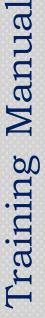
# **National Training**

# on "Seed Certification" (January 08-12, 2024)







Organized by: Government of India Ministry of Agriculture & Farmers Welfare Department of Agriculture & Farmers Welfare National Seed Research & Training Centre GT Road, Collectry Farm Varanasi- 221106 Phone: 0542-2370222, Fax: 0542-2370298 Website: www.nsrtc.nic.in NATIONAL TRAINING ON SEED CERTIFICATION (JANUARY 08-12, 2024)

**Course Director** 

Sh. Manoj Kumar, IAS Director

## **Training Coordinators**

**Sh. Javesh Kumar** Junior Seed Analyst **Dr. A. K. Verma** Senior Seed Analyst

Organized by:



Government of India Ministry of Agriculture & Farmers Welfare Department of Agriculture & Farmers Welfare

## NATIONAL SEED RESEARCH AND TRAINING CENTRE VARANASI-221106 (UTTAR PRADESH)

Tel: 0542-2370222, Fax: 0542-2370298 E-mail: <u>dir-nsrtc-up@nic.in</u> Website: www.nsrtc.nic.in

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Compiled & Edited by:

Sh. Javesh Kumar, Junior Seed Analyst Dr. A. K. Verma, Senior Seed Analyst Er. M. K. Vishwakarma, Seed Processing Engineer Dr. M. P. Yadav, Seet Technologist Mrs. Ekta Kumari, Senior Seed Analyst Sh. Anil Varma Nalla, Junior Seed Analyst

NATIONAL SEED RESEARCH AND TRAINING CENTRE VARANASI-221 106 (UTTAR PRADESH) भारत सरकार राष्ट्रीय बीज अनुसंधान एवं प्रशिक्षण केन्द्र कृषि एवं किसान कल्याण मंत्रालय कृषि एवं किसान कल्याण विभाग जी.टी. रोड, कलेक्ट्री फार्म, पोस्ट आफिस इन्डस्ट्रीयल इस्टेट, वाराणसी 221 106 (उ.प्र.)



GOVERNMENT OF INDIA NATIONAL SEED RESEARCH & TRAINING CENTRE Ministry of Agriculture & Farmers Welfare Deptt. of Agriculture & Farmers Welfare G.T. Road, Collectry Farm, P.O. Industrial Estate, Varanasi-221106 (U.P.)

## FOREWORD

Seeds are the foundation of agriculture. Technology has modernized much of farming's day-to-day operations, but without a steady supply of high-quality seed, yields and crop quality would be greatly decreased. Seed quality plays an important role in the production of agricultural and horticultural crops. Characteristics such as trueness to variety, germination percentage, purity, vigor and appearance are important to farmers planting crops. Achieving and maintaining high seed quality is the goal of every professional seed producer.

Seed Certification is a process designed to maintain and make available to the general public continuous supply of high quality seeds and propagating materials of notified kinds and varieties of crops, so grown and distributed to ensure the physical identity and genetic purity. Seed certification is a legally sanctioned system for quality control of seed multiplication and production. The end users need a wide product choice and seed industry today is set to work with a 'farmer centric' approach and is market driven. However, there is an urgent need for Seed production agencies to transform themselves in tune with the industry in terms of infrastructure, technologies, approach and the management culture to be able to survive in the competitive market and to enhance their contribution in the national endeavor of increasing food production to attain food & nutritional security.

In this context, National Seed Research and Training Centre, Varanasi has organize five days National Training on "Seed Certification" during 08<sup>th</sup> to 12<sup>th</sup> January, 2024, the training course is design to suit the need of officials from Central/State government Institution & Public sector undertaking who are engage in the field of Seed Certification.

This training manual comprises all the latest information pertaining to Seed Certification system in India. I hope this compilation will serve as a useful resource book and guide to all participants.

## National Training

## On

## Seed Certification

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## NSRTC at a glance...

**National Seed Research and Training Centre (NSRTC), Varanasi** established under Govt. of India, Ministry of Agriculture& Farmers Welfare, Department of Agriculture and Farmers Welfare, during October 2005.

The prime objective of establishment NSRTC is to have a separate National Seed Quality Control Laboratory, which is serving as **Central Seed Testing Laboratory (CSTL)** as well as to act as **Referral laboratory** for hon'ble court of the entire country.

Further, this **CSTL** has to coordinate and monitor the functioning of all the **notified State Seed Testing Laboratories** presently available in our country in order to obtain Uniformity in Seed quality Regulation at National level.

More importantly for facilitating International seed Movement, our CSTL the member laboratory of International Seed Testing Association (ISTA), ZURICH, Switzerland and expected to become accreditated Laboratory very soon and thereafter will be eligible for issuing International seed movement certificates on behalf of Government of India.

NSRTC is the National Centre for Training Human resources for the officials who are all involved in the **Seed Quality Control, Seed Law Enforcement and stake holders of Seed Industry.** 

In order to fulfill the mandate, NSRTC organize National trainings, workshops, National seed congress for the benefit of personnel involved in seed development and quality control programme and stakeholders of seed industry for updating their knowledge and skills.

The NSRTC is situated under greater periphery of the Holy city Varanasi, which is located 7 KM away from heart of city towards south – west on Varanasi - Allahabad GT road, Collectry farm, surrounded by Banaras Hindu University (6 km), Indian Institute of Vegetable Research (20kms) and well linked by Air, Train and Road.

#### **PRIME OBJECTIVES:**

- To have a separate National Seed Quality Control Laboratory, which is serving as Central Seed Testing Laboratory (CSTL).
- To act as **Referral laboratory** for hon'ble court for the entire country w.e.f 1.4.2007 onwards.
- Member laboratory of International Seed Testing Association (ISTA), Switzerland,
- Centre for testing all transgenic crop seeds etc., in future
- To organize National and International seed related conferences, symposium and trainings for the benefit of personnel who are involved in seed development and quality control programme and stakeholders of seed industry.

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• Centre for training human resource on all seed related aspects.

## VISION:

Our vision is to

- Contribute integrated approach towards quality seed availability.
- Have separate National Seed Quality Control Laboratory as CSTL.
- Maintain uniformity in seed testing and seed quality control at National level.
- Make Seed Industry in India globally competitive.

## **MISSION:**

Our mission is to lead and engage in downstream programmes on Seed Science and Quality Control to disseminate the values of seed production and availability of quality seed to the need of National and International seed community.

## STRATEGY:

NSRTC pursues its Mission and Goals through

- Integrated approach and system -based programs on seed quality control and act as Referral Lab for the hon'ble Court.
- Strengthening Seed Technological Research in seed production disciplines of major crops.
- Total seed quality management through systemic seed certification and law enforcement process.
- Interaction with stake holders of seed industry, officials of seed certification and law enforcement, seed producers and other seed organizations that share's NSRTC mission.
- Continued efforts in improving / updating knowledge and skill of human resources involved in seed certification and quality control as a training human resource on all seed related aspects
- In order to meet out these visions and mission's strategy the NSRTC is housed in a modern building with all latest infrastructural facilities, equipments and machineries, excellent conference/ seminar hall, workshop /class rooms, exclusive ISTA member laboratories, museum, well stocked library.

## Staff strength:

The Ministry of Finance sanctioned of 23 posts for National Seed Research and Training Centre, Varanasi for making the centre functional so as to meet out the mandate.

NSRTC is especially designed for continuous dissemination of knowledge of seed and thereby improve skill, competency and scientific soundness of individuals engaged in seed development programme. NSRTC regularly organizes training on various aspects of seed for the officials working in Seed Certification Agencies (25 in number), Seed Testing Laboratory (147 in number), Seed Law Enforcement Agencies, Agricultural Universities and other institutes dealing with seeds. The NSRTC, Central Seed Testing Laboratory acts as a referral lab under clause 4(1) of the Seeds Act, 1966. CSTL, NSRTC is testing more than 20,000 samples per year and performs at par with ISTA (International Seed Testing Association) with regard to seed testing net work in the country.

## National Seed Testing Laboratory as Central Seed Testing Laboratory

The testing of seed material will be flowing from different State Seed Corporations as well as Seed Producing Organizations for physical purity, seed health and at later stage genetic purity that is mostly required in referral cases. At present the mandate of Central Seed Testing Laboratory (CSTL) is to receive 5% samples from seed producing organizations all over the country. In addition, CSTL act as a Nodal centre for coordinating the activities of Seed Quality Control programmes on behalf of Government of India in accordance with the Act and Rules with the State Notified Seed Testing Laboratories.

## **Grow Out Test**

NSRTC have been allotted 10 hectares of land out of which the office premises have been constructed in about 2.5 hectares of land and remaining land have been kept reserve for organizing Grow Out Test for which Green House/Poly House and other necessary facilities have been created.

## NSRTC is geared to go Global

NSRTC is a globally competitive Institute in Seed Science and Quality control, marching ahead with:

- To promote the availability of quality seed to meet the challenges of Science based Agriculture.
- Making of promising Technologies reach the seed entrepreneurs and other stakeholders through innovative Trainings, Conferences, Workshops & Symposia.
- Establishing uniformity in Seed production & Quality Control programmes at National level.
- Innovative curriculum planning and implementation to make Seed Science & Research more vibrant and responsible to match the vision and needs of present and future.

Manoj Kumar, IAS Director, NSRTC

# Seed Legislation System in India

Dr. Rajesh Kumar ICAR-Indian Institute of Vegetable Research P.B. 01, P.O. Jakhini-Shahanshahpur, Varanasi-221 305, U. P.

#### Introduction

Seed is the fundamental and important input for a successful agriculture production. Agricultural sector is very much dependent on the timely availability and quality of seeds for a fruitful harvest. Quality seed alone is estimated to contribute 15-20% to total crop production. In India, agriculture is the main occupation, and thus provide rich opportunities for the seed market. India is one of the largest potential seed market in the world. According to a report, the Indian seeds market reached a value of US\$ 3.6 Billion in 2017, exhibiting a CAGR of around 17% during 2010-2017, which is further expected to grow at a CAGR of 14.3% during 2018-2023, reaching a value of more than US\$ 8 Billion by 2023. The Indian seed industry is the fifth largest seed market in the world, accounting for 4.4% of global seed market after the U.S. (27%), China (20%), France (8%) and Brazil (6%). In terms of global trade, India is almost self-sufficient in flower, fruits and vegetables and field crops seeds. Thus, it is essential to increase the production and distribution of quality seeds. Seed quality gets more significance in view of emerging biotic and abiotic stresses, issues related to quality and phytosanitary measures, competition in domestic/international markets and emerging food needs.

It is important to maintain its purity and quality through various stages of seed production i.e. Breeder, foundation, registered and certified seed. Measures of seed legislation with respect to quantity and quality were initiated in the country by establishment of NSC (1963) under Ministry of Agriculture (seed sector was majorly under public sector).Government of India had framed and brought out different legislations to protect the quality of seeds and planting materials - Seeds Act (1966), Seed Rules (1968), Seed (Control) Order (1983), New Policy on Seed Development (1988), Plants, Fruits & Seeds (Regulation of import into India), 1989, The PPV & FR Act (2001), Essential Commodities Act including Seeds (1955), National Seed Policy (2002) and Seed Bill (2004) to take care of seeds right from the production to marking, labeling and marketing levels to maintain the quality standards as prescribed. These laws are framed in order to make available quality seeds to a common farmer and train them to approach authority for justice.

#### Seed Legislations by Government of India

#### Seeds Act (1966)

#### The Seeds Act (1966) has a total of 25 Sections, mentioned as under:

- 1. Enacted by Parliament for the <u>whole of India</u> to regulate seeds.
- <u>Definitions</u> and seeds of food crops, oil crops, cotton seeds, seeds of cattle fodder and all types of vegetative propagating material are included (16 Clauses). <u>Clause 11 says</u> - "seed" means any of the following classes of seeds used for sowing or planting-

(i) seeds of food crops including edible oil seeds and seeds of fruits and vegetables;

(ii) cotton seeds;

(iii) seeds of cattle fodder;

And includes seedlings, and tubers, bulbs, rhizomes, roots, cuttings, all types of grafts and their vegetative propagated material, of food crops or cattle fodder.

#### Amendment of Section 2 of 1966

#### The Seeds (Amendment) ACT, 1972 [9th September, 1972]

In section 2 of the Seeds Act, 1966, in clause (11), after sub-clause (iii), the following sub-clause shall be inserted, namely:- (iv) jute seeds.

3. Constitution of a **Central Seed Committee** to advise the Central and State Governments on matters arising out of the administration of this act and carry out other functions assigned to it by the Act.

There are 7 clauses in this section -

#### Clause 2: The Committee shall consist of the following members, namely:-

- i. A Chairman to be nominated by the Central Government;
- ii. 8 persons to be nominated by the Central Government to represent such interests that Government thinks fit, of whom not less than two persons shall be representatives of growers of seed;
- iii. One person to be nominated by the Government of each of the States.
- iv. Other clauses deals with the formation of sub-committees, their tenure, making bye-laws for fixing the quorum and regulating its own procedure, etc.
- v. Establishing a Central Seed Laboratory as well as State Seed Laboratory to carry out seed analysis of notified variety. [2 clauses State Govt. may establish or declare State Seed Lab.].
  - 3. Empowerment of the Central Seed Committee to notify any variety found suitable as per the Act after notification in the Official Gazette for different states or different areas.
  - 4. Empowerment of the committee to fix the minimum limits of germination and purity of seed for a variety to be notified as well as for marking or labeling a seed lot to be sold commercially.
  - 5. Regulation of sale of seeds of notified varieties by compulsory labeling, revealing the true identity of the variety, germination as well as purity.
  - 6. Constituting a certification agency for undertaking the process of certification. The State Government or the Central Government in consultation with the State Government may establish a certification agency for the State to carry out the functions entrusted to the certification agency by or under this Act.

#### The Seeds (Amendment) ACT, 1972 [9th September, 1972]

#### Insertion of new sections 8A to 8E

After section 8 of the principal Act, the following sections shall be inserted, namely:

The Central Seed Certification Board

**8A.(1)** The Central Government shall establish a Central Seed Certification Board (hereinafter referred as Board) to advise the Central Government and the State Governments on all matters relating to certification and to co-ordinate the functioning of the agencies established under section 8.

8A. (2) The Board shall consist of the following members, namely:-

- (i) A Chairman, to be nominated by the Central Government;
- (ii) Four members, to be nominated by the GoI from the persons employed by the State Governments as Directors of Agriculture;
- (iii) Three members, to be nominated by the GoI from the persons employed by the AUs as Directors of Research;
- (iv) 13 persons, to be nominated by the GoI to represent such interests as that Government thinks fit, of whom not less than 4 persons shall be representatives of seed producers or tradesmen.

8A. (3) <u>A member of the Board shall</u>, (unless his seat becomes vacant either by resignation or otherwise,) <u>be entitled to hold office for two years from the date of his nomination:</u>

Provided that a person nominated under clause (ii) or clause (iii) of sub-section (2) shall hold office only for so long as he holds the appointment by virtue of which his nomination was made.

#### Other Committees

8B. The Board may appoint as many Committees as it deems fit consisting wholly of the members of the Board or wholly of other persons or partly of members of the Board and partly of other persons as it thinks fit to exercise such powers and perform such duties as may be delegated to them, subject to such conditions as it may think fit, by the Board.

8C. No proceeding of the Board or any Committee thereof shall become invalid merely by reason of the existence of any vacancy therein or any defect in the constitution thereof.

#### **Procedure for Board**

8D. The Board may, (subject to the previous approval of the Central Government), make bye-laws for the purpose of regulating its own procedure and the procedure of any Committee thereof and the conduct of all business to be transacted by it or such Committee.

#### Secretary and other officers

8E. The Central Government shall-

- (i) appoint a person to be the Secretary of the Board, and
- (ii) provide the Board with such technical and other staff as the Central Govt. considers necessary."

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9. Power of certification agency to recommend notification of suitable variety and grant of notification certificate provided the seed meets minimum limits of germination and purity.

Section 9 has 3 sub-sections:

#### The Seeds (Amendment) ACT, 1972 [9th September, 1972]

#### Amendment of section 9

In section 9 of the principal Act,-

 (i) in sub-section (3), for the words, brackets, letter and figure "minimum limits of germination and purity specified for that seed under clause (a) of section 6", the words " prescribed standards" shall be substituted;

(ii) to sub-section (3), the following proviso shall be added, namely:-

"Provided that such standards shall not be lower than the minimum limits of germination and purity specified for that seed under clause (a) of section 6."

10. Empowerment to the Certification agency for **revocation of certificate** if the agency is convinced that holder has obtained certificate (under Sec. 9) by misrepresentation or not complied with the conditions.

#### Opportunity of show cause is given.

11. Provision for an appeal by the holder on payment basis to express before an appellate authority. (Sec 11 has 3 sub-sections).

Any person aggrieved by a decision of a certification agency under Sec 9 or Sec 10, may appeal to authority specified by the State Govt. within thirty days from the date on which the decision is communicated to him and on payment of such fees as may be prescribed:

On receipt of an appeal under sub-section (1), the appellate authority shall, after giving the appellant an opportunity of being heard, dispose of the appeal as expeditiously as possible.

Every order of the appellate authority under this section shall be final.

- 12. Appointment of a seed analyst to undertake seed testing.
- 13. Appointment of seed inspector who is deemed to be a public servant within the meaning of section 21 of the Indian Penal Code (45 of 1860).
- 14. Empowerment of seed inspector to draw samples from any seller or a purchaser and verify the quality by sending samples to a seed analyst in the seed testing laboratory. (5 sub-sections & 5 Clauses). <u>Examine records, registers, docs... seize</u>.

Where the Seed Inspector takes any action under clause (a) of sub-section (1), he shall call at least two persons to be present at the time when such action is taken and take their signatures on a memorandum to be prepared in the prescribed form and manner.

The provisions of the Code of Criminal Procedure, 1898 (5 of 1898), shall, apply to any search or seizure under this section as they apply to any search or seizure made under the authority of a warrant issued under section 98 of the said Code.

- 15. Laying-out of procedure for seed sample collection and other rules. The section also entrust inspector with the power to break open any seed container or door of any premises where such seed may be kept for sale, under those circumstances when owner refuses to cooperate. The whole operation has to be done in presence of two witnesses with their signatures on a memorandum. (5 sub-sections).
- 16. Responsibility of Seed analyst to report the results in a specified format after analysis of the seed samples to Seed Inspector as well as the seller/ purchaser. Complainant if dissatisfied with the result can apply to the court for sending samples to Central Seed Testing Laboratory. Central seed laboratory shall thereupon send its report to the court in the prescribed format within one month from the date of receipt of the sample.

The report sent by the Central Seed Laboratory shall supersede the report given by the Seed Analyst.

- 17. Restriction on import and export of seeds of notified varieties. Any variety imported or exported should meet the minimum limits of seed germination and purity marked or labeled on the container.
- 18. The Central Govt., on recommendation of the Committee, Recognize seed certification agencies of foreign countries for the purpose of this act.
- 19. Penalty or punishment or both for those who do not comply with the provisions of the act and also prevent seed inspectors from executing his power. <u>First offence with fine which may extend to five hundred rupees</u>, and if previously convicted, imprisonment up to 6 months, or fine of Rs.1000/- or both.
- 17. Forfeiture of property (seeds) belonging to any person convicted under this act, due to contravention (breach) of the procedures under this act.
- 18. Punishment for offences committed by companies or any corporate. All who was in-charge of, when the time the offence was committed and was responsible to the company shall be deemed to be guilty of the offence and punished accordingly.
- 19. Protection of Government action taken in good faith, i.e. no prosecution or legal proceeding will lie against Government or any Government Officer for anything that is done in good faith.
- 20. Power for Central Govt. to give directions to any state govt. for smooth conduct of the act.
- 21. **Exemption** Non-application of the act to the seed exchange by the farmers without any brand name.
- 22. Power of Government to make rules to carry out various functions of Central Seed Committee, Central Seed Laboratory, Certification Agency and Seed Inspectors.

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#### The Seeds (Amendment) ACT, 1972 [9th September, 1972]

#### Amendment of section 25

In section 25 of the principal Act,-

(a) in sub-section (2), after clause (f), the following clause shall be inserted, namely:-

"(ff) the standards to which seeds should conform,";

(f) the form of application for the grant of a certificate under section 9, the particulars it may contain, the fees which should accompany it, the form of the certificate and the conditions subject to which the certificate may be granted;

(b) in sub-section (3), for the words "in two successive sessions, and if, before the expiry of the session in which it is so laid or the session immediately following", the words "in two <u>or more</u> successive sessions, and if, before the expiry of the session immediately following the session or the successive sessions aforesaid" shall be substituted.

#### Seed Rules, 1968

The rules were framed to implement various legislations given under Seeds Act, 1966. It contain**11** sections (39 rules).

#### I. Preliminary (Rules 1 to 2: Short titles & Definitions)

This section provides short title, definitions of various terminology used under the seed rule.

**II. Central Seed committee** <u>(Rules 3 to 4:</u> Functions of the Central Seed Committee & Travelling and Daily Allowances Payable to Members of the Committee and its sub- Committees)

This section describes the specific functions entrusted to the committee by the act such as recommendation for Seed Testing fee, advice on the suitability of seed testing laboratory, recommendation for the procedure and standards for seed certification and testing. Also the rules provide details of traveling and daily allowances payable to the members of the committee.

#### III. Central Seed Laboratory (Rule 5 - Functions)

This section describes the specific functions entrusted to the Central Seed Laboratory such as coordinating with State Seed Laboratories for uniformity in test results, collecting data on quality of seeds available in the market and any other function assigned to it by the Central Government.

## IV. Seed Certification Agency (Rule 6. Functions of the Certification Agency)

This section deals with the specific functions entrusted to the Certification Agency such as outlining the procedure for submission of applications, growing, harvesting and processing and storage of seeds indented for certification, maintaining a list of recognized nucleus seed breeders, inspections of seed production fields, seed processing plant and seed stores, grant of certificates.

**V. Marking or Labeling (Rules 7 to 12**: Responsibility for Marking or Labeling, Contents of the mark or label, Manner of marking or labeling the container under clause (C) of section 7 and clause (B) of section 17, Mark or Label not to contain false or misleading statement, Mark or label not to contain reference to the Act or Rules contradictory to required Particulars, and Denial of Responsibility for mark or label content prohibited)

Rules for marking or labeling of seed lots indented for certification have been provided in this section. The label should contain name of the person or agency that produced the seed and shall be responsible for the accuracy of information given in the unopened original container. The label should contain the name, the address of the person offering the sale of the seed, name of the variety, germination and purity level of the seed, net weight of the seed, date of seed testing and a statement if the seed is treated. Any transparent cover used solely for the purpose of packing during transport or delivery need not be marked or labeled.

**VI. Requirements for Certification (<u>Rule 13 to 14:</u> Requirements to be complied with by a person carrying on the Business referred to in Section 7 and Classes and sources of certified seed)** 

Three classes of certified seed have been specified in this section, viz. Foundation (progeny of breeder seed), Registered (progeny of foundation seed) and Certified (progeny of registered / foundation seed) and each class shall meet the specific standards. Certification agency has the discretion of producing certified seed from certified seed provided that it <u>does not exceed three generation</u> and the genetic purity is not significantly altered.

# VII. Certification of seeds (<u>Rules 15 to 17</u>: Application for the Grant of a Certificate, Fees and Certificate)

The detailed procedure of seed certification starting from applying for certification till the grant of certificate has been provided in this section. Application has been outlined by the certification agency containing the name and details of the applicant, the name of the seed to be certified, class and source of the seed, germination and purity and mark or label. A fee of Rs. 25 is levied for certification.

Once certified, the certification tag containing information such as name and address of the certification agency, name of variety, lot number, name and address of the producer, date of issue of its certificate and its validity, an appropriate sign, to designate certified seed. The <u>color of the tag shall be white for foundation</u>, <u>purple for registered and blue for certified seed</u>. The holder of certificate shall allow any seed inspector to enter and inspect the seeds kept for sale, registers or other documents.

#### The Seeds (Amendment) Rules, 1981 [10th June, 1981]

#### After rule 17 of the Seeds Rules, 1968, the following rule shall be inserted, namely:-

<u>17-A.</u> The Certification agency shall, before granting the certificate, ensure that the seed conforms to the standards laid down in the Manual known as "Indian Minimum Seed Certification Standards" published by the Central Seed Committee, as amended from time to time.

The amendment says certification agency shall ensure that the seed standards confirm to the minimum seed certification standards laid down in the manual known as Indian Minimum Seed Certification Standards published by the Central Seed Committee which is commonly called as Blue Book.

# VIII. Appeal (<u>Rule 18 to 19):</u> The form and manner in which and the fee on payment of which the appeal may be referred and Procedure to be followed by the Appellate Authority)

Provision for appeal has been provided by submitting a memorandum accompanied by a treasury receipt for Rs. 100. The appellate authority shall exercise all the powers which a court has, while deciding appeal under the code of civil procedure, 1908.

- <u>Rule 19:</u> Procedure to be followed by the Appellate Authority. In deciding appeals under the Act the appellate authority shall exercise all the powers which a Court has and shall follow the same procedure which a Court follows in deciding appeals from the decree or order of an original Court under the Code of Civil Procedure, 1908 (5 of 1908)
   The Seeds (Amendment) <u>Rules</u>, 1973 (30<sup>th</sup> June 1973)
- ✓ In rule 19 of the Seeds Rule, 1968 the words, 'shall exercise all the powers which a Court has and' shall be omitted.
- ✓ IX. Seed Analyst and Seed Inspectors (<u>Rule 20 23:</u> Qualifications of Seed Analyst, Duties of a Seed Analyst, Qualifications of Seed Inspectors and Duties of a Seed Inspector)
- ✓ The specific qualifications and duties of seed analyst and seed inspectors have been provided in this section. Seed analyst should possess a Master Degree in Agriculture/ Agronomy/ Botany/ Horticulture from a recognized University with at least one year experience in Seed Technology or possess a Bachelors degree in Agriculture/Botany from a recognized university with a minimum of three years experience in Seed Technology for this purpose. Seed analyst shall analyze the seed samples according to the provisions of the Act. Seed Inspector shall be a graduate in agriculture with at least one year experience in Seed Technology.

<u>**Rule 21:**</u> **Duties of a Seed Analyst. –** On receipt of a sample for analysis the Seed Analyst shall first ascertain that the mark and the seal or fastening as provided in clause (b) of sub-section (1) of section 15 are intact and shall note the condition of the seals thereon.

(2) The Seed Analyst shall analyze the samples according to the provisions of the Act and these rules.

(3) The Seed Analyst shall deliver the copy of the report of the result of the analysis to the persons specified in sub-section (1) of section 16.

(4) The Seed Analyst shall from time to time forward to the State Government the reports giving the result of analytical work done by him.

## The Seeds (Amendment) Rules, 1973 (30th June 1973)

- In rule 21 of the said rules for sub-rules (2) and (3) the following sub-rules shall be substituted, namely:
- "(2) The Seed Analyst shall analyze the samples in accordance with the procedures laid down in the Seed Testing Manual published by the Indian Council of Agricultural Research as amended from time to time."

- "(3) The Seed Analyst shall deliver in Form VII, a copy of the report of the result of analysis to the persons specified in sub-section (1) of Section 16, as soon as may be but not later than 30 days from the date of receipt of samples sent by the Seed Inspector under sub-section (2) of the Section 15".
- > Rule 23; clause h (Duties of a Seed Inspector) -
- > (h) perform such other duties as may be entrusted to him by the competent authority.
- > The Seeds (Amendment) Rules, 1973 (30th June 1973)
- In rule 23 of the said rules, in clause (h) for the words competent authority "the words" State Government shall be substituted.

#### The Seeds (Amendment) Rules, 1974 (29th April 1975)

#### After rule 23 of the said rules, the following rule shall be inserted namely:-

#### "23-A. Action to be taken by the Seed Inspector if a complaint is lodged with him:-

(1) If farmer has lodged a complaint in writing that the failure of the crop is due to the defective quality of seeds of any notified kind or variety supplied to him, the Seed Inspector shall take in his possession the marks or labels, the seed containers and a sample of unused seeds to the extent possible from the complaint for establishing the source of supply of seeds and shall investigate the causes of the failure of his crop by sending samples of the lot to the Seed Analyst for detailed analysis at the State Seed Testing Laboratory. He shall thereupon submit the report of his findings as soon as possible to the competent authority.

(2) In case, the Seed Inspector comes to the conclusion that the failure of the crop is due to the quality of seeds supplied to the farmer being less than the minimum standards notified by the Central Government, launch proceedings against the supplier for contravention of the provisions of the Act or these Rules."

#### Part X. Sealing, Fastening, Dispatch and Analysis of Samples

(<u>Rules 24 to 37</u>: Manner of taking Samples, Containers to be labeled and addressed, Manner of Packing Fastening and Sealing the Samples, Form of Order, Form of Receipt for Records, how to be sent samples to the Seed Analyst, Memorandum and Impression of seal to be sent separately, Addition of Preservatives to Samples, Nature and Quantity of the Preservative to be noted on the Label, Analysis of the Sample, Form of Notice, Form of Report, Fees and Retaining of the Sample)

The details of sampling, labeling, manner of packing and sealing the samples as well as its dispatch to the seed analyst has been provided.

- Samples of any seed shall be taken in a clean dry, moisture and leakage proof container and shall be carefully sealed.
- The label on any sample of seed sent for analysis shall bear:

a. serial number;

b. name of the sender with official designation, if any;

c. name of the person from whom the sample has been taken.

d. Date and place of taking the sample;

e. Kind or variety of the seed for analysis;

f. Nature and quantity of preservative, if any, added to the sample.

• The container of sample for analysis shall be sent to the Seed Analyst by registered post or by hand in a sealed packet enclosed together with a memorandum in Form V in an outer cover addressed to the Seed Analyst.

# It should be ensured that the sample reach the destination without any kind of damage/alterations/leakage.

- Whenever any **preservative** is added to a sample, the nature and quantity of the preservative added shall be clearly noted on the label to be affixed to the container.
- <u>Analysis of the sample</u> On receipt of the packet, it shall be opened either by the Seed Analyst or by an officer authorized in writing in that behalf by the Seed Analyst, who shall record the condition of the seal on the packet. Analysis of the sample shall be carried out at the State Seed Laboratory in accordance with the procedure laid down by the Central Government.
- **Form of notice:** In Form VI, the notice be given under clause (a) of sub section (1) of section 15 to the person from whom the Seed Inspector intends to take sample.
- **Form of report** : In Form VII, the result of the analysis be delivered.
- **Fees**: Rs. 10/- per sample of the seed analyzed.
- **Retaining of the sample:** The sample of any seed shall, under clause (c) of Sub-section (2) of section 15, be retained under a cool, dry environment to eliminate the loss of viability and in insect proof or rat proof containers (good quality of uniform shape & size). The containers shall be dusted with suitable insecticides and the storage room fumigated to avoid infestation of samples by insects.
- Part XI. Miscellaneous (<u>Rules 38 to 39:</u> Records and Form of Memorandum)
- The need to maintain stock record of seeds and record of the sale of seed have been provided in this section.
- **Records**: A person carrying on the business referred to in section 7 shall maintain the following records, namely:
- a. stock record of seed;
- b. record of the sale of seeds;
- **Form of Memorandum**: The memorandum to be prepared under sub-section (4) of section 14 shall be in Form VIII.

#### Seeds (Control) Order, 1983 [30th December, 1983]

In exercise of the powers conferred by section 3 of the <u>Essential Commodities Act</u>, <u>1955</u>, the Central Government hereby makes the **Seeds (Control) Order**.

<u>**Dealer to obtain license:-**</u>No person shall carry on the business of selling, exporting or importing seeds at any place except under and in accordance with the terms and conditions of license granted to him under this order.

**<u>Application for license:</u>**Every person desiring to obtain a license for selling, exporting or importing seeds shall make an application in duplicate in Form 'A' together with a fee of rupees fifty for license to licensing authority.

#### Grant and refusal of license:-

(1) The licensing authority may, after making such enquiry as it thinks fit, grant a license in Form 'B' to any person who applies for it under clause 4:

A license shall not be issued to a person-

(a) whose earlier license granted under this Order is under suspension, during the period of such suspension;

(b) whose earlier license granted under this Order has been cancelled, within a period of one year from the date of such cancellation.

(c) who has been convicted under the Essential Commodities Act, 1955 or any order issued thereunder within three years preceding the date of application.

(2) When the licensing authority refuses to grant license to a person who applies for it under clause 4, he shall record his reasons for doing so.

**Period of validity of license:-**Valid for three years from the date of its issue.

**Renewal of license:**-An application for renewal in duplicate, in Form C is made before the expiry of the license along with a fee of Rs.20/-

If any application for renewal is not made before the expiry of the license, but is made within one month from the date of expiry of the license, the license may be renewed on payment of additional fee of rupees twenty five, in addition to the fee for renewal of license.

- ✓ The seed dealer has to essentially display the stock position (opening and closing) on daily basis along with a list indicating prices or rates of different seeds.
- ✓ A cash or credit memorandum has to be given by the dealer to purchaser of seeds, compulsorily.

**Appointment of licensing authority -** The State Government is empowered with appointing a licensing authority, inspectors and mode of action for supply regulation.

**Appointment of Inspectors -** The State Government may appoint inspectors and define the area within which each such Inspector shall exercise his jurisdiction.

#### Inspection and punishment

 An Inspector may ask any dealer to give any information with respect to purchase, storage and sale of seeds; enter upon and search any premises where any seed is stored; draw samples of seeds meant for sale, export and seeds imported for confirmation of Quality Standards, seize or detain any seed in respect of which he has reason to believe that a contravention of this Order has been committed or is being committed and seize any books of accounts or document relating to any seed.

- The Inspector shall give a receipt, in respect of books of accounts or documents seized and return the seized books of accounts or documents after the purpose is solved.
- The provision of section 100 of the Code of Criminal Procedure, 1973 relating to search and seizure shall apply to searches and seizures.
- The Inspector shall report the fact of seizure to a Magistrate where-upon the provisions of sections 457 and 458 of the Code of Criminal Procedure, 1973 shall apply.
- Every person, if so required by an Inspector, shall be bound to offer all necessary facilities to him for the purpose of enabling him to exercise his power under this clause.
   Time limit for analysis Under this order the time period for completion of seed analysis in case of any doubt about quality is 60 days compared to 30 days under Seed Rules.

**Suspension/Cancellation of license:-**The licensing authority may, after giving the holder of the license an opportunity of being heard, suspend or cancel the license on the following grounds, namely:-

- (a) that the license had been obtained by misrepresentation as to a material, or
- (b) that any of the provisions of this Order or any condition of license has been contravened.

**Appeal -** Any person aggrieved by an order, may within sixty days (along with Rs. 20/-), appeal to authority as the State Government may specify, and the decision of such authority shall be final.

**Amendment of license -** Upon a written request along with Rs. 10/-, the licensing authority may amend the license of the dealer.

**Maintenance of records and submission of returns, etc -** Every dealer shall maintain books, accounts and records and submit monthly return (by 5<sup>th</sup> of every month) relating to his business as may be directed by the State Government.

#### New Policy on Seed Development, 1988

The policy was formulated to provide Indian farmers with access to the best available seeds and planting materials of domestic as well as imported.

- The policy permits the import of selected seeds under Open General License (OGL), to make available high quality seeds to farmers to maximize yield, increase productivity thereby farm income.
- The policy allows import under OGL of items such as seeds of oilseed crops, pulses, coarse grains, vegetables, flowers, ornamental plants, tubers, bulbs, cuttings and saplings of flowers.
- While the import of horticultural crops including flowers need recommendation from Directors of Horticulture, import of crop seeds require permission from ICAR.
- ICAR will direct MLT in various agro-climatic conditions at least for one season.
- Evaluation of important traits such as yield, pest resistance etc. needs to be done within 3 months of harvest after which importer shall apply to the DAC for permit.

• Within a month, DAC will process it and thereafter controller of Imports and Exports will issue a license.

The policy was immediately followed by an order by Government of India (Plants, Fruits and Seeds Order) for the purpose to regulate the import of agricultural items into India.

#### Plants, Fruits and Seeds Order

#### (Regulation of Import into India order) 1989

- Post entry quarantine facilities shall be established which shall be permitted to be released by Designated Inspection Authority.
- □ Import of any form of seed for consumption or sowing should carry a permit issued by the competent authority, and the import should be only through specified customs stations.
- □ The consignment shall be inspected by the Plant Protection Advisor.
- Amendments have been made for the above order during 1998, 2000 and 2001.
- With the liberalized trade in agriculture, as consequence to WTO agreements, Government thought of providing new legislative provisions under the new order.

## Plant Quarantine (Regulation of import into India) Order, 2003

- > The Order has now <u>replaced</u> the Plants, Fruits and Seeds order, 1989.
- The order has widened the scope of plant quarantine activities and has made pest risk analysis compulsory for imports
- The order includes provision for regulating the import of soil, moss, germplasm and GMO's for research, insects, microbial cultures and bio-control agents, timber and wooden logs
- The order prohibits import of commodities contaminated with weeds, alien species, and packaging material of plant origin unless the material has been treated.
- Agricultural imports are thus classified as: prohibited plant species, restricted species where import permitted only by authorized institutions and declarations and plant material imported for consumption or industrial processing permitted with phytosanitary certificate (to prevent spread of noxious pests).
- Pest risk analysis during post entry quarantine is compulsory.
- Import of germplasm has to be permitted by NBPGR and any other biological materials such as soil, microbes, moss etc. has to be permitted by Plant Protection Advisor.
- Notified entry points for import have been increased compared to PFS Order, 1989 (26 quarantine and fumigation stations located at 10 airports, 9 seaports and 7 land frontiers)
- Strengthening Plant Quarantine facilities, opening new quarantine stations, establishing advanced molecular diagnostic facilities for rapid pathogen detection, setting up of National Pest Risk Analysis unit are other important features of the Order.

#### Protection of Plant Varieties and Farmers' Rights Act, 2001

• Global realization on the role of plant genetic resources in development of superior crop varieties and use of many traditionally grown plants in development of medicines and various

industrial applications raised concerns for Conservation of Biological Diversity (CBD) which came into force in the year 1993.

- Government of India felt the need to provide protection to plant varieties which have tremendous commercial value after India became signatory to the Trade Related Aspects of Intellectual Property Rights Agreement (TRIPS) in the year 1994.
- The TRIPS agreement required the member countries to provide for protection of plant varieties either by a patent or by an effective *sui generis system or by any combination there of. The sui generis* system for protection of plant varieties was developed by India integrating the rights of breeders, farmers, and village communities. **The Protection of Plant Varieties and Farmers Right Act was thus formulated in the year 2001**.
- The PPV&FR Act covers all categories of plants except microorganisms.
- Crops important for India in the world trade, species of Indian origin, crops where India could benefit from introduction of new germplasm are the priorities.
- The act is unique in the world with inclusion of rights of farmers, breeders, and researchers.
- A variety can be registered for protection if it satisfies the criteria of Novelty, Distinctness, Uniformity and Stability (NDUS).
- Farmers can save, re-sow, exchange, share and sell farm produce of any protected variety except its commercial marketing with brand name.

## National Seed Policy, 2002

National Seed Policy was formulated in 2002 to raise India's share in the global seed trade by facilitating advanced scientific aspects such as biotechnology to farmers and in March 2002, first transgenic Bt cotton was approved for commercial cultivation in India.

- The policy encourages private sector participation in research and development of new plant varieties.
- The rights empowered to various bodies for regulating the quality of seeds produced, distributed and for providing variety protection as per the Seeds Act, 1966 and PPV & FR Act, 2001 <u>have been retained in the policy</u>.
- Promotion of seed village scheme to increase the production and make available the seeds in time as well as upgrading the quality of farmers' saved seeds.
- Establishment of a <u>National Seed Board</u> in place of Central Seed Committee and Central Seed Certification Board to undertake seed certification and advising Government on all matters related to seed planning and development. NSB will serve as the apex body in the seed sector.
- Setting up of National Seed Research and Training Centre (NSRTC, 2005) to impart training in seed technology.
- The Central Seed Testing Laboratory will be established at the National Seed Research and Training Center to <u>perform referral</u> and other functions as required under the Seeds Act.
- Development of a National Seed Grid to provide information on availability of different varieties of seeds with production details. Both public and private sector will be encouraged to join the grid for a clear assessment of demand and supply of seeds.

- All genetically engineered crops/varieties will be tested for environment and bio-safety before their commercial release, as per the regulations and guidelines of the Environment Protection Act (EPA), 1986.
- All seeds imported into the country will be required to be accompanied by a certificate from the Competent Authority of the exporting country regarding their transgenic character or otherwise.
- All importers will make available a small sample of the imported seed to the Gene Bank maintained by NBPGR.
- Incentives will be provided to the domestic seed industry to enable it to produce seeds of high yielding varieties and hybrid seeds at a faster pace to meet the challenges of domestic requirements.
- > The Department of Agriculture & Cooperation (DAC) will supervise the overall implementation and monitoring of the National Seeds Policy.

Few of Policy's other recommendations have been addressed in PPV &FR, Act, 2001 also. Major ones are maintenance of a National Register on seeds of varieties, establishing a national gene fund, disclosure of the variety's expected performance and provision for farmer to claim compensation in case of crop failure.

Further, aims of National Seed Policy such as development of infrastructure, ensuring supply of good quality seeds and facilitating the International seed trade are sought to be addressed through the proposed Seeds Bill, 2004.

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## Seed Sampling: Principles & Procedures

Sandeep Kumar Lal Principal; Scientist and Incharge, Seed Testing Laboratory Division of Seed Science and Technology ICAR- Indian Agricultural Research Institute, New Delhi Email: skl\_nsp@yahoo.com

Sampling involves the selection from the total population of a subset of individuals upon which measurements will be made; the measurements made on this subset (or sample) will then be used to estimate the properties (or parameters) of the total population. Only a small portion (quantity) of a seed lot can be examined in a laboratory. Therefore, it is important that this small portion is representative of the seed. Hence, drawing of representative sample correctly is fundamental in order to obtain uniform, accurate and reproducible results. The reliability of the interface made about the quality of the seed lot depends primarily on two components: the accuracy with which the sample represents the lot and the accuracy and precision of the laboratory test. It is observed in many cases that the variations in test results are due to the variation in the sampling. Hence, seed sampling is one of the basic components responsible for the accurate seed testing results. Therefore, utmost care is required for drawing the samples. No matter how accurately the laboratory tests are done, the results can only show the quality of the sample submitted for analysis; consequently the sample should accurately represent the composition of the seed lot.

In his foreword to the ISTA Handbook on Seed Sampling (1986), late Dr Arne Wold, Former Chairman of Bulking and Sampling Committee of ISTA wrote "Sampling of seeds is an important part of seed quality control. Correct sampling is a pre-requisite for the reliable estimation of the quality of a seed lot. Accurate description and detailed information of the sampling procedures are therefore necessary. Uniformity in sampling seed lots as well as drawing working samples is as important as uniformity in test methods in order to obtain accurate and reproducible results, Incorrect sampling may lead to misleading test results, discarding seed lots of high quality or to the approval of seed lots of low quality, which may reduce crop yield or even result in complete failure".

Seed sampling refers to the selection of a small portion of seed from a larger amount. It is aimed at obtaining a sample of a size suitable for tests, in which the probability of a constituent being present is determined only by its level of occurrence in the seed lot. While selecting samples, equal amounts should be drawn from different parts of the seed lot – from the top, middle and bottom of the lot. In general, seed lots are either in bags, in bulk or in stream. The amount or size of a seed lot determines how much sampling is required. This is called the sampling intensity. Seed Testing Laboratory personnel are not necessarily engaged in the sampling of seeds. But, nevertheless they should be well acquainted with the principles of seed sampling and should also be able to guide properly the persons engaged in this job.

## Objectives

- 1. Sampling is done to get a uniform and representative sample from a seed lot, to minimize the errors during seed testing. The size of the submitted sample required for testing is small as compared to the size of the lot. Therefore, care must be taken to ensure that the submitted sample represents the lot of the seed to be tested.
- 2. Hence it is essential that the samples be prepared in accordance to ISTA rules to ensure that the small sized sample should represent truly and in the same proportion all constituents of seed lot.

## Definitions

#### Seed lot

A seed lot is a specified quantity of seed that is physically and uniquely identifiable.

## Primary sample

A primary sample is a portion taken from the seed lot during one single sampling action.

## Composite sample

The composite sample is formed by combining and mixing all the primary samples taken from the seed lot.

#### Sub-sample

A sub-sample is a portion of a sample obtained by reducing a sample.

#### Submitted sample

A submitted sample is a sample that is to be submitted to the testing laboratory and may comprise either the whole of the composite sample or a sub-sample thereof. The submitted sample may be divided into sub-samples packed in different material meeting conditions for specific tests (e.g. moisture or health).

#### Duplicate sample

A duplicate sample is another sample obtained for submission from the same composite sample and marked "Duplicate sample".

## Working sample

The working sample is the whole of the submitted sample or a sub-sample thereof, on which one of the quality tests described in these ISTA Rules is made and must be at least the weight prescribed by the ISTA Rules for the particular test.

#### Sealed

Sealed means that a container in which seed is held and closed in such a way, that it cannot be opened to gain access to the seed and closed again, without either destroying the seal or leaving evidence of tampering. This definition refers to the sealing of seed lots, as well as of seed samples.

#### Self-sealing containers

The 'valve-pack' bag is a specific type of self-sealing container. It is filled through a sleeve-shaped valve which is automatically closed by the completion of filling the bag.

## Marked/labeled

A container of a seed lot can be considered as marked or labeled when there is a unique identification mark on the container, which defines the seed lot to which the container belongs. All containers of a seed lot must be marked with the same unique seed lot designation (numbers, characters or combination of both). Making of samples and sub-samples must ensure that there is always an unambiguous link between the seed lot and the samples and sub-samples.

## General principles of seed sampling:

A composite sample is obtained from the seed lot by taking primary samples from different positions in the whole seed lot and combining them. From this composite sample, sub-samples are obtained by sample reduction procedures at one or more stages forming the submitted sample and finally the working samples for testing. Sampling and sample reduction must be performed using appropriate techniques and equipment that is clean and in good condition.

- 1. Sampling should be carried out only by persons trained and experienced in seed sampling and employed by the official organizations.
- 2. The seed lot shall be so arranged that each individual container or part of the lot is conveniently accessible. Upon request by the sampler, the owner shall provide full information regarding the bulking and mixing of the lot. When there is definite evidence of heterogeneity, sampling should be reduced. In case of doubt, heterogeneity can be tested.
- 3. The size of the seed lot should also not exceed maximum seed lot size limits as prescribed in the rules, subject to a tolerance of 5%.
- 4. When sampling is being done by hand, great care should be taken to keep the fingers tightly closed around the seeds so that none may escape. Seed sampler may request that bags be emptied or partially emptied to facilitate sampling. The bags may then be refilled. This may be necessary, since it is impossible to obtain sample deeper than 400 mm that is, from the lower layers in bags and bins.
- 5. Other things being equal, a large sample is more representative of a lot than is a small sample. Moreover, if there is a choice as to whether to reduce a sample before sending it to the laboratory, the larger quantity should be submitted.
- 6. The sampler should determine that all seed bags sampled are identified as belonging to a single lot, either by a label or stencil mark on the bag.
- 7. The sampler must sample the minimum requisite number of bags from the seed lot. The sampling intensity must not be less than that prescribed below:

Weight of	Weight of lot (kg or	Number of primary samples			
individual	number of container)				
container in the					
seed lot					
>100 kg	Up to 500 kg	at least 5			
	501 - 3000 kg	1 for each 300 kg , but not less than 5			
	3,001 -20,000 kg	1 for each 500 kg , but not less than 10			
	20,001 kg and more	1 for each 700 kg , but not less than 40			
Note:					
Applicable for cont	tainers of more than 100 kg, o	or from streams of seed entering containers			
When sampling a se	eed lot of upto 15 containers,	regardless of their size, the same number of			
	primary samples shall be tak	ken from each container			
15 - 100 kg	1 - 4 containers	3 from each container			
Inclusive	5 - 8 containers	2 from each container			
	9 - 15 containers	1 from each container			
	16 - 30 containers	15 from the seed lot			
	31 - 59 containers	20 from the seed lot			
	60 or more containers	30 from the seed lot			
	• No	ite:			
• For seed pellets	, seed granules, seed mats an	nd tapes, small packets and reels, containers			
of less than 30,000 seed units must be combined to sample units that not exceeding					

 Table 1: Sampling intensity

• For seed pellets, seed granules, seed mats and tapes, small packets and reels, containers of less than 30,000 seed units must be combined to sample units that not exceeding 20,00,000 seeds. The sampling units shall be regarded as containers.

• For containers holding less than 15 kg of seed, containers must be combined into sampling units not exceeding 100 kg (20 Containers of 5 kg, 33 containers of 3 kg or 100 containers of 1 kg). The sampling units shall be regarded as containers.

When sampling a seed lot of upto15 containers, regardless or their size, the same number of primary samples shall be taken from each container.

- 8. Care must be exercised in reducing composite samples. Careless splitting of the sample cannot be expected to produce two similar portions.
- 9. Any seed known to have been treated with a poisonous fungicide should be identified so that the person who subsequently may handle the sample will be informed of the potential hazard.
- 10. While taking samples from machine sewed cotton bags, a few stitches at one of the top corners can be loosen broken and then this break can be closed with hand stapling device after the contents of the bag have been sampled or a self adhesive label shall be affixed to ensure proper sealing and to avoid a tampering.

- 11. The weight of the sample drawn should not be less than the weight of the submitted sample as prescribed in the ISTA rules.
- 12. Under seed law enforcement programme, only trained and experienced officials are authorized to undertake sampling and he has to give notice to such intention to the person from whom he intends to take sample. Three representative samples should be taken in the prescribed manner, which should be marked & sealed.
- One sample to be delivered to the person from whom it has been taken
- Second to be sent for analysis to the Seed Analyst of the area.
- Third to be retained for any legal proceedings.
- At least two persons should be present and obtain the signature of both the witnesses on form VIII of the Seed Rules.
- Sampler must verify the information provided on the label as per the requirements of the Seed Act.

Following information should be checked on label

- i. Kind
- ii. Variety
- iii. Lot Number
- iv. Date of Test
- v. Seller's name & address
- 13. In case of certified lots, sampler should check the following information on seed certification tag:
- Name & Address of certification agency
- Kind & Variety
- Lot No.
- Name & Address of certified seed producer
- Date of issue of the certificate & its validity
- Class & Designation of seed
- Period during which the seed shall be used for sowing
- 14. The seed lot should be so arranged that each individual or part of the lot is conveniently accessible.

#### Procedures for sampling a seed lot

#### Preparation of a seed lot and conditions for sampling

At the time of sampling, the seed lot shall be as uniform as practicable. If there is documentary or other evidence of heterogeneity, or the seed lot is found to be obviously heterogeneous, sampling must be refused/ stopped. In cases of doubt, heterogeneity can be determined. Seed may be sampled in containers or while entering the containers. The containers must be fit for purpose, e.g. must not damage the seed, and must be clean to avoid cross contamination. The containers must be labeled or marked before or just after sampling is

completed. The seed lot should be so arranged that each individual or part of the lot is conveniently accessible.

#### Obtaining primary samples

- The primary samples are drawn with the aid of suitable seed triers / or by hand in case of chaffy / non-free flowing seeds
- When defining the number and/or the size of primary samples, the seed sampler needs to ensure (besides meeting the minimum sampling intensity) that the minimum amount of seed required for the requested test(s) is sent to the testing laboratory and enough seed remains available for obtaining duplicate samples, if requested.
- Primary samples of approximately equal size shall be taken from a seed lot, irrespective of where in the lot or container the primary sample is taken.
- When the seed lot is in bags/ containers, the containers to be sampled shall be selected at random or according to a systematic plan throughout the seed lot. Primary samples shall be drawn from the top, middle and bottom of containers, but not necessarily from more than one position in any container, unless so specified.
- Closed paper bags may also be sampled in this manner. However, the holes in the paper bags should be closed with self-adhesive tape, duly signed by the sampler
- When the seed is in bulk or in large containers, the primary samples shall be drawn from random positions and depths with the aid of bin sampler.
- In case of chaffy seeds that have not been rendered free flowing, the primary samples are drawn by hand.
- When seed is to be packed in small or moisture-proof containers, (e.g. tins, or plastic bags), it should be sampled, if possible, either before or during the filling of the containers. When this has not been done, a sufficient number of containers shall be opened or pierced for abstraction of primary samples. The sampled containers shall then be closed or the containers transferred to new containers.
- Seeds are also sampled as it enters the containers, i.e. at the time processed seeds are being put into the containers, this can be done with the help of an automatic device or manually. A uniform quantity of seeds may be taken from the seed stream at specified intervals.
- Sampling seed lots of seed tapes and seed mats should be done by taking packets or pieces of tape or mat.
- The instruments being used must neither damage the seed nor select according to seed size, shape, density, chaffiness or any other quality trait. All sampling apparatus must be clean before use to prevent cross contaminations. Triers must be long enough so that the opening at the tip reaches at least half of the diameter of the container. When the container is not accessible from opposite sides, the trier must be long enough to reach the opposite side.

Sampling of seed lots may be done by one of the methods listed below.

a. **Automatic sampling from a seed stream**: Seed may be sampled by automatic sampling devices, provided that the instrument uniformly samples the cross section of the seed stream and the material entering the instrument does not bounce out again. It may be operated either

under manual or automatic control. However, the intervals between taking primary samples should be constant.

- b. **Manual sampling from a seed stream**: Seed streams may also be sampled by using manual instruments when fulfilling the requirements listed under a sampling stick.
- c. **Sampling stick**: Sampling stick (e.g. stick trier, sleeve type trier, spiral trier) consists of two parts, one of which fits loosely inside the other, but tightly enough so that seed or impurities do not slip between them. The outer part has a solid pointed end. Both parts have slots in their walls so that the cavity of the inner part can be opened and closed by moving the two parts against each other by either a twisting or a push-pull motion. The sampling stick may be used horizontally, diagonally or vertically. The spiral trier has slots in a spiral arrangement for their subsequent opening from the tip to handle and may only be used of a size smaller than *Triticum aestivum*. The sampling stick is inserted in the closed position into the container, gently pushing it so that the point reaches the required position. Further, the sampling stick is opened and slightly agitated to allow it to fill completely, followed by closing gently, withdrawing and emptying the primary sample into a container. Care should be exercised inclosing the sampling stick so that seeds are not damaged.
- d. **Nobble trier**: The Nobbe trier (dynamic spear) is a pointed tube with an opening near the pointed end, seed passes through the tube and is collected in a container. The minimum internal diameter of the Nobbe trier should be wide enough to allow the smooth and free flow of seed and contaminants through the trier. It is inserted at an angle of about 30° to the horizontal plane with the opening facing down and pushed until it reaches the required position and revolve it through 180°. Later, it is withdrawn with decreasing speed from the container, gently agitating the trier to help maintain an even flow of seed, and collect the seed sample coming in a suitable container.
- e. **Cargo sampler**: The cargo sampler (bulk sampler) consists of a special type of chamber that is fixed to a shaft. The lower part of the chamber is cone-shaped with a pointed end. To reach a greater depth, the shaft may be lengthened by screwing on successive extensions. There is a closing system in the chamber that may be a collar on the outside of the instrument, a wing connected to a door or a valve with a spring. Some cargo samplers can be closed before they are drawn back from the sampling position; others cannot be closed, so that the filled chamber is open during withdrawal.

For all species, the minimum inside diameter can be about 35 mm and the depth 75 mm. It is inserted in the closed position into the container and gently pushed vertically into the seed so that the point reaches the required *position*, pull the cargo sampler back about 10 cm or turn it (depending on the closing system), agitate it slightly to allow it to fill completely, gently close if possible and withdraw it and empty the primary sample into a container. Care should be exercised in closing the cargo sampler, so that the seeds are not damaged.

f. **Sampling by hand**: This method can be used for all species and may be the most suitable method for seed that may be damaged by the use of triers, seeds with wings, seeds with low moisture content, seed tapes and seed mats. For hand sampling seed in containers, all positions inside the containers must be accessible. Containers with layers which are not accessible from the regular opening may have to be cut open, sampled and repackaged. Containers may also be partially or completely emptied during the sampling process to gain

access to all positions in the containers. For sampling by hand, clean the hand and roll the sleeve up if necessary, insert the open hand into the container to the required position, close and withdraw the hand, taking great care that the fingers remain tightly closed about the seeds so none may escape, and empty the hand into a receiving pan.

#### Obtaining the composite sample

The primary samples are compared with each other during sampling. If they appear to be uniform, they are combined to form the composite sample, If not, the sampling procedure must be stopped. When primary samples are collected directly into one container, the content of this container may be regarded as the composite sample only if it appears uniform. If not, it must not be used for obtaining a submitted sample.

#### Obtaining the submitted sample:

The submitted sample of requisite weight or more may be obtained from the composite sample, either by repeated halving or by abstracting and subsequently combining small portions to an appropriate size. Obtaining subsamples such as seed moisture testing must be carried out in such a way that changes in moisture content are minimal. The composite sample can be submitted to the seed testing laboratory if it is of appropriate size for the tests to be conducted, or if it is difficult to mix and reduce the composite sample properly under warehouse conditions.

#### Obtaining the submitted sample for determination of moisture content:

Obtaining submitted samples of the required size for moisture testing must be carried out in such a way that changes in moisture content are minimal. Samples must be taken in the following way from the composite sample: first, mix the composite sample by either stirring it or by passing it through a mechanical divider and combining preferably once but not more than three times. Then, take a minimum of three subsamples from different positions and combine them to create the submitted sample for moisture testing.

#### **Obtaining duplicate samples:**

Duplicate samples, which were requested no later than at the time of sampling, must be prepared in the same way as the submitted sample.

#### Dispatch of the submitted sample

- The submitted sample should be sealed and marked with the same identification as the seed lot, in such a way that it establishes connection between the seed lot and the sample. The label should contain all the necessary details; such as variety, class of seed, quantity in the lot, to whom it belongs, name of the producer, seed treatment, date of harvesting and threshing, if known, sampled by, date of sampling and kind of tests required.
- For an ISTA International Seed Lot Certificate, the sample must be sealed. The additional information required as well as the name of any chemical treatment applied must be provided.
- After marking, samples should be packed so as to prevent damage during transit. For germination tests, it should be packed preferably in the cloth bags. Submitted samples for

germination tests, viability tests and health tests may only be packed in moisture proof containers if suitable storage conditions can be assured.

- For determination of seed moisture content, it should be packed separately in moisture proof containers from which as much air as possible has been excluded.
- Submitted samples shall be dispatched by the sampler to the seed testing laboratory without delay.

#### Procedure for obtaining the working sample

#### Minimum size of working sample

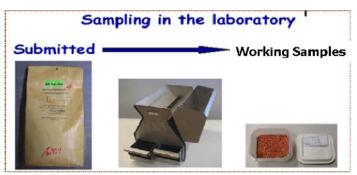
Minimum sizes of working samples are prescribed in the ISTA rules for each test. The working sample weights for purity analyses are calculated to contain at least 2500 seeds. These weights are recommended for normal use in purity test. The sample weights, for counts of other species are 10 times the weights recommended for purity analysis in column 4, subject to a maximum of 1000g. Working sample for all coated seeds except those defined as treated seeds must contain at least the number of seeds, pellets, granules as prescribed in the ISTA rules. If smaller sample is used, the actual number of pellets, seeds or granules in the sample must be reported.

#### Sample reduction methods

- If the seed sample needs to be reduced to a size equal to or greater than the size prescribed, the seed sample shall first be thoroughly mixed. The submitted/working sample shall then be obtained either by repeated halving or by abstracting and subsequently combining small random portions. One, two or more of these methods may be used in one sample reduction procedure. While using one of the dividers described for seed pellets the distance of fall must not exceed 250 mm.
- Only the spoon methods and the hand halving method may be used in the laboratory to obtain working samples for seed health testing where other samples or equipment may be contaminated by spores or other propagation material.
- For seed tapes and mats, take pieces of tape or mat at random to provide sufficient seeds for the test.
- After obtaining a working sample or half-working sample, the reminder shall be re-mixed before a second working sample or half-working sample in obtained.
- Sub-samples for moisture content determination may be taken in the following way; before taking the sub-sample, mix the sample by either stirring the sample in its container with a spoon or place the opening of the original container against the opening of a similar container and pour the seed back and forth between the two containers. Take a minimum of 3 sub samples with spoon from different positions and combine them to form a sub-sample of the required size. The seed may not be exposed to the air during sample reduction for more than 30 seconds.

## Methods for obtaining working samples

• The seed samples received in the Seed Testing Laboratory (Submitted sample) are required to be reduced to obtain working samples for carrying out various tests.

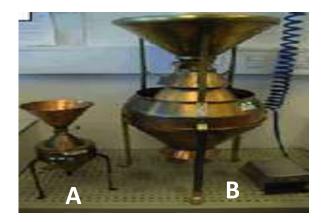


#### Mechanical divider method

This method is suitable for all kinds of seeds except some very chaffy seeds. The apparatus divides a sample passed through it into two or more through it into two or more approximately equal parts. The submitted sample can be mixed by passing it through the divider, recombining the parts and passing the whole sample through a second time, and similarly, a third time if necessary. The sample is reduced by passing the seed through repeatedly and removing parts on each occasion. This process of reduction is continued until a working sample of approximately, but not less than, the required size is obtained.

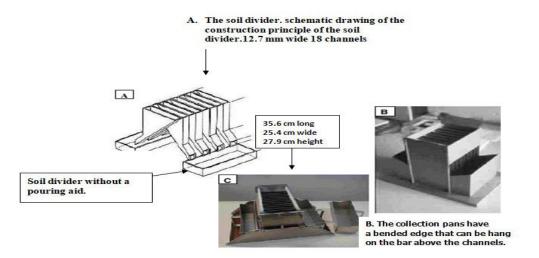
#### a. Conical divider.

The conical divider (Boerner type) consists of a hopper, cone, and series of baffles directing the seed into two spouts. The baffles form alternate channels and spaces of equal width. They are arranged in a circle and are directed inward and downward, the channels leading to one spout and the spaces to an opposite spout. A valve or gate at the base of the hopper retains the seed. When the valve is opened the seed falls by gravity over the cone where it is evenly distributed to the channels and spaces, then passes through the spouts into the seed pans. Dividers with more than 18 channels have been found to be suitable. Channels must be wide enough to allow the smooth free flow of seed and contaminants. The more channels and spaces, the better the accuracy. Typical conical dividers have about 15 channels and spaces.



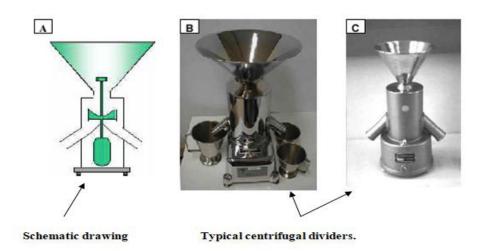
i. Small divider: 40.64 cm high and 15.24 cm in diameter, designed for small free-flowing seeds there are 22 channels and 22 spaces, each 7.9 mm wide.

- ii. Large divider: 81.28 cm high and 36.83 cm in diameter, designed for large seeds and grains, there are 19 channels and 19 spaces, each 25.4 mm wide.
  - **b.** Soil divider (Riffle divider): It is simpler divider, built on the same principle as the conical divider. However, the channels are here arranged in a straight row instead of a circle as in the conical divider. It consists of a hopper with 18 attached channels or ducts alternately leading to opposite sides, a frame to hold the hopper, two receiving pans and a pouring pan. The channels must be wide enough to allow the smooth free flow of seed and contaminants. The more channels, the better the accuracy. A minimum of 10 channels is required. While using the divider, the seed is placed evenly into a pouring pan and then poured in the hopper at approximately equal rates along the entire length. The seed passes through the channels and is collected in two receiving pans.



#### c. Centrifugal divider:

The centrifugal divider (Gamete type) makes use of centrifugal force to mix and scatter the seeds over the dividing surface. The seed flows downward through a hopper onto a shallow rubbercup or spinner. Upon rotation of the spinner by an electric motor, the seeds are thrown out by centrifugal force and fall downward. The circle or area where the seeds fall is equally divided into two parts by a stationary baffle so that approximately half the seeds fall in equally divided into two parts by a stationary baffle so that approximately half the seeds fall in one spout and half in the other spout. The centrifugal divider tends to give variable results, unless the spinner is operated after having poured the seed centrally into the hopper.



#### d. Rotary divider:

The rotary divider comprises a rotating crown base unit usually with 6 to 32 attached subsample containers, a vibration chute and a hopper. The seed is poured into the hopper and the rotary divider is switched on so that the crown/ base unit with the containers rotates with approx. 100 rpm and the vibration chute starts to feed the seed into the inlet cylinder of the rotating crown. The longer duration of the dividing operation, the better the accuracy.Thefeeding rate and therefore the duration of the dividing operation can be adjusted by the distance between the funnel of the hopper and the chute and the vibration intensity of the chute.

#### There are two principles:

- (i) The inlet cylinder feeds the seed centrally onto a distributor within the rotating crown distributing the seed to all containers simultaneously.
- (ii) The inlet cylinder feeds the seed de-centrally into the inlets of the containers rotating underneath the inlet cylinder so that the seed stream is subdivided into a lot of subsamples.

For this type of divider, mixing and dividing takes place in one operation.

#### e. Variable sample divider:

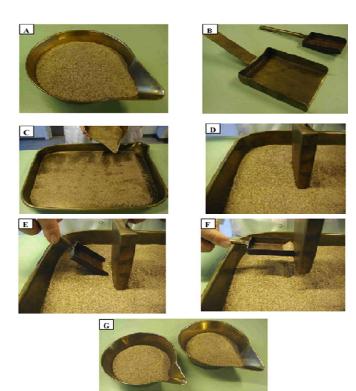
The variable sample divider consists of a pouring hopper and a rotating tube underneath that rotates with about 40 rpm. The tube distributes the seed stream from the pouring hopper onto the inner surface of a further hopper, which is well fitted into a third hopper all being concentric. In the second and the third hopper, there are slots that can be twisted against each other, resulting in wider or narrower slots. The effect is that a smaller percentage will pass through the slots. Either the smaller sample outside the hoppers or the bigger sample inside the hoppers can be used as the required sample. The position of the two hoppers in relation to each other can be adjusted accurately, resulting in pre-determined subsamples sizes.

#### f. Modified halving method:

The apparatus comprises a tray into which fits a grid of equal-sized cubical cells, open at the top and every alternate one having no bottom. After preliminary mixing, the seed is poured

evenly over the grid. When the grid is lifted, approximately half the sample remains on the tray. The submitted sample is successively halved in this way until a working sample, of approximately but not less than the required size, is obtained.

- g. Spoon method:
- The spoon method is recommended for single small-seeded species and for sample reduction for moisture determination or seed health testing sample reduction for seed health testing.
- For other tests, it is restricted to species with seeds smaller than *Triticumaestivum*; to the genera *Arachis*, *Glycine* and *Phaseolus*, and to tree genera Abies, Cedrus and Pseudotsuga. For all other species, it can only be used to obtain working samples in the laboratory for seed health tests.
- A tray, a spatula and a spoon with a straight edge are required. After preliminary mixing, pour the seed evenly over the tray; do not shake the tray thereafter.
- With the spoon in one hand, the spatula in the other, and using both, remove small portions of seed from not less than five random places. Sufficient portions of seed are taken to constitute a subsample of the required size.



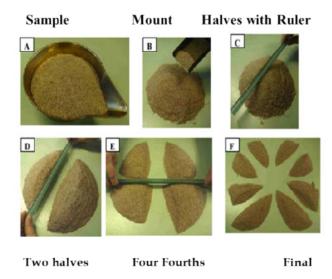
- A. Sample to be reduced.
- **B.** Two spoons
- **C.** A spoon is pushed vertically into the seed layer (as a substitute).
- **D.** Distributing the seed over the pan.
- **E.** With the second spoon the seed in front of the vertical spoon is collected.
- **F.** Both spoons are removed from the seed and the seed sample is transferred to a collection pan.
- G. Two sub-samples as the result.

#### h. The hand halving method:

This method is the most satisfactory method for chaffy and genera of tree and shrub seed. However, this method is restricted to:

 the following genera of chaffy seeds, Agrimonia, Andropogon, Anthoxanthum, Arrhenatherum, Astrebla, beckmannioa, Bouteloua, Brachiaria, Briza, Cenchrus, Chloris, DicghanthiumChloris, Digitaria, Echinochloa, Ehrharta, Elymus, Eragrostis, Gomphrena, Gossypium (linted seed only), Melinis, Oryza, Pennisetum (non glaucum), Psathyrostachys, Scabiosa, Sorghastrum, Stylosanthes (non guianensis), Trisetum, Urochloa;

- to the following genera of easily damaged fragile seeds: Arachis, Glycine and Phaseolus;
- and to the following genera and species of tree and shrub seeds: Acer, Aesculus, Ailanthus, Castanea, Cedrela, Corylus, Fagus, Fraxinus, Juglans, Liriodendron, Pinus cembra, Pinus pinea, Platanus, Populus, Quercus, Salix, Tectona, Ulmus.
- This method can also be used with the species, where all other dividing methods are extremely difficult or impossible to use. The steps involved are given below:
- The seed is poured evenly onto a smooth clean surface.
- Thoroughly mix the seed into a mound with a flat-edged spatula
- The mound is divided into half and each half is halved again, giving four portions. Each of the four portions is halved again giving eight portions which should be arranged in two rows of four.
- Combine and retain alternate portions: e.g. combine the first and third portions in the first row with the second and fourth in the second row. Remove the remaining four portions.
- Steps two, three and four are repeated using the retained portions from step four until the weight of sample required is obtained.



# Random Cups Method



- a. Schematic drawing of the tray, the cups and how to distribute the seed over the tray.
- b. A tray with cups and distributing a sample over the tray.
- c. The tray with the total sample distributed over the tray.
- d. The cups removed from the tray and emptied into a glass vessel.
- e. Cups of different size in one set.

To avoid variability in the results, the divider is operated

- i. Leveled by means of the adjustable feet.
- ii. Divider& four containers are checked for cleanliness.
- iii. A container is placed under each spout.
- iv. The whole sample is fed into the hopper; when filling the hopper, seed must always be poured centrally.
- v. The spinner is operated and the seed passes into the containers.

vi. Full containers are replaced by empty containers. The contents of the two full containers are fed into the hopper together, the seed being allowed to blend as it flows in. The spinner is operated.

vii. The procedure described in (vi) above is repeated at least once more.

*Storage of submitted samples before testing:* Every effort must be made to start testing a submitted sample on the day of receipt. Storage of orthodox seeds, when necessary, should be in a cool, well-ventilated room. Non-orthodox (i.e. recalcitrant or intermediated seeds should be tested as soon as possible after obtaining the submitted sample from the composite sample without any storage. Handling of the submitted sample and, if necessary, storage should be done under species specific optimum conditions.

Storage of samples after testing: The primary aim of storage of samples after testing is to be able to repeat the original tests carried out on the submitted sample. Therefore, storage conditions should be such that changes in the seed quality traits tested are minimal. For example, in the case of the purity test or other seed count, the sample should be stored in such a way that the physical identity is kept. In the case of germination, viability or health test of orthodox seeds the sample should be stored under cool and dry conditions. For such tests in recalcitrant and intermediate seeds of tropical and sub-tropical species, long term storage is not possible. For such seed of

temperate species storability depends on the fungal status and to some extent whether the seed is dormant or not. All factors pertaining to storage need to be determined on a species basis. Protection against insects and rodents may be necessary. When a re-test in a different testing laboratory is required, a portion shall be drawn from the stored sample and submitted to the designated testing laboratory. The remainder shall be retained in the store.

Table 2: Sample weights	of important field and	l vegetable crops
Table 2. Sample weights	of important field and	i vegetable clops

(1)	Minimum	Minimum weight for		
Crop	weight of seed	(2)	(2) (3)	
	lot (kg)	Submitted	Working sample	Working sample
		sample	for purity	for count of
		(gm)	analysis(gm)	other species
				seeds (gm)
		D CROPS		
D 11		nd Millet crops	40	400
Paddy	20,000	400	40	400
Wheat, Barley and Triticale	20,000	1000	120	1000
Oats	20,000	1000	100	1000
Maize	40,000	1000	900	1000
Sorghum	10,000	900	90	900
Pearl millet (Bajra) and	10,000	150	15	150
Common millet (Proso millet,				
Hog millet)				
Italian millet	10,000	90	9	90
Barn yard millet and Kodo	10,000	80	8	80
millet		80	0	00
Finger millet	10,000	60	6	60
Little millet	10,000	70	7	70
	Pu	lse crops		
Chickpea (Gram)	20,000	1000	1000	1000
Pigeon pea (Red gram, Arhar)	20,000	500	200	200
Green gram (Mung bean)	20,000	1000	120	1000
Black gram and French bean	20,000	1000	700	1000
Lima bean	20,000	1000	1000	1000
Lablab bean, Field bean,				
Indian bean (Sem) and Horse	20,000	1000	500	1000
gram (kulthi)				
Cowpea	20,000	1000	400	1000
Garden pea	20,000	1000	900	1000
Lentil	10,000	600	60	600
Chickling vetch	20,000	1000	450	1000

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Kidney bean (Moth bean)	20,000	750	75	750
	Oil	lseed crops		
Castor and Groundnut (pods)	20,000	1000	1000	1000
Groundnut (kernels)	20,000	1000	600	1000
Linseed	10,000	300	30	300
Niger	10,000	150	15	150
Rapeseed and mustard	10,000	160	16	160
Rocket salad (Tara mira)	10,000	40	4	40
Safflower	10,000	1000	180	1000
Sesame	10,000	70	7	70
Soybean	20,000	1000	500	1000
Sunflower (hybrids)	20,000	250	125	250
Sunflower (varieties)	20,000	1000	250	1000
	Fi	bre crops		
Cotton varieties (linted)	20,000	1000	350	1000
Cotton hybrids (linted)	20,000	250	25	250
Cotton varieties/ hybrid(delinted)	20,000	350	35	350
Jute (Patsan)	10,000	100	10	100
Roselle (Mesta)	10,000	700	70	700
Sunnhemp	10,000	700	100	700
	Fo	rage crops		
Birdwood grass (Dhaman)	20,000	25	3	25
Blue panic, Guinea grass, Setaria grass (Nandi grass) and Shaftal	10,000	25	2	20
Buffle grass	10,000	25	3	25
Cluster bean	20,000	1000	100	1000
Dharaf grass	10,000	-	-	-
Doob	10,000	25	1	10
Egyptian clover (Berseem) and Fenugreek (Methi)	10,000	60	6	60
Indian clover (Sweet clover, Senji)	10,000	40	4	40
Lucerne	10,000	50	5	50
Marvel grass and Para grass	10,000	30	3	30
Napier grass	10,000	150	15	150
Oats	20,000	1000	120	1000
Rice bean (Red bean)	10,000	-	-	-

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Stylo	10,000	70	7	70
Sudan grass	10,000	250	25	250
Teosinte	20,000	1000	900	1000
Velvet bean	20,000	500	50	500
Venezuela grass	10,000	25	5	25
Alfalfa (Lucerne)	10,000	50	5	50
Guinea grass	10,000	25	2	20
~	Green Manure a	nd Miscellaneous	Crops	
Dhaincha	20,000	900	90	900
Hemp	10,000	600	60	600
Indigo	20,000	600	60	600
Рорру	10,000	25	1	10
Sugar beet	20,000	500	50	500
Торассо	10,000	25	0.5	5
TODACCO		TABLE CROPS	0.0	
		ulb crops		
Garlic	40,000	-		
Mutiplier onion	40,000	250 bulbs	25 bulbs	250 bulbs
Onion	10,000	80	80	80
Onon	-			00
Proceeds approveds Calibra as		egetables (Cole cr		100
Brussels sprouts, Cabbage, Cauliflower, Broccoli,	10,000	100	10	100
Karamsag, Knol-kohl, Kohlrabi				
and Sprouting broccoli				
Chinese cabbage	10,000	40	4	40
Climitese cabbage	-	ceous vegetables		
Ash gourd Bottle gourd	20,000	700	70	700
Ash gourd, Bottle gourd and Winter squash,	20,000	700	70	700
Bitter gourd	20,000	1000	450	1000
Chow-chow	40,000	250	25	250
	-	_		
Ridge gourd	20,0000	1000	400	1000
Round gourd (Indian	20,000	1000	250	1000
squash),Snake gourd, Sponge				
gourd and Water melon	10.000	150	70	150
Cucumber, Long-melon, Musk	10,000	150	70	150
melon and Snap melon	10.000	250		
Little gourd	10,000	250	25	250
Pumpkin (Kashiphal)	10,000	350	180	350
Pointed gourd	10,000	700	70	700
Summer squash	20,000	1000	700	1000

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Green/Leafy vegetables							
Amaranthus	10,000	70	7	70			
Asparagus bean	20,000	1000	100	1000			
(vegetable cowpea)							
Celery	10,000	25	1	10			
Coriander	10,000	400	40	400			
Fenugreek (Methi) and Parsley	10,000	40	4	40			
Lettuce	10,000	30	3	30			
Parsnip	10,000	100	10	100			
Spinach	10,000	250	25	250			
Spinach Beet	20,000	500	50	500			
	Legumi	nous vegetables					
Broad bean	20,000	1000	1000	1000			
Cluster bean, Dolichos Bean,	20,000	500	50	500			
Field bean, sword bean Velvet							
bean							
French bean	20,000	1000	700	1000			
Goa bean, Jack bean	20,000	500	50	500			
Garden pea	20,000	1000	900	1000			
Lima bean and Scarlet runner	20,000	1000	1000	1000			
bean							
		t vegetables					
Carrot	10,000	30	3	30			
Celeriac	10,000	-	-	-			
Garden Beet	20,000	500	50	500			
Garden rhubarb	10,000	450	45	450			
Globe artichoke	20,000	1000	120	1000			
Jerusalem artichoke	20,000	1000	200	1000			
Radish	10,000	300	30	300			
Rat tail radish	10,000	300	30	300			
Sweet potato and Tapioca	40,000	250 roots	25 roots	250 roots			
Turnip	10,000	70	7	70			
Fruit vegetables							
Tomato (varieties)	10,000	70	7	70			
Tomato (hybrids)	10,000	7	7	7			
Brinjal, Sweet pepper Chillies	10,000	150	15	150			
(Hot Pepper),							
Okra (Bhindi)	20,000	1000	140	10 00			
Rat Tail Radish (Mungra)	10,000	300	30	300			

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# INFLUENCE OF ABIOTIC & BIOTIC FACTORS ON SEED QUALITY

# M.P. Yadav, Seed Technologist National Seed Research & Training Centre, Varanasi (U.P.)

Seed plays a vital role in agriculture and acts as a carrier of genetic potentialities of improved varieties. Conjunctive use of quality seeds along with other inputs will tap the superior genetic potentiality of high yielding varieties and bring benefits to the farmers. Hence, production of high quality seeds in a planned manner is very essential for increased agricultural production, thereby enhancing the economic status of the farmers. As the quality seed is very much essential and crucial input in agriculture, utmost care should be given in the process of seed production. Maintenance of seed quality at various stages of seed crop is highly essential because many abiotic and biotic factors act upon the crop growth and thus have their influence on seed quality factors Biotic and Abiotic Factors.

Ecological factors which affect dynamic change in a population or species in a given ecology or environment are usually divided into two groups: abiotic and biotic. "All the living organisms that inhabit an environment are called biotic factors" Biotic Factors are, in entirety, anything that affects living organism that is itself alive. Such things include animals which consume the organism in question, or the food that the organism consumes. As opposed to abiotic factors (non-living components of an organism's environment, such as temperature, light, moisture, air currents, etc.), biotic factors are the living components of an organisms environment, such as predators and prey. Biotic means something that is living. Biotic is the opposite of abiotic which means non-living. Biotic means relating to, produced by, or caused by living organisms. The term biotic may also refer to:

- Life, the condition of living organisms'
- Biology, the study of life
- Biotic Factors in ecology,
- Biotic material, which is derived from living organisms,
- Biotic potential, an organism's reproductive capacity, or
- Biotic Banking Brigade, an unofficial group of pie-throwing activists.
- In the world of Mass Effect Biotics are people capable of using their minds to manipulate mass effect fields.

**Abiotic factors** are geological, geographical, hydrological and climatological parameters. A **biotope** is an environmentally uniform region characterized by a particular set of abiotic ecological factors. Specific abiotic factors include:

• Water, which is at the same time an essential elements to life and a milieu

- Air, which provides oxygen, nitrogen, and carbon dioxide to living species and allows the dissemination of pollen and spores
- Soil, at the same time source of nutriment and physical support soil pH, salinity, nitrogen and phosphorus content, ability to retain water, and density are all influential
- Temperature, which should not exceed certain extremes, even if tolerance to heat is significant for some species
- Light which provides energy to the ecosystem through photosynthesis
- Natural disasters can also be considered abiotic

Abiotic components can be split into 3 main categories: climatic, eudaphic and social. Climatic factors include sunlight, humidity, temperature, atmosphere etc. Eudaphic factors are things to do with the nature of the soil and ground, such as the geology of the land and the soil type. Social factors include land use, water resources etc.

# I. Abiotic Factors on Seed Quality

**1.** Ecological Factors: Even though seed quality is influenced by numerous factors namely ecological factors, production factors and post harvest technology factors during seed production, the ecological factors (weather) are of paramount importance as mankind has no control over them. The weather parameters include temperature, light, rainfall, wind velocity and relative humidity, which play a vital role on the production of quality seeds.

i) Temperature: Of all the factors effecting seed production, temperature is considered as the most important one. In rice crop, prevalence of either high or low temperature during crop growth, affect the production of reproductive parts namely seed weight, size and seed yield. Seed dry weight in rice was stable over the temperature range from 21/16<sup>o</sup> to 30/25<sup>o</sup>C. *Jia et. al.* (1991) have stated that high temperature after heading of rice crop resulted in increased chaffiness by shortening the effective tillering stage. In wheat crop, Kernel weight decreased as temperature rose above 18<sup>o</sup>C. In maize, increased kernel abortion was reported up to 36% in high temperature condition (Cantarero et. al. 1999). In vegetable crops, the growth of warm season crops stops at 15<sup>o</sup> C and growth normally increased up to 40<sup>o</sup>C.

Temperature may be considered as a measure of intensity of heat energy. The range of maximum growth for most agricultural plants is between 15 and 40°C (59 to 140°F). The temperature of a place is largely determined by its distance from the equator (latitude) and the altitude. Based on the above the vegetations are classified as tropical, temperate, taiga, tundra and polar. Every plant community has its own minimum and maximum temperature known as their cardinal points. The table indicates the cardinal points of some of the common crops.

# Cardinal temperature of certain crops for germination

Сгор	Minimum <sup>0</sup> C	Optimum <sup>0</sup> C	Maximum <sup>0</sup> C
Wheat	4.5	20	30-32

National Training on "Seed Certification", January 08-12, 2024 National Seed Research & Training Centre, Varanasi (U.P.)

Barley	4.5	20	29-30
Oats	4.5	20	29-30
Maize	8-10	20	40-43
Sorghum	12-13	25	40
Rice	10-12	32	36-38
Tobacco	12-14	29	35

**Chilling injury -** Plants growing in hot climate are exposed to low temperature (which is above the freezing point) for some time, and are found to be killed or injured severely. Chlorotic condition or bands on leaves of sugarcane, sorghum and maize when exposed for 60 hours at 2 to 4<sup>o</sup>C.

**Freezing injury** – This is generally caused in plants growing in temperature region. When the plants are exposed to very low temperature, water is frozen into ice crystals in the intercellular spaces. The protoplasm of the cell is dehydrated and mechanical distortion takes place resulting in killing of the cells. Frost damage to potatoes, tea etc in winter in the hilly areas like Nilgiris.

**Suffocation –** During winter the ice or the snow form a thick cover over the ground and the crop suffers for want of oxygen. Ice in contact with roots inhibits diffusion of carbon dioxide and the respiratory products may become harmful to plants.

**Heaving** – Injury to plants is caused by a lifting upward of the plant along with the soil from its normal position in temperate regions where snowfall is a common phenomenon.

**Heat injury –** Very high temperature often stops growth. The plant faces incipient starvation due to high respiration rates. The plant is stunted and if such a condition persists for a long period the plant is killed. Direct temperature effects are noticeable in young seedlings and transplanted crops. High temperature cases sterility in flowers. The general effects of excessive heat are defoliation, premature dropping or fruit and in extreme cases death of plant.

**ii)** Light: The light requirement at each kind of crop should be known otherwise, when they are grown in other conditions, their seed quality will be affected. For instance, wheat varieties in general are photosensitive while improved rice varieties are photo insensitive. In maize, the shading period from 3 to 17 days after silking has significant effect on the number of abortive seeds.

**Light intensity** – In photosynthesis about one per cent of the light energy is converted into potential chemical energy. Very low light intensity reduces the rate of photosynthesis and may

even light intensity reduces the rate of photosynthesis and may even result in the closing of the stomata very high light intensities are detrimental to plants in many ways. It causes rapid loss of water resulting in the closure of stomata. The most harmful effect of high light intensity is the phenomenon of solarization in which all the cell contents are oxidized by atmospheric oxygen. This oxidation is different from respiration and is termed photo oxidation. Heliophytes (sun loving) and sciophytes (shade loving), the dry matter production is affected. Many species produce maximum dry matter under high light intensity if water is available in plenty.

**Quality of Light -** When white light is passed through a prism it is dispersed into wavelengths or different colours; violet 400-435 m $\mu$ ; blue 435-490; green 490-514; yellow 574-595; orange 595-626 and red 626-750. The principal wavelengths absorbed in photosynthesis are in the violet-blue and the orange-red regions. Among these short rays beyond violet such as infrared are detrimental to growth. Red regions. Among these short rays beyond violet such as infrared are detrimental to growth. Red light seems to be the most favourable light for growth followed by violet-blue.

**Duration of Light –** The length of the day has greater influence than the intensity. The response of plant to the relative length of day and night is known as photoperiodism. Plants which develop and produce normally when the photoperiod is greater than a critical minimum (more than 12 hours of illumination) are called 'long day plants' and those develop normally when the photoperiod is less than a critical maximum (less than 12 hours of illumination) are called 'short day plants.' Some plants are found to be unaffected by photoperiod and are called as ' day-neutral' plants. Plant characters like floral development, floral initiation, bulb formation, rhizome production etc., are all influenced by photoperiodism. Among the crop plants soybean, maize and millets are examples of short day flowering plants while sugar beet, wheat, and barley are long day plants and others are intermediate (day neutral) in that they flower at any day lengths.

**Direction or Light -** Shoots roots and leaves show different orientation to the direction of light. In temperate regions the southem slopes show better growth of crops than the northern slopes due to the direction of light contributing more sunlight towards the southern side.

**iii) Rainfall:** In the regions of moderate rainfall and humidity as compared to regions of high rainfall, good quality seeds can be obtained. Excessive rainfall interferes with the pollination, results in delayed maturity and precocious germination or vegetable seeds.

**iv) Wind Velocity:** Excessive wind during pollen production will result in poor seed set and ill filled seeds. Brown seed discoloration is caused by wind at flowering and occurrence milky white seed is increased by wind 14-21 days after heading in rice. (Ebata and Ishikawa, 1989). Wind affects the growth mechanically and physiologically. The sand and dust particles carried by the wind may damage plant tissues. Emerging seedlings may be completely covered or alternatively the roots of young plants may be exposed by strong winds. Winds may also cause considerable losses by inducing lodging, breaking or stalks and shedding of grains. The physiological effects of wind consist mainly in increasing transpiration as well as evaporation from the soil. Hot dry winds, may however adversely affect photosynthesis and hence productivity, by causing closure of the stomata

even when soil moisture is adequate. Moderate winds have a beneficial effect on photosynthesis by continuously replacing the carbon dioxide absorbed by the leaf surface.

**v) Relative Humidity:** Flowering, pollination and seed setting in temperate crops needed low humidity – dry weather at seed maturity and moderate to low humidity for sub temperate and topical crop. High humidity and temperature encourage production of diseased seeds. Warm dry climate is suitable for production of disease free seed.

**2. Season:** More extreme cases of unseasonal climate usually reduce the flowers and subsequently the seed set. Late spring frosts in temperate regions kill flowers and young fruits. An abnormal drought also has similar effects. Even if death or premature shed of whole fruits does not occur, a proportion of seeds may abort later.

**3. Irrigation:** Irrigation at critical growth phases is very much essential for the normal seed development and maintenance of seed quality. Water stress during critical phases like seed germination, seedling establishment, flowering, fruiting and seed development stages is detrimental to seed set, yield and seed quality.

**4. Soil nutrient and management factors:** The fertility of the soil in which a plant grows influences the chemical composition of the developing seed and consequently its metabolism and vigour during germination. Nitrogen and phosphorus availability can influence seed development and seedling vigour, but their effect varies among species. It is probable that inorganic nutrients stored in seed provide valuable reserves during early germination of seeds. Minor elements are also known to affect the seed development. Deficiency in soil boron causes 'Hollow heart' defect in pea seed. In rice seeds, copper deficiency heads to loss of viability. Calcium deficiency causes darkened plumule in groundnut. Mn/Mo deficiency leading to 'Marshy spots' in peas grown in calcareous soil and it leads to promotion of hard coated seeds and hard seededness.

**5. Post harvest handling of seeds:** Seeds of few species cannot tolerate excessive desiccation and can still retain their viability. It is very important to harvest the seed in proper time to get best quality of seeds. Harvesting at an early stage will cause damage to seeds during threshing and cleaning. If the harvesting of the seeds are not done in the right time and optimum moisture content and allowed to remain in the field until dead ripe stage, the seeds are likely to be exposed to weather elements like rain and temperature, besides insect and fungal pathogens and suffer mechanical damage.

#### I) Seed Drying

Excessive drying rates may cause stress cracks in the seed, because of unequal drying throughout the seed. Even if the seeds are not dries too fast, they may be injured by over drying and usually this will lead to increased susceptibility to mechanical damage of the seeds there by reduction in the quality of seed.

#### ii) Moisture Content

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Susceptibility to mechanical damage increases, as the moisture content decreases. However, safe moisture content varies among species. Large seed legumes are particularly sensitive and excessive injury begins to occur at moisture content below 15%.

#### iii) Mechanical Damage

Mechanization of harvest, handling and conditioning operation subject seed to physical forces that can do mechanical resistance of the seed covering and the seed as a whole will be exposed to crusting impacts, abrasion, and various types of cutting and shearing action. The failure of the seed coverings to protect the embryonic axis and nutritive tissue affects the germination capacity of the seed.

#### iv) Seed Storage

The storage sanitation is very much essential that will tell upon the quality of the seed. Based on the storability of the seeds, the storage condition should be provided. Matrinez et. al., (1994) revealed that most beans varieties stored for 180 days at 75% RH maintained high germination rates (74-98%) on seed quality.

#### **II. Biotic factors**

#### 1) Diseases

Seed borne diseases of different crops cause damage to the crop and reduce the seed yield and quality. They cause loss to the agricultural economy in different forms. Losses may be immediate with the first crop produced from the infected seed lot and it may be a long-term effect if the pathogens are able to survive in the soil debris and weed hosts. It will serve as a storehouse to spread the infection to the succeeding susceptible crops. Hence, production of disease free seeds is of prime importance.

#### 2) Pests

Considerable reduction in the seed quality can be occurred due to insect outbreak in considerable quantity. Especially in the stored products, the main sources of infestations are field infestation, infestation by migration, infestation through conveyance and storage buildings or structures, birds, bird nests, ants, rodents etc. They cause hollowing of the seeds. Kernels are reduced to mere powder. *Sitophilus oryzea* and *S. zeamais* are known to attack seeds in fields too. Adults cut circular holes. Heating takes place during heavy infestation, which will lower the seed quality.

#### 3) Weed seeds

Weeds are exceptionally tough plants and are able to reproduce e aggressively and quickly outnumber other desired plants. Especially in the seed production plots, the chances for contamination and admixture of weed seeds are more resulting in the reduction of seed quality. Right from the sources of production, processing, storage up to sowing of the seed by the cultivators, the scope of pollution remains. The weed plants affect the normal plant growth of a seed crop by becoming a competitor for space, light, water and nutrients and reduce the seed yield and vigour. During harvest, the weed seeds mix with seeds and thereby the recommended seed standards cannot be met.

#### Conclusion

Now-a-days seed has taken a new dimension and to improve the economy of our county, productions of high quality seeds are highly warranted. Hence, application of favorable biotic and abiotic factors in the process of seed production will not only yield us high quality seeds but also make our seed production a profitable venture.

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# SEED TESTING: AN OVERVIEW

# Dr. Sandeep Kumar Lal Principal; Scientist and Incharge, Seed Testing Laboratory Division of Seed Science and Technology ICAR- Indian Agricultural Research Institute, New Delhi

Seed testing has been developed to evaluate the planting value of seed for minimizing the risks of planting low quality seeds through assessment of seed quality prior to sowing. Seed testing cannot make the seed better, but appropriate measures can be taken based on test results to avoid hazards in agricultural production or remedy for poor seed quality. Seed quality is a concept, comprising of different attributes which are of interest to different segments of the industry - to the producer, the processor, the warehouseman, the merchant, the farmer, the certification authority and to the government or agency responsible for seed control. In all cases, the ultimate object of making a test is to determine the planting value of seed. Since seed is a living biological product, its behaviour cannot be predicted with the certainty that characterizes the testing of inert or non-biological material. The methods used must be based on scientific knowledge of seed and on the accumulated experience of seed analysis; the accuracy and reproducibility required depend on the purpose of the test.

Seed testing is of major importance for all those who produce, sell and use seeds. It is the science of evaluating the planting value of seed for minimizing the risk of planting low quality seeds and the primary aim of the seed testing is to get accurate and reproducible results regarding the quality status of the seed samples submitted to the seed testing laboratories. It refers to testing of the seed for the determination of its quality viz. the percentage composition of pure seed, weed seeds, seeds of other species, other cultivars, moisture content, germination, vigour health of the seed lot in question is regarded as seed testing. It helps to gain information regarding planting value of seed lots, which should be performed to obtain accurate and reproducible results across the laboratories. However, the main aim of the seed testing is to obtain accurate and reproducible results across the above-mentioned seed quality parameters of a seed lot, thereby enabling the farmer community to get quality seeds. Seed testing is conducted to achieve the following objectives:

- Planting purposes: To assess seed quality of seed lots, i.e. their suitability for planting
- **Labeling purposes:** To determine whether a seed lot meets established quality standards or labelling specifications or not?
- **Upgrading the seed quality**: To determine the need for drying and processing and specific procedures that should be used
- Seed Certification purposes
- Seed Law enforcement purposes
- To obtain results which can be used to compare the planting value of different lots
- To identify the seed quality problems and their probable causes

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- To determine the need for seed drying and processing and specific procedures that should be used.
- **Fixation of seed prices**: To establish quality and provide a basis for price and consumer discrimination among lots in the market.
- Predicting storability of seeds.

Seed testing or seed quality evaluation is a highly specialized job, which is an important aspect of seed programme in India for the purpose of certification and seed law enforcement. Therefore, seed testing laboratory is considered as the hub of seed quality control. Hence, it is necessary that seed testing laboratories are established, manned and equipped in a manner such that the samples received could be analyzed in the minimum possible time so that the quality control work and the needs of seed industry are effectively met. The important pre-requisites for ensuring good seed testing work include the following:

- Establishment of a STL for the seed industry and farmers.
- Availability of the sets of seed testing procedures, rules and manuals.
- Seed herbarium of the crop variety and weed seed species of the area for correct identification.
- Controlled-environment, rat-proof sample storage room for guard and test awaiting samples.
- Leaders with scientific background to serve the interests of lab and the seed industry (including farmers).
- A highly responsible and dedicated staff providing efficient service i.e. prompt analysis and team spirit among the employees.
- Uniformity in equipment, protocols and interpretation i.e. consistently good facilities and skilled analysts
- Promotion of research, leading to improvement of the whole seed programme, especially of testing procedures.

The importance of seed testing was realized more than 100 years ago for assured planting values, when the adulteration of vegetable seeds was practiced by mixing stone dust in some parts of the world, particularly in Europe, when Professor Friedrich Nobbe (Germany) in 1969 advocated that the seeds must be tested before sowing. Nobbe's hypothesis was based on scientific investigations made by him on the vegetable and flower seed samples offered for marketing in European Countries. This gave birth to the establishment of seed testing laboratories in European countries, USA and Canada during late eighties and early nineties. The establishment and development of seed testing laboratories in the developed world generated tremendous impact in the seed trade and the development of seed testing procedures. The International Seed Testing Association (ISTA) was established in 1924 and the first set of International Rules of Seed Testing were framed and published by ISTA during 1931.

At present, about 140 Seed Testing Laboratories are functioning in the country and testing more than 6 lakh seed samples annually, including 2 Central Seed Testing Laboratories at National Seed Research and Training Centre, Varanasi and Central Institute for Cotton Research, Nagpur (for GM cotton only).

#### **Consequences of faulty Seed Testing**

- 1. **Poor field emergence**: This may happen when the seed analyst is not well-versed with normal and abnormal seedling characteristics, leading to over estimation of germination test results.
- 2. **Rapid deterioration of germination during storage**: This may happen due to improper determination of seed moisture content, resulting in recommendation of a safe (low) M.C. of the submitted sample, based on which the producer stores the seed without further drying and hence leading to rapid decline in germination.
- 3. **Varietal admixtures / genetic impurity reported by the grower**: This may happen due to inability of the seed analyst in differentiation of other distinguishable varieties.
- 4. **Spread of obnoxious weeds:** This may happen due to inability of the seed analyst in identifying such weeds and/ or due to unavailability of reference material for weed identification.
- 5. **Spread of designated seed borne diseases**: This may happen due to inability of the seed analyst in identifying seed borne diseases and/ or due to unavailability of reference material. Besides, lack of infrastructure and training opportunities for the analysts. A correct assessment can either reject such lots (above the permitted level) or suggest the necessary seed treatment for control.

#### The basic components of seed testing include:

- a) Seed Sampling
- b) Physical Purity Analysis
- c) Determination of seed moisture content
- d) Germination Test

#### In addition, other important aspects of seed quality are also tested viz.

- Seed Health
- Seed viability
- Seed vigour
- Species & Variety Identification
- Testing of specified traits/ GM detection

#### PHYSICAL PURITY TESTING

Physical purity analysis provides information about the proportion of pure seed component in the seed lot as well as the proportion of other crop seeds weed seed and inert matter by weight in percentage for which seed standards have been prescribed. Thus, it helps in:

- 1. Improving the plant stand (by increasing the pure seed component).
- 2. Raising a pure crop (by eliminating other crop seed and weed seeds).
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- 3. Raising a disease free-crop (by eliminating inert matter).
- 4. In the use of seed drill (by selecting uniform particles).

#### There is a need for physical purity analysis for:

- 1. Seed Certification or Seed Law Enforcement Agencies to judge that the seed lot conforms to the prescribed standards.
- 2. Seed processing plants for using right kind of processing equipment.
- 3. Physical purity analysis is a pre-requisite for germination test because 'pure seed' component is used for germination testing.

#### Objective

The primary objective of physical purity analysis is to determine:

- 1. The percentage composition by weight of the sample being tested and by inference the composition of seed lot; and
- 2. The identity of various species of seeds and inert particles constituting the sample.

The definition of the various physical purity components in the ISTA Rules are as follows:

#### Pure seed

The pure seed shall refer to the species stated by the sender, or found to predominate in the test, and shall include all botanical varieties and cultivars of that species (even if immature, undersized, shrivelled, diseased or germinated, providing they can be definitely identified as of that species) unless transformed into visible fungal-sclerotia, smut balls or nematode galls.

#### Pure seed shall include:

a) Intact seed units (commonly found as dispersal units i.e. achenes and similar fruits, schizocarp, florets etc.) as defined for each genus or species;

b) Pieces of seed units larger than one half their original size.

# From the above main principles, certain exceptions are made for particular genera or species as follows:

- 1. Seed units of families *Leguminaceae*, *Cruciferae*, *Cupressaceae*, *Pinaceae* and *Taxodiaceae* with the seed coat entirely removed shall be regarded as inert matter. Separated cotyledons of *Leguminaceae*are regarded as inert matter, irrespective of whether or not the radicle-plumule axis and/or more than half of the testa may be attached.
- 2. In certain genera of family *Gramineae*.
- a. A minimum size of caryopsis is required i.e. in *Lolium, Festuca* and *Elytrigiarepens* a floret with a caryopsis one third or more of the length of palea measured from the base of rachilla is regarded as pure seed, but a caryopsis less than 1/3 the length of the palea is regarded as inert matter.
- b. The presence of caryopsis in spikelets and florets is not always obligatory.

- c. The separation of pure seed and inert matter is done by uniform blowing procedure. This method is obligatory for *Poa pratensis* and *Dactylisglomerata*.
- d. Multiple seed unit's (MSU) are left intact in the pure seed fraction e.g. Dactylis and Festuca.
- *e.* Attached sterile florets are not removed, but left attached and included in the pure seed fraction e.g.*Arrhenatherium, Avena, Chloris, Dactylis, Festuca, Holcus, Poa, Sorghum* and *Triticum spelta.*
- f. For certain genera appendages are left on the seed but reported if found to the extent of 1% or more, the percentage of such material must be shown on Analysis Certificate (example – paddy).

**Other crop seed:** Other crop seed shall include seed units of any plant species other than that of pure seed grown as crops. Multiple structures, capsules, pods are opened and the seeds are taken out and the non-seed material is placed in the inert matter.

**Weed seed:** Seeds, bulblets or tuber of plants recognized by laws, official regulations or by general usage shall be considered as weed seeds.

**Inert matter**: Inert matter shall include seed units and all other matter and structures not defined as pure seed, excluding other crop seed and weed seeds.

#### **General principles**

As per ISTA Rules, the working sample is separated into three components i.e. pure seeds, other crop seeds, and inert matter. The percentage of each part is determined by weight. All species of seed and each kind of inert matter present shall be identified, as far as possible and if required for reporting, its percentage by weight shall be determined.

# Equipments

Aids such as transmitted light, sieves and blowers may be used in separating the component parts of the working sample. The blower is to be used, for the uniform blowing method, for species of family Gramineae.

#### Other equipments required are:

- a. Dividers
- 1. Soil type
- 2. Boerner (work on gravitational force)
- 3. Gamete (works on centrifugal force and electrically operated)
- b. Balance: Electric or electronic balance are better due to their accuracy and quickness
- c. Blowers
- d. Diaphnoscope using reflected light are used to separate inert matter, such as empty florets of grasses.
- e. Sieves
- f. Sample pans, dishes, forceps, spatula and hand lens
- g. Seed herbarium of crop and weed seed

# Procedure

*Obtaining working sample:* Since the size of the working sample is minute as compared with the size of the seed lot to which it represent, it is therefore, very essential that the working sample should be obtained in accordance with the procedures. The working sample shall be either a weight estimated to contain atleast 2,500 seed units or not less than weight indicated i.e. 40 g for *Oryza sativa*. Boerner or soil type seed divider should be used to homogenize the submitted sample before reducing it to the size of working sample. The following guidelines need to be followed:

- a. Check the cleanliness of the divider and the container.
- b. Pour the entire contents of the submitted sample into the hopper of the divider.
- c. Allow the content of the submitted sample to pass through the main body of the divider. In case of 'Soil type' seed divider, this can be accomplished by tilting the hopper over the body of the divider while in case of 'Boerner' divider, by opening the gate-valve situated at the base of the hopper.
- d. Recombine the contents of both sample receiving pans and again pass it through the divider.
- e. Repeat this process twice in order to homogenize the submitted sample.
- f. Divide the submitted sample.
- g. Set aside the contents of one container.
- h. Divide the contents of the other container subsequently till the weight of working sample is obtained.

# Separation

- 1. Clean the work board, sample and purity dishes before starting the separation
- 2. Examine the working sample to determine the use of particular aid, such as blower or sieves for making separation.
- 3. After preliminary separation with the help of sieves or blower, place and spread the retained or heavier portion (A) on the purity work board.
- 4. With the help of spatula or forceps, draw working sample into thin line and examine each particle individually. The criteria used being the external appearance (shape, size, colour, gloss, surface texture) and/or appearance in transmitted light.
- 5. Separate out impurities such as other crop seeds, weed seeds and inert matter and place the impurities separately in purity dishes, leaving only the pure seed on the purity board.
- 6. Seed enclosed in fruits other than those indicated in pure seed should be separated and the detached empty fruit/appendages classed as inert matter.
- 7. Collect the pure seed in the sample pan.
- 8. Put the lighter portion (B) of the work board and examine under magnification for further separating into the requisite classes (other crop seed, weed seed and inert matter).
- 9. After separation, identify the other crop seed, weed seed and record their names on the analysis card. The kind of inert matter present in the sample should also be identified and recorded.
- 10. Weight each component, pure seed, other crop seed, weed seed and inert matter in grams to the number of decimal places shown below:

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S.No.	Wt. of working sample (g)	No. of decimal place required	Example
1.	Less than 1	4	0.9025
2.	1 to 9.990	3	9.025
3.	10 to 99.99	2	90.25
4.	100 to 999.9	1	902.5
5.	1000 or more	0	9025

11. Calculate the percentage by weight of each component to one decimal place only, basing the percentage on the sum of the weight of all the four components. Component of 0.05% to 0.1% are reported as 0.1%. The components of less than 0.05% shall be recorded as 'Trace'.

*Reporting results:* The results of purity test be given to one decimal place only and the percentage of all component must total 100. If the result for a component is nil, this must be shown as 0.0% in the appropriate space of the report form. The report should also include the kind of inert matter and the Latin names of the crop seed and weed seed found in the sample.

#### DETERMINATION OF OTHER SEEDS BY NUMBER

The object of the determination of other seeds by number is to estimate the number of seeds of other species stated by the applicant either generally (e.g. all other species) or by reference to one category of seeds (e.g. species scheduled as noxious in a certain country) in a seed lot. As and when there are 'Seed Standards' for other seeds (in the Indian context, we have standards for other crop seeds, weed seeds, objectionable weed seeds in a number of crops), in number/kg (not in percentage by weight as in physical purity analysis), the determination of other seeds by number is made under 'other determination'. In the International trade, it is mainly done for objectionable weed seeds.

The test could be complete, limited, reduced or reduced-limited.

- A complete test is one in which the whole working sample is searched for all other seeds present
- A *limited test* is one in which the search is limited to stated species only in the whole working sample.
- A *reduced test* is one in which only part of the working sample is examined.
- A *reduced-limited* test is one in which less than the prescribed weight of seed for a working sample is examined for stated species only.

**General principles:** The determination is made by count and expressed as number of seeds found in the quantity examined. When seeds found cannot be identified with certainly to the species level, it is permitted to report the genus name only.

**Apparatus:** Sieves, blowers and other mechanical devices can be used to aid the analyst in examining the sample and reducing the work involved.

# Procedure

Working sample

a) The size of the working sample shall be either a weight estimated to contain at least 25,000 seed units or the weight prescribed for submitted sample.

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If a species stated by the applicant is difficult to identify a minimum of one fifth of the prescribed working sample weight only need be examined for that particular species.

**Determination:** The working sample is searched either for seeds of all other species or of certain stated species, as required by the applicant. The number of seeds found of each species sought is counted. If the search is limited to certain stated species, the examination may be stopped when one or more seeds of one or all of the stated species (as appropriate to the applicant's requirements) have been found.

**Calculation and expression of results:** The result is expressed as the number of seeds belonging to each stated species or category found in the actual quantity examined. In addition the number per unit weight (e.g. per kilogram) may be calculated.

**Reporting results:** The actual weight of seed examined, and the scientific name and number of seeds of each species sought and found in this weight shall be reported, in addition number of seeds of each species per kilogram should be given. The certificate should mention complete test, limited test, reduced test or reduced-limited test.

Current methods being adopted in physical purity analysis are time consuming, cumbersome and require skilled manpower. New equipments and techniques are already being used in advanced seed testing laboratories. The uniform blowing procedure is new and widely used method for purity analysis, including purity separations and other seed determinations in the laboratories that test cereal seed crops, to separate empty florets and other lightweight inert matter from pure and well-developed seeds. Blower monitoring system using anemometer is also being developed to make this methodology more user-friendly. The Oregon State University Seed Laboratory has developed a purity testing system known as the **Ergovision System**, which is being used in purity testing of small seeded species such as grasses or vegetables. This system consists of a -optic light for image clarity, optimum magnification and an automated seed flow in the field of view. Microscope purity station with built-in transmitted light is used for inspecting seeds during purity examination, which is used in testing grass seeds, it consists of a diaphanoscope with transmitted light to determine whether seed contain caryopses or are empty. We can also judge the degree of endosperm development. Similarly, few models of seed scanner are available in the market today, consisting of a camera connected to a computer; and the system is used for identification of seed species. X-ray technology is being used to monitor the seed cleaning process to separate filled and empty seeds as well as to detect physical/ insect damage. Digital portable X-ray machines are already available in the market; it has an overall advantage to store all images electronically for later verification and decision making.

# DETERMINATION OF ODV (OTHER DISTINGUISHABLE VARIETIES)

The object of the determination of ODV is to estimate the number of seeds of other distinguishable varieties present in a seed lot of designated variety.

#### Field of application: The determination is valid only if

- (1) The seed standards are available
- (2) The cultivar is stated by the sender

(3) The authentic sample of the cultivar is available for comparison.

**General principles:** The determination shall be made only on the basis of readily apparent differences in the stable and well known morphological characters of the seed. Wherever, the difference has occurred due to physiological factors, such as frost, drought, immaturity, storage or due to any other reasons which have affected size, shape and luster of the seed the Seed Analyst should not classify those seeds as 'ODV'.

#### Apparatus and facilities

- i. Work board, Magnifier, Stereoscopic Microscope, Spatula and Forceps
- ii. Authentic samples of the cultivar.

**Procedure:** This determination must be made before conducting the physical purity analysis.

**Working sample:** The whole submitted sample must be used for making the determination. The analyst should, therefore, weigh the submitted sample and record the weight on the analysis card. The sample should be examined under magnification to determine the number of seeds of other cultivar present in the sample.

**Calculation and expression of the results**: The result of the test is expressed as the number of seeds belonging to the other cultivar found in the actual quantity examined. In addition, the number per unit weight (e.g. per kg) may be calculated.

**Reporting the results:** The actual weight of the seed examined and number of the seeds of the other cultivar present in the sample shall be reported on the Analysis Certificate. In addition, number per unit weight (e.g. per kg) may be reported.

**Scope and limitations:** The determination of the other distinguishable variety (ODV) seeds in a seed lot, as a seed quality attribute, came into existence in the year 1988, when the Ministry of Agriculture & Cooperation, Govt. of India published the Indian Minimum Seed Certification Standards (IMSCS) in which the maximum permissible limit of ODV seeds in 43 crop plants including cereals such as maize and paddy, pulses (most of the pulse crops such as bengal gram, pigeon pea, pea, cowpea, lentil, mungbean, urdbean etc.), oilseeds such as soybean and mustard and vegetable crops, such as okra were prescribed. As per today's rule status, all the certified seed lots of the above mentioned crops are to be tested for ODV and should conform to the prescribed standards before certification is granted.

#### GERMINATION TEST

In seed testing, germination has been defined as "the emergence and development from the seed embryo of those essential structures which, for the kind of seed tested indicate its ability to develop into a normal plant under favourable, conditions in soil". Laboratory germination begins with the primary root piercing the seed coat and ends when the seedling has developed to the stage that it can be evaluated according to the ISTA Rules. Hence, Germination is the emergence and development of the seedling to a stage where the aspect of its essential structures indicates whether or not it is able to develop further into a satisfactory plant under favorable conditions in the field (ISTA, 2021) The germination percentage indicates the proportion by number of seeds which have produced seedlings classified as normal under prescribed conditions.

**Object of the germination test**: To determine the germination potential of a seed lot, which can then in turn be used to compare the quality of different lots and also estimate the field planting value to predict performance of the seed and seedling in the field. **Principle**: Germination tests shall be conducted with a pure seed fraction. A minimum of 400 seeds are required. The test is conducted under favourable conditions of moisture, temperature, suitable substratum and light if necessary. No pre-treatment to the seed is given except for those recommended by ISTA.

**Number of replications**: Four replication of 100 seeds, A minimum of 3 replication of 100 seeds may be used under unavoidable situations or Eight or six replications of 50 seeds or Sixteen/twelve replication of 25 seeds according to the kind of and size of containers.

In certain circumstances, it may be necessary to test less than 400 seeds. In such cases, at least 100 seeds must be tested in replicates of 25 or 50. At the request of the applicant, a germination test can be carried out on 200 seeds, for issuance on a Blue International Seed Sample Certificate only. In this case, the number of seeds tested is less than 400 and must be reported under 'Other Determinations'

#### Materials required

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

# 1. **Sand**

**Size of sand particle**: Sand particles should not be too large or too small. The sand particles should pass through 0.80 mm sieve and retained by 0.05mm sieve.

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

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

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

2. **Paper**: Most widely used paper substrates are filter paper, blotter or towel (kraft paper). It should have capillary movement of water, at vertical direction (30 mm rise / min.). It should be

free from toxic substances and free from fungi or bacteria. It should hold sufficient moisture during the period of test. The texture should be such that the roots of germinating seedlings will grow on and not into the paper.

Type of paper	Basic Mass (gm/ m <sup>2</sup> )	Bursting Strength (Kg/ cm <sup>2</sup> ), min	Capillary rise (in mm), min	-	Ash % by mass (max.)
Filter Paper	130-135	1.0	30	6.0 - 7.5	1.2
Towel Paper	90-95	2.0	30	6.0 - 7.5	1.5

The general specifications of paper media are given below:

#### Methods

# Top of paper (TP)

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

# Between paper (BP)

The seeds are germinated between two layers of paper. The seeds are placed between two layers of paper and rolled in towels. The rolled towels are placed in the germinator in an upright position.

# Germination apparatus

**Germination cabinet / Germination room**: This is called chamber where in temperature and relative humidity are controlled. We can maintain the temperature, relative humidity and light required for different crops.

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

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

# Duration of testing

First count: Is approximate, but sufficient for an accurate seedling evaluation.

**Intermediate count**: In order to remove normal well-developed seedlings and seedlings/seeds presenting infection

**Final count**: May be shortened if the maximum germination is reached before the end or may be extended if seeds have slow germination.

The duration of the test is determined by the time prescribed for the, final count but the chilling, periods before or during the test, which is required to break dormancy, is not included in the test period. If at the end of the prescribed test period some seeds have just started to germinate, the test may be extended for an additional period up to 7 days. A test may be terminated prior to the prescribed time when the analyst is satisfied that the maximum germination of the sample has been obtained. The time for the, first count is approximate and a deviation of 1-3 days is permitted. The First count may be delayed to permit the development of root hairs in order to be certain that root development is normal, or may be omitted. Intermediate counts may be at the discretion of the analyst to remove seedlings, which have reached a sufficient state of development for evaluation, to prevent them becoming entangled. But the number of intermediate counts should be kept to a minimum to reduce the risk of damaging any seedlings that are not sufficiently developed. Seedlings may have to be removed and counted at more frequent intervals during the prescribed period of the test when a sample contains is infected with 'fungi or bacteria. Seeds that are obviously dead and decayed, and may, therefore, be a source of contamination for healthy seedlings, should be removed at each count and the number recorded.

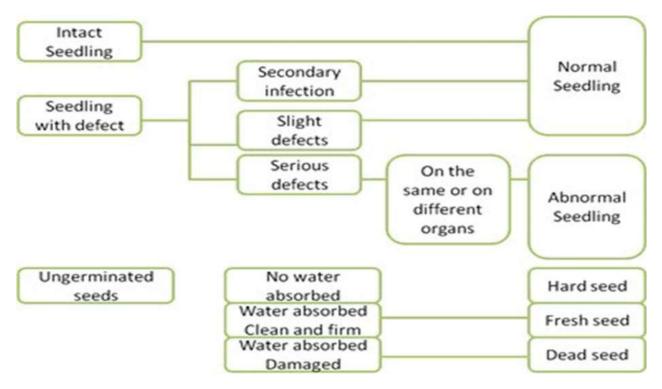
#### **Evaluation of germination test**

All seedlings have:

- a root system, which grows down into the soil;
- a seedling axis with the terminal bud, which grows up towards the light; and
- one, two or several cotyledons as lateral appendages to the seedling axis

The germination tests need to be evaluated on the expiry of the germination period, which varies according to the kind of seed. However, the seed analyst may terminate the germination test on or before the final count day or extend the test beyond the period depending on the situation.

First and second counts are usually taken in case of Top of Paper (TP) and Between Paper (BP) media; however, a single final count is made in case of sand tests. At the first and subsequent counts, only normal and dead seeds (which are source of infection) are removed and recorded. In evaluating the, germination test, the, seedlings and seeds are categorized into normal seedlings, abnormal seedlings, dead seeds, fresh ungerminated and hard seeds. The fresh ungerminated or hard seeds and abnormal seedlings should be evaluated at the end of germination. The stage of development of the essential structures must be sufficient to permit detection of any abnormal seedlings. It may also be necessary to remove the seed coat and separate the cotyledons in order to examine the plumule in species where essential structures are still enclosed at the end of the test.



**Normal Seedlings:** It is necessity to separate the normal seedlings, which are counted in the percentage germination, from any abnormal seedlings. Normal seedlings show the capacity for continued development into normal, plants when grown in good quality soil and under favourable conditions of water supply, temperature and light.

- **Intact seedlings** seedlings with all their essential parts well developed, complete, in proportion to each other and healthy.
- Seedlings with slight defects or deficiencies seedlings showing certain slight defects of their essential structures, provided they show an otherwise satisfactory and balanced development comparable to that of intact seedlings of the same test
- Seedlings with secondary infection A seedling that is decayed by fungi or bacteria is classified as normal, if it is evident, that the parent seed is not the source of infection (secondary infection), and if it can be determined that all the essential structures were present.

**Abnormal Seedlings:** Abnormal seedlings are those, which do not show the capacity for continued development into normal plants when grown in good quality soil and under favorable conditions of water supply, temperature and light.

**The causes of abnormalities include** mechanical injury of the embryo, Heat damage to the embryo, Chemical damage to the embryo, Deficiencies in the physiological make-up of the seed or embryo, Primary infection and disease of the seedling, and unknown causes.

If seeds are germinated in the laboratory under non-optimal conditions, the seedlings may show apparent abnormalities that are not due to damage or physiological disturbances of the embryo but to the germination conditions.

- In the restricted conditions of blotter envelopes, bent and twisted or even broken seedlings may be found;
- If the seeds are not planted in the correct orientation (radical-part of the seed downward) in rolled towels, the seedlings may have their roots initially growing upward and the coleoptiles or seedling axes growing downward;
- If the seedbed is too wet seedlings may show short retarded roots or decayed root tips;
- Chemicals in the germination medium, e. g. bleaches used in paper making, may suppress germination and/or cause abnormalities (mainly of the root).

In such cases, allowances must be made for the germination conditions and the sample should be retested, if necessary

The seedlings with the following defects shall be classified as abnormal:

*Damaged seedlings*; seedlings with any of the essential structures missing or so badly and irreparably damaged that balanced development cannot be expected e.g. seedlings with no cotyledons; seedlings with constrictions, splits, cracks or lesions which affect the conducting tissues of the epicotyl, hypocotyl or root; seedlings without a primary root of those species where a primary root is an essential structure, except for Pisum, Vicia, Lupinus, Vigna, Glycine, Arachis, Gossypium, Zea and all species' of Cucurbitaceae, when several vigorous secondary roots have developed to support the seedlings, in soil.

*Deformed seedlings:* Seedlings with weak development or physiological disturbances or in which essential structures are deformed or out of proportion; such as spirally twisted or stunted plumules, hypocotyls or epicotyls; swollen shoots and stunted roots; split plumules or coleoptiles without a green leaf; watery and glassy seedlings, or without further development after emergence of the cotyledons

*Decayed seedlings:* seedlings with any of their essential structures so diseased or decayed as a result of primary infection that normal development is prevented.

# Ungerminated seeds

**Hard seeds**: At the end of a germination test, hard seeds are counted and reported as such on the ISTA Certificate.

**Fresh seeds**: When 5 % or more of fresh seeds are believed to be present, their potential to germinate must be determined by dissection, tetrazolium or excised embryo. Those determined to have the potential to germinate are reported as fresh. Those determined not to have the potential to germinate are reported as dead.

After this determination, if there is any doubt as to whether the seed is fresh or dead, it must be classified as dead. If not already applied, measures must be taken to break dormancy if 5 % or more of fresh ungerminated seeds are found.

**Dead seeds**: Obviously dead (soft, mouldy) seeds are counted and reported as such on the ISTA Certificate. If it can be seen that a seed has produced any part of a seedling (e.g. the tip of the primary root) even though decayed at the time of assessment, it is counted as an abnormal seedling and not as a dead seed.

**Retesting:** The result of a test shall be considered unsatisfactory and shall not be reported and a second test shall be made by the same or an alternative method, under the following circumstances:

- When dormancy is suspected (fresh ungerminated seeds).
- When the result may not be reliable because of phytotoxicity or spread of fungi or bacteria.
- When there is difficulty in deciding the correct evaluation of a number of seedlings.
- When there is evidence of errors in test conditions, seedling evaluation or counting, a retest must be made using the same method or an alternative method.
- If a sample does not respond satisfactorily to the method selected, it will be necessary to retest it by one or more of the alternative methods.
- When the range for the 100-seed replicates exceeds the maximum tolerated range.
- When due to counting errors more than 5 seeds are lost or found during a germination test (i.e. ±1.25 % for a total of 400 seeds), then the test must be repeated. The result of a germination test must be reported as follows:
- the actual duration of the test (in days, excluding the period of special treatment or method used for promoting germination);
- the percentages, calculated to the nearest whole number of normal seedlings, hard seeds, fresh seeds, abnormal seedlings and dead seeds.
- If the result for any of these categories is found to be zero, it must be reported as '0'.
- If an applicant requests that the test be terminated when the sample reaches a pre determined germination percentage, before the final count, then only the percentage of normal seedlings is reported. The results of the other categories (abnormal seedlings, hard seeds, fresh seeds and dead seeds) must be reported as 'N', because they have not been determined

# Calculation and expression of results

The results are expressed as percentage by number. Germination rate is the average number of seeds that germinate over the five-day and 10-day time period.

Germination (%) = Number seeds germinated x 100 Number seeds on tray

# Methods to improve germination

For many species where hard seeds occur, some special treatment is essential. This treatment may be applied prior to the commencement of the germination test or, if it is suspected that the treatment may adversely affect non-hard seeds, it should be carried out on the hard seeds remaining after the prescribed test period. The treatments are as below:

**Soaking:** Seeds with hard seed coats may germinate more readily after soaking for up to 24-48 hours in water or for Acacia spp. after plunging seeds in about three times their volume of near boiling water until it cools. The germination test is commenced immediately after soaking.

**Mechanical scarification:** Careful piercing, chipping, filing or sand papering of the seed coat may be sufficient to break the dormancy condition. Care must be taken to scarify the seed coat at a suitable part in order to avoid damaging the embryo. The best site for mechanical scarification is that part of the seed coat immediately above the tips of the cotyledons.

Acid scarification: Treating with in concentrated Sulphuric acid ( $H_2SO_4$ ) is effective with some species (e.g.*Macroptilium* sp., *Brachiaria* sp., *Sesbania* sp.). The seeds are moistened with in the acid until the seed coat becomes pitted. Digestion may be rapid or take more than one hour, but the seeds should be examined every few minutes. After digestion, seeds must be thoroughly washed in running water before the germination test is commenced. In the case of *Oryza sativa* scarification may be performed by soaking the seed in one normal nitric acid (HNO3) for 24 hours (after preheating at 50 °C).

**Inhibitory Substances:** Naturally occurring substances in the pericarp or seed coat, which act as inhibitors of germination may be removed by washing the seeds in running water at a temperature of 250C before the germination test is made. After washing, the seeds should be dried back at a maximum temperature of 25°C (e.g.*Beta vulgaris*). Germination of certain species is promoted by removing outer structures such as involucre of bristles or lemma and palea of certain Poaceae (Gramineae).

**Disinfection of the seed:** For samples of *Arachis hypoagea* and *Beta vulgaris* only, a fungicide treatment may be applied before planting the seed for germination, when the seed lot is known not to have received such a treatment. When a fungicide pretreatment is used, the name of the chemical, the percentage of active ingredients and the method of treatment shall be reported on the certificate.

**Pre chilling:** In some seeds having physiological dormancy pre chilling is required for inducing germination. Replicates for germination are placed in contact with the moist substratum and kept at a low temperature for an initial period before they are removed to the temperature as shown in (ISTA Seed Testing Rules - Table 2). Agricultural and vegetable seeds are kept at a temperature between 5°C and 10°C for an initial period up to 7 days. Tree seeds are kept it a temperature between 3°C and 5°C, for a period, varying with the species, from 7 days to 12 months. In some, cases it may be necessary to extend the pre-chilling period or to re-chill. The pre-chilling period is not included in the germination test period but both the duration and the temperature should be reported on the analysis card

**Pre-drying:** The replicates for germination should be heated at a temperature not exceeding 400C with free air circulation for a period of up to 7 days before they are placed under the prescribed germination conditions. In some cases it may be necessary to extend the pre-drying period. Both the duration and the temperature should be reported on the Analysis Certificate.

# **Chemical Treatments:**

**Potassium nitrate (KNO<sub>3</sub>):** The germination substratum may be moistened with a 0.2% solution of KNO<sub>3</sub> (as indicated in ISTA Seed Testing Rules - Table 2). The substratum is saturated at the beginning of the test but water is used for moistening it thereafter. The use of this treatment should be noted on the analysis certificate.

The procedure for preparing solutions and soaking blotters is as follows:

i) Preparation of stock KNO<sub>3</sub>, solution (2%): Place 20 gms KNO<sub>3</sub> crystals in 1000 ml water shake until dissolved. This must be diluted before being used to soak blotters.

ii) Preparation of 0.2%  $KNO_3$  solution for soaking blotters: Add 90 ml water to 10 ml of stock solution

iii) Procedure for soaking blotters:

a. Take the blotters representing the sample and place into the prepared solution, (0.2%)-one at a time.

b. Turn blotters over in one movement, but ensuring that they are still free moving in the solution.c. Remove one at a time, in order of placing in solution and place on tray.

**Gibberellic acid (GA<sub>3</sub>):** Moisten the germination substratum with 50 ppm solution of GA, which can be prepared by dissolving 500 mg of  $GA_3$  in 1000 ml of water (Due to low solubility of  $GA_3$  in water; hence, it need to be dissolved small amounts of ethyl alcohol at first. Place the seed for germination under prescribed temperature conditions.

# DETERMINATION OF SEED MOISTURE CONTENT

As seed moisture and its management influences so many physiological seed quality parameters essential to seed quality. Seed moisture measurement appropriate to the purpose is needed in commerce and research. The optimum method for moisture testing depends upon:

- Chemical composition of seed
- Seed structure
- Moisture content level
- Degree of accuracy and precision required
- Constraints of time
- Technical expertise and cost

In order to measure the moisture content of seeds, methods can be broadly grouped in two categories:

- Direct method
- Indirect method

*Direct method:* Under this category, the seed moisture content is measured directly by loss or gain in seed weight.

These are:

- Desiccation method
- Phosphorus pentaoxide method
- Oven-drying method
- Vacuum drying method
- Distillation method
- Karl Fisher's method
- Direct weighing balance
- Microwave oven method

*Indirect method:* These are no so accurate; estimation is approximate, but convenient and quick in use. These are frequently used in seed processing plants, which measure physical parameters like electrical conductivity/ resistance of the moisture present in the seed.

The constant temperature oven drying method is the only practical method, approved by International Seed Testing Association (ISTA) and other organization to be used for routine seed moisture determination in a seed -testing laboratory.

*Constant temperature oven drying method:* The constant temperature oven drying method is broadly grouped into two categories:

- Low Constant Temperature Oven Method
- High Constant Temperature Oven Method

*Low constant temperature oven method:* This method has been recommended for seed of the species rich in oil content or volatile substances. In this method, the pre-weighed moisture bottles along with seed material are placed in an oven maintaining a temperature of  $103^{\circ}$  C. Seeds are dried at this temperature for  $17\pm1$  hr. The relative humidity of the ambient air in the laboratory must be less than 70 per cent when the moisture determination is carried out. At the end of prescribed period, cover the container, place in desiccator to cool for 30 to 45 minutes. It is recommended for seed species with high oil/volatile content *e.g.* onion, groundnut, mustard, chillies, soybean, cotton, linum, castor, til& tree species.

*High constant temperature oven method:* The procedure is the same as above except that the oven is maintained at a temperature of 130°C. The sample is dried to a period of four hours for*Zea mays*, two hours for other cereals and one hour for other species). In this method, there is no special requirement pertaining to the relative humidity of the ambient air in the laboratory during moisture determination.

# Essential equipment and supplies

- Constant temperature precision hot-air electric oven
- Weighing bottles/Moisture containers
- Desiccator with silica gel
- Analytical balance capable of weighing up to 1 mg
- Seed grinder/An adjustable grinding mill
- Tong
- Heat resistant gloves
- A brush/A steel brush

**Period of seed drying:** The prescribed period of seed drying shall be 17±1 hrs under low constant temperature. Seed drying period begins from the time oven returns to maintain the desired temperatures.

*Sample size:* The ISTA rules recommend that two replicates, each with 4 gm of seed be used for determination of seed moisture content. This seed sample weight may be modified to 0.2 to 0.5 gm per replicate, with precise weighing, for use in seed gene banks, to avoid unnecessary depletion of precious biological resources.

#### Procedure

- Seed moisture content determination should be carried out in duplicate on two independently drawn working samples.
- Weigh each bottle with an accuracy of 1 mg or 0.1 mg.
- First weigh the empty bottle/container with its cover.
- Grind the seed material (if need be, in seed species listed in Table 3), evenly using any grinder/grinding mill that does not cause heating and/or loss of moisture content.
- Mix thoroughly the submitted sample, using spoon, and transfer small portions (4 to 5 gm) of seed samples directly into weighing bottles/containers, by even distribution on bottom of the containers.
- Place the weighing bottles/containers in an oven, already heated to or maintaining the desired temperature, for the recommended period.
- At the end of seed drying period, weighing bottles/containers be closed with its lid/cover.
- Transfer the weighing bottles/containers to the desiccators having silica gel (self indicatingblue), to cool down for 40- 45 min.
- Weigh again the cooled weighing bottles/containers
- Calculate the seed moisture content.

#### Calculation of results

The moisture content as a percentage by weight (fresh weight basis) is calculated to one decimal place, by using of the formulae:

M2- M3

% Seed moisture content (mc) =----- x 100

M2- M1

Where

M1 = Weight of the weighing bottle/container with cover in gm

M2 = Weight of the weighing bottle/container with cover and seeds before drying

M3 = Weight of the weighing bottle/container with cover and seeds after drying

(Note: The seed moisture determination must be done in two replicates, with precise weighing i.e. up to three decimal places, using lightweight weighing bottles/containers.)

# **Reporting of results**

Seed moisture content is reported to the nearest 0.1% on ISTA analysis certificate. If the seed moisture content is determined using any moisture meter, the brand name and type of the equipment be mention on the analysis certificate, under column of other "other determinations". Reporting of range for which the moisture meter is calibrated is another requirement, on seed analysis certificate.

# **Benefits of Seed Testing:**

- 1. Seed testing has been utilized for the assessment of planting value of the seed lots for maximizing agriculture production.
- 2. Seed testing is considered as the hub of any successful seed programme.
- 3. It plays significant role in the conservation of plant genetic resources and plant quarantine.
- 4. Facilitate seed quality control operations through seed certification/ law enforcement.

# Constraints in Seed testing in India:

- Laboratory equipment, trained personnel and working conditions are important components in seed testing. However, some the SSTLs are not well equipped in terms of modern infrastructure and other facilities, which lead to faulty results.
- Variation between the results of state seed testing laboratories and CSTL w.r.t. referee samples
- Lack of regular training opportunities for the seed testing personnel
- Non-uniformity in SOPs leads to approval of poor-quality seed lots and rejection of good quality seed lots.
- Seed health is very important component of seed quality which has not been given due consideration.

# **Useful References:**

- 1. Agrawal P.K. (1993). Handbook of Seed Testing, Dept. of Agriculture and Cooperation, Ministry of Agriculture, Govt. of India, New Delhi, pp: 340.
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- 3. <u>https://www.seedtest.org/en/home.html</u>

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# **PPV& FR Act, 2001; An Overview** Prof. (Dr.) C.P. Sachan Ex. Nodal Officer- DUS Test Project CSAU Ag. Tech. Kanpur

The PPV&FR act 2001 fulfilled a mandatory requirement for India under the TRIPS, namely , enacting a legislation for effective protection of plant varieties (PVP) either by way of patent or a sui generis system stipulated under article 27.3(b) of the TRIPS agreement, a move welcome by Indian agriculture and seed sector. Since the public sector alone could no longer meet the crop improvement research and seed requirements of agriculture in India and the private industry had begun to play a substantial role in plant breeding .thePPV&FRAct is a valuable instrument, which confer important right on the registry as against the right of others . Significant responsibility lies on the PPV&FR authority and also on the central government to ensure that there is ample clarity and and no ambiguity in the enactment, rules and procedures, so that the PPV&FR registry perform its function effectively to achieve the objectives enshrined in the PPV&FR Act . Indian legislation is not only in conformity with International Union for the Protection of New Varieties of Plants (UPOV), 1978, but also have sufficient provisions to protect the interests of public sector breeding institutions and the farmers. The legislation recognizes the contributions of both commercial plant breeders and farmers in plant breeding activity and also provides to implement TRIPs in a way that supports the specific socio-economic interests of all the stakeholders including private, public sectors and research institutions, as well as resourceconstrained farmers.

## Aims & Objectives

- To establish an effective system for the protection of plant varieties, the rights of farmers and plant breeders and to encourage the development of new varieties of plants.
- To recognize and protect the rights of farmers in respect of their contributions made at any time in conserving, improving and making available plant genetic resources for the development of new plant varieties.
- To accelerate agricultural development in the country, protect plant breeders' rights; stimulate investment for research and development both in public & private sector for the development new of plant varieties.
- Facilitate the growth of seed industry in the country which will ensure the availability of high quality seeds and planting material to the farmers.

## **Rights under the Act**

- **Breeders' Rights** : Breeders will have exclusive rights to produce, sell, market, distribute, import or export the protected variety. Breeder can appoint agent/ licensee and may exercise for civil remedy in case of infringement of rights.
- **Researchers' Rights** : Researcher can use any of the registered variety under the Act for conducting experiment or research. This includes the use of a variety as an initial source of

variety for the purpose of developing another variety but repeated use needs prior permission of the registered breeder.

• **Farmers' Rights**: The Protection of Plant Varieties and Farmers' Rights Act (PPV&FR Act) seeks to address the rights of plant breeders and farmers on an equal footing. It affirms the necessity of recognizing and protecting the rights of farmers with respect to the contribution they make in conserving, improving and making Plant Genetic Resources (PGR) available for the development of new plant varieties.

The PPV&FR Act recognizes the multiple roles played by farmers in cultivating, conserving, developing and selecting varieties. With regard to developing or selecting varieties, the Act refers to the value added by farmers to wild species or traditional varieties/ landraces through selection and identification for their economic traits. Accordingly, farmers' rights encompass the roles of farmers as users, conservers and breeders. Farmers are granted nine specific rights, which are as under:

## Right 1: Access to seed [Section 39(1)(iv)]

Farmers are entitled to save, use, sow, re-sow, exchange, share or sell their farm produce, including seed of protected varieties, in the same manner as they were entitles to before the coming into force to the PPV&FR Act. However, farmers are not entitled to sell branded seed of a variety protected under this Act. Farmers can use farm saved seed from a crop cultivated in their own.

## Right 2: Benefit sharing [Section 26]

Plant breeders and legal entities including farmers who provide Plant Genetic Resources (PGR) to breeders for developing new varieties shall receive a fair share of benefit from the commercial gains of the registered varieties. Out of all the national plant variety protection laws enacted since 2001, the PPV&FR Act is the first that integrates a provision for access and benefit-sharing (ABS) along with Plant Breeder's Rights (PBRs). Accession of the genetic resource used in breeding is permitted under the Biological Diversity Act, 2002. However, the PPV&FR Act requires a breeder to make a sworn declaration on the geographical origin of the genetic resources used in the pedigree of the new variety, and its accession.

## Right 3: Compensation [Section 39(2)]

Registered seed must be sold with the full disclosure of their agronomic performance under recommended management conditions. When such seed is sold to farmers but fails to provide the expected performance under recommended management conditions, the farmer is eligible to claim compensation from the breeder through the intervention of the PPV&FR Authority.

## Right 4: Reasonable seed price [Section 47]

Farmers have the right to access seed of registered varieties at a reasonable and remunerative price. When this condition is not met, the breeder's exclusive right over the variety is suspended under the provision concerning compulsory licensing, and the breeder is obligated to license the seed production, distribution and sales of the variety to a competent legal entity. Most of the laws for plant variety protection have provisions on compulsory licensing of protected varieties to ensure adequate seed supply to farmers, and several of them also use unfair pricing as grounds for compulsory licensing.

**Right 5**: Farmers' recognition and reward for contributing to conservation [Section 39(i)(iii) & Section 45(2)(C)]

Farmers who have been engaged in PGR conservation and crop improvement, and who have made substantial contributions in providing genetic resources for crop improvement, receive recognition and rewards from the national gene fund. The gene fund receives resources from the implementation of the Act, which in turn are complemented by contribution from national and international organizations. The expenditures of the fund are earmarked to support the conservation and sustainable use of PGR, and in this way it can be considered to be a national equivalent to the global benefit-sharing fund operating within the International Treaty on Plant Genetic Resources for Food and Agriculture.

Since 2007, the Plant Genome Saviour/Community awards, associated with the national gene fund, has been rewarding farming communities and individual farmers for their contribution to insitu and on farm conservation to the selection of PGR. The Authority in consultation with Government of India, has established five Plant Genome Saviour Community Awards of Rs 10 Lakh each along with citation and memento to be conferred every year to the farming communities for their contribution in the conservation of Plant Genetic Resources.

In accordance with the Protection of Plant Varieties and Farmers' Rights (Recognition and Rewards from the Gene Fund) Rules, 2010 the Authority also setup ten Plant Genome Saviour Farmer Reward of Rs 1 Lakh each with citation & memento and also twenty Plant Genome Saviour Farmer Recognition annually from 2012-13 to the farmers engaged in the conservation of the Genetic Resources of the landraces and wild relatives of economics plants and their improvement through selection and preservation.

## Right 6: Registration of farmers' varieties [Section 39(1)(iii)]

The PPV&FR Act allows for the registration of existing farmers' varieties that fulfill requirements for distinctness, uniformity, stability and denomination, but does not include that of novelty. This right provides farmers with a one-off opportunity for a limited period of time, from the moment when a crop species is species is included in the crop portfolio under the PPV&FR Act for registration. Once registered, these varieties are entitled to all PBRs.

# Right 7: Prior authorization for the commercialization of essentially derived varieties [Section 28 (6)]

When farmers' varieties, whether extant or new, are used by a third party as source material for the development of an essentially derived variety, the farmers need to provide prior authorization for its commercialization. Such a process can allow farmers to negotiate the terms of authorization with the breeder, which may include royalties, benefit-sharing, etc.

Right 8: Exemption from registration fees for farmers [Section 44]

Under PPV&FR Act, farmers have the privilege of being completely exempted from payment of any kind of fees or other payments that are normally payable for variety registration; tests for distinctness, uniformity and stability (DUS), and other services rendered by the PPV&FR Authority; as well as for legal proceedings related to infringement or other causes in courts, tribunal, etc.

## Right 9: Farmer protection from innocent infringement [Section 42]

If a farmer can prove before court that he or she was not aware of the existence of any rights at the time of an infringement on any such rights, as detailed in the PPV&FR Act, he or she will not be charged. This provision is made in consideration of the centuries-old unrestrained rights that the farmers had over the seed of all varieties, the novel nature of the PPV&FR Act and the poor legal literacy of farmers.

#### Implementation of the Act

To implement the provisions of the Act the Department of Agriculture, Cooperation and Farmers Welfare, Ministry of Agriculture and Farmers Welfare established the Protection of Plant Varieties and Farmers' Rights Authority on 11" November, 2005. The Chairperson is the Chief Executive of the Authority. Besides the Chairperson, the Authority has 15 members, as notified by the Government of India (GOI). Eight of them are ex-officio members representing various Departments/ Ministries, three from SAUs and the State Governments, one representative each for farmers, tribal organization, seed industry and women organization associated with agricultural activities are nominated by the Central Government. The Registrar General is the ex-officio Member Secretary of the Authority.

#### **General Functions of the Authority**

Registration of new plant varieties, essentially derived varieties (EDV), extant varieties;

Developing DUS (Distinctiveness, Uniformity and Stability) test guidelines for new plant species;

Developing characterization and documentation of varieties registered;

Compulsory cataloging facilities for all variety of plants;

Documentation, indexing and cataloguing of farmers' varieties;

Recognizing and rewarding farmers, community of farmers, particularly tribal and rural community engaged in conservation and improvement;

Preservation of plant genetic resources of economic plants and their wild relatives;

Maintenance of the National Register of Plant Varieties and

Maintenance of National Gene Bank.

Registration of varieties

A variety is eligible for registration under the Act if it essentially fulfills the criteria of Distinctiveness, Uniformity and Stability (DUS). The Central Government issues notification in official Gazettes specifying the genera and species for the purpose of registration of varieties. So far, the Central Government has notified 157 crop species for the purpose of registration. To access the list, The PPV&FR Authority has developed "

#### Fees for registration

Application for registration of plant varieties should be accompanied with the fee of registration prescribed by the Authority. Fee for registration for different types of variety is as under:

S.No	Types of Variety	Fees for Registration
1	Extant Variety notified under section 5 of the Seeds Act, 1966	Rs 2000/-

2.	New Variety/Essentially Derived Variety (EDV)/ Extant Variety about which there is common knowledge (VCK)	Individual Rs. 7000/- Educational Rs.10000/- Commercial Rs.50000/-
3.	Farmers Varieties	No Fee

The Registration of a variety is renewable subject to payment of annual and renewal fee as notified in the Plant Variety Journal of India of the Authority and Gazette of India dated 15.06.2015.

#### **DUS Test Centers**

Authority has notified DUS test Centers for different crops with a mandate for maintaining and multiplication of reference collection, example varieties and generation of database for DUS descriptors as per DUS guidelines of respective crops. To access the list of DUS test Centers.

#### Certificate of Registration

The certificate of registration issued will be valid for nine years in case of trees and vines and six years in case of other crops. It may be reviewed and renewed for the remaining period on payment of renewal fees subject to the condition that total period of validity shall not exceed eighteen years in case of trees and vines from the date of registration of the variety, fifteen years from the date of notification of variety under the Seeds Act, 1966 and in other cases fifteen years from the date of registration of the variety.

#### **Benefit Sharing**

The benefit sharing is one of the most important ingredients of the farmers' rights. Section 26 provides benefits sharing and the claims can be submitted by the citizens of India or firms or non-governmental organization (NGOs) formed or established in India. Depending upon the extent and nature of the use of genetic material of the claimant in the development of the variety along with commercial utility and demand in the market of the variety breeder will deposit the amount in the Gene Fund. The amount deposited will be paid to the claimant from National Gene Fund. The Authority also publishes the contents of the certificate in the PVJI for the purpose of inviting claims for benefits sharing.

#### **Rights of Community**

It is compensation to village or local communities for their significant contribution in the evolution of variety which has been registered under the Act.

Any person/group of persons/governmental or non-governmental organization, on behalf of any village/local community in India, can file in any notified centre, claim for contribution in the evolution of any variety.

#### **Convention countries**

Convention country means a country which has acceded to an international convention for the protection of plant varieties to which India has also acceded or a country which has law of

protection of plant varieties on the basis of which India has entered into an agreements for granting plant breeders' rights to the citizen of both the countries. Any person if applies for the registration of a variety in India within twelve months after the date on which the application was made in the convention country, such variety shall, if registered under this Act, be registered as of the date on which the application was made in convention country and that date shall be deemed for the purpose of this Act to be the date of registration.

Plant Varieties Protection Appellate Tribunal

There is transitory provision by which it is provided that till the PVPAT is established the Intellectual Property Appellate Board (IPAB) will exercise the jurisdiction of PVPAT. Consequently the Plant Varieties Protection Appellate Tribunal (PVPAT) has been established by appointing Technical Member. All orders or decisions of the Registrar of Authority relating to registration of variety and orders or decisions of the Registrar relating to registration as agent or licensee can be appealed in the Tribunal. Further, all orders or decisions of Authority relating to benefit sharing, revocation of compulsory license and payment of compensation can also be appealed in the Tribunal. The decisions of the PVPAT can be challenged in High Court. The Tribunal shall dispose of the appeal within one year.

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# Identification of Objectionable Weed Plants and Seeds in Reference to Seed Certification Standard and their Management

S. K. Chakrabarty

**Principal Scientist** 

Division of Seed Science and Technology, ICAR-IARI, New Delhi-110012

Increase in seed trade has promoted increased transfer of seeds of crop varieties from one region and country to other. Association of seeds of other varieties and weeds in a seed lot is a well known. It not only reduces the quality of seed but its nature. Weeds disseminate to new habitat through the crop seeds. It is also common that weeds produce enormous number of seeds every generation.

Before seeds are marketed within or to outside the country, its testing is mandatory. In seed testing, it is essential to test the seed samples for physical purity including the presence of weed seeds. Through seed testing, contamination by weeds and other crop seeds are avoided. In modern seed testing, authentic identification of weed seeds in crop varieties is vital.

Government of India took several steps including framing and implementing seed legislations for regulation of quality of being used for further multiplication nd cultivation. The Seeds Act was enacted in 1966 and ensuing Seed Rules framed under The Seeds Act, were notified in 1968 and since then several amendments have been carried out in Seeds Act and Seed Rules from time to time for regulation of seed. Government of India has notified Indian Minimum Seed Standards [IMSCS] for ensuring the seed quality viz., Genetic purity, physical purity, germination, moisture, and seed health. The physical purity from seed testing point of view refers to physical or mechanical purity of seed lot. The results of purity analysis reflect the physical quality status of seed lot. These results are of great significance to seed processors and seed certification or seed law enforcement agencies (to judge whether the seed lot confirms to the prescribed standards or not). As per Indian Minimum Seed Certification Standards the physical purity of the seed lot consists of (i) pure seed (%) (ii) maximum inert matter (%) (iii) maximum other crop seeds (no/kg) and (iv) maximum weed seeds (no/kg) including maximum objectionable weed seeds (no/kg). As per ISTA Rules the working sample is separated into three components i.e., pure seeds, other seed, and inert matter. The percentage of each part is determined by weight. Other seeds are undesirable because:

- They are a source of weed introduction to new areas
- Once established, weeds are difficult to eradicate
- High contamination will lower purity content
- Risk of rejection due to lower seed standards

#### Need for accurate identification of weed seeds

Weed seeds as concomitant admixtures always affect physical purity of seed lots.Since the inception of seed testing, correct seed identification has been the basis for purity analysis. Seed collection, seed illustrations and descriptions of seed morphology have been valuable tools in the identification of unknown seeds. The experiences in the field of establishment and maintenance of

seed collections were presented at International Seed Testing Association [ISTA] workshops in Wageningen [Jensen, 1979d] and Budapest [Jensen, 2000]. Accurate identification of seeds, both crop seeds and weed seed contaminant is necessary for correct labeling of seed moving in commercial channels. There are many other group of plants in which seeds of one species may closely resemble to those of another species, some cases those may be undesirable or noxious weed seed, so seed analyst must be able to analyze and evaluate these structures. Internationally weed seed identification and its application in field of seed science and technology is administered by ISTA. In ISTA, identification of weed seeds, developing and adopting tools for physical purity analysis of seed lots is governed by ISTA Purity Committee. Working group on seed identification was initiated in the year 1977. It consisted of collection and identification of crop and weed species through seed morphology. Many working programmes were also initiated within the purity committee for preparation of universal list of weed species, in 2004 and 2006 two more working programmes were also initiated for identification of crop and weed seeds and documentation of digital images of weed species to aid identification of seeds. Identification of weed seeds in the seed lot requires a detailed knowledge of gross seed morphology of disseminule produced by plants. Identification of weed seeds is extremely important for seed quality analysis for the issue of Orange International seed lot certificate and Blue International seed sample certificate and also for routine seed quality analysis, seed certification etc., and for ISTA accreditation of seed testing laboratory.

#### Seed identification

It is sometime a difficult job to identify a seed of a crop or weed species. An in-depth knowledge of botany of a plant as well as its seed is necessary, for correct identification of a particular species. In systematic botany or taxonomy the closely related or similar type of plants are grouped into a single category. These groups are: family, genus, species etc. In seed identification the particular seed in question must be identified up to the species level.

The seed, a mature ovule consists of an embryo a protective covering and stored food as endosperm. The identification of seed is usually by comparison, comparing the seeds with a mental image of what something should be, with specimens in a reference collection or with illustration of seeds. In most cases, the useful clues for the identification of seeds come from the following characters:

- 1. The size, shape and colour of seeds
- 2. The nature, arrangement and pattern of markings that is lines, ridges, pits, projection on the seed surface
- 3. The shape and position of the attachment scar
- 4. The presence of wings, hairs or scales, spines etc
- 5. The internal structure, position and size of the embryo, presence or absence of the endosperm

Seed keys are developed based on characters pertaining to family, genus, and species. Once the seed is characterized for a particular family, identification of the seed could easily be made by studying the above-mentioned seed characters. Quite often it is difficult to identify the seeds as such. In such situation, growing it to a plant could aid in identification of seed. The original seed sample of the species is always helpful in identification of unknown unconventional crop and weed seeds.

#### Seed characteristics of some common families

- 1. *Gramineae:* seed unit is a caryopsis, a fertile floret a spikelet or a spike. The embryo lies on outside of the endosperm and visible near the base of caryopsis on dorsal side.
- 2. *Leguminosae:* Seeds vary greatly in size, shape and surface characters. The fruit may be one seeded in several-seeded pod.

#### a) Mimosoideae and Caesalpinodeae

- i) The seeds are elongate broad and flattened, the two faces being plane or only rounded, colour is varied from black to white and yellow
- ii) The hilum is very small, unspecialized and located at one end of the seed.

## b) Papilinoideae

- i) The seeds vary greatly in size, shape, colour and location of hilum and chalaza.
- ii) In hilum, there is a fine longitudinal groove or slit down the middle. The area may be minute, as in some of the clovers, or may be large enough to be seen without magnification as in vetch.
- iii) In some species the hilum is obscured by a persisting layer of corky tissue, as in cowpea and beans.
- 3. *Cruciferae*: The seeds unit may be a true seed, in indehiscent pod or a segment of a pod.
- a) The seeds are mostly spherical, or sometimes slightly flattened.
- b) The surface has reticulum or netting or lines or ridges.
- c) The seed surface is covered with microscopic pits. These pits are usually covered with a whitish film, giving the appearance of white spots on the surface.
- 4.*Polygonaceae:* The fruit or so-called seed is an achene which is three angled orflattened.The outer hull (pericarp) is hard, brown and glossy.

5.*Chenopodiaceae*: The seeds are flattened, circular or obovate in shape.

a) The embryo is either in the form of a ring or horseshoe.

6.*Caryophyllaceae*: The seeds are black or brown, thick and flattened.

- a) The scar lying on the edge.
- b) The surface is roughened by tubercles of various types which are arranged in definite pattern.
- 7. *Euphorbiaceae:* Seeds vary greatly in size, shape and surface configuration.
- a) The scar is a flattened area at the base. In some species the scar is obscured by caruncle (whitish corky outgrowth).
- b) Distinctive feature of the seed in this family is the presence of prominent raphe.
- 8. *Solanaceae:* The seeds are orbicular, oval or ovate. They are more or less flattened and may be thick or thin.
- 1. The embryo is curved with an abundant endosperm.

- 2. The seed surface may be smooth, or variously configured with a reticulum, broken lines or pits.
- 9. *Compositae:* The seed unit is an achene, which is an indehiscent, one seeded fruit. The top of the achene is usually depressed. In many species there is a fringe of fine bristles or scales around the outer rim.

#### Characteristic of some common weeds

#### Family: Caryophyllaceae

*Spergula arvensis*: The seed 1-15 mm diameter lens shaped, dull black, thin, flattish with winged. Embryo, Linear, 'U' shape without endosperm.

#### Family: Chenopodiaceae

*Chenopodium album* (Bathva): The seed is circular, flat, and round; diameter 1-11/2mm, colour black, smooth and shiny surface.

*Chenopodium murale* (Bathva): Similar to *C. album* but slightly bigger in size and dull in appearance.

#### Family:Convolvulaceae

*Convolvulus arvensis* (Field weed): The seed colour, dull grayish brown, length, 4 to 4½ mm; surface roughened with fine tubercles or short wavy lines. Back side convex and lateral plane, scar: inverted 'U' shape and at right angles to the seed's long axis.

*Ipomea hederacea*: The seed diverse in shape (trigonous wedge, two inner faces are equal): size (lanceolate, ovoid to globose surface; smooth and colour: brown black. Scar: horseshoe shape and usually parallel to long axis.

#### Family:Poaceae

*Avena fatua* (Wild oat): The seed consists of mature floret, narrowly cylindrical, tapering at apex, bears a twisted and bent dorsal awn, ventral side flat with fine grooves; colour: grey, brown or black, yellow to white.

*Panicum*species(grasses): The seed unit consists of one seeded spikelet. The grain surrounded by glumes (thin and papery). Lemma and Palea (hard, smooth and shiny, size: 1<sup>1</sup>/<sub>2</sub> to 2<sup>3</sup>/<sub>4</sub> mm usually lance shape).

*Setaria etalica*: The seed unit consists of one seeded spikelet. The grain surrounded by glumes (thin, papery, and smooth). Lemma and Palea (hard, smooth, and shiny)

## Family:Liliaceae

*Asphodelus tennuifolius* (wild onion): The seed 1<sup>1</sup>/<sub>4</sub> long, flattened elliptical three angled (sharp) acute and black (crustaceous) testa.

#### Family: *Papaeraceae*

*Fumaria parviflora*: Fruit very small, globose, one seeded, indehiscent nutlet, rugose when dry and rounded at the top with two pits, color usually green.

## Family: Papillionaceae

*Medicago sativa* (lucerne): The seed roughly oval (scar lies in broad indentation near one end or kidney shape twisted the alongaxes (scar lies in middle of a distinct notch). Colour greenish yellow or light brown, length 1<sup>1</sup>/<sub>2</sub> mm and width 2<sup>1</sup>/<sub>2</sub> mm to 3mm.

*Melilotus alba* (white sweet clover): The seed is identified by size (bigger length about  $2\frac{1}{2}$  mm and width  $1\frac{1}{2}$  mm), shape oblong to oval and translucent in appearance), and colour (golden yellow to light brown). Scar lies in shallow indentation near top.

#### Family: Polygonaceae

*Rumex*sp (wild spinach): Seed three sided acute as both ends, brown, spinning segments if present with long, fine teeth on the margins.

# Seed Standards [weed seeds (max.)] as per Indian Minimum Seed Certification Standards in field crops:

Referred table clearly depicts role of correct identification of weed seeds for certification vis-à-vis quality assurance purpose.

Сгор	Total wee	ed seeds	Objectiona seec		Remarks (Objectionable
	Foundation	Certified	Foundation	Certified	weed seeds)
Barley	10/kg	20/kg			
Paddy	10/kg	20/kg	2/kg	5/kg	Wild Rice
					(Oryza sativa L. var. fatua Prain)
Wheat	10/kg	20/kg	2/kg	5/kg	Convolvulus arvensis
					Phalaris minor
Maize	None	None	-	-	-
Sorghum	5/kg	10/kg	-	-	-
Pearl millet	10/kg	20/kg	-	-	-
Chickpea	None	None	-	-	-

Black gram	5/kg	10/kg	-	-	-
Green gram	5/kg	10/kg	-	-	-
Pigeonpea	5/kg	10/kg	-	-	-
Castor	None	None	-	-	-
Groundnut	None	None	-	-	-
Mustard	10/kg	20/kg	5/kg	10/kg	Argemone mexicana
Safflower	5/kg	10/kg	None	None	Carthamus oxyacantha
Soybean	5/kg	10/kg	-	-	-
Sunflower	5/kg	10/kg	None	None	Orobanche cumana
Cotton	5/kg	10/kg	-	-	-
Berseem	10/kg	20/kg	5/kg	10/kg	Chicorium intybus
Lucerne	10/kg	20/kg	5/kg	10/kg	Cuscuta spp.
Napier grass (slips)	-	-	None	None	Cirsium arvense Cuscuta spp. Sorghum halepense Agropyron repens Convolvulus arvensis
Oats	10/kg	20/kg	2/kg	5/kg	Avena fatua

#### Tools and resources for seed identification

An old Chinese proverb "One picture is worth of thousand words" is especially applicable to the identification of seeds. Enormous literature in the form of atlas, compendium and handbook are available for identification enumerating species of temperate region, but very miniscule information in this regard available for tropical species particular to Indian sub-continent. Digital weed seed atlas consisting of digital seed images and descriptions of species based on morphological keys will effectively supplement seed analyst for easy identification and significantly improves the efficiency of the seed testing laboratories. Seeds of some kind of plants are sufficiently distinctive that they are not easily confused with those of other kinds and their identification poses no problems. There are many groups of plant, in which seeds of one species

may closely resemble with seeds of other species. In some cases one of these may be a crop plant and the other an undesirable or designated weed species. The seed analyst must be able to analyze and evaluate the structures of such seeds in relation to those of other similar species; in such scenario digital weed seed atlas would become handy.

A guide with colour photos of 200 species including agricultural, horticultural and weed species published by National Institute of Agricultural Botany (NIAB), United Kingdom [Jones et al. 2004], a descriptive and illustrated seed-book on 175 weeds were worked out by French Group for Study and Control of Varieties and Seeds (GEVES), France in 2004. More than 1400 images of seeds and fruits have been made available online by the efforts of Dr. Arnold Larsen, Colorado state university, USA; the website [www.seedimages.com] includes colour images of seeds as well as descriptions and keys for identification of described species. Professor Miller McDonald and his staff at Ohio State University, USA, have placed number of seed images of cultivated and weed species with useful basic and advanced seed identification quiz. Dr. John H. Wiersema and his colleagues at Germplasm Resource Information Network (GRIN), United States Department of Agriculture/Agricultural Research Service have added over 3,800 seed/fruit or embryo images or drawings to GRIN, with plans to add another 5,000 in the future [Jensen, 2008]. A quick start weed guide (expert system) with seed identification keys which makes use of 21 externally visible characters (e.g., colour, shape, size and texture) using a computerized database has been developed to identify weed seeds prohibited/restricted by the Australian Quarantine and Inspection Service [Gupta et al., 2005]. Weed seed atlas was compiled depicting digital images of common agricultural weed seeds of USA with descriptions of seed characteristics for its effective usage in seed quality testing laboratories [Schuler, 2009]. In India weed seeds have been collected from crop fields, seed processing plants, marketplaces etc., characterized and identified (Chakrabarty and Tomar, 2005).

## 1. Seed herbarium or reference collection

A seed herbarium is a standardized collection of seed specimens that has a practical value to the seed analyst in aiding identification and for visual comparison of seeds. These seeds are collected by naturalists, classified, and stored. The easiest means to arrange the seed herbarium is to place it in alphabetical order. Families are arranged alphabetically. Genera within the family are then placed in alphabetical order. And then the species within each genera arranged by the alphabet. This arrangement makes it easy to retrieve specimens but gives no clue to the relationship between the specimen and other species, genera, or families

#### 2. ISTA reference

- ISTA International Rules for Seed Testing
   <u>https://www.seedtest.org/en/international-rules-for-seed-testing-\_content---1--1083.html</u>
- ISTA List of Stabilized Plant Names (7th Edition) <u>https://www.seedtest.org/upload/cms/user/ISTAListofStabilizedPlantNamesed.75.pdf</u>
- ISAT Universal List

https://www.seedtest.org/en/universal-list\_content---1--1446.html

 ISTA Handbook on Pure Seed Definition https://www.seedtest.org/en/productdetail------137.html

#### 3. Seed reference books

- Handbook on Tropical Species
- NIAB Seed Identification Handbook
- Identification of Crop and Weed Seeds
- Illustrated Taxonomy Manual of Weed Seed
- The Encyclopedia of Arable Weeds
- The illustrated guide to Weed Seeds of New Zealand
- The Digital Seed Atlas of the Netherlands
- A Manual for the Identification of Plant Seeds and Fruits
- Seed Purity and Taxonomy
- 4. Online resources
- USDA National Germplasm System
   <u>https://npgsweb.ars-grin.gov/gringlobal/taxon/abouttaxonomy</u>
- Seed Identification Guide (SIG) https://www.idseed.org/seedidguide/
- Colorado State University (requires paid subscription) <u>https://www.seedimages.com/Default.aspx</u>
- The OHIO State University
   <u>https://www.oardc.ohio-state.edu/seedid/</u>
- USDA Seed Images
   <u>https://frontrangeseedanalysts.weebly.com/usda-plates.html</u>
- Flower Seed Images
   <u>https://frontrangeseedanalysts.weebly.com/flower-seed-images-frsa-1995.html</u>
- Digital Plant Atlas
   <u>https://www.plantatlas.eu/</u>

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# VARIETAL RELEASE & NOTIFICATION SYSTEM IN INDIA

Prof. (Dr) C.P. Sachan CSAU Ag. & Tech Kanpur

#### Introduction

India is a fast-growing economy and agrarian country. Almost 65 percent of the Indian population depends on agriculture and its allied sectors to obtain employment and sustain livelihood. The seed is considered as a basic and key input in agriculture. High-quality seed production was the major concern in the Indian subcontinent till the 1960s.. In order to meet the food and nutritional demand of population and to *become* self-reliant in food grain production, Indian Government established All India Coordinated Crop Research Projects (AICCRPs) to produce a large number of varieties with assured seed quality in all major crops.

The production of high-quality seeds was one of the pillars to change the position of Indian agriculture into the new world order. The ultimate intention was to introduce the newly evolved high yielding cultivars to the resource-poor farmers for broad- spectrum cultivation in the area of their adoption.

Under such circumstances the Government of India ac1<now1edged seed an essential commodity under the Essential Commodities Act, 1955. On October 1964, Varietal Release System (VRS) came into existence with the formation of the Central Variety Release Committee (CVRC) at the national level, and State Variety Release Committees (SVRCs) at each state level. A Central Seed Committee (CSC) a statutory body was established under the Ministry of Agriculture, Cooperation and Farmers Welfare provided in the Seeds Act, 1966.

The functions of the CVRC were taken over by the CSC in 1969 to ensure the quality of seeds on sale and notification of the varieties. To perform the function at central level to release/notification, provisional notification and de-notification of cultivars, CSC constituted a Central Sub- Committee on Crop Standards, Notification & Release of Varieties for both Agricultural and Horticultural Crops, while to perform similar functions at state level, State Seed Sub-Committee (SSSC) was constituted.

#### Development of plant genetic material

Entries (pure lines/open pollinated varieties/composites/synthetics/hybrids etc.) are developed by the concerned plant breeders/agencies through breeding programs for the benefit of humankind. Different conventional (Introduction, selection, hybridization mutation & polyploidy followed by selection etc.) and advanced (tissue culture-based techniques like somaclonal variation, anther and pollen culture, marker assisted breeding, transgenic or genome editing techniques) breeding methods are being used by the different agencies to generate elite material for high yield potential, nutritional quality and other associated traits. Developed elite materials are being tested by the concerned plant breeder/s at their research station for three to four years in replications for stability and selected superior cultivars enter into the All India coordinated crop improvement projects (AICCIPs) trials for further testing in multi-environments across the country.

## All India Coordinated crop Improvement projects system of varietal testing

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First AICCIP was started in way back of 1957 by ICAR on maize crop for systemic testing of entries and for release of high yielding new maize varieties. In general, the three-tier system (IVT-AVTI-AVTII) of multi-location evaluation is used for three years except perennial fodder crops (requires four years-one for crop establishment and three for evaluation) in India. Multi locational trials are conducted by the Project Coordinator (PC)/ Project Director (PD) of AICCIPs with the help of concerned principle investigators. All AICCIP trials are well organized, systemic and conducted through a uniform testing procedure across the centers as per crop standard. It is a powerful system to screen large number of entries and recommend well-tested, superior, and adapted new cultivars to the end users.

#### Essential Parameters of a entry for Testing under AICCIP

1.Station trial or preliminary yield trial-Concerned plant breeder must perform station or regional trial and proposed entry must have undergone censorious evaluation process or screening (insect pests and diseases). Crop based quality parameters and tolerance to key abiotic stresses are also to be screened as per the requirement. Pre-coordinated trial data on yield, trait stability and other related agronomic traits must be available to the PC/PD in support of the relevance of entry.

2. The entry must have a high degree of genotypic stability, phenotypic uniformity, germination percentage and physical purity (as per the minimum seed certification standards).

3. The entry must have few distinct diagnostic traits which make it different to all remaining varieties. These distinct traits help to identification of variety during legal infringement (DUS testing).

4.All the information related to the development of entry *i.e.* parentage or pedigree should be available to the PC/PD by the concerned plant breeder/ agency. If the performance of entries are same in the coordinated trials, then preference will be given to the variety which has been developed by the using of diverse parents in breeding program.

5.Private companies can enter their material into the coordinated trial system as similar to other agencies but have to pay the prescribed fee for their entries as per guideline of the Government of India.

#### Prevailing Testing System(AICCP)

The AICCIP centers for various crops are located at ICAR institutes or State Agricultural Universities (SAUs) or other volunteer centers recommended by AICCIP workshop based on covered crop area, adaptability, and agro-climatic condition etc. Following steps are involved .

#### Initial varietal trial (IVT)

The time duration of the initial varietal trial (IVT) is one year. All the entries, which were superior to their respective station trials, would be introduced into the IVT. These entries would be used for multi-location trials along with checks. In general, three checks (national, zonal and local checks) are being used for efficient evaluation of entries across the centers. These checks cannot be replaced after the IVT. Maintenance of genetic purity, germination and physical purity of new material are the prime objectives of the concern plant breeder/agency. The IVT trials are conducted in such a manner that minimum difference of yield (5-10%) and other ancillary traits can be measured. The cultural practice(seed rate, date of sowing, row to row and plant to plant spacing; weed, fertilizer and water management etc) shall be strictly followed by the IVT centers as per guideline of PC/ PD.

The plot size of IVT is smaller than advanced trials. An IVT includes the maximum number of locations across the country to evaluate varietal adaptation and performance. A team of scientists (plant breeder, agronomist, pathologist, entomologists etc.) will monitor all the trials as per the recommendation of the PC. Each member of monitoring team submits their report to the PC based on their observation during trial monitoring. Entries which are superior over the best check in terms of yield and other related traits will be promoted into the advance varietal trial-I. The superiority is primarily decided based on yield potential and other related important traits such as quality traits.

#### Advance varietal trial-I

Based on superiority (5-10%) over the best performing check, superior entries will enter into the AVT-I from IVT. The number of tested entries in the AVT-I will be less than IVT. The plot size is large in AVT-I as compared to IVT, therefore data generated on yield and other ancillary traits will be more realistic, accurate and minimal chances of error. The number of testing locations should be more as compare to IVT in a given zone. During AVT-I, additional data on disease and or insect pest tolerance under artificial epiphytotic condition must be generated by the experts. Based on the performance of entry over the best performing check-in the respective zone, the superior entries would enter into the AVT-II.

#### Advance varietal trial-II (AVT-II)

All the requirements shall be fulfilled as similar to AVT-I. However, few additional data will be generated at AVT-II stage *i.e.* response of entries to different dates of sowing, seed rate, spacing between plant to plant and row to row (population density), behavior in different level of fertilizer and irrigation by sponsored agronomists; response of diseases and pests by the plant pathologists, crop quality parameters by the biochemists. The seed technology center will develop descriptors which help in the seed certification process. All the processed and analyzed data on yield and other related traits, across the locations/centers (cooperating and volunteer) shall be submitted to the PC. On the basis of these data, annual reports are being made in each crop. All the data of superior entries are comprehensively discussed in the annual workshop/national group meetings by the PC/project director. After completion of the AVT-II, the concerned breeders are informed to submit varietal proposal based on the performance of their entries during three years of evaluation.

#### Procedure for identification and Release

Based on three years performance, best performing test entries shall be identified in the annual group meet at the pre-defined institute/ university. The Zonal Coordinators and Principal Investigators attend the national group meet to provide wider aspects of information on the varieties. After the approval from Deputy Director General (Crop Science) of Indian Council of Agricultural Research (ICAR), a "Varietal Identification committee (VIC)" constituted in advance of national group meet. All the committee members (Table-1) shall be informed well in advance by the PC or PD. The VIC provides detailed information on recommended entries to the Central Sub-Committee on Crop Standards, Notification, and Release. This committee has sole right to release and notify the best-performing entry into national wise or zonal wise based on the recommendations of the VIC.

#### Table-1Varietal Identification Committee (VIC)

Representative

**Organizational** position

DDG (Crop Science)/ his or her nominee	Chairman
Project Coordinator/Project Director of AICCIP	Member Secretary
Director of Research of institute/SAUs of that region when the meeting is held	Member
Agricultural Commissioner (Department of Agriculture)	Member
One nominee of Seed organization (NSC, SSC)	Member
One representative of private seed agencies	Member
One representative of crop-based industries	Member
Project coordinator (seed technology)	Member
Two eminent scientists of that institute	Member

#### Major criteria for identification of the variety

**1.**The candidate variety must have a minimum of three years of yield and other ancillary trait data from multi-location coordinated trials.

**2.**At least two-year data on disease and pest reaction at a hot spot or artificial epiphytotic condition.

**3.** The candidate variety must have at least one-year data on agronomic performance like seed rate, dates of sowing, planting density, irrigation, and fertilization. In forage crops, three year rigorous evaluation must be done for annual crops (seed yield data for third year only) and four year for perennial crops (one year for crop establishment and other three years for evaluation).

**4.** The concerned breeder must have at least a minimum requirement of nucleus seed so that breeder seed can be generated easily.

**5.** The concerned plant breeder should have pure seed for planting of 5 ha area. If he or she did not match the requirement, then identification can only be postponed for one year.

#### Central Sub-Committee on Crop Standards, Notification, and Release

Central Sub-Committee on Crop Standards, Notification, and Release of Varieties appointed by Central Seed Committee under Section 3 of the seed act, 1966 during 1994. The committee comprised one chairman and 17 members(Table-2) .Central Sub-Committee releases varieties as per the benefit of the stake- holders and need of regional, zonal or national importance, and the State Seed Sub-Committee releases varieties beneficial for particular state. Notification of variety is compulsory on regulating the seed quality under the provision of Seed Act, 1966. Notification usually authorizes certified seed production throughout the country, by private or public seed multiplication organizations. Once the Central Sub-Committee accepts the proposal, the varieties will be released for the concerned agro-climatic zone/s (may cover one or more number of states or nation- ally). Simultaneously, it must be notified for seed certification purpose in the country. During the release, the concerned breeder must have a minimum amount of seed which can be sown at least ten-hectare area. Later on, seed multiplication is the responsibility of various seed agencies (NSC, SSC, private seed companies and progressive farmers, etc.).

S.N.	Representative	Organizaonal
1	Deputy Director General (Crop Sciences), ICAR	Chairman
2	Deputy Commissioner (QC) DAC & FW, GOI	Member Secretary
3	Directors of State Seed Certification Agencies, or their representatives	Member
4	Project Directors of Departments of Agriculture o all states, or their epresentatives	Member
5	Project Coordinators/Directors of AICCIPs	Member
6	Agricultural Commissioner, GOI	Member
7	Representatives of the seed industry, NSC, State Seed Corporations, Member private seed companies	Member
8	Representatives of ICAR, ICAR institutes, NGOs	Member
9	Progressive farmers	Member

#### Table-2 Central Sub Committee

#### Difference between released & notified varieties

S.N.	Released variety	Notified variety

1	It is not a statutory function under the Seed Act, 1966	Statutory function and variety will be registered under Section 5 of seed act 1966.
2	It cannot be used for seed certification	Only notified varieties to come under seed certification
3	No guarantee on seed quality for farmers	Only notified varieties to come under seed certification
4	Seed law enforcement agencies (seed inspector etc.) cannot draw and test seed samples	They have the right to draw and test seed samples
5	These are not assets of Govt. of India	They have the right to draw and test seed samples
6	Its main purpose is to make available the information of cultivar to the public and its area c adoption	The main purpose is seed quality regulation
7	Difficult to trace out the genesis	The notification of the varieties will help to trace out its genesis

## Central seed committee (CSC)

It is a legal body constituted by the Department of Agriculture, Cooperation and Farmers' Welfare (DAC&FW), Ministry of Agriculture and Farmers' Welfare (MoA&FW), Government of India to advise central and state government on matters related to the implementation of seed act, 1966 and other related functions

S.N.	Representative	Organizational
1	Secretary, DAC&FW, MoA& FW, GOI	Chairman
2	Additional Secretary (In charge Seeds), MoA8z FV	Member

## d committee (CCC)

	GOI	
3	Agricultural Commissioner, MoA& FW, GOI	Member
4	Deputy Director General (Crop Sciences), ICAR	Member
5	Joint Secretary (In charge Seeds), MoA& FW, GC	Member
6	Progressive farmers/ seed growers (4) nominate by the Central Government	Member
7	One representative from each State Govt.	Member
8	Director of National Seeds Project, MoA8z FW, G	Member Secretary

#### Major Empowerment of central seed committee

- The CSC has authority to release varieties (pure lines/hybrids/composites/synthetics) developed by central research institutes (ICAR/non-ICAR), AICCIPs, private or corporate sector, and other organization as per the scientific data authenticity for zonal basis (which may include more than one state) or at national level.
- The CSC has authority to approve proposals received from the State Variety Release Committees/State Seed Sub-Committees for varieties developed by the State Research Institutes but is considered suitable for areas outside the state (based on their performance).

#### State Seed sub-committee

The State Seed Sub-Committees are constituted by Central Seed Committee and are authorized to set up a State Seed Laboratory, State Seed Certification Agency (SSCA) and an Appeals Authority, and to appoint seed inspectors and seed analysts.

#### Empowerment of state seed sub committee

There are some rights which have been provided by the Central Seed Committee for proper functioning of seed chain in respective state in India. These empowerments are-

- The State Seed Sub Committee will advise the state government on all matters related to the execution of the Seeds Act, 1966.
- Planning for different crop varieties to be grown in different regions of the state, and to review the assessment of seed requirements.
- Considering the release of new varieties for the state and recommend their notification to the Central Seed Committee.

#### Need of notification

Since only notified varieties will be under the purview of Seed Law Enforcement, hence it is necessary to bring the seed of a particular crop variety under notification system. The seed inspector can only draw a sample from notified variety for analysis and ensure the seed quality. A released variety cannot come under seed chain without notification by the Gazette of India. The notification is made by the Central Government on the recommendation of the Central Seed Committee. Thus, notification is prerequisite for production of certified seed which ensures high quality of seeds to the farmers. The breeder seed can only be produced after the notification of variety and notified varieties enter into seed chain.

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#### **Denotification of varieties**

Released varieties can be denotified if they are not performing well in the area of their adoption or have been in cultivation for more than 15 years or are not much in demand. Denotification can be done based on the recommendation of central seed committee by the government of India.

#### Conclusion

There are several ways and means to increase the crop production and productivity, however using genetically pure and high-quality seed is first and prime objective in agriculture. Therefore the variety which will be used by farmers must have undergone several evaluations in order to ensure its stable yield potential, tolerance to biotic and abiotic stresses and these criteria are being fulfilled by a legal varietal release system. The main objective of the varietal release system in India is to intro- duce newly developed, high yielding varieties to the farmers for broad-spectrum cultivation in the area of their adoption and only those varieties will be notified which are superior to existing one.

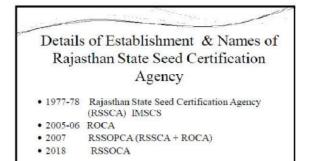
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# Field Inspection in Seed Certification-An Overview

Sh. B.S. Gupta

Ex-Seed Certification Officer,

Rajasthan State Seed & Organic Production Certification Agency, Jaipur





#### Details of Seeds Act, 1966

Seeds Act 1966 \_ Section 5 - Notification of Varieties Seeds Act 1966 \_ Section 6 - Labeling of Seed Seeds Act 1966 \_ Section 7 - Regulation of Sale of Seeds of Notified Kinds or Varieties Seeds Act 1966 \_ Section 8 - Establishment of Seed Certification Agency Seeds Act 1966 \_ Section 9 - Grant of Certificate by Certification Agency Seeds Act 1966 \_ Section 10 - Revocation of Certificate Seeds Act 1966 \_ Section 10 - Revocation of



The General Seed Certification Standards are applicable to all crops which are eligible for certification, and with field and seed standards for the individual crops, shall constitute the Minimum Seed Certification Standards. The word 'Seed' or 'seeds' as used in these standards shall include all propagating materials

I. 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 kind and varieties so grown and distributed as to ensure genetic identity and genetic purity. Seed certification is also designed to achieve prescribed standards

II. Certification Agency Certification shall be conducted by the Certification Agency notified under Section 8 of the Seeds Act, 1966.

III. Certified Seed Producer Certified seed producer means a person/organization who grows or distributes certified seed in accordance with the procedures and standards of the certification.
IV. Eligibility Requirements for Certification of Crop Varieties Seed of only those varieties which are notified under Section 5 of the Seeds Act, 1966 shall be eligible for certification. Classes and Sources of Seed\_\_\_\_\_

A. Breeder Seed Breeder seed is seed or vegetative propagating material directly controlled by the originating or sponsoring plant breeder of the breeding programme or institution and/or seed whose production is personally supervised by a qualified plant breeder and which provides the source for the initial and recurring increase of Foundation seed.

Breeder seed shall be genetically so pure as to guarantee that in the subsequent generation i.e. certified Foundation seed class shall conform to the prescribed standards of genetic purity. The other quality factors of Breeder seed such as physical purity, inert matter, germination etc. 13

shall be indicated on the label on actual basis. The Breeder seed shall be packed and supplied by the breeders in the form and manner indicated in **Appendix-I**. B. Certified Seed Certified seed shall be the seed certified by Certification Agency notified under Section 8 of the Seeds Act, 1966 or seed certified by any Certification Agency established in any foreign country provided the Certification Agency has been reorganized by the Central Government through notification in the Official Gazette. Certified seed shall consist of two classes, namely, Foundation and Certified seed and each class shall conform to the following description: 1. Certified Foundation seed shall be the progeny of Breeder seed, or be produced from Foundation seed which can be clearly traced to Breeder seed. Thus, Foundation seed can even be produced from Foundation seed. During the production of certified Foundation seed, the following guidelines shall be observed: B. Certified Seed Certified seed shall be the seed certified by Certification Agency patified under Section 8 of the Seeds Act. 1966 or seed Certified by any Certification Agency established in any foreign country provided the Certification agency has been reorganized by the Central Government through notification in the Official Gazette. Certified seed shall consist of two classes, namely, Foundation and Certified Seed and each class shall conform to the following description: 1. Certified Foundation seed shall be the progeny of Breeder seed, or be produced from Foundation seed which can be clearly traced to Breeder seed. Thus, Foundation seed can even be produced from Foundation seed. During the production of certified Foundation seed, the following guidelines shall be observed:

vegetatively propagated crops;
 apomictically reproduced crops;

- self-pollinated crops;
- often cross-pollinated and cross-pollinated crops, these being gene pools
- should not loose their genetic identity and purity if measures to safeguard the same are adequately taken;
- composite and synthetics;

- parental line increase of hybrids.

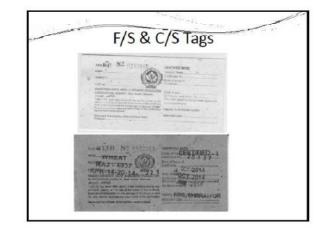
2. Production of Foundation seed stage-1 and II shall be supervised and approved by the Certification Agency and be so handled as to maintain specific genetic identity and genetic purity and shall be required to conform to certification standards specified for the crop/variety being certified.

3. (a) Certified seed shall be the progeny of Foundation seed and its production shall be so handled as to maintain specific genetic identity and purity according to standards prescribed for the crop being certified; (b) Certified seed may be the progeny of Certified seed provided this reproduction does not exceed three generations beyond Foundation seed stage-I and - it is determined by the Certification Agency that genetic identity and genetic purity will not be significantly altered; - and when the Certification Agency is satisfied that there is genuine shortage of Foundation seed despite all the reasonable efforts made by the seed producer. (c) Certification tag shall be of blue colour (shade ISI No. 104 AZURE BLUE) for Certified seed class. (d) Certified seed produced from Certified seed shall not be eligible for further seed increase under certification. Certification tags for such production which is not eligible for further seed increase under certification shall be super scribed with, "not eligible for further seed increase under certification". V

VI. Phases of Seed Certification Certification shall be completed in six broad phases listed as under:
(a) receipt and scrutiny of application
(b) verification of seed source, class and other requirements of the seed used for raising the seed crop;
(c) field inspections to verify conformity to the prescribed field standards;
(d) supervision at post-harvest stages including processing and packing;
(e) seed sampling and analysis, including genetic purity test and/or seed health test, if any, in order to verify conformity to

and/or seed health test, if any, in order to verify conformity to the prescribed standards; and (f) grant of certificate and certification tags, tagging and

(i) grant of certificate and certification tags, tagging and sealing.





#### WHY INSPECTION ARE NECESSARY

The primary objective in conducting field Inspections is to confirm that seed produced from a crop grown for seed purpose is of the designated variety, and that it has not been contaminated genetically and or physically beyond certain specified limits. Genetic contamination of a seed crop is prevented by permitting pollination by pollen from a specific desirable source recognized as the pollinator, and conversely, by preventing pollination by pollen from an undesirable or unrecognized source, through controlled pollination, physical or mechanical contamination in the field is avoided by preventing admixture during sowing and harvesting. Field inspections ensure that steps necessary to overcome genetic and physical contamination have been taken in time to make them effective.

#### The objective of field inspection is fulfilled by erifying that the seed crop is:

A. Raised from seed whose source is approved.

- B. Grown on a field area which satisfies the prescribed land requirements as to previous crop (s), to prevent contamination by volunteer plants and disease spread by pathogens.
- C. Provided with the prescribed isolation and or with the prescribed number of border rows in hybrid seed production.
- D. Planted in the prescribed ratios of female (seed) and male (pollinator) parents in the case of hybrid seed production.
- E. Properly rogued to remove contaminating factors such as pollen shedders in bajra and sorghum, shedding tassels in maize, crosses, off types, diseased plants/ears, objectionable weeds, and inseparable other crop plants so as to conform to the standards prescribed for these factors.

#### F. True to the varietal characteristics descriptive of that variety.

Harvested properly to avoid mechanical admixture. G. Grown in compliance with other special requirements for the

#### crop concerned.

The field observations made for these are compared with a set of prescribed norms called the Minimum Seed Certification Standards which are specific for each crop. The Minimum Seed Certification Standards specify the requirements for seed crops as to previous crops, isolation, varietal purity, other crop plants, objectionable weeds and freedom from certain designated diseases. They also specify the requirements for seed lots for physical qualities including pure seed, inert matter, other crop seed, weed seed, and objectionable weed seed, and for germination and entitled the"Indian Minimum Seed Certification Standards", published in September, 2013.

#### WHO SHOULD INSPECT WHAT

The authority of an agency to inspect a seed crop depends basically on whether the inspection is for certification under the Seeds Act 1966 or is only to assure production of high-quality uncertified seed. If the inspection is for official certification, only the officially notified agency for the concerned region under the Act. has the authority to perform the inspection. If the inspection is only to ensure high quality in uncertified seed, any qualified agency such as the seed producing or contracting agency may make the inspection.

If the inspection is for certification under the Act, the seed crop should be of the variety eligible for such certification. If the inspection is only to ensure high quality in uncertified seed any crop recognised by the qualified agency as for seed production can be inspected.

#### FIELD INSPECTION

#### GENERAL GUIDE LINES

Procedure for field inspections differ among crops and among growth stages of the same crop. The following broad principles on inspection methods are common to most crops and stages of growth.

- The number of inspections indicated in MSCS are the minimum and should be conducted at proper stage.
- The inspecting officer should ensure that he is guided by the producer to the correct seed field.
- Inspection of cross-pollinated crops at and after commencement of flowering should be made without prior intimation to the producer.
- 4. The producer or his representative should be requested to accompany to the field during the entire inspection and they be shown all the factors observed in the field and which will be recorded in the inspection report.

- 5. When seed fields of the same class/variety of the same producer are separated by less than 50 meters they can be considered as one field unit for inspection provided they are of same growth stage and level of conformity to standards. If they are separated by more than 50 meters, a separate inspection report shall be made for each unit.
- It is compulsory to observe it and its border areas before entering the fields, especially in tall crops like Bajra, Sorghum, Mustard etc. and crops requiring sizeable isolation distances around the outer boundary of the seed fields.
- If one third or more of a self pollinated/cross pollinated crop is so lodged that taking counts is difficult, the seed crop may be recommended for rejection.
- Walk through the entire seed field while taking field counts (it should not be localized to a portion or a few portions of a field) it should be randomly distributed all over the field.
- 14. A seed crop liable to be partially rejected due to inadequate isolation, further inspection of the efficient field (including the affected portion) should be continued according to the prescribed number and procedure and separate counts for the affected area should be mentioned in the inspection report.
- 15. If on the basis of first set of field counts, the seed crop doed not conform to the prescribed standards for any factor, a second set of counts should be taken for the concerned factor, provided the percentage of the first set of counts for that factor is more than maximum permissible limit but not more than twice the maximum permissible limit.
- 16. For seed crops involving two parental lines, even if two sets of counts in one parental line show that the field does not conform to the prescribed standards it is necessary to take counts in the other parental line.

Sr	.No	Crop	Number of Inspections	Stages of Inspection
	1	2	3	4
1.	5- 4- 1	Ragi, Paddy, Wheat, Cowpea Greengram, Blackgram, Redgram, Groundnut, Soyabean, Frenchbean, Amaranthus	2	Flowering to harvest
2.		Maize (a) Inbred line, single crosses ans hybrids (a) Composites, Synthetics and open pollinated varities	4 2	First before Flowering and three during silking stage First pre-Flowering and second during Flowering
3.	2	Hybrid Sorghum, Hybrid Bajra. Hybrid Sunflower and their parents	4	First before Flowering, second and third during Flowering & fourth during pre- harvesting.

- 9. If the plant population in a field is so thin that the entire population is less than the number of counts required entire population may be counted.
   10. Counting may be started from any pointed of the seed field but spotting a defect and trying to include/avoid it in the counts, is not desirable.
   11. Factors counted during inspection need not normally be pulled out, but be shown to the seed grower/farmer to rogue out such plants.
   12. If plants/heads of the designated factors which were pulled
  - out by the producer are lying on the ground within out skirts of the seed field, the producer should be directed to collect and remove them from the field.
- 13. If the seed field is found to be liable for rejection either in part or in full on account of inadequate isolation, the prescribed number of field counts for the entire are still to be taken for that inspection.
- 17. If on the basis of two set of counts the seed crop does not conform to the presented standards, further inspections need not be made unless the seed crop is eligible for re-inspection (after removal of contaminating factors). If the seed crop is not eligible for such reinspection then LIABLE FOR REJECTION and final inspection should be recorded in the inspection report.
- 18. If the factor present beyond the maximum permissible limit as verified by two sets of counts could not have already caused contamination of the seed crop or when contamination has already taken place; if removal of contaminating factors and contaminated materials could make the seed crop conform to the prescribed standards, their removal from the field may be recommended to permitted. Re-inspection to conform removal and conformity to standards must then be made when re-inspection is permitted and it should be shown in the inspection report.
- 19. Observations made during field inspection shall be directly recorded on inspection report on the spot and the signature of the cultivator or his representative on the field should be obtained on all copies of inspection report and, if he refuses to sign then it should be indicated in the inspection report as "Refused to Sign".

Sr.No.	Crop	Number of Inspections	Stages of Inspection
1	2	3	4
4	Open pollinated varieties of Sorghum, Bajra, Sunflower, Saffloer, Sesamum and jute	3	First pre-Flowering second during Flowering & third during pre- harvesting.
5.	Cotton (a) Hybrids (a) Varieties	4	First before Flowering Second and third during Flowering (Emasculation and crossing) fourth during picking of bolls. Flowering to harvest
6.	Castor (a) Hybrids (a) varieties	4 2	1# before Flowering 2 <sup>nd</sup> and 3 <sup>rd</sup> during Flowering 4 <sup>th</sup> at pre-harvest Flowering to harvest

	Sr.No.	Crop	Number of Inspections	Stages of Inspection
-	7.	Dhaincha	2	1 <sup>st</sup> before Flowering 2 <sup>nd</sup> at flowering and pod stage
	8.	All cucurbits and fruit vegetables(other than hybrids) viz, Brinjal. Bhindi, Tomoto, Chillies, Capsicum	3	1 <sup>st</sup> pre-Flowering, 2 <sup>nd</sup> during Flowering and fruiting 3 <sup>rd</sup> during mature fruit stage
	9.	Potato	3	1 <sup>st</sup> 45 days after sowing, 2 <sup>nd</sup> just before haulm cutting, 3 <sup>rd</sup> after haulm cutting.
	10.	Radish, Carrot and Trunip	3	1 <sup>st</sup> 20-30 days after sowing, 2 <sup>nd</sup> when lifted & replanted, 3 <sup>rd</sup> flowering.
	11.	Cumin, Coriander and Fennel	3	1 <sup>st</sup> before Flowering 2 <sup>nd</sup> 50% Flowering 3 <sup>rd</sup> Maturity

#### WHEN TO INSPECT The field inspection offered for seed certification are

conducted at following stages :

- (1) Vegetative or pre-flowering stage.
- (2) Flowering stage.
- (3) Post flowering and pre-harvest stage.

(4) Harvest stage.

#### FIELD COUNTS

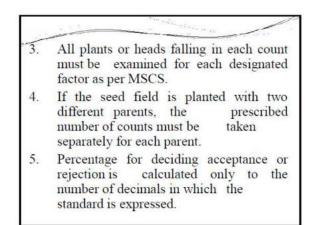
 The number of counts taken and the method of taking counts vary from crop to crop for all crops; five counts are taken for any area upto 5 Acre and an additional count is taken for every additional 5 Acre as given below:

Area of the fiedls crops	No. of Counts to be taken
Up to 5 acres	5
Above 5 to 10 acres	6
Above 10 to 15 acres	7
Above 15 to 20 acres	8
Above 20 to 25 acres	9
Above 25 to 30 acres	10

In any inspection if the first set of counts shows that the said crop does not conform to the prescribed standards for any factor, a second set of counts shall be taken for the factor. However, when the first set of counts shows a factor to be more than twice the maximum permitted, it is not necessary to take a second set of counts. Two sets of counts are called double counts. In any inspection if the first set of counts shows that the said crop does not conform to the prescribed standards for any factor, a second set of counts shall be taken for the factor. However, when the first set of counts shows a factor to be more than twice the maximum permitted, it is not necessary to take a second set of counts. Two sets of counts are called double counts.

- 2. Taking double sets of counts for a factor is :
- a) Necessary if in the first set of counts occurrence of the factor is more than the maximum permitted, but not more than twice the maximum permitted.
- b) Necessary if in the first set of counts occurrence of factor is equal two twice the maximum permissible level.
- c) Not necessary if in the first set of counts occurrence of the factor is less than or equal to the maximum permitted.
- d) Not necessary if in the first set of counts occurrence of the factor is more than twice the maximum permitted.

S.No.	Crop	No. of plants/ heads per count	Remarks
1.	Bhindi, Brinjal, Bulb crops Capsicum, Castor, Chilli, Colecrops, Cotton, Cucurbits, Maize, Groundnut, Potato, Redgram, Root crops, Teosinte, Tomato	100 plants	wide spaced and non tillering
2.	Beans, Cowpea, Gram, Leaf crops, Moong, Mustard, Peas, Sesamum, Sunhemp, Sunflower, Blackgram, Green Gram, Lentil, Niger	500 plants	Medium spaced and mon tillering
3.	Berseem, Jute, Lucerne, Mesta, Soyabean	1000 plants	Medium spaced and line sown
4.	Bajra, Barley, Oats, Paddy, Sorghum, Wheat, Ragi millets	1000 heads	Tillering crops



#### WHAT TO INSPECT

Basically sources of genetic and physical contamination must be observed and extent of their occurrence estimated.

Sources of contamination can broadly be classified as follows : A. OFF TYPES

Off types are the plants of the same species as that of the seed crop variety but morphologically of different characters eg. pigmentation, plant type, stem/ leaf shape and texture, size/colour of flower or fruit etc.

Similarly plants of other varieties of same crop are also included in off types. To designate a plant as off type it is necessary to trace it to any variety.

#### B. INSEPARABLE OTHER CROP PLANTS

Such type of plants whose seeds are similar in size, colour etc. and are difficult to separate from the seeds of seed crop by mechanical means are inseparable other crop plants.Such plants are counted if the growth stage of these plants is such that the maturity time resembles to the seed crop and may cause mechanical admixture at the time of harvesting/threshing.

Designated Insperable other crops
Oats, Wheat and Gram
Barely, Wheat and Gram
Barely, Oats and Gram

#### C. OBJECTIONABLE WEED PLANIS The plants of weed species harmful in the followering ways Size's hape of seeds are similar to crop seed which are difficult to remove by mechanical means. Growth habits has deterimental or competing effects on crop plants. Mode of spread, perpetuation, perennation or growth habit make eradication 3. difficult. 4. Plant parts are poisonous/injurious serves as alternate host for pests and diseases. Such plants are counted if the growth habit is similar to the seed crop thus causing admixture at the time of harvesting/threshing. Designated objectionable Weeds Crop Wild rice or red rice (Oryza sativa var) Paddy Rape, Mustard Satyanashi (Argemone mexicana) Cacurbits Wild Cucurbits Spp. Okra (Bhindi)

Lettuce

Berseem

Lucerne Methi Satyanashi (Argemone mexicana) Wild Cucurbits Spp. Wild Abelmoschus spp. Wild Lettuce (Lactuca serriola) Chicory or Kasni (Cicorium intybus) Dodder (Cuscuta spp) Senji (Melilotus spp)

#### D. DISEASES

Seed may carry seed borne, soil or air borne diseases. Economical and effective measures of some seed borne diseases are available. However, counts of each designated diseases should be mentioned in the inspection report.

#### E. ISOLATION

A proper designated isolation distance is compulsorily be maintained in the seed fields. All precautions should be taken so that produce of rejected area of the seed field on account of isolation is not mixed with that of the certified seed field. Threshing certificate if required may be given.

SI. No.	Сгор	Minimum distance () Foundation		
1	2	3	4	5
1.	Paddy, Wheat, Ragi Barley, Groundnut, Soyabean	3	3	Other varieties; the same variety no conforming to varietal purity requirements for certification. For loose smu susceptible whea from affected

SLNo	Сгор	Minimum distance Foundation	Isolation (meters) Certified	To be isolated by the distance in column 3 or 4 from fields of
2.	Maize (a) Inbred Line & Single crosses	400	-	Any maize with same kernel colour and texture, same inbred/single cross not conforming to varieta purity requirements for certification. Any maize with different kerne
	(b) Hybrids	-	200	colour and texture. Any maize kernel colour and texture colour same as that of seed parent. Maize of the same cross no
	- (c)Composite	-	300	conforming for certification. Any Maize with kernel colour or texture different from that of the seed parent.
	s Synthetics and open pollinated varieties.		200	Other varieties, the same variety no Conforming to varietal purity requirements for certification.

National Training on "Seed Certification", January 08-12, 2024
National Seed Research & Training Centre, Varanasi (U.P.)

-	SL No	Сгор	Minimum distance Foundation	Isolation (meters) Certified	To be isolated by the distance in column 3 or 4 from fields of
	3.	Sorghum (a) Hybrids	300	200	Other varieties of grain or dua purpose Sorghum, the same variety not conforming to varietal purity
			400	400	requirements for certification. Johnson grass (Sorghum halepense and forage sorghum with high tillering and grassy panicle.
			-	5	Sorghum hybrids with same mal parent and conforming to varietz purity requirement for certification.
		(b)Open pollinated Varieties	200	100	Other varieties of grain or dua purpose & same variety no conforming to varietal purit requirement for certification.
			400	400	Forage sorghum with high tillerin and grassy panicle, johnson gras (Sorghum halepense)

SL No.	Сгор	Minimum distance Foundatio n	Isolation (meters) Certified	To be isolated by the distance in column 3 or 4 from fields of
4.	Bajra (a) Hybrids (b) Open pollinated	1000 400	200 200	Other varieties, the same variety not conforming to varietal purity requirement for certification. Other varieties, the same variety not conforming to
5.	varieties Cowpea, Green	10	5	variety purity requirement for certification. Other varieties and fields of
	gram, Black gram, Bengal gram, Peas and beans		1000	same variety not conforming to the purity requirement for certification.
б.	Red gram	250	100	Other varieties and fields of same variety not conforming to the purity requirement for certification.

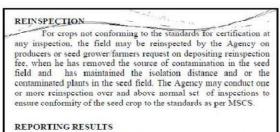
Sl. No.	Crop	Minimum distance Foundation	Isolation (meters) Certified	To be isolated by the distance in column 3 or 4 from fields of
10.	Sesamum	100	50	Other varieties of the same kind or same variety not conforming to the varital purity requirements for certification.
11.	Cotton (a) Parents of hybrids and varieties (b) Hybrids	-	30 5	Other varieties of the same species, fields of the same variety not conforming to the variat punity requirements fo certification, fields of othe species. Between the block of the parenta lines of the same hybrids.
12.	Jute	50 5	30 5	Other varieties of the same variety not conforming to varieta purity for certification. Fields of other species.

-	SL No.	Сгор	Minimum distance Foundation	Isolation (meters) Certified	
	17.	Potato	5	5	Other varieties, the same variety no conforming to varietal punity requirements for certification.
	18.	Cluster bean	10	5	Other varieties, the same variety no conforming to varietal purity requirements for certification.
	19.	Gourds (cucurbits) (a) Hybrids (b) Varieties	1500	1000 500	Fields of varieties including commercial hybrid of the same variety. Variety and the variety/hybrid not conforming to varietal purity requirements for certification.
	20.	Amaranthus	400	200	variety and the variety/hybrid noi conforming to varietal purity requirements for certification.

SI. No.	Crop	Minimum distance Foundation	Isolation (meters) Certified	To be isolated by the distance in column 3 or 4 from fields of
7.	Sunflower (a) Hybrids (b) Varieties	600 400	400 200	Fields of other varieties and fields of the same variety not conforming to the varietal purity requirement for certification and wild sunflower.
8.	Saffower and Niger	400	200	Fields of other varieties of the same kind or the same variety not conforming to the varietal purity requirement for certification.
9.	Castor	600	300	Other varieties of the same kind or not Varieties and hybrids conforming to the varietal purity requirement for certification
Seed production by modified method		1000	300	Other varieties of the same kind or not conforming to the varital purity requirement for certification.

SL No.	Crop	Minimum distance Foundation	Isolation (meters) Certified	To be isolated by the distance in column 3 or 4 from fields of
13.	Tomato Varieties Hybrids	200	25	Other varieties of the same species not conforming to varietal purity for certification. Other varietie of the same species not conforming to varietal purity for certification.
14.	Bhindi	500	250	Fields of other varieties, the same variety not conforming to varietal purity requirements for certification and wild Abelmoschus spp.
15.	Capsicum and chilli	500	250	Other varieties, the same variety not conforming to varietal purity requirements for certification, chilli from capsicum and vice versa.
16.	Brinjal (a)varieties (b)Hybrids	300 200	150 200	Other varieties, the same variety not conforming to varietal purity requirements for certification.

SL No.	Сгор	Minimum distance Foundation	Isolation (meters) Certified	To be isolated by the distance in column 3 or 4 from fields of
21.	Mustard (a) Self Compatible (b) Self Incompatible (c) Fields of Rocket salad and any of the other spp. of Genus Brassica.	200 100 100	50 50 50	Other varieties of Brassica, the same spp. & fields of the same variety not confirming to varietal purity requirement "do" "do"
22.	Femgreek	50	25	Other Variety and the same
23.	Cumin	800	400	variety not confirming to
24	Coriander	200	100	varietal purity requiremet
25.	Fennel/Ajwain	200	100	for certification
26.	Dhaincha	10	05	60 60



The results of the field inspection must be reported in the prescribed inspection report of the Agency & is to be signed by the seed grower/farmer also. A copy is to be given to him on spot.

Sometimes, even after following all regulations and observing normal field counts, an officer may some times observe defects which do not come in field counts. Under such conditions he may follow the suggested procedure :

1.	When patches or rows off types, shedders, shedding						
-	tassels objectionable weeds, inseparable other crop plants						
	heads or plants affected by diseases are noticed but not						
	come under field counts, separate observations such as						
	size of the patch number of rows etc. should be made.						
	reported and be shown on a map. The officer should exercise discretion and attempt to save the crop from rejection						
	by advising the grower to remove the defective patch						
	before contamination occures.						
2.	If the male/female parents in seed production involving						

- two parents have been irregularly planted, it should be recommended as "LIABLE FOR REJECTION". 3
- If the seed crop is grown as mixed, inter or companion crop other than prescribed norms, it should be recommended as liable for rejection.
- If the seed crop has failed partially or completely or is damaged by cattle, flood, drought etc. or the producer does not want to offer it for certification, the inspection report should still be prepared. 4.

#### HARVESTING.

Seed crop meeting the field standards after final field certification shall be properly harvested, threshed, dired and transported to the registered seed processing plant as per crop calender for processing and certification. during the above operations seed producer growers should take all necessary precautions to safe guard the seed quality.

- The Crop should be harvested at proper stage. (i)
- It should be properly dried, threshed so that no admixture (ii) takes place at threshing floor.
- All thresher or bags should be clean, bags are not old and torned. (iii)
- All stones, stalks, mud balls etc. should be removed for better (iv) processing
- Bags should not be over filled & not more than 100 Kg. (v) capacity.
- Care to be taken for Soyabean harvesting, threshing & packing. (vi)



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# Causes of Seed Quality Deterioration and Remedies to be Adopted

Dr.C.P.Sachan,

Ex Professor & Head,

Deptt. of Seed Science & Technology, CSAU Ag. & Tech. , Kanpur

Seed deterioration is an undesirable and detrimental attribute of agriculture. This process is a separate event from seed development and germination. Losses in seed quality occur during field weathering, harvesting and storage. Deterioration caused by field weathering is directly related to seed exposure to adverse conditions. Seeds are highly susceptible to damage and mechanical injury during post-harvest handling. Seed quality is depends upon initial seed quality, temperature, moisture content and mycoflora. Rapid deterioration occurs due to these environmental conditions make very difficult to maintain its viability during storage. However, the seed quality and viability during storage depend upon the initial quality of seed and the manner in which it is stored. Seed deterioration is associated with various cellular, metabolic and chemical alterations including lipid peroxidation, membrane disruption, DNA damage, impairment of RNA and protein synthesis and cause several detrimental effects on seed.As seed deterioration increases, seed performance is progressively decreases. Losses in seed quality occur during field weathering, harvesting and storage. Several factors contribute to the susceptibility for seed deterioration. The basic causes are temperature, relative humidity, seed moisture content and by invasion of and damage to tissues by microorganisms, insects.

## WHAT IS DETERIORATION?

Seed deterioration can be defined as "deteriorative alterations occurring with time that increase the seed's exposure to external challenges and decrease the ability of the seed to survive". Seed ddeterioration causes loss of seed quality with time.

It is a natural process which involves cytological, physiological, biochemical and physical changes in seeds. These changes reduce viability and ultimately cause death of the seed.

## **TYPES OF DETERIORATION**

Deterioration is evident as a decrease in percentage germination, while those seeds that germinate produce weak seedlings. Losses in seed quality occur during field weathering, harvesting and storage harvesting time of several crops depends on its maturity time and on physiological maturity. Harvesting stage influences the quality of seed, germination, vigor, viability and also storability Physiological maturity attainment is a genotypic character which is influenced by several environmental factors. Deterioration of seed in the field before harvest (field weathering) begins when the seed reaches physiological maturity and it extends till the seeds are harvested. The moisture content of physiological maturated seed is approximately 50- 55%. Because of the high moisture content, the seed cannot be harvested commercially and must remain in storage on the plant through a desiccation period till moisture levels are adequately low to permit mechanical

harvest without causing undue damage to the seed. This desiccation period vary from a few days to several weeks before the seed attains a harvestable moisture level, about 14%. During this post maturation, pre-harvest period weather conditions have a great influence on the quality of the harvested seed. Field conditions are rarely favorable for such storage. Seed quality is influenced by numerous factors that occur in the field before harvesting and during harvesting, drying, processing and storage. The losses are worsened if seeds are stored at high temperatures and high relative humidity conditions

## **Field Weathering**

The deterioration of seed quality, vigor and viability, due to high relative humidity and high temperature during the post-maturation and pre-harvest period is referred to as field weathering. Weathering occurs in the period between the attainment of physiological maturity till harvesting in the field. Deterioration caused by weathering is directly related to seed exposure to hot and humid conditions, rainfall, photoperiod after ripening are pre-harvest factors, cause seed quality loss. Among all these factors, influence of moisture on seeds during ripening appears to exert the major influence on predisposition to weathering. Adverse environmental conditions during seed filling and maturation result in forced seed maturation, which is associated with low yields.. Harvest delays beyond optimum maturity extend field exposure and intensify seed deterioration. Weathering not only lowers seed germination, but also increases susceptibility to mechanical damage and disease infection. Timely harvesting avoids prolonged exposure to moisture, and is the best means of avoiding weathering.

## Harvest and Post-harvest Deterioration

Seed quality is highly affected by harvesting and handling methods. Harvest and post-harvest deterioration comprises threshing, processing machinery, seed collection, handling, transporting and drying. Mechanical damage is one of the

major causes of seed deterioration during storage. Very dry seeds are prone to mechanical damage and injuries. Such damage may result in physical damage or fracturing of essential seed parts; broken seed coats permit early entry and

Easy access for microflora, make the seed vulnerable to fungal attack and reduce storage potential . Large seeded varieties are more sensitive to mechanical damage than small seeds.

#### Storage

Storability of seeds is mainly a genetically regulated character and is influenced by quality of the seed at the time of storage, pre-storage history of seed (environmental factors during preand post-harvest stages), moisture content of seed or ambient relative humidity, temperature of storage environment, duration of storage and

biotic agents . These environmental conditions are very difficult to maintain during storage. The seed storage environment highly influences the period of seed survival. After planting of deteriorate seeds, seedling emergence may be poor and ttransmission of pathogens to the new

crop may occur. Lower temperature and humidity result in delayed seed deteriorative process and thereby leads to prolonged viability period.

## MECHANISMS OF SEED DETERIORATION

Once seed deterioration has happened, this catabolic process cannot be reversed It is a sequence of events beginning with a chain of biochemical events, predominantly membrane damage and impairment of biosynthetic reactions, and then the resulting losses of various seed performance attributes, starting with reduced germination rate, reduced field emergence, increased numbers of abnormal seedlings and finally seed death. Structural changes associated with oxidation are reduced membrane fluidity, altered folding of DNA, lost elasticity of proteins and increased brittleness of the cellular matrix.

#### **Biochemical Manifestation of Seed Deterioration**

Seed deterioration is associated with various cellular, metabolic and chemical alterations including chromosome aberrations and damage to the DNA, impairment of RNA and protein synthesis, changes in the enzymes and food reserves and loss of membrane integrityy. Some of the major physiological and biochemical events of deterioration are presented below.

#### **Membrane** Degradation

It is extensively consented that loss in cellular membrane integrity is one of the primary causes for loss of viability. Under harsh storage conditions loss in membrane permeability leads to increased leaching of seed constituents and hence loss in

Viability. During seed deterioration, membrane degradation increase electrolyte leakage. Decline in seed germination, field emergence and seedling vigor is associated with high level of electrolytes leakage. Alterations of membrane systems, such as the tonoplast, plasmalemma and endoplasmic reticulum, result in diminishing of normal cell function and energy production.

#### **Enzymes Alterations**

Enzyme alterations, such as reduced activity of lipase, ribonuclease, acid phosphatase, protease, diastase, catalase, peroxidase,  $\Box$  and  $\Box$  amylase,

DNase and dehydrogenase enzymes. ROS and hydrogen peroxides are produced from several metabolic reactions and could be destroyed by the activity of scavenger enzymes like catalase and peroxides. Peroxides activity decreases substantially with ageing. Due to this seeds become more sensitive to the effects of oxygen and free radicals in membrane unsaturated fatty acids and produce lipid peroxidation products

such as monaldehyde and lipid conjugants.

## **Changes in Cell Chemical Constituents**

In deteriorate seeds significant decrease in protein, oil content and total sugars and increase in free fatty acids and reducing sugars showed that carbohydrates increased with decrease in protein

content in deteriorated seeds. Some studies indicated that oligosaccharide which has been associated in stabilizing membranes decreased during storage.

#### **Reduced Metabolic Activity**

High relative humidity hastens deterioration and results in reduction of nucleic acids with increased storage period. Metabolic activities of seeds were low in non-viable seeds than in viable seeds. Long term storage decreases the ability to form nucleic acids and nucleotides.

#### Free Radical Damage

Deterioration is partially associated to the accumulation of free-radicals produced by the metabolic process. Seed storage subjects lipids to slow consistent attack by oxygen, forming hydrogen peroxides, other oxygenated fatty acids and free radicals. The free radicals are unstable and may react and damage nearby molecules.

Oxygenated fatty acids in the absence of enzymes activity in the dry seed would

accumulate and damage cellular components and leads to deterioration of seeds. Lipid peroxidation and free radicals formation are the major causes for the deterioration of oil seeds in storage.

#### **Chromosome Aberrations**

One of the changes linked with seed ageing is aberration of chromosomes, sometimes pertained to as mutagenic effects. Some of the chromosome alterations in seeds comprise fragmentation, bridges, fusion, ring formation of chromosomes and variations in nuclear size. Some other causes of deterioration are:

- Decline in sugar content,
- Inability of ribosomes to dissociate, enyme degradation and inactivation (amylase, dehydrogenase, oxidases, phospholipase, glutamic acid decarboxylase),
- Formation and activation of hydrolytic enzymes,
- Starvation of meristematic cells,
- Increases in seed leachates and free fatty acid content,
- Reduced respiration, and accumulation of toxic compounds.

Moreover the main cause of seed damage, lipid peroxidation causes initial biochemical changes in seed that can be observed during storage. Auto oxidation of lipids and increase in the content of free fatty acids throughout storageperiod are the main reasons for rapid deterioration of seed of oil plants .In sunflower seeds, loss of viability is associated with an accumulation of malondialdehyde (MDA), Membrane disruption is one of the primary reasons attributed to seed deterioration. As a result, seed cells are not capable to hold their normal physical condition and function. Lipid peroxidation can result in not only destruction of the lipid itself, but also damage to cell membranes and other cellular components.

#### **Factor Affecting Deterioration**

The rate of seed deterioration is highly influenced by environmental (temperature, relative humidity and seed moisture content) and biological factors (such as fungi that create their own biological niche) Seed longevity is determined by seed moisture, temperature and seed attributes that are influenced by genetic and environmental interactions during seed maturation, harvesting and storage Several other factors such as environmental conditions during seed producing stage, pests, diseases, seed oil content, storage longevity, mechanical damages of seed in processing, fluctuations in moisture (including drought), weathering, nutrient deficiencies, packaging, pesticides, improper handling, drying and biochemical injury of seed tissue can affect vigor of seeds.

## Kind/variety of the Seed

The seed storability is considerably determined by the kind or variety of seeds. Some seeds are naturally short-lived, e.g., onion, soybeans, peanuts, etc., whereas some seeds like, tall fescue and annual rye grass, appear very similar but differ in storability. Genetic make-up of varieties also influences storability

## **Genotypic Factors**

Some types of seeds are inherently long lived; others are short lived, while others have an intermediate life span owing to their differences on genetic makeup.

## **Initial Seed Quality**

High initial viability of seeds maintains their quality in storage longer than those with less initial viability. Vigorous and undeteriorated seeds can store longer than deteriorated seeds. Seeds that have been broken, cracked, or bruised due to

handling deteriorate more rapidly in storage than undamaged seeds. Cracks in seeds serve as entrance to pathogens causing consequent deterioration. Seeds that have been developed under environmental stress conditions (such as drought, nutrient deficiency and high temperatures) become more susceptible to rapid deterioration.

## **Effect of Temperature**

High temperature hastened the rate of these biochemical processes triggering more rapid deterioration that resulted in rapid losses in seed having high moisture content. Seeds sensitivity to high temperatures is strongly dependent on their water content, loss of viability being quicker with increasing moisture content. Temperature is important because it influences the amount of moisture and also enhances the rate of deteriorative reactions occurring in seeds as temperature increases.

## **Effect of Moisture Content**

Deteriorative reactions occur more readily in seeds at higher moisture content and subsequently, this condition constitute hazard to the longevity of seed survival . seeds stored at high moisture content demonstrate increased respiration, heating, and fungal invasion resulting in reduced seed

vigor and viability. After physiological maturity the rate of seed quality loss depends on the degree of unfavorable environmental conditions surrounding the seed. Environmental moisture, predominantly intermittent or prolonged rainfall, during the post maturation and pre-harvest period, is quite detrimental to seed quality and cause rapid deterioration. When exposed to humid conditions (heavy rain), dried seeds can absorb enough moisture to reach 27% and subsequently expand in volume. At this moisture level, seed respiration is hastened. Cotyledonary reserves will be consumed, not only by the seed itself, but also by fungi allied with the seed. It has been reported that seed moisture content of about 6-8% is optimum for maximum longevity of most crop species. Below 4-6% seed moisture content lipid auto oxidation becomes a damaging factor and seeds become more susceptible to mechanical damage.

The moisture content of seed during storage is the most persuasive factor affecting the longevity. Storing seeds at high moisture content enhances the risk of quicker deterioration at shorter time. Seeds are hygroscopic in nature; they can pick up and releases moisture from and to the surrounding air. They absorb or lose moisture till the vapor pressure of seed moisture and atmospheric moisture reach equilibrium Control of relative humidity is the most important because it directly influences the moisture content of seeds in storage as they come to equilibrium with the amount of moisture surrounding them; a concept known as equilibrium moisture content. The lower the moisture content, the longer seeds can be stored provided that the moisture level can be controlled all through the storage period.

#### Effect of Organisms Associated with Seeds

## Bacteria and fungi:

There are several factors which favor infection fungi and promote their infestation such as moisture content of seed and interspace relative humidity, temperature, prestorage infection and storage pest. Most storage fungi belong to *Penicillium* and *Aspergillus* genera. They induce seed deterioration by producing toxic substances that destroy the cells of seeds.To minimize the risk of fungi invasion, seeds have to be stored at low moisture content, low temperature, and RH. Researches show that all storage fungi are completely inactive below 62% relative humidity and show very little activity below about 75% relative humidity upwards, ity. The storage bacteria requireat least 90% relative humidity for growth and therefore only become significant under conditions in which fungi are already very active.

#### Insect and Mites:

There is no insect activity at seed moisture contents below 8%, but if grain is

infected, increased activity may generally be expected up to about 15% moisture content. The optimum temperature for insect activity of storage insects ranges from 28 to 38°C. The temperatures below 17 to 22°C are considered unsafe for insect activity. Although it is usually preferable to control insect and mite activity by the manipulation of the seed environment, i.e., use of fumigants and insecticides. The main problem of chemical control is the adverse effect of chemicals on seed viability and vigor, and some of them are dangerous to handle.

However, fumigants which have been used successfully include methyl bromide, hydrogen cyanide, phosphine, ethylene dichloride and carbon tetrachloride in 3:1 mixture, carbon disulphide and naphthalene. Insecticides – used in seed storage include lindane and Malathion.

#### Provenance

Seeds obtained from different sources may show differences in viability and storability. Nevertheless, the seed begins its existence before it harvest and it is expected that seeds harvested in different pre-harvest condition.

# Fluctuating Environmental Conditions

Fluctuating environmental conditions are harmful for seed viability. Rapid changes in seed moisture content and temperature cause deleterious effect.

#### **Oxygen Pressure**

Recent researches on the role of a gaseous environment on seed viability indicate that increases in pressure of oxygen incline to decreases the viability period.

#### **Other Factors**

Factors besides those discussed above that affect seed storage life are the direct sunlight on the seed, number of times and kind of fumigation, effect of seed treatment, etc.

# Symptoms of Seed Deterioration

Seed deterioration is an inexorable, irreversible degenerative change in the quality of a seed. It is observable in their dropped performance during germination such as delayed seedling emergence, slower rate of seedling growth and development, loss of the capacity to germinate was the final phase of deterioration process and

final indication of vigor loss. Deteriorated seeds are also diminished resistance to environmental stresses during germination and early seedling growth. Some of the symptoms of the deteriorated seed are:

# Morphological Changes

Changes in seed coat color are apparently owing to oxidative reactions in the seed coat that are enhanced under conditions of high temperature and relative humidity.

#### Ultra Structural Changes

By electron microscopy examination, two broad patterns of coalescence of lipid bodies and plasmalemma extraction from the cell wall related with deterioration have been observed. Both of these events influence cell membrane integrity.

# Cell Membrane Changes

Some consequence of membrane damage includes:

- Breaks in the plasmalemma structure and its contraction from the cell wall,
- Fragmented endoplasmic reticulum lacking of polyribosomes,
- Lack of dicytosomes monosomes arbitrarily dispersed in the cytoplasm,
- Disintegration of mitochondria and plastids,
- Coalescence of lipid droplets,
- Condensation of chromatin and lobed nucleus,
- Lyses of membranes of lysomic structures.

#### Loss of Enzyme Activity

The most sensitive test for measuring early seed deterioration is tetrazolium (TZ) and glutamic acid decarboxylase activity test. Other oxidases enzymes such as catalase, peroxidase, amylase and cytochrome oxidase also correlated with seed deterioration.

#### **Reduced Respiration**

As seeds deteriorate, respiration becomes gradually pathetic and eventually leads to loss of germination. Reduction in respiration rate is closely associated with seed eterioration.

#### **Increase in Seed Leachates**

Increase in leachate content when soaked in water is often noticed symptom of deteriorated seeds. The leachate concentration can be measured by electrical conductance methods and by determines the soluble sugar content of the leachate.

# **Increase in Free Fatty Acid Content**

The hydrolysis of phospholipids leads to the release of glycerol and fatty acids, and this reaction hastens with increasing seed moisture content. The frequent accumulation of free fatty acids concludes in a decline in cellular pH and is detrimental to normal cellular metabolism

#### Methods for Testing Seed Deterioration

#### **Germination Test**

It is an analytical procedure to evaluate seed germination under standardized, favorable conditions. Standard germination testing includes media, temperature, moisture, light, dormancy breaking and germination counting standard for various crop seeds.

# Tetrazolium (TZ) Test

TZ test is extensively accepted as an accurate mean of estimating seed viability. This method was developed by Professor Georg Lakon in the early 1940s. It is quick method to estimate seed viability (Copeland and McDonald, 2001). This test

distinguishes between viable and dead tissues of the embryo on the basis of their relative respiration rate in the hydrated state.

# **Electrical Conductivity Test**

As seed deterioration progresses, the cell membranes become less rigid and become more water pincreasing electrical conductivity. It provides a rapid indication of seed viability for seed lots.

# Vital Coloring Test

The principle of this method is the differential coloration of live against dead tissues when exhibited to certain dyes such as sulfuric acid, indigo carmine and aniline dyes. These dyes stain the dead tissue blue and the live tissue leftovers unstained. This method is particularly useful for determining viability of tree seeds.

#### **Enzyme Activity Test**

These methods measure enzyme activity (such as lipase, amylase, diastase, catalase, peroxidase and dehydrogenase) of imbibed seeds as an indication of their viability.

#### Other Tests

Other testing methods are free fatty acid test, hydrogen peroxide test, indoxyl acetate test, fastgreen test, ferric chloride test, sodium hypochlorite test, excised embryo test and X-ray test. These methods were discussed by Copeland and McDonald (2001).

#### **Detrimental Effects of Seed Deterioration**

Some probable consequences of deteriorative changes in seeds are:

- Decreased percent germination;
- Reduction in vigor and viability;
- Degradation of cellular membranes and loss of permeability control
- ; Increased solute leakage;
- Impairment of energy-yielding and biosynthetic mechanisms;
- Reduced biosynthesis and respiration; Reduced germination rate and early seedling growth;
- Reduced rate of plant growth and development;
- Reduced storage potential;
- Decreased growth uniformity;
- Increased susceptibility to environmental stresses, especially during germination, emergence, and early seedling development;
- Reduced tolerance under adverse conditions
- Decreased yield;
- Decreased emergence percentage;
- Increased percentage of abnormal seedling;
- Loss of the capacity to germinate; and

Loss in seed weight results in decreasing the quality of seed produced, such as purple stained seeds (contaminated by fungi), wrinkled seeds, fissures

in the seed coat, insect damaged seeds, discoloration of seed, etc.

# CONCLUSION

Seed quality, germination, vigor and viability are highly influenced by environmental factors in field and storage. There was a distinct reduction in yield, seedling growth, loss of capacity to germinate and increased susceptibility to environmental stresses which cause numerous harmful effect on seed quality.

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# OECD Varietal Certification & its importance in context to India

Sh. B.S. Gupta

Ex Seed Certification Officer

&

Master Trainer OECD Seed Schemes

Rajasthan State Seed & Organic Certification Agency Pant Krishi Bhawan, Jaipur

#### Outline

- Introduction
- International (OECD) Seed Certification
- OECD Seed Schemes
- India: Participation
- Indian Vs OECD seed certification
- Status of Implementation
- Certification Charges
- Progress

#### Introduction

- The Organization for Economic Co-operation and Development (OECD) is an intergovernmental organization founded in 1961
- Act as a multilateral forum to discuss, develop and reform economic and social policies
- Promote sustainable economic growth and employment, a rising standard of living and trade liberalization

#### Contd...

- Inter-governmental Organization
  - 34 Member countries
  - Works with > 80 partner, developing and transition economies
     Works with > 30 international organisations (IOs)
- Multilateral Forum ...
  - Addresses economic, social, environmental, trade and agricultural challenges
  - Economic analyses ....

#### International (OECD) Seed Certification

- The OECD Schemes for the Varietal Certification referred as International (OECD) Seed Schemes
- Provides International framework for certification of seeds moving in International seed market
- Voluntary & Self financing programmes
- 61 participating countries

#### **OECD Seed Schemes: Objectives**

- To encourage the use of "quality-guaranteed" seed in participating countries
- To authorize the use of globally recognised labels and certificates for international seed trade
- · To facilitate the import and export of seed
- To enhance co-operation and understanding between
  - Importing and exporting countries
  - Public and private sector and other international organizations

#### OECD Seed Schemes

10

There are eight broad groups of crops

- Cereals
- Maize
- Sorghum
- Grasses and legumes
- Crucifers and Other Oil or Fibre Species
- Vegetables
- Fodder Beet and Sugar Beet
   Subterranean Clover and Similar Specie

#### **OECD Seed Schemes: Benefits**

- · Facilitate the International seed trade
- Provides a framework to develop seed production plans
- Internationally harmonized rules for seed certification
- Develops collaboration between the public and private sectors
- Regular exchange of information with other national certification agencies
- Improves competencies in domestic seed quality regulation system

#### India: Participation

- India: become member of OECD seed schemes in October 2008
- National Designated Authority (NDA): Joint Secretary (Seeds), Ministry of Agriculture, Government of India
- Participating in six seed schemes
- NDA is responsible for implementation of the schemes



Indian delegation at OECD Annual Meeting, Chicago, 2008

#### Implementation

- National Designated Authority is apex body for implementation of the schemes
- Registration of varieties eligible for certification in the national list of OECD
- · Certification for varietal identity and purity
- · Control plot tests
- Nine State Seed Certification Agencies identified as Designated Authorities for implementation of the schemes

Name of DA	Area of Operation
TSSOCA, Hyderabad	Telangana, Chattisgarh
RSSOCA, Jaipur	Rajasthan, Haryana, Punjab & MP
BSSCA, Patna	Bihar, Jharkhand, West Bengal , All North Eastern States including Sikkim and Andaman & Nicobar
MSSCA, Akola	Maharastra, Gujarat, Daman & Diu, Dadra & Nagar Haveli & Goa
USSCA, Dehradun	Uttarakhand, HP, Delhi, Jammu & Kashmir
APSSCA	Andhra Pradesh and Orissa
UPSSCA	Uttar Pradesh
KSSCA	Karnataka
TSSCA	Tamil Nadu, Pudduchery, Lakshadweep and Kerala

#### **OECD List of Varieties**

- It is an official list of varieties of NDA as eligible for certification
- A variety proposed to be added in the OECD List of Varieties for certification must:
  - Be distinct
  - $\checkmark$  Have an acceptable "value" in at least one participating country.
  - Be maintained
  - ✓ Be included on the National Official Catalogue of the country of registration of the variety
  - ✓ 245 varieties of 24 crops listed under OECD.

#### OECD varietal list: Criteria for inclusion

- Released and notified under the Seeds Act, 1966
- Filed for registration to PPV & FR Authority
- Tested under multi-location trails for two years in public system
- Export potential and Tested under multi-location trails including in-house trials for two years
- Tested outside the country for two years along with data



	OECD	Labels	
IDIAN CERTURICATION TACS	Indian seed category	OECD Seed category	Color code
	Breeder seed	Pre-basic	White with diagonal
	Foundation seed	Basic	White
	Certified seed	Certified 1st Generation	Blue
	Certified seed – II	Certified 2nd Generation and successive generations	Red
ALL ALL	Labelled seed	Not Finally Certified Seed	Grey

Indian seed certification	<b>OECD</b> Seed Certification	
1. Classes of seed		
Nucleus Seed: • Through maintenance breeding by the maintainers / breeders varietal characters checked. • Controlled and maintained by the maintainers/breeder. • Carries breeder's certificate. • Used for breeder seed multiplication.	Breeders Maintenance Material: • Checked against DUS Centers for the definite characters. • Carries Suppliers Labels. • Controlled and maintained by the maintainer/breeder. • Used for pre-basic seed multiplication.	
Breeder Seed: (Golden Yellow Tag)  Controlled by monitoring team of L. crop breeder, II. Representative of Director of Seed Certification/ Assistant Director of Seed Certification, III. Representative of NSC, ix. Farmers / producers representative Grow Out: Text is employed for certain crops. Produced through Breeder Seed Production Center (BSP) based on the Indent Used for foundation class seed multiplication.	Pre-Basic Seed: (White Label with diagonal Violet Stripe ) • Controlled by official certification authority (DA) + Maintainer. • Undertake pre-controlled test – • Can not be commercialized and not for sale. • Produced officially by the recognized • Institute/organization.	

Indian seed certification	OECD Seed Certification
Foundation Seed: (White Colour Tag) • Controlled by official seed certification agency directly and no role of maintainer. • need based GOT test • Produced through registered seed producers / growers. • Can be used for foundation stage I (F1) to foundation stage II (F2) multiplication on specific cases for the open pollinated varieties with specific approval from the Director of Seed Certification. • Can be used for certified stage of multiplication. • Initial validity period of 9 months from the date of test and subsequently six months for revalidation.	Basic Seed: (White Label) • Controlled by official certification authority • (DA) + Maintainer. • Undertake pre-control tests – • Can not be commercialized and not for sale. • Produced officially by the recognized Institute/organization. • No such validity period

Indian seed certification	OECD Seed Certification
Certified Seed: (Azure Blue Tag)	Certified Seed (C1) - (Blue Label ) (C2 ) - (Red Label )
Controlled by official seed certification agency directly and no role of maintainer. • need based GOT test • Produced through registered seed producers / growers. • Can be used for certified stage I (F1) to certified stage II (F2) multiplication on specific cases for the open pollinated varieties with specific approval from the Director of Seed Certification. • Can be used for certified stage II and commercial multiplication. • Initial validity period of 9 months from the date of test	<ul> <li>DA's and Controlling Authorities- under take quality control including post control test + provision of Patent Royalty to the Maintainers / Breeder's.</li> <li>Used for the commercial multiplication/sale.</li> <li>No such validity period is existing.</li> </ul>

Indian seed certification	OECD Seed Certification
Labeled Seed: (Opal green colour)	Not Finally Certified Seed : (Grey Label)
Produced by the producer himself and no role of certification agencies. > Label with all seed standards details and signed by the producer himself. > Producer himself responsible for varietal purity and seed standards.	Seed Which is to be exported from the country of production after field approval, but before final certification as basic or certified seed, shall be identified in fastened containers by the special label.
No such class of seed exist	Standard Seed: (Dark Yellow Label) > mainly exists in vegetable seed scheme. > declared by the supplier as being true to the variety and of satisfactory varietal purity. > It must conform to the appropriate conditions in the Scheme.

Indian seed certification	OECD Seed Certification
Eligibility of Varieties and Parental Constituents	
Varieties notified under Section (5) of the Seed Act, 1966 eligible for certification	<ul> <li>Registered in National catalogue of Varieties.</li> <li>Country shall have national list of varieties under the OECD Schemes. (DUS) (VCU at least in country)</li> </ul>
Field inspection & sampling	1
<ul> <li>Done by seed certification officials and supervised by supervising authority.</li> <li>There is no system of authorization of private inspectors / seed sampler in Indian system</li> </ul>	<ul> <li>Done by the officials of DAs</li> <li>Can also be done by authorized inspectors/ samplers and supervised by official supervisors.</li> <li>(5% check sampling done by official seed samplers.)</li> </ul>

(Specific Crop standards ) IMSCS	OECD
Previous Cropping	
Free from volunteer crop Eg. Groundhut- 2 years Sunflower- 1 year	Minimum time interval between seed crop and any other crop of same species Cruciter spp- 5 years Other spp- 2 years Hybrid seed may not be grown of the same field in successive years.
Isolation Distance	
No modification of Isolation distance is permitted (except maize)	Distances can be modified where there is sufficient protection from undesirable pollen or where the possibility of cross-fertilization is eliminated.
Seed standards	
Maximum permitted objectionable weed plats: 1. Foundation Seed : 0.010% 2. Certified Seed : 0.020% Insect damage- For both F.S. and C.S Maize and Legumes- 1% Other crops- 0.5 %	Specific permissible limit for designated diseases and weed seeds not indicated in OECO standards. No maximum permissible limits are indicated in case of insect damage.

Pre and Post Control Tests	
As per IMSCS, SCAs shall conduct GOT wherever it is a pre-requisite No provision of Pre and Post Control Tests	Pre control test is compulsory for Pre- basic and Basic seed. A part of every sample of Basic Seed and 5 to 10% of the Certified seed shall be checked in a post-control test conducted immediately or in the following season
Issue of Certificates	
For Breeder seed by the concerned Scientist in charge of Production. For Foundation and Certified class issued by the officers of SSCA.	The Designated Authority may issue certificates for each lot of Pre-Basic, Basic and Certified seed approved under the Scheme.
Blending of Lots	
No provision for blending	lots of certified seed of the same generation of one variety may be blended before or after export in accordance with the regulations of the Country. Provision for Re-packing and Re- labeling in another Country.

Status of
Implementation

September 2007	Submission of application to the Secretary General, OECD Secretariate
April 2008	OECD Evaluation Mission to India
June to July 2008	Indian delegation participated in the Annual Meeting of the OECD held in Chicago, USA.
October 2008	OECD Seed Schemes was accepted by the OECD council.
November 2008	Notification of the Joint Secretary (Seeds), Government of India Ministry of Agriculture as NDA for the OECD Seed Scheme

January, 2009	Tamil Nadu, Karnataka, Andhra Pradesh, Maharashtra,
	Rajasthan, Uttarakhand and Uttar Pradesh were nominated as Designated Authorities
March, 2009	Haryana, Bihar and Assam were subsequently identified as Designated Authorities.
June, 2009	Indian Delegation participated in the Technical Working Group and Annual Meeting of OECD at Paris
February , 2010	Training on OECD Seed Schemes for the members of DAs was organized by the Govt. of India at New Delhi by inviting Foreign Expert MR. David White, Seed consultant, OECD Coordinating Centre, United Kingdom( UK)
June, 2010	OECD Seed Scheme training in Canada under the leadership of Joint Secretary( Seeds), GOVT. of India, DAC, New Delhi
July , 2010	OECD Seed Schemes Workshop organized by the Govt. of India at NSRTC, Varanasi,

	tatus of Implementation
September, 2012	Workshop on OECD Seed Schemes held at Hyderabad by inviting members of DA.
January, 2013	Hands on Training programme on OECD Varietal Certification and seed testing at Bengluru.
April, 2015	Publication of book " OECD Varietal Certification in INDIA" By Ministry of Agriculture and Farmers Welfare Govt. of India, New Delhi"
October, 2015	Training of OECD Seed Schemes for the members of DA'S was organized by the GOVT. Of India at NSRTC, Varanasi
July, 2016	Telangana State Seed & Organic Certification Authority

July, 2016	Telangana State Seed & Organic Certification Authority organized the National level workshop
November & December, 2016	Telangana State Seed & Organic Certification Authority in collaboration with Govt. of India organized the second International level workshop
January, 2017	National Task Force on OECD Seed Schemes was constituted to accelerate implementation of OECD seed schemes
November,2017	Theoretical Workshop on OECD Varietal Certification at Hyderabad by inviting Foreign Experts.
April, 2018	Practical Workshop OECD Varietal Certification at Hyderabad by inviting Foreign Experts.
September, 2018	Workshop on Introduction of International ( OECD) Seed Certification at Dehradun ( Uttarakhand)
January, 2020	Training programme on OECD Varietal Certification in India organized by KSSOCA at Bengaluru.

	Crops		Number of varieti	es
In the world		Public	Private	Tota
200 species 49,899 varieties	Pulses	17		17
In India 20 species	Oilseeds	9	2	11
	Cotton	2	5	7
	Pearl millet	3	5	8
237 varieties Further listed	Barley	4		4
10 species	Rice	15	21	36
73 varieties	Wheat	5	1	6
	Sorghum	4	3	7
	Maize	1	12	13
	Total	160	78	237

<b>OECD</b> Certification	Charges
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S. No	Operational details	Grasses, legumes, cereals, maize and sorghum (Rs.)	Crucifer and other oil or fiber species seed (Rs.)	Vegetabl es seed (Rs.)
1	Registration charges for growers/sowing report	125	125	125
2	Field inspection charges/acre Varieties Hybrids	500 750	500 750	500-4000
3	Processing charges (per qtl.) Ginning and processing (cotton) kapas/qtl Post harvest supervision charges for 8 hrs	40	40 75	1000

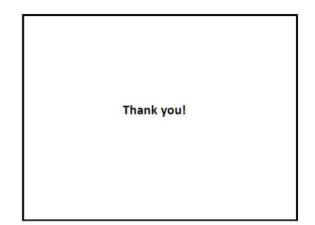
S.No	Operational Details	Grasses, legumes, cereals, maize and sorghum (Rs.)	Crucifer and other oil or fiber species seed (Rs.)	Vegetable s seed (Rs.)
4	Seed testing charges (per sample or actual)	400	400	400
5	Pre-control & post control test charges (per sample)	1000	1000	1000
6	Tag charges (per tag)	10	10	10
7	Varietal purity (DNA test) test charges (per sample or actual)	2000	2000	2000

Registration & Annual renewal charges of Seed Production Organisation is Rs.2000/-and 1000/- respectively.
 Registration & Annual renewal charges of Seed Processing Plant is Rs.3000/- and 1000/- respectively.

#### The progress on International (OECD) **Seed Certification**

- The TSSOCA has initiated the registration of area under the schemes from Kharif, 2016
- During the year 2016-17 an quantity of 17159.00 quintals of seed was certified under OECD seed schemes
- During 2017-18 a quantity of about 7000 quintals of seed was certified
- For 2018-19, a quantity of about 7000 quintals of seed was certified
- · For 2019-20, a quantity of about 1000 quintals of seed was certified

Certification from 20				3 to	2020 (8550	(A)	
S. No	Season	Crop	Variety	Class	Qiy. Carnified (Qn)	Remarks	
1.	Kharif 2013	Moong	RMG-268	Pro- banic	Na	Crop failure due VMV (Velle Monsicrirus)	
2.	Robi 2013-34	Wheat	Raj-3765	Pre-	16.00	KVK Abutar	
3.	Robi 2014-15	Wheat	Raj-3765	batic	16.00	KVK Abutar	
4.	2015-16	Moong	IPM 1-3	Pre- banic	1.80	KVK Abutar	
5.	Rahi 2016-17	Rape Seed (Hyula)	PAC-482	C/S		1. UPL Reward Production Agency. 2. Exported RUSSIA	
۴.		(Hysia)	PAC-40	C.S	About 1402Qts Raw Seed Shifted to TSSOCA for Purther Certification sofivities	1. UPL Rewari (Production Agency	
9.	Rabt 2017-18	Bybrid Manard	CORAL (PAC- 437)	c.s	About 250 Qt: Row Seed Shifted to TSSOCA for Further Certification activities	1. UPL Reward (Production Agency	
8.	Rabi 2018-19	Wheat	RAJ- 3765	Entic	1 sore area about 12 quintats	BARI, Durgapura, Jaipur	
9.	R481 2019-20	Hyola	PAC-405	G/S	250 quintels	UPL Rewari	
10.	R481 2020-21	Hutte	PAC-401	C/5	150 scres proposed	UFL Reward	



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# SEED CERTIFICATION SYSTEM IN INDIA

#### Dr. Sadashiva V,

# Director,

Karnataka State Seed and Organic Certification Agency, Bangalore, Karnataka

#### History of Seed Certification in India:

In 20<sup>th</sup>century, the newly developed varieties lost their identity due to genetic contamination and mechanical mixtures. To avoid this, agronomist and breeders started visiting the fields of progressive farmers and educated them to avoid mechanical mixtures and keep the seed genetically pure. This process slowly led to field inspection.

The farmers and the scientists thought that field inspection could be useful in maintaining genetic purity of crop varieties. But other problems started like to what extent the mechanical mixtures or genetic contamination should be permitted etc.

To overcome these problems representatives from USA and Canada met in Chicago Illinois in **1919** and organized the **International Crop Improvement Association (ICIA)**.

The ICIA, which later in **1969** changed its name to **Association of Official Seed Certification Agency (AOSCA)**, laid the beginning of modern day seed certification.

The field evaluation of the seed crop and its certification started with the establishment of **National Seeds Corporation in 1963.** 

A legal status was given to seed certification with the enactment of first **Indian Seed Act in the year 1966** and formulation of **Seed Rules in 1968**. The Seed Act of 1966 provided the required impetus for the **establishment of official Seed Certification Agencies by the States.** 

Maharashtra was the first State to establish an official Seed Certification Agency during 1970 as a part of the Department of Agriculture, whereas **Karnataka was the first State to establish the Seed Certification Agency as an autonomous body during 1974.** 

At present <u>22 States in the country have their own Seed Certification Agencies</u> under the Seed Act, 1966.

In India, seed certification is voluntary and labelling is compulsory.

#### Seed Certification:

Seed Certification is a legally sanctioned, scientifically and systematically designed process to secure, maintain, multiply and make available to farmers, seeds of superior plant varieties, so grown to ensure genetic purity, physical quality, high germinability and free from pest and diseases.

#### Purpose of Seed Certification:

To maintain and make available to the farmers, through certification, high quality seeds and propagating materials of notified kind and varieties. The seeds are so grown as to ensure genetic identity and genetic purity.

#### **Certification Agency:**

Certification Agency shall be formed under <u>Section-8 of the Seed Act, 1966</u>. **Principles of forming Seed Certification Agency:** 

- Seed Certification agency should be an **autonomous body**.
- Seed Certification Agency should not involve itself in the production and marketing of seeds.
- The **Seed Certification Standards and Procedures** adapted by seed certification agency should be **uniform**, throughout the country.
- Seed Certification Agency should have **close linkage** with the **technical and other related institutions**.
- Seed Certification Agencyshould be to **operate** on **no-profit no- loss basis**.
- Adequate trained staff in seed certification should be maintained by the Certification Agency.
- It should have **provision for creating adequate facilities** for ensuring timely and through **inspections**.
- It should serve the interests of seed producers and farmers/growers.

#### **Objective of Seed Certification:**

The **main objective of the Seed Certification** is to ensure the acceptable standards of seed viability, vigour, purity and seed health. A well-organized seed certification should help in accomplishing the following three primary objectives.

- The systematic increase of superior varieties
- The identification of new varieties and their rapid increase under appropriate and generally accepted names.
- Provision for continuous supply of comparable material by careful maintenance.

#### **Eligibility for Certification of Crop Varieties:**

- Seed of only those varieties which are notified under <u>Section-5 of the Seed Act, 1966</u> shall be eligible for certification.
- Breeder seed is exempted from certification.
- Foundation and Certified seeds come under certification.

# Seed Production System:

The Indian seed programme largely adheres to the **limited generations system** for seed multiplication in a phased manner.

The system recognizes Three generations namely **Breeder**, **Foundation and Certified seeds** and provides adequate safeguards for quality assurance in the seed multiplication chain to maintain the purity of the variety as it flows from the breeder to the farmer.

#### Generation System of Seed Multiplication:

The choice of a proper seed multiplication model is the key to further success of a seed programme which basically depends upon,

- a. The rate of genetic deterioration
- b. Seed multiplication ratio
- c. Total seed demand

# Three - Generation model

Breeder seed - Foundation seed - Certified seed Breeder Seeds:

- Breeder seed is the **progeny of nucleus seed** of a variety and is produced by the originating breeder or by a sponsored breeder.
- Breeder seed production is the mandate of the Indian Council of Agricultural Research (ICAR) and is being undertaken with the help of;
- 1. ICAR Research Institutions, National Research Centre and All India

Coordinated Research Project of different crops;

- 2. State Agricultural Universities (SAUs)
- 3. Sponsored breeders recognized by selected State Seed Corporations
- 4. Non-Governmental Organizations.
- 5. National Seeds Corporation (NSC)
- 6. State Farms Corporation of India (SFCI),
- 7. State Seeds Corporation (SSCs),
- 8. KrishiVigyanKendras (KVKs) etc.
- Colour of the tag **Golden Yellow**
- Genetic purity percentage 99.99%

# Foundation Seed:

- Foundation seed is the **progeny of breeder seed** and is required to be produced from breeder seed or from foundation seed which can be clearly traced to breeder seed.
- The responsibility for production of foundation seed has been entrusted to the NSC, SFCI, State Seeds Corporation, State Departments of Agriculture and private seed producers, who have the necessary infrastructure facilities.
- Foundation seed is required to meet the standards of seed certification prescribed in the Indian Minimum Seeds Certification Standards, both at the field and laboratory testing.
- Colour of the tag White
- Genetic purity percentage 99% Certified Seed:
- Certified seed is the **progeny of foundation seed** and must meet the standards of seed certification prescribed in the Indian Minimum Seeds Certification Standards.
- In case of self-pollinated crops, certified seeds can also be produced from certified seeds provided it does not go beyond three generations from foundation seed stage-I.
- Colour of the tag **Azure blue/**recently changed to **stain blue colour**
- Genetic purity percentage for varieties-98% and hybrids-95%

# Eligibility Requirements for Certification:

Any variety to become eligible for seed certification should meet the following requirement:

- General requirements- should be a notified variety under Section-5 of the Indian Seed Act, 1966. Should be in the production chain and its pedigree should be traceable.
- Field standards Field standards include the isolation requirements, offtype, spacing, designated disease, planting ratio, border rows etc.
- Specific requirements Presence of off-types in any seed crop, pollen-shedders in Sorghum, Bajra, Sunflower etc., Shedding tassels in maize crosses, disease affected plants, objectionable weed plants etc., should be within the maximum permissible levels for certification.
- Seed standards Minimum seed certification standards have been evolved crop-wise.

# Phases of Seed Certification:

Seed Certification is carried out in six broad phases listed as under:

- 1. Verification of seed source, class and other requirements of the seed used for raising the seed crop.
- 2. Receipt and scrutiny of application.
- 3. Inspection of the seed crop in the field to verify its conformity to the

prescribed field standards.

- 4. Supervision at post-harvest stages including processing and packing.
- 5. Drawing of samples and arranging for analysis and also for Grow Outs Test

for foundation seeds to verify conformity to the seed standards

6. Grant of certificate, issue of certification tags, labelling, sealing etc.

# 1. Verification of Seed Source

- Source verification of parental seed used in seed production forms the basis for multiplication under seed certification programme.
- During first inspection of seed plot/farm the seed certification officer will verify whether the seed used to raise the crop is from an approved source.

# 2. Receipt and Scrutiny of Application

- Any person, who wants to produce certified seed, shall register his name with the concerned jurisdiction of Seed Certification.
- There are 3 seasons under certification *viz.*, Kharif (June- September), Rabi (October-January) and Summer (February- May).
- The applicant shall submit three copies of the application to the Seed Certification office 10 days before the commencement of the season or at least at the time of registration of sowing report.
- On the receipt of application, the concerned officers from Seed Certification Agency will verify the time limit, variety, eligibility and its source, the class mentioned, remittance of fee etc.,

# 3. Field Inspection

- Objective in conducting field inspection is to verify the factors which can cause irreversible damage to the genetic purity or seed health.
- **Crop stages for inspection** The number of field inspections and the stages of crop growth at which the field inspections should be conducted vary from crop to crop. It depends upon duration and nature of pollination of the seed crop.
- ✓ Pre flowering stage
- ✓ Flowering stage
- ✓ Inspection during Post flowering and pre-harvesting stage
- ✓ Inspection during harvest
- Assessment seed crop yield:

It is necessary to avoid malpractices at the final stage before harvest operation. The seed certification officer is expected to fix the appropriate seed yield.

• Liable for rejection report:

If the seed crop fails to meet with any one factor as per the standards, then seed crop is recommended for rejection and the signature of the producer is obtained and sent to Deputy Director of Seed Certification within 24 hours.

#### • Re-inspection:

For the factors which can be removed without hampering the seed quality, the producer can apply for re-inspection to the concerned Deputy Director of Seed Certification within 7 days from the date of first inspection order. For re-inspection the inspection charge is collected. It's allowed only for self-pollinated crops.

#### 4. Post supervision of seed crop after harvest:

The post-harvest inspection of a seed crop covers the operations carried out at the threshing floor, transport of the raw seed produce to the processing plant, pre-cleaning, grading, seed treatment, bagging and post processing storage of the seed lot.

#### 5. Sample to Testing Laboratory:

- Sampling of seeds for laboratory tests: The Certification Agency's concerned officer shall draw a representative composite sample as per procedure specified in Seed Testing Manual.
- The composite sample will be divided into three equal parts, and one shall be sent for analysis to a notified Seed Testing Laboratory (L), the second part to the Seed Producer (P) and retain the third part as a Guard Sample (G).
- The Seed Testing Laboratory shall analyse the seed samples in accordance with the prescribed procedure and deliver the Seed Analysis Report to the Certification Agency as soon as may be, but not later than 30 days from the date of receipt of the samples.

# Grow Out Test:

The Certification Agency shall conduct grow-out test to determine genetic purity of a seed lot wherever it is a pre-requisite for grant of the certificate and also on the seed lots where a doubt has arisen about the genetic purity.

# 6. Grant of certificate, issue of certification tags, labelling, sealing

#### etc.

- Certified seed should be tagged within 2 months from the date of test. A validity period of 9 months is given from date of test for the certified seed lots.
- If the seed lots are not sold or used within the validity period, then there is a procedure for validation i.e. extension of validity to certified seeds.

# Validity Period of the Certificate:

- The validity period shall be **nine months** from the date of test at the time of initial certification.
- The validity period could be **further extended for six months** provided on retesting seed conforms to the prescribed standards in respect of physical purity, germination and insect damage for all seeds except vegetatively propagating material for which lot shall be re-examined for seed standards specified for respective crop.
- A seed lot will be eligible for extension of the validity period as long as it conforms to the prescribed standards.

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# Seed Processing, Seed Testing & Packaging in Certification System

# B.S. Gupta

Former Senior Seed Certification Officer

Rajasthan State Seed & Organic Production Certification Agency, Jaipur

# • Seed processing is mainly a cleaning process of removing the undesirable material i.e. inert matter,

- weed seed, other crop seed, damaged seed, light & chaffy material from the raw seed by mechanical means to obtain the good quality seed.
- Seed cleaning is done on the basis of differences in physical properties of good & undesirable seed i.e.
- 1. Seed size (Length, width and thickness)
- 2. Weight/density
- 3. Shape
- 4. Surface texture
- 5. Colour

# Steps of Seed Processing

- 1. Receipt/intake of raw seed from the seed growers
- 2. Physical verification of raw seed
- 3. Approval of seed processing plant
- Drying (if necessary)
   Pre-cleaning preparing seeds for basic cleaning
- Fre-cleaning preparing seeds for basic ci
   Basic seed cleaning/ grading
- Basic seed cleaning/ grad
   Fine cleaning/grading
- 8. Sampling & submission of sample to STL
- 9. Seed treatment
- 10. Tagging & bagging
- Issue of Certificate II under Section-9 of the seed Act 1066
- 12. Storage- stacking of finally packed seed.

#### Receipt/intake of raw seed from the seed growers

- The seed production Agency accept the raw seed from the seed growers at the seed processing plant as per the estimated yield given by the certification agency at the time of final field inspection.
- Each and every bag of raw seed should have been marked the specific identification code of the seed grower including the name of crop, variety, class & stage of seed.
- The production agency check the moisture and physical quality of raw seed with reference to admixture of other crop seed, insect damage, inert matter and after weighing issue receipt to seed grower for the quantity received.
- No raw seed intake will be entertained by the certification Agency after the cutoff date as per the crop calendar

#### Physical verification of raw seed

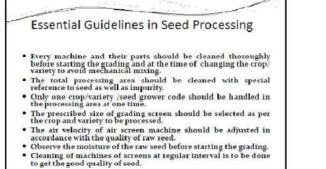
- Seed production Agencies submit the complete information of raw seed procured by them to the certification Agency just after the cutoff date of intake.
- Certification Agency after receipt of intake list of raw seed conduct the physical verification in order to check the information submitted by seed production Agencies.
- Only physically verified stocks are entertained for further seed processing and certification.

#### Approval of seed processing plant

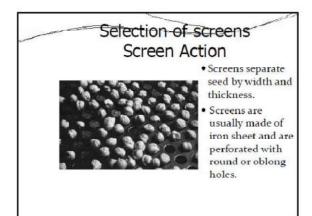
- The Production Agency after physical verification of raw seed request for the registration/renewal of their seed processing plant.
- The certification agency conduct the inspection of seed processing plant and evaluation is done with reference to essential parameters specified for the registration/renewal of evaluation. (format)

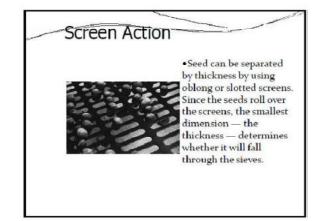
#### Essential requirements for the registration/ renewal of seed processing plant. (A) <u>Building</u> - The plant should be installed in a suitable building in a spacious processing shed having sufficient light facility, exhaust and ceiling fans, sufficient working space to provide storage of raw seed around the machine and to keep the graded seed, space for packating and movement of working personals. (B) <u>Storage</u> - Sufficient and separate godowns for storage of each category of seed i.e. raw seed, graded seed, packed seed, under size seed, packing material & chemicals. (C) <u>Machine & other equipmentis</u>. The processing machinery should have a sed of standard equipment i.e. predestore, seed gradet, indentied cylinder, gravity separator, seed treator, moisture meter, bag closer, vaccum cleaner, weighting machine. All the machines should be to aranged that seed flow continuously from

During the inspection of seed processing plants marks is to be allotted for specified essential requirements. Out of total 100 marks minimum 60 marks is required for the

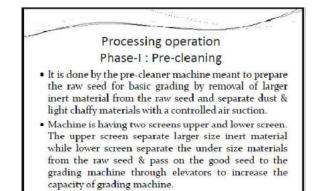








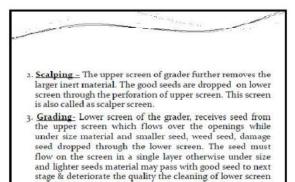
	-	eed Components Po um thickness/dia.c	
Shape of	seed	Upper screen	Lower screen
Round	ł	Round holes	Slotted holes
Oblon	g	*Slotted holes	Slotted holes



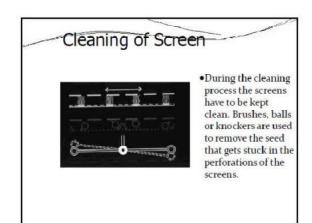
#### Processing Phase-II : Basic grading

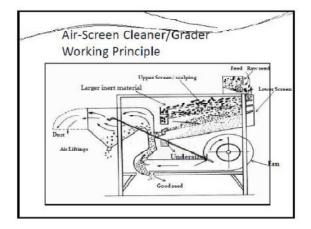
- It is done by the seed grader machine which is designed for essential process of grading on the basis of differences in the seed size and weight.
- The process of grading is operated in three ways.
- <u>Air suction -</u> The grading machine have two air systems (suction fans) designed as upper and lower air suction.
   (a) The upper air removes dust and light chaffy materials from the taw seed before they reach the upper screen. It is controlled by an air adjustable system.

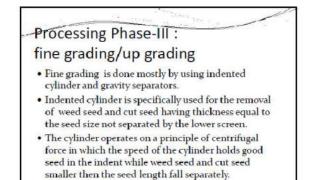
(b)The lower air suction removes the light seed and trashes from the lower screen. It is also controlled by an air adjustable system.



should be done at regular interval to avoid the clogging.







# Indented cylinder separator



 It consists of indents which lift particles that fit into the indents. Particles which do not fit drop out of the cell and fall downwards.

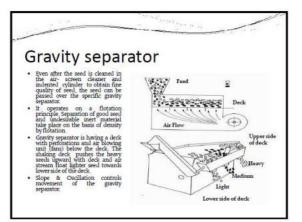
ood seed

Cut seed, Weed seed

#### Specific gravity separation



• Even after the seed is cleaned in the air-screen cleaner and the indented cylinder, it may be necessary to obtain higher-quality seed. In such cases, the seed can often be passed over the specific gravity separator.

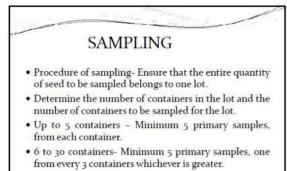


#### Formation of seed lot

- After grading certification Agency allot the lot no. to each identification code of fat In the IMSCS a procedure for formation of lot no. is defined to induce traceability of
- First part- Called the harvesting month & year code- for ex., NOV. 15 it explain the crop is harvested in the month of NOV aces
- Called the production location code and indicate State or Union Territory w d field was located for this purpose each State is allotted a permanent numer Rajasthan State the numerical code is 20. CSCB For R
- ode and shall indicate me tion Agency allot the regist ion Agency. For ex. Durgap ird par nt no
- 4. Fourth parts The seed production regimes, for etc. Longaptura, juput allotted plant no. 4. Fourth parts The seed product code it will indicate ultimate serial no. of an individual lot based on the tere seed of particular seed grower it e. on. All the four parts in the lot no shall be written in series with a dash (-) between First, Second, Third & Lot no: NOV iso science. NOV iso Seed charaveted in NOV acog as Seed corporated in Regarding plant on Durgspura, Jaipur or Seed processing land code taxes to the particular farmer.

#### Seed lot size

- Seed lot Is a physically identifiable quantity of seed which is homogeneous.
- The seed equal to the size of wheat the maximum size of seed lot will be 200 Qtls. subjected to the tolerance limit of 5 %
- The seed less than the size of wheat the quantity maximum size of seed lot will be 100 Qtls. subjected to the tolerance limit of 5 %
- The seed more than the size of maize the maximum size of seed lot will be 400 Qtls. subjected to the tolerance limit of 5 %



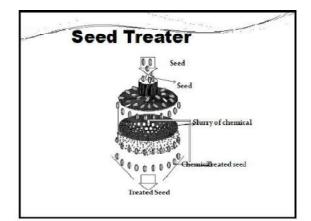
• More than 30 - Minimum 10 primary samples, one from every 5 containers whichever is greater.

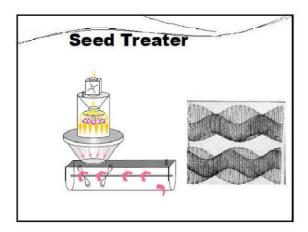
#### Sampling & submission of sample to STL

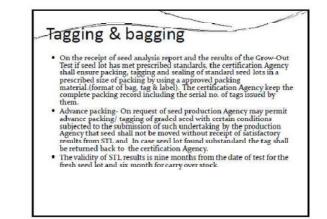
- The soon after completion of the seed processing certification Agency shall draw a representative composite sample as per procedure specified in Seed Testing Manual.
- similation and a representative composite sample as per proceedure specified in Seed Testing Manual. The quantity of seed samples so drawn shall be sufficient to provide three samples of the size of submitted sample. The composite sample will be divided into three equal parts and one shall be sent for analysis to a notified Seed Testing Laboratory, the second part to the seed producer and retain the third part as a guard sample. Primary sample Several individual samples are drawn from the different containers each such sample is called a primary sample. Composite sample All the primary samples drawn from one lot are combined to form a bulk and is called composite sample. Submitted sample. A portion of seed derived from the composite sample to be submitted for analysis to Seed Testing Laboratory is called submitted sample. After proper sealing the sample is to be sent to STL.

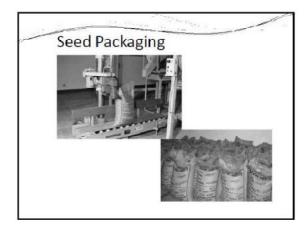
#### Seed treatment

- The application of chemicals i.e. fungicides, insecticides or a combination of both, to seed so as to disinfect them from seed or soil borne pathogens and storage insects & pests.
- Method of seed treatment-
- Dusting- The chemicals and seed are thoroughly mixed by mechanical mixer normally at the rate of 200 to 300 gm. Chemical/qtls.
- 2. Slurry- In this method the fungicide is applied to the seed in a soup like water suspension which is mixed with the seed in a special slurry treater. All foundation class seeds shall invariably be subjected to such treatment. To prepare a slurry 200 gm. of chemical mix with the 600 ml. of water for each qtls. of seed



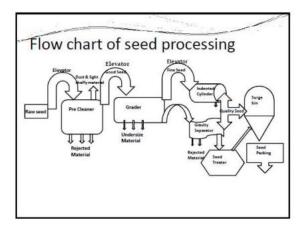


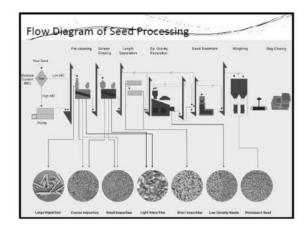


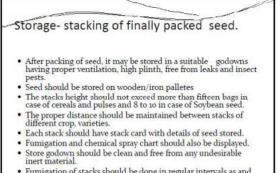


#### Issue of Certificate -II under Section-9 of the seed Act 1966

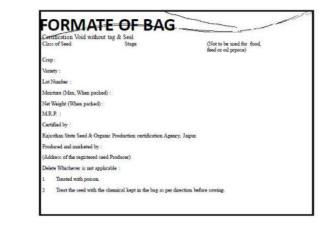
- On completion of all certification work the certification Agency shall issue a certificate II under Section-9 of the seed Act 1966 for each seed lot indicating all the required information's, regarding the seed standards, validity and serial no. of tags issued to that particular lot with the detail of seed producer.
- Now the seed shall be marketed by seed production Agency.





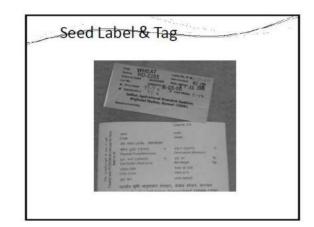


 Fumigation of stacks should be done in regular intervals as and when required.



#### SPECIFICATION FOR THE LABLE

•	TO BE PRINTED BY PRODUCTION AGENCY)
1	The length and breadth of the label shall be 15 X 10cm or proportionately small label may be used.
2	The content of the mark or label shall contain the following information namely.
a.	Label No.
Ъ.	Kind
c.	Variety
d.	Lot Number
e.	Date, Month and Year of test
f.	Valid up to
g.	Germination (Minimum )
h.	Physical Purity (Minimum)
b. c.d. e. f. g.h.i. j.k.l. m.	Genetic Purity (In case of variety) (Minimum)
j.	Net Weight
k.	Moisture, When packed, (Max)
1.	Name of the chemical used for seed treatment, If seed is treated
m.	Name and address of the person who offers for sale, sells or otherwise supplies the seed
n.	
o,	If seed has been treated, the following statement shall also be printed on the label
	"DO NOT USE FOR FOOD, FEED AND OIL PURPOSED"
1	The caution for mercurial's and similarly toxic substance shall be POISON in type size, Prominently displayed on the label in RED.
2	The colour Should resemble with opal green (IS No. 275).





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# Seed quality assurance system in India

# Nakul Gupta

# ICAR-Indian Institute of Vegetable Research, Varanasi 221305, U.P.

Quality seeds make possible higher production and productivity of crop, avoid the spread of the disease form one place to another place, enhance the income of the farmer and stabilize any seed company in the market. Quality seed alone can lead to 15-20 % increase in productivity. Therefore, it is of paramount important to monitor and test the quality of the seed viz. genetic purity, physical purity, germination, vigour, moisture content and seed health before its marketing and distribution.

Seed quality control and assurance is the combined efforts and activities undertaken to ensure that the seeds that are being produced for the end users (farmers) conform to mini- mum quality standards

#### Seed quality parameters and their testing:

- a. Genetic Purity- Genetic purity means trueness-to-type of the seed lot. It is important to assure the genetic identity which makes cultivars distinct. Genetic purity is best evaluated through a field trial in which the percentage of off-types in a seed lot is determined. Seed companies typically conduct variety trials each season to evaluate the genetic quality of contract lots; ideally, the seed lot is evaluated in comparison to the parent stock seed lot and competitors' lots of the same variety. The results of these variety trials are made available to the grower; this information is used as a tool to guide on-farm selection of the plants in the seed crop so the seed produced from that crop is true-to-type.
- b. Physical Purity-Physical purity evaluation consists of a examination of purity percentage. The purity exam determines the percentages by weight of pure seed, other crop seed, weed seed, and inert matter in a sample. The contracting seed company typically defines the purity standard for a particular seed crop and communicates this standard to the grower.
- c. Germination percentage- Germination/ viability testing determines the percentage of live seeds in a sample that have the potential to produce normal seedlings under favorable germination conditions. All seed sold commercially be tested by a certified lab within six months of sale and must meet minimum germination standards that are set for each major crop group. Dormant seed do not germinate in this test even when viable. For that tetrazolium (TZ) test is a quick biochemical test that evaluates seed viability based on seed respiration. This test is useful as it measures the percent live seeds in a sample regardless of the seeds' dormancy status.
- d. Vigor testing: Vigor testing moves beyond a simple assessment of germination by evaluating how quickly seed germinates and whether the germinating seeds and developing seedlings are "normal" and robust in the early stages of growth. Vigor tests measure the potential for rapid, uniform emergence of seeds under a wide range of field conditions.
- e. Seed health testing: It indicates whether the seed carries diseases that will have a significant impact on the health and productivity of the crop. In addition to seedborne pathogens, many other non-pathogenic fungi and bacteria can grow on seed surfaces, and high populations can

reduce seed viability and vigor. Proper harvest, processing, and storage methods are key to avoiding storage mold in seed lots.

Since seed is a global commodity, seed testing methods should be uniform in all over the world. International seed Testing Association (ISTA) develop, adopt and publish standard procedures for sampling and testing seeds, and to promote uniform application of these procedures for evaluation of seeds moving in international trade. It also involves in ISTA Accreditation Programme, issuing Analysis Certificates exclusively by ISTA Accredited Laboratories (Orange International Seed Lot Certificate, Blue International Seed Sample Certificate), promotion of research, training, publishing information in all areas of seed science and technology and cooperation with related organizations.

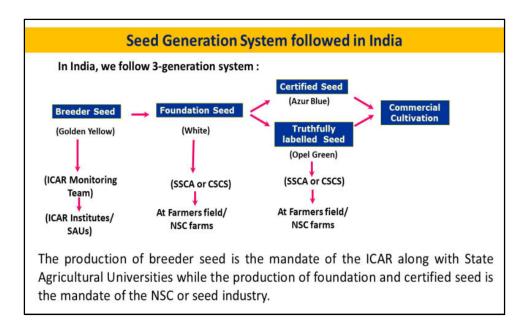
Plant breeders utilized the variability present in the population to breed new varieties which are high yielding, resistant to pest and diseases, better quality and high shelf life. Initial handful of seed which the breeder has in small quantity is referred as nucleus seed which is 100 % genetically pure. This is further multiplied through generation system so that it is made available to farmers as quickly as possible. During seed multiplication, varietal purity and identity needs to be maintained. Each multiplication cycle starts from the 'breeder seed'. If the breeder seed is not of high purity, the contaminants present get multiplied several times in the succeeding generations of foundation and certified seed production. The presence of contaminants may even lead to complete loss of the improved features of the cultivar. Prevention of contamination is a key to successful seed production programme. The quality of seed can be maintained by following generation system of seed production. Normally, In India three generation system of seed production is followed.

*Three-generation model:* Breeder seed - Foundation seed - Certified seed (cross-pollinated crops) eg: Cauliflower, Bottle Gourd, Radish, Turnip, Carrot, Palak, Onion

*Four-generation model:* Breeder seed - Foundation seed (I) - Foundation seed (II) - Certified seed (self-pollinated crops) eg; Brinjal, Tomato, Okra, Cowpea

*Five-generation model:* Breeder seed - Foundation seed (I) - Foundation seed (II) - Certified seed (I) - Certified seed (II) (may be adopted only in the event of an unfavorable/critical situation) eg: Garden Pea, Potato.

- Breeder seed is seed produced by the originating plant breeder or ICAR institution.
- Foundation seed is produced from breeder seed and is monitored by CSCA/SSCA.
- Certified seed is produced from either foundation seed and is monitored by CSCA/SSCA through certification. Certified seed crops must pass both field inspection and laboratory analysis.



The field must be planted from the proper class of seed, have appropriate isolation, and be free of problem weeds and diseases. After harvest, a sample of the seed crop must be sent to an official seed certification laboratory for germination and purity analyses. The seed must meet the standards set by the seed certifying agency. Seed that has passed the field inspection and the laboratory analysis can be tagged as Certified seed. In addition to the Certified tag there must also be an analysis tag with information on kind, variety, and quality parameters.

	Generation system of Hybrid seed production	
B	reeder Seed → Foundation Seed → C	Certified Seed
SI	Maintenance of SI line by Breakdown of SI	SI X SC
CGMS	<ul> <li>Maintenance of Parental lines</li> <li>A (Male sterile and use as female)- A X B</li> <li>B (Maintenance line)- Selfing</li> <li>R (Restorer line)- Selfing</li> </ul>	AXR
GMS	<ul> <li>Maintenance of Parental lines (Supression of Male sterility by temperature/ photoperiod/ chemicals)</li> <li>P1 – Selfing</li> <li>P2 - Selfing</li> </ul>	P1 X P2
HE	Maintenance of Parental lines by selfing	P1 X P2 By manual pollination

#### Seed certification

Seed certification is a legally sanctioned system for quality control of seed multiplication and production. Seed certification ensures that all seeds of the notified variety/kind sold in the country are of high physical and genetic purity, freedom from weed seeds and diseases and good germinability. It is carried by the state seed certification agency and National Seeds Corporations where the seed certification agency doesn't exists. The legal sanctity for seed certification is provided by Seeds Act, 1966 according to this Act, all the seeds of notified varieties/kinds when sold to farmers must meet the minimum standard of germination and physical purity. The seed should be packed in a suitable container and a label has to be affixed on the container. Information about germination, physical purity, variety, date of test, name of the seed producer has to be given on the label.

# History of Seed Certification in India

- The field evaluation of the seed crop and its certification started with the establishment of National Seeds Corporation in 1963.
- A legal status was given to seed certification with the enactment of first Indian Seed Act in the year 1966 and formulation of Seed Rules in 1968. The Seed Act of 1966 provided the required impetus for the establishment of official Seed Certification Agencies by the States.
- Maharashtra was the first State to establish an official Seed Certification Agency during 1970 as a part of the Department of Agriculture, whereas Karnataka was the first State to establish the Seed Certification Agency as an autonomous body during 1974.

In India, seed certification is voluntary and labelling is compulsory.

# **Objectives of seed certification**

The objective of seed certification is to ensure genuineness and quality of seed to the purchaser well-organized seed certification system helps in accomplishing the following three primary objectives of seed programmes

(a) The systematic increase of superior varieties

(b) The identification of new varieties and their rapid increase under appropriate and generally accepted names

(c) Provision of a continuing supply of comparable material by careful maintenance.

**Certification agency** - Certification shall be conducted by the Certification Agency notified under Section 8 of the Seeds Act, 1966. Presently there are 22 seed certification agencies in India. In those states where the seed certification agency is not present National Seeds Corporation is entrusted the responsibility of seed certification.

**Eligibility requirements for certification** - Seed of only those varieties which are notified under Section 5 of the Seeds Act, 1966 shall be eligible for certification. Any variety to become eligible for seed certification should meet the following requirement:

**General requirements** -The variety and the kind must be notified under section-5 of the Indian Seed Act, 1966. Should be in the production chain and its pedigree should be traceable.

**Field standards** - Field standards include the selection of site, isolation requirements, spacing, planting ratio, border rows etc.

**Specific requirements** - Presence of off-types in any seed crop, pollen-shedders in Sorghum, Bajra, Sunflower etc., Shedding tassels in maize crosses, disease affected plants, objectionable weed plants etc., should be within the maximum permissible levels for certification.

**Seed standards** – Indian minimum seed certification standards (IMSCS) have been developed by crop-wise.

# Certification shall be completed in six broad phases:

- 1. Receipt and scrutiny of application
- 2. Verification of seed source
- 3. Field inspections to verify conformity to the prescribed field standards.
- 4. Supervision of post-harvest stages including processing and packing
- 5. Analysis of seed samples including genetic purity and seed health test
- 6. Grant of certificate and certification tags, tagging and sealing

#### **Process of Seed certification**

**1) An Administrative check on the origin of the propagating material:** Source seed verification is the first step in Seed Certification Programme. Unless the seed is from approved source and of designated class certification agency will not accept the seed field for certification, thereby ensuring the use of high quality true to type seed for sowing of seed crops.

**2)** Field Inspection: Evaluation of the growing crop in the field for varietal purity, isolation of seed crop is to prevent out-cross, physical admixtures, disease dissemination and also ensure crop condition as regards to the spread of designated diseases and the presence of objectionable weed plants etc. the number of field inspection depends on the nature of pollination and generally two field inspection in self pollinated crops, three field inspection in cross pollinated crops and Four field inspection in hybrid seed production are carried out. The stage of inspection is also very important. Normally, field inspection are out at least three stages vegetative stage, flowering stage and maturity stage,

#### **Field Counts**

Field inspections are made of the standing seed crops, while later inspections and tests are done in the laboratory on representative seed samples collected from seed lots. These lots come from the seed crops that were inspected and approved at the field stage for conformity to prescribed standards. The primary objective of field inspections is to confirm that seed produced from a crop grown for seed purposes, is of the designated variety, and that it has not been contaminated genetically and/or physically beyond certain specified limits. Representative sample of plants taken at random from a seed plot for recording the observation on off types, pollen shedders, diseased plants, inseparable other crop plants

Area of the field in acres	Number of counts to be taken
Up to 2 ha	5
Above 2, up to 4 ha	6
Above 4 ha, up to 6 ha	7
Above 6 ha, up to 8 ha	8
Above 8 ha, up to 10 ha	9

Table1: The number of counts taken for different area under seed certification

Note: One additional count is taken for every additional 5 acres (2 ha)

**Number of plants /count** - Number of plants/heads or tillers to be included in one count depend upon the type of crop involved.

# Table 2. Number of plants/count to be taken for various crops.

Crops	Number plants/count
Okra, Eggplant, Capsicum, Chilli, Tomato, Potato, Cole crops, Cucurbit Root crops, Bulb crops Castor, Cotton, Groundnut, Maize, Pigeo pea,Sunflower	
French beans, Dolicas beans, Cluster beans, Cowpea, Garden pe Fenugreek, Spinach, Amaranth, Vegetable Mustard, Coriander	500 Plants
Lucerne, Berseem, Jute, Soybean	1000 Plants

#### 3) Conduct of lab test for seed quality evaluation:

If the seed plot passes the field requirement. The seed inspector draws the sample of the seed lots and sends it to the seed testing laboratory where the seed quality tests are done.

#### Phases of Seed Certification

Seed Certification is carried out in six broad phases listed as under:

- 1) Receipt and scrutiny of application
- 2) Verification of seed source, class and other requirements of the seed used for raising the seed crop
- 3) Inspection of the seed crop in the field to verify its conformity to the prescribed field standards
- 4) Supervision at post-harvest stages including processing and packing
- 5) Drawing of samples and arranging for analysis to verify conformity to the seed standards
- 6) Grant of certificate, issue of certification tags, labeling, sealing etc.

#### Validity Period of the Certificate

The validity period shall be nine months from the date of test at the time of initial certification. The validity period could be further extended for six months provided on retesting seed conforms to the prescribed standards in respect of physical purity, germination and insect damage for all seeds except clonally propagating material for which lot shall be re-examined for seed standards specified for respective crop. A seed lot will be eligible for extension of the validity period as long as it conforms to the prescribed standards.

#### References

Indian Minimum Seed Certification Standards. http://agricoop.nic.in/imagedefault/seed/INDIAN\_MINIMUM\_SEED\_CERTIFICATION\_ST ANDARDS.pdf NSRTC. <u>www.seednet.gov.in</u>

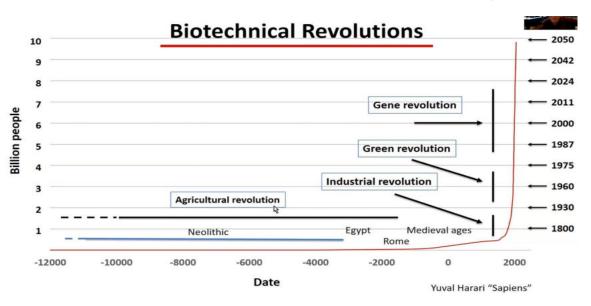
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# Modern Techniques for Testing of Transgenic Plants and Biosafety Issues

Dr. Aravind Kumar Konda Scientist (Biotechnology)

ICAR-Indian Institute of Pulses Research, Kalyanpur, Kanpur-208024

Sustaining an ever-growing population has proven to be a formidable challenge, but various advancements and revolutions throughout human history have played a role in bringing this objective within reach. An examination of history reveals that the human population began to increase only after humans began cultivating crops. As a result, they began to transition from a nomadic lifestyle to a settled existence in various regions that were appropriate for crop production, which ultimately led to the formation of civilizations. On the contrary, an abrupt increase in population occurred during the industrial revolution and persisted until the green revolution, as illustrated in the accompanying figure. Man initiated the domestication of untamed species during the agricultural revolution by selecting suitable plant varieties and combining their characteristics via crossing. This domestication has produced commodities in the new world that are easier to process, more productive, and edible than their untamed counterparts. This is supported by phenotypic alterations from the progenitor species to the current form, particularly in crops such as maize, wheat, and rice, the three most significant sources of carbohydrates for humans. The cereals underwent selection with a greater emphasis on production traits (e.g., increased grain yield, increased productive portions per plant) as opposed to survival traits (e.g., seed dispersal efficiency, thick protective seed coats). Throughout the green revolution, which commenced in the mid-20th century, ongoing endeavors were made to select plants that exhibited greater responsiveness to inputs including fertilizers, water, and so forth. Additionally, significant emphasis was placed on developing genetic resistance to both biotic and abiotic factors that posed challenges to crop cultivation. Thus, intensive agriculture gained traction and resulted in the development of genotypes that exhibited enhanced environmental adaptability and resistance to limiting stresses. Numerous novel plant breeding techniques were implemented in conjunction with established methods, including tissue culture-induced somaclonal variations, wide hybridization aided by embryo rescue and other inventive methods to surmount post-fertilization obstacles, induced variability via mutation breeding, and somaclonal variations induced through tissue culture, among others. However, crossability barriers imposed restrictions on the ability to transfer and combine characteristics from distinct genetic stocks, particularly between unrelated genera. The gene revolution has effectively resolved this challenge by enabling the transfer of desired regulatory sequences and genes into cultivated genotypes, thereby imparting desired traits through a process known as genetic transformation. Thus, modern biotechnology offers a novel instrument for the extremely precise reproduction of plants and animals.



#### Fig. 1 Different revolutions that have helped in sustaining human population growth

Transgenic plants are defined as those in which genetic engineering techniques have been used to modify the DNA of the plants in order to introduce a non-native trait. Therefore, a transgenic plant is composed of one or more genes that have been inserted artificially via transformation techniques. Once it has been determined that the introduced (engineered) genes are stably integrated, exhibiting the desired phenotype, and conferring it, the plant is deemed transgenic. Therefore, genetically modified (GM) crops exhibit characteristics of specificity and accuracy regarding modifications introduced, provide the flexibility to determine the incorporation of desirable qualities into well-adapted varieties with minimal disruption, and undergo a rapid process in comparison to traditional breeding methods. To sustain an expanding population, produce more desirable products, and generate plants with desired characteristics

#### A brief history of transgnenic plants

- In 1983, first genetically engineered (antibiotic resistant) plant tobacco developed
- In 1987, Plant Genetic Systems, founded by Marc Van Montagu and Jeff Schell, genetically engineered insect-resistant (tobacco) plants by incorporating cry genes from *Bacillus thuringiensis (Bt)*.
- In 1994, FlavrSavr tomato produced by silencing the polygalactouronase gene, the first genetically modified crop approved for sale in the U.S.
- In 1995, Monsanto introduced the NewLeaf variety of potato, its first genetically modified crop. Resistant to Colorado potato beetle due to cry genes.
- In1996, 1st genetically modified flower moon dust, bluish coloured carnation, was introduced by inducing the gene for delphinidin production from petunia and transferred to the carnation.
- In 2000, Vitamin A-enriched golden rice, developed by the addition of three beta-carotene biosynthesis genes namely 'psy' (phytoene synthase) from daffodil plant and 'crt-1' gene from the soil bacterium Erwinia auredorora and 'lyc' (lycopene cyclase) gene from wild-type rice endosperm.
- In 2013, Robert Frarley, Mark Van Montagu and Marry Dell Chilton were awarded world food prize for developing first transgenic plant.

(e.g., increased yields, crops that endure pests and diseases, and increased shelf life), transgenic plants are required. In order to introduce a new function (gain of function approach), a fully self-

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contained unit of function (gene along with its own regulatory sequences such as promoter, terminator, and enhancer) could be introduced into transgenic plants; likewise, to eliminate a trait (loss of function approach), post-transcriptional silencing or knockout of the existing gene could be utilized. As a boxed item, a concise history of the transgenic plants is included.through transformation procedures. The plant is considered transgenic only after it is established that the introduced (engineered) gene(s) are stably integrated, expressing and conferring the expected phenotype. Thus genetically modified (GM) crops are specific and precise with respect to changes made, allows flexibility to decide addition of traits into existing well adapted varieties with least disturbance and it is a fast process compared to conventional breeding approaches. Transgenic plants are needed to produce plants with desired traits including increased yields, crops that last longer and withstand pests and diseases, to feed the growing population and to produce more desirable products. Transgenic plants could be produced to introduce a new function (gain of function approach) by introducing a completely self contained unit of function (gene along with its own regulatory sequences such as promoter, terminator and enhancers) or to eliminate a trait (loss of function approach) by silencing post-transcriptionally or by knocking out the existing gene. A brief history of the transgenic plants is provided as box item.

In accordance with the genetic elements employed in the transformation process, four distinct varieties of transgenic plants have been created: transgenics, in which the selected elements and/or gene are foreign to the recipient (e.g., Bt transgenics); cisgenics, in which the donor gene and all regulatory sequences of the transgene are from the same crop species or cross-breedable species as the host; and intragenics, where the donor gene and all regulatory sequences are identical copies of the host's native gene cassette, including their regulatory sequences integrated in the host plant in the normal-sense orientation; and intragenics, within this category, gene In this scenario, promoters and terminators of distinct genes can regulate gene-coding sequences (with or without introns), so long as the transgene-regulating genes are present in the same pool of cross-breedable genes and genome-edited plants. While the introduced gene constructs are temporarily stored within the plant, the modification they induce is permanent and inherited. Mendelian segregation ensures that introduced gene constructs are eradicated in subsequent generations, leaving the final product genotype devoid of any introduced genes.

**Principles of Genetic Engineering:** As previously stated, the process of developing transgenic organisms entails the in vitro manipulation of DNA through the utilization of recombinant DNA technology procedures and methods to generate new or novel gene constructs. Therefore, restriction enzymes and ligases are employed in order to generate recombinant DNA molecules from the isolated component sequences (including but not limited to the promoter, terminator, gene, and any cis elements such as enhancers, introns, transcript stabilizers, and so forth). After the recombinant molecule, which is typically a self-contained expression unit, has been synthesized, it is transported to the plant cell of interest to undergo integration, expression, and sustained inheritance.

**Steps in gene transformation:** Mainly there are four steps

- 1. Make the transgene construction. This is typically done in *E. coli* though other microbes may be used.
- 2. Deliver the intended DNA into cells of the recipient organism
- 3. Select transformed cells using selectable markers such as antibiotic resistance (tetracycline or kanamycin resistance), herbicide resistance (such as glyphosate, glufosinolate), or visual marker genes (*gus/gfp*) or through positive selection.
- 4. Regenerate whole organism from transformed cells and confirm for the integration, expression and stable transmission of the introduced gene cassette through different molecular methods.

**Transfer of recombinant DNA into plant cells:** Diverse methodologies exist for the introduction of the engineered gene construct into plant cells. In general, they are categorized as either biological or physical routes. As vehicles, the biological approach will employ alternative organisms, including plant viruses (with the substitution of certain native genes) and Agrobacterium (specifically, Agrobacterium tumefaciens and A. rhizogenes). Physical methods involve the utilization of physical phenomena for the purpose of delivery, as the name suggests. Several strategies, including microinjection, have been tested and validated; however, gene cannons, biolistics (particle delivery systems, or PDS), and electroporation (typically of protoplasts) are the most prominent examples in this category. Microlaser, Silica Carbide Fibers, and Pollen Tube Pathway.

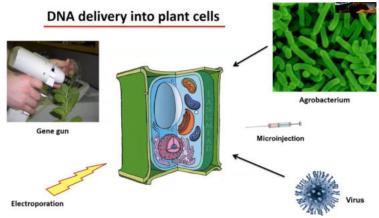
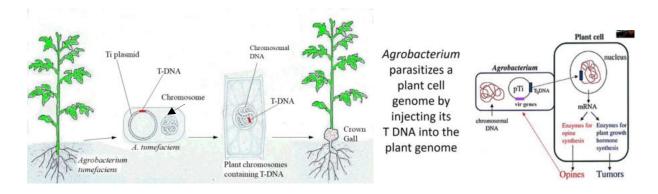


Fig. The main DNA delivery systems into plant cells

Agrobacterium-mediated transformation is widely utilized among the transformation methods and approaches owing to its ability to deliver typically one or two copies of the transgene construct in a clear and precise manner. Agrobacterium, commonly known as crown galls, is a soilborne pathogen that induces tumor development in a variety of plants. Agrobacterium was found to be the causative agent of this malignancy through the transformation of cells located in the crown region of the plant using a fragment of DNA, commonly referred to as T-DNA for transfer DNA. Left and right border sequences, which are 24–25 base pairs in length and distinguish T-DNA from other DNA, are responsible for limiting the sequence that is conveyed to plants. This T-DNA is a component of the megaplasmid carried by Agrobacterium; it is known as Ti-plasmid (for tumor-inducing plasmid) and it contains genes implicated in the production of opines and plant growth hormones. The continuous division of cells that receive T-DNA at the junction of the transformation process is attributed to the existence of growth hormone-producing genes, ultimately resulting in the development of a tumor. Furthermore, the transformed cells commence opinoid production and secretion; Agrobacterium will utilize these substances as an energy source. As a consequence, Agrobacterium is commonly referred to as "nature's genetic engineer," given that it modifies a plant cell to produce sustenance for itself.



# Fig:3. Agrobacterium, a soil bacterium that causes crown gall disease in plants, is a nature's genetic engineer. It processes and transfers a piece of DNA (T-DNA) from its plasmid and tansfoms plant cells into factories that make and export opines which in turn will be utilized by Agrobacterium as food.

By utilizing this inherent capability of Agrobacterium, transgenic plants have been produced. As specified by the breeder, the engineered or modified Agrobacterium will contain the desired gene in addition to selectable markers delimited by border sequences within the T-DNA. Subsequently, this T-DNA will be introduced into the plant. After entering the cell, the T-DNA proceeds to the nucleus, where it is arbitrarily integrated into the genome of the plant cell. It begins to manifest itself upon integration, allowing the transformed cells to withstand selection pressure and consequently become susceptible to selection.

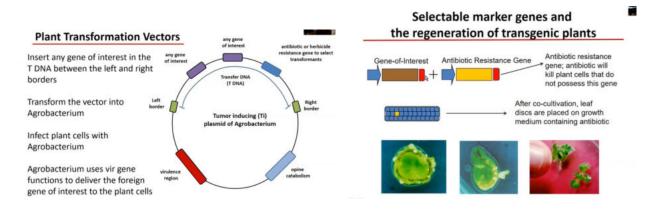


Fig.4 A typical plant transformation vector used in Agrobacterium mediated transformation and how the selectable marker genes are useful in allowing only the transformed cells survice and multiply during the in vitro selection procedure.

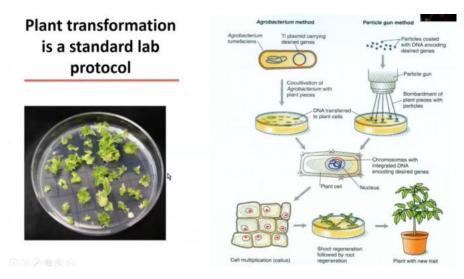


Fig.5 A typical plant transformation protocol followed in the lab

**Applications of transgenic plants:** Transgenic plants possess a vast array of potential uses, extending beyond the development of commercially viable traits to addressing fundamental inquiries into gene function. The emergence of numerous genomic instruments has led to a deluge of data concerning the function of genes and diverse gene networks in the development of characteristics. In order to determine the functional significance of the chosen genes, one may employ either a gain-of-function or loss-of-function strategy. This shall be accomplished through the generation of transgenics in which the gene function is nullified or by expressing the genes in a genotype devoid of it. Therefore, the development of transgenics has evolved into an essential component of functional genomics and has become a critical resource for molecular biologists conducting strategic, applied, or fundamental research. The following are several conspicuous instances where transgenic technology has been implemented in vegetation. This list is merely illustrative of the possible applications and is not exhaustive.

#### Imparting resistance to biotic stress

1) Insect resistance - eg: BT crops

2) Virus resistance -eg: rainbow papaya

#### Imparting resistance to abiotic stress

1) Herbicide resistance - eg: roundup ready soyabean

#### Improvement of crop yield and quality

1) Extended shelf life of fruits -eg: Flavr savr tomato

2) Improved nutrition -eg: Golden rice

3) Improved coloration -eg: Japanese blue roses

#### Production of low-cost pharmaceuticals

1) Edible vaccines -eg: Rabies virus g-protein in tomato

#### 2) Essential proteins -eg: Soy bean, maize

Insect resistance (e.g., Bt cotton, the only commercially transgenic crop cultivated in India), nutritional enhancement (e.g., rainbow papaya), virus resistance (e.g., rainbow papaya), herbicide resistance (e.g., roundup-ready crops), and delayed fruit ripening (e.g., flavr savr tomato) are the five most prominent examples that have been utilized across crops and countries. Thus far, the primary focus has been on modifying input traits. However, the forthcoming field-ready exploitation of second-generation transgenic plants is anticipated to enhance output traits. In the future, transgenic plants are anticipated to be utilized in the production of molecules that contribute to the betterment of humanity (Fig. 6). The primary transgenic crops that are cultivated globally consist of cotton, maize, soybeans, potatoes, canola, and brinjal. Numerous nations have implemented these crops for commercial purposes (see Figure 7). A comprehensive compilation of commercially viable transgenic crops and the specific traits that have been engineered is presented in a recent review by Kumar et al. (2020). An illustration from this review is depicted in the figure that follows (Fig. 8).

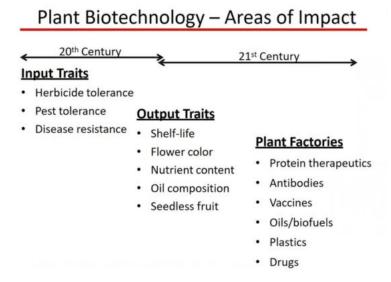
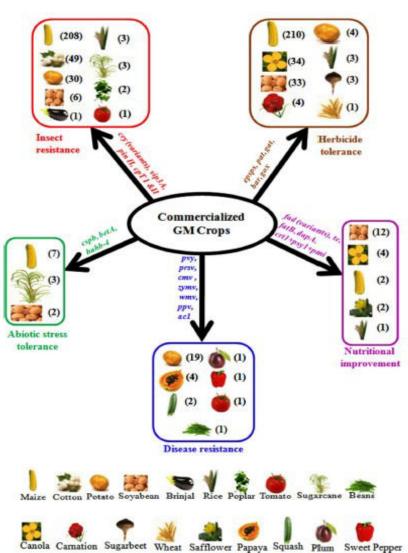


Fig. 6 Traits incorporated in transgenic crops

Crop Name	Countries
Cotton	Argentina, Australia, Brazil, Colombia, India, Mexico, Paragu ay, South Africa, USA
Maize	Brazil, Canada, Colombia, Cuba, European Union, Honduras, Paraguay, Philippines, South Africa, USA, Portugal, Vietnam
Soyabean	Argentina, Bolivia, Brazil, Canada, Chile, Costa Rica, Mexico, Paraguay, South Africa, USA, Uruguay
Potato	Canada, USA
Canola	Canada, USA, Australia, Chile
Brinjal	Bangladesh

#### Fig 7. Major transgenic crops and the countries growing them

Fig. 2 Diagrammatic representation of commercialised transgenic (GM) crops with various improved traits viz., insect resistance, herbicide tolerance, nutritional improvement, disease resistance and abiotic stress tolerance. Numbers in parentheses represent numbers of commercialised transgenic events in the particular crop. The traitwise major genes employed for GM crop development are given beside the arrows. cry (variants)-crystal proteins; vip3avegetative insecticidal protein; pin II-proteinase inhibitor II; cpt I and II-Cowpea trypsin inhibitor; esps-5-enolpyru-vylshikimale-3-phosphale synthase; par-phosphinothricin acetyltransferase; gar-glyphosate acetyltransferase; barbialaphos resistance; govglyphosate oxidoreductase; fad (variants)-flavin adenine dinucleotide; te-thioesterases; fatB-dapA-dihydrodipicolinate synthase; crtl-Calreticulin; psyl-phytoene synthase; pmi-phosphomannose isomerase; pvy-coat protein of potato virus Y: prsv-coat protein of papaya ring spot virus; cmv-coat protein of cucumber mosaic virus; zymv-coat protein of zucchini yellow mosaic virus; wmv-coat protein of watermelon mosaic virus; ppv coat protein of plum par virus; Ac1-encoding viral replication protein (Rep) from bean golden mosaic virus



(Source: Taken from Kumar et al., 2020)

# Fig 8. Diagrammatic representation of the main target traits manipulated, commercialized transgenic crops, number of events released and the major genes used for imparting the traits

# Molecular techniques for identification of transgenic plants

There are several molecular techniques used for testing transgenic plants. These techniques help in confirming the presence of the desired transgene and determining its expression level and stability. Here are some commonly used molecular techniques for testing transgenic plants:

1. Polymerase Chain Reaction (PCR): PCR is a widely used technique to amplify specific DNA sequences. It can be used to detect and confirm the presence of transgenes in plant samples. Specific primers designed to anneal to the transgene sequence are used to amplify the target DNA, and the amplified product is visualized on an agarose gel.

2. Southern Blotting: Southern blotting is a technique used to identify and characterize specific DNA sequences. It involves the transfer of DNA fragments from an electrophoresis gel to a solid support, typically a nylon membrane. The transferred DNA is then hybridized with a labeled probe specific to the transgene sequence, allowing for its detection.

3. Northern Blotting: Northern blotting is similar to Southern blotting but is used to detect and analyze RNA expression. It can be used to determine the expression level of the transgene in plant tissues by hybridizing the transferred RNA fragments with a labeled probe specific to the transgene mRNA.

4. Western Blotting: Western blotting is a technique used to detect specific proteins. It can be used to determine the expression and stability of transgene-encoded proteins in transgenic plant samples. Protein extracts from plant tissues are separated by gel electrophoresis, transferred to a membrane, and probed with a specific antibody against the transgene-encoded protein.

5. Real-time Quantitative PCR (qPCR): qPCR is a highly sensitive technique used to measure the expression level of a gene of interest. It can be used to quantify the amount of transgene mRNA in plant tissues, allowing for the evaluation of transgene expression levels.

6. Next-generation sequencing (NGS): NGS technologies enable high-throughput sequencing of DNA, RNA, or both. It can be used to obtain a comprehensive analysis of the transgenic plant genome and transcriptome, including the identification and characterization of transgene insertions and their expression patterns.

These molecular techniques are commonly used in combination to fully characterize and evaluate the presence, expression, and stability of transgenes in plants.

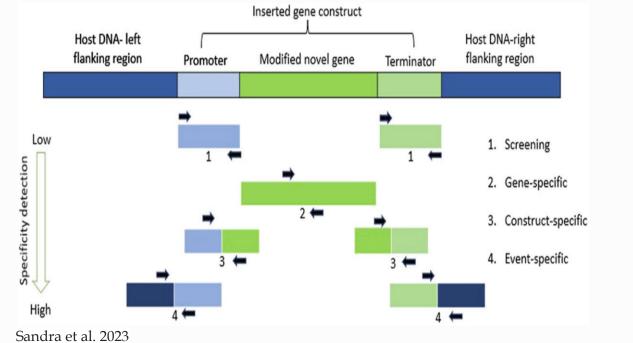
## Methods of GM Seed Testing

There are four different levels of GM assessment in seed samples as mentioned below:

• GM detection: Performing a seed screening in order to identify the existence of transgenic seedlings. Primers are employed to identify the presence of generic genetic elements (GMOs) in a

seed lot by detecting a general genetic element, such as the constitutive promoter or a selection marker, which is commonly present in all GMOs.

- Gene-specific detection: By utilizing primers that are specific to the gene sequences and testing for a particular transgene, it is possible to identify a particular GMO. The identification of a specific transgene being examined in the seeds is suggested by the presence of amplified fragments.Gene construct-specific detection: Specific gene constructs within a seed batch are identified. Primers specific to sequences of the promoter/terminator and a portion of the gene accomplish this.
- Event-specific detection: Detection of a gene event requires knowledge of the flanking sequences, or the sequence of the host genome that is in close proximity to the targeted gene construct. Utilizing primers designed to detect the unique integration site of a particular GMO, the event is identified.



Take home message from the discussion shall be:

- Transgenic technology has become an integral part of plant breeding programmes for precise modification of the genetic architecture. Genome editing a new tool in the armory.
- ✓ Transgenic plants are helpful in creating both gain of function and loss of function traits.
- Transgenic technology has led to greater understanding of the plant life, cellular mechanisms, physiology of traits, complicated gene regulations.
- ✓ We have come a long way from understanding to transform the organisms to exploiting the technology. Single gene of interest (GOI) to multiple genes (upto 30 kb) transferred and stably inherited. Even minichromosomes could be moved.
- ✓ Moved from input traits to making plants as factories to meet the human needs

- ✓ With so many programmes on integrated genomics now we have many candidate genes for imparting traits – even the complicated ones (QTLs)
- ✓ Still there is a need to identify suitable regulatory sequences to direct expression of GOI for realizing the intended phenotype in the plant
- ✓ Still there are limitations in realizing transgenic plants at will in crop plants.
- Social licensing is an issue that needs to be addressed for full exploitation of the TG technology

During the discussion, most of the examples of transgenic crops, traits manipulated and the methods of detection will be dealt in detail with suitable schematic diagrams, genes involved, methods followed and the present status. Examples will be taken from our lab experiences and they will be discussed at length to fix the concepts, principles and procedures. Also, during the course of the interaction class, the issues and concerns raised with respect to commercial release of the transgenic plants into the environment will be discussed.

## Further reading:

Kumar, K., Gambhir, G., Dass, A. *et al.* 2020. Genetically modified crops: current status and future prospects. *Planta* **251**, 91-97 (2020). <u>https://doi.org/10.1007/s00425-020-03372-8</u>

Low LY, Yang SK, Kok DA et al. 2018. Transgenic plants: Gene constructs, vector and transformation method. DOI:10.5772/intechopen.79369

Basso MF, Arraes FBM, Grossi-de-Sa M, et al. 2020. Insights into genetic and molecular elements for transgenic crop development. *Front. Plant Sci.* 11:509.doi: 10.3389/fpls.2020.00509 Sandra, N., Ravishankar, K.V. and Chidambara, B., 2023. Molecular Techniques for Testing Genetic Purity and Seed Health. In *Seed Science and Technology* (pp. 365-389). Springer, Singapore.

Reader is recommended to watch the following video for getting a good overview on plant biotechnology.

https://ucdavis.mediaspace.kaltura.com/media/Lecture+20+-+SSC+100+-+11+14+2016/0\_jvzav0jq

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# SEED CERTIFICATION THROUGH ONLINE USING SATHI PORTAL

Dr. Sadashiva V,

Director,

Karnataka State Seed and Organic Certification Agency, Bangalore, Karnataka

# SATHI – Seed Authentication, Traceability and Holistic Inventory

**SATHI** is a **digital chain** envisioned and created by **Department of Agriculture & Farmer's Welfare, Govt. of India** in partnership with **National Informatics Centre** (NIC) which will provide **holistic approach** to cover complete seed life cycle over **multiple seed generations** starting from nucleus to breeder seed, breeder to foundation seed, foundation seed to certified seed and distributing the seeds to farmers through licensed dealers.

# It Will

- Enhance and ensure the quality and purity of seed through complete digital platform of seed life cycle.
- **Increase** the **accountability** with the help of seed traceability through multiple modules.
- **Improve** the **on-field efficiency** of SCO by **reducing the human error** and integrating technology to the system.
- Generate GIS based MIS reports powered by Bharat Map Interface.
- **Reduce transactional time** for registration, approval, access to field inspection reports, lab testing reports and certification.

# **Salient Features**

Here are some of its salient features which will take seed production and inventory to new heights:

- GIS Report based on Bharat Map Interface.
- Provision of wallet service.
- Offline friendly and device agnostic mobile application.
- Quality inspection for the quality check of the inspection process.
- System generated sample slip on processed verification data.
- Online forwarding of the samples to seed testing laboratory.
- Issuing of tag certificate based on digital tag register.

# NUCLEUS TO BREEDER SEED MANAGEMENT:

- SATHI assists us to execute the following with ease by integrating the following to its system:
- Breeder Seed indent generation to ICAR, SAU, and Breeder Seed Production Centre.
- Registration of Breeder Seed Production Centre.
- Allocation of breeder seed through programmed functionality.
- Issuing of Breeder Seed Labels.
- Lifting of allocated breeder seed by indentors.

• Registration of indentors and submission of indents.

# BREEDER TO FOUNDATION AND CERTIFIED SEED CERTIFICATION:

SATHI allows you to access and handle all the data from any device, from anywhere by having the following features in its module:

- Registration of Seed Grower, seed producing agency & seed processing plant breeder to certified seed certifications:
- Verification of seed source, class and other requirements of the seed used for raising the seed crop.
- Field inspections to verify seed to the prescribed field standards using offline mobile app.
- Processing and verification of certified seed (can be merged and told that data will be uploaded into database.)
- Sampling, testing and issue of tag for foundation seed/certified seed.
- Billing and Accounting module to process information related to management of fees and pro forma updates.
- Permission module to allow SPAs/ SPPs for interstate permission, small size bag allocation, venturing with other marketing firms.
- Applying unique and distinct tags, QR code to the lots by the help of Tag Register.
- Reducing wastage of breeder seeds, foundation to certified seeds by Downgrading of Lot-Class.

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# SEED CERTIFICATION WITH REFERENCE TO THE SEEDS ACT, 1966 & THE SEED RULES, 1968

R. Harichandan, M.Sc(Ag.) Seed Certification Officer, OSSOPCA, Bhubaneswar E Mail: <u>singhamrh.seed@gmail.com</u> Phone No. 7205096115

**Seed means** any of the following classes of seeds used for sowing or planting: 1. seeds of food crops including edible oil seeds and seeds of fruits and vegetables; 2. Cotton seeds; 3. Seeds of cattle fodder; 4. Jute seed and includes seedlings, and tubers, bulbs, rhizomes, roots, cuttings, all types of grafts and other vegetatively propagated material, of food crops and cattle fodder.

Seed has been **adopted readily** as the basic input for crop production since the inception of agriculture and importance of good seed has been accepted for increasing crop production and productivity.

The **eternal nature** of agriculture has dignified seed as the most credible agri-input throughout the world and the scientific concept of seed quality has gathered momentum with the pace of time.

The **duality of demand** for quantity and quality of foodstuff and the undesirable impact of deteriorating climate are forceful events to expedite new dimensions of the seed system.

The fast growing **scientific developments** in various fields around the world are increasing the pre-existing vastness of agriculture and thus the seed sector is subject to modern challenges every day.

The seed to seed system being burdened with the **infinite expectations** of one and all is no more a mere barter business but encompasses volumes of experiments from ancient practices to modern drones.

The **concept of seed quality** and the importance of quality seed are known to the most ignorant farmer and thus it is a compulsion for all to make available the quality seeds as per the changing needs to satisfy the mass.

The **contrast variations** of the agro-climatic zones of the Indian sub-continent and the limitations in adaptability of the kinds or varieties are of grave concern for maintaining the tempo of revolution in agriculture.

The **evolution of the concept of seed quality regulation** in India has emerged from the situation of food shortage and passed through food surplus with the establishment of National Seeds Corporation and State Seeds Corporations.

With the enactment of the Seeds Act, 1966 and subsequent promulgation of the Seed Rules, 1968, the seed quality regulation through **compulsory labeling and voluntary certification** have been achieved to increase the production, supply and marketing of quality seeds in India.

Further, the declaration of **seed as an essential commodity** and the **compulsory seed dealer licensing** and law enforcement system boosted the seed quality control system in

addition to the seed quality assurance system achieved through compulsory labeling and voluntary certification.

Out of twenty five sections of the Seeds Act, 1966, five sections are related to seed certification. These are the Section 8, Section 9, Section 10, Section 11, and Section 18. Similarly, out of eleven parts of the Seed Rules, 1968, four parts are related to seed certification. These are Part IV, Part VI, Part VII and Part VIII. Some other sections of the Seeds Act, 1966 and some other parts of the Seed Rules, 1968 are helpful in understanding the seed quality regulation and seed certification.

## **Establishment of Seed Certification Agency**

As per the provisions of the Section 8 of the Seeds Act, 1966 the State Government or the Central Government in consultation with the State Government may, by notification in the Official Gazette, establish a certification agency for the State to carry out the functions entrusted to the certification agency by or under this Act.

Maharashtra was the first state of India to establish an official Seed Certification Agency during 1970 as a part of the Agriculture Department and Karnataka was the first state to establish Seed Certification Agency as an autonomous body during 1974. The Seed Certification Agency in Odisha started functioning from 1978. An up to date list of seed certification agencies can be obtained from any authentic source.

# Constitution of the Central Seed Certification Board

As per the Seeds (Amendment) Act, 1972 dated 9<sup>th</sup> September, 1972, there is provision of the Central Seed Certification Board to advise the Central Government and the State Governments on all matters relating to certification and to co-ordinate the functioning of the agencies established under section 8. The constitution, composition and function of the Board have been elaborated from Section 8A to 8E. The notifications related to constitution and proceedings of the Board can be obtained from any authentic source.

# **Evolution of Indian Minimum Seed Certification Standards**

The seed certification and seed testing are guided by the Indian Minimum Seed Certification Standards (IMSCS), the Hand Book of Seed Testing and the recently introduced Indian Seed Certification Working Manual.

As per the provisions of Section 6 (a) of the Seeds Act, 1966, the Central Government may, after consultation with the Committee and by notification in the Official Gazette, specify the minimum limits of germination and purity with respect to any seed of any notified kind or variety.

As per the provisions of the Section 7 (b) of the Seeds Act, 1966, no person shall, himself or by any other person on his behalf, carry on the business of selling, keeping for sale, offering to sell, bartering or otherwise supplying any seed of any notified kind or variety, unless such seed conforms to the minimum limits of germination and purity specified under clause (a) of section 6.

As per the provisions of the Section 9 (3) of the Seeds Act, 1966, on receipt of any application for the grant of a certificate, the certification agency may, after such enquiry as it

thinks fit and after satisfying itself that the seed to which the application relates conforms to the minimum limits of germination and purity specified for that seed under clause (a) of section 6, grant a certificate in such from and on such conditions may be prescribed.

As per the Seeds (Amendment) Act, 1972 dated 9<sup>th</sup> September, 1972, in sub-section (3) of Section 9, for the words, brackets, letter and figure " minimum limit of germination and purity specified for that seed under clause (a) of section 6", the words "prescribed standards" shall be substituted.

As per the provisions of the Rule 17 of the Seed Rules, 1968, there is provision for grant of certificate by the certification agency. Further, according to the Seed (Amendment) Rules, 1981 No. 18-48 / 81-SD dated 10th June, 1981, Rule 17-A has been inserted as: "The certification agency shall before granting the certificate, ensure that the seed conforms to the standards laid down in the Manual known as "Indian Minimum Seed Certification Standards" published by the Central Seed Committee, as amended from time to time."

The first IMSCS was published in the year 1971 and was made part of the Seed Rules, 1968 through the amendment No. 18-48 / 81-SD dated 10th June, 1981. These standards were subsequently revised in 1988 for rectifying few of the disparities and also incorporated contemporary information generated in seed technology research. Further the IMSCS have been revised in 2013 (Part A and Part B) incorporating several advancements in the field of seed science and technology. Subsequent developments through notification can be obtained from authentic source.

## Phases of Seed Certification

Out of 33 paragraphs of the General Seed Certification Standards of the IMSCS, 2013; Part VI describes the phases of seed certification according to which the seed certification works shall be completed in six broad phases such as 1. Receipt and scrutiny of application; 2. Verification of seed source, class and other requirements of the seed used for raising the seed crop.3. Field inspections to verify conformity to the prescribed field standards; 4. Supervision at post-harvest stages including processing and packing, 5. Seed sampling and analysis, including genetic purity test and /or seed health test, if any, in order to verify conformity to the prescribed standards and 6. Grant of certificate and certification tags, tagging and sealing.

# 1. Receipt and scrutiny of application.

The application form for seed production under the seed certification programme has been attached to the Seed Rules, 1968 as the Form I. With the passage of time several new requirements have been added to this application and in recent on-line system (SATHI) it has been tried to bring uniformity all over the country. The present application has three basic components such as: (a) Personal information of the seed grower, (b) crop information regarding the crop / kind, variety and class etc. and (c) the land / field information about the location of the seed crop.

Additional information / documents / requirements: Identity proof of the seed grower, land records, beneficiary list in case of group registration, payment details of registration and field inspection fees, signature of the seed grower and signature of the seed producer are the additional requirements submitted at the time of receipt and scrutiny of application.

The seed grower shall be an individual person or an organization / department like National Seeds Corporation or State Agriculture University or private seed companies who is desirous of producing certified seed in accordance with the procedures and standards of certification as per the provision of the paragraph III of IMSCS. There is no minimum or maximum limit for the area offered for certification, provided the certified seed production meets all the prescribed requirements. However, one unit of certification shall not exceed 10 hectors and this shall be ensured during receipt and scrutiny of application as per the paragraph VIII and IX of IMSCS.

# 2. Verification of seed source, class and other requirements of the seed used for raising the seed crop.

For verification of seed source used for seed production including class of the seed whether breeder, foundation or certified class has been used for raising the seed crop, one or more relevant evidence such as certification tags, seals, labels, seed containers, purchase / sale records etc. as may be demanded shall be submitted according to paragraph V and paragraph VII of IMSCS. The crop variety of the source seed shall be compulsorily be notified kind or variety notified under Section 5 of the Seeds Act, 1966 as per the provisions of the paragraph IV of IMSCS.

Further validity period of the source seed lot used for seed production and adaptability of the kind or the variety shall be taken into consideration. During verification of source, it shall be checked for three generation seed multiplication such as Foundation stage I to Foundation stage II to Certified stage I or Foundation stage I to Certified stage II. Production of Foundation stage I and II shall be approved by the certification agency. The production of Certified stage II may require approval of the certification agency. Since there is no scope for supervision by certification agency, it is faithfully accepted that the seed crop has been raised from the claimed source seed.

## 3. Field inspections to verify conformity to the prescribed field standards.

According to the paragraph XI of IMSCS, field inspection shall be performed by technically trained personnel authorized by the certification agency to verify those factors which can cause irreversible damage to the genetic purity or seed health of the seed. After completion of the field inspection, a copy of the report shall be handed over to the seed grower or his / her representative. In online system of field inspection, an auto generated report is available in the public domain for reference and a message shall be sent to the registered mobile of the seed grower for information and subsequent action. The IMSCS clearly describes the number of inspections to be carried out for different crops at different growth stages. The factors for which observations are taken during field inspection including isolation distance vary from crop to crop.

The provision of re-inspection has been provided in paragraph XII of the IMSCS where additional inspection over and above the minimum number of inspections may be taken up to ensure conformity of the seed crop to the prescribed standards. In case of re-inspection, a reinspection fee may be demanded by the certification agency. The Certification Agency has the authority to refuse certification of any seed production field that does not conform to the Minimum Standards prescribed for that particular crop according to the provision of the paragraph XXX of the IMSCS.

When a seed field is not found meeting the prescribed standards for the class for which it has been registered but conforms to the prescribed standards to the immediate lower class, the Certification Agency may accept such seed fields for certification to the immediate lower class provided request has been made to this effect by seed producer. However, downgrading of the seed class shall not be applicable in case of hybrids and their parents as per the paragraph XXVII of IMSCS.

## 4. Supervision at post-harvest stages including processing and packing.

The seed crop meeting field standards for certification shall be harvested (cutting of seed crop), threshed and transported to the seed processing plant in accordance with the guidelines issued by the certification agency. During these operations, the seed grower will take all precautions to safeguard the seed from admixture and other causes of seed deterioration as per the paragraph XIII of the IMSCS.

At this stage supervision is generally over looked at the seed grower's field / premises and the seed that is transported to the seed processing plant is supposed to be the faithful produce of the seed field certified. Again supervision at the seed processing plant is taken up either during receipt of the seed stock or during seed processing or both. Seed processing which includes cleaning, drying, treating, grading and other operations to improve the quality of the seed shall be done by using specified screens as per Appendix-VII and VIII of the IMSCS. The seed processing plants where there is no facility for drying, the seed stocks shall be received at approved seed moisture content.

The seeds shall be treated or a packet of seed treating material is placed inside the seed container as per the provision of paragraph XX of IMSCS. Further, the details of the seed treating chemical are depicted on the container and the label as per the Notification No. S.O. 767 (E) dated 6<sup>th</sup> November, 1991 under the Seeds Act, 1966.

The seed container shall be labeled as per the provisions of the clause (c) of Section 7 and clause (b) of Section 17 of the Seeds Act, 1966. The particulars of the label shall be as per the provisions of Section 6 (b) of the Seeds Act, 1966, the Notification No. S.O. 767 (E) dated 6<sup>th</sup> November, 1991 under the Seeds Act, 1966 and the Rule 8 of the Seed Rules, 1968. Apart from this label, certain information are written on the body of the seed container as per the provisions of the Rule 9 (3) of the Seed Rules, 1968.

It may be noted here that the name and address of the person who offers for sale, sells or otherwise supplies the seed shall be written on the label. Further, the name and address of the person who is responsible for its quality shall be written on the label as per Rule 8 (v) of the Seed Rules, 1968.

It is worth of mention here that the person whose name appears on the label shall be responsible for the accuracy of the information required to appear on the label so long as seed is contained in the unopened original container as per Rule 7 of the Seed Rules, 1968.

The packing size of the seed containers is pre-approved. Any additional packing size may require approval of the seed certification agency. In case of re-packing after receipt of seed

testing result as per paragraph XXIX of IMSCS, the seed quality at the time of re-packing shall be good otherwise dispute may arise as there is no provision for re-testing at this stage.

The lot size shall be as per the Appendix - V and construction of the seed lot number shall be as per the Appendix – VI of the IMSCS.

# 5. Seed sampling and analysis, including genetic purity test and /or seed health test, if any, in order to verify conformity to the prescribed standards.

According to the Rule 21(2) of the Seed Rules, 1968 the Seed Analyst shall analyse the samples according to the provisions of the Seeds Act, 1966 and the rules of the Seed Rules, 1968. The Seed (Amendment) Rules, 1973 No. 7 (17) / 69 – Seeds Dev. Dated 30<sup>th</sup> June, 1973 described that the Seed Analyst shall analyse the samples in accordance with the procedures laid down in the Seed Testing Manual published by the ICAR, New Delhi amended from time to time. In this Seed Testing Manual, the procedure for sampling has been described in detail.

When a seed lot does not meet the prescribed seed standards, the certification agency on request of the seed producer may permit re-cleaning, re-sampling and re-testing. The re-cleaning, re-sampling and re-testing shall be permitted only once as per the provision of the paragraph XXIV of the IMSCS.

The Certification Agency has the authority to refuse certification of any seed lot that does not conform to the Minimum Standards prescribed for that particular crop according to the provision of the paragraph XXX of the IMSCS.

The validity period of a seed lot shall be nine months from the date of test (date of issue of the seed testing result) at the time of initial certification. The validity period could be further extended for six months provided on re-testing seed conforms to the prescribed standards in respect of (i) physical purity, (ii) germination and (iii) insect damage for all seeds except vegetatively propagating material for which lot shall be re-examined for seed standards specified for respective crop. A seed lot will be eligible for extension of the validity period as long as it conforms to the prescribed standards as per the provisions of paragraph XXXI of IMSCS. The procedure for extension of validity period is given in Appendix – XII of IMSCS.

When a seed lot is not found meeting the prescribed standards for the class for which it has been registered but conforms to the prescribed standards to the immediate lower class, the Certification Agency may accept such seed lots for certification to the immediate lower class provided request has been made to this effect by seed producer. However, downgrading of the seed class shall not be applicable in case of hybrids and their parents as per the paragraph XXVII of IMSCS.

## 6. Grant of certificate and certification tags, tagging and sealing.

The certificate is granted under Section 9 (3) of the Seeds Act, 1966. The certificate shall be in the Form II according to the Rule 17 of the Seed Rules, 1968. The person to whom the certificate is granted shall attach a certification tag to every container of the certified seed and shall follow the provisions in respect of labeling as per the Rule 17 (i) of the Seed Rules, 1968. The colour of the certification tag shall be white for foundation seed and blue for certified seed as per Rule 17 (iii) of the Seed Rules, 1968. The container of the certified seed shall carry a seal of such material and in such form as the certification agency may determine and no container

carrying a certification tag shall be sold by the person if the tag or seal has either being tampered with or removed as per Rule 17 (iv) of the Seed Rules, 1968.

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# **Methods of Genetic Purity Testing**

# Nakul Gupta

# Scientist

# ICAR-Indian Institute of Vegetable Research, Varanasi 221305, U.P.

Genetic purity of seed is the most important parameter of seed quality as it determines the authenticity of seed and yield potential of variety/hybrid. In general, the negative impact on seed yield, consistency and quality is due to the presence of a large proportion of off-types. In the scientific production of seeds, special attention is paid to every step to maintain the authenticity of the variety."Genetic Purity of a seed lot is determined on the basis of distinct morphological characteristics of the variety expressed at seed, seedling and plant level by comparing its submitted sample with authentic sample under identical environmental condition."

## Source of contamination

Generally, all these kind of impurities occurs as result of delusion or inaccuracy during seed multiplication, harvesting, drying, handling or packaging of seed. For example-

- Seed fertilized by foreign pollen, especially in open pollinated crops
- Mutation
- Unclean harvesting equipment
- Carelessness at the processing
- Mistakes in bagging and tagging
- Mechanical mixture of seeds of other varieties during sowing, harvesting or storage In some cases, two additional sources of contamination have appeared as a result of the particular method of seed production used in hybrids.
- Incidental collection of male rather female inbred.
- Sibs (seed produced by self or sib-pollination among female lines.

All of these sources of contamination are preventable through prudent management. Sibs are the main concern in hybrids and they are difficult to avoid therefore it may cause a large proportion of contamination. Sibs originates from the failure of crossing management such as emasculation, male sterile or self-incompatibility.Examination to determine the genesis of the variety can be conveniently divided into the following three groups:

- Laboratory examinations
- > Tests in glasshouses or growth chambers.
- > Field trial and field inspection.

## In that article, we discussed Grow-Out-Test, which is part of the field testing and inspection.

## **Grow-Out-Test**

The grow out test is a widely used method for testing the genetic purity of seed samples. This test involves planting seeds in a controlled environment and observing the resulting plants for any off-types or impurities. The grow out test is a non-destructive test, meaning that the seeds can be used for planting after the test is complete. This test is relatively easy to perform and can be conducted in a short amount of time. However, the test may not be reliable in all situations, as environmental factors can affect the growth of the plants. The main purpose of the magnification test is to establish the authenticity (true-to-type) of the variety. The determination is based on the observation of

plant characteristics of a variety that are least environmentally influenced and are highly heritable. In general, differences between varieties are most pronounced under favorable growing conditions. In plot trials, the condition must be set up in such a way that the genetic differences to be examined are as clear as possible.

The amount of seed to be used on a plot is calculated on the basis of qualities of the seed. Before planting, seeds easily distinguished in the laboratory to be of another cultivar are separated, and their portion of the seed quantity is calculated. If possible, questionable seeds can also be separated, which can be seeded separately and examined in greater detail.

The different samples of the same cultivar are seeded in adjacent plots, with typical samples at appropriate intervals. This is especially important for cross-fertilized crops, where the examination, for the most part, is based on a comparison between the samples to be tasted and the standard sample.

The field plot must be carefully observed during the growing session, the appearance of each of the same cultivars is compared to the others, and particularly to the standard sample. The consistency of the stand is also considered. It is particularly worth noting when shooting and/or flowering starts, and how long each period lasts.

## Sampling:

**Submitted sample:**The submitted sample for grow out test is drown simultaneously with submitted sample for other test. The sample size will vary depending on the plant species.

Сгор	Size of sample (g)
Genera with seed size similar to pearl millets	100
Genera with seed size similar to Beta vulgaris	250
Sorghum, rice, wheat and other genera of similar seed size	500
Maize, cotton, groundnut, soybean and other of similar seed size	1000

**Table 1:** Recommended sample size for growth test submission:

*Note:* The amount of the submitted sample is doubled if it's necessary to determine genetic purity at both the seed and plant levels.

**Working Sample:**In order to observe the acceptable off-type plants specified as the minimum seed certification standard in the optimal population, i.e., at least 400 plants, the size of the working sample mostly depends on the test weight and germination percentage of the crop.

Table-2: Number of plants require for grow out test

Maximum permissible Off-types (%)	Number of plants required per sample for
	observation
0.10	4,000
0.20	2,000
0.30	1,350
0.50	800
1.00 and above	400

## **Planting Instructions:**

- 1. To make sure that an approximately similar number of plants of the same species or cultivar are established in all plots, the weight of the seed sown should be adjusted in the event that the germination of the sample being sown exhibits significant variation.
- 2. It must be carefully checked to make sure that it doesn't already contain seed from a previous sample before adding another sample to a seed drill.
- 3. For each sample, there should be a minimum of two replicate plots. a fallback strategy or an alternative region inside the same field.
- 4. Any realistic size for the plots is acceptable as long as there are sufficient plants present to make the calculation with the required level of precision.
- 5. If the seed is planted *in situ*, it should ideally be mechanically planted in rows.
- 6. Plants and rows should be separated from one another sufficiently to allow for the development of the features being studied.
- a. Cereals, legumes and oil plants: Every plot should be seeded with a convenient number of rows. The recommended row spacing for flax and cereals is 200 to 250 mm, whereas the ideal row spacing for the other species listed below is 400 to 500 mm. The following number of plants per meter of row need to be thought of as ideal:

Crop	Plants/Meter	Crop	Plants/Meter	Crop	Plants/Meter
Linum	100	ViciaFaba	10	Pisum	30
Cereals	60	Other Vicia	30	Lupinus	30
Brassica	30	Papaver	50	Glycine	30

Table-3: Number of plants per meter of row:

The specification for different crops given in the Indian minimum seed certification standards are given in the Table-3. The certification agency may, however, change these specification, if deemed necessary.

S. No	Сгор	Row length (meters)	Plant to plan distance (cm	Space betwee rows (cm)	Space between plots (cm)	No. of replications
1.	Wheat, barley, oats	6	2	25	50	2
2.	Pea, cowpea	6	10	45	90	2
3.	Chickpea, green grar black gram	6	10	30	60	2
4.	Maize	10	25	60	90	2
5.	Hybrid cotton	5	10	45	45	2
6.	Paddy:					
	very early to medium	6	15	20	45	2
	late and very late	6	25	30	60	2
7.	Pearl millet	6	10	60	90	2
8.	Sorghum	6	10	45	60	2

Table-4: Spacing specifications

a. *Herbage plants:*It is advised to use rows that are between 300 and 450mm apart and measure approximately 15m in length overall.

Where it is possible to discern between two or more cultivars through the examination of single plants, a special plant approach should be applied. Single plants are often grown by sowing each seed separately in a greenhouse or laboratory. The plants are moved onto field plots once they have reached an appropriate size. Under ideal circumstances, it may be feasible to sow the seed in place, in which case seedlings are separated into single plants. Plants should be spaced apart by at least 600mm in both directions.

b. *Root and Other Crops:* Root and other crops grown spaced in rows.Each plot should include at least two rows, with a total length that will provide 400 or more plants for analysis. In order to grow about the same number of plants in the test and control plots, the sowing rate should be modified because both transplanting and thinning are potential sources of error. Only when it is deemed to be absolutely required is it possible to thin out or transplant from another part of the plot.

# **Recording of observations:**

Throughout the entire growth season, observations should be conducted, and any differences from the control sample should be noted. Plants that are easily identifiable as being of a different cultivar, species, or as aberrants should be counted and noted.

# 1. Estimating the number of plants

When possible, the number of plants in the plot should be counted or estimated, ideally while the plants are being studied. This is required in order to give the field plots test's estimated percentage of aberrant individuals.

Evaluation in conjunction with check counting is used for unthinned crops like grains. The plot contains at least two repeating locations where the number of individuals per meter of row is counted. The total number of plants in the plots can be computed using these counts. The best time to do this activity is after the plants have fully emerged but before they begin to tiller.

It is highly impractical to count the number of plants on unthinned plots in perennial, strongly tillered species, such as herbage seeds. In these species, the quantity of aberrant plants may be expressed as a function of area, number of seeds dispersed, or another appropriate metric.

# 2. Taking observations

The minimum number of plants that needs to be examined are given in following table. The minimum number is dependent on maximum permissible off-types.

Maximum types (%)	permissible	Of Minimum genetic purity (%)	Number of plants required possible sample for observation
	0.10	99.9	4,000
	0.20	99.8	2,000
	0.30	99.7	1,350
	0.50	99.5	800
1.0	0 and above	99.0 and below	400

Table-5: Minimum number of plants to be observed in GOT

# **Expression of Results:**

- a. Seeds and Seedlings: The findings of the determination of the seedlings are provided as a percentage of the number of typical seedlings evaluated.
- b. When possible, the number of plants discovered to be of different cultivars, different species, or aberrant shall be calculated as a percentage of the total number of plants tested.

In the case of herbage plants and related species, when cultivated in rows with broad spacing, it is difficult to quantify the total number of plants inspected per plot. The outcome may be stated as the number of divergent plants produced by the weight of seed dispersed.

The mean and other statistics can be calculated when characters are measured. It is frequently difficult to describe properly all off-types in cultivars of cross-fertilizing species, such as rye, root crops, herbage plants, etc. In this situation, any estimations of percentage impurity should be accompanied by relevant comments about the veracity of test samples.

Tolerance may be applied by using the reject table given below.

Standard	Reject numbers fo	r sample size of
	800	400
99.5 (1 in 200)	8	*
99.0 (1 in 100)	16	8
95.0 (5 in 100)	48	24
90.0 (10 in 100)	88	44
85.0 (15 in 100)	128	64

Table-6 Reject number for prescribed standards and sample size:

\*Indicates that the sample size is too small for a valid test.

When nothing is worthy of special comments is found the results may be reported as "*The results of the field plot examination of this sample revealed nothing to indicate that varietal purity is unsatisfactory.*"

# **Reporting of results**

- The percentage of other species and cultivars of off-type plants must be indicated in the growout test findings.
- Results must be reported as such if the sample is discovered to be a cultivar different from the one specified by the sender.
- The report must specify that the sample contains a mixture of several cultivars if there are more than 15% plants from another cultivar.
- If no information deserving of special commentary is discovered, the report must mention that the sample's grow-out test findings showed nothing to suggest that the cultivar or species name provided by the sender is inaccurate.

# Advantage:

- It is cheapest way to examine reasonable number of plants.
- It is possible to examine a large number of plots and for each plot it is possible to check large number of plants.
- The plants are examined during the whole period of growth. Some characters are more prominent at one time of the year than another, and the sample may therefore, be examined several times during the session.

## Disadvantage:

- The result are not available until 4 to 12 months after the seed was receives for testing.
- The conventional grow-out test, which relies on morphological markers, is time- and spaceconsuming and frequently does not allow for the clear identification of genotypes. For a quick assessment of seed purity, molecular markers are crucial. Biochemical and molecular markers can be employed to support grow-out test results and mitigate their drawbacks.

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# Detection of seed borne pathogens

Dr. B. K. Sarma

# Department of Mycology and Plant Pathology, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi

Seed borne pathogens present a serious threat to seedling establishment. Close association with seeds facilitates the long-term survival, introduction into new areas and widespread dissemination of pathogens. Under greenhouse conditions, the risks of significant economic losses due to diseases are great because factors including high populations of susceptible plants, high relative humidity, high temperatures and overhead irrigation, promote explosive plant disease development. Under these conditions, the most effective disease management strategy is exclusion which is accomplished by using seed detection assays to screen and eliminate infested seed lots before planting. The following will explore the current state of seed detection technology and include recent advances. A summary of the features of each assay is presented.

#### Conventional seed detection assays

Testing seeds for plant pathogens can be a difficult task. Unlike infected vegetative plant tissues, infested seeds can be asymptomatic, making visual detection impossible. Additionally, pathogen populations on seeds may below, and infested seeds may be non uniformly distributed within a lot. Many detection assays exist for different seed borne pathogens, however, few satisfy the minimum requirements for adequate seed tests. Ideally, seed assays should be sensitive, specific, rapid, robust, inexpensive and simple to implement and interpret. Seed assays have been developed based on different technologies including visual examination; selective media; seedling grow-out tests and serological techniques. While these tests have been used for many years, some of them have shortcomings that make them less than ideal. Brief descriptions of these assays including their advantages and disadvantages are discussed below. *Visual examination* 

In some cases infected seeds display characteristic symptoms, including discoloration and shriveling. Examples of such seed borne diseases include purple seed stain (*Cercosporakikuchii*), and advanced stages of Phomops is seed decay (*Phomopsislongicolla*) of soybean (*Glycine max*), and Cylindrocladium black rot(*Cylindrocladiumparasiticum*) of peanut (*Arachis hypogeae*). In these cases seed lot infestation can be reduced by using automatic devices that sort seeds based on visual of physical characteristics. These systems usually display low detection sensitivity.

Additionally, seeds infested by fungi, bacteria and viruses may display no macroscopic symptoms, making visual or physical inspection of seeds useless as a detection assay. *Selectivemedia* 

A direct method of testing seeds is by allowing pathogens to grow from them onto appropriate artificial media. This can be done by directly plating surface-sterilized seed samples or seedwash liquid onto artificial media, followed by incubation under adequate conditions. Once a pathogen is isolated it can be identified by its cultural or biochemical characteristics e.g. the production of a bluish-green fluorescent pigment on King's B medium in the case of fluorescent Pseudomonas spp. or the production of dark, muriform conidia in the case of Alternariaspp. Unfortunately, seeds may be contaminated by saprophytic microorganisms (non pathogens) that grow as well as, or better than target organisms on nutrient-rich, artificial media. The excessive growth of saprophytic organisms including *Rhizopusspp., Penicillium* spp., and yeasts make it impossible to identify pathogens that may be present. The inability to identify the unique characteristics of the target pathogens in the presence of contaminating microorganisms lead to inaccurate assessments of seed lot infestation. To overcome this problem, selective artificial media are developed that use antibiotics, fungicides, selected carbon and nitrogen sources and other inhibitory compounds to retard the growth of non target micro flora while allowing the pathogen to grow. Many selective and semi selective media have been developed for seed borne fungi and bacteria. Unfortunately, development of such media is time consuming and requires specific knowledge of the nutritional requirements and chemical tolerances of the target organism, relative to the non target seed micro flora. Employing selective media also requires 2 to 4 d for pathogen growth and the test operator mustbe familiar with the range of cultural characteristics associated with the pathogen. Finally, while selective media can be applied for certain bacteria and fungi, it cannot be applied for non culturable obligate parasites, e.g., viruses, nematodes and certain fungi and bacteria.

#### Seedling grow-out assay

The seedling grow-out assay is a direct measure of the seed lot's ability to transmit a disease. To conduct this assay, seed lot samples are planted under greenhouse conditions conducive to disease development and after germination, seedlings are observed for the development of symptoms. Seedling grow-out is one of the most applicable and widely used seed detection assays but for successful implementation, infected seedlings must display obvious and characteristic symptoms. Unfortunately, this is not always the case as some diseases have no distinct symptoms, e.g., wilting, chlorosis, etc. Another drawback of the seedling grow-out

assay is that large seed samples (10,000 to 50,000 seeds in the case of bacterial fruit blotch (*Acidovoraxavenae* subsp. *citrulli*) of watermelon (*Citrullus lanatus*) must be tested to statistically ensure that one infested seed can be detected. In addition to losses associated with the destructive testing of expensive seeds, assaying this quantity of seeds requires large areas of greenhouse space and adequate labor for assay set up and evaluation. The seedling grow-out assay is also time consuming, requiring up to weeks for seedling germination and symptom development. Finally, seed test evaluators must be familiar with the symptoms associated with each disease. This can be difficult since each disease has a range of possible symptoms that are influenced by environ-mental conditions. Hence, for the seed-ling grow-out assay, greenhouse to ensure consistent results. In large greenhouses this can be a challenge and it can lead to erroneous test results. Also, because of the variations in seedling symptom expression it is often necessary to isolate the pathogen from suspected seedlings for confirmation. These extra steps further prolong the time required to complete the seedling grow-out assay. Residual contamination and cross-contamination between spatially separated seed lots are also issues of concern under greenhouse conditions.

#### Blotter method

The collected seed samples need to analyze for the presence of major seed borne fungal the pathogens by blotter method following the International rules for Seed Testing. Seeds are tested for each variety maintaining four replications. Twenty-five seeds are placed on three layers of moist blotting paper (Whatman No.1) in each glass petridish. The petridishes are incubated at 25±1°C under 12/12 hrs light and darkness cycle for 7 days. Each seed is observed under stereomicroscopes in order to record the presence of fungal colony and bacterial ooze 7 days after incubation based on growth habit. In doubtful cases temporary slides are prepared from the fungal colony observed under compound microscope. Appropriate keys are consulted for identification of the fungi and bacteria. The results are presented as percent incidence for individual pathogen. Germination of the seeds is also recorded. Each individual incubated seed is observed under stereomicroscope in order to record the incidence of seed borne fungi. Most of the associated pathogens were detected by observing their growth characters on the incubated seeds on blotter paper. For proper identification of fungi temporary slides are prepared from the fungal colony and observed under compound microscope and identified with the help of Keys. The fungi from the incubated seeds are also transferred to PDA when needed. The culture is incubated at 25±10°C for 3-7 days. Temporary semi permanent slides are

prepared from the fungal colony and observed under compound microscope. The fungi are identified with the help of different books, manuals and publications. The results are presented as percent incidence for individual pathogen. Acronyms written on the reverse.

#### Rolled paper towel method

Germinability of the seeds are determined in the laboratory at room temperature (30±2°C). 200 seeds are randomly taken from each variety and 40 seeds are placed between a pair of moist paper towels. There should be replications for each variety. The towels are rolled and the ends are closed by threads and covered by polyethylene paper to prevent drying. After 10 days of incubation period observations pertaining to (a) % germination, (b) Non germinated seed (hard seed and rotten seed), (c) Post-emergence death, (d) Shoot length (e) Root length (f) Vigor Index and (g) Incidence of different organism needs to be recorded. For determination of organisms some portion of the fungi growth on the infected seeds are taken with the needle and observed under compound microscope. For determination of seedlings vigour 10 seedlings (normal / abnormal) are randomly selected from each paper and their individual shoot and root length is measured. Length of shoot is measured from the base of the stem up to the growing point of the youngest leaf. Similarly, length of root is measured from the starting point of the root to the largest available lateral root apex. Vigour of the seedling is determined by the following formula:

Vigour Index = (mean of root length+ mean of shoot length) × percentage of seed germination.

## Agar plate method

In the agar plate method, two hundred seeds are tested for each maintaining replications. Surface disinfected seeds (0.1% mercuric chloride) are plated on the PDA medium and the plated seeds are usually incubated for 5-7 days at 22-25°C under 12h altering cycles of light and darkness. At the end of the incubation period, fungi growing out from the seeds on the agar medium are examined and identified. Identification is done based on colony characters and morphology of speculation structures under a compound microscope. In the agar plate method more than one type of fungal colonies are produced. In this case, identification is done on the most frequently occurring colony present in all the petridishes and then the second most frequent, the third most frequent and soon. Thereafter, the identification of the different colonies are done visually and then under a stereomicroscope and followed by an examination

of the fruiting structures under a compound microscope. Once the identification is done, the colonies are assigned names and their acronyms written on the reverse [16].

#### Serology-based assays

Serological seed assays rely on antibodies (polyclonal or monoclonal) generated against unique antigens on the surfaces of plant pathogens. Antibodies bind strongly and specifically to their antigens and can subsequently be detected by the enzymatic digestion of substrates or fluorescent tags. Serology-based seed tests have several formats including the widely applied enzyme-linked immunosorbent assay (ELISA) and immune fluorescence microscopy. Serological assays do not require pure isolations of the pathogen and, hence, are applicable to biotrophic and necrotrophic seed borne pathogens. Currently serology is the most widely used detection assay for seed borne viruses and it has proven to be sensitive and robust. Serology has also been widely used for the detection of bacterial and fungal plant pathogens, but the unavailability of species-specific antibodies is a limitation. Additionally, the detection thresholds of serology-based assays vary significantly based on the quality of the antibody and the testing format. Finally, with serology-based assays it is possible to detect nonviable pathogens which results in erroneous (false-positive) interpretation.

#### PCR-based assay

PCR-based assays exhibit very higher levels of sensitivity than any other conventional techniques. They require extraction of PCR-quality DNA from the target organisms in the background of saprophytic organisms and inhibitory seed-derived compounds when applied to seed tests.PCR consists *in vitro* enzymatic amplification of an initial quantity of target DNA from any living organisms including fungi, bacteria and viruses. However, due to its specificity, speed and sensitivity it has been used to diagnose many seed borne pathogens. However, high capital costs and technical expertise for establishing PCR capabilities is major obstacle in the PCR-based detection technique. The second major obstacle in successful implication of this method is false negatives (inhibition of PCR reaction by various compounds contained in seeds) and false positives (amplification of DNA from non-viable cells) which restricts the accurate detection of the pathogen. Along with this its incapability in distinguishing between viable and non-viable cells is also one of the major constraints of this method.

## Some Important Bacterial Seed Borne Pathogens

#### Crops

**Pathogens** 

Wheat	Pseudomonas syringaepv. syringae, Xanthomonas campestrispv. translucens		
Maize	Pantoeastewartiisubsp. stewartii, Clavibactermichiganensis		
	subsp. nebraskensis		
Rice	X. oryzaepv. oryzae, X. oryzaepv. oryzicola, Acidovoraxoryzae		
Bean	P. syringaepv. phaseolicola, Curtobacteriumflaccumfacienspv.		
	flaccumfaciens, Xanthomonas campestris pv. phaseoli and		
	X. fuscans var. fuscans		
Soybean	P. syringaepv. glycinea		
Chickpea	Rhodococcusfascians		
Cereals	Rathayibactersp.		
Alfalfa	C. michiganensissubsp. insidiosus		
Tomato	Pseudomonas syringaepv. tomato (tomato), P. syringaepv.		
Pepper	syringae, Xanthomonas spp., Clavibactermichiganensissubsp.michiganensis		
Carrot	Xanthomonas campestris pv. carotae		
Onion	Pantoeaananatis, Burkholderiacepacia		
Crucifers	Xanthomonas campestris pv. campestris, P. syringaepv.alisalensis(broccoli)		
Pseudo	<i>monas</i> spp. (crucifers)		
Cucurbits	P. syringaepv. lachrymans, Acidovoraxcitrulli		
Lettuce	Xanthomonas campestris pv. vitians		

# Some Important Fungal Seed Borne Pathogens

<u>Crops</u>	Diseases	Pathogens
Wheat	Loost smut Karnal smut Flag smut	Ustilago segetum var. tritci Neovossia indica Urocystisagropyri
Chickpea	Ascochyta blight Wilt	Ascochyta rabiei Fusarium oxysporumf.sp.ciceri
Crucifers	Grey and black leaf Spot	Alterniabrassicae A. brassicicola
Rice	Bunt False Smut Stackburn	Neovossia horrid Ustilaginoidea virens Pyriculariaoryzae Trichoconiellapadwickii
Cotton	Anthracnose Wilt	Colletotrichum indicum F.oxysporumf.sp.vasinfectum
Maize	Black kernel rot Cob rot Southern leaf blight	Botryodiplodiatheobromae Fusarium moniliformae Drechlera maydis
Pearl millet	Downy mildew Smut	Sclerosporagraminicola Tolyposporiumpenicillsriae

Sorghum	Anthracnose	Colletotrichum graminicola
	Kernel or grain smut	Sphacelothecasorghi
	Downy mildew	Peronosclerosporasorghi
Soybean	Anthracnose	Colletotrichum dematium
	Pod & stem blight	Phomopsis sojae
	Purple seed stain	Cercosporakikuchii
Cucumis spp.	Anthracnose	Colletotrichum lagenarium
Brinjal	Fruit rot	Phomopsis vexans
Onion	Damping off	Botrytis allii
	Downy mildew	Peronospora destructor
	Purple blotch	Alterniaporri
	Stemphylium Blight	Stemphylium vesicarium
Pepper chilies	Anthracnose	Colletrotrichumcapsici
	Or ripe fruit rot	
Tomato	Buck eye rot	Phytophthora parasitica
	Damping off	Phythiumaphanidermatum
	Early Blight	Alterniasolani
	Late blight or	Phytopthorainfestans
	Fruit rot	

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# Management of seed borne pathogens

Dr. B. K. Sarma

# Department of Mycology and Plant Pathology, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi

Diseases can have a significant effect on production of specialty seed crops. Seed growers must pay attention to diseases that affect the vegetative growth stage of the crop, as well as those that affect the reproductive growth stages (flowering and seed formation). Some diseases, such as Verticillium wilt of spinach, become symptomatic only when the crop enters the reproductive stage; these diseases are more important to seed growers than to vegetable growers (unless the vegetable crop also has a flowering stage, e.g., tomato or potato). While vegetable growers are concerned primarily with the pathogens that affect marketable yield and quality, seed growers must also learn how to diagnose and manage seed borne pathogens and the microorganisms that affect seed quality. Pathogens usually remain viable for longer in seed than in vegetative parts of the plant or in the soil. Seeds are a major means of survival of some plant pathogens and of introducing new pathogens to a field or region.

Seed borne disease refers to the particular plant diseases that are transmitted by seed. In some cases the transmission on seed is insignificant compared to the population of disease organisms that exist in soil or on weed species. In other cases, the transmission on seed is the primary means by which a disease spreads. While we are cautious about any type of disease on seed, it is this latter set of diseases that we must be most vigilant in controlling. Planting infected seed may result in a widespread distribution of disease within the crop, and an increased number of initial infection sites from which the disease can spread. As an example, consider the development of ascochyta blight in a chickpea crop. Since there is a high rate of seed-to-seedling transmission of this disease, even a small percentage of infected seed can result in significant seedling infection in the field. For a seed lot with 0.1 per cent ascochyta infection (one infected seed in 1,000 seeds) and a planting density of three to four plants/ft.2, 175 infected seedlings per acre could potentially result. This is a substantial amount of early infection for such an aggressive disease.

The diseases that are caused by fungal pathogens which persist (survive) in the soil matrix and in residues on the soil surface are defined as soil borne diseases. Thus the soil is a reservoir of inoculums of these pathogens, the majority of which are widely distributed in agricultural soils. Diseases of plants are caused primarily by three types of pathogens: bacteria, fungi, and viruses. Despite that fungi comprise the largest group of pathogens, the bulk of seed-specific diseases are caused by bacteria or viruses. This is due to the fact that bacteria and viruses are more adept at entering and then travelling through the veins of the plant, a phenomenon known as 'systemic infection,' and from the vascular system may make their way into the developing embryos of seeds. The risk of seed-borne disease infection varies widely by crop, disease, and location. Many diseases will only become a problem if grown in a region or environment conducive to the disease. Commonly diseases present on seed may also be soilborne or air-borne and the ultimate fate of the crop may be as dependent on the variety

resistance and crop management practices as on the presence of seed-borne inoculum. There are additionally many microorganisms present on seeds that have no known negative effects and some feel may hold potential positive effects, although there is no current research documentation. It is still important to start with high-quality, clean seed. In select instances the spread of specific pathogens from seed may introduce the disease to the system with devastating effects. Such is the case of Watermelon Fruit Blotch, bacteria that can be seed- or soil-borne and difficult to manage once in the system; particularly in the warm and humid regions.

It is recommended to have seed tested at an accredited laboratory to assess the levels of seed-borne diseases. Commercial laboratories can test pulse seed to determine the level of seed-borne fungi that cause ascochyta blight, anthracnose, botrytis seedling blight and grey mould, and sclerotinia. A parallel test for germination will indicate whether seed quality has been affected by such factors as immaturity, disease, mechanical injury and chemical damage. Testing for vigour may also be beneficial, as it serves as an indication of how seed will respond in less than ideal growing conditions. The best way to ensure success of a disease-management program is to adapt it to the diseases expected and to use integrated disease-control measures. Among these measures are resistant or tolerant varieties, crop rotations, fungicides, nematicides, and suggested agronomic practices. The success of any one or all of these measures may depend on how carefully you scout your crops. Because periodic scouting increases the likelihood that disease controls will be applied properly, it can help prevent loss through disease and unnecessary use of pesticides.

Disease pathogens restricted to the seed coat are treatable by external application of antimicrobial agents such as bleach, acid, trisodium phosphate, or other commercial products. Rarely do these treatments effect 100% sterilization, but they can greatly reduce levels of pathogens. These types of treatments are typically used for the class of non-seed-specific diseases in which seed borne transmission is minor compared to the levels of inoculum already present in soil due to crop debris. An example of such a disease is cucurbit scab (*Cladosporium cucumerinum*), a fungal disease which tends to flare up in wet years on fields that have grown cucurbits repeatedly.

Seed should be tested for germination to determine its suitability for planting. Germination can decrease in the bin over the winter, especially if the seed was immature or damaged at harvest. It is a good investment to re-test seed for germination in the spring, if quality was questionable in the fall, increasing the seeding rate will compensate for low germination, but only to a certain extent. If the reduced germination was a result of disease, an increased seeding rate can introduce more disease into the field. In general, seed treatments may have either systemic or contact modes of action. Controlling fungi that are carried within the seed requires a systemic product (i.e. smut in barley), whereas contact or protectant products are adequate for surface-borne or soil-borne fungi. Systemic seed treatments are diluted quite quickly within the plant once the seed germinates and is actively growing. Some treatments will protect a young seedling against early leaf disease or root rot infection, but in most cases, seed treatments are no longer effective after seedling emergence. Seed treatments are used on many crops to control a variety of pests. These are commonly used to ensure

uniform stand establishment by protecting against soil borne pathogens and insects. Seed treatments can often be used to control pathogens that occur on or in the seed. These are not the only available method to control a particular pest; should be compared to alternative pest control measures for cost, efficacy, safety, and so on. Seed treatments can often be supplemented with other control measures to achieve satisfactory results.

# Types of seed borne pathogens

Seed borne disease refers to the particular plant diseases that are transmitted by seed. In some cases the transmission on seed is insignificant compared to the population of disease organisms that exist in soil or on weed species. In other cases, the transmission on seed is the primary means by which a disease spreads. While we are cautious about any type of disease on seed, it is this latter set of diseases that we must be most vigilant in controlling. For the purposes of this article, we will call them seed-specific diseases.

Diseases of plants are caused primarily by three types of pathogens: *bacteria, fungi,* and *viruses.* Despite that fungi comprise the largest group of pathogens, the bulk of seed-specific diseases are caused by bacteria or viruses. This is due to the fact that bacteria and viruses are more adept at entering and then travelling through the veins of the plant, a phenomenon known as 'systemic infection,' and from the vascular system may make their way into the developing embryos of seeds.

Fungi, in contrast, tend to be restricted to the outer layers of the plant, where they initiate infection by means of air-borne spores and then proceed to spread by attacking nearby cells of the outer layers. Fungi are much less likely to enter the vascular system of the plant, and thus infect seed mostly when they either 'crawl' all the way to seed on the outside of the plant, or else send out spores that land on the seed. In either case, the fungal spores are on the outside of the seed, in the layers of the seed coat. Spores on the seed coat are more prone to either dry up and die, or else to get sloughed off with the seed coat during seed germination, thereby failing to cause disease on the next generation of plants.

## Strategies for management of seed borne pathogens

Disease management tactics are either preventive (actions taken to avoid or reduce the likelihood of disease problems) or curative (treatments that eliminate or reduce the effects of a particular disease after it has become established). Because there are few effective curative practices available to organic farmers, organic farmers focus their disease management efforts primarily on preventive cultural practices. Such practices include planting pathogen-free seed, planting in fields of low inoculum potential and in locations with good air movement, adopting wide row spacing, orienting the crop rows to maximize air movement between rows, and tying or staking seed crops to improve air circulation and reduce humidity in the canopy. If feasible, consider using drip or furrow irrigation instead of overhead irrigation, or irrigate earlier in the day to allow the canopy to dry before nightfall.

Some significant pathogens of seed crops are soil borne, such as Fusarium wilt of spinach. To manage soil borne pathogens, it is important to know the cropping history of the field and to adopt appropriate crop rotations. A rotation of 6 to 15 years, depending on the

susceptibility of the spinach cultivar, is required to control Fusarium wilt in spinach seed crops. Some soil borne pathogens affect more than one crop, e.g., the fungus that causes Verticillium wilt of spinach can also infect potato, so it is important to avoid growing other crops in the rotation that may be alternative hosts to soil borne pathogens that affect the seed crop.

Strict management of, and screening for, seed borne pathogens of vegetable crops is critical to maintaining high seed quality. Even low levels of seed contamination can cause epidemics of some diseases when infected seed is planted in the field. For example, the tolerance level for contamination of crucifer seed with the causal agent of black rot, *Xanthomonas campestris* pv. *campestris*, is 0 contaminated seeds in 10,000 to 50,000 seeds (depending on the market or country in which the seed will be distributed).

Seeds contaminated with a pathogen can be treated physically (e.g. hot water) or chemically (e.g. bleach) to destroy inoculum or reduce the incidence of infection. Some physical and chemical treatments may reduce seed quality (germination, vigor, and/or longevity), so it may be important to test a particular seed treatment on a small sample of seed and check for possible phytotoxicity to the seed before treating an entire seed lot. Hot water treatment can only be used on some crops, such as brassicas, carrots, tomatoes, peppers, and lettuce, but even on those crops very precise parameters must be followed for hot water treatment to avoid damaging the seed. There are a number of biological and natural disease management products coming onto the market that are approved for use on organic farms, but it must be noted that the efficacy of these bio control products may vary among sites, crops, and diseases, reflecting the complexities and particulars of interactions amongst the host, pathogen and environment. Therefore, planting pathogen-free seed, when possible, is always preferable to trying to eradicate a pathogen from seed.

Disease pathogens restricted to the seed coat are treatable by external application of antimicrobial agents such as bleach, acid, trisodium phosphate, or other commercial products. Rarely do these treatments effect 100% sterilization, but they can greatly reduce levels of pathogens. These types of treatments are typically used for the class of non-seed-specific diseases in which seed borne transmission is minor compared to the levels of inoculum already present in soil due to crop debris. An example of such a disease is cucurbit scab (Cladosporium cucumerinum), a fungal disease which tends to flare up in wet years on fields that have grown cucurbits repeatedly.

Seed-specific disease pathogens that reside inside the seed, which are typically bacteria or viruses, cannot be eliminated by surface sterilization. Because they're often inside the embryo itself, these pathogens are almost certain to divide and spread to cause infection when that seed germinates and grows. They cannot be eradicated by external application of chemicals; however, they are susceptible to the one agent that can penetrate the interior of the seed, which is heat. The number one method for sterilizing seed is to treat it with either wet or dry heat, which penetrates to the core of seed. Heat kills the majority of bacterial and fungal pathogens, and bacterial pathogens are particularly sensitive to heat. Wet heat, in the form of hot water, is more effective than dry heat, and thus the most common method for treatment of seed disease is hot water of 122°F (50°C) for 20-25 minutes. We have found in our own lab, though, that

temperatures of 118°F (47°C) are equally effective for most pathogens and less damaging to the seed.

Hot water is commonly used for treatment of most small seeds, but is less effective and more difficult to use for large seeds. Large seeds tend to be damaged by wetting and re-drying, are more difficult to penetrate fully with heat, and are so bulky as to make it difficult to efficiently wet and dry them. Unfortunately, viral pathogens are generally not susceptible to heat, although dry heat has been shown to have some efficacy against certain tomato viruses. Solutions of bleach or trisodium phoshphate are sometimes used to remove surface infections of virus in pepper and tomato seed. In general, though, viral pathogens are quite difficult or impossible to remove from seed, and thus virus-diseased plants in a seed field are almost always pulled up and destroyed immediately.

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# Duties and responsibilities of seed certification inspector

Dr. Rajesh Kumar

**Principal Scientist** 

ICAR-Indian Institute of Vegetable Research Varanasi, Uttar Pradesh

India is a agriculture based country. Agriculture play a major role in countries GDP. After independence our agriculture production and productivity grows many times which make the country self-sufficient. At present main task before all the organization engaged with the farmers and agriculture is to maintain and increase the production and productivity which is only possible by maintaining soil health and supplying good quality inputs to the farmer on time.

Among all those inputs required in agriculture "quality seed" of improved varieties is most important. Many organizations are responsible to make available such seed to the farmer before sowing. On one side Agriculture research institute are developing many improved varieties of most of the crops and gat them notified under the seed act so that good quality seed of these varieties will be available the seed producing organizations. On other side these seed producing organizations multiplied the good quality seeds of deferent varieties and make available to the farmer on time. For control of quality of such crops and varieties seed responsibility is given to state seed certification agencies established under seed act 1966 at state level.

These state seed certification agency works in accordance with the procedures laydown in Indian Minimum Seed Certification Standards 2013. Field level officers of these agencies go to field of seed plot for verification of field standards and after field inspection they advise the seed grower for quality improvement. If seed crop found to be substandard they propose seed crop field for rejection.

For those seed plots which are up to the standard at field level the field officer of state seed certification agency advise to seed producer farmer about precaution to be taken during harvesting, threshing and transportation to the assigned processing plant so the that purity of seed lot will be maintained. After seed lot received at processing plant, in the same manner, officer of state seed certification agency who is assigned to that processing plant will visit the plant and assure that quality of seed will be maintain as per the guideline given in the Indian minimum seed certification standard 2013 and instruction given by the superior office will also be fallowed so that seed will not loses its identity and quality, till its final tagging and sealing is done.

Before we know Duties and responsibilities of seed certification inspector we shell know what seed is so that we can differentiate seed form grain.

Botanical definition of seed is that "It is fertilized and fully develop embryo". Definition of seed in agriculture science is "It may be a part of a plant which is used to develop a new plant it may be root stem or a stick or fertilized and fully develop embryo". We shall only remember the second definition of seed for seed certification. Six main characters differentiate the seed from grain. If these six characters are present then only we can say that it is seed otherwise seed will lost its identity. These six characters are as under.

- 1. Physical appearance & Uniform Size.
- 2. Germinability.
- 3. Seed Vigor.
- 4. Genetic Purity.
- 5. Genetic Identity.
- 6. Free From Seed Born Diseases.

If above six character are present than only it comes under the category of seed otherwise it is grain. All the time, during the process of seed certification, you will remember that these six characters will not be ignored.

# Duties and responsibilities of seed certification inspector:-

It is must for seed certification inspector to perform their duties in accordance to the procedure laydown in Indian Minimum Seed Certification Standard 2013 and working manual of their certification agency. According to the procedure laydown in Indian Minimum Seed Certification Standard 2013 seed certification must be performed in six steps, which are as under :-

- 1. Verification of parental seed material.
- 2. Receipt of farmer wise seed production application, Scrutiny of application forms and registration of suitable forms.
- 3. Field inspections to verify conformity to the prescribed field standards.
- 4. Supervision at post-harvest stages including processing and packing.
- 5. Seed sampling in accordance to the procedure laydown in seed testing manual and sanding the samples to seed testing laboratory for analysis to verify conformity to the prescribed seed standards for genetic purity test and other prescribed testing for seed standard.
- 6. After receipt of seed testing reports from the laboratory if results meat the prescribed seed standard grant of certificate and certification tags tagging and sealing will be carried out as per the guideline given in Indian Minimum Seed Certification Standard 2013 and working manual of their certification agency.

Official of the certification agency shell always keep in mind that goal of the certification agency is to make the good quality seed available to the farmer on time.

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# Post Harvest Handling of Seeds

Er. M. K. Vishwakarma

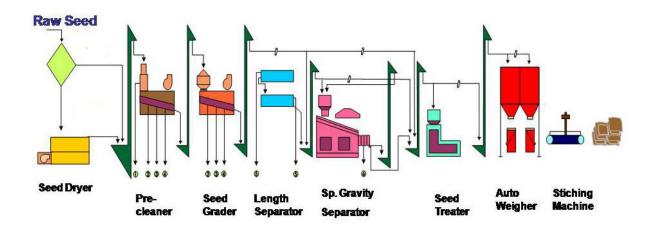
Seed Processing Engineer, National Seed Research & Training Centre, Varanasi

Seed is one of the most important inputs for sustainable agriculture. Quality of seed affects both yield and credibility in the market. Unlike in grain, extreme care and vigilance is required in seed to avoid mechanical mixing of crop varieties during post harvest stages such as threshing, winnowing, drying, Pre-cleaning, grading, packaging, storage and marketing. Many a time carelessness as well as ignorance at any stage cause colossal loss in seed quality and market value. Hence in-depth knowledge of post harvest care and improvement in physical purity of seed is most important.

The objective of seed processing is to achieve clean, pure seeds of high physiological quality (germinability) which can be stored and easily handled during succeeding processes, such as pretreatment, transport and sowing. Processing includes a number of handling procedures, where applicability differs e.g. according to seed type, condition of the seeds after harvest and potential storage period.Seed cleaning typically consists of a series of processes during which impurities are gradually removed and the seed lot concurrently achieves a progressively higher purity (Fig-1). The type, order, and adjustment of the processes depend on seed type and type of impurities.During seed processing, contaminants are removed to a level that meets the industry wide minimum seed certification standards, failing which, they may be discarded or blended with a relatively better lot of the same variety. Contaminants are removed by procedures utilizing machines which exploit the differences in physical characteristics of the desirable seed and other components in the mixture. These physical properties include but are not limited to length, width, thickness, shape, density, terminal velocity, drag coefficient, reflectivity, surface texture, electrical conductivity and resilience. Seed separators are designed to utilize the difference in a single physical property or a combination of physical properties of the seed.

## **Concept of Separation Processes:**

Separation and purification of materials forms an important process in post harvest handling of agricultural products. Naturally occurring processes are inherently mixing processes and have led to the reverse procedure of separation processes which are becoming the most challenging categories of engineering problems. Mechanical separations are applicable to heterogeneous mixtures. Broadly, a separation processes a mixture of substances in two or more products which differ from one another in composition. The separation is caused by the addition of a separating agent which may be in the form of energy. Need for separation accounts for the most of the production cost of a pure substance. Often separation itself can be the key function of the entire process e.g. grain cleaning. To a large extent man's ability to ease food shortage depends upon his technical knowledge and capacity to extract and separate essential food materials from the new or inexpensive sources. From the above considerations, it is apparent that much careful thought and effort must go into understanding and improvement of various separation processes.



# (1) Large Impurities (2) Coarse Impurities (3) Small Impurities (4) Light Impurities (5) Short Impurities (5) Low Density Seed

Fig-1: Flow diagram of modern seed processing

## Methods of Seed Separation:

Improvement in seed separation technology from simple hand picking and domestic hand screen to present day methods runs parallel to the story of civilization. A modern seed processing involves moving the field produce through a series of machines which perform specific operations and pass on the product to the next machine after discharging the reject. A well designed seed processing plant is laid out to permit by passing any machine without interrupting the product flow. Many types of seed cleaning machines are used to remove contaminants from the harvested-threshed seed.

## Air-Screen Cleaner:

The air-screen cleaner is the most widely used machine. It is an essential unit operation in seed processing plant. The simplest mechanical method of separating particulate solids, the class to which most agricultural seeds and food grains belong, is by passing than over screens which are stationary or reciprocating and are set at a slight downward slope, so that small particles will pass through and larger materials will tail over them. In combination with air-fans or blowers, the screen machine provides adequate conditioning for some seed crops. Such machines work by taking advantage of dimensional and aerodynamic differences. Agricultural screens are constructed of perforated metal or woven wire mesh. Hole shapes in perforated screens are usually round, triangular, oblong or rectangular, Openings in wire screens are square or rectangular, their size being represented by mesh numbers. Round hole screens are identified by a number denoting diameter of the perforation. In India, these numbers indicate the diameter in millimeters. Rectangular or oblong holes in perforated screens are identified by two numbers describing the width and length of the slot. Selection of the screen depends on the seeds being handled. Screen opening sizes used for different crops have been prepared and are available in literature. Screens with various sizes and shapes of holes drop some particles and retain others depending mainly on the width and thickness of particles and, to a lesser extent on their length.

Pneumatic separators or air columns exploiting aerodynamic differences are used to remove dust, chaff or other light contaminants. The air system in air-screen machine operates in this manner. As a finishing machine it can remove light, immature, shriveled or damaged seeds from already cleaned good seed lots. Air screen combinations are extensively used in grain combining and threshing.

The air screen machine in general employs three cleaning elements: aspiration, scalping and grading. The light seeds and chaffy materials are removed from the seed through aspiration. In scalping operation, the good seeds are dropped through top screen opening and the larger materials (trash, clods etc.) are carried over the screen into the rejection spout. In grading operation, the good seed ride over screen openings, while smaller particles (under size, cut shriveled, broken seeds) drop through.

Feed hoppers of air screen cleaner cum grader are of three types: Roll feed hopper consists of a container to receive the seed, hopper flights and auger to spread the seed across the width of the hopper and a revolving fluted roll in the bottom of the hopper that feeds and even steady flow of seed to the top screen and distributes the seed across the full screen width. In roll feed brush hopper a rotating shaft pulls trash of seeds down to the revolving fluted roll and a tough fibre brush to prevent clogging. In the metering hopper a shaft width specially bent rod is used to spread the seed. Other special purpose variants are designed to handle special seeds.

## **Principles of operation:**

In a typical two screen seed cleaner cum grader, as the seed is delivered by the feed hopper the air blast removes light weight seed and chaff, scalping screen remove material larger than the crop seed; grading screen dropout material smaller than the crop seed. In a four screen machine, the 4 screen do the following operations: (a) 1<sup>st</sup> screen- scalping, (b) 2<sup>nd</sup> screen- grading, (c) 3<sup>rd</sup> screen- close scalping, (d) 4<sup>th</sup> fine grading. At the seed drop off the gravity screens they fall through the lower air separation to remove residual light seed and trash.

# Length Separator:

Length separators are designed to lift and remove the short fraction from a varied length mixture by exploiting the difference in the largest dimension of the product and the reject. These are two types of length separators, the indented disc separators and indented cylinder separators. Both lift out short particles out of a seed mixture with a given pocket or indentation and a relatively cleaned product is pushed further. The indented disc separator consists of a series of indented discs, mounted together on a rotating horizontal shaft. Each disc is designed with an open centre and numerous undercut recesses on each face. The broken seeds and the material shorter than the crop seed are lifted by the indents and are delivered into a trough at the side of the machine. Discs of increasing pocket sizes are normally provided on the shaft so that the particles of increasing lengths are removed selectively. The long seed that does not match the pockets is pushed by the incoming seed through the open centre of the disc and is discharged at the outlet.

The indented cylinder separator consists of a rotating cylinder and an adjustable trough. The inner surface of the cylinder has closely spaced indents. The seed mass to be handled is fed at one end and lies at the bottom of the cylinder. As the cylinder rotates on its axis the short seeds are lifted from the mixture by indents. Thus at some point before reaching the top of the rotation, the seeds fall out from the indents, because of the tilting of the later. Actually, the seeds resting in the indents lose balance and are eventually received in the adjustable trough from where they are conveyed out by an auger. The long seed which is not lifted by the indents gradually move through the cylinder end are discharged to a separate spout at the other end of the cylinder. The quality of separation depends on the position of the trough and the speed of the cylinder.

# **Specific Gravity Separator:**

A specific gravity separator consists of two key components - air chest and the deck. Air chest houses fans and motor. The deck is mounted above the chest. The deck is a rectangular or triangular table covered with a porous cloth or wire mesh and inclined in two directions. The gravity separator classifies components of a mixture mainly according to density. Separation is caused in two steps. Seed mixture introduced at the back of the porous deck is stratified by the low pressure air coming through the deck. Low density particles tend to float and form a layer at the top and the high densityparticles sink to the bottom layer. Fractions of intermediate density, assume intermediate position. For proper identification of different density fractions, the seed lot must be well screened before hand so that all particles are of the same size. The seed should be dust free. An aspiration canopy is installed above the feed corner to further suck up any residual dust. The oscillating motion of the deck moves the high density particles laterally towards the uphill side at the deck. Simultaneously the floating low density material moves downhill by gravity. As the seed mixture layers travel from the feeding corner to the discharge end of the deck, a continuous gradation of particles takes place ranging from the low density ones at the lower side of the deck to the high density ones at the upper side. Adjustable splitters divide the output into number of density fractions needed. For deck covering a closely woven material for small seeds and a coarse weave for large seeds is used. Typical covering materials are small hole perforated metal and wire mesh. The coverings are supported by a deck frame, which serves as the top of the air chamber and helps to equalize the flow of air through the seed mass. Feed rate, air flow rate, deck angles and frequency of stroke are major adjustments. These adjustments are interrelated.

## Seed Refining:

To further refine the seed, machines have been developed to take advantage from additional differences in physical properties. The electrostatic separator exploits the difference in the electrical characteristics of the seeds and contaminants. The quality of separation depends on the relative availability of the components in the seed mixture to conduct electricity or to hold electrical charge on surface. A spiral separator senses the ability of components to roll. This is very simple machine and operates completely by gravity. It has no moving parts and needs no prime mover. The endless draper belt separator utilizes surface texture differences to separate rough seeds from the smooth ones. A magnetic separator requires certain pre treatment of the feed mixture. Iron power or a magnetic fluid is added. Variation in seed coat characteristics is utilized. The iron is selectively adsorbed by rough, broken, cracked porous or sticky components making them more reactive than the smooth components. A colour separator acts on differences in reflective properties. The components of the mixture must be cingulated for individual sensing by the photoelectric cells. To scale up the throughput multi-channel machines are required.

#### New Emerging Technologies:

Modernization of agriculture causes demand for higher quality seeds and invites application of new technologies to seed conditioning. This needs removal of all contaminants even when the physical property difference is very slight. This emphasis has led to the investigation of measurement system for physical properties and development of systems for improved seed conditioning. With the advent of microprocessors and the rapidly expanding application of technology, seed conditioning is beginning to benefit as the use of computers is integrated into the new equipments. Machine vision system (MVS) is being used for seed conditioning. The feasibility of the application was shown for identifying seeds of different colour, size and shape. The MVS can also be used to detect stress cracks in certain seeds. There appears a need to develop expert systems for modern seed processing and once a system is made available, the performance and the status of an average worker can be raised to the level of an expert.

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# PROBLEMS AND PROSPECTS OF SEED CERTIFICATION

R. Harichandan, M.Sc(Ag.) Seed Certification Officer, OSSOPCA, Bhubaneswar E Mail: <u>singhamrh.seed@gmail.com</u> Phone No. 7205096115

**Seed means** any of the following classes of seeds used for sowing or planting: 1. seeds of food crops including edible oil seeds and seeds of fruits and vegetables; 2. Cotton seeds; 3. Seeds of cattle fodder; 4. Jute seed and includes seedlings, and tubers, bulbs, rhizomes, roots, cuttings, all types of grafts and other vegetatively propagated material, of food crops and cattle fodder.

With the enactment of the Seeds Act, 1966 and subsequent promulgation of the Seed Rules, 1968, the seed quality regulation through compulsory labeling and voluntary certification have been achieved to increase the production, supply and marketing of quality seeds in India.

Further, the declaration of seed as an essential commodity and the compulsory seed dealer licensing and law enforcement system boosted the seed quality control system in addition to the seed quality assurance system achieved through compulsory labeling and voluntary certification.

The purpose of seed certification is to maintain and make available to the farmers, through the process of certification, high quality seeds and propagating materials of notified kind and varieties to ensure genetic identity and genetic purity. Seed certification is also designed to achieve prescribed standards.

The problems and prospects are perennial with any system but our effort shall aim at reducing the problems and enhancing the prospects. The seed certification system is not immune from these effects. The problems and prospects of seed certification to be discussed here are mainly of field operation related and a sound knowledge can be helpful in fulfilling the purpose of seed certification.

# SEED PRODUCER RELATED PROBLEMS AND PROSPECTS

**Production Programme:** In seed certification system, seed producer is a major component. The seed producer takes up seed production. All the activities of seed production are under taken by the seed producer starting from obtaining source seed to marketing of certified seed. The risk of business is burdened by the seed producer. When the certified seed business progresses then the seed producer increases seed production through increase in area and increase in quantity and vice versa. Thus the prospect of seed certification lies with the progress of seed producer.

The seed certification system is highly dependent on the seed producers to take up seed production programs for both financial achievement and technical involvement. The selection of new crops / varieties has been the prerogative of the seed producers and unless the seed producer is proactive, prospective certification system becomes a mute spectator.

**Grower Selection:** Seed Grower selection is a critical component of seed production. Unless the seed producer selects progressive seed growers, the seed production programme suffers.

**Notified Variety:** The seed of those varieties which are notified under Section 5 of the Seeds Act, 1966 shall be eligible for certification. This compulsory nature of certified seed production poses problem of adoption of unsuitable new varieties when the ruling varieties are old and discouraged.

**Seed Economy:** If any seed producer / seed grower desires to sow a larger area by economizing a seed rate, permission shall be obtained from the certification agency. The provision of seed rate economy is a widely accepted concept to use less seed to cover more areas under seed production and accordingly the unit packing size shall be uniformly designed for unit area of one acre. The research output in this regard shall be approved by competent authority and shall be easily available to the seed grower / seed producer to avoid any confusion.

**Production of Foundation Stage II and Certified Stage II:** For production of Foundation stage II permission shall be taken from the certification agency and this is practiced for production of Certified stage II also.

**Seed Processing:** The seed processing units where majority of post-harvest seed certification works are carried out shall be adequate and efficient in space and machinery to facilitate early upgrading and subsequent maintenance of seed standards. The crop / variety specific requirement for processing has gone far ahead and the mandatory requirement of seed grader has been coupled with voluntary addition of pre-cleaner and gravity separator. Further addition of modern equipment for seed processing is the need of the hour to match the advanced seed processing units of the private sector.

**Seed Storage Facilities:** The seed storage facilities are neither controlled nor ambient in majority of cases and due to existence of varied climatic conditions during post-harvest operations, the natural process of seed quality deterioration is likely to be accelerated. Some times during re-packing (4kg. / 5kg.) of passed seed lots it is observed that there is visible presence of insects and damage due to insects is more than the permissible limit for the crop. Use of HDPE bags or bags of any other material other than cotton and jute might be adding to quick seed quality deterioration. Most of the seeds are hygroscopic in nature. The moisture content of raw seeds / processed seeds and the storage facilities may inhibit or invite unwanted situations to the seeds. Unless proper sanitary measures are taken, the certified seeds are subject to rapid loss of seed quality.

**Labeling of seed containers:** The concept of producer label is well known. Sometimes ambiguity arises due to writing on the seed containers. Some people are confused that if producer label is attached to the seed container then writing on bags is required or not. Further, where the contents of the producer label are entirely written on the bags then attachment of producer label is required or not. Rule 9(3) of the Seed Rules, 1968 describes about this concept but writing on the bags and attachment of the label go simultaneously in many seed containers.

**Tagging and Sealing:** The seed producer to whom the certificate is granted shall attach a certification tag to every container of the certified seed. The container of the certified seed shall carry a seal of such material and in such form as the certification agency may determine and no container carrying a certification tag shall be sold by the person if the tag or seal has either being tampered with or removed.

#### CERTIFICATION AGENCY RELATED PROBLEMS AND PROSPECTS

**Uniformity:** The seed certification practices or processes or procedures shall be uniform in all the seed certification units in a state and any relaxation in one part of the state may create uproar in other parts. Due to the introduction of on line system of seed certification, seed production, seed certification, supply and distribution of certified seeds have been user friendly. The SATHI approach may bring wider acceptability and uniformity to the seed certification system. The uniformity in seed certification system among the states and even within a state is essential for growth and stability of the system. The seed certification agencies of different states shall meet and share the common and differences.

**Field Inspection:** Field inspection shall be conducted without prior notice to the seed producer as per the provisions of paragraph XI of IMSCS. However, it is a common practice to inform the seed producer / seed grower before field inspection. Whether all the fields of the seed crop are inspected or it is done at random is a major concern. The minimum number of prescribed field inspections shall be conducted and re-inspection may be done as per the situation.

**Seed Sampling:** Seed sampling is a major step for seed quality regulation. Random sampling following proper sampling intensity shall be followed. Any deviation may invite un-warranted situation.

**Seed Processing Schedule:** The Certification Agency shall prepare and communicate seed processing and packing schedule to all certified seed producers soon after the certification of seed crops at field stage and the seed producer shall adhere to the schedule specified by the Certification Agency. However, re-scheduling may be accepted by the Certification Agency on the request of seed producer on genuine grounds as per Paragraph XV of IMSCS. This practice is exactly reverse in most cases. Completion of seed processing as per the certification calendar is a hard task for seed producers and a lot of extensions in cut of dates are done to complete the process.

**Threshing Certificate:** The threshing certificate has not been mentioned as provision anywhere in the seed legislations. But it is a good step towards post harvest seed quality supervision. Where threshing certificate is issued after verification of quantity and quality of raw seeds, then it ensures seed quality to great extent. Marking or sealing of raw seed bags is a cumbersome process but in certain cases this shall be practiced.

**Genetic Purity:** Genetic purity is a more discussed and less understood concept in seed certification. Some people are confused with ODV and genetic purity. The invisible loss of genetic purity may have visible loss in production and productivity. A number of closely related varieties have been developed, released and notified which are morphologically so similar that the breeders are confused to distinguish.

Since, the genetic purity of any kind / variety is an extreme requirement for genuineness of the cultivars, the genetic purity of the breeder seeds shall be such to achieve 99% genetic purity for foundation class and 98% or so genetic purity for the certified class of seeds. The complementary laboratory tests for testing genetic purity shall be adopted as an easy, quick and cheap alternative to the conventional grow-out test. The infra structure and trained man power shall be available in every state.

**Validity Period:** The nine month validity period calculation from the date of test in the certification system looks favorable for the delayed tested seed lots though natural deterioration of seed starts from attaining physiological maturity. The provisional requirement of emergence testing of seed at the dealer's sale point is a check for loss of desired plant population. Further, the re-validation of seed lots shall be easy for the seed dealers to ascertain the usefulness of the left over seeds.

**Seed Quality Complain:** Any complain of seed quality under the Rule 23 (A) of the Seed Rules, 1968 is not defective but deficient as there is no provision of specific mode of complain and also there is no specific mode of enquiry and reporting either to penalize the seed producer or to provide any compensation to the affected farmer.

**Seed Law Enforcement:** The seed law enforcement applied to certified seed and a labeled seed looks different as the seed standards for certification samples are more inclusive than the seed standards of labeled seeds. Further, there is no mention of the names of tests required under any particular situation in seed law enforcement samples. It is commonly accepted that the seed shall be sold within the validity period and at the same time it is commonly apprehended that the seed might have lost some of the seed standards within the validity period. There shall be periodic evaluation of seed quality within the validity period at least for internal assessment.

**Seed Certification Personnel:** Availability of adequate number of trained / skilled technical certification personnel is a vital requirement.

**Organic Certification:** There is vast scope for organic certification and the future of seed certification agencies may rely on the organic products certification.

# SEED GROWER RELATED PROBLEMS AND PROSPECTS

**Multiple Seed Producers:** An individual seed grower shall take up the seed production under a single seed producer. Involvement of multiple seed producers is a problem.

**Sub letting of seed production:** The seed grower to whom the seed producer has allocated source seed shall not sub let the seed production programme to other farmers.

**Rented land and tenant farming:** When an individual seed grower uses others land for seed production then it shall be clear that it is a rented / borrowed / lease hold or any such type of land but not sub letting of seed production.

**Seed Production Field:** The seed grower shall cultivate the seed crop at the designated fields and the seed production area shall match to the registered area and quantity of source seed used.

**Scattered Fields:** In case of seed producers like state seeds corporations, several individual seed growers are involved in seed production. Since seed certification has evolved through time, the early selection of seed growers years back was scattered over large area. Further, the seed plots of individual seed growers are well apart from each other beyond the provision of not less than 50 meters.

**Seed rate and area covered:** The standards for use of specific seed rate for different crops shall be followed and the area covered shall be proportionate to the corresponding seed rate.

**Quality of Source Seed:** The quality of source seed plays an important role in seed production. The seed producer shall obtain good quality source seed for certified seed production.

**Unsuitable Varieties:** The land of an individual seed grower may be suitable for a particular category of crop and / or varieties. If suitable crop / variety is not available with the seed producer then seed production and seed certification face problem.

**Multiple Varieties:** As per the managerial ability of an individual seed grower, the crop / varieties shall be allocated. Multiple varieties of the same crop in the same season poses serious threats to seed quality.

**Multiple Classes of Varieties:** Sometimes an individual seed grower is allocated with foundation and certified class of seed of the same variety or different varieties which causes problems in seed quality maintenance.

**Production Practice:** A uniform production practice shall be followed for the seed crop. For example, normal transplanting or mechanical transplanting or line transplanting or SRI or line sowing etc. shall be followed properly.

**Agronomic Practices:** Though all the agronomic practices aim at higher production and there is no such standard agronomic practice applicable for seed crop, still then the normal agronomic practice and crop care shall be taken up by the seed grower.

**Differential Growth:** The seed grower shall take care to maintain uniformity of the seed crop throughout the seed production fields. Differential growth may cause inconvenience during field inspection and it may have adverse effect on seed quality.

**Rouging at proper stage and time:** Rouging of off types, other crop plants, weed plants, diseased plants etc. shall be shall be done at proper stage and time. Delay may lead to improper rouging and subsequently causing visible and invisible deterioration of seed quality.

**Harvest at proper crop stage:** The seed grower shall harvest the crop at proper stage of maturity of the seed crop. Both early harvest and late harvest have detrimental effects on seed quality.

**Farm Facilities:** The seed grower shall possess minimum required farm facilities such as barn yard, shed and shelter, storage facilities for the certified seeds to be handled before transport to the seed processing plant.

**Use of Farm Machineries:** Due to several reasons a number of farm machineries are in use in agriculture which is also used in seed production. There is no standard operating procedure for use of these farm machineries for seed crops. Certain farm machineries and their care less use cause serious damage to the quality of seed. Where combine harvester is used for seed crop adequate care shall be taken for seed drying.

**Lodging and other damages:** The seed crop fields suffering from lodging or any other form of crop damage before final field inspection are reported by the certification personnel. However, any damage after final field inspection shall be cared by the seed grower himself. The produce from such fields shall not be bulked with good seeds.

**Poor Packing:** The seed grower shall pack the raw seeds preferably in new bags or in well cleaned old bags for transport to the seed processing plant. Packing in un-cleaned, old and weak bags may lead to spill of seeds that may cause loss of seed and loss of seed quality.

**Joint Transport:** Two or more seed growers of a particular village or locality may resort to transport the raw seeds jointly in one vehicle to reduce transport cost. This may cause mixing of seed bags of one variety or different varieties causing serious threat to seed quality.

**Malpractice:** Sale of good quality produce of seed crop at higher prices and purchase of ordinary grain at lower prices and subsequent supply to the seed processing plant is a harmful practice which shall be prohibited.

**Isolation Distance:** Isolation distance prescribed for the crop shall be followed strictly. In most of the cases it causes invisible damage to the seed quality. Rejection due to lack of isolation distance is a serious threat to seed production in several crops. The seeds collected from the rejected fields shall be kept away from seeds of the certified fields.

**Crop Calendar:** The seed grower shall follow a crop calendar for all the activities of seed production. The seed producer shall guide the seed growers and advice of the seed certification personnel shall be followed for maintenance of seed quality.

The competitive ability of the certified seeds with the labeled seeds looks weaker due to compulsory notification of the kind / variety for certification and some other reasons therewith. Sometimes it looks that the labeled seeds of some local seed producers are far away from the reach of the seed law enforcement. The production including procurement, processing and testing; supply and distribution and trade and commerce of the labeled seeds shall be known to the enforcement authority to maintain a healthy choice of the common farmer.

Some policy matters like subsidy and any other policy of the Central Government and State Government may create problems or become prospective for certified seed production.

I as a resource person have my own problems and prospects. Whatever vast the experience may be, I belong to a particular state and have certain knowledge about few known states but lack information about all over the country. Further, the problems and prospects may vary from state to state and change with the passage of time.

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# **Determination of Seed Viability**

# A. k. Verma Sr. Seed Analyst, NSRTC, Varanasi

#### Seed Viability

"Seed viability can be defined as the ability of the embryo to live, grow and develop into a seedling under favorable environmental conditions".

or

Seed viability refers to state of aliveness

#### **Objectives of seed viability**

- To obtain quick estimation of viability of seed samples or of individual seeds remain ungerminated at the end of germination test.
- To determine the rapidly viability of the seeds of certain species which germinate very slowly or show high degree of dormancy.

#### Factors affecting seed viability

#### 1. Internal factors

- ✓ Immature and small seeds within a seeds ,within a seed lot do not store as well as mature and large seeds within a seed lot (Wien *et al*)
- ✓ Several kinds of environmental stresses during seed development, and prior to physiological maturity, can reduce the longevity of seeds.
- ✓ The physical condition and physiological state of seeds greatly influence their life span.
- ✓ Seeds that have been broken, cracked, or even bruised deteriorate more rapidly than undamaged seeds (McDonald 1985; Priestley 1986)

#### 2. Genetic factor

Seeds of some species are genetically and chemically equipped for longer storability than others under similar conditions.

Most long-lived seeds belong to species possessing hard, impermeable seed coats.Seeds of canna (Sivoriet *et al.*, 1968), Lotus (Wester 1973), and Lupinus (Porsild and Harrington 1967) have been reported to be viable even after 500 years.

Seeds of other species are characteristically short lived, these include vegetables such as lettuce, onion, and parsnip and also agronomic crops such as Rye. Generally seed species possessing high oil content do not store as well as those with low oil content. For ex, whole wheat seeds contain only about 3% oil, but their embryo portion has about 27% oil. Seeds of different species may also be chemically similar but have different storability due to differences in genetic potential. For example, Chewings Fescue and annual rye grass seeds are similar in appearance and chemical composition; however rye grass seeds have much better storability under comparable conditions. Genetic differences in storage potential are not limited to seeds of different species, It also occur among cultivars. The bean cultivar black Valentine stores better than Brittle wax (Toole and Toole 1953).However the environment strongly alters the genetic potential for seed longevity.

# Relative humidity and temperature Temperature

At a temperature of 0° c, formation of intracellular ice crystals can disrupt membrane integrity & contribute to seed deterioration. However Seeds with moisture levels below 14% do not form ice crystals. It should be noted, however, that at 14% initial moisture, seeds stored in cold rooms below 0°c will likely gain moisture. Most cold rooms have a high relative humidity & seeds achieve equilibrium with relative humidity after a brief period of storage. Thus seeds stored at low temperature must be in conditions in which the relative humidity is controlled or placed in moisture –proof containers to avoid increase in moisture content & increased deterioration.

# Seed Moisture

Seeds contain moisture above 14% begin to exhibit increased respiration, heating, and fungal invasion that destroy seed viability more rapidly. Below 5% seed moisture, a breakdown of membrane structure hastens seed deterioration. This probably a consequence of reorientation of hydrophilic cell membranes due to loss of the water molecules necessary to retain their configuration. Thus, studies standardized that storage of seeds Cereal (10-12 %), Pulses (7-8 %), Vegetables (4-5 %), Oilseeds (7-8 %) appears to be ideal; for maximum longevity.

# VIABILITY TESTS

- Standard Germination test
- Tetrazolium test
- Excised embryo test
- Fast green test
- Conductivity test

# **1. STANDARD GERMINATION TEST**

The emergence and development of seedling to a stage where the aspects of its essential structures indicate whether or not it is able to develop further in to a satisfactory plant under favorable conditions in soils (ISTA,1985).

# STEPS OF GERMINATION TEST

- Putting of seeds
- > Keeping in germinators at optimum condition
- Period of test -Days to count -Ist and II nd count
- Seedling evaluation
- Calculation of results
- Reporting of results

# **SEEDLING EVALUATION:**

- CONCEPT: Evaluation should be done only after all essential structures are fully expressed & evaluate as NS, AS, HS, FUG & dead seeds
- Normal seedlings (NS): Seedlings showing continued capacity for development into normal plant when grown in good quality soil under favorable conditions
- NS Categories (ISTA)
- Intact seedlings :Seedlings with essential structures well developed in all proportions, healthy, showing balanced growth
- Slight defective Seedlings : Seedlings with slight defects in their essentials structures provided they show normal vigorous, balanced growth in comparison with intact seedlings
- Seedlings with secondary infection : Seedlings with clear evidence of secondary infection are classified as NS provided all essential structure are otherwise normal.

Seedlings with secondary infections even if seriously decayed or diseased are considered as normal

# 2. Tetrazoloium test

Tz is a biochemical test and one of the quick methods to predict seed viability developed by Lakon (1942) in Germany.

**Viability**: Seed viability indicates that a seed contains structures and substances enzyme system which give it the capacity to germinate under favorable condition in the absence of dormancy.

# **Objectives**:

1. To obtain quick estimation of viability of seed samples or of individual seeds remained ungerminated at the end of germination test.

2. To determine the rapidly viability of the seeds of certain species which germinate very slowly or show high degree of dormancy.

# **Equipments and chemicals required:**

a. One percent solution (W/V) of 2, 3, 5 Triphenyl tetrazolium chloride (TZ) or bromide.

b. Potassium dihydrogen phosphate.

c. Disodium hydrogen phosphate.

Conditioning Media: Blotter, paper towel or beaker.

Cutting or piercing devices: Razor blade, dissecting knives and needles.

Staining dishes: Watch glasses/petridishes.

Magnifying devices: Hand lens and stereoscopic microscope.

# Preparation of buffer solution

Solution 1 – dissolve 9.078 g KH2PO4 in 1000 ml water

Solution 2 – dissolve 11.876 g Na<sub>2</sub>HPO<sub>4</sub> in 1000 ml water

Mix 400 ml of solution 1 with 600 ml of solution 2 to get a liter buffer solution of neutral pH.

To get 1% of TZ solution, dissolve 1 g of TZ salt in 100 ml of buffer solution. (The one percent solution is used for seeds that are not bisected through the embryo, while the 0.1 percent solution is used for seeds in which the embryo is bisected. Other low concentration such as 0.2 percent and 0.5 percent are some time used instead of 0.1 percent solution).

**Straining:** The prepared seed should be placed in suitable container (small beaker, Petri-dishes, watch glass, etc.) and place these container in a dark ward place. The staining time varies for different kinds of seed, different methods of preparation and different temperature (less than one hour to approximately eight hours).

A sample is satisfactorily stained when tissue develops interpretable staining characteristics and the analyst can sense embryo conditions. When observations indicate that a sample has stained sufficiently, the TZ solution should be discarded and observation can be made.

**Principle**: when the seeds are soaked in colorless solution of 2, 3, 5 triphenyl tetrazolium chloride (TZ) or bromide. it interferes with the reduction process of living cells within the seed tissue and accepts hydrogen ions from the dehydrogenase enzymes. Due to hydrogenation, (H+ ions transfer) triphenyl tetrazolium chloride get reduced into a red coloured compound, non diffusible substance called formazan. In the living cells. Since, the reactions takes place within the respiring (living) cells and the formazan is no diffusible a clear topography of living and nonliving areas within the seed can be developed by using proper procedure. For this reason, the test is designated as the topographical tetrazolium test.

The reaction as follows:

$$C_{0}H_{3} Cl \qquad N = N - C_{6}H_{5}$$

$$C_{0}H_{3} Cl \qquad 2e+2H^{+} C_{0}H_{3} C \qquad + H^{+}Cl \qquad + H^{+}Cl \qquad N = N - C_{6}H_{5}$$

$$2,3,5 = triphenyl tetrazolium chloride \qquad 2,3,5 = triphenyl formazan$$

(forms a clear solution in water)

2,3,5 – triphenyl formazan (a red stable, no diffusible substance)

**Evaluation of sample**: The sample is ready for evaluation when it is stained. Observe the staining pattern and calculate the percentage of viable seed.

#### 1. On the basis of staining of embryo

- a. Embryo completely stained- viable.
- b. Embryo unstained-non viable.
- c. Plumule or radical unstained-non viable.

#### 2. Assessment on the basis of cotyledon

- a. Complete staining-viable.
- b. Unstained-non viable.
- c. Necrosis -evolution on the basis of category.

#### 3. Assessment on the basis of necrosis

- a. Unstained tissue at the attachment of the embryo-non viable.
- b. Unstained tissues are away and are not connected with embryo-viable.

#### 4. Assessment on the basis of colour intensity

- a. Dark red -vigours seed.
- b. Pink colour -weak seed.
- c. Dark red fractured- non viable.

#### 5. Specific evaluation

#### A. Germinable seeds of cereals

- a. Well developed embryo with an fractured normal cherry red stain.
- b. Necrosis with the upper or lower ends of the scutellum.
- c. Radical unstained but embryonic axis stained.

# **B.** Non germinable seeds f cereals

- a. Whole embryo unstained.
- b. Scutellum node unstained.
- c. Major area of coleoptiles unstained.

# C. Germinable seeds of legumes/oil seeds

- a. Non fractured red coloured embryo and cotyledon.
- b. Normal red coloured embryo with only one normal cotyledon.

c. Normal red coloured embryo with half or more than half of both the cotyledons attached to embryo are of red colour.

#### D. Non germinable seeds of legumes

- a. Embryo completely unstained.
- b. Fracture at radical or plumule with dark red line.
- c. Plumule or radical tip unstained.

- d. More than <sup>1</sup>/<sub>2</sub> part of both the cotyledons attached to embryo are colourless.
- e. Attachment of embryo to cotyledon is unstained.

**Calculation:** the results are reported as percentage of viable seeds in relation to total seed tested.

#### Advantages of TZ:

1. Quick estimate of viability can be obtained (within 12-20 hrs.)

2. When the seed is dormant or very slow in germination, a viability test is extremely useful.

3. Seeds are not damaged (in dicot only) in analysis, therefore they could be germinated. **Disadvantages of TZ**:

- 1. It is difficult to distinguish between normal and abnormal seedlings.
- 2. It does not differentiate between dormant and non dormant seeds.

#### 3. Excised embryo test

- The excised embryo test is similar to germination tests in that it measures the quality of the seed by their actual germination.
- In addition it allows some measure of the embryo dormancy to be made, by counting those seeds which, although not growing normally, have grown slightly, remained firm and have kept their color for the test period.
- The test is not valid for previously germinated seeds and must not be applied to samples which contain any dry germinated seeds.
- The success of the test requires considerable skill and experience in the operator and the ISTA rules restrict it to only a few species

# 4. Fast green test

- ∠ The fast green test reveals physical fractures in the seeds such as corn.
- $\swarrow$  seeds are soaked in a 0.1% fast green solution for only 15-30 seconds.
- ✓ During this period, the fast green penetrates any area of the seed coat which has been fractured and stains the endosperm green.
- After the soak period, the seeds are washed and the fractures then become apparent (visible) in the seed coat.

# 5. Conductivity test

- The conductivity test is a biochemical test, which measures the amount of electrolytes, which leach through the seed coat or fruit coat of the intact seed.
- > A higher conductivity may indicate a low viable seed lot.
- > The expected readings for a conductivity test will vary greatly from crop to crop.
- > It is most useful for peas, soybean samples, and a lesser degree for corn.

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NATIONAL SEED RESEARCH & TRAINING CENTRE G. T. Road, Collectry Farm, Varanasi- 221106 (U.P.) Tel: 0542 2370222 E-mail: <u>dir-nsrtc-up@nic.in</u> Website: www.nsrtc.nic.in