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DETERIORATION OF SEED QUALITY AS REVEALED BY TOPOGRAPHICAL TETRAZOLIUM TEST IN FIELD BEAN

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A study to determine seed quality of stored field bean cv. Co. 1 by topographical tetrazolium test in comparison with the standard germination test revealed that the germinability differed significantly due to the seed treatments and storage containers. The percentage of germinable seeds in tetrazolium test showed close association with the results of standard germination test. Besides, this test has elegantly and picturesquely characterised the damages occurring in seeds due to deterioration.

A rapid, reliable and reproduceable, method to determine the germination potential coupled with vigour of seed is required for the seed industry over and above the existing standard germination test. The tetrazolium test is the one which simultaneously and rapidly estimates the potential germination and soundness of individual embryos (Moore, 1961; 1962) as well as diagnosis the causes for embryo disturbances (Moore, 1966).

In this biochemical test, 2, 3, 5 triphenyl tetrazolium chloride molecules react with hydrogen atoms released by the dehydrogenases enzymes which are involved in the respiration process of living tissues and results in the production of a water insoluble oil-soluble red pigment, formazan. This makes it possible to distinguish the red colour living tissues from the colourless dead ones.

This study was undertaken to assess the location and nature of disturbances within the embryo tissues as influenced by pre storage seed treatments, storage containers and period of storage).

MATERIALS AND METHODS

Ths seeds of field bean *(Lablab Purpureus* (L.) Sweet var. *Lignosus* (L.) prain), cv. Co. 1, commonly known as mochai, treated with seed protectants and stored in different containers for 40 months under ambient temperature and relative humidity conditions formed the material for this study. The details of pre-storage treatments were as follows:

a] Seed protectants :

T_0 — Untreated [control]

T_1 — Captan 75per cent WDP and DDT 50 per cent WP at 2 g aud 200 mg, respectively dissolved in 5 ml of water per kilogram of seed

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T_3 — Captan at the rate mentioned above + Activated Clay @ 1:100 ratio (w/w)

T_4 — Coating with red earth paste at 1:100 ratio [w/w]

b] Containers :

C_1 — Fresh gada cloth bag of 14X10 cm size

C_2 — 700 gauge thick polyethylene bag of 14X10 cm size [Heat sealed]

Topographical tetrazolium test

A 0.5 per cent aqueous solution of 2,3,5-triphenyl tetrazolium chlorid of pH 7.0 was prepared by dissolving 5 g of the salt in 1000 ml of sorensen's phosphate buffer solution. Each test was carried out with 4 X 100 seeds, sampled with a soil divider. The seeds were preconditioned by keeping the seeds in moist paper towels overnigth, so that they absorb moisture slowly without causing damagee to cotyledon. The seed coat is removed without damaging the embryo. The seeds were, then completely immersed in tetrazolium solution and kept in darkness for 45 min at 40°C in an incubator. After staining, solution was decanted and seeds were thoroughly rinsed with water and spread on the petridish under water. The seeds were thoroughly examined with a magnifying lens and classified into germinable and non-germinable seeds based on the staining pattern (Delonche *et al*(1962). The mean of four replication was expressed as percentage to the nearest whole number.

Standard germination test

The germination test was carried out using "between papertowel medium" (ISTA, 1976), at 25°C ± 0.5°C temperature and 90 ± 3 per cent relative humidity with 4 X 100 seed spaced uniformly. After 7 days' the percentage germination was calculated based on the number of normal seedlings produced.

RESULTS AND DISCUSSION

The differences between mean germination percentage of seeds as evaluated by the topographical staining pattern, were highly significant due to seed treatments and storage containers. The interaction of seed treatments and container was also highly significant (Table 1) The seeds treated with captan and activated clay recorded high germination and was superior to those from other treatments. The untreated control and seeds treated with red earth were inferior, recording 53 per cent germination each. The seeds stored in 700 gauge polyethylene bag (moisture vapour-proof containner) recorded 92 per cent germination and was superior to those stored in cloth bag (moisture pervious container). The germination-potential of seeds determined by this method agreed very closely to

Table 1. Viability of 40 month-old field bean seed Cv. Co1

	Tetrazolium test						Standard germination est		
	Germinable [%]			Nonegerminable [%]			Germinatian [%]		
	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean
T_0	17	89	53	83	11	47	25	90	58
T_1	32	95	64	68	5	36	55	100	78
T_2	50	98	74	50	2	26	55	95	75
T_3	22	84	53	78	16	47	45	95	70
Mean	30	92	—	70	8	—	45	95	—

CD [P=0.05]

Treatment	3**	2**	5**
Container	2**	1**	3**
Treatment X Container	4**	3**	7**

Table 2. Staining pattern (mean values in percentage) of field bean seeds CV. Co 1 stored for 40 months

Tetrazolium staining pattern	C_1				C_2			
	T_0	T_1	T_2	T_3	T_4	T_1	T_2	T_3
1. Germinable–Seed Completely stained not overly intense		10	20	32	14	80	85	90	61
2. Germinable–Minor unstained areas on cotyledons		4	7	10	4	5	5	5	10
3. Germinable–Extreme unstained tip of radicle unstained, minor unstained areas on cotyledons		3	5	8	4	4	5	3	13
4. Non–germinable–Radicle unstained		40	38	40	20	–	–	–	–
5. Non–germinable–unstained area in region where plumule is located		9	6	4	20	–	–	–	–
6. Non–germinable–Series of unstained areas on upper portion of radicle hypocotyl axis		8	5	3	24	–	–	–	–
7. Non–germinable–More than one half of cotyledon tissue unstained		–	–	–	–	5	3	1	3
8. Non–germinable–Milky red areas of cotyledon		–	=	=	–	5	1	–	2
9. Non–germinable–More than extreme tip of radicle unstained more than one–half of cotyledon tissue unstained		15	10	3	10	–	–	–	6
10. Non–germinable–Juncture af radicle–hypocotyl axis and cotyledon unstained		10	9	=	4	–	–	–	4
11. Non–germinable–Seed completely unstained		1	–	–	=	–	–	–	=

the results of germination by standard germination test (Tagie 1). Agrawal et. al. (1973) found a clase association between the viability percentage obtained by the tetrazolium method and the germination percentage in corn, paddy day wheat.

The classification of germinable and non-germinable seeds based on topographical staing pattern revealed that the majority of the non-germinable seeds showed necrotic areas either in the primary axis or in the cotyledons ot both as detailed in the Table 2. It was observed that the seeds treated with red earth and stored in polyethlene bag developed necrotic areas in the radicle as well as in the cotyledon. however, this type of injury was not observed in seeds treated with other seed treating chemicals. It was also interesting to note that the radicle portion of the seeds stored in cloth bag was heavily damaged during storage as compared with their counterparts stored in polyeth ylene bag. Oxley (1948) suggested exhaustion of organic matter particularly the depletion of respiratory substrate (went and muntz, 1949) is the cause for loss of viability.

Evaluation of seeds into germinable and non-germiuable and vigorous and less vigorous ones could be accomplished with a relatively short period of time, not evceedihg 24 hrs by this method. Moore (1967) is of the opinion than the tetrazolium topographical method is a powerful diagnostic tool for detecting and analysing seed injury.

REFERENCE

AGRAWAL, P. K., J. L. Karihaloo, S. M. M. Ahmed and P. C. Gupta, 1973. Predicting germinability in maize, wheat and paddy seeds on the basis of tetrazolium test, Seed Res., 1: 83-85

DELOUCHE, J. C. T. wayne Still, M. Raspet and M. Leinhard. 1962. The terazolium test for viability, Tech. Bull. No. 51, Mississipi agric. Expt. Sta. Mississippi State Univ. PP. 1-63

INTERNATIONAL, Rules for Seed Testing. 1976. International Seed Testing Association. Seed Sci. & Technol. 4: 3-49.

MOORE, R P. 1961. Tetrazolium evaluation of the relationship between total germination and seed quality, Proc Ass. Off. Seed Analysts, 51: 127-130.

MOORE, R. P. 1962. Tetrazoljum, as a Universally acceptable quality test of viable seed. Proc. Int. Seed Tese. Ass. 27: 794-805.

MOORE. R. P. 1966., Tetrazolium tests for diagnosing causes for seed weakness and for Predicting and understanding performance, proc, Ass. Off. Seed Analysts, 56: 70-73.

MOORE, R. P. 1967. Freeze injury to seed corn as evaluated in tetrazolium and growth test. proc. Ass. Off. Seed Analysts. 57: 138-140.

OXLEY, T. A. 1948. The Scientific principles of grain storage. Northern publishing Co., Liverpool.

WENT, F. W. and P. A. Muntz, 1949. A Long - term tesf of seed Longevity. El. Aliso, 2: 63-75.