INDEX

Course :	GPB 243	**Credit :**	3(1+2)
Course title :	Principles of Seed Technology	**Semester :**	IV

Sr. No.	Topic	Date	Page No.
1	Seed production in major cereals : Wheat and Rice		
2	Seed production in : Sorghum and Bajra		
3	Seed production in : Maize.		
4	Seed production in major pulses : Green gram and Black gram		
5	Seed production in pulses : Pigeonpea and Lentil		
6	Seed production in pulses : Gram and Field pea		
7	Seed production in major oil Seeds : Soybean, Rapeseed & Mustard		
8	Seed production in major vegetable crops : Brinjal and Tomato.		
9	Seed production in vegetable crops :Chilli and Okra.		
10	Seed production in vegetable crops : Onion		
11	Seed production in : Pumpkin, Bottle gourd		
12	Seed production in : Bitter gourd, Ridge gourd, Sponge gourd		
13	Seed sampling and testing procedure		
14	Physical purity test		
15	Seed moisture test		
16	Germination test – types of germination		
17	Germination test – different methods of germination		
18	Seed viability test		
19	Seed and seedling vigour test		
20	Genetic purity test : Grow Out Test		
21	Genetic purity test : Electrophoresis		
22	Seed certification : Procedure		
23	Field inspection, preparation of field inspection report		
24	Visit to seed production farms of cereal crops		
25	Visit to seed production farms of oilseed crops		
26	Visit to seed production farms of pulse crops		
27	Visit to seed production farms of fiber crops		
28	Visit to seed testing laboratories		
29 & 30	Visit to seed processing plant		

CERTIFICATE

This is to certify that Mr./ Miss _____
Reg. No. _____ has successfully completed all the practicals of the Course GPB 243: Principles of Seed Technology of B.Sc. (Hons.) in Agriculture, during the IVth Semester of academic year 2018-19

Place: A. C.

Date :

(Course Teacher)
Botany Section

EXERCISE NO. 1

SEED PRODUCTION IN MAJOR CEREALS : WHEAT AND RICE

Wheat

Land – Medium to Black, well drain and free from weeds. During previous season / year same crop should not be grown in the same field.

Isolation – Wheat is self-pollinated crops but 1 to 4 per cent cross pollination is reported. From other varieties of same crop : 3 meter

From smut affected plot : 180 meter.

Sowing time – First fortnight of November

Seed – 100 -125 kg/ha. Seed should be obtained from a authentic sources decided by seed certification ~~Agronomy.~~ Agency .

Fertilizer dose- 120 : 60 : 40 NPK Kg/ha.

Field Inspection : 2/3 times, before flowering, at flowering & before harvesting

Roughing: Off types, objectionable weeds, pest and disease infected plant should be removed as and when noticed.

Harvesting and yield – Harvesting is done when crop is fully matured. Take maximum care to avoid mixing of seed during harvesting threshing bagging etc.

Yield: 20-30 qtls/ha.

Germination – 85 % , physical purity – 98 %, moisture = 8 to 12 %

Rice :

The rice is offenly self-pollinated crop with less than 0.1 % natural out crossing.

- In rice WA (Wild Abortive) source of cytoplasm is used in hybrid seed production.
- 'A', 'B' 'R' line approach (Three Line Breeding) for rice hybrid has been developed by the scientist "Yuan Long Ping" in China during 1973 -first time in the world.
- For commercial hybrid development in rice, there are Four different approaches.

1. Three line method or CMS system
2. Two line method or PGMS / TGMS system.

3. One line method or Apomixis system
4. Chemically induced made sterility method.

- The scientists from China Dr. Yuan Long Ting. developed first hybrid rice in the world using CGMS source during 1977.
- The source of cytoplasm used in hybrid seed production of rice is WA (Wild Abortive).
- Dr. Yuan Ling Ping and his team in Hainan island of Southern China developed the practical usable CMS system in rice during 1970.
- Dr. Yuan Ling Ping, reported the first attempt of identification of Three Line Approach for hybrid rice during 1973.
- Later on Dr. Yuan Ling Ping developed first hybrid based on CGMS in the year 1977 using the "Wild Abortive" male sterile cytoplasm
    - Accordingly there are four approaches for hybrid rice development.

1] Three line method or CMS system

2] Two line method or PGMS / TGMS system.

3] One line method or Apomixis system

4] Chemically induced male sterility method.

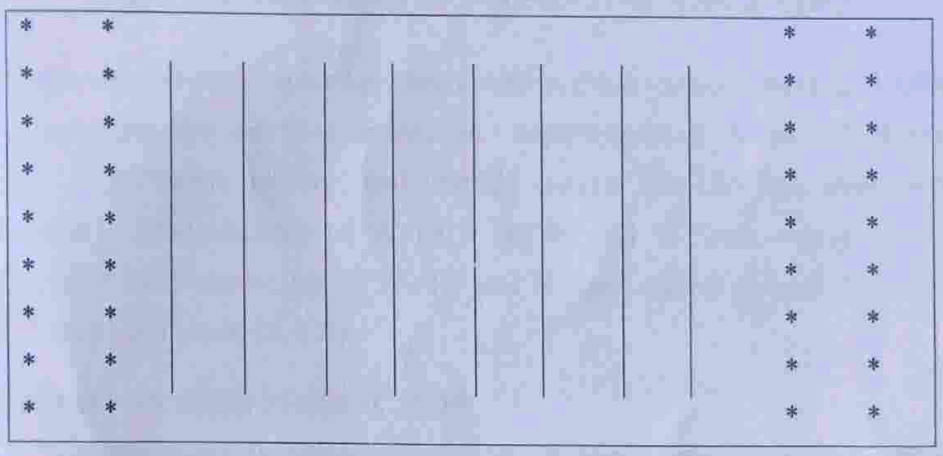
CERTIFIED (AxR) RICE HYBRID SEED PRODUCTION (CGMS)	
	Sahyadri— 4
Parentage	IR-58025 A x KJTR-4
Year of release	2006
Land requirement	The previous crop should not be rice or preferably fallow land is selected for seed production. Free from *Oryza sativa* var fatua
Sowing time	Sowing of seed in nursery so adjusted to attain simultaneous flowering of both seed parent and pollinator parent. Wind direction is to be considered for effective pollen available by wind to MS lines.
Sowing methods / Planting geometry	The seedlings should be of 21. to 25 days old age. For achieving proper synchronization, transplanting the seedling of right age is a must. Transplanting of seedlings

	across the wind direction to facilitate nicking and simultaneous flowering of seed parent (A) and pollinator parent (R) for effective pollination. The male parent (R) is sown in 2 - 3 sowing dates (staggered sowing).
Synchronization	Synchrony of flowering of male and female parent is a important factor to be considered. If not considered the : seed set on female parent is affected due to improper pollen supply by male parent during flowering. In view of this , it is aptly said that half the success is achieved if one is able to achieve good synchronization. Thus, For synchrony of flowering the periodical 3-4sowings are made for (R) male parent. Also transplanting of seedlings of different ages is done. The flowering maybe advanced by 2 - 3 days by urea (3 % solution) spray. Draining water from field may delay flowering.
Spacing (cm)	Seed parent (MS line) - 15 x' 1 5 cm. Pollinator (R) = 30 x 15 cm. [As shown in Figure]
Staggering	Staggered planting in nursery of male (pollinator) parent in 2 - 3 sowing dates staggered planting.
Planting ratio Female : Male &Seed rate kg/ha	Female and male parents are planted in 10:2with seed rate of 8:2 ratio with seed rate of 20: 15 Female : Male 6 : 1 ratio with seed rate of 25 : 10 Female : Male. The row ratio should be such that the male row would be able to pollinate the female rows most effectively.
Fertilizers Kg/ha	150:60:50 NPK kg/ha.
Method of improving seed setting in hybrid seed production	
1) Supplementary pollination	Natural cross pollination is supplemented with pollination using rope pulling method which is done 3 -5 times on calm clay during anthesis period.
.2) Flag leaf clipping	Enhancing out crossing rate is one of the key factor to increase the seed yield. Normally flag leaves are erect and longer than panicle and they come in way of easy pollen

	dispersal thus affecting out crossing rate. Thus, flag leaves are clipped off when the main culm are in blooming stage, which helps in uniform pollen movement and wide dispersal pollen grains to give better seed setting as the crop is often self pollinated.
3) G. A. application	Most of WA based CMS lines have imperfect panicle exertion with 10 to 15 % spikelet are enclosed in flag leaf and are not available for out crossing. Thus, GA_3 spraying (20 ppm) for Two' to Three times is applied on seed parents at initial heading stage to improve their panicle exertion.
Isolation Distance (Meter)	100 meter or isolation period of 21 days is maintained.
Rouging	Rouging of off types, volunteer plants, wild rice plants,
Spacing (cm)	Seed parent (MS line) - 15 x' 1 5 cm. Pollinator (R) = 30 x 15 cm. [As shown in Figure]
Staggering	Staggered planting in nursery of male (pollinator) parent in 2 - 3 sowing dates (staggered planting).
Planting ratio Female : Male &Seed rate kg/ha	Female and male parents are planted in 10:2with seed rate of 8 : 2 ratio with seed irate of 20 : 15 Female : Male. 6 : 1 ratio with seed rate of 25 : 10 Female : Male. The row ratio should be such that the male row would be able to pollinate the female rows most effectively.
Fertilizers Kg/ha	150:60:50 NPK kg/ha.
Method of improving seed setting in hybrid seed production	
1) Supplementary pollination	Natural cross pollination is supplemented with pollination using rope pulling method which is done 3 -5 times on calm clay during anthesis period.
.2) Flag leaf clipping	Enhancing out crossing rate is one of the key factor to increase the seed yield. Normally flag leaves are erect and longer than panicle and they come in way of easy pollen dispersal thus affecting out crossing rate. Thus, flag leaves

	are clipped off when the main culm are in blooming stage, which helps in uniform pollen movement and wide dispersal pollen grains to give better seed setting as the crop is often self pollinated.		
3) G. A. application	Most of WA based CMS lines have imperfect panicle exertion with 10 to 15 % spikelet are enclosed in flag leaf and are not available for out crossing. Thus, GA_3 spraying (20 ppm) for Two' to Three times is applied on seed parents at initial heading stage to improve their panicle exertion.		
Isolation Distance (Meter)	100 meter or isolation period of 21 days is maintained.		
Rouging	Rouging of off types, volunteer plants, wild rice plants,		
	plants affected by disease and stem borer. The pollen shedders from female parent.		
	Minimum 2 field inspections. One at prior to flowering. Ear emergence and second between dough and maturity stage.		
	0.20		
	0.02 *Qryza satiya var fatua*	Objection able	0.50
Harvesting	Harvest the male parent first separately to avoid chances of mechanical mixture. Later on female parent seed separately.		
Seed yield Q/ha	5 to 15 Q/ha,		
AFTER HARVEST			
Seed moisture %	13		
Germination %	80		

NOTE : Maximum pure seed (97%) and inert matter (2 %) is recommended as physical purity standard.



* *Plants of Male Parental Line. – Plants of Female Parental Line The Figure (Above) Showing the Planting method of Hybrid Seed Plot

Certified Hybrid Seed Production of Rice :

THE RICE HYBRIDS DEVELOPED SO FAR,

USING CGMS "WA"CYTOPLASM

	Rice	Commercial Hybrids	State
1.	Sahyadri - 1	IR - 58025 A x BR 827 - 35 - 3 - .1 - 1 R (1998)	Maharashtra
2.	Sahyadri-2	IR - 58025 A x KJTR -2 (2004)	
3.	Sahyadri -3	IR- 58025 Ax P-CJTR-3 (2005)	
4.	Sahyadri -4	IR - 5 8025 A x KJTR -4(2006)	
5.	APHR-1	IR - 58025 A x Vajram.(1994)	
6.	APHR-2	IR -.62829 A x MTU -9992 (1994)	A./P.
7.	CORH-1	IR-62829AxIR-10198 (1994)	Tamil Nadu
8.	CORH-2	IR-58025AxC2OR	-,, -
9.	KRH-1	IR - 58025 A x IR- 9761 (1994)	Karnataka
10.	KRH-2	IR 58025 A xKMR 3(1996)	- ,, -
11.	CNRH-3	IR-62829AxAjay (1.995)	W.Bengal
12.	DRRH - 1	IR-58025 AxIR-40750 (1 996)	A.P.
13.	Pant Sankar Dhan- 1	IR -58025 AxURPR193-133(1997)	U.P.

******

EXERCISE NO. 2

SEED PRODUCTION IN : SORGHUM AND BAJRA

A] Foundation and certified seed production of sorghum varieties, hybrids, synthetics and composites). Nucleus Seed Production = Maintenance of 'A' line (MS line) (A x B), 'B' line (maintainer line) and Fertility restorer line (R) line, Breeder Seed Production = Multiplication of 'A' (A x B), 'B' and 'R' lines. Foundation Seed Production = Seed production of 'A', 'B* and 'R' lines. Certified Seed Production = Hybrid seed production (A x R)

FOUNDATION SEED PRODUCTION

Seed production (multiplication) of 'A', 'B' and 'R' lines. Seed production (multiplication) of' A',

- The A line is a male sterile line.
- It is maintained by crossing with maintainer line ie B line (A x B crossing

SORGHUM. *Sorghum bicolor* L. Mench.		
	H.Y.V	Hybrids
Land requirement	Free from striga weed, Johnson and forage grass. During Previous year same crop should not be there in the field	
Varieties/ Hybrids	KHARIF:SPV-946, SPY - 475 RABi:M-35-I,Sei.3, Pnule yashoda, Mauli	KHARIF:CSH-16, CSH-17, CSH-18 & CSH-23. RABI:CSH-15(R), CSH-19(R)
Sowing time	KHARIF : up to 15th July, RABI: = 15th September to 30th October..	
Sowing methods	By drilling.	By drilling.
Spacings (cms)	45 x 15	45x15
Planting ratio Female : Male		On row basis Female : male 4:2with 4 border rows of male parent all around seed plot.
Seed rate kg/ha	10 to 12	Female = 8 Male = 4
Fertilizers Kg/ha	40:60:60 at sowing and 40:00:00 after one month of sowing.	
Isolation Distance	Foundation = 300	Foundation = 300

	Certified = 200, 400 meter.	Certified - 200, 400 meter
(Meter)	From Johnson grass & , forage sorghum	From Johnson grass & forage sorghum
Field inspection	Minimum 3	Minimum 4
Off types %	Foundation = 0.05 Certified = 0.10	Foundation = 0.05 Certified -0.10
Pollen shedders % when MS line is used	-	Foundation = 0.05 Certified -0.10
Objectionable weeds %	F = 0.05, C = 0.10 Johnson grass, forage sorghum ie. Sudan grass	
Objectionable disease %	F = 0.05, C = 0.10 Grain smut, head smut, sugary disease ie. Ergot	
Seed yield Q/ha	30-40	04-06
AFTER HARVEST		
Seed moisture %	12.0	
Germination %	Minimum 75.0	
Seed borne disease	Grain smut, head smut, sugary disease i.e. Ergot	
Major pests	Shoot fly, stem borer, midge fly, aphids, caterpillar	

NOTE : Maximum pure seed (98 %) and inert matter (2 %) is recommended as physical purity standard.

- The commercial hybrids are developed by utilizing cytoplasmic genetic male Sterility.
- Stephan and Holland (1954) discovered cytoplasmic genetic male sterility [CGMS] in sorghum due to interaction between rail o cytoplasm and kafir nuclear factor.
- There are 4 sources of male sterile cytoplasm ie. A, A_2, A_3& A_4.
- The A1 (Milo cytoplasm) sources of male sterile cytoplasm is utilized commercially.
- 'A', 'B', 'R' line approach (Three Line Breeding) for sorghum hybrid has been developed and utilized for hybrid seed production.
- The commercial hybrids are mostly based on Milo Cytoplasm.

MAINTENANCE BREEDING OF SORGHUM.

1. Maintenance of male sterile line "A"
2. Maintenance of maintainer line "B"
3. Maintenance of restorer line "R".

DIFFERENT SORGHUM HYBRIDS 'RELEASED

Sr No	Hybrid	Parentage	Year	Place
1	CSH-1	CK60AxIS-84	1964	NRCS Hyderabad
2	CSH-2	CK 60 Ax IS -3691	1965	NRCS Hyderabad
3.	CSH-3	2219 Ax IS -3691	1970	NRCS Hyderabad
4	CSH-4	1036 Ax SWARNA	1973	NRCS' Hyderabad
5	CSII-5	2077 A x CS- 3541	1974	INK^sHyderabad
6	CSH-6	2219 A xCS-3541	1977	NRCS Hyderabad
7	CSH-7	36 Ax 168	1977	Parbhani
	CSH-7(R)	36 Ax 168	1977	Parbliani
8	CSH-8	36AxPD3-l-ll	1977	Parbhani
	CSH-8(R)	36AxPD3-l-ll	1977	Parbhani
9	CSH-.9	296 A x CS - 3541	1982	NRCS Hyderabad
10	CSH-10	296 Ax SB -1085	1984	Dharwad
11	CSH-11	296 Ax MR- 750	1986	ICRISAT Hyderabad
12	CSH-12	296 A xM 148/138	1986	UAS, Dharwad
	CSH-12 (R)	296 AxM148/138	1986	UAS, Dhanvad
13	CSH-13	296A.xRS-29	1992	NRCS Hyderabad
	CSH-13 (R)	296AxR3-29	1991	NRCS Hyderabad
14	CSH-14	AKMS14AxRS-29 150	1994	PKV,Akola
15	GSH-15	104 Ax RS-585	1992	NRCS Hyderabad
	CSH-15(R)	104AxRS-585	1992	NRCS Hyderabad
16	CSH-16	27AxC-43	1997	NRCS Hyderabad
17	CSH-17	AKMS14AxRS-673	1998	NRCS Hyderabad
18	CSH-18	IMSL9AxIndore~12	1999	Indore
19	CSH-19	104Ax R-354	2000	PKV.-Akola

20	CSH-20 (MF)	2219AxUPMC-503	2005	Pantnagar Multi cut forage
21	CSH-21 (Fodder)	MLSA848 x MLR -34	2005	Mahindra Seed
22	CSH-22 (SS)	ICS 38 A x SSV - 84 Sweet Sorghum	2004	NRCS Hyderabad
23	CSH-23	MS.7A-X R627	2007	NRCS Hyderabad

DIFFERENT SORGHUM HYBRIDS 'RELEASED'

Sr No	VARIETIES	Parentage & breeding method used	Year	Place	Remarks
1	CSV - 1	Selection from IS 3924	1968	NRCS Hyderabad	
2	CSV -2	IS 3922 x Karad local	1974	NRCS Hyderabad	
3	CSV -.3	IS 2954 x 13 P 53	1974	NRCS Hyderabad	
4	CSV -4 (CS 3541)	IS 3675 x IS 3541 Pedigree selection	1974	NRCS Hyderabad	
5	CSV -5	IS 3687 x Aispuri local	1974	NRCS Hyderabad	
6	CSV- 6	IS 3922 x Aispuri local	1974	NRCS Hyderabad	
7	CSV-7(R)	IS 2950 xM 35-1	1978	AICRP Hyderabad	
8	CSV~8(R)	•R24xR16	1977	AICRP Hyderabad	
9	CSV -.9	CS 3541 Tall mutant	1982	AICRP Hyderabad	
10	CSV.- 10	SB 1066 x CS 3541	1983	Udaipur	
11	CSV -11	SC 108-3 xCS 3541	1985	ICRISAT	
12	CSV -12 (SPV 462)	2947 x 232) x CO 22	1985	Coimbatore	
13	CSV -13	(IS 12622 x 555) x [(IS3612x2219B) x M35-1]	1988	ICRISAT	
14	CSV -14 (R)	M 35 - 1 x (CS 2947 x CS 2644) x.M 35 - 1	1992	AICRP Hyderabad	
15	CSV -15	SPV 475 x SPV 462	1996	AICRP Hyderabad	
16	SPV- 96	Selection from 148x512		AICRP Hyderabad	
17	SPV -102	Selection from (148 x		NRC	
		512) x (148x370)		Hyderabad	
1.8	SPY -126	Tall mutant from CSV -4		AICRP Hyderabad	
19.	SPY -351	Selection from SC108-3xCS3541		1CRISAT Hyderabad	..
20	CSV -17	SPY 946 x SPY 772	2003		
21	CSV- 18	CR-4 x IS 18370	2004		
22	M35-1	Selection from local Bedar area. Pure line selection)	1938	Mohot (Solapur)	

23	Swati	SPY 86 x M 35 - 1 Pedigree selection	1984		
24	Selection -3	Pedigree selection from local land races	1994	Solapur ,	
25	Phule Yashoda	Pedigree selection from local land races. Pure line selection	4998	Rahuri	
26	Phule Maulee	Pedigree selection from local land races. Pure line selection	1999 *	Rahuri	
27	Chitra (SPY1546)	SPV655xRSLG112	2005	Rahuri	
		HURDA SORGHUM			
	Uttara	Selection from local Germplasm	2005	Rahuri	
		FORAGE SORGHUM			
	Phule Amruta	RSSV 2 x SPY 462	2003	Rahuri	
		SWEET SORGHUM			
	SSV 84	Selection from IS 23568	1991	Rahuri	
	CSV19SS	RSSV 2 x SPY 462	2004	Rahuri	

Bajra

PEARLMILLET Bajra) *Pennisetum glaucum* (L.) R. Br.		
	H.Y.V	Hybrids
Land requirement	Free of volunteer plants, No previous crop of bajra.	
Sowing time and methods	Drilling of sowing method 2nd fortnight of June [Kharif] Drilling of sowing method 2nd fortnight of January [Summer]	
Varieties / Hybrids	WCC-75,ICTP-8203, ICMV - 221 RHR - 1 (composite) and Raj - 171	MH-179.ICMH-451, Shraddha, Saburi, Shanti, MLBH-267, Mahalaxmi, GK- 1004, No. 7626.
Spacings (cm)	45x15	75x20
Planting ratio Female : Male	------	6 Female : 2 male
Seed rate kg/ha	3 kg	Female =1.5 Male = 0.75
Fertilizers Kg/ha	60 : 3,0 : 30	
Isolation Distance (Meter)	Foundation =1000 Certified = 200	Foundation = .1000 Certified- 200; (5 meter for other hybrid programme involving same parent)
Field inspection	3	4
Off types %	Foundation = 0.05 Certified = 0.10	Foundation = 0.05 Certified = 0.10-
Pollen shaders % when MS line is used		Foundation = 0.05 Certified = 0.10
Objectionable weeds %	----------	
Objectionable disease %	F 0.05 for downy mildew 0.02 for ergot C 0.10 for downy mildew & ergot C 0.04 for grain smut	F 0.05 for downy mildew 0.02 for ergot C 1 .0 for downy mildew & ergot
Seed yield Q/ha	15 - 20	03 - 04

AFTER HARVEST	
Seed moisture %	12.0
Germination %	Minimum 75.0
Seed borne disease	Wilt, leaf spot, yellow mosaic virus, sterility. Mosaic virus
Major pests	Shoot borer, pod borer

PEARL MILLET HYBRID SEED PRODUCTION:

1. Burton (1958) identified cytoplasmic genetic male sterility with Tifton cytoplasm in bajra.
2. 'A', 'B', 'R' line approach (Three Line Breeding) for bajara hybrid has been developed and utilized for hybrid seed production.
3. The commercial Bajara hybrids are mostly based on Tifton Cytoplasm.

	WCC-75	Composite from world germplasm		ICRISAT	
45	RHRBH 8609 (Shraddha)	RHRBI Ax RHRBI 138	1990	MPKV Rahuri	Highly popular
46	RHRBH 8924 (Saburi)	RHRBSAx RHRBI 458	1995	MPKV Rahuri	Highly popular
47	PUSA 605	846 Ax PPMI 69	1997	IARI	Moderately popular
48	PUSA 415	576 Ax PPMI 85	1999	IARI	Moderately popular
49	RHRBH 9808 (Shanti)	RHRBBAx RHRBI 1314	2007	MPKV Rahuri (Dhule)	Very good bread quality

EXERCISE NO. 3

SEED PRODUCTION IN MAIZE

MAIZE (*Zea mays*)		
Land requirement	Maize performs best on well leveled piece of land with good drainage. Undulated and uneven fertility gradients in soil will create the problems of uniform emergence and vigour. Sandy loam to silky loam soils with 5.5 to 7.5 PH is suitable for proper cultivation. The land should be free from volunteer plants and objectionable weeds.	
Varieties / Hybrids	Early = Kiran, Panchganga Madhuri Midlate = Manjari, Navjot, Prabhat, Umber popcorn Late = Dhaval, African tall	Deccan hybrid- 109 Pusa hybrid - 2, MMH- 133 DMH-107, MMH-69 BIO 9681, SSF 9374, JK 2492, PRO-311, Trishalata for rabi.
Sowing time	Kharif = June, Rabi = October, Summer = 2nd fortnight of January	
Sowing methods	Drilling	Drilling =
Spacing (cms)	**60 x 20**	75 x 22.5
Planting ratio Female : Male	----------	6 : 2 Single hybrid 4 : 2 Double hybrid
Seed rate kg/ha	15 - 20	Female = 12, Male = 5
Fertilizers Kg/ha	120:60:40	
Isolation Distance (Meter)	a) For different kernel colour = F : 600, C: 300 b) For same kernel colour = F : 400, C: 200 c) For Teosinate = F : 600, C : 200 d) Same inbreed/hybrid field = F : 400, C : 200 not confirming MS CS	
Field inspection	2	4
Off types %	Foundation & Certified =1.0	Foundation 0.20 Certified = 0.50
Pollen shaders % when MS line is used	------	Foundation 0.50 Certified = 1.00 for shedding tassel also

Objectionable weeds %		
Objectionable disease %	Downy mildew, Helminthosporium, stalk rot, ear rot &. kernel rot	
Seed yield Q/ha	25-30	Inbreeds =30 Single Hybrid = 6-8. Double Hybrid =10-25.
AFTER HARVEST		
Seed moisture %	12.0	
Germination %	Minimum 90.0	F = 90, C = 90
Seed borne disease	Wilt, leaf spot, yellow mosaic virus, sterility mosaic virus	

MAIZE HYBRID SEED PRODUCTION

Different terminologies

1) Single cross hybrid: It is a cross between two inbred lines i.e. F_1 hybrid between Ax B.
2) Three way cross hybrid : It is a first generation of a cross between a single cross and inbred line ie. (A x B) x C. Means three inbreeds are involved.
3) Double cross hybrid : It is a first generation of a cross between two. single crosses i.e. (A x B) x (C x D). Means four inbreeds are involved.
4) Double Top cross hybrid : It is a first generation of a cross between an inbred line and an open pollinated composite variety ie. A x (OPV) open pollinated composite variety.
5) Multiple cross hybrid : It is a combination of more than four inbred lines. They are of high adaptability, ie. (A x B) x (C x D) x (E x F)x (GxH).

THE VARIOUS STEPS INVOLVED IN MAIZE HYBRID SEED PRODUCTION ARE:

a) Maintenance if inbred lines (parental lines).
b) Production of single cross hybrid.
c) Production of double cross hybrid.
d) Production of multiple cross hybrid.

TABLE showing Isolation blocks need if all three generations of seed multiplication are being taken at one place.

	Hybrid Type	Number	Isolation Block Requirements
1.	Single cross hybrid (A xB)	Three	First two isolations for two inbreed lines, breeder seed and foundation seed production. Third isolation for certified seed production (A x B),
2.	Three Way Cross (A xB) x C	Five	Three isolations for three inbreed lines. One isolation for F_1 seed production (A x B) as foundation seeds. One isolation for production of certified seed (A x B) x C.
3.	Double cross hybrid (A x B) x (C x D)	Seven	Four isolations for four inbreed lines ie. A, B, C, & D. One isolation for FI seed production (A x B) as foundation seeds. Two isolation for seed production of two parental single cross hybrids ie. (AxB) & (CxD). One isolation for certified double cross hybrid ie. (A x B) x (CxD).
4.	Double Top Cross (A xB)xOPV	Five	Three isolations for two inbreed and one OPV lines. One separate isolation for FI seed production ie. (A x B) as foundation seeds. One isolation for production of certified seed [(A x B) x OPV]

DETASSELING IN MAIZE :

The maize crop is a monoecious nature i.e. Male and female inflorescences are located on different parts of the same plant. This condition is advantageous for emasculation.

1. After 50 days of sowing the female line are strictly observed or watched for appearance of tassel.
2. In some cases shedding of tassel may start in the flag leaf itself.
3. Therefore the whorl of ieaf sheath may be opened and tassel is removed.
4. The tassel (male flower) are removed before anthesis and this operation is

called Detasseling.

5. The silk (stigma)of detasseled female plant is allowed to pollinate with desired male parent. .
6) Therefore from time to time first tassel appearance, Detasseling work has to be continuously carried without fail until the last tassel on female line is removed.

PRECAUTIONS WHILE DETASSELING:

- It should be ensured that entire pollen bearing part is removed from seed parent,
- Do not hold the tassel too low on the stalk as otherwise plant tops may be pulled out.
- Detasseling should carried out without interruption irrespective of any type of hindrance.
- Female row should be cleared of all suckers, lodged and damaged plant if any, Start detasseling every day from the same side to identify escaping plant
- One should check detasseling, after detasseler has started the work.

SEED PRODUCTION OF SYNTHETIC VARIETIES IN MAIZE:

SYNTHETIC VARIETY: A variety synthesized by crossing inter - se a number of genotypes selected for good combining ability in all possible combinations with subsequent maintenance of variety by open pollination is known as synthetic variety (Allard, 1960), The seed production of synthetic variety involves following steps.

A] Synthesis of synthetic variety & nucleus seed production.

B] Breeder seed production.

C] Foundation and certified seed production.

SEED PRODUCTION OF COMPOSITE VARIETIES IN MAIZE:

The term composite variety refers to a germplasm composite which is commonly used to designate a broad group of materials mixed together in many different ways, and include breeding materials put together on basis of desirable characters such as yield potential, maturity, disease resistance etc. The seed production of composite variety has different four steps.

A) Synthesis of composite.
B) Breeder seed production
C) Foundation seed production
D) Certified seed production.

EXERCISE NO. 4, 5 & 6

SEED PRODUCTION IN MAJOR PULSES : GREEN GRAM AND BLACK GRAM, PIGEONPEA AND LENTIL, GRAM AND FIELD PEA

Land requirement – Medium to Black, well drained
Sowing Time – 3rd week of June to 1st week of July
Other details –

Particulars	Pigeon pea	Gram	Greengram	Blackgram
Seed rate kg/ha	15-25	60-100	15-20	15-20
Spacing cm	45x10 60x20	30x10	10	10
Isolation distance B/F meter C	200 100	10 5	10 5	10 5
Germination %	75	85	75	75
Fertilizer dose. NPK Kg/ha.	25:50:00	25:50:00	20:40:00	20:40:00
Inspections	2/3	2/3	2/3	2/3
Roughing	Off types, diseased plant should be removed as noticed.	Off types, diseased pl. should removed as noticed.	Off types, diseased plant should be removed as noticed.	Off types, diseased plant should be removed as noticed.

EXERCISE NO. 7

SEED PRODUCTION IN MAJOR OILSEEDS : SOYBEAN, RAPESEED & MUSTARD

1	Land	Soybean	Mustard / Rapeseed
	Requirement	Medium to black, well drained, previous season soybean grown land should be avoided	Medium to black ,well drained, previous season mustard crop grown land should avoided
2.	Spacing (cm)	45 x 10	45 x 15
3.	Fertilizer NPK, Kg/ha.	50:75:00	50:25:00
4.	Seed Rate (kg/ha)	75 kg / ha	5 kg / ha
5.	Isolation distance in meter	3 (all stages)	50 (F), 25 (C)
6.	Germination %	70 %	70 %
7.	Inspections	2/3	2/3
8.	Roughing	Off types, diseased plant should be removed as when noticed.	Off types plants should be roughed out before anthesis.

EXERCISE NO. 8

SEED PRODUCTION IN MAJOR VEGETABLE CROPS : BRINJAL AND TOMATO.

Foundation and Certified Seed Production of Brinjal [*Solanum melongena* Linn.]		
	FOUNDATION	**CERTIFIED**
	Hybrids are developed by hand emasculation and pollination. In such crop on hand pollination produces 1000 to 1200 seeds in single fruit. Therefore it will not be expensive to produce hybrid seed. The flower is quite amenable for rapid emasculation and pollination. Both suitable male and female parents are sown side by side. During flowering, The flower bud which may open during next morning are selected for emasculation. The emasculation is done in evening of earlier days. The buds are opened with the help of forceps and the stamens are removed. These buds are bagged in a butter paper to avoid contamination. Next day morning the desired pollens collected in Petri dish from male parental flowers. The pollens from Petri dish are dusted with the help of brush on stigma of the bud emasculated during evening. The pollinated buds are again bagged with different colour bag to identify separately.	
	The land selected should be free from volunteer plants, that means no previous brinjal crop in the same field. The land should be fertile, rich in organic matter.	
	Manjarigota, Vaishali, Pragati,RHR-B~9-2-l-l. Pusa purple Round, Pusa purple Long, Arka kusumakar, Arka shirish, Pusa kranti Arka navneet	Pusa hybrid -5 Pusa hybrid -6 Krishna (MPKV)
Source of seed	Breeder seed	Foundation seed
Source rate	350 -450 gram /ha. The 6 to 7	3 50 -450 gram /ha. The 6

kg/ha	nursery bed of 7.5 mt long, 1.20 mt wide and 10 -15 cm height for one hectare planting.	to 7 nursery bed of 7.5 mt long, 1.20 mt wide and 10-15 cm height for one hectare planting.
Isolation Distance (Meter)	200 meter	100 meter
Field inspection	Minimum 3 First before flowering, Second during flowering, Third after fruit matures/ before harvesting.	
Roughing	Off types may be removed- depending upon the varietal characteristics before flowering. Further rouging will be based on fruit shape, size, colour. Blight and little leaf disease affected plants are to be removed as and when noticed immediately.	
Objectionable weeds %	----------	----------
Objectionable disease %	Phorriopsisblight Foundation = 0.10	Certified = 0.50
Off types %	Foundation =0.10	Certified =0.20
Germination %	Foundation = 70	Certified = 70
Seed moisture%	Foundation = 08	Certified =08
Harvesting, Yield & seed extraction	The harvesting is done when the fruits are fully ripe and colour changes to yellow. After harvesting the outer fruit covering is peeled out and the flesh with seeds is cut in to thin slices. These slices are soaked in water till the seeds are separated. The material is allowed to stand overnight in this condition due to which seed separation from pulp become easier. After separation the seeds are dipped into the water. The seeds which float on water should be rejected. Later on the seeds are dried in partial shade to moisture content of 8%. The 8 quintals of seed/ha can be achieved.	
Purity %	Foundation = 98	Certified = 98
Innert matter	Foundation = 02	Certified = 02

Tomato

Tomato = *Lycopersicun esculentum* Mill.

It is self pollinated crop.

FOUNDATION /CERTIFIED SEED PRODUCTION OF TOMATO HYBRID/VARIETY	
	The hybrid development is specially in Hybrid. In variety seed production remaining all points required is same.
Varieties / r/brids	Varieties :Pusa ruby. Bhagyashree, Phule raja, Rajashree, Dhanashree, Arka meghali, Arka vikas, Arka saurabh, Pusa rohini, Pusa gaurav Hybrids : TH - 2312 (PAU 1991), Vasundhara (MAU)
Hybrid development	Hybrids are developed by hand emasculation and hand pollination. Both male and female parents are sown side by side. During flowering, cone of anthers having short filaments are removed and bilobed stigma is pollinated.
Source of seed	Foundation seed of both female and male parents are to be used.
Land requirement	Land should be free from seed borne disease. No previous tomato crop in the selected field.
Sowing time	Kliarif = July, Rabi = September, October
Sowing methods	Transplanting in the evening hours to avoid mortality.
Spacing (cm)	90 x 30 in *Kharif*
Fertilizer Kg/ha.	Sowing = 50 : 50 : 50 and 50:00:00 after one month of sowing.
Seed rate kg/ha	0.50 on [20 raised beds of 2.0 x 1.25 meters]
Isolation Distance (Meter)	200 meters for foundation. 100 meters for certified.
Staking	Staking of plants improves the yielding ability.
Field inspection	Minimum Three. First Before flowering, Second during Flowering time and Third at Maturity &Fruit ripening .
Rouging	Firstly on the basis of growth characters, off types should be removed. Secondly at the time of flowering on the basis of flower character, off types should be removed.

	And thirdly, the rouging should be taken up Vigorously on the basis of plant foliage and fruit characters. The plant fails to show truthfulness to the variety should be removed from the seed plot. Disease affected, virus affected and insect pest affected plant be removed as and when observed immediately.
Off types%	Foundation = 0.10, Certified = 0.20
Pollen shedders % when MS line is used	----------
Objectionable weeds %	----------
Objectionable disease %	Foundation = 0.10 Certified = 0.50 Early blight, Leaf spot, Tobacco Mosaic Virus.
Harvesting & Seed extraction	For seed extraction from tomato, there are three methods. 1) Fermentation 2) Alkali treatment and 3) Acid treatment. [These methods are studied in detail in HORT-232] .Complete ripened fruits should be harvested and crushed under feet in wooden boxes and allowed to ferment for 24 to 48 hours. Later seeds are washed with water through a sieve and dried in sun. On large scale HCL can be used for fermentation one liter for 1 00 kg fruits. On an average, 50 to 200 kg fruits are required for one kg of seed.
Seed yield Q/ha	
AFTERHARVEST	
Seed moisture %	Foundation = 08 Certified = 08
Germination %	Foundation = 70 Certified = 70
Seed borne disease	Bacterial wilt
Major pests	Cut worm, Jassids, Fruit borer, Root-knot-nematodes.

EXERCISE NO. 9

SEED PRODUCTION IN VEGETABLE CROPS :CHILLI AND OKRA

Foundation and certified seed production of Chilly (varieties and hybrids).

- Chilly [*Capsicum annum_var accuminatum* Linn.]
- Chilly is often cross pollinated crop.
- The pungency in chilly is due to "oleoresin *capsicin*" which is volatile alkaloid.
- In small chilies the capsicin content is more (highest) as compare to large.,
- The capsicin content in chilly is considered to be inversely proportional to the size of fruit.
- The chilly has a very good medicinal value.

Foundation and Certified Seed Production of Varieties [Chilly]		
	Foundation	Certified
Land requirement	There are no requirements as to previous crop, however, the land should be free from volunteer plant. The land selected for seed production the soil should be well drained and aerated.	
Varieties	Parbhani tejas, Konkan kirti, Musalwadi, Agnirekba, Phule sai, Phule mukta, Surakta, Jayanti, Pusa jwala, Sankheswari-32, Arka basant, Arka gaurav, Arka mohini	
Source of seed	Breeder seed	Foundation seed
Fertilizer Kg/ha.	FYM = 25 tons for rainfed and 50 tones for irrigated. 175 Kg each Ammonium sulphate and Single super phosphate + 100 Kg Potassium sulphate at the time of transplanting. Top dressing of 175 kg Ammonium sulphate 40 - 45 days after transplanting.	
Seed rate kg/ha.	1 to 1 .2 Kg/ha for raising the nursery. Sowing in the raise beds of 2.0 x 1 .25 meter, 25 such beds are sufficient for transplanting of one ha.	
Isolation Distance (Meter)	400 meter	200 meter
Field inspection	Three field inspections. First before flowering, Second at the time of flowering and Third at pod maturity stage.	
Rouging	The off types plants are removed at least thrice during crop growth. Firstly 'before flowering on the basis of plant characters. Secondly, at the time of flowering on the basis of flower characters. Thirdly at the time of pod maturity on the basis of pod characters. At all the stages, disease & insect pest affected, virus affected plants are removed as soon as they are noticed.	
Objectionable weeds %	None	
Objectionable disease %	Anthracnose, Leaf blight.	
Off types %	None	None

Germination %	60	60
Seed moisture%	08.00	08.00
Yield		
Purity %	98.00	
Innert matter	02	

OKRA/BHENDI /LADIES FINGURE (varieties and hybrids).

Foundation and Certified Seed Production of Varieties		
	Foundation	Certified
Land requirement	The selected land should not have previous crop of okra, so that it will be free from volunteer plants. The land should be free from wild okra. The soil should have rich in organic matter with good water holding capacity. The soil should not be low lying, marshy and heavy clay and should be well leveled.	
Varieties	Pusa sawani, Pusa 8makhmali, Phule utkarsha, Parbhani kranti , Arka anamica, HYBRID: Phule kirti	
Fertilizer kg/ha	50 : 50 : 50 NPK during sowing. 50 N after one month of sowing	
Seed rate kg/ha/Spacing	8 to 10 kg for *Kharif* with spacing of 60 x 30 cm, 10 to 15 kg for Summer with spacing of 45 x 30 cm	
Isolation Distance (Meter)	Foundation = 400	Certified = 200
Field inspection	Three Field inspections. First before flowering. On basis of foliage characters, off types are to be removed.	
Rouging	Volunteer plants, Wild okra plants, Other varieties plant, Other crop plants, Disease affected plants, especially Yellow Vein Mosaic Virus affected plants, Insect pest affected plants are to rouged out from seed production field. The off type plants are easily distinguishable on the basis of plant height, stem characters, pigmentation, flower shape and size.	
Objectionable weeds	Wild okra	
Objectionable disease	Yellow vein mosaic	
Off types %	Foundation- 0.10	Certified - 0.20
Germination	65%	
	08%	
Harvesting and Threshing, Yield	The dried pods are harvested. The varieties with angular pods which open along suture should be harvested promptly to avoid shattering. The pods are generally picked by hand. Pods are threshed and seeds are separated. The 10 quintal seed/ha can be achieved.	
Purity %	99%	
Innert matter	01%	

EXERCISE NO. 10

SEED PRODUCTION IN VEGETABLES CROPS : ONION

FOUNDATION AND CERTIFIED SEED PRODUCTION OFONION (VARIETIES AND HYBRIDS).

- The onion is highly cross pollinated crop.
- The CGMS male sterility is exploited in onion for hybrid seed production.
- In onion seed production, Bulb to seed is most commonly accepted method.
- Mostly Bulb to seed method is used for seed production, because it permits selection of true to type and healthy bulb selection & seed yields are comparatively very high.
- Onion is a biennial crop in seed production and requires two seasons to produce seeds.
- In onion the pungency is due to volatile oil known as allyl - propyl –disulphate

Seed to seed method	Bulb to seed method
This is also called as in - situ method.	In first season the bulbs are harvested, lifted, stored and replanted in next season.
In first season the bulbs are kept in the field in the winter so that next season the seed can be produced.	This method can be practiced as annual, biennial method.
Advantage of this method is cost is reduced arid no problem of storage of bulb.	In annual method seeds are sown in June - July and transplanted in Aug-September.
The disadvantage is that the seed yield is low & pure seed production is not possible since there is no opportunity of selection of standard bulb size.	The bulbs are ready by Oct - Nov
	The bulbs are harvested, cured. Here the bulbs are sorted for genetic purity and replanted in another field by end of Nov
This method is followed in *kharif* and	Bolting takes place in February and seeds

poor keeping quality varieties.	are ready for harvest by May.
	In biannual method nursery is sown in Oct - Nov and transplanted in Dec - Jan.
	The bulbs are lifted, sorted and True to Type bulbs are stored upto Sept - Oct and replanted in another field.
	As this method require 1.5 year, ii is known as biannual method.
	The advantage is that seed yield are more with quality seeds.
	The disadvantage is that more time is involved and becomes costly affair.

Foundation and Certified Seed Production of Onion		
	STAGE - I	**STAGE - II**
Land requirement	The selected land should not have previous crop of onion. The soil should have rich in organic matter with good water holding capacity. Tho soil should not be low lying, marshy and heavy clay and should be well leveled.	
Varieties	Red coloured : N - 53, Baswant-780, Pusa red, Agri Found Light Red (AFLR), Agri Found Dark Red (AFDR), White coloured : Phule safed, N - 2 - 4 - 1 Yellow coloured : Phule Suvarna	
Seed rate kg/ha, Spacing	8 to 10 kg on raised beds of 7.5 mt long, 1 .2 mt wide and 10 cm height. 30x20cm.	15 Quintal bulbs of 3 - 4 cm diameter. 45 x 30 cm.
Source of seed	Foundation = Breeder seed Certified = Foundation seed	Foundation = Breeder Certified = Foundation
Sowing time & Sowing method	In plains, October / November. Transplanting of 8 to 10 week old seedlings. Relatively high temperature and long photoperiod is essential for bulb formation.	
Fertilizers kg/ha	80 : 60 : 80 NPK, Half N and full P, K at sowing,	

	Remaining half 'N' in two split doses as top dressing 30 - 40 and 70 days after transplanting/ sowing.	
Isolation Distance (Meter)	Foundation = 05 meter. Certified = 05 meter.	Foundation = 1000 meter. Certified = 500 meter.
Field inspection	Minimum Four inspections. First Early stage of growth, Second, Digging out of bulbs, Third, Transplanting of bulbs and Fourth during flowering.	
Roughing	Depending upon the foliage colour, plant type or late maturing off types are to be removed. After harvesting of bulbs, the bulbs should be carefully sorted out. Such off types are thick neck, doubles, bottlenecks which do not confirm to varietal characters.	
	HARVESTING OF BULBS :	
Harvesting, Curing and storage of bulbs	• The maturity is indicated by dropping of the tops just above the bulb.	
	• Irrigation should be stopped and harvesting of bulbs is done.	
	• After curing 3-4 weeks, the tops above neck are cut leaving 2.5 cm portion so that neck is not exposed.	
	• At this stage remove injured and rotten bulbs including pre-mature bulbs.	
	• The selected good bulbs are stored at proper condition as furnished,	
	1) The bulb should be v/ell matured, dried and cured before storage.	
	2) The storage should be well ventilated.	
	3) The storage should be done in shallow trays with perforated bottoms.	
	4) Storage temperature should range 0 to 4.50 C until 3 to 4 weeks prior to replanting.	
	5) Malic hydrazide solution (3 gram in one liter water) should be sprayed 15 days before harvest for effective storage of bulbs.	

	HARVESTING OF SEED :	
	• The seeds are ready to harvest when first head (umbel) gets blackened	
	• 2 to 3 pickings are necessary to harvest all heads at just right stage.	
	• When the seed become black then seed heads are cut or snapped off, keeping a small portion of stalk attached.	
	• Seeds heads after harvest should be thoroughly dried.	
	• Air circulation is important while drying the seed heads.	
	• Hence the trays or analyses should be fitted only to a depth of 15 cm.	
	• The heads are threshed when seed separates easily from them.	
	Much of the seeds falls from capsule during drying.	
	* For proper cleaning of seed, the seeds are dipped in water for not more than 3—5 minutes and then they are dried under sun or through artificial means.	
	The seed should be dried 6 - 8% moisture for safe storage	
Objectionable disease %	----------	
Off types %	Foundation = 0.10	Foundation = 0.01
	Certified =.0.20	Certified = 0.05
Germination %	Foundation = 70 %	Foundation = 70 %
	Certified = 70%	Certified - 70 %
Seed moisture?	----------	08 %
Yield	15 -20 Quintals bulbs.	10 Quintals of seed.
Purity %	98 %	
Innert matter	02%	

EXERCISE NO. 11 & 12

SEED PRODUCTION IN PUMPKIN, BOTTLE / BRITTLE/ SPONGE / RIDGE GOURD

Bottle gourd [*Lagenaria siceraria*], Ridge gourd [*Luffa acutangula*], Cucurbits

Foundation and Certified Seed Production of Varieties [Bottle gourd & Ridge gourd, Cucurbits]		
	Foundation	**Certified**
Land requirement	There are no requirements as to previous crop, however, the land should be free from volunteer plant. The land selected	
	For seed production, the soil should be well drained and aerated.	
Sex expression and Sex ratio	1) Gourds are cross pollinated crops. 2) Sex expression and Sex ratio is a typical varietal character. 3) They are however modified by environment. 4) Lower soil fertility, Higher temperature, Longer light period all these conditions induces maleness. 5) Certain gases and chemicals also affect the sex. 6) Auxins and anti-auxins, both at proper concentration modify the sex. 7) Giberllic acid at higher concentration reduces maleness, but at lower concentration of 10 - 25 ppm increases female flowers. 8) Malic hydrazide, Ethrel and Boron & Molybdenum spray suppress the male flower and increase in female flower & fruit set.	
Varieties 1) Bottle gourd	Samrat, Pusa meghdoot. Pusa sandesh, Arka bahar, Punjab komal, Pusa manjari	
1) Ridge gourd	Phule Sucheta, Konkan Harita, PusaNasdar, Pusa Sadabaha.IIHR – 8.	
Source of seed	Breeder seed	Foundation seed

Seed rate kg/ha	For Kharif 3—4 Kg/ha seed having 90 % germination. For Summer 4.5 - 5.5 Kg/ha seed having 90 % germination.	
Isolation Distance (Meter)	1000	500
Field inspection	Three. First before flowering, Second at the time of flowering & Third at Fruit ripening.	
Rouging	The off types plants are removed at least thrice during crop growth. Firstly before flowering on the basis of plant characters. Secondly, at the time of flowering on the basis of flower characters. Thirdly at the time of fruit maturity on the basis of fruit characters. At all the stages, disease & insect pest affected, virus affected plants are removed as soon as they are noticed. The weeds are removed as and when required by weeding.	
Staking	The crop should be staked for better yield and quality of fruits especially in rainy season. The crop can be staked with bamboos or coles. These are fixed at proper distance and ropes are tied, on the poles at 60 cm distance.	
Objectionable weeds %	None	
Objectionable disease %	None	
Off types %	None	
Harvesting, Extraction of seed	1) The seed crop is ready for harvest when the fruits have turned pale yellow or golden. 2) The seeds are obtained by cutting individual fruit in half, longitudinally and scrapping them out with knife. 3) At the time of extraction the seed will not separate completely from the pulp surrounding it. 4) Its adherence to this material is broken by any one of following methods. . a) Fermentation b) Mechanical means and c) Chemical extraction. 5) 110- 130 Kg/ha seed yield may be obtained.	
Purity. %	98	
Inert matter	02	
Germination %	60	
Seed moisture%	08	

EXERCISE NO. 13
SEED SAMPLING AND TESTING PROCEDURE

Definitions

1. Seed lot: It is the specified quantity of uniformly blended seed which is physically identifiable with known origin & history and designated by a proper number or mark is known as seed lot. **or** A seed lot is a specified quantity of seed physically identifiable in respect of which an international analysis certificate may be issued known as seed lot.

2. Sampling: It is the process of obtaining a seed sample suitable from the whole seed lot for different tests in which the same constituents are present as in the seed lot and in the same proportion is known as seed sampling.

The object of seed sampling is to obtain a sample of a size suitable for tests, in which the same constituents are present as in the seed lot and in the same proportion. The quantity of seed tested in the laboratory is minute compared with the size of the seed lot which it is intended to represent. To obtain uniform and accurate results in seed testing it is essential that the samples to be taken and prepared with care and in accordance with the methods prescribed in the ISTA rules. Every effort must be made to ensure that the sample sent to the seed testing laboratory exactly represents the composition of the seed lot. Likewise, in reducing the sample in the laboratory, every effort must be made to obtain a working sample that is representative of the sample submitted. Thus, the rules are not only the guide lines for seed testers, but as far as sampling is concerned, also for inspectors and other authorized persons who do the sampling in the ware houses.

Types of sample

Primary sample: A small quantity of seed taken from processed seed lot, with the help of sampling equipments is called as primary sample. For Taking primary sample, processed seed lot to be sample should be homogenous. For small containers, 100 kg weight is taken as basic unit & the small containers combined to form sampling unit not exceeding this weight. e.g 20 container of 5 kg each, 33 containers of 3 kg each or 100 containers of 1 kg each. For sampling process, each unit is regarded as 1 container.

Sampling intensity or minimum no. of samples to be taken from containers or bulks is as below.

Sampling intensity for seed in container		**Sampling intensity for seed in bulk**	
Number of containers	Minimum of primary samples	Seed lots size (Kg)	Minimum of primary samples
Up to 5	5 (At least each container)	Up to 50 kg	3
6 to 30	Minimum 5 (At least 1 from 3 containers)	51-500 kg	5
31 to 400	Minimum 10 (At least 1 from 5 containers)	501-3000 kg	Minimum 5 (At least 1 from the each 300 kg lot)
More than 400	Minimum 80 (At least 1 from the every 7 containers)	3001 to 20000 kg	Minimum 10 (At least 1 from the each 500 kg lot)
-	-	20001 to 40000 kg	Minimum 40 (At least 1 from the each 700 kg lot)

All these primary samples are then mixed thoroughly to constitute composite sample.

Composite sample: The sample formed by combining and mixing all the primary samples is known as composite sample. The size of composite sample is 10 times more than required submitted sample.

Submitted sample: The sample submitted to the seed testing laboratory which comprises the composite sample reduced as necessary called submitted sample.. Before taking submitted sample the composite sample should be thoroughly mix and reduced up to prescribed weight with the help of dividers or repeated halving methods.

Working sample: A reduced sample taken from the submitted sample in the laboratory on which one of the quality tests is made is known as working sample..

PRINCIPLES AND PROCEDURE FOR SAMPLING THE LOT

Before sampling of a lot is carried out the sampler should be satisfied that the lot shows no evidence of heterogenity. In cases of doubt, heterogenity can be determined with the heterogenity test.

The size of the lot shall also not exceed certain limits. This means that in agricultural seeds the lot shall not exceed 10,000 kg, for the large seeded species 20,000 kg.

A third requirement for sampling is that the lot shall be in bags or other containers, sealed and labelled or marked for identification by a single lot designation. This is because an International seed lot certificate may not be issued in respect of loose seed.

At the time of sampling all containers must be labelled or marked to show a lot identification corresponding to the lot identification of the certificate. The containers shall be sealed or seen to be sealed by the sampler.

Practical No.1

Title : Drawing a primary samples and preparation of composite samples.

Equipment: Stick seed trier, Nobbe trier, bin sampler, sampling pans, bucket, pan balance, weight box.

A) Triers or probes

Study different types of seed triers.

1) Stick seed trier: It is the most commonly used sampling instrument. It consists of a hollow brass tube inside a closely fitting outer shell or sleeve which has solid pointed end. The tube and sleeve have been slots in their walls so that when the tube is turned until the slots in the tube and sleeve are in line, seeds can flow into the cavity of the tube, when the tube is given half turn the openings are closed. It may be used horizontally or vertically. However, when seedlot is in bulk, vertical insertion is more practicable.

2) Bin sampler: It is used for sampling the seedlot stored in bins. It is similar in construction to stick trier but it is much larger ranging upto 1600 mm in length and 38 mm in diameter with 6 to 9 slots.

3) Nobbe trier: It is a pointed tube long enough to reach the centre of the bag with an ova! hole near the pointed end. The total length of the trier is 500 mm with internal diameter of 14 mm. It is used for drawing samples from bags.

B) Sampling by hand : In addition to seed trier, sample can also drawn by hand when seedlot is of chaffy and non free flowing seeds as in cynadon, chloris, panicum etc

Procedure:

1. Use proper seed trier for drawing primary sample. In case of chaffy seeds, draw sample with hands.
2. Draw primary samples from different containers or different portions of seedlot in bulk selected at random. While drawing primary sample, representative portion from top, middle and bottom of container or bulk be included.

3. Check the trier for presence of previous seed if any and then close the trier.
4. Insert the trier in the container or in the bulked seed in closed condition.
5. Open the trier and turn it 2 to 3 times gently so as to allow it to fill it with seed completely.
6. Close the trier, remove it from the bag or bulked lot and empty its content in bucket or sampling pan.
7. Draw required number of primary samples according to the size of the seedlot.
8. Mix all primary samples together so as to form composite sample.

The sampling should be varied from top, middle and bottom of the bags. To sample the bottom of standing bags they may be raised off the floor placed on to pot other bags. In certain species especially chaffy seeds, sampling by hand is satisfactory method. It is difficult this method to sample deeper than 400 mm. It is impossible to obtain sample from the lower layer in bags and bins. In such cases the sampler may take special precautions such as requesting that same bags to be emptied or partly emptied to facilitate sampling and then be refilled. When sampling is done by hand great care should be taken to keep the fingers tightly closed above the seeds so none may escape.

For seed lot in bags that are uniform in size the following **sampling intensity** shall be regarded as the minimum requirement.

Upto 5 Containers	-	Sample each container and always take at least 5 primary samples.
6 to 30 containers	-	Sample at least 1 in every 3 containers, but never less than 5
31 to 400 container	-	Sample at least 1 in every 5 containers, but never less than 10.
More than 400 containers	-	Sample at least 1 in every 7 containers, but not less than 80.

When sampling seed lot in bulk, in containers of different or very small sizes, the following sampling intensity is the minimum requirement.

a) Upto 500 Kg	:	At least 5 individual samples
b) 501 to 3000 Kg	:	One individual sample for each 500 kg but not less than 5 sample
c) 3001 to 20,000 kg	:	One individual sample for each 500 kg but not less than 10 samples.

d) More than 20,000 kg : One individual sample for each 700 Kg but not less than 10 samples.

If the primary samples appear uniform they shall be combined and mixed to form the composite sample. From that the submitted sample is obtained by one of the laboratory methods referred to below, using larger equipment if necessary. If it is difficult to mix and reduce the sample properly under warehouse conditions the entire composite sample shall be forwarded to the seed testing laboratory for reduction. If the composite sample is of appropriate size it may be regarded as the submitted sample without reduction.

Practical No.2

Title: Preparation of submitted sample

Material: Composite sample, piece of cloth or paper, sheets, cloth bags, lables polythene bags.

Procedure:

1) Take a piece of cloth or sheet of paper and spread it on the floor.
2) Mix thoroughly the composite sample and weigh it. Pour it on cloth or paper.
3) Spread the sample uniformly on the paper or cloth.
4) Divide the sample in 4 parts by passing the palm diagonally (in x manner) through the sample,
5) Take two opposite portions of the sample and mix them weigh them. Discard other two portions of the sample
6) Repeat steps from Sr.No.3 to 5 3-4 times till required weight of sample is obtained.
7) Take a clean cloth bag and fill it with submitted sample prepared.
8) Fill in the sample coupon in the Performa given below.
9) Insert the sample coupons in the bag, close the bag by proper stitching and seal it.
10) Send the sample to the seed testing laboratory
11) If seed moisture is to be drawn, draw additional quantity of seed and put it in separate polythene bag with label and seal it. Send it with main sample.

Dispatch of submitted sample:

Sample need to be dispatched to STL as early as possible along with detail information such as name of person who draw the sample from lot, name of crop, variety, stage, code no, lot size, date of sampling and types of tests required. Apart from this, two reference sample are prepared. One reference sample is to be stored by SCA office and the third sample handed over the concerned seed producer. Office sample of seed lot passed in seed testing is stored for two years.

Preparation of working sample in the laboratory

As the size of the submitted sample is larger than the actual quantity of seed required for carrying different tests. It is necessary to reduce the submitted sample, to working sample required for carrying different tests. Part of the submitted sample is required to be stored for retesting if required. The working sample weights for purity analysis are calculated to contain at least 2500 seeds but subject to maximum 1000 gm.

PRINCIPLES AND PROCEDURE FOR SAMPLING IN THE LABORATORY

The submitted sample received in seed testing lab is registered and designated by a code number. Submitted sample is tested for determination of seeds of other crop, weed, objectionable weeds, objectionable diseases and other distinguishing varieties by number. Three working samples of the submitted sample, which passes the seed certification standard by number are prepared.

Several methods and apparatus are available to reduce the submitted sample to the size of the working sample. Minimum weights of submitted and working sample are as fixed in ISTA rules.

Standard for seed lot and samples size required for different crops

Crop	Maximum lot size (kg)	Submitted Sample (gm)	Working Sample (gm)	Working Sample Size for ODV (gm)
CEREALS				
Sorghum	10,000	900	90	900
Pearl Millet	10,000	150	15	150
Wheat	20,000	1,000	120	1,000
Rice	20,000	400	40	400
Maize	40,000	1000	900	1000
Finger Millet	10,000	60	6	60

OIL SEEDS				
Groundnut Pods	20,000	1,000	1,000	1,000
Groundnut Kernals	20,000	1,000	600	1,000
Sunflower Variety	20,000	1,000	250	1,000
Sunflower Hybrid	20,000	250	125	250
Linseed	10,000	150	15	150
Sesame	10,000	70	7	70
Soybean	20,000	1,000	500	1000
Rapseed	10,000	100	10	100
Mustard	10,000	160	16	160
Castard	20,000	1,000	500	1,000
PULSES				
Pigeon Pea	20,000	1,000	300	1,000
Chickpea	20,000	1,000	1,000	1,000
Greengram	20,000	1,000	120	1,000
Blackgram	20,000	1,000	150	1,000
Horsegram	20,000	800	80	800
Lablab bean	20,000	1,000	500	1,000
Cowpea	20,000	1,000	400	1,000
Pea	20,000	1,000	900	1,000
Lentil	10,000	600	60	600
Vegetables				
Tomato Variety	10,000	70	7	7
Tomato Hybrid	10,000	7	7	7
Onion	10,000	80	8	80
Brinjal	10,000	150	15	150
Chilli	10,000	150	15	150
Okra	20,000	1,000	140	1000
Coriander	10,000	400	40	400
Bottle gourd	20,000	700	70	700
Ridge gourd	20,000	1,000	400	1,000
Radish	10,000	300	30	300

Preparation of working sample:

1. Mechanical divider method

The sample is mechanically divided by the seed divider. Seed dividers are of following types.

a) The conical divider (Boerner type divider)
b) The soil divider (multiple slot divider)
c) Centrifugal divider (Gamet divider)

Mechanical Divider Method : In this method, the seed sample is divided mechanically with the help of the seed divider. When a seed sample is passed through the divider. It is divided into two approximately equal parts. This process is repeated

3 to 4 times, each time removing half portion till working sample of desired weight is obtained. This method is suitable for all kinds of seeds except for extremely chaffy seeds. There are three types of mechanical seed dividers in use.

a) Conical divider (Boerner)

b) Garnet seed divider (centrifugal)

c) Soil type divider

a) Conical (Boerner type) divider: It consists of a hopper cone and series of baffles directing the seed into two spouts. The baffles form a alternate channels and spaces. The channels are united to one spout and spaces to other spout. It is available in two sizes i.e. small for small seeds and large for large seeds.

b) Gamet seed divider: Its working is based on the use of centrifugal force. It consists of a hopper, shallow rubber cup called spinner rotated by an electric motor, baffles and two spouts. When the seed is put into the hopper and electric moter started, the centrifugal force, first mixes the seed and scatters the seed over the dividing surface. Then the seed flows downward on to the rotating spinner which throws them out by centrifugal force. The seeds fall on stationary baffles which separates them in two equal parts. The seeds pass out through the spouts.

c) Soil type divider: Working of this divider is based on the same principles as the conical divider. It consists of a hopper, straight rows of channels, a frame to hold the hopper, two receiving pans and a pouring pan.

Practical No.3

Title: Preparation of working sample with conical divider.

Material: Submitted sample, balance, weight box, conical divider.

Procedure:

1) Check the conical seed divider and its parts for presence of seed of previous sample.
2) Close the passage with guiding valve.
3) Put the dividing pan below each spot.
4) Pour submitted sample into the hopper and open the guiding valve. Seed will be collected into two receiving pans.
5) Mix seed from both receiving pans
6) Put empty receiving pans below the spouts. Close the guiding valve and pour mixed seed sample into the hopper.
7) Open the sliding valve. Collect seed from one of the receiving pans for further reduction. Discard seed from other pan.
8) Repeat the procedure given under point No.6 and 7 3 to 4 times till working sample of desired weight is obtained.

Weigh the working sample. Record the observations.

1. Name of crop seed
2. Weight of submitted sample
3. Weight of working sample obtained.

Practical No.4

Title: Preparation of working sample with garnet divider

Material: Garnet divider, balance, weight box, submitted sample

Procedure:

1. Adjust the level of garnet divider with the help of adjustable feet.
2. Check the divider and its containers for cleanliness.
3. Put receiving pans under both spouts.
4. Weigh the submitted sample and pour it into the hopper. Switch on the divider. See that seed is collected into two pans.
5. Mix the seed of both pans together.
6. Reduce the seed sample by repeating the procedure, given under steps 3 and 4, for 3 to 4 times and discarding seed from one pan each time.
7. Weigh the working sample and record observations.

Observations:

1. Name of the seed sample
2. Weight of submitted sample taken
3. Weight of working sample prepared

Practical No.5

Title: Preparation of working sample with soil type divider

Material: Soil type divider, submitted sample, balance, weight box.

Procedure:

1. Weigh the submitted sample
2. Check the soil type divider and its pans for previous seed and dirt. Put the receiving pans one under each chamber.
3. Empty the content of submitted sample in pouring pan and pass the sample into the hopper and see that seed is collected in two receiving pans.
4. Mix the seed of both pans. Put the receiving pans under each chamber.
5. Pour the seed sample into the hopper with the help of pouring pan.
6. Take fraction of seed sample from one pan for further reduction and discard seed fraction from other pan.
7. Repeat procedure given under steps 5 to 6 3 to 4 times. Weigh sample and record

Observations:

1. Name of seed sample
2. Weight of submitted sample
3. Weight of working sample

2. Modified halving method

The apparatus comprises a tray into which fits a grid of equal sized cubical, open at the top and every alternate one is halving no bottom. The seed is poured evenly over this grid. When the grid is lifted approximately half the sample remains on the tray. This can be repeated until a working sample of approximately but not less than the required size is obtained.

Practical No. 6

Title: Preparation of working sample by modified halving method.

Material: Tray with grid of cubical cells, submitted sample, balance, weight box, sample pan.

Procedure

1. Mix the submitted sample thoroughly and weigh it.
2. Take a tray and place on it a grid of uniform sized cubicles alternately closed bottom.
3. Pour the seed with the help of piece of paper uniformly even the surface of the tray.
4. Lift the grid and take the seed fallen in the tray for further reduction.
5. Repeat procedure given under steps at Sr.No.3 to 4 till working sample of desired weight is obtained.
6. Weigh the working sample and record observations.

Observations:

1. Name of the seed sample
2. Weight of the submitted sample taken
3. Weight of the working sample obtained

3. Random cups method

2) Random cup method :

This method is more suitable for seeds requiring a working sample upto 10 gms provided that

i) Seeds are not of extremely chaffy structure and

ii) Seeds do not bounce or roll (e.g. brassica spp). In this method, 6 to 8 small cups are placed at random on a tray. After, preliminary mixing the seed is poured uniformly over the tray. The seed that fails into the cups is taken as working sample.

Practical No.7

Title: Preparation of working sample by random cup method.

Material: Submitted sample, 6 to 8 small plastic cups, tray balance, weight box, sampling pan.

Procedure:

1) Mix the submitted sample thoroughly
2) Weigh the submitted sample
3) Take a tray and place cups on it by keeping uniform distance between them.
4) Pour seed with the help of piece of paper uniformly over the surface of tray.
5) Collect the seed fallen in the cups, mix it and weigh. If the weight of seed is more or less closer to the desired weight of working sample, take it as working sample. Otherwise repeat the procedure given under steps No.4 and 5 till working sample of required weight is obtained.
6) Record the observations.
   a) Name of Seed sample
   b) Weight of submitted sample taken
   c) Weight of working sample obtained

4. Spoon method

This method is only permitted for small seeded species. After preliminary mixing the seed is poured evenly over the tray in the same way as it is poured in the random cup method. Do not shake the tray thereafter. With the spoon in one hand and the spatula in the other and using both small portions of seed are removed from not less than 5 random places.

Practical No.8

Title : Preparation of working sample by spoon method.

Material: Tray, spatula, spoon, balance, weight box, submitted sample taken.

Procedure:

1) Mix the submitted sample well and weigh it.
2) Take an empty tray and place it on the table.
3) Pour submitted sample over the tray uniformly and gently. Do not disturb the tray
4) Take spoon in right hand and hold spatula in left hand and draw a small portion of sample from one part. Draw minimum five samples from randomly selected pots.
5) Mix the portions and weigh the sample.
6) Repeat this till working sample of desired weight is obtained.
7) Weigh the working sample and record observations.

Observations

1. Name of the seed material
2. Weight of submitted sample taken
3. Weight of working sample obtained.

Exercise -

Arrange different equipments of seed sampling for identification and draw the diagram of equipments.

******

EXERCISE NO. 14

PHYSICAL PURITY TEST

The purity test is done with the objectives

1. To determine the composition of sample by dividing each sample into 4 components namely pure seeds, other crop seed, weed and inert matter and to judge the quality of seed sample on the basis of proportion of pure seed and other components as per prescribed norms of SCA.
2. To identify objectionable weed seeds and other crop seeds found in sample and to give them botanical names.
3. To determine eligibility of seed sample for seed certification.
4. To get the pure seed for further seed tests like germination.

Materials

Seed blower, purity work board, forceps, magnifying lens, spatula, dishes, sieves, needles and balance etc.

Procedure

1. The working sample of desired weight is prepared.
2. Use seed blower, if seed sample is chaffy or grass species after adjusting air flow.
3. Place the working sample on a board or glass plate and with the help of forceps, needles and magnifiers, separate out the seed sample into following components.

i) Pure seed

ii) Other crop seed,

iii) Inert matter

iv) Weed seed

i) **Pure seed:** Pure seed refers to the seed of species which is stated by sender or found to be dominant in the seed lot. Such seeds are immature, undersized, shriveled, achenes or similar fruits, diseased seeds, germinated seeds, intact seed unit or diseased seed unless transformed into fungal sclerotia, smut balls or nematode galls be regarded as pure seed provided they can be identified as that species, of pure seed.

Note: Piece of seed unit longer than half of original size should be considered as pure seed (provided it can be authentically be identified as of that crop).

ii) **Other crop seeds:** It includes seed and seed like structure of any plant species other than that of pure seed. The distinguishable characteristics set out for pure seed should be applicable to other crop seed except certain weed species which are classified separately.

iii) **Inert matter:** It includes seed and seed like matters; mainly pieces of broken or damage seeds, achenes and caryopsis, empty glumes, other matter mainly soil, sand, stone, chaff, stems, leaves, pieces of bark, flowers, fungi bodies etc.

iv) **Weed seed:** The seeds, bulblets or tubers of plants recognized as weeds by official regulations(objectionable weeds) or by general usage(common weeds).

4. After complete separation of components of sample, retain the pure seed on purity work board for rechecking. After re-checking the pure seed separate other seeds and inert matter.
5. Weigh the each of the three components.

Wt. of working sample (g)	The number of places of decimals upto which each component needs to be weighed.
Less than 1 gm	4
1 to 9.999 gms (but less than 10 gms)	3
10 to 99.99 gms (but less than 100gms)	2
100 gms to 999.9 gms	1
Greater than 1000 gms	0

(Note : After weighing each component, they should be properly marked & retained for future reference. Only pure seed component (minimum 400 number of seeds should be used for germination test.)

6. Calculate the percentage of each component on the basis of the sum of weights of the components and not on the basis of the original working sample. The sum total of percent of all components should be 100.

Calculate the percentage value of each component on the basis of total of sum of weights of all components and not on the basis of the original sample.

$$\text{i) Pure seed (\%)} = \frac{\text{Wt. of pure seed}}{\text{Total wt. of all seed components}} \times 100$$

$$\text{ii) Inert matter (\%)} = \frac{\text{Wt. of inert matter}}{\text{Total wt. of all seed components}} \times 100$$

$$\text{iii) Other crop seed (\%)} = \frac{\text{Wt. of other crop seed}}{\text{Total wt. of all seed components}} \times 100$$

$$\text{iv) Weed seed (\%)} = \frac{\text{Wt. of weed seed}}{\text{Total wt. of all}} \times 100$$

7. If percentage of seed of any other crop species or weeds together is more than 0.1 per cent or if the number of seeds is more than 20, separate out all seeds of that species from working sample as well as submitted sample.
8. Reporting results :

a) Results of purity analysis is to be given in one decimal place.
b) The total of percentage of all components must be 100.
c) If percentage components are less than 0.05 per cent, then it is to be reported as trace.
d) The percentage of each components is shown in the analysis sheet at proper space.
e) If the results are nil, it is to be shown as 0.00 per cent.
f) Latin names of pure, weed and other seeds must be reported.

Errors in Purity Analysis

1. **Moisture:** Variation in weight due to moisture may occur, while sample is being analyzed or even it is left on desk for some time, which affects pure seed than the inert matter. Hence, the analysis should be completed without loss of time.
2. **Calculation error :** It is commonly overlooked. To avoid it, care should be taken in regard to weighing of purity fraction to the requisite decimal places accurately and later in calculating the percentage of various components.

Laboratory work : Find out various components of working sample given to you and calculate the percentage of each and give your opinion about the sample.

EXERCISE NO. 15
SEED MOISTURE TEST

Moisture content of seed is one of the important factors affecting viability and quality of seed. It is loss in weight when the seed is dried or the quantity of water collected when it is distilled. It is expressed as a percentage of the weight of the original sample.

Methods of Moisture Determination

The basic methods are-

1. **Drying without heat**

Samples are dried without heat or moderate heat in vaccum using phosphorus pentoxide (P_2O_5) as desiccant.

2. **Lyophilization**

(Freeze dried)- Biological materials are frozen and water removed by sublimation in vaccum.

3. **Reversibility method**

a) **Red drying**

This method determines drying time and temperature so that loss of weight by decomposition is accounted for.

b) **Karl Fisher Titration Method**

In this method water is extracted from finely ground seed with methyl alcohol and then determined by titration by a special reagent. This is most accurate method.

However, these methods require much time, equipments and high skills of operation and hence not practically used.

4. **Hot Air Oven Method**

Method is most practical and commonly used for moisture determination.

Objective: To determine moisture content of a given sample.

Material

Grinding mill, hot air oven, chemical balance, crucible with lid, dessicator, spoon, trays and seed sample.

Procedure

1. Take 4 to 5 gms of duplicate working sample for determination of moisture from submitted sample accurately.
2. Crops of larger seed size (e.g. cotton, maize, sorghum, paddy, wheat, etc) are ground with grinding mill in such a way that at least 50 per cent of the ground material should pass through a wire sieve of 0.5 mm meshes and not more than

10 per cent remain on a wire sieve. For leguminous crop seeds (e.g. pea, soybean, chickpea etc) coarse grinding is necessary i.e. 50 per cent ground material should pass through sieve with 4 mm meshes.

3. If moisture content of seed is more than 17 per cent (Rice-13% soybean-10%) pre-drying is obligatory. Similarly, very moist seed of maize (above 25%) and others, samples should be dried at 70°C for 2 to 5 hours depending on initial water content.
4. Weigh the clean and dry crucible with lid accurately.
5. Put the ground seed sample prepared earlier (4-5 gm) in a crucible with help of spoon and again take the weight of crucible with lid very accurately.
6. Place the crucible rapidly in hot air oven as under
   i) In low constant temperature oven method, keep the container at temperature 103°C ± 2°C and dry for 17+1 hours (e.g. onion, chillies, soybean, radish and brinjal etc),
   ii) In high constant temperature oven method, keep the material at 130°C ± 2°C for 2 ± 1 hrs.

Table. Oven dry method recommended for different crops

Crop	Oven dry method
Rice, wheat, pearly millet, maize, sorghum, chickpea, lathyrus, pea, pigeonpea	High constant temperature (130 ± 2°C)
Groundnut, rape seed and mustard, soybean, sesame, linseed, castor and cotton	Low constant temperature (103 ± 2°C)

7. Remove the crucible with lid and cool in dessicator
8. Weigh the crucible with lid and contents.
9. Calculate the percentage of moisture content in seed sample by using formula-

$$\text{Moisture\%} = \frac{M_2 - M_3}{M_2 - M_1} \times 100$$

$$= \frac{90 - 85}{90 - 40} \times 100$$

$$= \frac{5}{50} \times 100 = 10\%$$

Where,

M_1 = Weight of empty crucible with lid
M_2 = Weight of crucible with seed sample
M_3 = Weight of cructible with seed sample and lid after drying
M_2-M_1 = Weight of sample
M_2-M_3 = Loss in weight after drying

Laboratory work-

Determine the moisture percentage of sample given to you.

****

EXERCISE NO. 16

GERMINATION TEST: TYPES OF GERMINATION

Germination is the awaking of the dormant embryo. In mature angiospermic seeds, embryo lies in the dormant stage. When physiological activities are ceased. As soon as favourable conditions are available dormancy is broken and germinating begins, thus it is resumption of active growth of the embryo after a period of dormancy.

Changes during germination:

1. Swelling of seed due to imbibition i.e. water is absorbed through cell wall by diffusion and osmosis.
2. Bursting of seed coat due to swelling.
3. Dilution of stored food material within seed.
4. Initiation and activation of physiological activities such as respiration & secretion of enzymes.
5. Digestion of complex insoluble food reserves to soluble forms by enzymatic activities.
6. Assimilation of these soluble food material at meristemic area to provide energy for cellular activity & growth.
7. Emergence of radicle and plumule through seed coat.
8. Growth of seedling by process of cell division, enlargement & differentiation at growing point.

When seed placed in soil gets favourable conditions, radicle grow vigorously and comes out through micropyle and fixes seed in the soil. Then either hypocotyl or epicotyl begins to grow.

Essential structure of seedling.

1. The root:

The first root of germinating seedling mostly in dicotyledonous is primary root. It is commonly white, slender & elongates rapidly. Later on, numerous root hairs are usually produced to this primary root. At later stage, secondary roots are produced as either lateral from the primary root itself or as adventitious roots emerging from other parts (i.e. hypocotyls) of seedling. In monocotyledons, the primary root does not survive long and replaced by secondary roots or seminal roots e.g. gramineae and does not produce lateral roots. Main function of root systems are to anchor the plant in soil, to absorb water and dissolved salts and to conduct these to cotyledons and the shoot.

2. Cotyledon:

Cotyledons from the part of embryo within seed. They act as photosynthetic organs in epigeal germination. Mostly they provide nutrients (which were either stored or photosynthesized by them) to seedling. In monocotyledons, it is divided into two isolated parts with different functions such as the shield shaped scutellum for the absorption of stored food and the sheath like coleoptile which protects the shoot apex (while emerging through soil) and coleorhiza which protects root apex (while growing into soil)

3. Hypocotyl:

The part of seedling axis immediately above the primary root & up to the point of attachment of cotyledons is called hypocotyl. In epigeal germination, the hypocotyl elongates & bring cotyledons above the soil.

4. Epicotyl:

The part of seedling axis between point of attachment of cotyledons and that of first foliage leaf (or pair of leaves) is known as epicotyl. In hypogeal germination, it elongates & brings the shoot with 1st foliage leaves into the light above the soil surface.

5. Shoot apex:

The upper end of seedling axis is called the shoot apex. This main shoot growing point consists apical meristem and leaf initials. The developing leaves envelop it and from terminal bud.

Types of Germination:

1. Hypogeal germination:

When cotyledons remain **below soil surface** due to rapid elongation of epicotyl (portion of embryo above cotyledons) then it is termed as hypogeal germination. It occurs with the majority of monocotyledons (e.g. gramineae/ poaceae), some large seeded legumes (e.g. Pea, bean, gram) and some trees like mango, jack fruit, coconut & arecanut.

2. Epigeal germination:

When cotyledons pushed **above soil surface** due to rapid elongation of hypocotyls (portion of embryo below cotyledons), then it is termed as epigeal germination. It is observed in horticultural & woody plant species e.g. Cotton, cucumber, castor, sunflower, groundnut, guar, gourds, tamarind & French bean.

3. Vivipary:

Germination of seed inside the fruit attached to the mother plant (which also nourishes the seedling at initial stages just after germination) is known as 'Vivipary' and it is observed in many plants which grows along sea coasts e.g. Mangrooves and also in agaves (e.g. bubils). When radicle of such seedling elongates, swells in lower part and gets stouter, it separates from parent plant also due to increase in weight and falls on ground vertically in such a way that the radicle goes into soft mud and the plumule remain above the soil level.

Pre-harvest sprouting:

Sprouting of seed due to high moisture on the matured plants standing on the field is known as pre harvest sprouting and it is different than vivipary. e.g. Groundnut, Bajra.

Hypo - epigeal germination:

A dicot species leaves one cotyledon beneath the soil as hypogeal germination while the other cotyledon comes out above soil as epigeal germination, e.g. Paperomia peruviana.

Factors affecting germination:

Following factors are essential for normal germination of seed.

1. Water (Moisture): It enables the resumption of physiological activities, swelling of seed, due to absorption of moisture and causes bursting of seed coat and softening the tissue due to which embryo awake and resumes its growth.

2. Temperature: A suitable temperature is necessary for proper germination. Germination of seed does not take place beyond certain minimum and maximum temperature i.e. 0 °C and above 50°C. optimum temperature range for satisfactory germination of seed is 25 to 30°C.

3.Oxygen: It is essential during germination for respiration and other physiological activities which are vigorous during the processes.

4. Light: It is not considered as essential for germination and it takes place without light. The seedling grow more vigorously during darkness rather in light. However, for survival of germinating seedling, light is quite essential. Germination of Nicotiana tabacum, sorghum helapense, cynadon dactylon and chloris gayana need light but it is essential for lettuce, while jowar, bajra, pea and bean are neutral in the requirement of jowar for germination.

5. Substratum -Substratum is the medium used for germinating seeds in the laboratory. It may be absorbant paper (blotting paper, towel paper, tissue paper) soil and sand, Substratum should be free from toxic substances. It should not act as a medium for growth of micro organism.

B) Internal:

1. Food and auxin: Embryo feeds on the stored food material until young seedling prepare its own food. Auxins are the growth promoters hence quite essential during the germination.

2.Viability: All seeds remain viable for certain definite period of time and thereafter embryo becomes dead. It depends on maturity of seed, storage conditions vigour and presents and type of species. Generally, it is for 3 to 5 years and they remain for more than 200 years also as in lotus.

3. Dormancy: it is failure of mature viable seed to germinate under favourable conditions of moisture. Many seeds do not germinate immediately after their harvest, they require rest period for certain physiological activities.

Germination in laboratory test is the emergence and development from the seed embryo of those essential structures which, for the kind of seed being tested, indicate ability to develop into a normal plant under favorable conditions in soil.

The primary objective of this test is to gain information with respect to field planting value of the seed and provide results which can be used to compare the value of different seed lots.

Materials:

1) Working seed sample (400 pure seeds separated during physical purity test).
2) Seed counter.
3) Germination papers such as filter / towel / blotting / crape kraft papers Or sand / soil to be used as substratum for germination as per methods.
4) Germinators with thermometers: For germination of seeds under controlled (temperature, light, relative humidity and oxygen) conditions within specified period.
5) Other materials such as petriplates with lids & cotton wool, germination boxes with sand / silica, wax paper & rubber bands as per germination methods.
6) Red pencil, forceps, magnifying lens.

******

EXERCISE NO. 17

GERMINATION TEST – DIFFFERENT METHODS OF GERMINATION

Methods of Germination testing

At least four hundred seeds should be tested for germination. Seed selected for germination should be from 'pure seed' component separated in purity analysis and should be counted without discrimination as to size or appearance, by hand, counting boards or by vacuum seed counter.

1. Top of paper (T.P.)

In this method seeds are germinated on top of one or more layers of paper which are placed either in enclosed transparent Petri dishes or boxes and are kept in an incubator or germinator. Moistened porous paper or absorbent cotton can be used as base for paper or even as an immediate substratum.

2. Between paper method (B.P.)

The seeds are germinated between two layers of germination paper which are placed directly on germination trays in cabinet or room type germinator or in metal, plastic or glass boxes. In former method, relative humidity in the cabinet, or room should be maintained to the saturation. The paper can be folded or rolled and placed in an upright position. Metal, glass or plastic frames can be inserted between papers to ensure ventilation. Moistened porous paper or absorbent cotton can be used as base for the paper or even immediate substratum. However, paper should not be too wet to form water film if pressed with finger.

3. Germination in sand

Seeds are either planted in uniform layer of moist sand and then covered with loose sand 1 to 3 cms. Deep or seeds are pressed into the surface of the sand. Amount of water is added e.g. cereals except maize may be germinated to 50 per cent or its water holding capacity while larger seeds legumes and other to 60 percent.

4. Germination in soil

Soil or an artificial compost is used instead of sand. This method is used to

conform the evaluation of seedlings, in doubtful cases and testing samples which produce seedlings with phototoxic symptoms when germinated on paper or sand. Soil should be kept wet.

Procedure for germination Test

I. Germination on towel paper

1. Take rectangular germination paper (crape craft paper) and soak it in water, remove excess water.
2. Put it on polythene paper slightly bigger than germination paper.
3. Place seeds of given sample on germination paper with the help of counting board in four replications of 100 seeds each.
4. Cover the seeds with another moist germination paper and roll along with polythene paper and tie both ends of roll by rubber bands.
5. Keep the count of seedlings on the prescribed day and report the percentage of normal, abnormal, dead, hard and fresh ungerminated seeds.

II Germination in Petri-dish

1. Take germination paper (blotting) and prepare round pieces as per inner diameter of dishes.
2. Place cotton wool at the bottom of dish and cover the piece of blotting paper, add water till paper becomes wet and remove excess water from the dish.
3. Put either 50 or 25 seeds in each dish on moist paper at proper distance.
4. Cover petri-dish with lid and put it in germinator/incubator maintained at appropriate constant temperature.
5. Take the germination count and calculate the germination percentage.

III. Germination in sand and soil

1. Take earthen or plastic pots filled with sand or soil
2. Add water to obtain sufficient moisture in soil/sand
3. Put the seeds of variety to be tested at appropriate depth with proper spacing.
4. Cover the seeds with soil or sand and give water if necessary and put them in germinator at appropriate constant temperature.

Observe that following from the germinated seeds and report the results.

1. Normal seedling

Seedling which shows the capacity for continued development into normal plants when grown in good quality soil and under favourable conditions of water supply, temperature and light. Following seedlings may be treated as normal seedlings.

a) Seedlings with well developed system of root with primary root intact hypocotyl epicotly and a normal plumule and cotyledons.

b) A well developed primary leaf within or emerging through the coleoptile in monocotyledons.

2. Abnormal seedlings

Which do not show the capacity for continued development into normal plants when grown in good quality soil under favorable conditions of water supply, temperature and light.

Following seedlings may be treated as abnormal.

a) Seedlings without cotyledons, constrictions, splits cracks and lessions.

b) Seedlings without primary root

c) Seedlings without damaged and stunted root and plumules, coleoptile without primary leaves.

d) Seedlings with decayed essential structure and discoloration.

3. Hard seed

The seeds belonging to leguminoseae and malavaceae family which remain hard at the end of prescribed period of test. Because they have not absorbed water due to impermeable seed coat are called hard seed.

4. Fresh ungerminated seeds

Seeds other than hard seeds which remain firm and viable even after appropriate treatment for breaking dormancy are classified as fresh ungerminated seeds.

5. Dead seeds

Seeds at the end of test period are neither hard nor fresh and have not produced seedlings, classified as dead seeds.

Table. **Minimum seed certification standard (%) for germination test**

Foundation and certified	Crop
	FIELD CROPS
65	Cotton (Linted)
70	Cotton (Delinted)
70	Soybean, sunflower, groundnut, castor, groundnut
75	Sorghum, pearl millet, minor millets, pigeonpea, moong, lathyrus lentil, fieldpea
80	Rice, sesame, linseed, safflower, niger, horsegram, jute, berseem, Lucerne
85	Wheat, barley, triticale, oat, chickpea, rape seed and mustard
90	Maize
	VEGETABLE CROPS
60	Cucurbits, capsicum, chilli, spinach, carrot, sugarbeet
65	Okra, cauliflower
70	Cluster bean, brinjal, radish, tomato, fenugreek, cabbage, onion
75	Indian bean, cowpea, French bean

Laboratory work-

Calculate the percentage of each sample given to you and give your conclusion about sample.

***

EXERCISE NO. 18

SEED VIABILITY TEST

SEED VIABILITY : The capacity of seed to show living properties like germination and growth ie. normal seedling under favourable environmental conditions is called as seed viability.

Methods of seed viability tests:

1) Biochemical Test ie Tetrazolium test 2) Embryo excision test (EET)

3) Accelated ageing test (AAT) 4) Other methods includes

1) Indigo Carmine Test (IC)

2) Radiographic method (X ray contrast)

3) Glutamic Acid Decarboxylase (GADA) test

4) Seed lechate conductivity Test (SLC)

5) Seed Crushing Test (SC)

1] TETRAZOLIUM TEST : Lakon (1942)

Object

Object of the biochemical test is to determine quickly the viability of seeds of certain species which germinates slowly by regular germination process. By reason the principle of evaluation and its indicator, the test is designated as "the topographical tetrazolium test".

Principle

In a biochemical test the reduction process which takes place in living cells are made visible by the reduction of an indicator. The indicator used in the tetrazolium test for seeds is a colourless solution of the tetrazolium salt which is imbibed by the seed. Within the seed tissues it interferes with the reduction process of living cell and accepts hydrogen from the dehydrogenses. By hydrogenation of the 2, 3, 5 triphenyl tetrazolium chloride, a red stable and non-diffusible substance, triphenyl formagane, is produced in living cells. This makes possible to distinguish the red colored living parts of seeds from a colourless dead ones. In addition to completely stained viable seeds and completely unstained non-viable seeds, partially stained seeds may occur. Varying "proportions of necrotic tissues occur in different parts of these partially strained seeds. Localisation and spread of necrosis in the embryo and on endosperm and the intensity of colour determine whether such seeds are classified as viable or non-viable".

Procedure :

→ each 4 replications of 100 seeds each from the pure seed fraction of physical purity test.

→ To facilitate penetration of tetrazolium test.

→ the seeds are fully immersed in distilled water or kept in paper towel for 18 hrs.

General Directions

Reagents

A 1% aquaceous solution (pH 6.5 - 7.0) of tetrazolium chloride or Bromide is used. If the pH of the distilled water is not with in the range of 6.5 - 7.0, the tetrazolium salt should be dissolved in Buffer solution. The buffer solution is prepared as follows.

Solution

Solution 1 : Dissolve 9.078 g of $KHgPO_4$ in 1000 ml. of water.

Solution 2 : Dissolve 11.876 g of $Na2HPO_4 .2H2O$ in 1000 ml of water.

Take 400 ml. of solution 1 and 600 ml of solution 2 and mix them together, to make a litre of buffer solution prepared as above and dissolve 10 gms of tetrazolium salt. This gives a tetrazolium solution of pH 7.0

Procedure

Each 4 replications of 100 seeds each from the pure seed fraction of physical purity test. To facilitate penetration of Tetrazolium solution, the seeds are fully immersed in distilled water or kept in paper towel for 18 hrs. The testa of the dicot is removed and the monocot is exposed by dissecting the seed longitudinally or laterally. The seeds are then completely immersed in 1% tetrazolium solution for 3 hrs. During treatments two preparations are kept in darkness at 20°C. After termination of the Tetrazolium test, the solutions is decanted and the preparation are mixed with water prior to evaluation. For examination the preparations are spread on a plate and kept wet throughout the determinations. The seeds are evaluated with the help of magnifying devices. Individual seed is evaluated as viable or dead on the basis of staining pattern in embryo.

Calculation:

The results are reported as percentage of viable seeds in relation to total seeds tested.

2] Embryo excision test

The objective of excised embryo test is to determine quickly the viability of tree seeds which normally germinate slowly or show dormancy under the prescribed methods to such an extent that a complete germination test requires more than 60 days.

→ Take 50 seeds ~~from~~ randomly from pure seeds.

→ The seeds are soaked for 1 to 4 days slowly running water or in standing water at temp below 25°c or in standing water at room temperature, at least two changes of water per days.

→ Hard outer covering, if any are removed before soaking the seeds. The seeds having hard seed coats are craked before soaking.

→ The embryos are excised from soaked seeds under moderately sterile condition in clean.

→ The instruments & working surface should be sterilized with 70% ethanol solution in water.

→ Seed coats should be carefully cut with a scalpel or razer blade & embryo excised with scalpel.

→ The embryos ~~must~~ must be touched as little as possible

→ Instruments must be cleaned & sterilized before every excision.

→ Those seed damaged by excision should be discarded & replaced by one of the extra seeds of working sample.

Method:

The test is performed in four replicates of fifty seeds drawn at random from pure seed fraction of purity test. The seeds are soaked for one to four days slowly running water or in standing water at a temperature below 25oC or in standing water at room temperature, at least with two changes of water per day. Hard outer covering if any are removed before soaking the seeds. The seeds having hard seed coats are cracked before soaking.

The embryos are excised from soaked seeds under moderately sterile conditions in a clean drought proof room e.g. below sheet of glass fixed about 200 mm above working surface. The instruments and working surface should be sterilized with 70 % ethanol solution in water.

Seed coats should be carefully cut with a scalpel or razar blade and embryos excised with scalpel. The embryos must be touched as little as possible. Instruments must be cleaned and sterilized before every excision. Those seeds damaged by excision should be discarded and replaced by one of the extra seeds of working sample.

INCUBATION:

The excised embryos should be placed on top of filter paper and kept under normal conditions of light and moisture at a constant temperature of 20 to 25 o^c for fourteen days.

As far as possible, the whole working sample should start incubation at the same time.

If heavy mould infection develops, a sterile re-test must be made.

Evaluation

The embryos should be examined daily and the test terminated as soon as distinct differentiation between viable and non viable embryos can be made, upto a maximum of 14 days.

Embryos mechanically damaged by excision can be distinguished from non viable embryos by localized discoloration of tissue into one of non-viable categories listed below, they shall be classified as viable.

The following categories shall be considered viable.

a) germinating embryos

b) Embryos with one or more cotyledons exhibiting growth or greening.

c) Embryos remaining firm, slightly enlarged and either white or yellow according to species.

d) Embryos of conifers that exhibit curvature of hypocotyls.

Following categories should be considered non-viable

a) Embryos, which rapidly develops severe mould, deterioration and decay.

b) Degenerated embryos

c) Embryos exhibiting extremely brown or black discolouration.

d) Dead or embryoless seeds.

Calculation and expression of results

The number of seeds considered viable is determined in each of four replicaticates. Maximum tolerated rages for replication differences are the same as for germination test. The average percentage is calculated to the nearest whole number and reported.

EXERCISE NO. 19

SEED AND SEEDLING VIGOUR TEST

Seed vigour is the sum of those properties of seed which determine the potential level of activity and performance of seed during germination & seedling emergence under a wide range of field conditions.

The test for determination of seed vigour

1. Direct Tests:

1. Brick gravel test
2. Paper piercing test

2. Indirect tests:

1. First count method
2. Speed of germination
3. Seedling growth rate
4. Seedling length
5. Seedling dry weight
6. Vigour index length
7. Vigour index mass
8. Tetrazolium Test

1. Direct tests:

a) Brick gravel test: A porous brick gravel of 2 to 3 mm size is used. About 30 mm layer of moist gravel is placed above the seed. This layer impedes the emergence of weak, partially diseased seedlings as well as coleoptile injured seedlings. Vigorous seedlings are these emerged from layer of brick gravel.

b) Paper Piercing Test: This test involves the use of sand plus a special paper disk through which seedlings penetrate. It is used for cereal crops in which seeds are placed on top with 1.25 cm moist sand and covered with special paper and kept for eight days.

2. Indirect tests:

1. First count

The number of normal seedlings counted at the first count ($4/5^{th}$ day) represents the faster germinating seeds. Higher percentage of normal seedling during the first count indicates the seed vigour.

2. Speed of germination

Number of germinated seeds are counted every day from the first day and the cumulative index is made by the formula.

$$n1/1 + n2/2 \ldots\ldots + nx/x = N$$

Where,

n1... nx are the number of seed germinated on day 1 to day x.

1 ... x are the number of days.

3. Seedling growth rate

Twenty seeds are placed in straight line on a paper towel moistened with distilled water and kept at an angle of 75 in a germinator at optimum temperature. Only 10 competitive normal seedlings are selected for observation. The remaining seedling are removed. For the next 10 days the length of each seedling is measured daily in cm. Seedling growth rate is determined by dividing the mean increase in length from each previous measure by the number of days the seedling had been in the germinator. Sum of each count at the end of the test period is expressed as seedling growth rate (Copeland, 1976).

4) Seedling length

Length of 10 normal seedling grown in moist towel paper kept at optimum temperature is measured in cm on the day of final count. The lot showing maximum seedling length is considered as vigorous.

5. Seedling dry weight

The weight of seedling excluding the cotyledon is taken on 10th day after oven drying at 100 C for 24 hr in g. The lot exhibiting the maximum seedling dry weight is considered as vigorous.

6. Vigour index length

A combination of standard germination test with seedling length provides broad evaluation of seedling vigour, seed lot with high vigour index is considered as vigorous. (VI I) = Germination × seedling length on the day of final count. (Root leng. + Shoot length).

7. Vigour index mass

Vigour index in terms of mass is determined by the multiplication of germination percentage with seedling dry weight on the day of final count.

8. Tetrazolium Test

This test is used as a viability test in which seed samples, to be tested for vigour test are washed to remove any traces of fungicides and then soaked for 16 to 20 hours in water at 30°C. The seeds are then cut-longitudinally from distal end towards base leaving two halves at base in joined condition. Shallow cuts are made through the pericarp of the seed half. Such seeds are soaked in 1% solution of TZ' salt for 24 hours

at 30°C. Sometimes, antibiotic compounds (Streptomycin/ penicillin) may be added at low concentration to prevent microbial infection.

However, this test differs from viability test in evaluation pattern. In viability test, seeds are evaluated into two categories viz. viable and non-viable. However, here they are evaluated in several categories. The aleurone cells become red where as dead cells remain unstained. Seeds are classified into.

A. 100-75 per cent of total aleurone surface stained-High vigour.

B. 75-27 per cent of total aleurone surface stained-Medium to low vigour.

C. Less than 25 per cent of total aleurone surface stained-Poor vigour.

Factors affecting seed vigour

1. **Seed size:** Bolder seeds produce vigorous seedlings
2. **Seed endosperm:** Well developed endospermic seeds show more vigour.
3. **Seed coat :**Papery seed coat shows greater vigour than thick seed coat.
4. **Genetical factors:** Hybrid seed produce much vigorous seedlings than parental seeds.
5. **Seed age:** Fresh seed having proper dormancy shows more vigour than old.
6. **Germination condition:** Under favourable conditions of germination, seeds show better vigour.

****

EXERCISE NO. 20

GENETIC PURITY TEST : GROW OUT TEST

I. Object

To determine the genetic purity of a given seed lot of a released cultivar and the extent to which the submitted sample conforms to the prescribed standards.

II. Sampling

The samples for grow-out test are to be drawn simultaneously with the samples for other quality tests and the standard procedure shall be followed.

The size of the submitted sample will be as follows.

1000 g - for cotton, groundnut, soybean and species of other genera with seeds of similar size.

500 gm - for sorghum, wheat, paddy and species of other genera with seeds of similar size.

250 gm - Beta and species of other genera with seeds of similar size.

100 gm - For bajra, jute and species of all other genera

250 tubers, stalks/ roots, corms, seed potato, sweet potato and other vegetatively planting propagating crops.

III) Procedure

While raising the desired population, standard and recommended cultural practices (e.g. field preparation, size of the plot, row length, distance between rows, distance between plants, irrigation, fertilization etc.) in respect of individual crops to be followed both for the unknown sample and its control.

The possibility to prove the genuineness of a cultivar by grow-out test is based on hereditary characteristics of the plants. Usually the cultivar differences are more distinct if growth conditions are favourable. Crop should be so grown that the genetical difference expresses themselves as clearly as possible. In self-fertilizing species the individual of a cultivar may be theoretically identical whereas the individual of a cultivar in cross-fertilizing species may not be genetically similar, but comprise a number of types. Therefore it is easier to determine the cultivar purity in self-fertilizing species than in cross-fertilizing species where the examination or greater part are based on the mutual comparison between the samples to be tested and the standard sample. Hence it is essential to sow the various samples of the same cultivar in successive and standard sample are sown at suitable intervals (for example one standard sample for every ten samples to be tested).

The size of plots, length etc. will differ crop to crop. However, the specifications for certification agency may change the specification considered necessary.

Sr. No.	Crops	Row length (m)	Plant to plant distance (cm)	Space between rows (cm)	Space between plots (cm)
1.	Wheat, barley oat	6	2	25	50
2.	Pea, cowpea	6	10	45	90
3.	Chickpea, green gram black gram	6	10	30	60
4.	Maize	10	25	60	90
5.	Hybrid cotton	5	10	45	45
6.	Paddy				
	a) Very early to medium	6	15	20	45
	b) Late and very late	6	25	30	60
7.	Pearl millet	6	10	50	90
8.	Sorghum	6	10	45	60

The seed rate may be adjusted depending on the germination percentage of individual samples and the sowing may be done by dibbling. Subsequent thinning is not recommended.

The test crop could be raised along with the control either in the areas recommended for the variety or in off-season nurseries. The authentic control sample from the origination plant breeder/breeding Institute is to be maintained by the testing station/Agency following standard procedures. A minimum of two hundred plants from control sample would be raised along with the test crop.

IV. Observations

a) All plants are to studied keeping in view the distinguishing characters described for the cultivar both in the test crop as well as the control. Necessary corrections may be incorporated if the control is found to be heterogeneous.

b) Observations are made during the full growing period, or for a period specified by originating breeding Institute add deviations from the standard sample of the same variety are recorded. At suitable development stage of the plots are examined carefully, and plants which are obviously of other cultivar are counted and recorded.

The specification of the field plot, row length etc. may be determined from the information given in para. II above. And on the basis of the number of plants required for taking observations is dependent on maximum permissible off types (minimum genetic purity) which are as follows

Maximum permissible Off-type (%)	Minimum genetic purity (%)	Number of plants required for sample for observation
0.10	99.9	4000
0.20	99.8	2000
0.30	99.7	1350
0.50	99.5	800
1.00 and above	99.0	400

V. Calculation interpretation and reporting of the result

Percentage of other cultivar, other species or aberrant found may be calculated upto first place of decimal.

While interpreting the result, USB of tolerance may be applied by using the reject table given below at serial No. VII.

VI. Analysts for grow out test

The analysts employed for conducting 'grow-out test' should possess the basic qualification as identified under seeds Rule, 1968.

VII. Reject number for prescribed standards and sample size

Genetic purity (%)	Reject number for sample size(number of plants) of	
	100	400
99.5	0.5	2
99.0	1.0	8
95.0	5	24
85.0	15	64

VIII. Result: Result is reported as percentage of genetic purity.

IX. Conclusion: The sample with genetic purity less than the MSCS is rejected.

****

EXERCISE NO. 21

GENETIC PURITY TEST : ELECTROPHORESIS

Electrophoresis

Electrophoresis is a technique which separate a mixture of protein into distinct band in a get that has been placed into an electrical field. This separation into distinct band is due to differences in size (molecular wt.) and charge of the involved.

Objective

Verification of variety by electrophoretic mobility of protein on poly acrylamide gel.

Equipment

Slab gel electrophoresis unit, power supply unit, aspirator, pH meter, mortar pestle, centrifuge, illuminator tray, razor blade and eppendorf tube or test tube.

Preparation of samples

Single seed soybean is decoated, crushed with mortar pestle and transferred to eppendorf tube or test tube.

Defattening

Crushed seed of soybean is treated with 5 ml mixture of chloroform, methane and acetone (2:1:1) for a period of 24 hours for defattening. The solution should be changed 2-3 times. Defatted material is dried by leaving the tube open over night at room temperature.

Extraction of protein

Defatted material is treated with extraction medium. Extraction medium varies from crop to crop.

Soybean

TRIS hydroxyl methyl amino methane (1.21 g) is dissolved in 70 ml distilled water. pH of this solution is maintained at 7.5 by adding concentrated HCI drop wise. The volume is made up 100 ml with distilled water. This extraction medium (0.5 ml) is added in eppendorf tubes containing defatted material. The mixture is left for 2-3 hr. at room temperature after stirring. It is centrifuged at 10°C at 10,000-15,000 rpm for 10-30 minutes and the supernatant is decanted and used for loading.

Preparation of gel

30% Acrylamide solution

1.0 g bis acrylamide for running and 2.0 g for stacking gel is dissolved in distilled water with 75 g acrylamide and the final volume is made up 250 ml with distilled water.

TRIS HCI buffer

22.69- g TRIS for running gel and 7.26 g for stacking gel is dissolved in 50 ml distilled water. pH of the TRIS HCI buffer for running gel is adjusted at 8.8 and for stacking gel 6.8 with HCI . The final volume is made up 100 ml with distilled water.

10% SDS Solution

10 g sodium dodecyl sulphate (SDS) is dissolved in distilled water with constant stirring and heating and the volume is made up 100 ml with distilled water.

5% APS solution

0.5 g ammonium per sulphate (APS) is dissolved in distilled water and the volume is made up 10 ml with distilled water.

Filling of cassette

Running gel is poured in between the plates of the cassette with the help 6Fa syringe in such a way that no air bubble is trapped in the gel solution. Three fourth part of the cassette should be filled with running gel. A layer of distilled water is overlayed on the running gel with the help of pipette. This gel is kept under fluorescent light for one hour for polymerization. The water layer is poured off after polymerization of running gel. Stacking gel is poured on the running gel and an acrylic comb is placed inside the cassette to make required number of wells. The comb is removed after half an hour when the gel has polymerized. The wells thus prepared are washed with tank buffer or distilled water.

Electrophoresis

The cassette with gel is fixed into the electrophoresis unit. The lower and upper tank of the unit is filled with electrode buffer.

Table Constitution of electrode buffer for electrophoresis of different crops

Chemical	Pearl millet	Sun flower	Soybean	Cotton	Wheat
TRIS(g)	3.0	9.0	3.0	9.0	-
SDS(g)	-	3.0	-	3.0	-
Glycine (g)	14.4	42.3	14.4	42.3	16
Distilled water (I)	0.5	3.0	0.5	3.0	2.0
acetic acid	-	-	-	-	160ml
PH	8.3	8.3	8.3	8.3	3.2
Make up of volume with distilled water (1)	5.0	-	5.0		150 ml of above solution to 3 litre

Wells of the polymerized gel are loaded with required quantity of protein sample with the help of pipette.

Table required quantity of protein sample to be filled in well for electrophoresis

Protein sample	Pearl millet	Sunflower	Soybean	Cotton	Wheat
µl	50	5	40	10	10

Tracking dye

Bromophenol dye (2-3 drops) is added to the electrode buffer in upper tank.

Power supply

The electrophoresis unit is connected to the power supply with anode (-) to the lower reservoir and cathode (+) to upper reservoir. The power supply should be cut down when the tracking dye reaches the bottom of the gel.

Fixing

The plates of cassette are removed carefully. Next morning the gel is rinsed with water after removal of TCA.

Staining

The staining solution for different crops are prepared as per details given below and stored in amber colour water.

Soybean

1.25 g commassive blue + 227 ml methanol + 46 ml acetic acid + distilled water to make the volume 500 ml.

The staining tray is filled with stain and the gel is placed in it. It is incubated till the bands are developed.

Destaining

For soybean destaining is done with a solution of 50 part methanol, 75 part acetic acid and 100 part distilled water.

Interpretation of protein banding pattern

After staining of the gel, it is placed over a trans illuminator to see the banding pattern. Relative mobility of each protein (band) is calculated by the following formula

$$\text{Relative mobility (Rm)} = \frac{\text{Distance travelled by protein}}{\text{Distance travelled by tracking dye}}$$

On the basis of Rm value and thickness of the band a zymogram is drawn on a paper to show the banding pattern.

The varieties are verified on the basis of banding pattern

1. By measuring Rm of bands.
2. Total number of bands.
3. Presence or absence of specific band.
4. Intensity of band.
5. Difference in banding pattern in comparison to authentic zymogram of the variety under test.

****

EXERCISE NO. 22

SEED CERTIFICATION : PROCEDURE

Seed Certification: It is legally sanctioned system for quality control of seed multiplication and production.

PHASES OF SEED CERTIFICATION: The seed certification has six broad phases.

1) Receipt and scrutiny of application.
2) Verification of seed source, class and other requirements of seed for raising the seed crop.
3) Field inspections to verify conformity to the prescribed field standards.
4) Post harvest supervision seed crops.
5) Seed sampling and analysis to verify conformity to prescribed seed standards..
6) Grant of certificate & certification tags, tagging and sealing of the container / bags.

PROCEDURE FOR SEED CERTIFICATION :

The seed certification procedure involves 6 different steps.

1: Registration of seed plot with SSCA by the seed grower: The seed grower has to register the seed production plot (foundation & certified) with the concerned District Seed Certification Officer (DSCO) of State Seed Certification Agency (SSCA). This registration is to be done prior to sowing or within 15 days of sowing of seed production plot.

2: Verification of Seed Source : For the production of foundation or certified seeds, the seed producer must produce a documentary evidence to seed certification agency to establish the source of seed with which the field is sown.

3: Field Inspection to Confirm the Prescribed Field Standards : Field inspection is one of the important steps in seed certification because many identifications including Varietal identification are possible only in the field. The seed crop is checked for proper isolation from other crops to prevent harvesting a mixture of seeds. Two to four field inspections are recommended to be done during seed production of different crops.

4: Supervision at Harvesting and after Harvesting : Proper supervision is required at harvesting and threshing to prevent mixing of seed crop with other varieties. Therefore, the grower notifies the certification agency his intension to

commence harvesting and threshing. If machines are used for harvesting and threshing, they are inspected and approved for cleanliness before being operated. The threshed produce (raw seed) is kept in temporary storage in new bags for preventing its damage by insects and pests before it is processed. Seed processing equipment is checked for proper cleanliness and then raw seeds are cleaned, sized and treated with suitable fungicides and insecticides. Seed lots are then bagged and proper identification marks are affixed on bags.

5: Seed sampling and Testing in Seed Testing Laboratory : After seeds are processed and bagged, the samples are drawn by SSCA official and submit to seed testing laboratory for testing. Seed sample testing is done for germination, moisture and physical purity to check whether the seed lots meets the seed standards for certification.

6: Tagging and Sealing : Based on the field inspection and seed testing, the certification agency issues certification tag, if the seed lots meet the minimum standards laid down under the seed act. The seed bag is properly sealed with the certification tag.

SEAL

Rs..... only

SEED CERTIFICATION AGENCY

MAHARASHTRA STATE

APPLICATION FOR CERIFIED SEED PRODUCTION PROGRAMME

(USE SEPARATE APPLICATION FOR DIFFERENT CROP/ VARIETY)

1. Full Name :
2. Location
   a) Address :.....
   b) Village Name : .....
   c) Post Office :.....
   Pin code :.....
   d) Taluka :.....
   e) District :.....
   f) Ref. Phone No.....
   g) Near by S.T Stand.:.....km.....
   h) Near by Rly Stn :..... km.....
3. Crop Details
   a) Crop Name :.....
   b) Variety :.....
   c) Hybrid /Improved.....
   d) Male seed :.....
   e) Female Seed
4. Seed Source Details :
   a) Name of Product :.....
   b) Source Seed :.....
   c) R.O. Number :.....
   d) Lot. Number :.....
   e) Other information :.....
5. Load Details
   a) Survey / Gut Number :.....
   b) Local Name :.....
   c) Area ha. :.....
   Acre :.....
6. Farmer`s Name : North.....
   : South ....., East.....
   : West .....
7. Seed to be Produced : Found I / Found II / Certi I / Certi II
8. Sowing Date : ...../...../...../
9. Name of Agency :.....
   :.....
10. Fee details : Registration Fee : Challan No.....
    : Inspection Fee : Date...../...../
    : Late Fee :
    : Total : Date ...../...../.....

Application's Signature.....

FOR OFFICE USE ONLY

Registration No. : ..... Dist Code

No.of Units : ..... Taluka Code :

Total Challan Amount :.....

Crop Code : Agency Code

.....

Note :- Incomplete forms will be rejected

****

EXERCISE NO. 23 to 27

A. VISIT TO SEED PRODUCTION PLOTS

1) Students will visit the seed production plots of Serials, Oilsedds, Pulses & Fibre Crops Note down the observations in field inspection report.

2) Get acquainted with characters of improved varieties/hybrids.

FIELD INSPECTION REPORT

Maharashtra State Seed Certification Agency Divi./ /No.

(seed certification report for..... Ha.area)

1. Name of seed grower / Producer.....Report No..... Village..... Taluka..... District..... Date of inspection.....
2. Survey No. of Seed Plot..... Time : From..... to.....
3. Location of farm.....
4. Previous crop : Kharif..... Rabi..... Summer
5. Name of crop..... Variety.....
6. Sour of seed..... Class and quantity of seed.....
7. Total acreage under seed production.....
8. Acreage of filed inspection
9. Sowing date.....
10. Spacing .....
11. Stage of seed crop during inspection .....
12. Isolation distance (mts) :
    a) North..... b) South.....
    c) East..... d) West .....
13. Name and stage of growth of contaminants.....

14. Field count (No. of plants/ heads-100/500/1000):-

Count No.	Number of heads / plants				Remarks i.e. names of contaminants
	Off types	Other crops	Weeds	Affected by seed borne diseases	
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					
Total					
Average					
%					

15. Crop condition.....

16. Quality of seed production work.....

17. Thus this crop confirm the standards of seed certification.....

18. Estimated seed yield (qtl. /ha).....

19 .Remarks.....

20. Was the seed grower or his representative present at the time of inspection.

Signature of seed grower/
his representative

Signature of inspector
Name :
Designation

B. VISIT TO HYBRID SEED PRODUCTION FARMS

1) Visit the nearest hybrid seed production farm. Record observations in the inspection report.

2) Get acquainted with the characters of parental lines of hybrid.

The concept of hybrid vigour given shull (1098) has universdal application in all biological sytems: the development of hybrids in maize in 1930 toiggred the pace of utilizing heterotic advantage in other crops resulted in release of chain of commercial hybrids. The basic biological requirements for successful hybrid seed production are the presence of hybrid vigour, elimination of fertile pollen in female plant (CMS system), adequate pollination and fertility restoration (RHAsystem) in hybrid seed. Once these biological requirements are meet, a practical programme for development of hybrid seed can be formulated as under.

1. **Types of crosses** : single , Double,
2. **Procurement of seed**; An essential requirement for hybrid seed production prgogramme is adequate supply of breeder/ foundation seed of parents. The types of parents required for multiplication of seeds as foundation / breeder depends on the system used for elimination of fertile pollens from female parents. In cytoplsamic genetic system requires three lines- A, B and R line.
3. **Field selection**: in the selected field in previous year same crop should not have been grown.
4. **Isolation**: proper isolation as per crop should be maintained.
5. **Synchronization**: The perfect or near perfect synchronization of flowering between parental lines i.e. A and R line is the first requirement for successful hybrid seed production. The synchronization behavior is highly influenced by temperature and thus variation is synchronization has been observed in many crops.

   The synchronization in parental lines to some extent (1-2 days) can be induced by agronomic management like application of additional dose of to one of the parent (late), deep sowing of seeds of early parent and spraying of hormones responsible for early flowering.

7. **Planting ratio** : The female and male planting ratio is strongly influenced by the efficiency of pollinators and availability of visible pollens.
8. **Pollination** : Pollination is carried out by wind or insect in hybrid seed production. If natural pollination is not sufficient supplementary pollination should be done.

   Other agronomical practices remain same

Students will visit hybrid seed plot fill the inspection repot

FIELD INSPECTION REPORT

Maharashtra State Seed Certification Agency Divi./ /No.

(seed certification report for..... Ha.area)

1. Name of seed grower / Producer.....Report No.....
   Village.....Talika.....District..... Date of inspection.....
2. Survey No. of Seed Plot..... Time : From..... to.....
3. Location of farm.....
4. Previous crop : Kharif..... Rabi..... Summer
5. Name of crop..... Variety.....
6. Sour of seed..... Class and quantity of seed.....
7. Total acreage under seed production.....
8. Acreage of filed inspection
9. Sowing date.....
10. Spacing .....
11. Stage of seed crop during inspection .....
12. Isolation distance (mts) : a) North..... b) South.....
    C) East..... d) West .....
13. Name and stage of growth of contamitants.....
14. Field count (NO. of plants/ heads-100/500/1000):-

Count No.	Number of heads / plants				Remarks i.e. names of contaminants
	Off types	Other crops	Weeds	Affected by seed borne diseases	
1					
2					
3					
10					
Total					
Average					
Percent					

15. Crop condition.....
16. Quality of seed production work.....
17. Thus this crop confirm the standards of seed certification.....
18. Estimated seed yield (qtl. /ha).....
19. Remarks.....
20. Was the seed grower or his representative was present at inspection time.,

Signature of seed grower/
his representative

Signature of inspector
Name :
Designation

Exercise No. 27 :

VISIT TO SEED PRODUCTION FARMS OF FIBER CROPS

COTTON	H.Y.V	Hybrids
Land requirement	**Well drained leveled land Free from wilt disease & volunteer plants. Isolated field from other varieties of same species.**	
Varieties / Hybrids	Deshi = Y1, JLA –794, AKA–7 . American= LRA-5166, JLH-168, Rajat	**H x H** = H-6, H-8, H-10, NHH – 44, PKVHy-3,4, &5. **H x B** = NHB – 12 **H' x A** = DH – 9.
Sowing time	Rainfed = June, Irrigated = 2nd fortnight of May & June.	
Sowing methods	By drilling.	By dibbling.
Spacings (cms)	Deshi = 45 x 22.5. American = 60 x 30.	Female = 150 x 150 Male = 100 x 100.
Planting ratio Female : Male	--------[see Fig-1 for Hybrid Seed Production]	On area basis female to male ¾ : ¼ or 4/5 : 1/5 , Approximately 1200 hills of female and 600 hills of male parent for one ha. Is required.
Seed rate kg/ha	Deshi = 10 – 12 . American = 8 – 10.	Female = 0.50 Male = 0.25
Fertilizers Kg/ha	100:50:50 plus two foliar spray of Urea / DAP (15 – 20 gm/liter ie. 2%) during boll formation	
Isolation Distance (Meter)	Foundation = 50 Certified = 30 5 meter between parents of hybrid and field of other varieties.	Foundation = 50 30 meter & 5 meter between parents of hybrid and field of other varieties. Foundation =50 meter.
Field inspection	Minimum 3. The first & second at flowering and third at maturity.	Minimum 4. The first & second at flowering, third at maturity & forth 15 days prior to harvesting.
Off types %	Foundation = 0.10 Certified = 0.50	Foundation = 0.10 Certified = 0.50
Pollen shaders % when MS line used	----------	Foundation = 0.05 Certified = 0.10
Objectionable weeds %	**Ranbhindi, Holly hock**	
Objectionable disease %	----------	
Seed yield Q/ha	Seed cotton Deshi = 8 to 10. American = 10 to 12.	Seed cotton 2 to 4.
AFTER HARVEST		
Seed moisture %	10.0	
Germination %	Minimum 65.0	

Weed seed No/Kg	Foundation = 05	Certified = 10
Other crop seed No/Kg	Foundation = 05	Certified = 10
Other disease	Fusarium wilt, anthracnose.	
Major pests	Boll worms (pink, American & spotted), Jassids, aphids and thrips.	

H= *G. hirsutum*, B= *G. barbadense*, H'= *G. herbaceum*, A= *G. arboreum*

NOTE : Maximum pure seed (98 %) and inert matter (2 %) is recommended as physical purity standard.

Certified Cotton Hybrid Technology

Male Parent $1/4^{th}$ area	Female Parent $3/4^{th}$ area

Male Parent $1/5^{th}$ area	Female Parent $4/5^{th}$ area

CERTIFIED COTTON HYBRID SEED PRODUCTION	
	Cotton hybrid H_4 [Released in 1970, 1st Cotton Hybrid in World]
Parentage	Gujarath 67 x American nectariless.
Land requirement	Well drained leveled land, Free from wilt disease & volunteer plants. Isolated field from other varieties of same species.
Sowing time	Rainfed : June, Irrigated : 2nd fortnight of May & June.
Sowing methods	Dibbling
Spacings (cms)	Female : 150 x 150 , Male : 100 x 100.
Staggering	Male parent (pollinator) need to be sown with a specific interval for three times ie. One week before, One week after and with female parent. It is essential for continuous supply of male flowers for pollination of hand emasculated flowers of female parent.
Planting ratio Female : Male	On area basis = 2:1, 3:1 or 4:1 female : male parent lines or area basis.
Seed rate kg/ha	Female = 0.50 Male = 0.25
Fertilizers Kg/ha	100 : 50 : 50 at sowing and foliar spray of urea / DAP (15 to 20 gm in one liter ie. 2% during boll formation.
Isolation Distance (Meter)	Certified = 30 meter, 5 meter between parents of hybrid and field of other varieties.
Field inspection	Minimum 4 First before flowering, second & third at during flowering and fourth one during picking of cotton in female parent Factors to be considered are Isolation distance, Volunteer plants, Off type plants, weed plants. During last inspection eliminate selfed bolls.

Rouging	Timely rouging of off plants, Diseased plant should begin at seedling stage. The subsequent rouging for off types and severely diseased plants should be done at square initiation stage.
GA_3 treatment	GA_3 treatment @ 50 to 100 ppm is given to the stigma of female parent, so that the seed setting efficiency is improved.
Crossing Programme	**Doak (1939)** gave the technique of crossing should be followed. The crossing programme should be organized during first eight weeks of the reproductive stage only. During this time the crop should not suffer due to excessive or scanty irrigation facilities. **Hand Emasculation** : Emasculate the optimum bud before its opening in afternoon session (2 to 6 pm). Cover the emasculated flower with butter paper bag or straw tube. Ensure perfectness in emasculation and remove such buds which have escaped during emasculation when they open. **Hand Pollination** : Open the emasculated bud by removing butter paper bag. Pollinate the emasculated bud during next day morning (10am to 1 pm) with pollens of desired male parent. Cover the pollinated bud with red tissue paper bag.
Off types %	Certified = 0.50
Objectionable weeds %	Certified = 0.10 Ranbhendi, Holly hock.
Objectionable disease %	Certified = 0.10 Fusarium wilt, Anthracnose.
Harvesting (Picking)	Mature and completely opened crossed boll having red tissue paper bag are collected and sun dried for two days. Later on store them in specially marked gunny bag. The ginning is done so that the lint and seed cotton is separated.
Seed yield Q/ha	04 – 06
AFTER HARVEST	
Seed moisture %	10.0
Germination %	Minimum 65.0
Major pests	Boll worm, jassids and aphids.

NOTE : Maximum pure seed (98%) and inert matter (2%) is recommended as physical purity standard.

	Deshi cotton	American cotton
Introduction	----------	---------
Pure line selection	Gaorani, AKA – 5.	Buri 1007, SRT – 1
Mass selection	----------	---------
Pedigree selection [After hybridization]	Y – 1, PA – 141(Namdeo), PA - 183(Savata), PA – 255, PA – 402 (Vinayak) AKH – 4 AKA – 7 , AKA – 8 AKA 8401, Jyoti, JLA – 794	JLH – 168, RHC – 0688, NH – 452 (Renuka), NH – 545 AKH – 081, AKH – 8828 AKH – 84635 (Rajat), LRA – 5166, PH – 348 (Yamuna), DHY – 286, PH – 93 (Nagnath) Egyptian Cotton : Suvin.
Mutation Breeding	----------	MCU 7, MCU 10, Indore 2
Biotechnology	Bollguard, MECHBT – 12 , MCEHBT – 162.	
Commercial Hybrids	Hand Emasculation & Pollint	GMS / CGMS based
H x H hirsutum x hirsutum	H – 4, H – 6, H – 8, H – 10, NHH – 44, PKVHy – 2	(GMS) Saguna (CGMS) PKVHY – 3, & PKVHY – 4, PKVHY – 5
H x B hirsutum x barbadens	Varlaxmi, Savitri, DCH – 32, DHB – 105, TCHB – 213, HB – 224	----------
H x A herbaceum x arborem	G. Cot. DH – 7, PHA – 46, DDH – 2.	----------

DIFFERENT COTTON HYBRIDS with their parentages:

S.N.	Hybrid	Parentage	Year
	G. hirsutum x G. hirsutum Hybrids (H x H)		
	H_4	Gujarat 67 x American nectariless	1970
	H_6	G. Cot – 100(Vishnu) x G. Cot – 10	1989
	H_8	G. Cot. – 10 x Surat dwarf	
	H_{10}	B – 68 – 2 x LRA – 5166	1995
	NHH – 44	Bicanery Nerma – 1 x AC – 738	1983
	AHH – 468 (PKVHY-2)	AK – 32 x DHY – 286 – 1	1981
	PKVHY – 3*	MSCAK – 32 A x DHY – 286 –1 R	
	PKVHY – 4 *	MSCAK – 32 A x AKH – 07 R	1996
	PKVHY – 5*	MSCAK – 053 A x AKH – 02 R	
	Phule – 492	RHC – 003 x RHC – 004	2000
	Godavari	Buri nectariless x MCU – 5	1975

	PHH 316 (Hyb. ganga)	PH – 93 x PH – 325	
	G. Cot HY – 8	G. Cot – 10 x Surat dwarf	
	G. hirsutum x G. barbadense Hybrids (H x B)		
	Varalaxmi	Laxmi x SB – 289 – E	1972
	Savitri	Kop 203 x SB 1085 – 6	1973
	NHB – 12	NS – 15 x SB – 289 – E	1989
	Phule – 388	RHC 006 x RHC – 001	2001
	DCH – 32	DS 28 x SB– 425(yf)	
	G. herbaceum x G. arboreum Hybrids (H' x A)		
	DH – 9	4011 x 824	1988
	PHA – 46	Ph – 1 x PA – 146	1996
	G. Cot DH – 7	Sujay x G – 27	
	Intra specific (G. arboreum x G. arboretum)		
	AAH – 1	GMS – 1 x HD – 226	

* CGMS based hybrid, (yf) = Yellow flowered

EXERCISE NO. 28

VISIT TO SEED TESTING LABORATORIES

Objective of visit:

Seed testing laboratory is hub of quality control, which deals with evaluating planting value of a seed lots. To carry out these responsibilities effectively. Seed testing laboratories should be manned and equipped well.

Set up of seed testing laboratory

The seed testing laboratories are under the control of the Government. To perform various tests timely and to send the results to growers well in advance, for easy and timely marketing, the work of seed testing laboratory is divided into six sections.

1. Office section
2. Purity section
3. Germination section
4. Genetic purity section
5. Seed health section
6. Storage section

1. Office section: This section maintains the accounts and records of samples and forwards the results in prescribed proforma. Procedure of Routing Seed Sample in Laboratory: The sample received in the laboratory are first entered in a register and assigned, laboratory test numbers (registration number). The details such as sender's name, address, name of crop/variety plot number, date of dispatch and receipt of sample and kind of tests to be carried out. The sample is given office test number called code number to avoid identity of sample. Half quantity of original sample is preserved by storage section. It is used in case of dispute.

Following are different steps through which the sample is routed in seed testing laboratory.

1. Registration of sample
2. Mixing and dividing of sample & preparing working sample
3. Moisture and seed weight test
4. Purity analysis
5. Germination test
6. Seed health test
7. Checking the results
8. Issue of certificate Equipment used in laboratory:

I. Sampling:

1. Sleeve type triers
2. Boerner, Gamet precision, soil type seed dividers
3. Sample pans, buckets and containers

II. Purity equipments

1. Purity work board
2. Forcep and spatula
3. Magnifier
4. Aluminium dishes
5. Seed blower
6. Set of sieves
7. Sample trays

III. Germination equipments:

1. Seed germinator
2. Thermometers
3. Counting boards
4. Soil and sand boxes
5. Germination boxes
6. Petri dishes
7, Germination paper towel/ blotting
8. Wash bottles
9. Sample trays
10 .Sprinklers

IV. Viability test equipments:

1. Sampling dishes
2. Cutting and piercing devices
3. Oven
4. Droppers and
5. Dispensing bottles
6. Magnifiers
7. Magnifires
8. Stereoscopic microscope

V. Moisture test equipments :

1. Oven
2. Incubator
3. Autoclave
4. Sieves
5. Ultraviolet lamp
6. Stereoscopic microscope
7. Grinding Mill
8. Dessicators
9. Analytical balance
10. Moisture meter
11. Sampling pan

VI. Seed health testing equipments:

1. Stereoscopic microscope
2. Oven
3. Incubator
4. Refrigerator
5. Sieves
6. Autoclaves
7. UV Lamp

VII. Other apparatus: Seed cleaning machine i.e. air screen cleaner machine (laboratory model), spiral / disc gravity separator, aspirator, Hygrometer.

Seed Testing

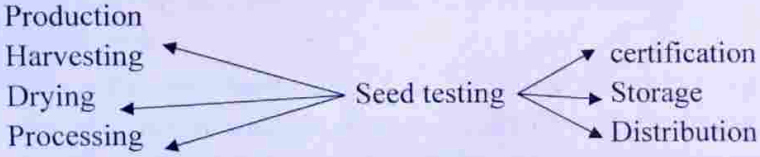
Testing of seed evaluates planting value and the authenticity of seed lot. It furnishes information of different quality attributes such as -

1. **Physical Purity :** It is essential for determination of minimum per cent of pure seed and maximum per cent of weed seed, other crop seed and inert matter. Purity test provides information for the specific processing & reprocessing requirements.
2. **Germination :** Optimum germination is required to maintain optimum plant population with recommended seed rate at specific spacing for high yield.
3. **Seed vigour & viability:** Vigorous & viable seeds are required to establish optimum vigourous plant population. These attributes denote capacity of normal seedling production under comparatively poor & stress conditions.

4. **Seed health:** Seeds free from seed born diseases are essential to avoid epidemics. Similarly seeds free microbes, insects and pests maintain optimum plant population. It indicates need for protection treatment.
5. **Objectionable weed:** Pure seeds should not be contaminated with objectionable weeds so as to check their spread.
6. **Genetic purity:** For uniform performance & quality of produce, seed used for sowing must be genetically pure.
7. **Seed moisture :** Optimum seed moisture is helpful in guiding of packaging & storage requirements. During maturity, this information is useful for deciding drying procedure.

Seed samples are send for seed testing must be sent in cloth bags with detailed information regarding the stage of seed production, variety, lot number, sample number, kind of test to be conducted, sender's name and address, date of dispatch, code no. of seed processing plant. For seed moisture test, sample seeds need to be sent in moisture proof packages (700 gauge polythene envelope)

Seed test results are useful in truthful labeling and quality certification programme. These results may cause rejection of seed lots for distribution or for further multiplication if it does not confirms minimum prescribed standards of purity, seed health, seed vigour etc. This record is also useful as a evidence against sellers and concerned seed production if they are selling low quality seed as a high quality seed to consumers. Seed testing is anchor in all seed programmes for utilization, storage and distribution.



Treating :

For giving label as a Certified seed to the given lot by SSCA, it is necessary to meet/ fulfill all prescribed standards for all seed quality attributes i.e. physical & genetic purity, germination, seed moisture, seed vigour, seed health etc. If it does not fulfill prescribed standards even for any one of the seed quality attribute, then the given seed lot is rejected for seed certification.

1) Visit the seed testing laboratories and write the report.
2) Enlist the various equipments used in laboratory and draw diagram.
3) Location of seed testing laboratory:
4) Name and designation of incharge:
5) Different testes carried out
6) List of crops tested:
7) Annual capacity of laboratory:

***

EXERCISE NO. 29 & 30

VISIT TO SEED PROCESSING PLANTS

SEED PROCESSING:

Harvested seed contains moisture and impurities such as shrivelled, broken and chaffy seed; diseased/insect infested hence, processing of seed is essential.

Seed processing refers to follow the steps required for preparation of harvested seed for marketing viz. drying cleaning, grading, seed treatment, bagging, tagging and sealing.

Objects:

1. To dry seeds to safe moisture level
2. To separate out inert material, weed seeds, other crop seeds and damaged seeds.
3. To maintain uniform size of seed
4. To treat the seed with protective chemicals in upgrade the seed quality.

I. Seed Drying: It is necessary to lower down the seed moisture content to safe moisture limit in order to maintain seed viability and vigour. It avoids deterioration by checking fast growth of molds and micro-organisms activities. It also helps to store for long time.

Methods: The main methods are –

i) Natural drying ii) Sun drying iii) Forced air drying

i) Natural Drying : During the physiological maturity, the moisture is transferred from seed to the atmosphere and crop become ready for harvest. It is also known as field drying.

ii) Sun Drying : Seed can be dried in sun and stored directly. During sun drying, seed should be turned and altered occasionally for uniform drying. While sun drying, seeds should not be spread on wet, dirty and kuchha threshing yard. Similarly produce of one crop variety should be dried at a time to avoid mechanical mixture.

iii) Forced Air Drying: In this system air (natural or heated) is forced into seeds. The air passing through damp seeds picks up water. The evaporation cools the air and seed. The heat necessary for evaporating the water comes from the temperature drop of the air. There are three methods -

a) Natural air drying - Natural air is used

b) Drying with supplement heat-small quantity of heat is used to raise temperature about 5 to 10 °F for reducing relative humidity.

c) Heated air drying-drying air is heated by 110 ° F. First two methods take long time (1 to 2 weeks or more) to reduce moisture level. They are used in Western countries in India, third method i.e. heated air drying is used.

Procedure

1. Change seed into bin to a recommended depth with uniform distribution.
2. Operate drier at recommended temperature which is done manually or by setting a thermostat to desired temperature.
3. After drying, allow blowing air to pass through seed without heat to bring seed temperature down to air temperature or 50 ° F.

The different systems of air forced drying are-

a) Wagon drying: Special type of batch drying with heated air. In this system, the seed is loaded directly to the specially constructed wagon. It is drawn to the drier and connected. 3-4 wagons can be dried at once. After drying wagons are disconnected and seed is cooled with small fan.

b) Bag Drying : This is suitable when many varieties are handled simultaneously or when seed lots are small in size and received in bags. Excellent air flow with minimum static pressure is possible due to drying bag is only one sack deep.

c) Box Drying : They are modified bag driers. This method helps to maintain identity of seed into in spite of bulk handling. The boxes are prepared locally and fitted with perforated metal or woven wire bottoms. After heating boxes are removed from drier and stored temporarily.

d) Multiple bin storage drying : This enables the drying of several lots of seed simultaneously using same drying fans. Alternatively, different lots of seeds could be dried. This is specially useful to dry two or more kinds of seeds at a time.

II. Cleaning and Grading : It is the process of separation of undesirable materials viz. inert matter, weed seeds, other seeds, light and chaffy seeds, damaged and deteriorated seed from desirable seed material. It is done on the basis of physical properties of seed and undesirable material. Seeds of different species and inert matter differ widely in respect of length, width, shape, weight and surface thus these forms the basis of seed cleaning.

Methods of cleaning seeds:

1. Methods of preparing seeds for seed cleaning (Pre-conditioning and pre-cleaning operations).
2. Basic seed cleaning operations
3. Upgrading the quality of cleaned seed

1. **Pre-conditioning** refers to shelling debearding etc. which is required for basic seed cleaning, while pre-cleaning refers to removal of particles such as stones, clods, trash etc. It is done by using various equipments such as scalpers, debearders, huller-scarifier, maize sheller and buckmorn machine.

2. Basic cleaning refers to actual cleaning and grading of seeds. It is done by an air screen machine (air screen cleaner) Different types of screens with sieves are used for different crops.

3. Upgrading of seed is done by removing specific contaminants or by size grading. The process is followed after basic cleaning and refers to upgrading of seed.

III. **Seed Treatment**: It refers to the application of fungicide or insecticide or both to seed so as to disinfect them from seed borne or soil borne pathogenic organisms and storage insects.

Advantages:

1. Prevention of spread of plant diseases.
2. Protection from seed rot and seedling blight
3. Improves germination
4. Protection from storage insects -
5. Controls soil insects

Type of Seed Treatments:

1. Seed disinfection: It refers to eradication of fungal spore established within seed coat or deep tissues. Fungicide treatment should penetrate the seed to kill fungus.

2. Seed disinfestations: It refers to destruction of surface borne organisms that have contaminated the seed surface. Chemical dips, soaks, dust, clurry or liquid can be practiced.

3. Seed protection : To protect seed and seedling from decay by soil micro organisms seed treatment is given. The seeds which are broken or infested by disease organisms must be treated, similarly, if seeds are required to be sown or planted under unfavourable conditions needs to be treated. In general, to avoid infection or infestation of disease organisms, seed treatment is given.

IV. Seed Sampling (As per Ex. No.11)

V. Bagging, tagging and handling: After seed treatment, seeds are packed/bagged into containers of specified net weight.

It consists of:

1. Filling of seed bags to an exact weight
2. Placing leaflet regarding improved cultivation practices
3. Attaching labels, certification tags on the seed bag and sewing with tag or label.
4. Storage or shipment of bag.

1) Visit the seed processing plant and write the report.
2) Draw the diagrams of important seed processing equipments.

Collect the following information

1) Name of owner of seed Processing plant:
2) Registration No.
3) Location:
4) Crops processed:
5) Equipments available :
6) capacity of plant:
7) processing sequence :

***