged after 4 days are counted. The germination percentage is computed by counting the number of normal seedlings as in germination test. Higher the germination percentage greater is the vigour.

6. Accelerated Ageing Test

Principle : The accelerated ageing test has been developed at the Seed Technology Laboratory, Mississippi State University, USA for determining the storage potential of seed lots. The ageing process is accelerated by subjecting the seeds to high temperature and relative humidity in a chamber before standard germination. The seed lots that show high germination in accelerated ageing test are expected to maintain high viability during ambient storage as well. Thus, ageing test gives an indication of the performance of the seed lot during ambient storage. Tests conducted at Pantnagar with Bragg soybean seeds have shown positive relationship between 3 days accelerated ageing test (42- 45°C temperature, 95-100% R.H.) and viability after 6 months of ambient storage (Gupta, 1980). However, Perry (1984b) reported inconsistency in accelerated ageing test results and not well related to field emergence of maize and soybean. The test also suffers from fungal growth on seeds at high temperature and humidity (Agrawal, 1987). This test is recommended for soybean seeds.

Apparatus and equipment : Acceleratged aging chamber, equipment for germination test, seed samples, tight jar, muslin cloth, wire mesh etc.

Procedure : One hundred seeds each in four replications are tied in a fine muslin cloth. The tied seeds are placed in jar on a wire mesh. The lower part of the jar is filled with water. There should not be a direct contact between water and the seed. The jar is covered with the lid and sealed with parafin wax to make it air tight. The jar is then placed in the accelerated aging chamber maintained at 45 ± 2°C temperature for 3-5 days. The jar is removed after this period and the seeds are cooled in a dessicator. The seeds are then tested in a normal germination test specific to different crops. The percent germination gives level of seed vigour. Higher the germination percentage greater is the vigour of the seed.

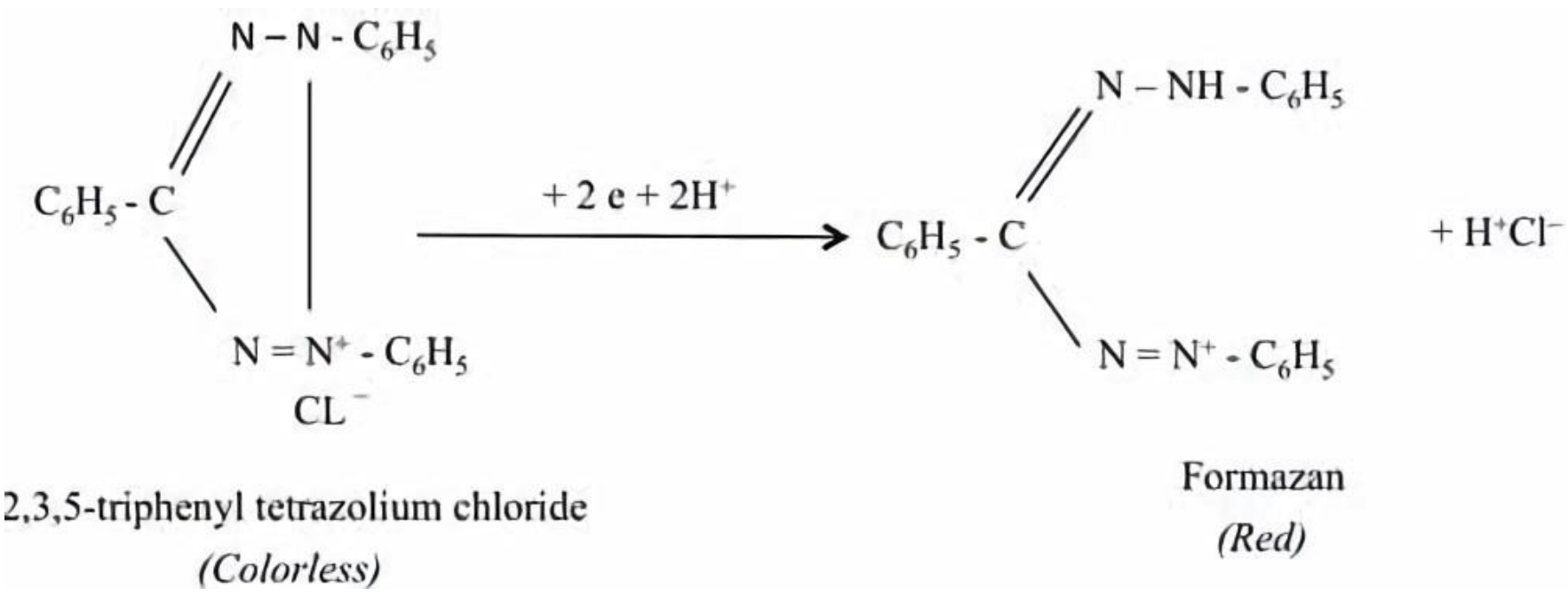
Future Role of Seed Vigour Testing

Seed vigour is an important component of seed quality and satisfactory levels are necessary in addition to traditional quality criteria of moisture, purity, germination and seed health to obtain optimum plant stand and high production of crops. As agricultural and horticultural techniques become progressively more sophisticated, the need for high vigour seeds will increase and testing standards, similar to those reconginzed for germination will be required (Perry, 1984b). The technology of seed vigour testing has not been perfected so far, so much so that there is not a single universally accepted seed vigour test method. Research is needed to further refine the current seed vigour test methods and to develop new methods which are more related to field/storage conditions.

Tetrazolium Testing

Tetrazolium (TZ) testing is a rapid method(can be finished within less than two days) for the evaluation of seed viability.
This method has been widely used by seed scientists to assess germination potentials, to determine the extent of seeddamage, and to evaluate seed vigor and/ or other seed lot problems.

The principle of TZ testing is based on the presence of dehydrogenase activity in viable seed tissues during the respiration process. Dehydrogenase can catalyze thecolorless 2,3,5 triphenyl tetrazolium chloride solution into a red dye formazan (Figure 1). Therefore, living tissues of seeds that imbibe tetrazolium chloride willbe stained red, while dead tissues will retain their natural color (Figure 2).



2,3,5-triphenyl tetrazolium chloride
(Colorless)

Formazan
(Red)

Figure 1. The principle of TZ testing (Elias et al., 2012).

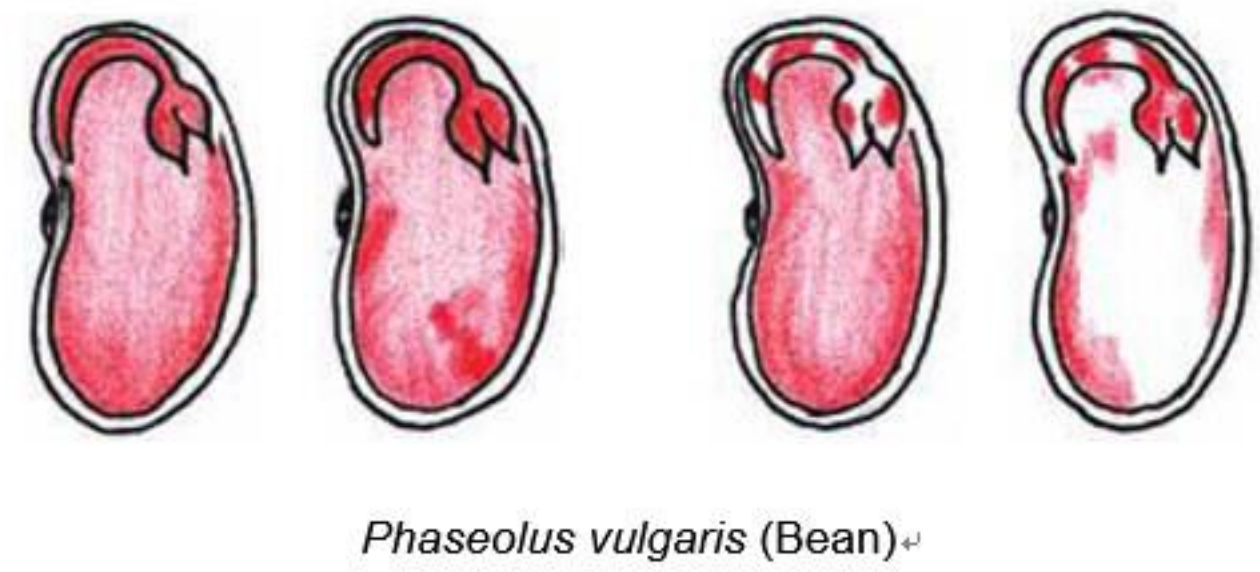


Figure 2. Examples of TZ testing for seed viability test (Elias et al., 2012). (Left two: viable seeds; Right two: non-viable seeds)

A standard TZ testing assay involves the following steps:

Preparation of dry seed. The seed coats of many species are best to be treated by mechanical abrasion so that water or solutions can penetrate into the interior tissues of the seed.

Moistening. This step aims to hydrate the seeds to activate the respiratory enzymesand to soften the tissues for cutting and piercing.

Preparation for staining. Cut or pierce the

seed to facilitate the absorbing of TZ solution by internal tissues.

Staining. Soak the cut seeds in TZ solution for certain periods of time.

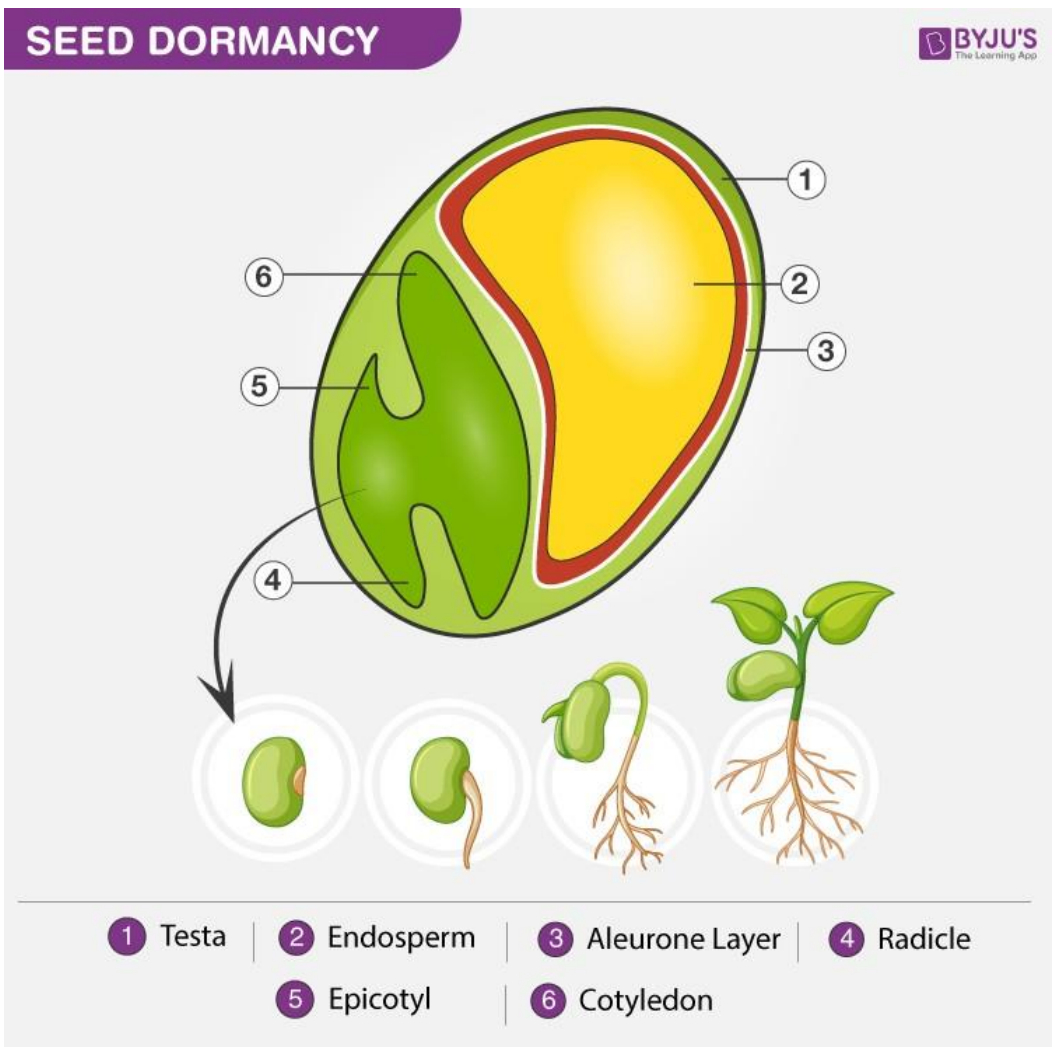
Evaluation of staining patterns. Examine the seed for a color change in the embryo.

Seed Dormancy

Seed dormancy can be defined as the state or a condition in which seeds are prevented from germinating even under the favourable environmental conditionsfor germination including, temperature, water, light, gas, seed coats, and other mechanical restrictions.

The main reason behind these conditionsis that they require a period of rest beforebeing capable of germination. These conditions may vary from days to monthsand even years. These conditions are the combination of light, water, heat, gases, seed coats and hormone structures.

Reasons or Causes of the Seed Dormancy



There are certain major causes for the seed dormancy. Listed below are the fewreasons for the seed dormancy.

Light

Temperature

Hard Seed Coat

Period after ripening Germination

inhibitors Immaturity of the seed

embryo

Impermeability of seed coat to water

Impermeability of seed coat to oxygen

Mechanically resistant seed coat Presence of

high concentrate solutes

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Molecular Plant
Review Article

Two Faces of One Seed: Hormonal Regulation of Dormancy and Germination

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http://dx.doi.org/10.1016/j.molp.2015.08.010

ABSTRACT

Seed plants have evolved to maintain the dormancy of freshly matured seeds until the appropriate time for germination. Seed dormancy and germination are distinct physiological processes, and the transition from dormancy to germination is not only a critical developmental step in the life cycle of plants but is also important for agricultural production. These processes are precisely regulated by diverse endogenous hormones and environmental cues. Although ABA (abscisic acid) and GAs (gibberellins) are known to be the primary phytohormones that antagonistically regulate seed dormancy, recent findings demonstrate that another phytohormone, auxin, is also critical for inducing and maintaining seed dormancy, and therefore might act as a key protector of seed dormancy. In this review, we summarize our current understanding of the sophisticated molecular networks involving the critical roles of phytohormones in regulating seed dormancy and germination, in which AP2-domain-containing transcription factors play key roles. We also discuss the interactions (crosstalk) of diverse hormonal signals in seed dormancy and germination, focusing on the ABA/GA balance that constitutes the central node.

Keywords: seed dormancy, germination, ABA, GA, auxin, crosstalk

Shu K., Liu X.-d., Xie Q., and He Z.-h. (2016). Two Faces of One Seed: Hormonal Regulation of Dormancy and Germination. Mol. Plant. 9, 34–45.

INTRODUCTION

Seed dormancy is crucial to plant survival and ensures that seeds germinate only when environmental conditions are optimal. It thus is an adaptive trait in numerous seed-plant species, enabling wild plants to survive under stressful conditions in nature (Finkelstein et al., 2008). Most crops have been domesticated from wild species and show decreased levels of seed dormancy compared with their wild relatives, which ensures higher emergence rates after sowing (Lenser and Theissen, 2013; Meyer and Purugganan, 2013). However, the inappropriate loss or release of seed dormancy results in the rapid germination of freshly matured seeds or even pre-harvest sprouting (vivipary) in crops (Figure 1), causing substantial losses in yield and quality in agricultural production in addition to problems including post-harvest management and subsequent industrial utilization (Simsek et al., 2014).

Induction, maintenance, and thereafter release of seed dormancy are important physiological processes in seed plants. The ecological significance of seed dormancy includes preventing germination out of season, and consequently decreasing competition within species and ensuring plant survival under stressful conditions. As a complex and mysterious biological question, seed dormancy has attracted increasing attention from multi-disciplinary researchers, including plant biologists, crop geneticists, breeders, and food scientists. Nevertheless, it remains ‘‘one of the least understood phenomena in seed biology’’ (Finkelstein et al., 2008), despite considerable progress over past decades (Graeber et al., 2012; Rajjou et al., 2012). In this review, we summarize the mechanisms underlying the regulation of seed dormancy and germination, and focus on the emerging findings concerning the phytohormone network controlling this transition, mostly from studies with the model plant *Arabidopsis thaliana*.

Published by the Molecular Plant Shanghai Editorial Office in association with Cell Press, an imprint of Elsevier Inc., on behalf of CSPB and IPPE, SIBS, CAS.

Figure 1. Representative Image of the Pre-harvest Sprouting Phenotype of Rice in the Field.
Pre-harvest sprouting of crops often occurs when mature plants encounter prolonged rainfall and high humidity during the harvest season, which decreases yields and grain quality and also causes problems in industrial process. Red arrows indicate sprouting seeds on panicles.

DISTINCT PROCESSES OF SEED DORMANCY AND GERMINATION

Seed dormancy and germination has been studied intensively and extensively in the past; however, what constitutes seed dormancy at the molecular level remains largely unknown. Here, we attempt to address this question from a new viewpoint based on recent progress.

Seed dormancy ensures that seeds germinate at the appropriate time. Therefore, during maturation, the embryo must be kept in a quiescent state, mobilizing almost no stored nutrients and undergoing no cell division or elongation. In this quiescent state, germination-promoted genes are not actively expressed. Therefore, the radicle does not penetrate the testa and endosperm. It is now widely recognized that the chromatin structure determines gene expression and thereby regulates multitudinous developmental processes. In recent years, many genes associated with chromatin remodeling have been reported to regulate seed dormancy and germination (Liu et al., 2007; Saez et al., 2008; Wang et al., 2011a; Cho et al., 2012; Zheng et al., 2012). Emerging evidence shows that ABA (abscisic acid) is also involved in chromatin remodeling (Chinnusamy et al., 2008). For example, the histone methyltransferase gene *KYP/SUVH4* is repressed by ABA (Zheng et al., 2012), while histone acetyltransferase *HvGNAT/MYST* is induced by ABA (Papaefthimiou et al., 2010), and the epigenetic regulators *HUB1* and *RDO2* are strikingly up-regulated during the induction of seed dormancy (Liu et al., 2011). These investigations indicated that the epigenetic regulatory-related genes possess key roles during seed maturation, which thereafter affect the seed dormancy establishment process (Figure 2).

We propose that subsequently, during the germination process, seed dormancy may be related to a characteristic chromatin structure in certain regions of chromosomes in the seed, where germination-promoted genes cannot be activated even in the presence of related transcription factors because their binding sites are unavailable due to steric hindrance, with phytohormones also involved in this process. In contrast, dormancy release leading to germination is a process in which the chromatin structure is modified by cold stratification or after-ripening treatments, making the germination-promoted genes available for transcription, resulting in cell elongation and division, seed coat and endosperm rupture, and finally emergence of the radicle when conditions are favorable.

Although dormancy is established during seed maturation, whereas exogenous ABA application (or even maternal ABA in the plant during seed development) only inhibits seed germination but fails to induce seed dormancy; only ABA synthesized by the seed can establish dormancy (Kucera et al., 2005). Thus, the differently localized ABA in plant tissues possesses distinct effects on seed dormancy or germination. In addition, ABI5 is an important positive regulator in the ABA-signaling pathway, and its loss-of-function mutant *abi5* is insensitive to ABA-mediated inhibition of seed germination; however, *abi5* does not show altered seed dormancy (Finkelstein, 1994; Brocard-Gifford et al., 2003; Finkelstein et al., 2008). Furthermore, DOG1 (Delay of Germination 1) is a key player in the induction and maintenance of seed dormancy, but ABA sensitivity is unchanged in *dog1* (Nakabayashi et al., 2012). A new study demonstrated that DOG1 mediates a conserved coat-dormancy mechanism including the temperature- and gibberellin (GA)-dependent pathways (Graeber et al., 2014). Subsequent studies suggested the importance of epigenetic regulation for DOG1. Histone demethylases LDL1 (LYSINESPECIFIC DEMETHYLASE LIKE 1) and LDL2 repress seed dormancy by regulating *DOG1* (Zhao et al., 2015), and chromatin remodeling of DOG1 is also involved in dormancy cycling (Footitt et al., 2015). Furthermore, the histone methyltransferases KRYPTONITE (KYP)/SUVH4 and SUVH5 repress *DOG1* and *ABI3* transcription during seed maturation (Zheng et al., 2012) (Table 1). These studies demonstrated that the DOG1-mediated regulation pathway might be distinct from the ABA and/or GA pathway (Figure 2). These observations suggest that distinct signaling pathways may be adopted in the regulation of seed dormancy and seed germination.

ABA AND GA, THE MAJOR DETERMINANTS: NEWLY EMERGING EVIDENCE

It is widely recognized that ABA and GA are the primary hormones that antagonistically regulate seed dormancy and germination (Gubler et al., 2005; Finkelstein et al., 2008; Graeber et al., 2012; Hoang et al., 2014; Lee et al., 2015a). During seed maturation, endogenous ABA accumulates in the seed, inducing and maintaining seed dormancy and thus preventing

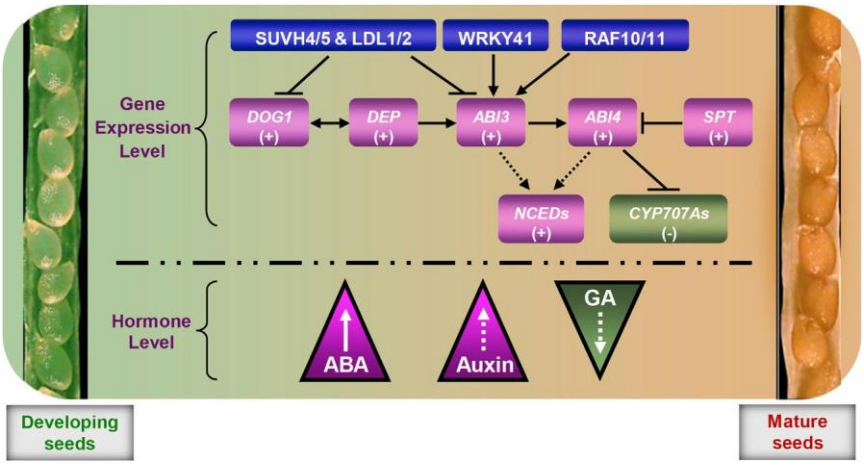

Figure 2. Changes in Accumulation of Key Hormones and Expression of Key Regulators during Seed Maturation.

Several key regulators are involved during the seed maturation process, and constitute a complex network. At the gene expression level, transcription levels of the two important ABA catabolic genes *CYP707A1* and *CYP707A3* are down-regulated, while the ABA biosynthesis genes including the *NCEDs* genes are up-regulated, by ABI4 and other regulators, thus ABA accumulates to initiate dormancy. The other key dormancy-controlling regulator genes, including *ABI3*, *ABI4*, *DOG1*, *DEP*, and *SPT*, are activated during seed maturation to induce and maintain primary seed dormancy, and some of these genes interact with each other to regulate seed dormancy levels. Among them, SUVH4, SUVH5, LDL1, and LDL2 negatively regulate *DOG1* and *ABI3* transcription, while WRKY41 and RAF10/11 directly control *ABI3* expression. At the phytohormone level, ABA accumulates and seed dormancy is initiated, established, and maintained during seed development. However, the genetics of whether auxin biosynthesis is up-regulated and GA biosynthesis is down-regulated is not yet understood. The active lines with upward arrows indicate the change of ABA level, while dashed lines indicate the changes of auxin and GA level. The symbol (+) indicates the elevated transcription level, while (–) indicates the decreased expression level during seed maturation. The black arrows and bars indicate the positive and negative regulatory roles, respectively.

vivipary (Figure 2). In contrast, before the onset of the germination process the endogenous ABA level in the seed is down-regulated, while the GA content is up-regulated with imbibition and stratification treatments.

ABA is a major inducer and protector of seed dormancy (Vaistij et al., 2013). Seeds of typical ABA-deficient mutants germinate faster than the wild-type (Frey et al., 2011), and transgenic plants constitutively expressing the ABA biosynthesis gene maintain deep seed dormancy (Martinez-Andujar et al., 2011; Nonogaki et al., 2014). Conversely, ABA catabolism mutants accumulate high ABA levels and thus cause hyperdormancy in seeds (Matakiadis et al., 2009) (Table 1). In addition to ABA biogenesis, the ABA-signaling-dependent pathway also affects seed dormancy. During the seed germination process, ABA signaling must be desensitized, whereby the membrane-associated transcription factor peptidases S1P (Site-1 Protease) and S2P, process the bZIP17 protein from the endoplasmic reticulum (ER) to the Golgi and then to nucleus; and subsequently, the activated bZIP17 regulates the downstream transcription of ABA-signaling negative regulators (Zhou et al., 2015a). ABA acts through the PYR/PYL/RCAR–PP2C–SnRKs signaling cascade (Cutler et al., 2010; Hubbard et al., 2010). The PP2C proteins, ABI1 and ABI2, bind to the ABA receptors to inhibit signaling. Their dominant-negative mutants *abi1-1* and *abi2-1* show reduced seed dormancy due to the failure of interaction between the mutated proteins and receptors (Ma, 2009; Park et al., 2009). Another PP2C protein, HONSU, also acts as a negative regulator of seed dormancy by concurrently inhibiting ABA signaling and activating GA signaling (Kim et al., 2013), suggesting that HONSU is a key factor in mediating the ABA and GA crosstalk concerning seed dormancy. Unexpectedly, a newly identified PP2C gene, *RDO5* (*Reduced Dormancy 5*), shows the strongly reduced seed dormancy phenotype, but its ABA sensitivity and content remain unchanged (Xiang et al., 2014). Further genetic and bioinformatics analysis demonstrated that RDO5 regulates seed dormancy through mediating the transcription of the PUF family of RNA binding genes, *APUM9* (*Arabidopsis PUMILIO 9*) and *APUM11* (Xiang et al., 2014). This evidence suggests that the RDO5-mediated regulation pathway is distinct from the ABA-signaling pathway, and further detailed investigation is needed.

As the major downstream component of ABA signaling, ABI3 is a main regulator of seed dormancy and germination (Bentsink and Koornneef, 2008). *ABI3* expression is regulated by *DEP* (*DESPIERTO*), which is involved in ABA sensitivity during seed development, and *dep* seeds show complete dormancy loss (Barrero et al., 2010). WRKY41 regulates *Arabidopsis* seed dormancy also through directly controlling *ABI3* transcription during seed maturation and germination (Ding et al., 2014). Another key component in the ABA-signaling pathway, ABI4, was also described as a positive regulator of primary seed dormancy (Shu et al., 2013). Subsequent studies demonstrated that MYB96, the ABA-responsive R2R3-type MYB transcription factor, positively regulates seed dormancy and negatively regulates germination through mediating expression of *ABI4* and ABA biogenesis genes, including *NCED2* and *NCED6* (Lee et al., 2015a, 2015b) (Table 1). Furthermore, a study showed that calcium also regulates seed germination by affecting *ABI4* transcription that controls ABA signaling (Kong et al., 2015). These studies demonstrated the key regulatory roles of positive regulators in ABA signaling during the transition from seed dormancy to germination.

Although ABI5 has no effect on seed dormancy and does not affect dormancy level (Finkelstein, 1994), this transcription factor negatively regulates seed germination (Piskurewicz et al., 2008; Kanai et al., 2010), suggesting the distinct signaling pathways for ABA-mediated seed dormancy and ABA-inhibited seed germination discussed above. A recent study showed that the MAP3K (mitogen-activated protein kinase kinase kinase) genes, *RAF10* and *RAF11*, regulate seed dormancy by affecting *ABI3* and *ABI5* transcription (Lee et al., 2015c). At post-transcription level, PKS5 (SOS2-like Protein Kinase 5, also known as CIPK11 or SnRK3.22) phosphorylates the special residue

Gene name	Dormancy level of mutant	General description of genes	References
ABI3	Decreased	Positively regulates ABA signaling and represses seed germination	Finkelstein, 1994
ABI5	Not changed	Positively regulates ABA signaling and represses seed germination	Brocard-Gifford et al., 2003; Finkelstein, 1994; Finkelstein et al., 2008
ABI4	Decreased	Positively regulates ABA signaling and represses seed germination	Shu et al., 2013; Kong et al., 2015
NCED5	Decreased	ABA biosynthesis gene, and the ABA content is decreased	Frey et al., 2011
CYP707A1/2	Enhanced	ABA-inactivated gene, and ABI4 negatively regulates its transcription	Millar et al., 2006; Matakiadis et al., 2009; Shu et al., 2013
GA1/2	Enhanced	GA biosynthesis genes, and the GA content is decreased in mutants	Lee et al., 2002
GA2oxs	Decreased	GA-inactivated genes, and the GA content is up-regulated in mutants	Yamauchi et al., 2007
RGL2/SPY	Enhanced	GA signaling is blocked in mutants	Jacobsen and Olszewski, 1993; Lee et al., 2002
MYB96	Decreased	Decreases *ABI4* and some ABA biogenesis gene transcription	Lee et al., 2015a, 2015b
DOG1	Enhanced	ABA sensitivity of *dog1* seeds is unchanged	Nakabayashi et al., 2012; Graeber et al., 2014
SUVH4/SUVH5	Enhanced	Repress *DOG1* and *ABI3* transcription	Zheng et al., 2012
LDL1/LDL2	Enhanced	Repress seed dormancy by negatively regulating *DOG1*	Zhao et al., 2015
WRKY41	Decreased	Directly promotes *ABI3* transcription	Ding et al., 2014
RAF10/RAF11	Decreased	Directly enhances *ABI3* transcription	Lee et al., 2015c
DEP	Decreased	Promotes *ABI3* transcription	Barrero et al., 2010
SPT	Decreased in *Ler*, while enhanced in *Col* background	Opposite roles in *Ler* and *Col* ecotypes	Belmonte et al., 2013; Vaistij et al., 2013
ARF10/ARF16	Decreased	Directly promote *ABI3* transcription	Liu et al., 2013b
BIN2	Not mentioned	Phosphorylates and stabilizes ABI5 to enhancing ABA signaling	Hu and Yu, 2014
PKS5	Not mentioned	Phosphorylates ABI5 (Ser42) and controls transcription of ABA-responsive genes	Zhou et al., 2015b
HONSU	Enhanced	As a PP2C protein, and impairs ABA signaling	Kim et al., 2013
RDO5	Enhanced	ABA sensitivity and content remain unchanged	Xiang et al., 2014
ABI1/2	Decreased	Dominant-negative mutants, and thus the mutated proteins cannot interact with ABA receptors	Ma, 2009; Park et al., 2009
CHO1	Decreased	Acts upstream on ABI4 genetically	Yamagishi et al., 2009; Yano et al., 2009
OsAP2-39	Decreased	Promotes *OsNCED1* and *OsEUI*, and thus enhances ABA biogenesis and impairs GA accumulation	Yaish et al., 2010
DDF1	Decreased	Directly promotes *GA2ox7* and thus decreases GA content	Magome et al., 2008

Table 1. Key Genes Involved in Seed Dormancy and Germination.

(Ser42) in ABI5 and controls transcription of ABA-responsive genes and, consequently, precisely regulates ABA signaling and the germination process (Zhou et al., 2015b) (Table 1). Altogether, the endogenous ABA level and ABA signaling positively regulate seed dormancy and therefore negatively regulate seed germination, and some key genes are involved in these physiological processes (Figure 2).

Another key phytohormone, GA, breaks dormancy and stimulates germination by antagonistically suppressing ABA-triggered seed dormancy (Gubler et al., 2005; Graeber et al., 2012). High GA levels or GA signaling promote seed germination, possibly from the secretion of hydrolytic enzymes to weaken seed testa structure (Holdsworth et al., 2008), but the detailed mechanisms, especially in *Arabidopsis*, are largely

unknown. GA-deficient mutants, such as *ga1* and *ga2*, show strong seed dormancy and fail to germinate without exogenous GA treatment (Lee et al., 2002; Shu et al., 2013). In contrast, mutants defective in GA2-oxidases (GA2ox), which deactivate bioactive GA, show decreased seed dormancy (Yamauchi et al., 2007). Similarly, mutations in DELLA genes including *RGL2* (*RGA-LIKE2*) and *SPY* (*SPINDLY*), negative regulators of the GA-signaling pathway, can rescue the non-germination phenotype of *ga1* (Jacobsen and Olszewski, 1993; Lee et al., 2002). Furthermore, DELLAs also maintain the seed embryo in a quiescent state by restricting cell-cycle progression through repression of the activities of TCP14 (Teosinte branched1/Cycloidea/Proliferating cell factor) and TCP15 (Resentini et al., 2015), further supporting the ''quiescent state'' hypothesis of the embryo described above.

KEY ROLES OF AP2 DOMAIN-CONTAINING TRANSCRIPTION FACTOR IN SEED DORMANCY REGULATION

The ABA/GA balance determines the fate of a seed: high endogenous ABA and low GA levels result in deep seed dormancy and low emergence, while low ABA and high GA levels induce preharvest sprouting. Therefore, the ABA/GA balance must be strictly regulated. There are two major aspects of the ABA/GA balance: the balance of hormone levels and the balance of the signaling cascades. The question arises: in the ABA–GA interaction, which is cause and which is effect? It has been reported that ABA is involved in the suppression of GA biogenesis (Seo et al., 2006), and GA also negatively regulates ABA biogenesis during seed germination (Shu et al., 2013; Oh et al., 2007). Therefore, ABA and GA may interact as both cause and effect during this process. However, the molecular mechanisms involved in precisely controlling the ABA/GA balance were largely unknown up to now, with AP2 domain-containing transcription factors found to possess the pivotal roles.

Numerous previous studies demonstrated that ABI4 is a versatile factor that regulates many signaling pathways, including the responses to ABA, glucose, sucrose, ethylene (ET), and salt stress (Wind et al., 2013). Interestingly, ABI4 also positively regulates ABA catabolism genes, but negatively affects GA biogenesis genes; thus, the loss of *ABI4* function increases the expression of GA biosynthesis genes but decreases the expression of GA-inactivation genes, together leading to decreased primary seed dormancy in *abi4* (Shu et al., 2013). As an AP2 domain-containing transcription factor, ABI4 directly binds to the promoters of *CYP707A1* and *CYP707A2*, which function in ABA catabolism, subsequently promoting ABA accumulation. However, no direct targeting of GA metabolism genes by ABI4 has been detected so far, suggesting that ABI4 may not bind directly to the promoters of GA biogenesis genes but may recruit or activate an additional seed-specific transcription factor to repress the transcription of GA biogenesis genes. Nevertheless, in sorghum, SbABI4 and SbABI5 can directly bind to the promoter of *SbGA2ox3*, likely activating its expression and affecting seed dormancy (Cantoro et al., 2013). Further investigations of ABI4-repressed GA signaling may identify the missing link in the ABI4–GA signaling crosstalk.

It is noted that *ABI4* transcription is regulated by the transcription factor SPT (SPATULA), which is also a key factor in seed dormancy regulation pathways; its role depends on the ecotype background (Vaistij et al., 2013); furthermore, the expression of *SPT* is increased during seed maturation (Belmonte et al., 2013), suggesting that the *SPT–ABI4* module takes the key role during dormancy establishment and maintenance (Figure 2). Similarly, another AP2 domain-containing transcription factor, CHO1 (CHOTTO1), positively regulates seed dormancy and, more interestingly, acts upstream on ABI4 genetically (Yamagishi et al., 2009; Yano et al., 2009). In rice, a model monocot, the AP2 domain-containing transcription factor OsAP2-39, directly promotes transcription of the ABA biosynthesis gene *OsNCED1* and expression of the GA-inactivating gene *OsEUI* (*ELONGATED UPPERMOST INTERNODE*), thus enhancing ABA biogenesis and impairing GA accumulation (Yaish et al., 2010). Consequently, the transgenic overexpression of *OsAP2-39* leads to increased seed dormancy (Yaish et al., 2010). These phenotypes have been documented in GA-deficient mutants (Richter et al., 2013), indicating that OsAP2-39 plays a pivotal role in regulating the ABA/GA biogenesis balance. DDF1 (DELAYED FLOWERING 1), a further player and another AP2-class transcription factor, directly promotes transcription of the GA-inactivation gene *GA2ox7*, and thus remarkably decreases endogenous GA content (Magome et al., 2008). The next player, EBE (ERF BUD ENHANCER), also an AP2 domain-containing transcription factor, has been shown to positively regulate seed dormancy (Mehrnia et al., 2013). Altogether, these AP2-containing transcription factors negatively regulate GA biogenesis while positively regulating ABA biogenesis.

Consequently, we propose that the AP2 domain plays a critical but cryptic role in the dual regulation of ABA and GA biogenesis in fine-tuning seed dormancy and germination (Figure 2). It is speculated that a DNA motif may be among these regulators and may possess undiscovered functions regarding seed dormancy regulation, especially for the ABA/GA balance concerning biogenesis and/or signaling levels. Furthermore, because these genes positively regulate endogenous ABA and decrease GA levels, transgenic overexpression lines show deep dormancy levels and other undesirable agronomic traits, given that the optimal endogenous hormone levels are essential to normal plant development. Consequently, the negative regulation of these transcription factors (ABI4, DDF1, OsAP2-39, and CHO1) is important for normal seed dormancy, and these factors must be strictly regulated at the mRNA and protein levels by these unknown negative regulators. Finally, further screening for suppressors of these mutants (e.g. *abi4*, *ddf1*, *osap2-39*, and *cho1*) might provide important information about the genetic network of the AP2 family in seed dormancy and germination.

AUXIN: A NEW MASTER PLAYER IN SEED DORMANCY

The phytohormone auxin is involved in almost all aspects of plant development and in response to a multitude of environmental cues (Zhao, 2010). Previously, auxin alone was not considered a key regulator of seed germination, although it participates in crosstalk with ABA (Wang et al., 2011a). Exogenous auxin

application suppresses seed germination under high salinity (Park et al., 2011), indicating that this hormone plays an important role in seed dormancy and germination in response to environmental stimuli. Earlier studies revealed that IAA (indole-3-acetic acid) can delay seed germination and inhibit pre-harvest sprouting in wheat (Ramaih et al., 2003); ABA represses embryonic axis elongation during seed germination also by potentiating auxin signaling (Belin et al., 2009); and a next study suggested that after-ripening treatment-mediated dormancy release is associated with decreased seed sensitivity to auxin (Liu et al., 2013a). All these observations imply that auxin may play a role in regulating seed dormancy and germination.

Emerging genetic data show that auxin protects and strictly regulates seed dormancy alongside ABA (Liu et al., 2013b). Evidence for this conclusion is provided by the dormancy variation among seeds with altered auxin synthesis genes. Auxin-overproducing transgenic *iaaM-OX* seeds show higher IAA levels compared with wild-type seeds, while *yuc1/yuc6* seeds show lower IAA content. Consistently, *iaaM-OX* seeds exhibit strong seed dormancy, while *yuc1/yuc6* seeds show the opposite phenotype. Phenotypic analysis demonstrated that nearly all auxin-signaling mutants, including *tir1/afb3* and *tir1/afb2*, show a decreased seed dormancy level. These observations reveal a close positive correlation between auxin content/signaling and seed dormancy, as also found for ABA.

What is the mechanism by which auxin controls seed dormancy? Detailed genetic and biochemical evidence shows that ABI3 is required for auxin-mediated seed dormancy and germination. When auxin levels are low, the auxin-responsive transcription factors *ARF10* and *ARF16* are repressed by AXR2/3. Thus, the expression of *ABI3* cannot be activated by ARF10/ARF16, and seed dormancy cannot be maintained. In contrast, when auxin levels are high, ARF10 and ARF16 are released to activate *ABI3* transcription, and seed dormancy is maintained. Since ARF10 and ARF16 likely do not directly bind to the *ABI3* promoter (Liu et al., 2013b), they may recruit or activate an additional seed-specific transcription factor(s) to stimulate *ABI3* expression; thus, further screening of dormancy mutants is needed to identify the missing link in the *ARF10/16–ABI3* signaling cascade. In summary, auxin affects ABA signaling to achieve its physiological effect (Liu et al., 2013b). Whether auxin also affects *ABI4* and *ABI5* is a worthwhile project in future studies.

In addition, many endogenous and environmental signals can also affect auxin content and distribution, thus shaping plant development (Vanneste and Friml, 2009). This poses an interesting question: do the same signals affect auxin biogenesis or signaling to regulate seed dormancy during seed development?

DIVERSE REGULATORS: OTHER PHYTOHORMONES INVOLVED IN SEED DORMANCY AND GERMINATION

In addition to ABA, GA, and auxin, nearly all other phytohormones are also likely involved in modulation of seed dormancy and germination, including ethylene (ET), brassinosteroids (BRs), jasmonic acid (JA), salicylic acid (SA), cytokinins (CTKs), and strigolactones (SLs).

ET breaks seed dormancy and promotes seed germination by counteracting the effect of ABA (Arc et al., 2013b; Corbineau et al., 2014). Mutations in positive regulators of the ET signaling pathway result in deep dormancy, while the negative regulator *ctr1* (*Constitutive Triple Response 1*) seeds germinate more rapidly (Subbiah and Reddy, 2010). Several studies have demonstrated that ET negatively affects ABA biogenesis and signaling (Cheng et al., 2009; Linkies et al., 2009). Previous studies showed that ET may affect seed germination through an ABA/GA-independent pathway (Linkies and Leubner-Metzger, 2012), and ABA and ET regulate seed dormancy by the antagonistic effect, which is mediated by key factors, such as *SNL1* (*SIN3-LIKE1*) and *SNL2* (Wang et al., 2013), suggesting a diversification of seed dormancy regulation mechanisms during evolutionary history. Interestingly, a recent study showed that the ET receptors ETR1 (Ethylene Response 1) and ETR2 possess contrasting roles for ABA biosynthesis during seed germination under salt-stress conditions, which may be independent of ET signaling (Wilson et al., 2014). However, whether and how ET affects GA biogenesis and signaling regarding seed dormancy and germination is largely unknown so far.

During seed germination, BR-deficient or BR-signaling mutants show stronger responses to ABA compared with wild-type seeds, indicating that BR overcomes the inhibitory effect of ABA on germination (Steber and McCourt, 2001). BR was found to promote seed germination in opposition to ABA partly through an MFT (MOTHER OF FT AND TFL1)-mediated pathway, which forms a negative feedback loop to modulate ABA signaling (Xi and Yu, 2010; Xi et al., 2010). A further elegant study demonstrated that BIN2 (Brassinosteroid Insensitive 2), a key repressor of the BR-signaling pathway, phosphorylates and stabilizes ABI5 protein to mediate ABA signaling during seed germination, whereby BR treatment represses the BIN2–ABI5 interaction, thus antagonizing ABA-mediated inhibition (Hu and Yu, 2014). However, the detailed mechanisms underlying the BR–GA crosstalk are elusive. For example, does BR induce GA biogenesis or enhance GA signaling during germination? More research on the effect of BR on seed dormancy is clearly needed.

SA is a plant hormone mainly associated with various defense pathways. Circumstantial evidence suggests that SA also regulates seed germination as a bifunctional modulator. SA inhibits germination by inhibiting the expression of GA-induced a-amylase genes under normal growth conditions (Xie et al., 2007). However, it promotes germination under high salinity via another pathway that reduces oxidative damage (Lee et al., 2010b). CTKs promote seed germination by antagonizing ABA, specifically by down-regulating *ABI5* transcription (Wang et al., 2011b). Further study demonstrated that CTKs antagonize ABA signaling by inducing ABI5 protein degradation (Guan et al., 2014). These observations highlight the importance of *ABI5* at both mRNA and protein levels, and ABI5 is the pivot involved in CTK–ABA crosstalk. It is noteworthy that although CTKs have positive effects on germination, CTK-receptor mutants exhibit lower dormancy levels compared with wild-type seeds (Riefler et al., 2006). These inverse effects suggest that there are distinct pathways in the CTK-mediated seed germination

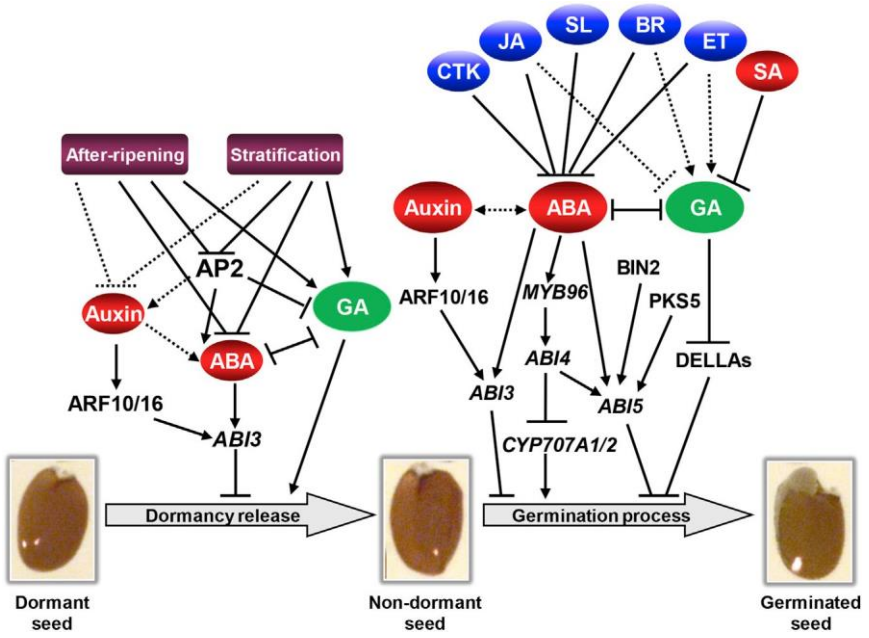

Figure 3. Preliminary Network of Phytohormone Functions in Seed Dormancy and Germination.

Dormancy release and germination of the seed are two separate but continuous phases. Freshly matured seeds are dormant and contain high levels of ABA and probably auxin, and low GA contents, resulting from changes in hormone biogenesis during seed development, as described in Figure 2. As the first phase in seed germination, after-ripening or stratification treatments break seed dormancy (dormancy release) by regulating ABA, GA, and auxin biogenesis and/or signals. These three hormones may interact to precisely control seed dormancy. In particular, ABA and auxin positively regulate seed dormancy in an interdependent manner, with auxin promoting *ABI3* transcription. Furthermore, AP2 domain-containing transcription factors, including ABI4, DDF1, OsAP2-39, and CHO1, positively regulate seed dormancy by promoting ABA biogenesis and repressing GA biogenesis/accumulation. The remaining question is whether AP2 domain-containing transcription factors also regulate auxin biogenesis and/or signaling. After seed dormancy is broken, non-dormant seeds initiate germination in the second phase. Different hormones affect this process by regulating the ABA/GA balance at either the biogenesis or signaling levels. The transcription factors ARFs, MYB96, ABI3, ABI4, and ABI5, the downstream target genes including *CYP707A1* and *CYP707A2*, and the GA-signaling negative regulators DELLAs play key roles in this process. Being a key factor, ABI5 was regulated precisely at transcription and post-transcription levels (ABI4 enhancing its expression while BIN2 and PKS5 phosphorylate ABI5). As the final step of seed germination, GA induced, but ABA inhibited, the rupture of the seed coat and enabled the radicle to penetrate the coat and complete emergence. The ABA/GA balance is the core determinant node in both steps. Arrows indicate positive regulation and bars indicate negative regulation.

regulation pathway. In future investigations, whether CTK affects GA biogenesis and/or signaling during the transition from dormancy to germination will be a pertinent topic.

Exogenous JA application delays seed germination (Nambara et al., 2010), indicating that JA has an inhibitory effect on the germination process. Interestingly, however, JA separately represses the transcription of ABA biosynthesis genes and promotes ABA-inactivating genes (Jacobsen et al., 2013), suggesting an antagonistic effect between JA and ABA. Consistent with this hypothesis, *coi1-16* and *jar1*, two JA-signaling mutants, show an ABA hypersensitive phenotype during germination (Fernandez-Arbaizar et al., 2012). However, it remains unclear why the role of JA in seed germination is sometimes contradictory. SLs are a small class of carotenoid-derived compounds that regulate many aspects of plant development, through the signaling pathway with D53 (*DWARF 53*) as a repressor (Umehara et al., 2008; Jiang et al., 2013; Zhou et al., 2013). SLs are host-derived germination stimulants for the seeds of parasitic weeds (Cook et al., 1966). They also trigger seed germination in other species, evidently by reducing the ABA/GA ratio (Toh et al., 2012). Furthermore, some key components in the SL signaling pathway affect seed germination, including *SMAX1* (*Suppressor of More Axillary Growth2 1*) in *Arabidopsis* (Stanga et al., 2013) and *OsD53* (Jiang et al., 2013; Zhou et al., 2013), which is the homolog of *SMAX1* in rice. However, the precise regulatory mechanisms underlying SLs need further investigation.

In summary, these plant hormones, including ET, BRs, JA, SA, CTKs, and SLs, regulate seed dormancy and germination, most likely by mediating the ABA/GA balance, although the interactions among these hormones and GA need further investigation, and some known detailed mechanisms are only the tip of the iceberg. In addition to these phytohormones, other small molecular compounds, including ROS (reactive oxygen species) and NO (nitric oxide), are involved in regulating seed dormancy and germination. ROS and NO synergistically break seed dormancy and probably act upstream of ABA (Bykova et al., 2011; Arc et al., 2013a). Thus, hormones and signaling compounds precisely regulate seed dormancy and germination through an integrated network of interactions with the ABA/GA balance as the central node (Figure 3).

In addition to phytohormones, various environmental cues determine the appropriate timing for seed germination, also by mediating the ABA/GA balance. Light is a major environmental factor during seed germination, increasing the expression of GA anabolic genes, *GA3ox1* and *GA3ox2*, and repressing the expression of *GA2ox2*, a GA catabolism gene (Cho et al., 2012). Previous studies demonstrated that blue light represses seed germination through enhancing the transcription of ABA biosynthetic genes and impairing the expression of ABA catabolic genes (Gubler et al., 2008; Barrero et al., 2014). After-ripening can also break seed dormancy, which negatively regulates ABA biogenesis (Figure 2). The transcription level of the ABA catabolism gene *CYP707A2* increases following after-ripening (Millar et al., 2006). Temperature is another environmental factor that influences seed dormancy both during seed maturation and in the soil by regulating the ABA/GA biogenesis balance (Footitt et al., 2011; Kendall et al., 2011). Temperature variation during seed maturation affects

primary seed dormancy by regulating coat permeability, which is a regulatory mechanism distinct from ABA/GA pathways (MacGregor et al., 2015). In addition, although the detailed mechanisms underlying the pre-harvest sprouting phenotype of *TaMFT*-RNAi plants are elusive, the homolog of *TaMFT* in *Arabidopsis*, *MFT*, is a pivotal factor that fine-tunes the ABA/GA-signaling balance (Xi et al., 2010; Nakamura et al., 2011). Consequently, seed dormancy is the integrated result of endogenous and environmental factors that regulate the ABA/GA balance, in either hormone accumulations or hormone signaling cascades.

CONCLUDING REMARKS AND PERSPECTIVES

Owing to forward- and reverse-genetic approaches in the model plant *Arabidopsis* and recent advances primarily in rice, rapid progress has been achieved in the field of seed dormancy and germination. Although certain key factors that regulate this important transition have been identified, and we know that plant hormones regulate seed dormancy and germination through a complex network (Figure 3), several open questions remain to be addressed.

First, the germination process includes two sequential steps: rupture of the seed coat and emergence of the radicle. Previous studies demonstrated that in cereal grains the starch granule deposition, hydrolase activity, and protein catabolism are important to seed germination, and thus the transcription level of genes encoding a-amylases are key determinants (Hong et al., 2012; Shaik et al., 2014). However, in *Arabidopsis* the precise molecular mechanisms underlying the rupture of the seed coat and endosperm processes remain largely unknown. Consequently, further detailed screening of the key genes involved in both of these stages is worthwhile.

Second, ABA is the key inducer of seed dormancy, and ABA represses GA biogenesis. Nevertheless, we still know little about changes in GA biogenesis during seed maturation (Figure 2). With the development of hormone detection assays, we can precisely investigate the amount of phytohormones in different tissues, even in single cells (Chen et al., 2011), which allows investigation of the kinetics of GA biogenesis over a time course during seed dormancy establishment.

Third, as the central node of seed dormancy and germination, where within the seed are the molecular activities of ABA and GA localized? Are ABA and GA synthesized de novo at these two sites? Pioneering studies developed a ''seed coat bedding'' assay, which was employed to demonstrate that ABA is synthesized de novo in the seed coat in an RGL2-dependent manner and thus represses germination of the embryo (Lee et al., 2010a; Lee and Lopez-Molina, 2013). Subsequently, where in the seed is GA biogenesis located?

Fourth, the possible increase in auxin levels during seed maturation raises an important question: what are the molecular mechanisms monitoring the auxin pathway during seed development? The key auxin biosynthesis genes *YUC1*, *YUC2*, and *YUC6* reach peak levels during the later stages of seed development (Liu et al., 2013b), suggesting that auxin biosynthesis may be enhanced during seed maturation. It will be interesting to investigate how the *YUC* genes are activated to fine-tune auxin biosynthesis during seed maturation.

Fifth, because ABA and auxin act synergistically to positively regulate seed dormancy, GA and auxin therefore may antagonistically function in seed dormancy. However, the detailed mechanisms underlying these synergistic and antagonistic effects are also largely elusive at the molecular level, including the precise interactions among ABIs, DOG1, DEP1, SPT (Figures 2 and 3), and downstream targets of these transcription factors, which directly function in seed germination.

Finally, given that environmental cues, such as light and temperature (Lim et al., 2014), regulate seed dormancy and germination through the ABA/GA biogenesis and signaling pathways, it is quite possible that environmental factors also affect auxin and other hormone pathways during seed germination. In this field, epigenetic effects are of particular interest because both hormonal and environmental cues are involved in epigenetic modifications. Breakthroughs concerning these regulatory mechanisms will provide a more complete network of hormone-mediated seed dormancy and germination, in addition to new solutions for controlling pre-harvest sprouting in crops.

FUNDING

This work was supported by the Natural Science Foundation of China (Grants 90817102, 91317308 and 91117018) and China Postdoctoral Science Foundation funded project (2014M552377).

ACKNOWLEDGMENTS

We thank Dr. Chengcai Chu and Dr. Pengfei Wang of the Institute of Genetics and Developmental Biology, Chinese Academy of Sciences, for providing the pictures of pre-harvest sprouting rice and *Arabidopsis* siliques. We apologize to colleagues whose work could not be discussed and cited because of space limitations. No conflict of interest declared.

Received: June 18, 2015
Revised: August 10, 2015
Accepted: August 13, 2015
Published: September 4, 2015

REFERENCES

Arc, E., Galland, M., Godin, B., Cueff, G., and Rajjou, L. (2013a). Nitric oxide implication in the control of seed dormancy and germination. Front Plant Sci. 4:346.

Arc, E., Sechet, J., Corbineau, F., Rajjou, L., and Marion-Poll, A. (2013b). ABA crosstalk with ethylene and nitric oxide in seed dormancy and germination. Front Plant Sci. 4:63.

Barrero, J.M., Millar, A.A., Griffiths, J., Czechowski, T., Scheible, W.R., Udvardi, M., Reid, J.B., Ross, J.J., Jacobsen, J.V., and Gubler, F. (2010). Gene expression profiling identifies two regulatory genes controlling dormancy and ABA sensitivity in *Arabidopsis* seeds. Plant J. 61:611–622.

Barrero, J.M., Downie, A.B., Xu, Q., and Gubler, F. (2014). A role for barley CRYPTOCHROME1 in light regulation of grain dormancy and germination. Plant Cell 26:1094–1104.

Belin, C., Megies, C., Hauserova, E., and Lopez-Molina, L. (2009). Abscisic acid represses growth of the *Arabidopsis* embryonic axis after germination by enhancing auxin signaling. Plant Cell 21:2253–2268.

Belmonte, M.F., Kirkbride, R.C., Stone, S.L., Pelletier, J.M., Bui, A.Q., Yeung, E.C., Hashimoto, M., Fei, J., Harada, C.M., Munoz, M.D., et al. (2013). Comprehensive developmental profiles of gene activity in regions and subregions of the *Arabidopsis* seed. Proc. Natl. Acad. Sci. USA 110:E435–E444.

Bentsink, L., and Koornneef, M. (2008). Seed dormancy and germination. Arabidopsis Book 6:e0119.

Brocard-Gifford, I.M., Lynch, T.J., and Finkelstein, R.R. (2003). Regulatory networks in seeds integrating developmental, abscisic acid, sugar, and light signaling. Plant Physiol. 131:78–92.

Bykova, N.V., Hoehn, B., Rampitsch, C., Hu, J., Stebbing, J.A., and Knox, R. (2011). Thiol redox-sensitive seed proteome in dormant and non-dormant hybrid genotypes of wheat. Phytochemistry 72:1162–1172.

Cantoro, R., Crocco, C.D., Benech-Arnold, R.L., and Rodriguez, M.V. (2013). In vitro binding of *Sorghum bicolor* transcription factors ABI4 and ABI5 to a conserved region of a GA 2-OXIDASE promoter: possible role of this interaction in the expression of seed dormancy. J. Exp. Bot. 64:5721–5735.

Chen, M.L., Huang, Y.Q., Liu, J.Q., Yuan, B.F., and Feng, Y.Q. (2011). Highly sensitive profiling assay of acidic plant hormones using a novel mass probe by capillary electrophoresis-time of flight-mass spectrometry. J. Chromatogr. B Analyt. Technol. Biomed. Life Sci. 879:938–944.

Cheng, W.H., Chiang, M.H., Hwang, S.G., and Lin, P.C. (2009). Antagonism between abscisic acid and ethylene in *Arabidopsis* acts in parallel with the reciprocal regulation of their metabolism and signaling pathways. Plant Mol. Biol. 71:61–80.

Chinnusamy, V., Gong, Z., and Zhu, J.K. (2008). Abscisic acid-mediated epigenetic processes in plant development and stress responses. J. Integr. Plant Biol. 50:1187–1195.

Cho, J.N., Ryu, J.Y., Jeong, Y.M., Park, J., Song, J.J., Amasino, R.M., Noh, B., and Noh, Y.S. (2012). Control of seed germination by light-induced histone arginine demethylation activity. Dev. Cell 22:736–748.

Cook, C.E., Whichard, L.P., Turner, B., Wall, M.E., and Egley, G.H. (1966). Germination of witchweed (*Striga lutea* Lour.): isolation and properties of a potent stimulant. Science 154:1189–1190.

Corbineau, F., Xia, Q., Bailly, C., and El-Maarouf-Bouteau, H. (2014). Ethylene, a key factor in the regulation of seed dormancy. Front Plant Sci. 5:539.

Cutler, S.R., Rodriguez, P.L., Finkelstein, R.R., and Abrams, S.R. (2010). Abscisic acid: emergence of a core signaling network. Annu. Rev. Plant Biol. 61:651–679.

Ding, Z.J., Yan, J.Y., Li, G.X., Wu, Z.C., Zhang, S.Q., and Zheng, S.J. (2014). WRKY41 controls *Arabidopsis* seed dormancy via direct regulation of ABI3 transcript levels not downstream of ABA. Plant J. 79:810–823.

Fernandez-Arbaizar, A., Regalado, J.J., and Lorenzo, O. (2012). Isolation and characterization of novel mutant loci suppressing the ABA hypersensitivity of the *Arabidopsis* coronatine insensitive 1-16 (coi1-16) mutant during germination and seedling growth. Plant Cell Physiol. 53:53–63.

Finkelstein, R.R. (1994). Mutations at 2 new *Arabidopsis* Aba response loci are similar to the Abi3 mutations. Plant J. 5:765–771.

Finkelstein, R., Reeves, W., Ariizumi, T., and Steber, C. (2008). Molecular aspects of seed dormancy. Annu. Rev. Plant Biol. 59:387–415.

Footitt, S., Douterelo-Soler, I., Clay, H., and Finch-Savage, W.E. (2011). Dormancy cycling in *Arabidopsis* seeds is controlled by seasonally distinct hormone-signaling pathways. Proc. Natl. Acad. Sci. USA 108:20236–20241.

Footitt, S., Muller, K., Kermode, A.R., and Finch-Savage, W.E. (2015). Seed dormancy cycling in *Arabidopsis*: chromatin remodelling and regulation of DOG1 in response to seasonal environmental signals. Plant J. 81:413–425.

Frey, A., Effroy, D., Lefebvre, V., Seo, M., Perreau, F., Berger, A., Sechet, J., To, A., North, H.M., and Marion-Poll, A. (2011). Epoxycarotenoid cleavage by NCED5 fine-tunes ABA accumulation and affects seed dormancy and drought tolerance with other NCED family members. Plant J. 70:501–512.

Graeber, K., Nakabayashi, K., Miatton, E., Leubner-Metzger, G., and Soppe, W.J. (2012). Molecular mechanisms of seed dormancy. Plant Cell Environ. 35:1769–1786.

Graeber, K., Linkies, A., Steinbrecher, T., Mummenhoff, K., Tarkowska, D., Tureckova, V., Ignatz, M., Sperber, K., Voegele, A., de Jong, H., et al. (2014). DELAY OF GERMINATION 1 mediates a conserved coat-dormancy mechanism for the temperature- and gibberellin-dependent control of seed germination. Proc. Natl. Acad. Sci. USA 111:E3571–E3580.

Guan, C., Wang, X., Feng, J., Hong, S., Liang, Y., Ren, B., and Zuo, J. (2014). Cytokinin antagonizes abscisic acid-mediated inhibition of cotyledon greening by promoting the degradation of abscisic acid insensitive5 protein in *Arabidopsis*. Plant Physiol. 164:1515–1526.

Gubler, F., Millar, A.A., and Jacobsen, J.V. (2005). Dormancy release, ABA and pre-harvest sprouting. Curr. Opin. Plant Biol. 8:183–187.

Gubler, F., Hughes, T., Waterhouse, P., and Jacobsen, J. (2008). Regulation of dormancy in barley by blue light and after-ripening: effects on abscisic acid and gibberellin metabolism. Plant Physiol. 147:886–896.

Hoang, H.H., Sechet, J., Bailly, C., Leymarie, J., and Corbineau, F. (2014). Inhibition of germination of dormant barley (*Hordeum vulgare* L.) grains by blue light as related to oxygen and hormonal regulation. Plant Cell Environ. 37:1393–1403.

Holdsworth, M.J., Bentsink, L., and Soppe, W.J. (2008). Molecular networks regulating *Arabidopsis* seed maturation, after-ripening, dormancy and germination. New Phytol. 179:33–54.

Hong, Y.F., Ho, T.H., Wu, C.F., Ho, S.L., Yeh, R.H., Lu, C.A., Chen, P.W., Yu, L.C., Chao, A., and Yu, S.M. (2012). Convergent starvation signals and hormone crosstalk in regulating nutrient mobilization upon germination in cereals. Plant Cell 24:2857–2873.

Hu, Y., and Yu, D. (2014). BRASSINOSTEROID INSENSITIVE2 interacts with ABSCISIC ACID INSENSITIVE5 to mediate the antagonism of brassinosteroids to abscisic acid during seed germination in *Arabidopsis*. Plant Cell 26:4394–4408.

Hubbard, K.E., Nishimura, N., Hitomi, K., Getzoff, E.D., and Schroeder, J.I. (2010). Early abscisic acid signal transduction mechanisms: newly discovered components and newly emerging questions. Genes Dev. 24:1695–1708.

Jacobsen, S.E., and Olszewski, N.E. (1993). Mutations at the spindly locus of *Arabidopsis* alter gibberellin signal-transduction. Plant Cell. 5:887–896.

Jacobsen, J.V., Barrero, J.M., Hughes, T., Julkowska, M., Taylor, J.M., Xu, Q., and Gubler, F. (2013). Roles for blue light, jasmonate and nitric oxide in the regulation of dormancy and germination in wheat grain (*Triticum aestivum* L.). Planta 238:121–138.

Jiang, L., Liu, X., Xiong, G., Liu, H., Chen, F., Wang, L., Meng, X., Liu, G., Yu, H., Yuan, Y., et al. (2013). DWARF 53 acts as a repressor of strigolactone signalling in rice. Nature 504:401–405.

Kanai, M., Nishimura, M., and Hayashi, M. (2010). A peroxisomal ABC transporter promotes seed germination by inducing pectin degradation under the control of ABI5. Plant J. 62:936–947.

Kendall, S.L., Hellwege, A., Marriot, P., Whalley, C., Graham, I.A., and Penfield, S. (2011). Induction of dormancy in *Arabidopsis* summer annuals requires parallel regulation of DOG1 and hormone metabolism by low temperature and CBF transcription factors. Plant Cell 23:2568–2580.

Kim, W., Lee, Y., Park, J., Lee, N., and Choi, G. (2013). HONSU, a protein phosphatase 2C, regulates seed dormancy by inhibiting ABA signaling in *Arabidopsis*. Plant Cell Physiol. 54:555–572.

Kong, D., Ju, C., Parihar, A., Kim, S., Cho, D., and Kwak, J.M. (2015). *Arabidopsis* glutamate receptor homolog3.5 modulates cytosolic Ca^{2+} level to counteract effect of abscisic acid in seed germination. Plant Physiol. 167:1630–1642.

Kucera, B., Cohn, M.A., and Leubner-Metzger, G. (2005). Plant hormone interactions during seed dormancy release and germination. Seed Sci. Res. 15:281–307.

Lee, K.P., and Lopez-Molina, L. (2013). A seed coat bedding assay to genetically explore in vitro how the endosperm controls seed germination in *Arabidopsis thaliana*. J. Vis. Exp. 81:e50732.

Lee, S., Cheng, H., King, K.E., Wang, W., He, Y., Hussain, A., Lo, J., Harberd, N.P., and Peng, J. (2002). Gibberellin regulates *Arabidopsis* seed germination via RGL2, a GAI/RGA-like gene whose expression is up-regulated following imbibition. Genes Dev. 16:646–658.

Lee, K.P., Piskurewicz, U., Tureckova, V., Strnad, M., and Lopez-Molina, L. (2010a). A seed coat bedding assay shows that RGL2-dependent release of abscisic acid by the endosperm controls embryo growth in *Arabidopsis* dormant seeds. Proc. Natl. Acad. Sci. USA 107:19108–19113.

Lee, S., Kim, S.G., and Park, C.M. (2010b). Salicylic acid promotes seed germination under high salinity by modulating antioxidant activity in *Arabidopsis*. New Phytol. 188:626–637.

Lee, H.G., Lee, K., and Seo, P.J. (2015a). The *Arabidopsis* MYB96 transcription factor plays a role in seed dormancy. Plant Mol. Biol. 87:371–381.

Lee, K., Lee, H.G., Yoon, S., Kim, H.U., and Seo, P.J. (2015b). The *Arabidopsis* MYB96 transcription factor is a positive regulator of ABI4 in the control of seed germination. Plant Physiol. 168:677–689.

Lee, S.J., Lee, M.H., Kim, J.I., and Kim, S.Y. (2015c). *Arabidopsis* putative MAP kinase kinase kinases Raf10 and Raf11 are positive regulators of seed dormancy and ABA response. Plant Cell Physiol. 56:84–97.

Lenser, T., and Theissen, G. (2013). Molecular mechanisms involved in convergent crop domestication. Trends Plant Sci. 18:704–714.

Lim, S., Park, J., Lee, N., Jeong, J., Toh, S., Watanabe, A., Kim, J., Kang, H., Kim, D.H., Kawakami, N., et al. (2014). ABA-INSENSITIVE3, ABA-INSENSITIVE5, and DELLAs interact to activate the expression of SOMNUS and other high-temperature-inducible genes in imbibed seeds in *Arabidopsis*. Plant Cell 25:4863–4878.

Linkies, A., and Leubner-Metzger, G. (2012). Beyond gibberellins and abscisic acid: how ethylene and jasmonates control seed germination. Plant Cell Rep. 31:253–270.

Linkies, A., Muller, K., Morris, K., Tureckova, V., Wenk, M., Cadman, C.S., Corbineau, F., Strnad, M., Lynn, J.R., Finch-Savage, W.E., et al. (2009). Ethylene interacts with abscisic acid to regulate endosperm rupture during germination: a comparative approach using *Lepidium sativum* and *Arabidopsis thaliana*. Plant Cell 21:3803–3822.

Liu, Y., Koornneef, M., and Soppe, W.J. (2007). The absence of histone H2B monoubiquitination in the *Arabidopsis* hub1 (rdo4) mutant reveals a role for chromatin remodeling in seed dormancy. Plant Cell 19:433–444.

Liu, Y., Geyer, R., van Zanten, M., Carles, A., Li, Y., Horold, A., van Nocker, S., and Soppe, W.J. (2011). Identification of the *Arabidopsis* REDUCED DORMANCY 2 gene uncovers a role for the polymerase associated factor 1 complex in seed dormancy. PLoS One 6:e22241.

Liu, A., Gao, F., Kanno, Y., Jordan, M.C., Kamiya, Y., Seo, M., and Ayele, B.T. (2013a). Regulation of wheat seed dormancy by after-ripening is mediated by specific transcriptional switches that induce changes in seed hormone metabolism and signaling. PLoS One 8:e56570.

Liu, X., Zhang, H., Zhao, Y., Feng, Z., Li, Q., Yang, H.Q., Luan, S., Li, J., and He, Z.H. (2013b). Auxin controls seed dormancy through stimulation of abscisic acid signaling by inducing ARF-mediated ABI3 activation in *Arabidopsis*. Proc. Natl. Acad. Sci. USA 110: 15485–15490.

Ma, Y. (2009). Regulators of PP2C phosphatase activity function as abscisic acid sensors. Science 324:1264–1268.

MacGregor, D.R., Kendall, S.L., Florance, H., Fedi, F., Moore, K., Paszkiewicz, K., Smirnoff, N., and Penfield, S. (2015). Seed production temperature regulation of primary dormancy occurs through control of seed coat phenylpropanoid metabolism. New Phytol. 205:642–652.

Magome, H., Yamaguchi, S., Hanada, A., Kamiya, Y., and Oda, K. (2008). The DDF1 transcriptional activator upregulates expression of a gibberellin-deactivating gene, GA2ox7, under high-salinity stress in *Arabidopsis*. Plant J. 56:613–626.

Martinez-Andujar, C., Ordiz, M.I., Huang, Z., Nonogaki, M., Beachy, R.N., and Nonogaki, H. (2011). Induction of 9-cis-epoxycarotenoid dioxygenase in *Arabidopsis thaliana* seeds enhances seed dormancy. Proc. Natl. Acad. Sci. USA 108:17225–17229.

Matakiadis, T., Alboresi, A., Jikumaru, Y., Tatematsu, K., Pichon, O., Renou, J.P., Kamiya, Y., Nambara, E., and Truong, H.N. (2009). The *Arabidopsis* abscisic acid catabolic gene CYP707A2 plays a key role in nitrate control of seed dormancy. Plant Physiol. 149:949–960.

Mehrnia, M., Balazadeh, S., Zanor, M.I., and Mueller-Roeber, B. (2013). EBE, an AP2/ERF transcription factor highly expressed in proliferating cells, affects shoot architecture in *Arabidopsis*. Plant Physiol. 162:842–857.

Meyer, R.S., and Purugganan, M.D. (2013). Evolution of crop species: genetics of domestication and diversification. Nat. Rev. Genet. 14:840–852.

Millar, A.A., Jacobsen, J.V., Ross, J.J., Helliwell, C.A., Poole, A.T., Scofield, G., Reid, J.B., and Gubler, F. (2006). Seed dormancy and ABA metabolism in *Arabidopsis* and barley: the role of ABA 8^0-hydroxylase. Plant J. 45:942–954.

Nakabayashi, K., Bartsch, M., Xiang, Y., Miatton, E., Pellengahr, S., Yano, R., Seo, M., and Soppe, W.J. (2012). The time required for dormancy release in *Arabidopsis* is determined by DELAY OF GERMINATION1 protein levels in freshly harvested seeds. Plant Cell 24:2826–2838.

Nakamura, S., Abe, F., Kawahigashi, H., Nakazono, K., Tagiri, A., Matsumoto, T., Utsugi, S., Ogawa, T., Handa, H., Ishida, H., et al. (2011). A wheat homolog of MOTHER OF FT AND TFL1 acts in the regulation of germination. Plant Cell 23:3215–3229.

Nambara, E., Okamoto, M., Tatematsu, K., Yano, R., Seo, M., and Kamiya, Y. (2010). Abscisic acid and the control of seed dormancy and germination. Seed Sci. Res. 20:55–67.

Nonogaki, M., Sall, K., Nambara, E., and Nonogaki, H. (2014). Amplification of ABA biosynthesis and signaling through a positive feedback mechanism in seeds. Plant J. 78:527–539.

Oh, E., Yamaguchi, S., Hu, J., Yusuke, J., Jung, B., Paik, I., Lee, H.S., Sun, T.P., Kamiya, Y., and Choi, G. (2007). PIL5, a phytochrome-interacting bHLH protein, regulates gibberellin responsiveness by

binding directly to the GAI and RGA promoters in Arabidopsis seeds. Plant cell 19:1192–1208.

Papaefthimiou, D., Likotrafiti, E., Kapazoglou, A., Bladenopoulos, K., and Tsaftaris, A. (2010). Epigenetic chromatin modifiers in barley: III. Isolation and characterization of the barley GNAT-MYST family of histone acetyltransferases and responses to exogenous ABA. Plant Physiol. Biochem. 48:98–107.

Park, S.Y., Fung, P., Nishimura, N., Jensen, D.R., Fujii, H., Zhao, Y., Lumba, S., Santiago, J., Rodrigues, A., Chow, T.F.F., et al. (2009). Abscisic acid inhibits type 2C protein phosphatases via the PYR/PYL family of START proteins. Science 324:1068–1071.

Park, J., Kim, Y.S., Kim, S.G., Jung, J.H., Woo, J.C., and Park, C.M. (2011). Integration of auxin and salt signals by the NAC transcription factor NTM2 during seed germination in *Arabidopsis*. Plant Physiol. 156:537–549.

Piskurewicz, U., Jikumaru, Y., Kinoshita, N., Nambara, E., Kamiya, Y., and Lopez-Molina, L. (2008). The gibberellic acid signaling repressor RGL2 inhibits *Arabidopsis* seed germination by stimulating abscisic acid synthesis and ABI5 activity. Plant Cell 20:2729–2745.

Rajjou, L., Duval, M., Gallardo, K., Catusse, J., Bally, J., Job, C., and Job, D. (2012). Seed germination and vigor. Annu. Rev. Plant Biol. 63:507–533.

Ramaih, S., Guedira, M., and Paulsen, G.M. (2003). Relationship of indoleacetic acid and tryptophan to dormancy and preharvest sprouting of wheat. Funct. Plant Biol. 30:939–945.

Resentini, F., Felipo-Benavent, A., Colombo, L., Blazquez, M.A., Alabadi, D., and Masiero, S. (2015). TCP14 and TCP15 mediate the promotion of seed germination by gibberellins in *Arabidopsis thaliana*. Mol. Plant 8:482–485.

Richter, R., Behringer, C., Zourelidou, M., and Schwechheimer, C. (2013). Convergence of auxin and gibberellin signaling on the regulation of the GATA transcription factors GNC and GNL in *Arabidopsis thaliana*. Proc. Natl. Acad. Sci. USA 110:13192–13197.

Riefler, M., Novak, O., Strnad, M., and Schmulling, T. (2006). *Arabidopsis* cytokinin receptor mutants reveal functions in shoot growth, leaf senescence, seed size, germination, root development, and cytokinin metabolism. Plant Cell 18:40–54.

Saez, A., Rodrigues, A., Santiago, J., Rubio, S., and Rodriguez, P.L. (2008). HAB1-SWI3B interaction reveals a link between abscisic acid signaling and putative SWI/SNF chromatin-remodeling complexes in *Arabidopsis*. Plant Cell 20:2972–2988.

Seo, M., Hanada, A., Kuwahara, A., Endo, A., Okamoto, M., Yamauchi, Y., North, H., Marion-Poll, A., Sun, T.P., Koshiba, T., et al. (2006). Regulation of hormone metabolism in *Arabidopsis* seeds: phytochrome regulation of abscisic acid metabolism and abscisic acid regulation of gibberellin metabolism. Plant J. 48:354–366.

Shaik, S.S., Carciofi, M., Martens, H.J., Hebelstrup, K.H., and Blennow, A. (2014). Starch bioengineering affects cereal grain germination and seedling establishment. J. Exp. Bot. 65:2257–2270.

Shu, K., Zhang, H., Wang, S., Chen, M., Wu, Y., Tang, S., Liu, C., Feng, Y., Cao, X., and Xie, Q. (2013). ABI4 regulates primary seed dormancy by regulating the biogenesis of abscisic acid and gibberellins in *Arabidopsis*. PLoS Genet. 9:e1003577.

Simsek, S., Ohm, J.B., Lu, H., Rugg, M., Berzonsky, W., Alamri, M.S., and Mergoum, M. (2014). Effect of pre-harvest sprouting on physicochemical changes of proteins in wheat. J. Sci. Food Agric. 94:205–212.

Stanga, J.P., Smith, S.M., Briggs, W.R., and Nelson, D.C. (2013). SUPPRESSOR OF MORE AXILLARY GROWTH2 1 controls seed germination and seedling development in *Arabidopsis*. Plant Physiol. 163:318–330.

Steber, C.M., and McCourt, P. (2001). A role for brassinosteroids in germination in *Arabidopsis*. Plant Physiol. 125:763–769.

Subbiah, V., and Reddy, K.J. (2010). Interactions between ethylene, abscisic acid and cytokinin during germination and seedling establishment in *Arabidopsis*. J. Biosci. 35:451–458.

Toh, S., Kamiya, Y., Kawakami, N., Nambara, E., McCourt, P., and Tsuchiya, Y. (2012). Thermoinhibition uncovers a role for strigolactones in *Arabidopsis* seed germination. Plant Cell Physiol. 53:107–117.

Umehara, M., Hanada, A., Yoshida, S., Akiyama, K., Arite, T., Takeda-Kamiya, N., Magome, H., Kamiya, Y., Shirasu, K., Yoneyama, K., et al. (2008). Inhibition of shoot branching by new terpenoid plant hormones. Nature 455:195–200.

Vaistij, F.E., Gan, Y., Penfield, S., Gilday, A.D., Dave, A., He, Z., Josse, E.M., Choi, G., Halliday, K.J., and Graham, I.A. (2013). Differential control of seed primary dormancy in *Arabidopsis* ecotypes by the transcription factor SPATULA. Proc. Natl. Acad. Sci. USA 110:10866–10871.

Vanneste, S., and Friml, J. (2009). Auxin: a trigger for change in plant development. Cell 136:1005–1016.

Wang, L., Hua, D., He, J., Duan, Y., Chen, Z., Hong, X., and Gong, Z. (2011a). Auxin Response Factor2 (ARF2) and its regulated homeodomain gene HB33 mediate abscisic acid response in *Arabidopsis*. PLoS Genet. 7:e1002172.

Wang, Y., Li, L., Ye, T., Zhao, S., Liu, Z., Feng, Y.Q., and Wu, Y. (2011b). Cytokinin antagonizes ABA suppression to seed germination of *Arabidopsis* by downregulating ABI5 expression. Plant J. 68:249–261.

Wang, Z., Cao, H., Sun, Y., Li, X., Chen, F., Carles, A., Li, Y., Ding, M., Zhang, C., Deng, X., et al. (2013). *Arabidopsis* paired amphipathic helix proteins SNL1 and SNL2 redundantly regulate primary seed dormancy via abscisic acid-ethylene antagonism mediated by histone deacetylation. Plant Cell 25:149–166.

Wilson, R.L., Kim, H., Bakshi, A., and Binder, B.M. (2014). The ethylene receptors ETHYLENE RESPONSE1 and ETHYLENE RESPONSE2 have contrasting roles in seed germination of *Arabidopsis* during salt stress. Plant Physiol. 165:1353–1366.

Wind, J.J., Peviani, A., Snel, B., Hanson, J., and Smeekens, S.C. (2013). ABI4: versatile activator and repressor. Trends Plant Sci. 18:125–132.

Xi, W., and Yu, H. (2010). Mother of FT and TFL1 regulates seed germination and fertility relevant to the brassinosteroid signaling pathway. Plant Signal Behav. 5:1315–1317.

Xi, W., Liu, C., Hou, X., and Yu, H. (2010). MOTHER OF FT AND TFL1 regulates seed germination through a negative feedback loop modulating ABA signaling in *Arabidopsis*. Plant Cell 22:1733–1748.

Xiang, Y., Nakabayashi, K., Ding, J., He, F., Bentsink, L., and Soppe, W.J. (2014). Reduced Dormancy5 encodes a protein phosphatase 2C that is required for seed dormancy in *Arabidopsis*. Plant Cell 26:4362–4375.

Xie, Z., Zhang, Z.L., Hanzlik, S., Cook, E., and Shen, Q.J. (2007). Salicylic acid inhibits gibberellin-induced alpha-amylase expression and seed germination via a pathway involving an abscisic-acid-inducible WRKY gene. Plant Mol. Biol. 64:293–303.

Yaish, M.W., El-Kereamy, A., Zhu, T., Beatty, P.H., Good, A.G., Bi, Y.M., and Rothstein, S.J. (2010). The APETALA-2-like transcription factor OsAP2-39 controls key interactions between abscisic acid and gibberellin in rice. PLoS Genet. 6:e1001098.

Yamagishi, K., Tatematsu, K., Yano, R., Preston, J., Kitamura, S., Takahashi, H., McCourt, P., Kamiya, Y., and Nambara, E. (2009). CHOTTO1, a double AP2 domain protein of *Arabidopsis thaliana*, regulates germination and seedling growth under excess supply of glucose and nitrate. Plant Cell Physiol. 50:330–340.

Yamauchi, Y., Takeda-Kamiya, N., Hanada, A., Ogawa, M., Kuwahara, A., Seo, M., Kamiya, Y., and Yamaguchi, S. (2007). Contribution of gibberellin deactivation by AtGA2ox2 to the suppression of germination of dark-imbibed *Arabidopsis thaliana* seeds. Plant Cell Physiol. 48:555–561.

Yano, R., Kanno, Y., Jikumaru, Y., Nakabayashi, K., Kamiya, Y., and Nambara, E. (2009). CHOTTO1, a putative double APETALA2 repeat transcription factor, is involved in abscisic acid-mediated repression of gibberellin biosynthesis during seed germination in *Arabidopsis*. Plant Physiol. 151:641–654.

Zhao, Y. (2010). Auxin biosynthesis and its role in plant development. Annu. Rev. Plant Biol. 61:49–64.

Zhao, M., Yang, S., Liu, X., and Wu, K. (2015). *Arabidopsis* histone demethylases LDL1 and LDL2 control primary seed dormancy by regulating delay of germination 1 and ABA signaling-related genes. Front Plant Sci. 6:159.

Zheng, J., Chen, F., Wang, Z., Cao, H., Li, X., Deng, X., Soppe, W.J., Li, Y., and Liu, Y. (2012). A novel role for histone methyltransferase KYP/SUVH4 in the control of *Arabidopsis* primary seed dormancy. New Phytol. 193:605–616.

Zhou, F., Lin, Q., Zhu, L., Ren, Y., Zhou, K., Shabek, N., Wu, F., Mao, H., Dong, W., Gan, L., et al. (2013). D14-SCFD3-dependent degradation of D53 regulates strigolactone signalling. Nature 504:406–410.

Zhou, S.F., Sun, L., Valdes, A.E., Engstrom, P., Song, Z.T., Lu, S.J., and Liu, J.X. (2015a). Membrane-associated transcription factor peptidase, site-2 protease, antagonizes ABA signaling in *Arabidopsis*. New Phytol. 208:188–197.

Zhou, X., Hao, H., Zhang, Y., Bai, Y., Zhu, W., Qin, Y., Yuan, F., Zhao, F., Wang, M., Hu, J., et al. (2015b). PKS5/CIPK11, a SnRK3-Type protein kinase, is important for ABA responses in *Arabidopsis* through phosphorylation of ABI5. Plant Physiol. 168:659–676.

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should be minimum seed loss

Upgrading should be possible for any particular quality

There should be have more efficiency

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Inert materials

Common weed seeds

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Sequence of operations are based on characteristics of seed such as shape, size, weight, length, surface structure, colour and moisture content. Because each crop seed possesses individually seed structure. Therefore, sequence of operation will be applied proper equipments. However, It is also involvedstages following as

Drying

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Conditioning

Cleaning

Separating or Upgrading

Treating (Drying)

Weighting

Bagging

Storage or Shipping

SEED STORAGE

What is seed storage - preservation of seed with initial quality until it is neededfor planting.

Stages of Seed Storage

The seeds are considered to be in storagefrom the moment they reach physiological maturity until they germinate or until they are thrown away because they are dead or otherwise worthless.

The entire storage period can be conveniently divided into followingstages.

Storage on plants (physiological maturityuntil harvest).

Harvest, until processed and stored in a warehouse.

In - storage (warehouses)

In transit (Railway wagons, trucks, carts, railway sheds etc.).

In retail stores.

On the user's farm.

Introduction

The ability of seed to tolerate moisture loss allows the seed to maintain the viability in dry state. Storage starts in the mother plant itself when it attains physiological maturity. After harvesting the seeds are either stored in ware housesor in transit or in retail shops. During the old age days , the farmers were used farm saved seeds, in little quantity, but introduction of high yielding varieties and hybrids and modernization of agriculture necessitated the development of storage techniques to preserve the seeds.

The practice of storing the seeds starts from the ancient days itself, following simple and cheap techniques e.g. Placingthe seeds in salt, red earth treatment to red gram etc. But the same practices are not hold good for the present day agriculture, because

large quantity to be stored exchange

of varieties and speciesexchange of

genes

The type of material to be stored decidesthe techniques to be followed for safe storage. Now a day's storage technique changed from ordinary go-down storage to cryogenic tank storage and even gene storage.

Objective of seed storage

To maintain initial seed quality viz., germination, physical purity, vigour etc., all along the storage period by providing suitable or even better conditions.

Since the main objective of seed storage is maintenance of an acceptable capacityfor germination and emergence, it can only be accomplished by reducing the rateof deterioration to the degree required to maintain an acceptable level of quality forthe desired period.

Purpose of seed storage

Seed storage is the maintenance of high seed germination and vigour form harvestuntil planting. Is important to get adequate plant stands in addition to healthy and vigourous plants. Every seed operation has or should have a purpose.

The purpose of seed storage is to maintain the seed in good physical and physiological condition from the time they

are harvested until the time they are planted. Seeds have to be stored, of course, because there is usually a periodof time between harvest and planting.
During this period, the seed have to be kept somewhere. While the time interval between harvest and planting is the basicreason for storing seed, there are other considerations, especially in the case of extended storage of seed.

Seed suppliers are not always able to market all the seed they produce during the following planting season. In many cases, the unsold seed are “carried over”in storage for marketing during the second planting season after harvest.
Problems arise in connection with carryover storage of seed because somekinds, varieties, and lots of seed do not carryover very well.

Seeds are also deliberately stored for extended periods so as to eliminate the need to produce the seed every season. Foundation seed units and others have found this to be an economical, efficient procedure for seed of varieties for which there is limited demand. Some kinds of seed are stored for extended periods to improve the percentage and rapidity of germination by providing enough time for

a “natural” release from dormancy. Regardless of the specific reasons for storage of seed, the purpose remains thesame maintenance of a satisfactory capacity for germination and emergence.The facilities and procedures used in storage, therefore, have to be directed towards the accomplishment of this purpose.

In the broadest sense the storage periodfor seed begins with attainment of physiological maturity and ends with resumption of active growth of the embryonic axis, i.e., germination. Seedsare considered to be physiologically andmorphologically mature when they reachmaximum dry weight. At this stage
dry-down or dehydration of the seed is well underway. Dry-down continues after physiological maturity until moisture content of the seed and fruit decreases toa level which permits effective and efficient harvest and threshing. This stage can be termed as harvest maturity.
There usually is an interval of time between physiological maturity and harvestable maturity, and this interval represents the first segment of the storage period. Any delays in harvesting the seed after they reach harvest maturityprolongs the first segment of the storage

period – often to the detriment of seed quality.

The second segment of the storage period extends from harvest to the beginning of conditioning. Seed in the combine, grain wagon, and bulk storage ordrying bins are in storage and their qualityis affected by the same factors that affectthe quality of seed during the packaged seed segment of the storage period. The third segment of the storage period begins with the onset of conditioning and ends with packaging. The fourth segmentof the storage period is the packaged seed phase which has already been mentioned. The packaged seed segment is followed by storage during distribution and marketing, and finally by storage on the farm before and during planting.

The control that a seedsman has over the various segments of the storage period for seed varies from a high degree of control from harvest to distribution, to much less control during the postmaturation-preharvest, distribution-marketing, and on-farm segments. Despite variable degrees of control over the various segments of the storage period, the seedsman’s plans for storage must take into consideration all

the segments. The things that can be done must be done if the quality of theseed is to be maintained.

SEED CERTIFICATION

Definition: Seed certification is a legally sanctioned system for quality control ofseed multiplication and production.

Purpose of seed certification

The purpose of seed certification is to maintain and make available to the public, through certification, high quality seeds and propagating materials of notified kindand varieties so grown and distributed as to ensure genetic identity and genetic purity.

IMPORTANCE OF CERTIFIED SEED

Certified seed is the starting point to a successful crop as well as an importantrisk management tool.

Here are the top 10 reasons to use certified seed:

You’re getting clean seed

Certified seed is grown under stringent production requirements and has minimalweed seeds or other matter.

You’re getting varietal purity

Certified seed uses systems to maximize genetic purity Off-types, other crop seeds,and weeds are guaranteed to be minimized

You’re getting guaranteed qualityassurance

Third party inspections in the field and at the processing plant ensure that all quality assurance requirements have beenmet. You can rest easy knowing your seedis what you expect it to be and can back up your assurances to others.

You’re getting access to new opportunities

Many end-users are requiring specific varieties for their products.Using certifiedseed can open the door to new opportunities and greater sales by providing proof of varietal identity.

You’re getting new genetics Improved

traits like better yield, pest resistance, drought tolerance, herbicide tolerance, and much more are delivered to

farmers in certified seed. Years of research and development went into these traits and they can only be reliablyaccessed through certified seed use.

You’re getting substance behind yourword

The blue tag is proof that you used certified seed to maintain the value traitsof the crop. It’s your assurance to grain buyers and others that what you are delivering is what you say it is.

You’re getting a better deal on crop insurance

Certified seed use can, in some cases, allow you to get a better deal on crop insurance premiums. Insurers know that certified seed means a crop with reducedrisk.

You’re getting maximum use of other inputs

You want the best genetics and purest fields to ensure you are making the most of your input dollars. Certified seed meansyou’re not wasting time and other inputs on a crop that won’t make grade.

You’re getting access to premium markets

Proper inputs make for a good crop, butseed is the only input that can get you more than higher yields. Use of certifiedseed can be your ticket to premium markets like tofu soybeans or high stability canola and MORE.

You’re getting traceability

Food safety and traceability are important considerations in agriculture. You can only be sure of your product if you know its origins. Certified seed is the key to thatknowledge: production of this seed is carefully controlled under a quality assurance system right from the very beginning. Using certified seed will allow you to capitalize on a whole history of traceability measures.

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Seed Germination Types

The three main types are:
(1) Hypogeal Germination (2) Epigeal Germination and (3) Vivipary (Viviparous Germination).

Type # 1. Hypogeal Germination:

In this kind of germination, the cotyledonsdo not come out of the soil surface. In such seeds the epicotyl (i.e., part of embryonic axis between plumule and cotyledons) elongates pushing the plumule out of the soil. All monocotyledons show hypogeal germination . Among dicotyledons, gram, pea , groundnut are some common examples of hypogeal germination.

In monocotyledons (e.g., wheat, maize, rice, coconut) radicle and plumule come out by piercing the coleorrhiza and coleoptile respectively. The plumule growsupward and the first leaf comes out of thecoleoptile. The radicle forms the primary root which is soon replaced by many fibrous roots.

Type # 2. Epigeal Germination:

In seeds with epigeal germination, the cotyledons are brought above the soil dueto elongation of the hypocotyl. In castor, cotton, papay, onion (Figs. 4.7., 4.8), flat green leaf like cotyledons can be seen in the young seedlings. Here the cotyledons,besides food storage, also perform photosynthesis till the seedling becomes independent. In some other plants like bean, the cotyledons being thick, do not become leaf-like; they shrival and fall off after their food reserves are consumed by the seedling.

Type # 3. Vivipary (Viviparous Germination):

Vivipary is the phenomenon of giving birthto young ones in advanced stage of development. It occurs in mammals (among animals) and mangrove plants. Inmangrove plants (e.g., Rhizophora, Sonneratia, Heritiera) the seeds cannot germinate on the ground because of the excessive salt content and lack of oxygenin marshy habitat. In such plants seed dormancy is absent.

The embryo of the seed (present inside the fruit) continues growth while the latteris attached to the parent plant. Hypocotyl elongates and pushes the radicle out of the seed and the fruit. Growth continues till the hypocotyl and radicle become several centimetres long (more than 70 cm in Rhizophora). The seedling becomesheavy.

As a result it breaks its connection with the fruit and falls down in the salt rich muddy water in such a position that the plumule remains outside the saltish waterwhile the tip of the radicle gets fixed in themud. This protects the plumule. The radicle quickly forms new roots and establishes the seeding as a new plant.

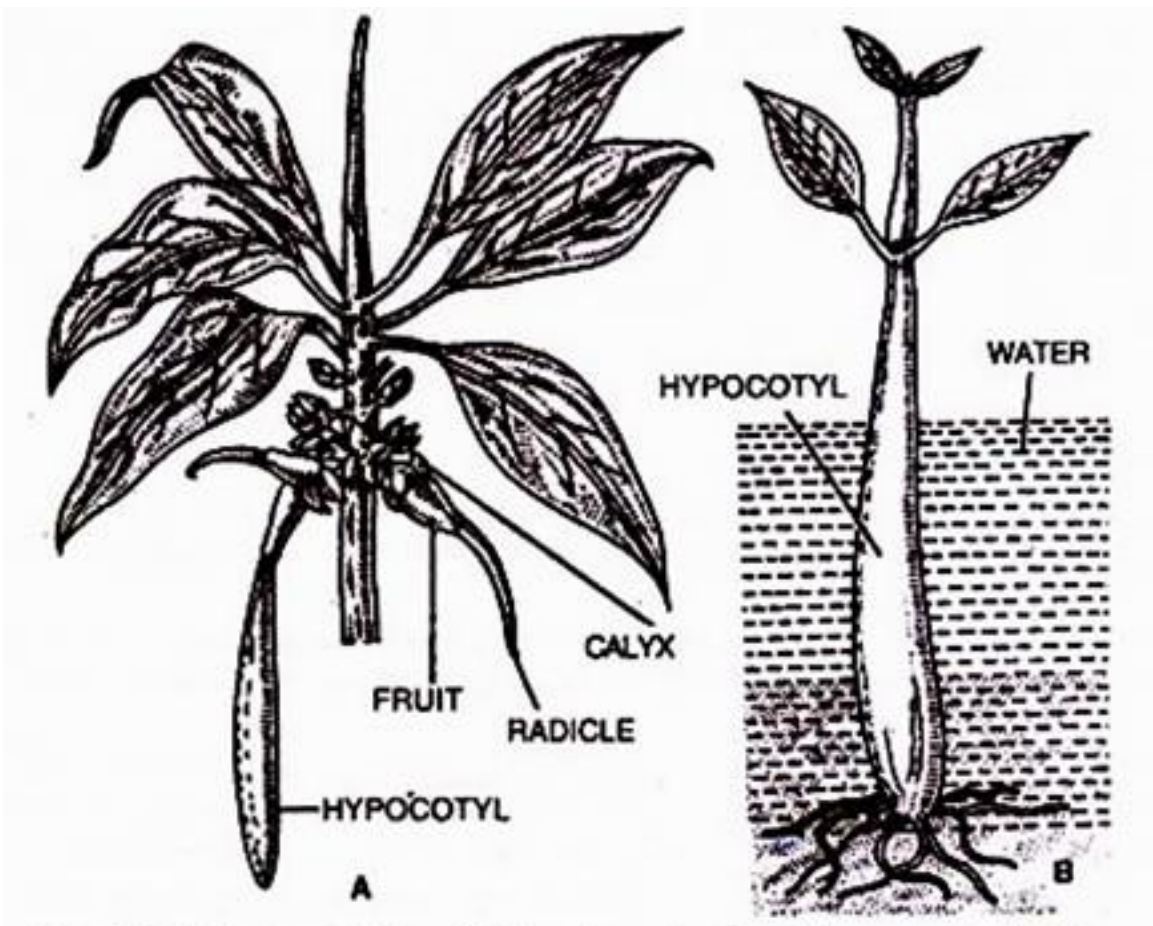

Fig. 4.9. Vivipary. A. twig of *Rhizophora* showing viviparous germination, **B.** A seedling has become established on tidal soil.

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Weighting

Bagging

Storage or Shipping

15. SEED VIGOUR TESTING

P.C. GUPTA

Seed vigour is an important quality parameter which needs to be assessed to supplement germination and viability tests to gain insight into the performance of a seed lot in the field or in storage. Several definitions have been offered to explain seed vigour. Looking into the complexity of the situation, the ISTA congress in 1977 adopted the definition of seed vigour as " the sum total of those properties of the seed which determine the level of activity and performance of the seed or seed lot during germination and seedling emergence". Although differences in physiological attributes of seed lots can be demonstrated in the laboratory, it was recommended that the term should be used to describe the performance of seeds when sown in the field (Perry, 1984a).

As the germination test is conducted in an optimum condition specific to different species, it is not always possible to get an idea of the performance of a seed lot in the field on the basis of germination test in the laboratory. It is mainly because of the reason that field conditions are seldom optimum and the emerging seeding suffers from one or the other kind of stress. In many cases seed lots having similar laboratory germinations may give widely differing field emergence values. Similarly, two seed lots having the same germination percentage in the laboratory may age differently when stored under ambient condition. These two situations indicate the incompletences of germination test in assessing the performance of a seed lot in the field or storage. This offers scope and possibility to determine vigour of a seed lot so that its field and storage performance can be assessed.

Seed vigour is still a concept rather than a specific property of a seed or seed lot. Several factors like; genetic constitution, environment and nutrition of mother plant, maturity at harvest, seed weight and size, mechanical integrity, deterioration and ageing and pathogens are known to influence seed vigour (Perry, 1984a). Therefore, care has to be exercised in selecting a seed vigour test to do the job. Two criteria have been employed by the ISTA seed vigour committee to evaluate the performance of seed vigour test methods for different crops :

(i) Reproducibility of vigour method

(ii) The relationship between vigour test results and seedling emergence in field soil.

There is no universally accepted vigour test for all kinds of seeds. The determination of following vigour tests will be useful in gaining additional information on seed quality.

1. Growth Tests

Principles : Growth tests are based on the principle that vigorous seeds grow at a faster rate than poor vigour seeds even under favourable environments. Vigorous seeds rapidly germinate, metabolize and establish in the field. Therefore, any method used to determine the rapidity of growth of the seedling will give an indication of seed vigour level.

Apparatus and equipment : All the equipments and materials needed to conduct a germination test are required. Additionally, a top loading balance and an air oven are also required.

Procedure

(a) First count : The test is done along with the regular germination test. The number of normal seedlings, germinated on the first count day, as specified in the germination test for each species, are counted. The number of normal seedlings gives an idea of the level of seed vigour in the sample. Higher the number of normal seedlings greater is the seed vigour.

(b) Seedling growth rate and dry weight : The seedlings are grown either in laboratory, green house or field. In laboratory, in between rolled towel paper method should be followed. Ten seeds are planted in the centre of the moist towel papers in such a way that the micropyles are oriented towards bottom to avoid root twisting. The rolled towel papers are kept in the germinator maintained at a temperature recommended for crop in reference. After a specified period of time (5-10 days) towel papers are removed and five seedlings are selected, their length is measured and mean seedling length is calculated. Seed lots producing the taller seedlings are considered more vigorous than the seed lots producing shorter seedlings. For dry weight determination, the seedlings are removed and dried in an air oven at 100°C temperature for 24 hours. The seedling dry weight provides additional information for assessing seed vigour.

(c) Speed of germination : One hundred seeds each in four replications are planted in recommended substratum for germination. The substratum is kept in a germinator maintained at recommended temperature for the crop in reference (Table 5.1). Number of seedlings emerging daily are counted from day of planting the seeds in the medium till the time germination is complete. Thereafter a germination index (G.I.) is computed by using the following formula :

$$G.I. = \frac{n}{d}$$

where, n = number of seedlings emerging on day 'd'

d = day after planting

The seed lot having greater germination index is considered to be more vigorous.

Example

Seed lot A — No. of seedlings= 0,0,0,40,30,12,7, counted

Day of counting = 1,2,3,4,5,6,7

Seed lot B — No. of seedlings =0,0,0,0,30,42,21 counted

G.I. of Seed lot A

$$\frac{0}{1}+\frac{0}{2}+\frac{0}{3}+\frac{40}{4}+\frac{30}{5}+\frac{12}{6}+\frac{7}{7}$$

$$= 10 + 6 + 2 + 1$$

$$= 19$$

G.I. of Seed lot B

$$\frac{0}{1}+\frac{0}{2}+\frac{0}{3}+\frac{0}{4}+\frac{30}{5}+\frac{42}{6}+\frac{21}{7}$$

$$= 6 + 7 + 3$$

$$= 16$$

In this example seed lot A has greater G.I. (19) than seed lot B (16), so seed lot A is more vigorous than seed lot B.

(d) Seed vigour index (S.V.I.) : This is calculated by determining the germination percentage and seedling length of the same seed lot. Fifty seeds each in four replications are germinated in towel papers as prescribed for the crop species in germination test. While evaluating the number of normal seedlings at the time of final count, the seedling length of 5 randomly selected seedlings are also measured. Seed vigour index is calculated by multiplying germination (%) and seedling length (mm). The seed lot showing the higher seed vigour index is considered to be more vigorous (Abdul-Baki and Anderson, 1973).

Example

Seed lot	% germination	Seedling length, mm	Vigour index
A	96	85	8160
B	95	76	7220
C	94	71	6674

In this example seed lot A is the most vigorous and seed lot C the least vigorous as they have the highest and the lowest values of seed vigour index, respectively.

2. Conductivity Test

Principle : Weakening of cell membrane in poor vigour seeds causes leakage of water soluble compounds like sugars, amino acids, electrolytes etc. when immersed in water. On the other hand, fresh seeds having intact membrane leach less quantity of these chemicals. The measurement of electrical conductivity (EC) of the leachate by a good and sensitive conductivity meter gives an accurate estimation of membrane permeability. The EC has been positively correlated with the emergence percentage of peas and broad beans (Mathews and Bradnock, 1968). The value of this test appears to be restricted to the large seed species of the Leguminoceae (Perry, 1984b).

Apparatus and equipment : Conductivity meter, beaker, 0.1% mercuric chloride, distilled water, seed sample, wash bottle an tissue paper.

Procedure : A seed sample of 2-5 gram is weighed and surface sterilized with 0.1% $HgCl_2$ for 5-10 minutes. The sample is washed thoroughly in distilled water. The clean seeds are immersed in 100 ml of water at 25 ± 1°C temperature for 10-12 hours. After this the seeds are removed with a clean forcep. The steep water left is decanted and is termed as leachate.

The conductivity meter is warmed for about 30 minutes before testing. First the conductance of distilled water is measured in a beaker. The electrode is then cleaned with a tissue paper and conductance of the leachate is read. The electrode is thoroughly washed using a wash bottle and wiped with a clean tissue paper before reusing. While recording the conductance, the lower bulb of the electrode should be fully emerged in the leachate. To get the EC of leachate the reading of distilled water is substracted from the sample reading. The value is then corrected for the temperature and multiplied by the cell constant factor. The reading is expressed as mu mhos/cm/g of seed. Lower the value of EC greater is the seed vigour.

3. Hiltner Test (Brick gravel test)

Principle : The test was developed by Hiltner in Germany in 1917. He observed that the seeds of cereal crops affected by Fusarium disease were able to germinate in regular test but were not able to emerge from brick gravels of 2-3 mm size. Compared to this, healthy seeds were able to emerge from the brick gravel (Robersts, 1972). The principle is that the weak seedlings are not able to generate enough force to overcome the pressure of brick gravels, so this method can be used to differentiate vigour levels in cereal seeds. Perry (1984b) found this method reproducible and associated with field emergence in case of wheat.

Apparatus and equipment : Germination box, aluminium tray, sand, sand marker brick gravel of 2-3 mm size, germinator, seed sample.

Procedure : The sand is seived, moistured and filled in the germination box leaving about 3 cm empty at the top. One hundred seeds are placed in each box in the impressions made by a sand marker. After this 2-2.5 cm of porous brick gravel is spread over the seeds. The box is kept in the germinator at appropriate temperature. After the period required for germination, the box is removed and the seedlings which have emerged through the brick gravel layer are counted. The percentage of emerged seedlings are used to compare seed vigour of different lots. The test should be repeated 3-4 times to get authentic value.

4. Paper Piercing Test

Principle : The principle of paper piercing test is similar to that of brick gravel test. High vigour seed lots are expected to produce strong seedlings which can pierce a particular type of paper while seedlings of poor vigour lots may not be able to pierce the paper. Therefore, the seedlings which emerge by piercing the paper

are more vigorous than those which are not able to emerge through the paper.

Apparatus and equipment : All the material required for conducting germination test in sand boxes or trays plus the special paper which should have the following characteristics :

(a) Basic weight = 90 g/m^2

(b) Thickness = 0.4 mm

(c) Bulk = 4

(d) Dry bursting strength = 0.3 kg/cm^2

(e) Breaking length = 1000-5000 mm

(f) Filtering speed = 500 ml/minute

(g) Wet bursting strength = 150 mm

(h) Ash content = 0.1%

(i) Fibre composition = Chemical wood pulp with high alpha percentage

Procedure : The cereal seeds are placed on 1.5 cm moist sand in a tray or sand box. The seeds are covered with specially selected dry filter paper, which is then covered with 2 cm of moist sand. After this, the sand boxes/trays are kept in a germinator maintained at 20°C temperature for 8 days. After 8 days sand boxes/trays are taken out and seedlings emerging above the paper are counted. A seed lot having maximum number of seedlings coming out of paper is considered to be most vigorous. *The test is highly dependant on the quality of paper and should be used when such papers are available.*

5. Cold Test

Principle : The cold test has been developed in USA to evaluate the seed vigour of maize (corn). In USA when the corn is planted in late spring, the soil is humid and cold. The weak seeds do not germinate and establish. Therefore, to simulate the actual field conditions witnessed at the time of corn planting, cold test

has been developed. The test aims to differentiate between weak and vigorous seed lots by subjecting them to low temperature prior to germination at optimum temperature. The test has been criticized for using field soil which greatly varies from place to place.

Apparatus and equipment : Aluminium tray, field soil, sand marker, germinator, seed sample.

Procedure : After grinding and properly sieving the soil is filled in tray upto 2 cm depth. Fifty seeds are placed over the sand and covered with another 2 cm thick layer of soil. The soil is compacted and enough water is added to make the soil about 70% of its water holding capacity. The temperature of the water should be 10°C. After watering the trays are covered with polythene bags and placed in the refrigerator maintained at 10°C temperature for one week. After one week the trays are removed and placed in the germinator at 25°C temperature. The seedlings emerged after 4 days are counted. The germination percentage is computed by counting the number of normal seedlings as in germination test. Higher the germination percentage greater is the vigour.

6. Accelerated Ageing Test

Principle : The accelerated ageing test has been developed at the Seed Technology Laboratory, Mississippi State University, USA for determining the storage potential of seed lots. The ageing process is accelerated by subjecting the seeds to high temperature and relative humidity in a chamber before standard germination. The seed lots that show high germination in accelerated ageing test are expected to maintain high viability during ambient storage as well. Thus, ageing test gives an indication of the performance of the seed lot during ambient storage. Tests conducted at Pantnagar with Bragg soybean seeds have shown positive relationship between 3 days accelerated ageing test (42- 45°C temperature, 95-100% R.H.) and viability after 6 months of ambient storage (Gupta, 1980). However, Perry (1984b) reported inconsistency in accelerated ageing test results and not well related to field emergence of maize and soybean. The test also suffers from fungal growth on seeds at high temperature and humidity (Agrawal, 1987). This test is recommended for soybean seeds.

Apparatus and equipment : Acceleratged aging chamber, equipment for germination test, seed samples, tight jar, muslin cloth, wire mesh etc.

Procedure : One hundred seeds each in four replications are tied in a fine muslin cloth. The tied seeds are placed in jar on a wire mesh. The lower part of the jar is filled with water. There should not be a direct contact between water and the seed. The jar is covered with the lid and sealed with parafin wax to make it air tight. The jar is then placed in the accelerated aging chamber maintained at 45 ± 2°C temperature for 3-5 days. The jar is removed after this period and the seeds are cooled in a dessicator. The seeds are then tested in a normal germination test specific to different crops. The percent germination gives level of seed vigour. Higher the germination percentage greater is the vigour of the seed.

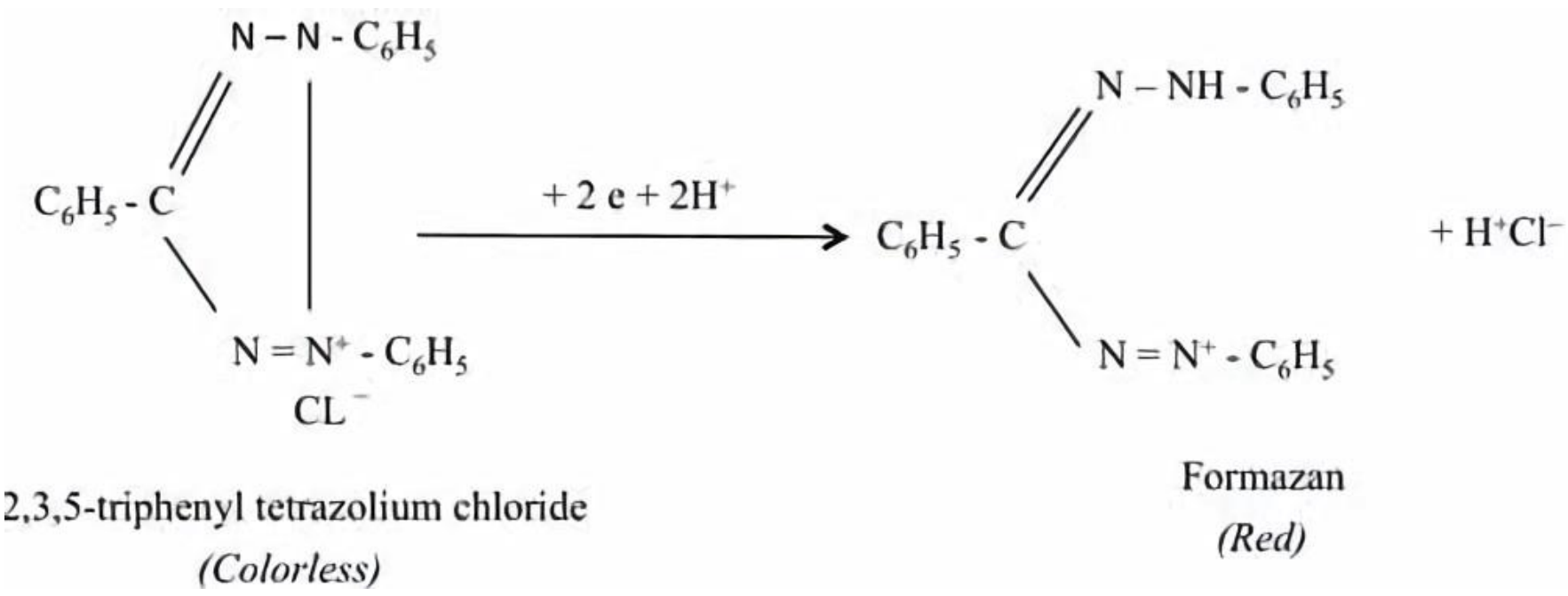
Future Role of Seed Vigour Testing

Seed vigour is an important component of seed quality and satisfactory levels are necessary in addition to traditional quality criteria of moisture, purity, germination and seed health to obtain optimum plant stand and high production of crops. As agricultural and horticultural techniques become progressively more sophisticated, the need for high vigour seeds will increase and testing standards, similar to those reconginzed for germination will be required (Perry, 1984b). The technology of seed vigour testing has not been perfected so far, so much so that there is not a single universally accepted seed vigour test method. Research is needed to further refine the current seed vigour test methods and to develop new methods which are more related to field/storage conditions.

Tetrazolium Testing

Tetrazolium (TZ) testing is a rapid method(can be finished within less than two days) for the evaluation of seed viability.
This method has been widely used by seed scientists to assess germination potentials, to determine the extent of seeddamage, and to evaluate seed vigor and/ or other seed lot problems.

The principle of TZ testing is based on the presence of dehydrogenase activity in viable seed tissues during the respiration process. Dehydrogenase can catalyze thecolorless 2,3,5 triphenyl tetrazolium chloride solution into a red dye formazan (Figure 1). Therefore, living tissues of seeds that imbibe tetrazolium chloride willbe stained red, while dead tissues will retain their natural color (Figure 2).



2,3,5-triphenyl tetrazolium chloride
(Colorless)

Formazan
(Red)

Figure 1. The principle of TZ testing (Elias et al., 2012).

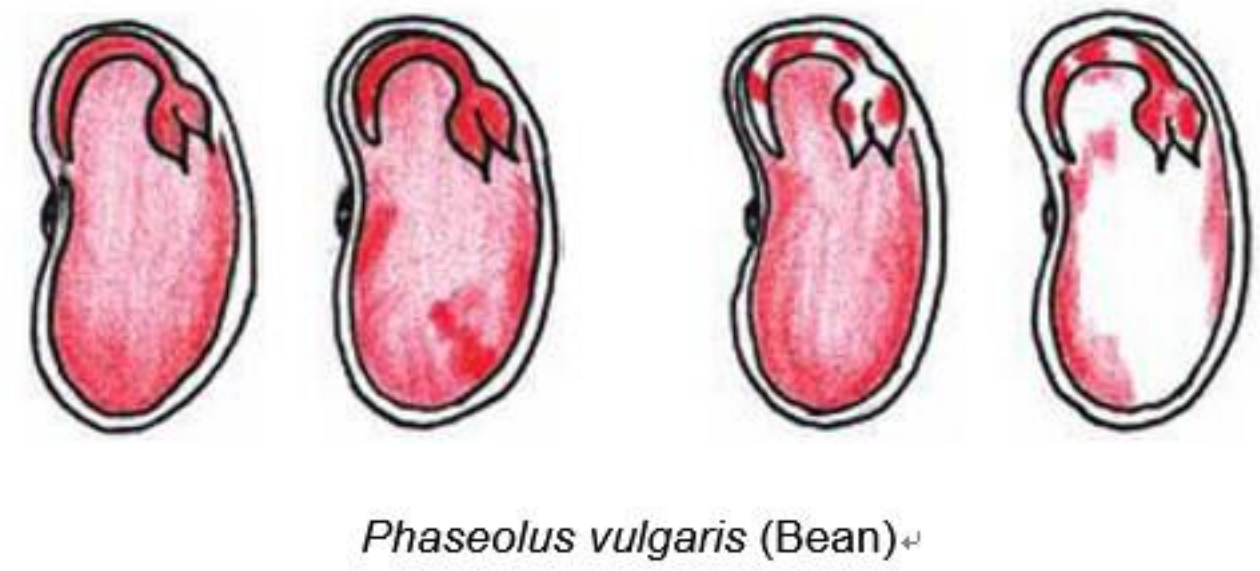


Figure 2. Examples of TZ testing for seed viability test (Elias et al., 2012). (Left two: viable seeds; Right two: non-viable seeds)

A standard TZ testing assay involves the following steps:

Preparation of dry seed. The seed coats of many species are best to be treated by mechanical abrasion so that water or solutions can penetrate into the interior tissues of the seed.

Moistening. This step aims to hydrate the seeds to activate the respiratory enzymesand to soften the tissues for cutting and piercing.

Preparation for staining. Cut or pierce the

seed to facilitate the absorbing of TZ solution by internal tissues.

Staining. Soak the cut seeds in TZ solution for certain periods of time.

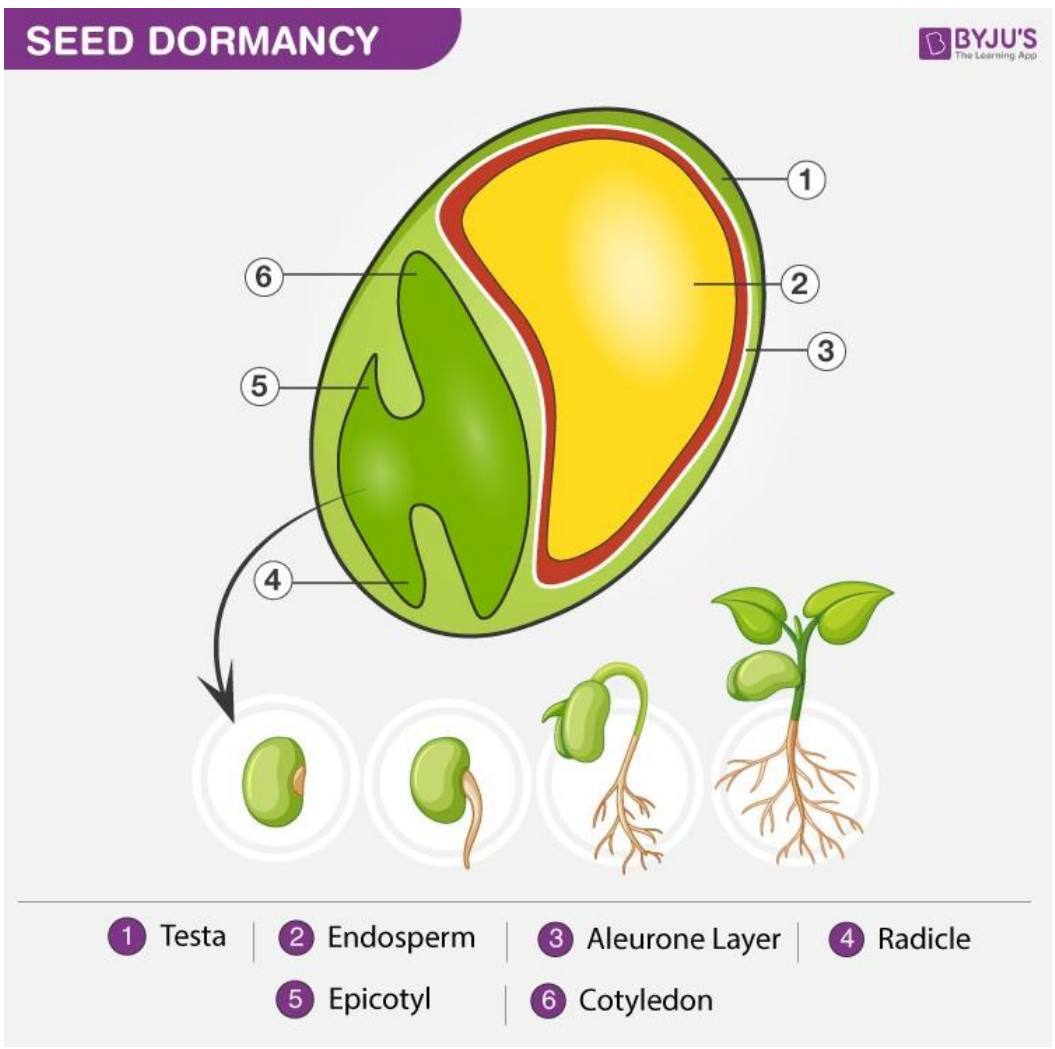
Evaluation of staining patterns. Examine the seed for a color change in the embryo.

Seed Dormancy

Seed dormancy can be defined as the state or a condition in which seeds are prevented from germinating even under the favourable environmental conditionsfor germination including, temperature, water, light, gas, seed coats, and other mechanical restrictions.

The main reason behind these conditionsis that they require a period of rest beforebeing capable of germination. These conditions may vary from days to monthsand even years. These conditions are the combination of light, water, heat, gases, seed coats and hormone structures.

Reasons or Causes of the Seed Dormancy



There are certain major causes for the seed dormancy. Listed below are the fewreasons for the seed dormancy.

Light

Temperature

Hard Seed Coat

Period after ripening Germination

inhibitors Immaturity of the seed

embryo

Impermeability of seed coat to water

Impermeability of seed coat to oxygen

Mechanically resistant seed coat Presence of

high concentrate solutes

Seed dispersal

Seed dispersal is the movement, spread or transport of seeds away from the parent plant. Plants have limited mobility and rely upon a variety of dispersal vectors to transport their propagules, including both abiotic vectors such as thewind and living (biotic) vectors like birds.

Seeds can be dispersed away from the parent plant individually or collectively, aswell as dispersed in both space and time. The patterns of seed dispersal are determined in large part by the dispersal mechanism and this has important implications for the demographic and genetic structure of plant populations, as well as migration patterns and species interactions. There are five main modes ofseed dispersal: gravity, wind, ballistic, water, and by animals. Some plants
are serotinous and only disperse their seeds in response to an environmental

stimulus. Dispersal involves the letting goor detachment of a diaspore from the main parent plant.

Animal dispersal (zoochory)

Many animal-dispersed fruits are dispersed by vertebrates—especially certain mammals and birds, although fishand reptiles can also act as dispersal agents—or ants. Vertebrate-dispersed fruits and seeds may be fleshy, or may have fleshy coverings; ant-dispersed seeds often have nutrient-rich appendages.

Wind dispersal (anemochory)

Fruits and seeds that are wind-dispersed frequently have modifications that help slow their descent to the ground and increase the chances that they will be blown laterally by air currents, so that theydo not land directly beneath or next to their parent plant. Seed modifications for wind dispersal can include small size and/or light weight, wings, hairs, and/or inflation.

One of the most obvious modificationsfor wind-dispersal is the wing. Winged fruits are common in the fossil record

beginning in the Paleogene. Winged fruitsor seeds often have a single wing, in which case the wing may be asymmetrical, or offset to one side of the fruit or seed. If they have more than one wing, the wings many be regularly arranged around the fruit or seed. Often, the structure of the wing or wings will cause a seed or fruit to spin or rotate as itfalls (known as autorotation, i.e., self-rotation). Maples (Acer) produce familiar wind-dispersed fruits that spin as they fall. If you live in a neighborhood withmaple trees, you can observe this yourself; watch the mature fruits as they fall from a tree on a windy day, or pick up the fallen mericarps (fruit halves) and drop them to watch them spin as they fall.

Water dispersal (hydrochory)

Plants that live in wetland environments or near the ocean may have buoyant, or floating, fruits or seeds. Cranberries (*Vaccinium* species) are low-growing plants found in boggy environments. Their bright red berries arenot particularly sweet, and thus probablynot terribly attractive to animals. Cranberries do, however, float, which aidsin their dispersal in wetland habitats. It has been hypothesized that cranberries

evolved from ancestors that had more palatable, animal-dispersed fruits. Humans take advantage of the berries' buoyancy during commercial production,as cranberry bogs can be flooded so thatthe floating berries can be more easily collected.

Some plants with floating fruits or seeds can disperse long distances over the ocean. The most obvious example of this is the coconut palm (*Cocos nucifera*), which has large, fibrous fruits that canfloat to and colonize oceanic islands. Similarly, legumes in the genus *Adenanthera* produce large, buoyant seeds; each seed harbors an air pocket, which enhances it ability to float.

Explosive dispersal (autochory or bolochory)

Amongst the more novel and exciting ways in which seeds are dispersed is through ballistic or explosive dispersal. Inthis method of dispersal, the fruit forciblyejects the seed(s), scattering them for a short distance. The common garden plant (also known as balsam, touch-me-not, and jewelweed, amongst other names) is one such plant. It produces capsules. When ripe, an animal

brushing by the plant can cause the capsule to open instantly, scattering theseeds. Another plant with dramatic explosive seed dispersal is the squirting cucumber (*Ecballium elaterium*), which ejects its seeds as the fruit detaches fromits stalk.

SEED CERTIFICATION

Definition: Seed certification is a legally sanctioned system for quality control ofseed multiplication and production.

Purpose of seed certification

The purpose of seed certification is to maintain and make available to the public, through certification, high quality seeds and propagating materials of notified kindand varieties so grown and distributed as to ensure genetic identity and genetic purity.

IMPORTANCE OF CERTIFIED SEED

Certified seed is the starting point to a successful crop as well as an importantrisk management tool.

Here are the top 10 reasons to use certified seed:

You’re getting clean seed

Certified seed is grown under stringent production requirements and has minimalweed seeds or other matter.

You’re getting varietal purity

Certified seed uses systems to maximize genetic purity Off-types, other crop seeds,and weeds are guaranteed to be minimized

You’re getting guaranteed qualityassurance

Third party inspections in the field and at the processing plant ensure that all quality assurance requirements have beenmet. You can rest easy knowing your seedis what you expect it to be and can back up your assurances to others.

You’re getting access to new opportunities

Many end-users are requiring specific varieties for their products.Using certifiedseed can open the door to new opportunities and greater sales by providing proof of varietal identity.

You’re getting new genetics Improved

traits like better yield, pest resistance, drought tolerance, herbicide tolerance, and much more are delivered to

farmers in certified seed. Years of research and development went into these traits and they can only be reliablyaccessed through certified seed use.

You’re getting substance behind yourword

The blue tag is proof that you used certified seed to maintain the value traitsof the crop. It’s your assurance to grain buyers and others that what you are delivering is what you say it is.

You’re getting a better deal on crop insurance

Certified seed use can, in some cases, allow you to get a better deal on crop insurance premiums. Insurers know that certified seed means a crop with reducedrisk.

You’re getting maximum use of other inputs

You want the best genetics and purest fields to ensure you are making the most of your input dollars. Certified seed meansyou’re not wasting time and other inputs on a crop that won’t make grade.

You’re getting access to premium markets

Proper inputs make for a good crop, butseed is the only input that can get you more than higher yields. Use of certifiedseed can be your ticket to premium markets like tofu soybeans or high stability canola and MORE.

You’re getting traceability

Food safety and traceability are important considerations in agriculture. You can only be sure of your product if you know its origins. Certified seed is the key to thatknowledge: production of this seed is carefully controlled under a quality assurance system right from the very beginning. Using certified seed will allow you to capitalize on a whole history of traceability measures.

Hormonal Regulations During Seed Dormancy:

(i) Gibberellins:

The germination of both dormant and non-dormant seeds has been shown to be stimulated by applied GAs. The stimulatory effect of GAs has been widely reported in seeds where dormancy or quiescence is imposed by different mechanisms like incomplete embryo development, mechanically resistant seed coats and presence of germination inhibitors.

Based on these observations, Amen postulated that GAs play a universal role in seed germination. Endogenous GA levels have been found to undergo change in relation to dormancy breaking conditions.

There are reports which show that low temperature treatment of dormant seeds lead to increased level of GA-like substances and germination. Dormancy induced in hazel (Corylus avellana) seeds by dry storage can be overcome either by GA or by cold treatment or stratification.

From the observation that GA content increases when the hazel seeds are transferred from the stratification temperature (5°C) to the germination temperature (30°C), Williams et al., postulated that the dormancy-breaking effect of chilling is to activate the GA-producing mechanism, whereas the subsequent synthesis takes place at higher germination temperature.

It is generally observed that the cereal seeds are not able to germinate after harvest and storage for 1 -3 months at room temperature is necessary to allow maximum germination.

Thus, dormancy in these seeds can be removed by dry storage after-ripening period during which the embryos acquire the capacity to produce GA. A study on GA synthesis by germinating barley by Radley suggests that the synthesis of GA by the embryo scutellum is regulated by the levels of sugars present in the endosperm.

He proposed that the inhibition of GA synthesis by the accumulation of sugars could serve an important regulatory role in cereal seed germination. Premature germination may be prevented by sugars present in the ripened grains whereas depletion of endosperm sugars during dry after-ripening would permit the production of GA and subsequent germination.

In the germination of photo dormant seeds, light influences GA metabolism. Both positively photoblastic seeds (e.g., Lactuca sativa) and negatively photoblastic seeds (e.g., Phacelia tenacetifolia) germinate under non-inductive conditions when treated with GA. It was shown that red light illumination of lettuce seeds could increase GA-like substances.

(ii) Cytokinins:

It was discovered that the application of exogenous cytokinins can counteract the blockage of GA-induced germination or enzymatic processes by naturally-occurring germination inhibitors. This would suggest that the hormonal control of seed dormancy and germination involves a balance between stimulatory and inhibitory compounds within the seed.

Based on this concept, Khan proposed a model in which the gibberellins assume the primary role in germination, whereas cytokinins and inhibitors are essentially permissive and preventive, respectively. Burrows (1975) has suggested three major sites at which cytokinins are involved in seed germination.

These are:

(a) Control at the gene level—a cytokinin-receptor protein has been isolated from pea bud chromatin and there is a direct interaction between cytokinins, its receptor protein and chromatin. Binding of cytokinins to ribosomes has also been confirmed,

(b) Control at the translational level—presence of cytokinin in different amino acid specific tRNA suggests that there is a specific relationship between cytokinins and tRNA and they have action on protein synthesis. Cytokinin- binding sites on higher plant ribosomes may constitute the control mechanism for determining which tRNA species are permitted access to the codon.

(c) Regulation of membrane permeability—cytokinins affect the permeability of cell membranes in a wide variety of plant tissue including seed. This is supported by the fact that cytokinins can influence many phytochrome-controlled processes and that phytochrome is involved in altering the selective permeability of cell membrane,

(iv) Regulation of GA levels—there is considerable evidence that red light stimulates the release of GA from etiolated chloroplasts and it is suggested that the primary effect is increased permeability of etioplast membrane.

Since cytokinin effect is similar to red-light effect, it seems possible that they can control membrane permeability and release of GAs. This hypothesis can explain as to why the low levels of GAs which are normally without effect on seed germination become effective when combined with cytokinins. The cytokinin-inhibitor antagonism model proposed by Khan (1975) is quite consistent with this hypothesis.

(iii) Abscisic Acid:
Several reports indicate that endogenous ABA levels drop in seeds which are subjected to a dormancy-breaking treatment like cold stratification and germination can also be inhibited by exogenous ABA.

The addition of ABA specifically inhibits the synthesis of certain enzymes which play a key role in germination. One of the earliest reports of the effect of ABA is its inhibition of GA-induced hydrolase synthesis in barley aleurone layers. Inhibition of hydrolase synthesis by ABA appears to be at the level of translation rather than transcription.

Ho and Varner (1976), have postulated that ABA might de-repress a regulator gene or interact with a regulator RNA or protein to inhibit the translation of a-amylase RNA. It appears that ABA inhibits the translation of specific mRNA species.

(iv) Ethylene:
Seeds have been shown to produce ethylene during germination. In castor bean, three peaks of ethylene production have been detected and those ethylene maxima coincide with the rapid growth phases of the seedlings. Non-germinating seeds, on the other hand, produce low levels of ethylene. Ethylene production pattern indicates the emergence of hypocotyl-radicle and subsequent growth of these organs.

It was noticed that any substance which broke seed dormancy also stimulated ethylene production, generally reaching the peak value within 24 hours of germination.

Germination rate of seeds is enhanced by exogenous ethylene treatment. In cocklebur promoters like gibberellin and

cytokinin may influence ethylene production by seeds, as a means to stimulate germination.

(v) Bud Dormancy:

The seeds are not the only organs which show the failure to germinate even under environmental conditions favourable for growth. The meristematic regions such as buds of woody or herbaceous plants also show dormancy and fail to grow. The winter temperatures prove to be lethal to the temperate herbaceous and woody perennials.

Most woody plants have developed for their buds a dormancy mechanism which helps them to survive winter cold. In the annual growth cycle of the temperate woody plants, the bud break usually takes place in spring.

Dormancy of buds may be defined as cessation of growth showing a rest condition. These buds in early spring, however, do not show any physiological dormancy and only the warm temperature of spring is needed to activate the buds into growth. In the early stages of growth, the buds do not have photosynthetic activity and thus depend on stored reserves.

Ultimately, the young leaves are formed in buds which attain photosynthetic capacity.

It is generally observed that the terminal growth of many woody plants of temperate zone ceases with the onset of cold weather. At some point after the cessation of shoot growth, physiological dormancy, also called rest, develops in the shoot.

This type of dormancy induced by rest condition is quite different from the dormancy under apical dominance displayed by-axillary buds which are prevented from growing by the presence of shoot tip.

If the rest condition of the shoot is largely advanced, removal of shoot tips will fail to release apical dominance and axillary bud burst does not occur. The intensity of bud rest has been shown to vary seasonally.

Hatch and Walker (1968) have shown that the rest intensity in peach increases from time when it can be first detached in September to a maximum in November, it then declines to a less pronounced rest by late December.

The rest period of several buds can be eliminated by chilling temperatures with an optimum about 5-7o C. Temperatures near 0°C seem to be too cold for physiological chilling purpose, whereas temperatures above 10°C appear to be too warm for the chilling process.

The amount of chilling required to fulfil the chilling requirement varies widely in different species which need either low or high chilling treatment.

But once the chilling requirement is satisfied, growth starts again by breaking of dormancy. Since the chilling requirement is generally met in winter with the return of warmer temperature of spring, bud burst and shoot elongation can start once again and the annual shoot growth cycle begins.

Some plants with very high chilling requirements are characterized by late growth and blooming. These plants receive part of their chilling in autumn and early winter. They have been shown to receive an additional period of chilling till

late winter and early spring, and are able to grow later in spring resulting in late blooming.

Such delayed growth in spring is possibly due to the variation in the ability of some cultivars to resume growth at lower temperatures than other species. For example Populustremeloides and Betula papyrifera are trees capable of initiating growth when daily minimum low temperatures are below 0°C, while Acerrubrum and Fraxinus nigra have much higher minimum temperature requirements.

In studies on the site of rest within the bud it has been shown that bud scales have some influence on bud dormancy but it may not be the only factor Swartz et al, have demonstrated that the removal of the bud scales may permit bud growth when rest is not strongly developed, but scale removal has little influence when the buds are in profound rest.

Thus, it can be indicated that the rest influence possibly resides in the meristematic part of the bud.

Hormonal relationships of bud dormancy in woody plants have been established whereby hormones have been shown to play a major role in regulating dormancy.

Of the different naturally-occurring plant hormones, ABA, gibberellins and cytokinins are positively implicated in the control of dormancy whereas IAA or ethylene has not been shown to participate in a direct manner. Hemberg (1949), first enunciated the hypothesis that specific inhibitory substances play an essential role in bud dormancy.

Subsequently, it was found that endogenous gibberellins and cytokinins overcome dormancy in a range of woody species and in organs such as potato tubers. It was further postulated that bud dormancy may be controlled by a balance between endogenous inhibitors such as ABA, and the growth-promoting hormones, especially the GAs.

The formation of resting buds and the onset of dormancy is promoted by short-days in many woody species, it seems reasonable to suggest that the cessation of growth and formation of resting buds are due to an inhibitory factor produced by the leaves under SD condition which is transmitted to the apex Philips and Wareing (1958), followed seasonal changes in the growth substance content of buds and leaves of Acer pseudoplatanus and noted changes in growth-promoting and growth- inhibiting activity correlated with the onset of bud dormancy in trees growing under natural conditions with shoot apices of Populus tremula cuttings, Eliasson (1969), observed a marked SD-induced reduction in GA-like activity.

A balance between promoter and inhibitor levels is an important factor in the control of bud dormancy. Exogenous applications of gibberellins and cytokinins are well known to be effective in overcoming dormancy of tree buds but the effect has not been confirmed in all plants studied so far.

On the other hand, the formation of resting buds by the application of ABA has been achieved in several woody species like Betula pubescens, Acer pseudoplatanus and Ribes nigrum.

Experiments in which ABA and GA were applied together to tree buds indicate that the growth-inhibitory action of ABA can be largely overcome by increasing concentrations of GA. Thus, experiments with externally applied ABA, GA, and kinetin give some support to the hypothesis that bud dormancy may be regulated by an interaction between these endogenous hormones.

More recently, ABA level has been measured in buds of several species from the onset of rest in autumn through the breaking of rest in late winter and early spring. In some cases, positive correlations between ABA contents and

intensity of dormancy have been established, but in others the relationship is less certain.

Based on the present state of knowledge. Powell has concluded that ABA in buds of many woody plants declines during the cold winter months, although it is not certain whether cold temperatures are responsible for this decline.

It is also well established that growth-promoting hormones tend to fall to low levels in shoots late in the growing season. He has argued then, that dormancy may be due to insufficient growth hormones and not due to an inhibitor.

But the failure of growth-promoting substances to promote growth when rest type of dormancy has already started suggests that dormancy is due to the gradual increase of some kind of an inhibitory influence, which Hormonal Regulations During Seed Dormancy:

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Seeds have been shown to produce ethylene during germination. In castor bean, three peaks of ethylene production have been detected and those ethylene maxima coincide with the rapid growth phases of the seedlings. Non-germinating seeds, on the other hand, produce low levels of ethylene. Ethylene production pattern indicates the emergence of hypocotyl-radicle and subsequent growth of these organs.

It was noticed that any substance which broke seed dormancy also stimulated ethylene production, generally reaching the peak value within 24 hours of germination.

Germination rate of seeds is enhanced by exogenous ethylene treatment. In cocklebur promoters like gibberellin and cytokinin may influence ethylene production by seeds, as a means to stimulate germination.

(v) Bud Dormancy:

The seeds are not the only organs which show the failure to germinate even under environmental conditions favourable for growth. The meristematic regions such as buds of woody or herbaceous plants also show dormancy and fail to grow. The winter temperatures prove to be lethal to the temperate herbaceous and woody perennials.

Most woody plants have developed for their buds a dormancy mechanism which helps them to survive winter cold. In the annual growth cycle of the temperate woody plants, the bud break usually takes place in spring.

Dormancy of buds may be defined as cessation of growth showing a rest condition. These buds in early spring, however, do not show any physiological dormancy and only the warm temperature of spring is needed to activate the buds into growth. In the early stages of growth, the buds do not have photosynthetic activity and thus depend on stored reserves.

Ultimately, the young leaves are formed in buds which attain photosynthetic capacity.

It is generally observed that the terminal growth of many woody plants of temperate zone ceases with the onset of cold weather. At some point after the cessation of shoot growth, physiological dormancy, also called rest, develops in the shoot.

This type of dormancy induced by rest condition is quite different from the dormancy under apical dominance displayed by-axillary buds which are prevented from growing by the presence of shoot tip.

If the rest condition of the shoot is largely advanced, removal of shoot tips will fail to release apical dominance and axillary bud burst does not occur. The intensity of bud rest has been shown to vary seasonally.

Hatch and Walker (1968) have shown that the rest intensity in peach increases from time when it can be first detached in September to a maximum in November, it then declines to a less pronounced rest by late December.

The rest period of several buds can be eliminated by chilling temperatures with an optimum about 5-7o C. Temperatures near 0°C seem to be too cold for physiological chilling purpose, whereas temperatures above 10°C appear to be too warm for the chilling process.

The amount of chilling required to fulfil the chilling requirement varies widely in different species which need either low or high chilling treatment.

But once the chilling requirement is satisfied, growth starts again by breaking of dormancy. Since the chilling requirement is generally met in winter with the return of warmer temperature of spring, bud burst and shoot elongation can start once again and the annual shoot growth cycle begins.

Some plants with very high chilling requirements are characterized by late growth and blooming. These plants receive part of their chilling in autumn and early winter. They have been shown to receive an additional period of chilling till late winter and early spring, and are able to grow later in spring resulting in late blooming.

Such delayed growth in spring is possibly due to the variation in the ability of some cultivars to resume growth at lower temperatures than other species. For example Populustremeloides and Betula papyrifera are trees capable of initiating growth when daily minimum low temperatures are below 0°C, while Acerrubrum and Fraxinus nigra have much higher minimum temperature requirements.

In studies on the site of rest within the bud it has been shown that bud scales have some influence on bud dormancy but it may not be the only factor Swartz et al, have demonstrated that the removal of the bud scales may permit bud growth when rest is not strongly developed, but scale removal has little influence when the buds are in profound rest.

Thus, it can be indicated that the rest influence possibly resides in the meristematic part of the bud.

Hormonal relationships of bud dormancy in woody plants have been established whereby hormones have been shown to play a major role in regulating dormancy.

Of the different naturally-occurring plant hormones, ABA, gibberellins and cytokinins are positively implicated in the control of dormancy whereas IAA or ethylene has not been shown to participate in a direct manner. Hemberg (1949), first enunciated the hypothesis that specific inhibitory substances play an essential role in bud dormancy.

Subsequently, it was found that endogenous gibberellins and cytokinins overcome dormancy in a range of woody species and in organs such as potato tubers. It was further postulated that bud dormancy may be controlled by a balance between endogenous inhibitors such as ABA, and the growth-promoting hormones, especially the GAs.

The formation of resting buds and the onset of dormancy is promoted by short-days in many woody species, it seems reasonable to suggest that the cessation of growth and formation of resting buds are due to an inhibitory factor produced by the leaves under SD condition which is transmitted to the apex Philips and Wareing (1958), followed seasonal changes in the growth substance content of buds and leaves of Acer pseudoplatanus and noted changes in growth-promoting and growth- inhibiting activity correlated with the onset of bud dormancy in trees growing under natural conditions with shoot apices of Populus tremula cuttings, Eliasson (1969), observed a marked SD-induced reduction in GA-like activity.

A balance between promoter and inhibitor levels is an important factor in the control of bud dormancy. Exogenous applications of gibberellins and cytokinins are well known to be effective in overcoming dormancy of tree buds but the effect has not been confirmed in all plants studied so far.

On the other hand, the formation of resting buds by the application of ABA has been achieved in several woody

species like Betula pubescens, Acer pseudoplatanus and Ribes nigrum.

Experiments in which ABA and GA were applied together to tree buds indicate that the growth-inhibitory action of ABA can be largely overcome by increasing concentrations of GA. Thus, experiments with externally applied ABA, GA, and kinetin give some support to the hypothesis that bud dormancy may be regulated by an interaction between these endogenous hormones.

More recently, ABA level has been measured in buds of several species from the onset of rest in autumn through the breaking of rest in late winter and early spring. In some cases, positive correlations between ABA contents and intensity of dormancy have been established, but in others the relationship is less certain.

Based on the present state of knowledge. Powell has concluded that ABA in buds of many woody plants declines during the cold winter months, although it is not certain whether cold temperatures are responsible for this decline.

It is also well established that growth-promoting hormones tend to fall to low levels in shoots late in the growing season. He has argued then, that dormancy may be due to insufficient growth hormones and not due to an inhibitor.

But the failure of growth-promoting substances to promote growth when rest type of dormancy has already started suggests that dormancy is due to the gradual increase of some kind of an inhibitory influence, which