DEGRADING OF SEED QUALITY FOLLOWING 8 MONTHS IN STORAGE OF THE RICE, MAIZE AND SOYBEAN BASIC GRADE SEED

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ABSTRACT

Most of the seed producers in Vietnam are farmers and small seed companies. They have not yet the standard storehouses to reserve seed of agricultural crops but commonly store seeds in uncontrolled temperature and moisture brick storehouses (BSH). This study identified influences of that method of seed preservation on seed quality of rice, maize and soybean seeds during storage. The results showed that seed moisture content is increased from start of storage to the eighth month and the seed quality is degraded compared to Vietnam standard of crop seed. Moisture contents of rice seed after 6 months, maize seed after 4 months and soybean after 3 months storage were higher than that of the standard seed. Seed vigor and viability were evaluated by TZ and seedling growth rate test. The degradation of vigor and viability value appeared in all three kinds of seed, but the strongest is a soybean seed degradation, from 97.0% down to 15.67% if used longitudinally cutting procedure. The TZ test method gives similar result as germination test does so that TZ test can be used to evaluate germination capacity of rice, maize and soybean seed with high accuracy. Germination percentage of three kinds of seed is reduced after 8 months in BSH. So that rice seed after 8 months, maize seed after 7 months and soybean after 4 months storage lost grade of basic seed compared to Vietnam standard for crop seed. The germination rate is different when using different tests with different substrates and methods with the same crop seeds. A large amount of fungi lives on seed surface and increases through storage months in BSH. In this study, no fungi were identified inside seeds except in soybean seed. After 8 months storage 100% of soybean seeds were infected with pathogens. Treating seeds with proper fungicides before packing and storage to protect seed and young seedling from many seedborne pathogens is necessary.

Key words: Seed, germination, vigor, viability, seed health, degrading, deterioration, storage, seed quality, Vietnam standard of crop seed, basic seed

Abbreviations: TZ, tetrazolium • Cla, cut laterally • Clo, cut longitudinally • BSH, brick storehouse • MC, seed moisture content

1. INTRODUCTION

In the agricultural production, seeds serve as reproductive units for begining of agricultural production process, therefore the selection and use of high quality seed of adapted superior varieties are very important. Using good quality seeds can increase yields by 5-20%. The level of this increase is directly proportional to the quality of seed (that is sown) (Leroy, 2000). Seed quality depends on many factors during seed sowing process, field management, post harvesting, seed processing and storage. Seed producers could store their seed for selling in the next season or even after some years. There are some storage methods as cryogenic, hermetic and containerized storage. Purpose of seed storage is to preserve planting stocks from one season to the next. In some cases the objective of seed storage is to maintain seed quality overtime for improved plant breeding programs (Mew *et al.*, 1994; Mew *et al.*, 2005). The storage life of seed is strongly influenced by type and condition of the seed storage such as seed characters and storage condition. Storage conditions consist of temperature, humidity. (Oren, 1978). In Viet Nam, some seed companies and seed producers lack standard storage with appropriate equipment and suitable conditions. Commonly, seeds are packed in woven plastic bags placed in the iron containers (100 kg), which are in the brick storehouse (BSH), under uncontrolled temperature and humidity. In this method, storage condition depends on the environment such as humidity and temperature. In order to identify the degradation level of seeds in this process, we conducted an experiment with three kinds of the basic seeds rice, maize and soybean during 8 months of storage.

2. MATERIALS AND METHODS

Materials

The study was carried out with three crop seeds: modern rice KD18 variety, maize open pollination VN2 variety and soybean inbred DT84 variety. The experiment was conducted from February to September 2006 at the BSH of Hanoi University of Agriculture.

Storage conditions

The seeds were packed in woven plastic bags 1 kg /bag. Ten bags were kept in an iron container (100kg) and three seed lots were placed in BSH. The storage conditions were similar to that of the environment outside: an average humidity 79.5% and an average temperature of 25.9^{o}C during 8 months in 2006.

Sampling

Seed quality was tested monthly from harvest until the eighth month. Primary samples were drawn from the seed lot (within containers) and mixed into a composite sample and then reduced to a submitted sample and a working sample used for testing (Schmidst, 2000, p.8-9)

Seed moisture content (MC) test

Moisture content is expressed as a percentage of the weight of the original sample (ISTA 1996). Moisture content was measured by taking moisture tester, the average of three readings (AOSA, 2004; CFIA, 1997; ISTA, 1996).

Vigor and viability tests

In the tetrazolium (TZ) test method, dry seeds are cut to expose the embryo. There are two cut methods lateral (Cla) and longitudinal (Clo). The solution of 0.1% tetrazolium chloride, a colorless dye, is applied to the embryo. After a suitable period, about 1/2 hour, the seed is examined for the appearance of a red color, indicating respiratory activity. The percentage of seed that shows respiratory activity via the color change indicates the percentage of viability. Two other methods to evaluate seed vigor are the Hiltner method (Brick Grit Test) and the Seedling growth rate (AOSA, 2004; CFIA, 1997; ISTA, 1996).

In the seedling growth rate test method, basic seed were weighed to determine their increase in dry weight. Seedling growth rate is correlated with vegetative development in the field (Copeland and McDonald, 1995).

Germination test

The study conducted on the substrates (petri dishes, sandy tray), with three replications, each replication saved 100 grains (Leroy, 2000). One week after germination, germinated seeds were counted and the percentage of germination is calculated as:

Germination percentage = No. seed “normal germinants”/ total of sown seed

Seed health test

Only pathogen infected seeds were tested by agar plates to identify seedborne fungi. Seed pre-treated by NaOCl (sodium hypochlorite). Healthy seed percentage is the number of infected pathogens per total number of seed tested (Leroy, 2000).

3. RESULTS AND DISCUSSION

The seed moisture content is the most critical factor in keeping high rate of seed germination and viability during storage. The seed must be dried to safe moisture content before storage. In sealed conditions, the seed moisture changes less than that in an open storehouse. The change of seed moisture content through 8 months of storage in the BSH indicated that seed moisture of rice, maize and soybean increased by 2.72% to 7.84%, with seed moisture fastest increasing in soybean seed.

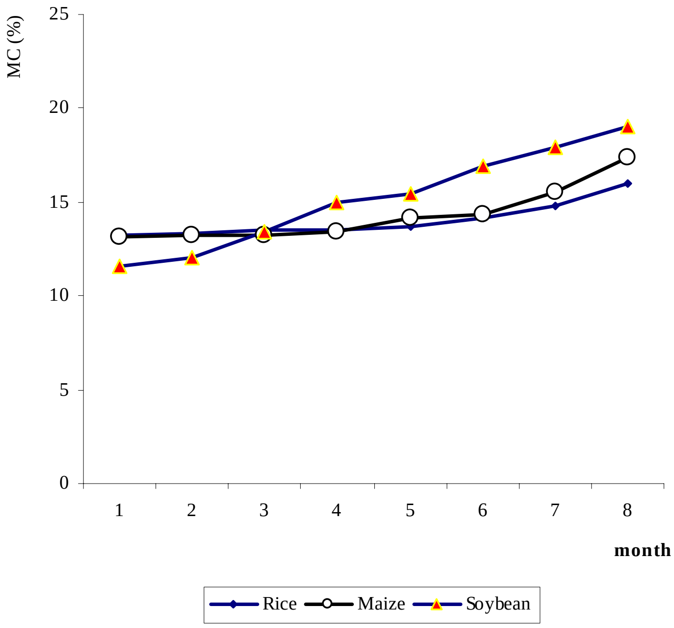

Fig. 1. Seed moisture content of rice, maize and soybean in BSH conditions.

The moisture content increased negligibly from the first to the sixth month in rice seeds and from the first to the third month in maize seeds respectively then increased faster to over 15.97 % for rice and 17.37% for maize seeds at the eighth month. The quality standard of crop seed in Vietnam (TCVN-1766:2004), for basic rice seed is not over 13.5% and the quality standard of crop seed in the Ministry of Agriculture and Rural Development (MARD) (10 TCN 312:2003) for seed of open pollinated maize is not over 13% in normal bags and for soybean not over 12%. Rice seed lost its grade after 6 months, maize after 4 months and soybean after 3 months of storage. The rice seed standard of IRRI is 14% for breeder seed, basic seed and certified seed (Mirsa et all, 1994; 2005) so that rice seed MC in this experiment is ensured. The moisture of soybean seed increased steadily from the first to the eighth month causing soybean seed loss of the germination capacity faster than rice and maize seed. This result agrees to other studies (ISTA, 1996).

The seed MC rapidly increased because seed stored in BSH are influenced by environmental condition. The average air humidity and temperature from February to September 2006 are in Fig. 2. Average temperature in all months is above 20^{o}C and the humidity is over 70%. These conditions are linked with seed moisture.

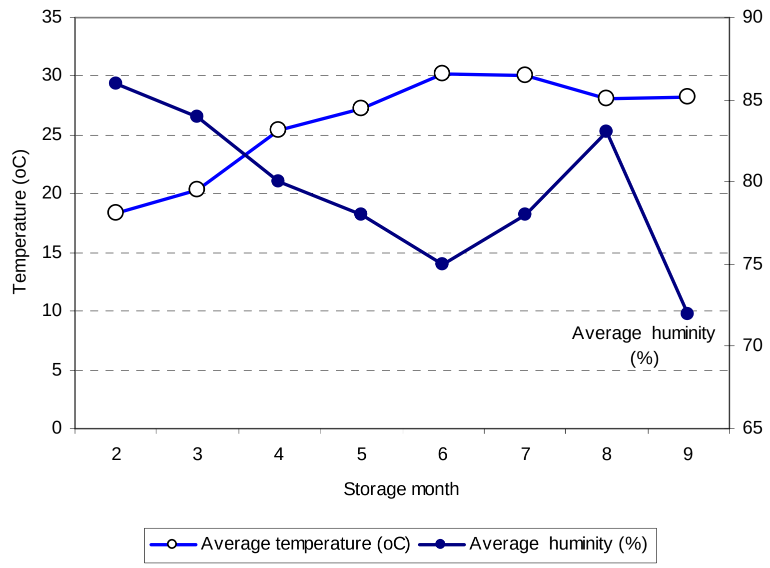

Fig. 2. Monthly humility and temperature pattern at the experiment location.

Germination test to measure seed quality evaluated the seed germination ability under optimal conditions. In this study germination rate of seed reduced along 8 months of storages in BSH.

The results of germination rate were different when using was different testing methods and substrates with the same kind of basic seed. Table 1 shows that in the first month, rice seeds have germination percentage highest on the petri dishes 99.0%, next Hiltner 98.0% and lowest in sand only 62.67%. Maize seed also showed result similarly but soybean seeds germinated highest in sandy substrate and Hiltner 95 – 97% and lowest on the Petri dishes 85.33%. In the sixth month test on the Petri dishes rice seed germination rate is 81.33%, on the sand (37.33%) and Hiltner is 90.0% respectively. For maize that is 86.33% and for soybean that is 3.33%..This result indicated that to test germination rate for different crop seeds different methods and substrates have to be used. Petri substrate is suited for rice and maize seed testing and sand substrate is suited for soybean seed testing.

Table 2. Germination rates (%) of rice, maize and soybean seed following 8 months in storage by petri, sandy substrate and Hiltner test method.

Seed	Method test	Month							
		1	2	3	4	5	6	7	8
Rice	Petri	99.00	98.33	95.33	92.33	90.67	81.33	80.33	78.67
	sandy	62.67	59.67	59.33	52.67	51.33	37.33	35.33	33.67
	Hiltner	98.00	93.67	89.33	88.33	90.00	90.00	77.67	65.33
Maize	Petri	100.00	99.67	99.00	95.00	93.67	90.33	82.67	81.00
	sandy	85.00	76.67	75.33	49.33	30.67	28.67	28.33	28.00
	Hiltner	100.00	99.67	99.33	98.00	89.67	86.33	84.33	4.00
Soybean	Petri	85.33	83.67	82.67	36.00	15.33	3.33	0	0
	sandy	97.33	94.67	92.33	50.67	48.33	11.33	0	0
	Hiltner	95.33	91.67	84.33	37.67	25.33	9.33	0	0

The Hiltner method could be most suitable substrate for germination test of rice, maize and soybean seed because firstly all three kinds of seed well germinated on this substrate during 8 months; secondly this substrate may be related to the field conditions.

The germination ability deterioration is different for three kinds of seed as soybean seeds deteriorated strongest. Soybean seeds reduced germination very fast from the third storage month and loss completely germination after the seventh month in BSH. Maize and rice seed were degraded from basic seed down to certified seed of the Vietnam standard of the crop seed classes for rice and maize of the germination rate factor. Quality standard of crop seed of the Vietnam (TCVN-1766:2004) (MARD, 2005), germination percentage of basic rice seed must be not less than 80 % and quality standard of crop seed of MARD (10 TCN 312:2003), seed standard of open pollinated maize with germination percentage must be not less than 85 % in normal bag and soybean seed not less than 70 %. So that rice seed after 8 months, maize seed after 7 months and soybean after 4 months of storage lost grade of basic seed compared to Vietnam standard of crop seed.

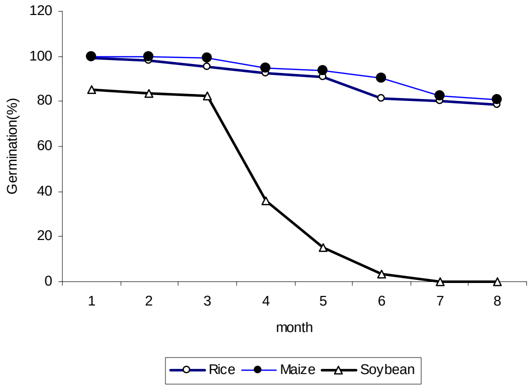

Fig. 3. Germination rate of three kinds of rice, maize and soybean seed on the Petri dishes during storage time.

For the same crop seeds, germination rates were different when using different methods of testing. The fig. 4 is illustrated that for soybean seeds as using the sand substrate the percentage of germination is 97.33%, using petri dishes the rate is 85.33% in first month storage. The rates of germination are the same after 8 months independently of which method of testing to be used.

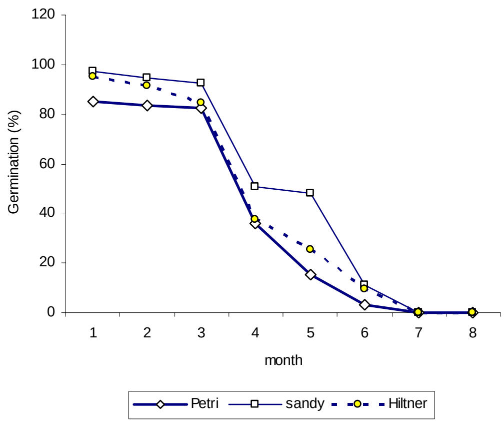

Fig. 4. Germination rate of the soybean seed on the three substrates (petri dishes, sandy and hiltner) in different storage times.

The assessment of the seed vigor and viability was conducted by two methods TZ test and seedling growth rate test. The TZ test is used throughout in the world to estimate seed viability, germination and vigor because its result can be extremely valuable and rapidly tested. This study was carried out with two grain cutting procedures that are cut laterally *(Cla)* and cut longitudinally *(Clo)*. The purpose was to identify seed viability following storage time and differences between two grain cutting procedures of rice, maize and soybean. Because cutting procedure could be influenced by TZ penetration and staining of test seed so that can be influenced to testing results.

Table 2. The seed vigor by TZ testing (% vigor seed).

Seed	Cutting method	Month							
		1	2	3	4	5	6	7	8
Rice	Cla	100.00	99.33	97.00	95.67	94.67	94.67	92.33	92.00
	Clo	98.00	98.00	95.67	92.33	91.00	91.67	88.33	87.67
Maize	Cla	100.00	99.67	99.00	96.33	93.33	93.33	91.67	91.33
	Clo	97.67	97.33	95.67	94.33	90.33	90.33	89.33	89.33
Soybean	Cla	100.00	97.33	96.00	91.33	88.33	77.00	52.67	37.67
	Clo	97.00	97.00	95.67	89.67	81.33	65.67	49.33	15.67

Notice: cut laterally (Cla) and cut longitudinally (Clo)

The seed viability and vigor were estimated by number seed stained and changed into red formazan color at normal stain (entire seed evenly stained, slight damage to root tip acceptable and slight damage to cotyledons). The strongest degradation vigor value is of soybean seed from 97.0% down to 15.67% if cut longitudinally, leaving seed intact at top of cotyledons(Clo), the method of the cut laterally and remove distal end of cotyledons(Cla) showed vigor seed percentage higher than that of Clo.

The same method cut grain trends reducing vigor of the seed shows in the Fig. 5. The soybean seed lost vigor from the third month of storage and nearly lost completely vigor at eighth month. Although rice and maize seed are degraded, the viability percentage is still over 87% (Mew *et al*., 2005).

The results evaluated by TZ test are very closes with results of the germination evaluation as illustrates in Fig. 6, that mean possibly to use TZ test to evaluate germination capacity of rice, maize and soybean seed with high accuracy.

For vigorous seeds it is possible to efficiently synthesize new biochemicals and rapidly transfer these new products to the emerging embryonic axis, resulting in increased dry weight accumulation (Copeland McDonald, 1995). Seed sown on the petri dishes, each kind of crop seed will collect 100 seedlings to dry at 80^{o}C degree during 24 hours then weighted by precision scales, dry weight of 100 seedlings (Table 4).

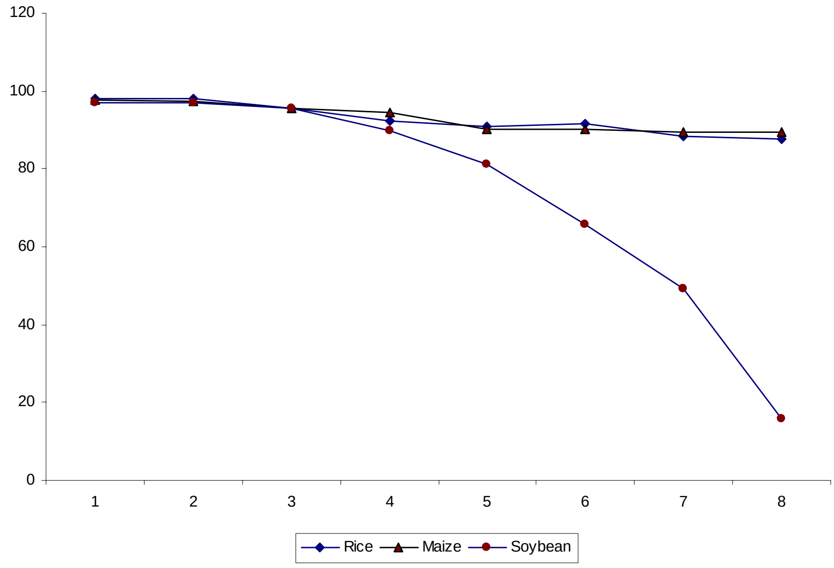

Fig. 5. Degrading of seed vigor through storage months.

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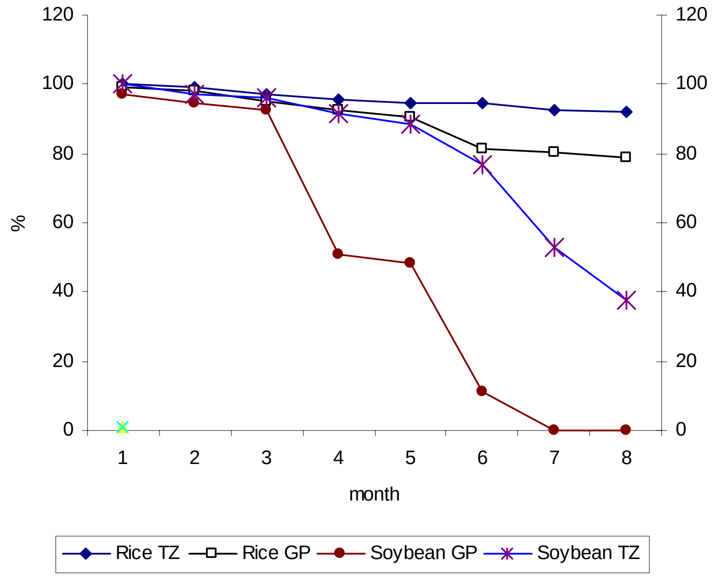

Notice: GP, germination percentage

Fig. 6. Comparison of the TZ and germination test result with rice and soybean seed.

Table 4. Seedling dry weight by seedling grown rate test method (g).

Seed	Month							
	1	2	3	4	5	6	7	8
Rice	3.05	2.58	2.15	1.82	1.46	1.74	1.30	1.25
Maize	27.03	26.76	25.85	17.46	16.23	15.53	13.65	13.27
Soybean	16.28	13.13	12.72	6.96	3.96	1.62	0	0

Dry seedling weight is also reduced through storage months and soybean reduced fastest and no more seedlings at seventh month. This test is also necessary in Vietnam because seedlings are too small, damaged or diseased not suitable for planting although they have high germination percentage. This result is in agreement to that of Chiu *et al*. (2003) and seedling dry weight of Vu Van Liet, Le Thi Thanh (2005).

Fungi living on seeds will cause damages to seed quality as vigor and germination capacity, seedborne infestation causes field diseases. Some damages are faster deterioration, delaying emergence. Emergence of seed, especially soybean seedlings in the field is frequently less than predicted by standard germination; one cause of this emergence failure is pathogen in the seed or in the soil. In pathogenic tests the agar was used for identification of seedborne fungi with two experiments one surface –disinfected by pretreating NaOCl and other entreating, the number of infected seed took in accounts of infection rate (Table 5).

Table 5. Seed health by agar plate procedure (% infected seed / total seeds).

Seed	Cutting	Month							
		1	2	3	4	5	6	7	8
Rice	TNa	0.00	0.00	0.00	0.33	0.00	0.20	0.33	0.67
	ETNa	29.67	43.33	51.00	99.00	99.33	100.00	100.00	100.00
Maize	TNa	0.00	0.00	0.00	0.33	0.33	0.00	0.67	0.67
	ETNa	30.67	44.67	49.33	98.67	99.67	100.00	100.00	100.00
Soybean	TNa	3.33	3.67	4.67	8.00	23.33	56.67	100.00	100.00
	ETNa	50.33	60.67	68.33	100.00	100.00	100.00	100.00	100.00

Notice: Pre-treat seed by NaOCl (sodium hypochlorite) (TNa) entreating (ETNa)

There is a large amount of fungi living on surface and their number increases through storage months in air - conditioned room (ACR). These fungi could be harmful for seedling when environment conditions favor their growth. Fungi-infected seed when sown on the field with favorable conditions such as high moisture and temperature will develop into damaged plants. In order to prevent fungi infection in seeds it should apply technologies in seed production, harvest, seed processing as planting in areas free from pathogen, treating fields with proper fungicide, disinfecting storage room and seeds before packing to protect seed and young seedling from many seedborne pathogens

No many fungi living inside seeds were identified in the study except for soybean seeds. In the first month only 3.33% of seed number infected, the rate of infection increased to 100% at the seventh month of storage. One explanation is that soybean seeds absorbed much humidity and created favourable condition for development of fungal spores (University of Illinois, 1998).

4. CONCLUSION

Seed moisture content through 8 months storage in BSH condition indicated that seed moisture of rice, maize and soybean increased by 2.72 to 6.07%, fastest increasing in soybean seed.

The results of seed vigor and viability evaluated by TZ test are similar to those of used germination test so that TZ test can be used to evaluate germination capacity of rice, maize and soybean seeds with high accuracy.

Germination test is very important to evaluate seed quality. For different crops different methods and substrates have to be used in seed germination test. Petri substrate is suited for testing rice and maize seeds and sand is suited for testing soybean seed. The Hiltner method could be the most suitable for germination test of rice, maize and soybean seed because firstly seeds of all all three crops well germinated on this substrate during 8 months; secondly this substrate may be related to field conditions.

Seedling growth rate test used to estimate vigor and viability is meaningful because seedling which are too small, damaged or infected with diseases should not be used for planting although seeds have high germination percentage.

Seed health test indicated that there is a large amount of fungi living on seed surface and increases through storage months in BSH. There is not many fungi living inside seeds except in soybean seeds. Treating seeds with proper fungicides before packing and storage to protect seed and young seedling from many seedborne pathogens is necessary.

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