INITIATIVE ON Genebanks

Comparative seed longevity study to customize monitoring intervals for seed viability test in *Megathyrsus maximus*

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Contents

Introduction

Seed genebanks play a vital role in conserving genetic diversity by preserving seeds for extended periods, often spanning decades or even centuries. Seed longevity is a complex trait that depends on numerous factors, and it varies among species and populations (Gianella et al., 2022). Each species exhibits a unique ageing rate that affects how long its seeds can be stored while remaining viable (Walters et al., 2005; Ellis et al., 2018; Colville and Pritchard, 2019). Understanding these ageing characteristics is essential for establishing optimal storage conditions tailored to the needs of each species (Probert et al., 2009).

Routine operations in genebanks like accession management, seed viability monitoring, and conservation are important practices aimed at enhancing operation efficiency while reducing costs. Understanding how to maximize the longevity of seeds both before and during storage as well as the relative longevity of different seed lots in storage can lead to cost savings by extending the intervals for viability monitoring and rejuvenation (Hay et al., 2021).

There has been limited research on the seed storage of many forage grasses and information about the storage behaviour of species is also insufficient (Chin and Hanson, 1999). The longevity of the forage seeds depends on the species, their initial seed viability and seed moisture content and storage temperature (https://cropgenebank.sgrp.cgiar.org/index.php/crops-mainmenu-367/forage-grasses-mainmenu-271/conservation-mainmenu-284/seed-bank-mainmenu-288). It is also clear that monitoring a large number of accessions from various species was a significant challenge when the longevity status was not known. In this study, we aimed to evaluate the potential duration of seed longevity in *Megathyrsus maximus* to determine the appropriate monitoring interval.

Methodology

Experimental procedures

In 2023, eight accessions of *Megathyrsus maximus* were selected for the study from conservation plots at the ILRI field genebank located at Zwai site. After this selection step, the plant management were well managed to ensure harvesting of high quality viable seeds. Following this, fresh seeds were harvested from each accession in May 2024. The harvested seeds were dried to the optimum moisture content in the drying room. After drying the seeds were scarified using concentrated H2SO4 for five minutes before ageing test to break dormancy from the seed. A standard protocol was used for the seed storage experiments (Hay et al., 2015: 2019; Whitehouse et al., 2015) in batches of eight accessions.

To make the seed at an equilibrium moisture level, the seeds were rehydrated using a non-saturated Lithium Chloride solution at the temperature of 20°C and relative humidity of 47% in an open Petri dish. When the seeds were at the equilibrium moisture content (ideally 14 days), the samples were labelled, put in a laminated aluminium foil pouch and transferred to the ageing box (at 45°C and 60% relative humidity). The box was sealed to avoid movement of air and the samples were drawn from the box at a given interval for the germination test. A single pouch was withdrawn at intervals of 1, 4, 8, 12, 16, 20, 24, 28, 32, 36, 40, 44, 46, 50 and 54 days. Once all the germination data had been collected, the germination percentage was plotted against the storage period for each accession, and probit analysis was conducted to determine seed longevity parameters.

Table 1. List of accessions used in the experiment for seed longevity study

Species	Acc. No.†	DOI
Megathyrsus maximus	22533	10.18730/G1FG1
Megathyrsus maximus	22543	10.18730/G1FVC
Megathyrsus maximus	22558	10.18730/G1GBW
Megathyrsus maximus	22564	10.18730/G1GJ=
Megathyrsus maximus	12784	10.18730/FRN5Z
Megathyrsus maximus	2118	10.18730/G00XV
Megathyrsus maximus	11084	10.18730/FQ0VP
Megathyrsus maximus	10619	10.18730/FPNDT

† Refers to ILRI genebank accession number.

Germination test and seedling evaluation

Germination tests were conducted using four replicates of 20 seeds each. Seeds were arranged on agar media in 90 mm diameter Petri dishes. The Petri dishes with seeds were incubated at an alternative temperature of 20/30°C with 12 hours light/12 hours dark. The germination test was conducted based on intervals. The evaluation of seedlings was done following ISTA guidelines (ISTA 2019).

Results

The initial viability of the seeds assessed across all accessions was recorded at over 80%. However, it was observed that as the duration of the ageing treatment increased, there was a corresponding decline in seed viability for each accession (Figure 1). There was a notable variation among the different accessions in terms of the duration it took for seed viability to decrease to 50%. Specifically, accession 11084 exhibited a relatively quick decline, reaching 50% of initial viability in 11.01 years. In contrast, accession 10619 showed a significantly slower decline, with viability dropping to 50% over 16.84 years (Annex 1).

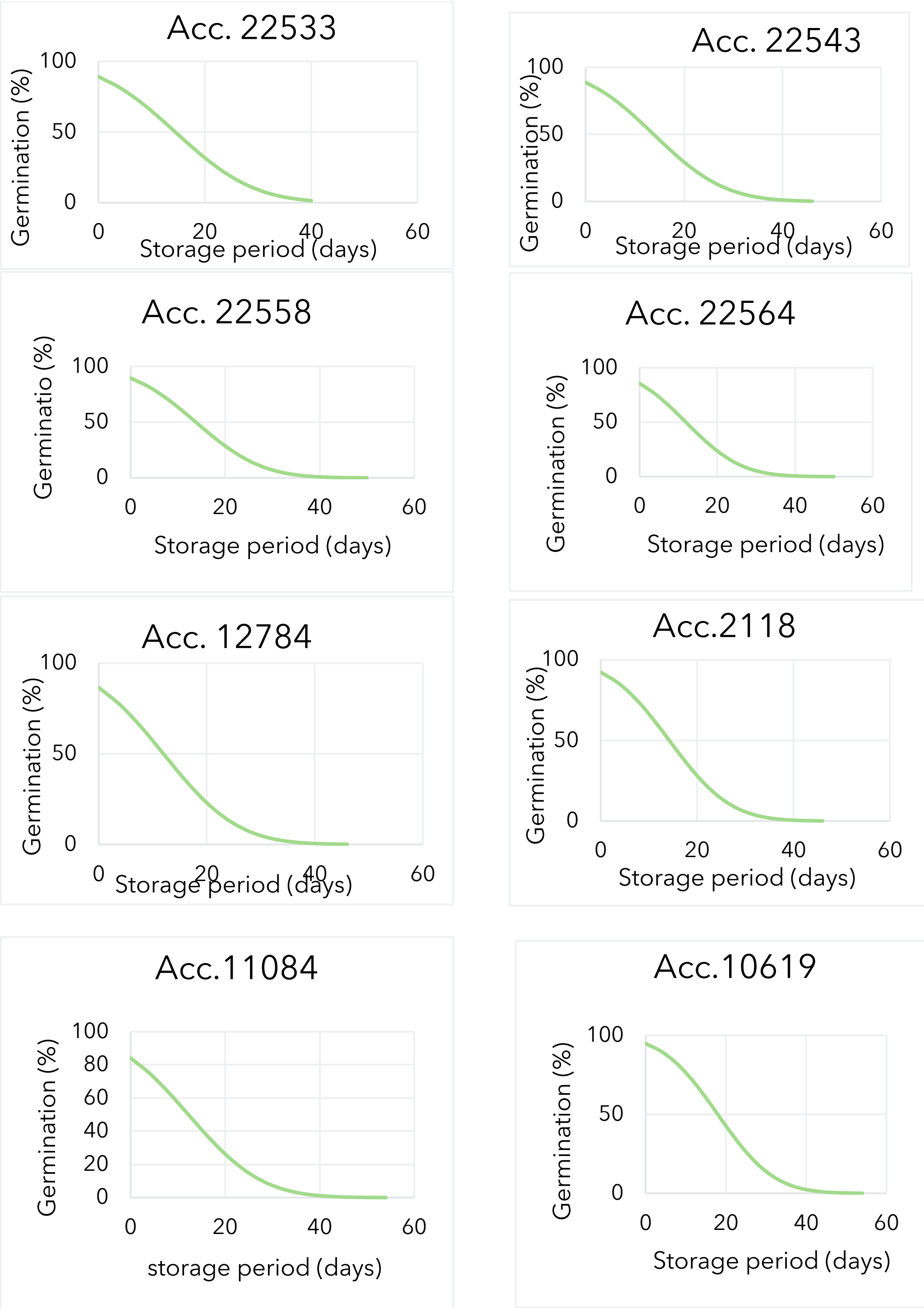

Figure 1. Survival curves fitted by probit analysis for eight accessions of *Megathyrsus maximus* seeds aged at 60% relative humidity and 45°C.

Discussion

According to FAO (2014) guidelines the viability monitoring interval was set at one-third of the time when viability declines below the threshold. In this study, variation in longevity was observed among the accessions. Justice and Bass (1978); Priestley et al. (1985) and Roberts and Ellis (1989), reported relatively little variation in the longevity of seeds from different seed lots within the same species and concluded the desire to know the full extent to which seed longevity can vary across diverse taxa. This suggests variation of tolerance level to the ageing process among the accessions. The variation in the initial viability between accessions might be varied due to the seed maturity status of each accession during harvesting.

Generally, for genebank management, it is important to know when viability will fall below the threshold to plan regeneration. Understanding how long the seeds remain viable also help to optimize monitoring intervals for better tracking of the seed quality in storages at the most critical stages of their lifecycle. The result will contribute to efficient conservation and germination strategy of seeds, providing a solid foundation for the effective management of this species.

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Annex 1. Estimate of longevity parameter for the viability to decline to 50% of the initial germination percentage for eight accessions of *Megathyrsus maximus* seeds

Accession	Ki	Estimate p50 (years)	δ	SE	Lower 95%	Upper 95%
22533	1.238	13.78	0.914	0.475	12.83	14.68
22543	1.217	13.26	0.911	0.468	12.32	14.15
22558	1.255	13.27	0.909	0.454	12.36	14.13
22564	1.059	11.62	0.910	0.487	10.64	12.54
12784	1.102	11.71	0.907	0.472	10.76	12.6
2118	1.428	13.64	0.899	0.419	12.81	14.45
11084	1.000	11.01	0.918	0.504	9.99	11.96
10619	1.622	16.84	0.909	0.435	15.98	17.67

Ki = Initial viability (probit), δ = is the standard deviation of the distribution of deaths in time, SE = standard error.

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