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Overcoming seed germination problems of traditional vegetables after cold storage

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Introduction

Fruit and vegetables are major sources of essential nutrients and, consequently, are important components of diverse diets. This has led health experts to recommend that people eat at least 400 grams of fruit and vegetables a day (WHO/FAO, 2005). Unlike tomato, cabbage and other global vegetables, traditional vegetables—predominantly grown by family farms—are often characterized as nutrient-dense with high nutritional value (Yang and Keding, 2009; Hughes and Ebert, 2013). Growing and including traditional vegetables in the diet not only combats malnutrition, but also generates income for small-scale farmers and improves resilience of their production systems (Ebert, 2014). Despite the potential of traditional vegetables to combat malnutrition and poverty, and despite the wealth of traditional knowledge about these species, many remain underutilized due to a lack of appropriate seed production, drying and storage technologies, and availability and access to quality seed. This project aims to overcome known germination hurdles of two traditional vegetables: okra (*Abelmoschus esculentus* (L.) Moench), and water spinach (*Ipomoea aquatica* Forssk).

Okra is widely cultivated under tropical and subtropical conditions and in warm temperate regions. It is a popular crop in West Africa, Brazil, India, the Philippines and Thailand (Siemonsma and Kouamé, 2004). Although the longevity of okra seeds is improved after drying and storage of seed at sub-zero temperatures (Doijode, 2001), okra germplasm often has poor field establishment, which may be caused by hardseededness, a condition that prevents seed from absorbing water due to an impermeable seed coat; without moisture, the seed fails to germinate. Genotype, position of pods in the plant, time of pod harvest, seed moisture content and micronutrient applications have been reported to impact okra seed germination (Purquerio et al., 2010; El Balla et al., 2011; Mohammadi et al., 2012). Studies are needed to determine the optimum seed moisture content for long-term storage without affecting seed germination and to define seed priming treatments to overcome hardseededness.

Water spinach or kangkong, a member of the Convolvulaceae, originated in tropical Asia and is an important leafy vegetable in South and Southeast Asia, but is also known in Oceania, tropical Africa, and South and Central America (Westphal, 1994). Germination rates are said to vary with seed coat color, being highest in the black-seeded types (Westphal, 1994). Hardseededness also varies with genotype and might be influenced by the length of time between cutting the plants and threshing. Germination rates of water spinach are often low (<60%) due to hardseededness induced by long storage periods (Grubben, 2004). Induced hardseededness is more pronounced in genebanks as seed is considerably drier to extend storage life compared with seed dealt with in commercial trade (Ellis et al., 1985).

Material and Methods

One accession each of water spinach originating from Thailand (VI050476) and Taiwan (VI054533) were sown in September 2012 and harvested in March 2013. Similarly, one okra accession each from Thailand (VI046536) and Zambia (VI50958) were sown in March 2013 and harvested in June. Freshly harvested water spinach fruits were dried for two weeks in a screenhouse, then crushed, followed by manual seed extraction. Cleaned seed of okra and water spinach was dried for 3 and 8 days, respectively, in the Genetic Resources and Seed Unit (GRSU) dehumidified drying room at 18 ^{o}C and 15% RH to 6% seed moisture content (SMC). Harvested okra fruits were also dried in the GRSU screenhouse for two weeks during July 2013, followed by manual seed extraction and cleaning and further drying as described for water spinach. Once the target SMC of 6% was reached, the germination rate of water spinach and okra seed was determined and seed was quickly packed into small size aluminum foil bags and heat-sealed. Sub-samples were stored in cold rooms operated at 5 ^{o}C (medium-term storage conditions) and -15 ^{o}C (long-term storage conditions) for a 6-month period.

At the end of the 6-month storage period, seed priming of water spinach and okra seed was conducted in August 2013 and February 2014, respectively. The following priming treatments were applied for two batches of seed: T1 - control; T2 - 24 h soaking in water at room temperature (28 ±2 ^{o}C); T3 - partial removal of seed coat followed by 24 h soaking in water; T4 - soaking in rice vinegar (≥4.5% acidity) for 2 h; T5 - soaking in KNO_3 solution (0.3%) for 1 h; T6 - 24 h soaking in water, followed by quick surface drying and then drying in a dehumidified chamber for 72 h. These treatments were chosen for ease of application at farm level. The germination rate of primed seed was determined in the laboratory with the between-paper method after incubation of seed for 3 weeks in a growth chamber (model ST3-2, Saint Tien Co. Ltd.) at 30 ^{o}C for water spinach and alternating temperatures of 20/30 ^{o}C for okra, with 8 hours light and 16 hours darkness. A second batch of primed okra and water spinach seed was sown in seedling trays with peat moss for field transplanting to evaluate stand establishment. Vigor of the plants was assessed based on a 1-5 scale (1 = very weak; 2 = weak; 3 = medium; 4 = strong; 5 = very strong) three weeks after field transplanting.

Results and Discussion

The two accessions of both crops showed a marked difference in initial germination rate prior to storage confirming observations made by El Balla et al. (2011) for okra and Westphal (1994) for water spinach on the importance of the genotype for seed germination. The water spinach accession from Thailand (VI050476) had a very low initial germination rate of 4% compared to 77% for the accession from Taiwan (VI054533; Fig. 1). Okra seeds of an accession from Thailand (VI046536) had an initial germination rate of 26% compared with 90% of the accession from Zambia (VI050598; Fig. 2). Seed priming of water spinach and okra seed was conducted after 6 months of storage at 5 ^{o}C and -15 ^{o}C. The germination rate of water spinach seed from Thailand (VI050476) remained very low at 4% and 1% after 6 months of storage at 5 ^{o}C and -15 ^{o}C, respectively (T1; control; Fig. 1). Partial removal of the seed coat followed by 24 h soaking in water (T3) elevated the germination rate substantially to 82% and 85% after 6 months of storage at 5 ^{o}C and -15 ^{o}C, respectively. The germination rate of seed from Taiwan (VI054533) increased from 77% prior to storage to 92% and 93% after 6 months of storage at 5 ^{o}C and -15 ^{o}C, respectively. Seed priming of the latter did not have any additional beneficial effect on the germination rate. Partial seed coat removal of water spinach seeds followed by 24 h soaking in water (T3) resulted in the highest germination rate under both laboratory and screenhouse conditions, and led to the highest survival rate and the most vigorous plants after a growth period of three weeks in the field (data not shown).

Storage temperature of okra seed had a major impact on the germination rate. While germination rate of seed stored for a 6-month period at 5 ^{o}C was low for the accession from Thailand (18%) and Zambia (20%), the germination rate reached 99% and 96%, respectively, when seed was stored at -15 ºC (Fig. 2). Seed priming was highly beneficial for seed stored at 5 ºC (T3), but was not required when seed was stored at sub-zero temperatures. A similar pattern was obtained when the germination tests were conducted under screenhouse conditions (data not shown). Similar to water spinach, priming with T3 resulted in the highest survival rate of okra plants under screenhouse conditions. However, vigor of plants did not statistically differ among treatments (data not shown).

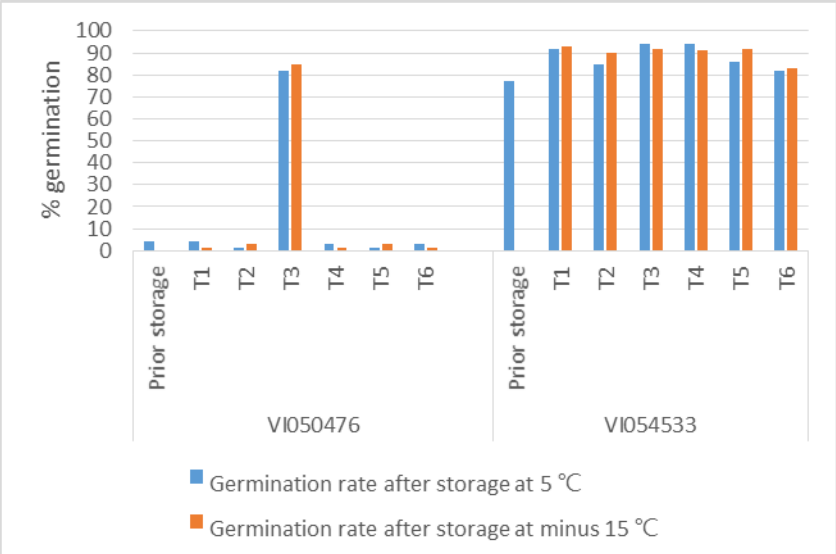

Figure 1: Germination rate (%) of seed of two water spinach accessions prior to storage and after a 6-month storage period and effect of different priming treatments: T1 = control; T2 = 24 h soaking in water; T3 = partial removal of seed coat and 24 h soaking in water; T4 = soaking in household vinegar for 2 h; T5 = soaking in KNO_3 solution (0.3%) for 1 h; T6 = 24 h soaking in water, followed by quick surface drying and then drying in a dehumidified chamber for 72 h.

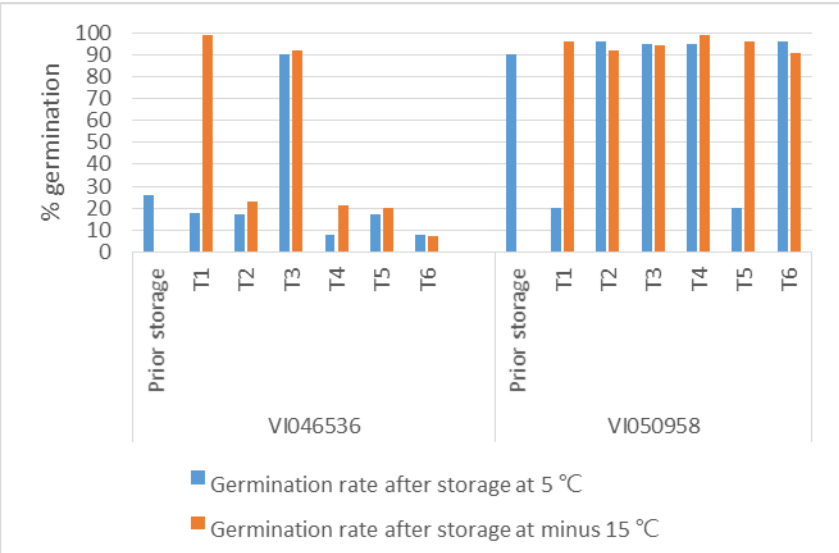

Figure 2: Germination rate (%) of seed of two okra accessions prior to storage and after a 6-month storage period and effect of different priming treatments (treatment details are given under Fig. 1).

Conclusions and Outlook

The results obtained indicate major genotypic differences in the germination rate of the genebank accessions chosen for both crops. This explains why researchers and farmers working with genebank accessions and commercial lines of these two crops often complain about poor germination and field establishment of some of the lines. In the case of seed coat imposed dormancy observed with water spinach seeds from Thailand (VI050476), partial removal of the seed coat followed by 24 h soaking in water (T3) elevated the germination rate substantially under both storage conditions as measured under laboratory and screenhouse conditions and led to the highest survival rate and the most vigorous plants in the field. Priming treatment T3 proved to be beneficial for both water spinach genotypes. Storage temperature of okra seed had a major impact on the germination rate. Storage at -15 ^{o}C boosted the germination rate markedly, making priming treatments obsolete. It remains to be seen whether the observed differences between storage conditions persist over longer storage periods.

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