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Long-term seed storage and inbred-lines recovering

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Maize seeds can be stored for long-term under optimal environmental conditions such as low temperature and relative humidity (Bilia et al. 1994). An inbred-line established from maize flint JD (Sementes Agroceres, Brazill) S2 population named family 41 resulted from sibling crosses of S5 progenies during 1991/1992, remained storage for 24 years. Seeds were bagged in kraft paper packages, sealed and identified (inbred-line number/year/crossing type). These packages were allocated in hermetically closed plastic boxes and stored in cold chamber at 10ºC and 40% relative humidity. To verify seed viability and to recover 41 family inbred-lines, a germination test was carried out into boxes containing *Sphagnum* under BOD at 27ºC (Table 1). Seed germination occurred in a week and 54.5% presented fungal contamination. Certainly fungi were present on the seed, once the seeds were not treated before storage. Apparently, fungi remained in a state of dormancy due not suitable environment to development. During seed germination the conditions favored fungi growth, which weakened and killed many seedlings. The surviving seedlings (Table 2) were transferred to vessel containing soil substrate and vermiculite (1: 1) and after one week acclimatization, the plantlets were transplanted to the field. Despite the high degree of homozygosis, the inbred-lines showed some phenotypic variation mainly during flowering time. Crosses type were decided for each line according to the number of plants. For the inbred-line 41122 sibling was carried out to the first ear and the second ear was self-fertilized. Inbred-lines 4124, 41234, 41233 and 41121 were self-fertilized. The inbred-line 41122 was the most precocious, emitting ears and tassels 79 and 81 days, respectively, and the flowering time occurring 89 days afterwards germination. The lineage 41233 flourished at 92 days and 41121 at 94 days afterwards germination. In the field lineage 41122 showed the highest number of surviving plants with completely normal phenotypic features (Table 2). Inbred-lines 4124, 41234, 41233 and 41121 exhibited phenotypic modifications such

as wrinkled leaves and reduced size and slow-growth. Further the recovered inbred-lines described here will be multiplied and used to cytogenetic characterization - knobs composition - and genome size determination. As observed, even going through a long period of storage, maize seeds are still viable for lines propagation and multiplication. This shows the importance of storage conditions for maintaining the longevity of maize seeds.

Table 1. Data about seed germination of line family 41.

Germination August 26, 2016		
Line number	**Seed numbers**	**Germinated seeds**
412323/ 89-1 / 1991	12	1
412321/ 89-1 / 1991-1 /1992	22	12
412421/ 89-1/ 1991	29	1
412422/ 89-1/ 1991	18	2
412331/ 89-1/ 1991	19	1
411211/ 89-1/ 1991	18	0
411211/ 89-1/ 1992	18	0
411232/ 89-1/ 91-2/ 1992	23	0
Total	159	17
Germination September 5, 2016		
41121/ 88-1/ 91-1/ 1992	24	3
41242/ 88-2/ 91-1/ 1992	13	9
41113/88-1/ 91-1/ 1992	10	9
41122/ 88-1/ 91-1/ 1992	20	17
41234/ 88-1/ 91-1/ 1992	26	24
4123/ 3-90 B-1/ 91	10	4
41233/ 1-90 B-1/ 1991	13	3
4124/ 1-90 B-1/ 1991	22	13
412321/ 89-1/ 1991	23	0
Total	161	82
	∑ = 320	∑ = 99

Table 2. Surviving lines that were transplanted into the field.

Line number	Seedling numbers in the field	Surviving seedlings
41122	16	11
4124	10	1
41234	13	1
41233	2	1
4123	3	0
41121	1	1
Total	45	15