Maximizing Regeneration Intervals in Plant Germplasm Collections

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Why maximize regeneration intervals?

Plant germplasm collections contain original seed collected in the field, donated by cooperators, purchased from markets or other sources, or exchanged with other genebanks. Once the seed is obtained and designated as an accession, every effort should be made to preserve the genetic variability within the accession as it was received. Seed must be regenerated to maintain adequate seed stocks for distribution and to maintain viable seed, though this is a costly operation with a risk of loss of original genetic variability. Variability can be lost by inadequate numbers of plants for regeneration, inadequate sampling to select seed for regeneration, environmental impact during seed production, uncontrolled cross-pollination, seed production differences among plants, seed mixtures during harvesting or cleaning operations, etc. The goal of plant germplasm collections should be to maximize regeneration intervals, so that accessions are regenerated as few times as possible and genetic variability is retained.

Maximize use of -18 C cold storage

Seed of species stored at -18 C have been reported to take 25 (peanut) to several hundred years (*Trifolium* campestre) to have germination reduced to 50% (Walters et al., 2005). At the USDA, ARS, Plant Genetic Resources Conservation Unit (PGRCU), Griffin, GA, freshly-regenerated seed samples are split. The bulk of seed from each accession is placed in heat-sealed foil bags in -18 C storage. Samples to handle distribution needs for several years are maintained in coffee bags at 4 C in 25% RH. These distribution samples are replenished as needed from the bulk samples maintained in -18 C. Storage of the bulk distribution samples in -18 C cold storage will reduce the need for seed regeneration.

Table 1. Accessions in -18 C cold storage

Crop	Accessions	%
Sorghum	21,203	62.2
Warm-season grass	5,056	75.9
Pepper	4,683	99.9
Mung bean	4.044	96.2
Legume	3,026	100.0
Okra	2,969	100.0
Peanut	2,600	28.3
Annual clover	2,136	100.0
Watermelon	1,871	100.0
Cucurbit	1,395	100.0
Sesame	1,213	100.0
Eggplant	993	100.0
Cowpea	834	10.4
Gourd	485	100.0
Castor bean	374	100.0
Total	**55,370**	**65.1**

Produce enough seed

Curators must plan to produce enough seed during regeneration to handle all requests that will occur during the viable life of that seed. Use past, current, and anticipated future demand to determine the seed amount needed. Past demand has shown that since 1988 from 700 (mung bean) to 12,400 (grasses) seeds are the most that have been distributed from any one accession per crop (Table 2). Specific accessions continually in high demand should have larger amounts of seed produced than accessions with little demand. Curators need to anticipate demand by continually observing the current and future research directions of their crops.

Table 2. Accessions with most distributions since 1988

Crop	Accession	Trait	Distributions No.	Distributions Seed
Watermelon	PI 189225	Fruit blotch resistant	96	2,400
	PI 595203	WMV2 resistant	96	2,400
Pepper	PI 159236	TSWV resistant	95	1,900
Sorghum	PI 550614		65	6,500
Grass	PI 196257	vetiver grass	62	12,400
Legume	PI 337098	velvet bean	59	1,475
Peanut	PI 565448	cv. Florunner	43	1,075
Cucurbit	PI 438698		38	950
Cowpea	PI 612607	CMV+BCMV resistant	35	1,750
Annual clover	PI 251563	crimson clover	33	3,300
Sesame	PI 165021		21	1,050
Okra	PI 105442	core collection	15	750
Mung bean	PI 291365	core collection	14	700

Produce high-quality seed

Initial seed quality impacts the longevity of seed maintained in cold temperature storage (Ellis and Roberts, 1980). By putting high-quality rather than low-quality seed into storage, the stored seed will remain at an acceptable level of viability longer. For many species, >65% germination in storage is acceptable viability. Regeneration of accessions with stored seed at higher levels of viability (>85%) is an unneeded expense and could result in loss of genetic variability or possible seed mixtures. Potential seed quality in regeneration plots will vary greatly with species and environment, as many wild species will never approach 100% germination. For all species regenerated since 2002 for the Griffin seed bank, 68% of regenerated accessions had >70% germination and 87% had >50% germination. Accessions with poor germination were regenerated again to obtain better quality seed.

Distribute seed sparingly

Users often want large amounts of seed to more rapidly achieve their research objectives. However, most genebanks can not afford to maintain large seed quantities for each accession. To retain adequate seed supplies for other users and to prevent the need for constant regeneration, genebank managers should provide only a representative sample of each accession requested. Distribution amounts range from 10-12 seeds per accession for difficult-to-regenerate legumes and wild peanuts to 200 seeds per accession for poor-germinating, warm-season grass species.

Table 3. Seed numbers distributed per request

Crop	No. Seed Distributed per Accession
Annual clover	100
Castor bean	25
Cowpea	50
Cucurbit	25
Eggplant	50
Gourd	25
Grass, warm-season	200
Guar	50
Hibiscus	50
Legumes	10-25
Mung bean	50
Okra	50
Pearl millet	200
Peanut, cultivated	25
Peanut, wild	12
Pepper	5-20
Sesame	50
Sorghum	100
Watermelon	25

References

Ellis, R.H., and E.H. Roberts. 1980. Annals of Botany 45:13-30.

Walters, C., L.M. Wheeler, and J.M. Grotenhuis. 2005. Seed Science Research 15:1-20.