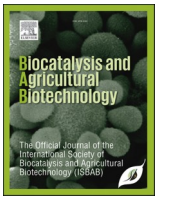
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Germination response of South African cannabis landraces to accelerated aging: Implications for seed storage

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ABSTRACT

This study investigated the impact of accelerated aging on seed germination in five South African cannabis landraces: Ladysmith Ugwayi wesiZulu ('L1') and Iswazi ('L2'), Bergville Natal ('B2') and Ugwayi wesiZulu ('B1'), and Msinga Ugwayi wesiZulu (M1). Seeds were subjected to aging at 42 °C for 0 (control), 24, 48, 72, 96, and 120 h. Seed viability assessed using a tetrazolium chloride (TTC) test, electrolyte conductivity, pH, and water activity were measured over five days. Germination tests were conducted at 30/25 °C in Petri dishes in the laboratory and validated in a greenhouse, with daily recording of germination percentage (GP), germination rate index (GRI), and coefficient of velocity of germination (CVG). Accelerated aging significantly ($p < 0.05$) reduced seed viability, GP, GRI, and CVG while increasing electrolyte leakage and water activity. Landrace responses varied with 'B1' having maintained the highest viability (100 % unaged, 94.41 % after 120 h), whereas 'M1' showed the lowest (70.63 % after aging). Electrolyte conductivity was highest in 'M1' (393.2 μS cm^{-1} g^{-1} after 120 h), while 'L2' exhibited the highest water activity (0.724 after 120 h). Germination declined significantly under prolonged aging, with 'L2' and 'B2' exhibiting over 50 % reductions in GP. The findings demonstrate that accelerated aging impairs seed quality through electrolyte leakage and moisture uptake demonstrated by water activity, with landrace-dependent sensitivity. These results emphasize the need for controlled storage to preserve cannabis seed viability and germination performance, particularly under conditions of elevated temperature and humidity common to many smallholder farming systems.

1. Introduction

Seed aging is a progressive phenomenon characterized by various physical, physiological, and biochemical alterations that ultimately compromise viability and lead to deterioration (Lakshmi et al., 2021). This decline is exacerbated by unfavorable environmental factors encountered both during production and storage, with lipid peroxidation playing a major role in reducing seed vigour

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and quality (Zheng and Ma, 2014; Kandpal et al., 2016; Feng et al., 2018). Several factors, including genetic makeup, mechanical injuries, moisture and oil levels, temperature fluctuations, humidity, pests, and oxygen availability, contribute to the deterioration rate. Among these, temperature and relative humidity are particularly significant (Kapilan and Thiagarajah, 2015; Bijanzadeh et al., 2017; Lakshmi et al., 2021).

A major driver of seed deterioration is oxidative stress, which compromises cellular integrity by damaging membranes, breaking down proteins, and degrading nucleic acids (Zheng and Ma, 2014). This metabolic decline is gradual yet irreversible, posing challenges to seedling establishment and overall agricultural productivity (Bijanzadeh et al., 2017; Nazari et al., 2020). To mitigate this, appropriate storage conditions, particularly those regulating temperature and humidity, are essential for extending seed longevity (Mohammadi et al., 2011; Feng et al., 2018). Given the slow nature of natural seed aging, accelerated aging tests serve as an effective method to simulate prolonged storage effects in a shorter period (Feng et al., 2018).

Accelerated aging is known to impair genetic stability, diminish vigour, and hinder early plant growth in various crop species (Hussein et al., 2012). It is widely used to evaluate storage viability and to identify resilient cultivars adapted to regions with high temperatures and humidity (de Lima et al., 2019). While these effects are well-documented in many crops, their specific impact on *Cannabis sativa*, particularly traditional landraces, remains poorly understood. This presents a significant research gap, especially in resource-constrained areas like South Africa, where suboptimal storage conditions can accelerate seed deterioration and compromise future cultivation (Mohammadi et al., 2011).

Seeds with high oil content, such as cannabis, are particularly prone to deterioration due to lipid instability (Suriyong et al., 2015). Cannabis seeds typically contain 25–35 % oil and 20–25 % protein, making them susceptible to degradation, much like sunflower, peanut, and soybean seeds (Suriyong et al., 2015). Under uncontrolled storage conditions, hemp seed viability can drop to 70–80 % within two years (Small and Brookes, 2012). Despite this, there is a significant gap in research on the aging process of cannabis seeds,



Fig. 1. The geographic origins of the five collected cannabis seeds.

particularly in South African landraces.

South Africa ranks among the leading cannabis producers around the world, with small-scale farming concentrated in the Eastern Cape and KwaZulu-Natal (Gastrow, 2003; Peltzer and Ramlagan, 2007). These communities typically lack access to controlled storage facilities, exposing seeds, particularly oil-rich cannabis, to rapid deterioration. Additionally, recent legalization enabling cannabis cultivation for medicinal and industrial use (Citti et al., 2018; Gouws, 2017), have increased the demand for viable, high-quality seeds.

This study explores how accelerated aging affects the germination performance of local cannabis landraces, with a focus on identifying genotype-specific deterioration patterns and key storage thresholds. It seeks to offer a foundation for future efforts, such as the development of storage protocols by agricultural extension services, while also supporting the preservation of indigenous genetic diversity, a valuable asset for climate-resilient breeding initiatives.

2. Methods and materials

2.1. Landrace collection

This study utilized seeds from five distinct cannabis landraces, obtained from various cannabis-growing regions in KwaZulu-Natal, as demonstrated in Fig. 1. To maintain consistency in naming and facilitate standardization, each landrace was assigned a pseudonym: Ladysmith Ugwayi wesiZulu ('L1') and Iswazi ('L2'), Bergville Natal ('B2') and Ugwayi wesiZulu('B1'), and Msinga Ugwayi wesiZulu ('M1').

The seed samples were stored at ambient temperature in the Horticultural Sciences Laboratory of the University of KwaZulu-Natal, Pietermaritzburg in South Africa (latitude: 29.636311 and longitude: 30.409060). Dark-coloured seeds, indicative of maturity, were selected for the experiment, while greenish seeds, linked to immaturity (Miraji et al., 2021), were excluded. Additionally, any seeds with visible defects were excluded from the seed batch.

2.2. Experimental design

The study was initially conducted in a laboratory and later validated in a greenhouse. Simulated seed aging experiments followed a factorial completely randomized design with five cannabis landraces and six aging durations (Langa et al., 2024b). Each experiment was replicated three times, using 90 Petri dishes (laboratory) and polystyrene trays (greenhouse) (Langa et al., 2024a). Seeds were subjected to accelerated aging (AA) for 0 (AA0: non-aged control), 24, 48, 72, 96, or 120 h (AA24-AA120), following procedures by Mandizvo and Odindo (2019). Seeds were placed on screen trays inside inverted plastic boxes (11 × 11 × 3.5 cm) containing 100 mL saturated NaCl solution beneath a copper mesh tray (10 × 10 × 3 cm). The boxes were sealed and incubated in a germination chamber (Labcon L.T.I.E., South Africa) at 42 ± 0.3 °C.

2.3. Seed viability test

The method followed was according to the approach outlined by Mandizvo and Odindo (2019), Langa et al. (2024a, 2024b) and involved testing seeds using 2,3,5-triphenyltetrazolium chloride (TTC) (Sigma Aldrich, Merck KGaA, 64271 Darmstadt, Germany). TTC is a fast and reliable method for assessing seed viability, producing a colour change in living tissues through dehydrogenase enzyme activity (Prudente and Paiva, 2018; Virgens et al., 2019). The percentage of viable seeds was calculated by dividing the number of viable seeds by the total number of seeds tested and then multiplying the result by 100.

2.4. Electrical conductivity and pH test

Following procedures by Mandizvo and Odindo (2019), 1 g of aged seed from each landrace (aged for 24, 48, 72, 96, or 120 h) was soaked in 10 mL of distilled water; unaged seed served as the control. Electrolyte conductivity (EC) and pH were measured initially and after 2, 12, and 24 h using a waterproof pH/EC meter (HANNA Instruments). Readings were taken in triplicate, with the probe rinsed between measurements to prevent cross-contamination. The electrical conductivity was calculated by dividing the obtained value by seed mass and expressed as $\mu S\ cm^{-1}\ g^{-1}$.

2.5. Water activity test

Water activity (a_w) was measured in seeds subjected to accelerated aging for 0, 24, 48, 72, 96, and 120 h using an AquaLAB CX-2 water activity meter (Decagon Devices Inc., Pullman, WA, USA). This non-destructive method provides a rapid and accurate measure of moisture content in hygroscopic materials, including orthodox seeds, due to its close relationship with water potential (φ) (Baldet et al., 2009). It reflects the equilibrium relative humidity (ERH) at which the seed's moisture stabilizes (Kavak and Eser, 2009), with a_w calculated by dividing ERH by 100. Readings were taken three times per sample.

2.6. Germination test

Following Langa et al. (2024a), germination tests used 30 seeds per cannabis landrace in Petri dishes (laboratory) and compost-filled trays (greenhouse). Daily germination counts over 5 days (laboratory) and 14 days (greenhouse) were used to calculate

germination percentage (GP,%), germination rate index (GRI, seeds/day), and coefficient of velocity of germination (CVG,%) following Khalaki et al. (2019), Awasthi et al. (2016), and Sobarzo-Bernal et al. (2021), respectively.

2.7. Data analysis

The data were subjected to analysis of variance (ANOVA) using GenStat®, 20.1 Edition (VSN International, Hamel Hempstead, UK, 2020) at the 5 % significance level. Tukey's test was used to separate the means of significantly different variables (Langa et al., 2024a).

3. Results

3.1. Seed viability

Seed viability varied significantly among landraces and treatments ($p < 0.05$), while their interaction was not significant. The results revealed that seed viability decreased with accelerated aging (Fig. 2). Seeds of 'B1' exhibited the highest seed viability, with means of 100 % (control) and 94.41 % (aged), compared to the other landraces. Meanwhile, the unaged seeds of 'L1' and 'L2' exhibited the lowest mean for seed viability at 92.22 %, respectively. The aged seeds of 'M1' also showed the least seed viability mean of 70.63 %.

3.2. Electrical conductivity of seed leachate and pH

The landraces, treatments, and their interaction demonstrated significant differences ($p < 0.001$) in pH and electrolyte conductivity in cannabis seeds. The results revealed that the EC showed increased electrolyte leakage with seed aging in all landraces (Fig. 3). 'M1' had the lowest EC amongst the landraces of 172.8 and 201.4 μS cm^{-1} g^{-1} under control and AA24, respectively. Landrace 'B2' had the lowest EC of 216.6, 226.2, 237.7, and 258.3 μS cm^{-1} g^{-1} under AA48, AA72, AA96, and AA120, respectively. Meanwhile, 'B1' had the highest EC of 252.4, 253.6, and 256.1 μS cm^{-1} g^{-1} under control, AA24, and AA48, respectively. 'M1' had the lowest EC at shorter aging durations but showed the highest values at extended periods, recording 309.5, 358.0, and 393.2 μS cm^{-1} g^{-1} under AA72, AA96, and AA120, respectively.

The results revealed that pH decreased with seed aging in all landraces (Fig. 4). Generally, 'M1' recorded the lowest pH irrespective of the accelerated aging period compared to other landraces, whereas 'B1' recorded the highest pH.

3.3. Seed water activity

The landraces, treatments, and their interaction demonstrated significant differences ($p < 0.001$) in water activity (aw) in cannabis seeds. The results revealed an increasing trend in water activity of cannabis landraces with the increasing accelerated aging period (Fig. 5). 'L2' exhibited the lowest water activity of 0.561 under control compared to the other landraces, and 'B1' had the highest mean of 0.638. 'L1' showed the lowest water activity of 0.590, 0.615, 0.646, 0.680, and 0.692 under AA24, AA48, AA72, AA96, and AA120, respectively. Meanwhile, 'B1' recorded the highest water activity of 0.684, 0.693, and 0.699 under AA24, AA48, and AA72, respectively. Landrace 'L2' also exhibited the highest water activity of 0.720 and 0.724 under AA96 and AA120, respectively.

3.4. Germination indices

Under both laboratory and greenhouse conditions, germination percentage (GP), germination rate index (GRI), and coefficient of

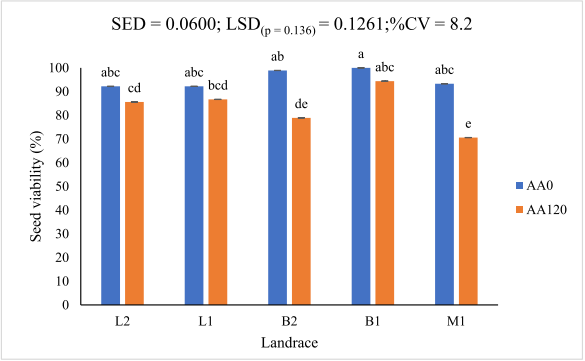


Fig. 2. Seed viability among different cannabis landraces ($p = 0.136$) in response to accelerated aging treatments (AA0: control, non-aged seeds; AA24-AA120: seeds aged for 24–120 h; number of replicates = 3). Columns sharing the same letter are not significantly different (Tukey's test). SED: standard error of differences; LSD 5 %: least significant difference at 5 % level; % CV: coefficient of variation (Langa et al., 2024b).

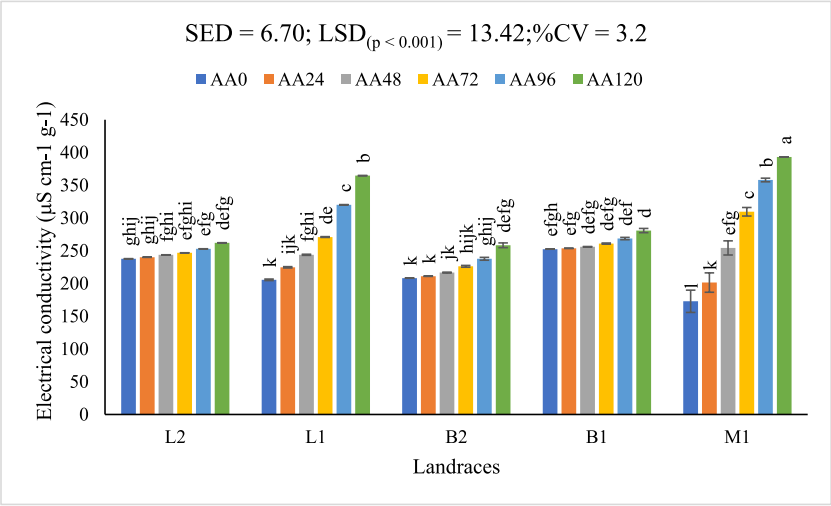


Fig. 3. Electrical conductivity among different cannabis landraces ($p < 0.001$) in response to various accelerated aging treatments (AA0: control, non-aged seeds; AA24-AA120: seeds aged for 24–120 h; number of replicates = 3). Columns sharing the same letter are not significantly different (Tukey's test). SED: standard error of differences; LSD 5 %: least significant difference at 5 % level; % CV: coefficient of variation (Langa et al., 2024b).

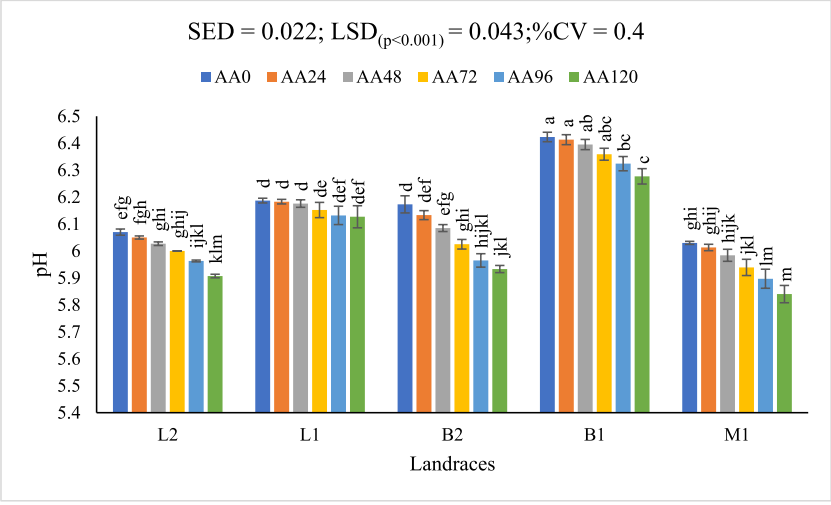


Fig. 4. The pH of different cannabis landraces ($p < 0.001$) in response to various accelerated aging treatments (AA0: control, non-aged seeds; AA24–AA120: seeds aged for 24–120 h; number of replicates = 3). Columns sharing the same letter are not significantly different (Tukey's test). SED: standard error of differences; LSD 5 %: least significant difference at 5 % level; % CV: coefficient of variation (Langa et al., 2024b).

velocity of germination (CVG) exhibited significant variations among landraces and treatments ($p < 0.001$). In the laboratory, landrace-treatment interactions significantly influenced GRI and CVG ($p < 0.05$) but had no significant effect on GP ($p > 0.05$). Conversely, in the greenhouse, these interactions had a significant impact on GP and GRI ($p < 0.05$), whereas no significant effect on CVG ($p > 0.05$) was observed.

3.4.1. Germination

The results revealed a significant reduction in cannabis landrace seed germination with increasing accelerated aging duration under both laboratory and greenhouse conditions (Fig. 6). Under laboratory conditions, 'L2' consistently exhibited the highest germination percentage among the landraces, while 'M1' had the lowest, regardless of aging duration. A 50 % reduction in germination was recorded in 'L2' and 'B2' after AA120, with 'L2' declining from 82.22 % (control) to 32.22 % and 'B1' from 75.56 % to 24.83 %.

Under greenhouse setting, 'B2' and 'L2' exhibited a general decline in germination with increasing aging duration, although 'L2' showed a slight increase at AA48 (67.78 %) compared to AA24 (64.44 %). Similarly, 'L1' showed a declining trend but had higher

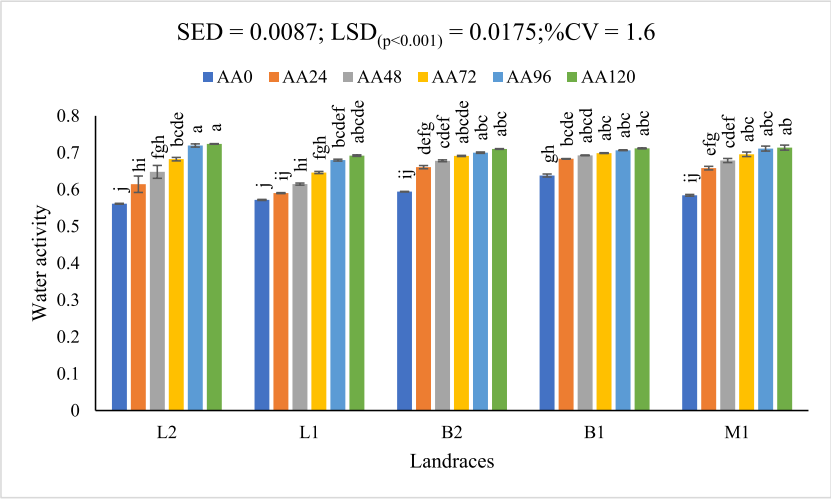


Fig. 5. The water activity of various cannabis landraces ($p < 0.001$) under different accelerated aging treatments (AA0: control, non-aged seeds; AA24–AA120: seeds aged for 24–120 h; number of replicates = 3). Columns sharing the same letter are not significantly different (Tukey's test). SED: standard error of differences; LSD 5 %: least significant difference at 5 % level; % CV: coefficient of variation (Langa et al., 2024b).

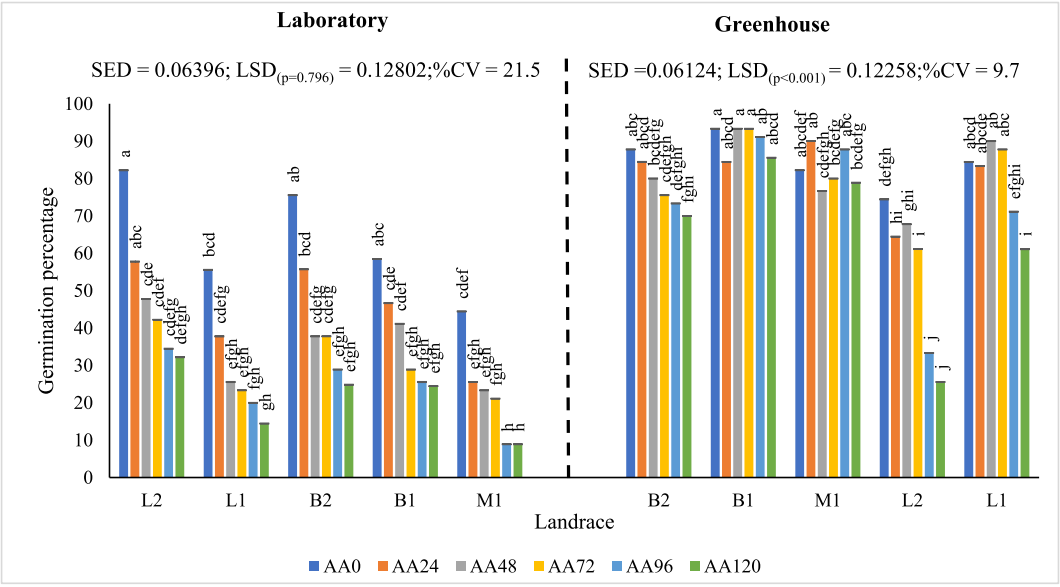


Fig. 6. The germination percentage of various cannabis landraces under laboratory ($p = 0.796$) and greenhouse ($p < 0.001$) conditions in response to different accelerated aging treatments (AA0: control, non-aged seeds; AA24–AA120: seeds aged for 24–120 h; number of replicates = 3). Columns sharing the same letter are not significantly different (Tukey's test). SED: standard error of differences; LSD 5 %: least significant difference at 5 % level; % CV: coefficient of variation (Langa et al., 2024b).

germination at AA48 (90.00 %) and AA72 (87.78 %) compared to control (84.44 %). 'B1' demonstrated overall higher germination percentages compared to other landraces, with control, AA48, and AA72 all maintaining 93.33 %, exceeding the 84.44 % recorded at AA24. In 'M1', a non-linear response was observed, with AA24 (90.00 %) and AA96 (87.78 %) showing higher germination than the control (82.22 %).

3.4.2. Germination rate index

The results showed a general decline in the germination rate index (GRI) with increasing accelerated aging duration under both laboratory and greenhouse conditions (Fig. 7). Under laboratory conditions, 'L2' consistently exhibited the highest GRI among the landraces, though it declined as aging duration increased. Conversely, 'M1' showed the lowest GRI under most treatments, except at AA48, where 'L1' recorded the lowest GRI (2.872 seeds day^{-1}). Some fluctuations were observed as 'B2' exhibited a higher GRI at

AA120 (3.717 seeds day^{-1}) than at AA96 (3.467 seeds day^{-1}), while 'M1' had a slightly lower GRI at AA96 (0.972 seeds day^{-1}) compared to AA120 (0.983 seeds day^{-1}). 'B1' recorded its lowest GRI at AA72 (3.161 seeds day^{-1}), compared to 3.644 at AA96 and 3.239 at AA120.

Under greenhouse conditions, GRI varied by landrace and aging duration but generally declined with prolonged aging. 'B2', 'M1', and 'L2' showed a consistent decrease in GRI, though they exhibited slightly higher values at AA120 compared to AA96. Specifically, GRI at AA120 for 'B2', 'M1', and 'L2' was 7.266, 6.278, and 1.625 seeds day^{-1}, respectively, compared to 6.192, 4.723, and 1.376 seeds day^{-1} at AA96. Similarly, 'L1' showed a declining trend, though it had a lower GRI at AA24 (5.121 seeds day^{-1}) compared to AA48 (7.515 seeds day^{-1}). 'B1' also exhibited fluctuations, recording a lower GRI at AA24 (12.466 seeds day^{-1}) compared to 12.815 at AA48 and 14.819 at AA72.

3.4.3. Coefficient of velocity of germination

The results demonstrated that accelerated aging reduced the coefficient of velocity of germination (CVG) under both laboratory and greenhouse conditions, with variations depending on landrace and aging duration (Fig. 8). Under laboratory conditions, CVG generally declined with increased accelerated aging, though landraces exhibited different levels of resilience. 'B2' recorded the highest CVG under control (61.70 %) and AA48 (40.85 %) compared to other landraces. 'L2' exhibited the highest CVG under AA24 (48.45 %), AA72 (39.99 %), and AA120 (42.35 %), while 'B1' had the highest CVG under AA96 (41.8 %). Conversely, 'L1' exhibited the lowest CVG under AA48 (34.27 %), AA96 (27.88 %), and AA120 (29.06 %). 'M1' also had the lowest CVG under control (40.22 %) and AA24 (37.73 %), while 'B1' recorded the lowest CVG under AA72 (31.88 %).

Under greenhouse conditions, CVG generally declined as the duration of accelerated aging increased. However, 'B2', 'M1', and 'L2' exhibited slightly higher CVG under AA120 compared to AA96, with values of 22.50 %, 17.57 %, and 17.59 % under AA120, respectively, compared to 20.20 %, 16.65 %, and 13.50 % under AA96. For 'L1', CVG declined with aging duration but showed a temporary increase at AA48 (21.29 %) compared to AA24 (18.66 %). Similarly, 'B1' recorded a higher CVG at AA72 (35.86 %) compared to AA24 (33.71 %) and AA48 (31.92 %).

4. Discussion

The current study examined how different cannabis landraces responded to various treatments for accelerated aging (AA), a simulation of seed aging procedure in a short time. The AA indicator is acknowledged as a reliable measure of seed vigour and storability since it correlates with field emergence (Kumar et al., 2019). The aging process induced the deterioration of seed viability, increased electrical conductivity of seed leachate and water activity, and decreased the potent of hydrogen of seed exudates. It also appears that accelerated aging treatments caused a significant decrease in germination characteristics in the current study under laboratory and greenhouse conditions, as further discussed in the succeeding subsections.

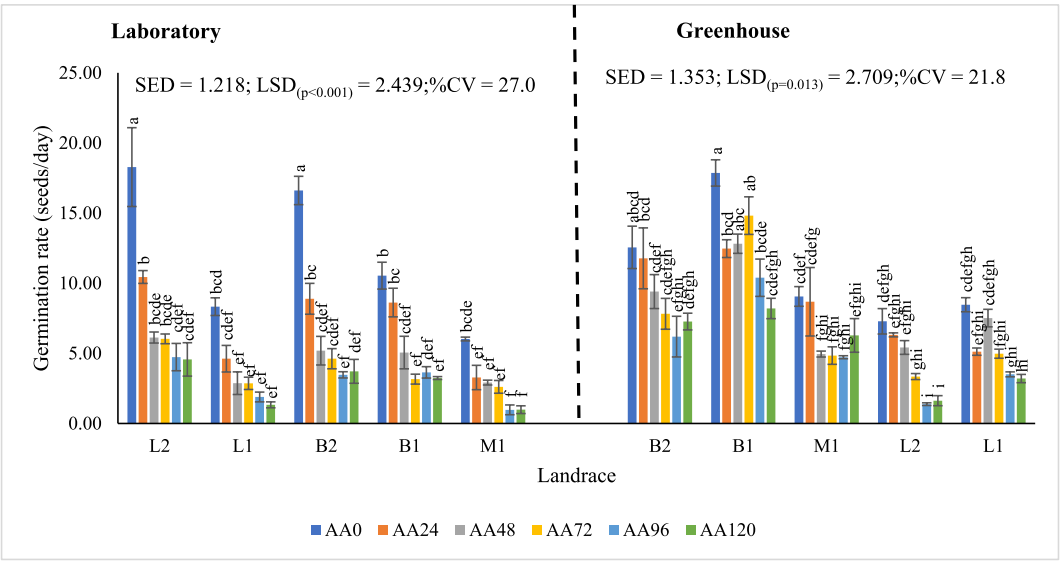


Fig. 7. The germination rate index of various cannabis landraces under laboratory ($p < 0.001$) and greenhouse ($p = 0.013$) conditions in response to different accelerated aging treatments (AA0: control, non-aged seeds; AA24-AA120: seeds aged for 24–120 h; number of replicates = 3). Columns sharing the same letter are not significantly different (Tukey's test). SED: standard error of differences; LSD 5 %: least significant difference at 5 % level; % CV: coefficient of variation (Langa et al., 2024b).

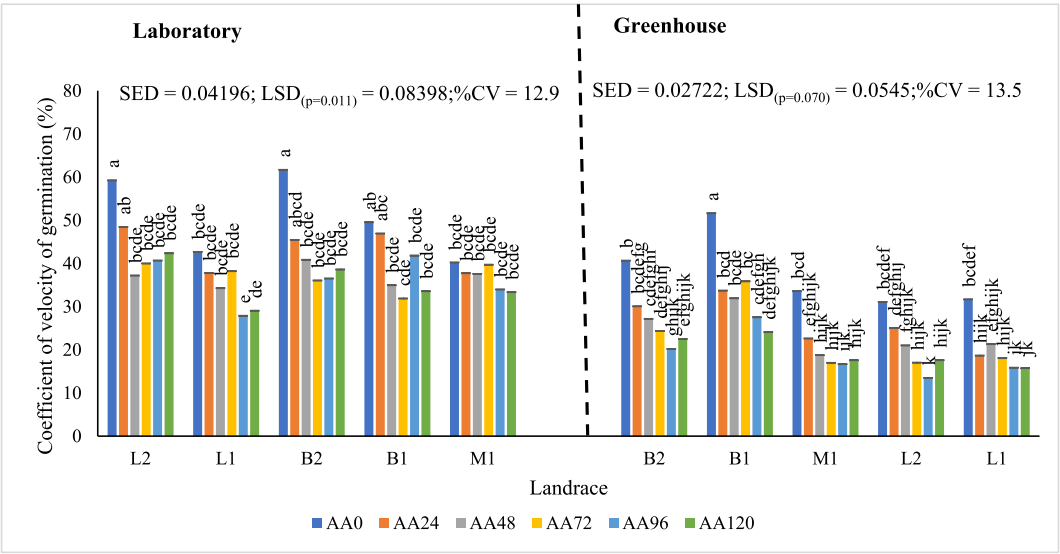


Fig. 8. The coefficient of velocity of germination of various cannabis landraces under laboratory (p = 0.011) and greenhouse (p = 0.070) conditions in response to different accelerated aging treatments (AA0: control, non-aged seeds; AA24-AA120: seeds aged for 24–120 h; number of replicates = 3). Columns sharing the same letter are not significantly different (Tukey's test). SED: standard error of differences; LSD 5 %: least significant difference at 5 % level; % CV: coefficient of variation (Langa et al., 2024b).

4.1. Seed viability

The study found that seed aging reduced viability and vigour, primarily due to intracellular changes and increased solute leakage, which impaired membrane stability and germination capacity. Increased solute leakage during imbibition indicates that the seed cannot establish coherent membranes during the rehydration of the seed, leading to a loss of viability and vigour (Barreto and Garcia, 2017; Lerin et al., 2021). Al-Maskri et al. (2003), also attributed reduced seed viability and growth to solute leakage and lipid damage in carrot seeds. Additionally, Hussein et al. (2011) found that aging weakens key enzymes responsible for converting stored nutrients, further compromising seed viability. The effects of seed aging in the current study manifested in landrace 'M1' which had the lowest seed viability, germination rate, and percentage, along with higher electrical conductivity in laboratory conditions. These findings suggest that 'M1' is particularly sensitive to aging stress, necessitating controlled storage conditions with reduced temperature and humidity to preserve seed viability. Additionally, the control treatment consistently showed better germination results than the aged seeds under both laboratory and greenhouse conditions.

4.2. Electrolyte conductivity of seed leachate and pH

Seed leaching is a reliable indicator of seed viability, as higher solute leakage correlates with increased seed deterioration (Min and Kang, 2011). The current study found that electrical conductivity of seed leachate increased with aging across all landraces, indicating declining seed quality. During rehydration, solute leakage occurs due to membrane damage, making conductivity a key marker of seed deterioration (Bijanzadeh et al., 2017). These findings align with Hussein et al. (2012), who reported a linear increase in maize seed leachate conductivity with aging, and Govindaraj et al. (2017), who found similar trends in rice seeds.

As seeds deteriorate, cell membranes lose rigidity, becoming more water-permeable, allowing sugars, organic acids, and H+ ions to escape (Singh et al., 2017; Junior et al., 2019). This acidifies the surrounding solution, lowering pH, which was a trend observed in this study. The reduction in pH correlated with increased electrical conductivity, confirming membrane damage and possibly inhibiting enzyme synthesis required for germination. This aligns with Mandizvo and Odindo (2019), who observed pH decline in aged Bambara groundnut seeds, further linking acidity to membrane deterioration. The results highlighted that seed viability loss is driven by both increased solute leakage and pH reduction, affecting germination across landraces.

4.3. Water activity

Water activity is a key indicator of seed moisture suitability for storage. A level below 0.60 prevents microbial growth, making it ideal for oilseeds prone to heating and quality loss (Caddick, 2001). In this study, all landraces except 'B2' recorded water activity below 0.60 under control conditions. The increase in water activity with accelerated aging may be linked to the breakdown of storage oil (triacylglycerol [TAG]), which supports early seedling growth by providing carbon skeletons (Kelly et al., 2011). This explained the decline in germination rates as aging progressed. These findings align with Kumar et al. (2015), who reported rising water activity in aging soybeans, and Krishnan et al. (2004), who observed a similar trend in stored wheat seeds. The increase in water activity during

accelerated aging highlighted the need for proper storage conditions to maintain cannabis landrace seed viability. This is especially relevant for landraces with low storability, which may need to be prioritized in breeding programs or managed under stricter storage conditions. Additionally, regular monitoring of water activity can provide a practical means of tracking seed quality over time, aiding decisions related to conservation, distribution, and planting.

4.4. Germination indices

Seed aging led to a decline in germination indices, including germination rate and coefficient of velocity of germination, under both laboratory and greenhouse conditions. While germination consistently decreased with aging in laboratory conditions, fluctuations were observed in the greenhouse, suggesting potential seed dormancy. Notably, landraces such as 'M1' and 'L1' exhibited higher germination in some aged treatments than in their respective controls, e.g., AA24 and AA96 for 'M1', and AA48 and AA72 for 'L1', indicating potential dormancy release. This aligns with findings that moist heat from accelerated aging can break physiological seed coat dormancy by increasing hydrogen peroxide levels (Visscher et al., 2018; Smychniuk et al., 2020). The greenhouse, which simulated the actual cannabis growing conditions, may have enhanced dormancy expression in these landraces. This finding has implications for both storage and breeding efforts. It suggests that controlled aging may help overcome dormancy in specific seed lots, improving germination performance. Furthermore, genotypes showing this response, such as 'M1' and 'L1', may have traits worth investigating in breeding programs aimed at improving seedling emergence and seed storability.

The decline in germination characteristics also correlated with increased electrical conductivity (EC) of seed leachate, indicating membrane damage. Previous research links reduced germination to mitochondrial membrane deterioration, which depletes the energy needed for germination (Hussein et al., 2012). The observed decline in germination indices aligns with previous studies. Rastegar et al. (2011) found that accelerated aging significantly reduced soybean germination. Similarly, Tabatabaei (2015) reported reductions in germination percentage, germination index, and mean germination time in barley seeds. Ghahfarokhi et al. (2014) observed that wheat seed aging affected germination percentage, seedling viability, seed biochemistry, and electrical conductivity. Thus, consistent with research on other crops, our findings highlighted that accelerated aging impacts cannabis seed germination by affecting seed reserve mobilization and biochemical integrity.

However, research on the effects of accelerated aging in *Cannabis sativa* seeds remains limited, particularly with regard to the physiological and biochemical responses of genetically diverse landraces. Most existing studies on seed aging focus on major crops such as soybean (Rastegar et al., 2011), barley (Tabatabaei, 2015), and wheat (Ghahfarokhi et al., 2014), leaving a significant knowledge gap in under-researched, yet economically and medicinally important species like cannabis. Given the growing global interest in cannabis cultivation for medicinal and industrial purposes, understanding the factors that influence seed viability, storability, and germination behavior is critical for sustainable production and effective genetic resource conservation. Our study addresses this gap by evaluating how accelerated aging influences germination traits, seed viability, and physiological responses in South African cannabis landraces, thus offering baseline data for further research.

Overall, while some landraces displayed intermittent germination resilience at specific accelerated aging intervals, none consistently maintained both strong germination performance and optimal physiological stability markers (e.g., low water activity and electrical conductivity). Notably, 'B1' had the highest seed viability (ranging from 100 to 94.41 %) and maintained high germination percentage under greenhouse conditions (93.33 % under control, AA48, and AA72), along with an elevated germination rate index of 14.819 seeds·day^{-1} at AA72, compared to 12.815 at AA48 and 12.466 at AA24. However, its water activity remained above 0.60 from control through AA72, making it less suitable for long-term storage. This highlights the challenge of identifying genotypes with both stress tolerance and long-term storage stability, a trade-off not previously documented in cannabis. The findings suggest that current germplasm lacks this ideal combination, emphasizing the need for multi-trait screening and targeted breeding strategies. Future research should explore the physiological and molecular basis of these trade-offs to support more effective selection for seed storability.

5. Conclusions

Accelerated aging significantly negatively impacted the germination performance of cannabis landraces, leading to progressive seed deterioration. Reduced seed viability, lower pH, and declining germination indices were linked to increased electrolyte leakage, indicating membrane damage. The sensitivity of cannabis seeds to elevated temperature and humidity highlighted the importance of proper storage to prevent physiological and biochemical degradation. Therefore, maintaining high seed viability under prolonged, uncontrolled storage is crucial, as effective storage delays seed aging and preserves germination potential.

Although some landraces showed intermittent germination resilience at specific accelerated aging intervals, none consistently combined strong germination performance with favourable physiological markers (e.g., low electrical conductivity and water activity). These findings emphasize the importance of multi-trait screening and the challenge of selecting genotypes suitable for long-term storage.

While this study did not examine biochemical changes during germination of aged seeds, such insights are particularly important for oil-rich crops like cannabis, which are more susceptible to deterioration due to lipid instability. Future research should explore how accelerated aging influences reserve mobilization, lipid degradation, antioxidant activity, and other biochemical processes across diverse cannabis genotypes. Ultimately, optimizing storage strategies and breeding for seed longevity are crucial for sustaining seed quality in smallholder systems and adapting to climate-related storage challenges.

CRediT authorship contribution statement

Sabeliwe Langa: Writing – review & editing, Writing – original draft, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Conceptualization. **Lembe Samukelo Magwaza:** Writing – review & editing, Supervision, Resources, Conceptualization. **Asanda Mditshwa:** Writing – review & editing, Supervision, Conceptualization. **Samson Zeray Tesfay:** Writing – review & editing, Supervision, Conceptualization.

Data availability statement

Research data is available upon request and can be requested by contacting the corresponding author of the article.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Data availability

Data will be made available on request.

References

Al-Maskri, A.Y., Khan, M.M., Khan, I.A., Al-Habsi, K., 2003. Effect of accelerated ageing on viability, vigor (RGR), lipid peroxidation and leakage in carrot (*Daucus carota* L.) seeds. Int. J. Agric. Biol. 5 (4), 580–584.

Awasthi, P., Karki, H., Bargali, K., Bargali, S.S., 2016. Germination and seedling growth of pulse crop (vigna spp.) as affected by soil salt stress. Curr. Agric. Res. J. 4 (2), 159–170.

Baldet, P., Colas, F., Bettez, M., 2009. Water activity - an efficient tool for seed testing [online] agris.fao.org. Available at: https://agris.fao.org/agris-search/search.do?recordID=FR2021095924. (Accessed 5 January 2023).

Barreto, L.C., Garcia, Q.S., 2017. Accelerated ageing and subsequent imbibition affect seed viability and the efficiency of antioxidant system in macaw palm seeds. Acta Physiol. Plant. 39 (3), 1–8.

Bijanzadeh, E., Naderi, R., Nosrati, K., Egan, T.P., 2017. Effects of accelerated ageing on germination and biochemistry of eight rice cultivars. J. Plant Nutr. 40 (2), 156–164.

Bijlakshmi, C.J., Jijeesh, C.M., Seethalakshmi, K.K., 2021. Impact of accelerated aging process on seed quality and biochemical changes of Dendrocalamus sikkimensis gamble. Acta Physiol. Plant. 43, 1–9.

Caddick, L., 2001. Water activity and equilibrium relative humidity. https://storedgrain.com.au/wp-content/uploads/2020/09/Water-activity-and-equilibrium-rh-Len-Caddick.pdf. (Accessed 20 March 2023).

Citti, C., Braghiroli, D., Vandelli, M.A., Cannazza, G., 2018. Pharmaceutical and biochemical analysis of cannabinoids: a critical review. J. Pharmaceut. Biomed. Anal. 147, 565–579.

de Lima, F.B., dos Passos, A.M.A., da Silva, J.B., Alves, C.Z., Netto, D.A.M., Cotrim, M.F., 2019. Accelerated aging in sorghum genotypes. Biosci. J. 35, 450–458. https://doi.org/10.14393/BJ-v35n2a2019-41774.

Feng, L., Zhu, S., Zhang, C., Bao, Y., Feng, X., He, Y., 2018. Identification of maize kernel vigor under different accelerated aging times using hyperspectral imaging. Molecules (Basel) 23 (12), 3078.

Gastrow, P., 2003. Mind-Blowing: the Cannabis Trade in Southern Africa. Unpublished Paper. Institute for Security Studies, Cape Town.

Ghahfarokhi, M., Ghasemi, E., Saeidi, M., Kazafi, Z., 2014. The effect of accelerated ageing on germination characteristics, seed reserve utilization and malondialdehyde content of two wheat cultivars. J. Stress Physiol. Biochem. 10, 15–23.

Gouws, J., 2017. Cultivation of cannabis and manufacture of cannabis-related pharmaceutical products for medicinal and research purposes. Med. Control Council: Appl. Guideline V1, 2017.

Govindaraj, M., Masilamani, P., Selvaraju, P., Albert, V.A., 2017. Effect of accelerated ageing on germination and seedling vigour of manually and mechanically harvested and threshed rice seeds. Int. J. Agric. Sci. Res. 7 (4), 39–48.

Hussein, H.J., Shaheed, A.I., Yasser, O.M., 2011. Effect of accelerated aging conditions on viability of sunflower (*Helianthus annus* L.) seeds. Euphrates J. Agric. Sci. 3 (3), 1–9.

Hussein, H.J., Shaheed, A.I., Yasser, O.M., 2012. Effect of accelerated aging on vigor of local maize seeds in term of electrical conductivity and relative growth rate (RGR). Iraqi J. Sci. 53 (2), 285–291.

Junior, C.A., De Menezes, F.L.G., Vitoriano, J.D.O., Da Silva, D.L.S., 2019. Effect of plasma-activated water on soaking, germination, and vigor of *Erythrina velutina* seeds. Plasma Med. 9 (2), 111–120.

Kandpal, L.M., Lohumi, S., Kim, M.S., Kang, J.S., Cho, B.K., 2016. Near-infrared hyperspectral imaging system coupled with multivariate methods to predict viability and vigor in muskmelon seeds. Sensor. Actuator. B Chem. 229, 534–544.

Kapilan, R., Thiagarajah, M., 2015. Effect of aging on the germination characteristics and enzyme activity of sunflower seeds. Int. J. Res. Innovat. Earth Sci. 2 (6), 147–150.

Kavak, S., Eser, B., 2009. Influence of polymer coatings on water uptake and germination of onion (*Allium cepa* L. cv. *aki*) seeds before and after storage. Sci. Hortic. 121 (1), 7–11.

Kelly, A.A., Quettier, A.L., Shaw, E., Eastmond, P.J., 2011. Seed storage oil mobilization is important but not essential for germination or seedling establishment in arabidopsis. Plant Physiol. 157 (2), 866–875.
Krishnan, P., Nagarajan, S., Moharir, A.V., 2004. Thermodynamic characterisation of seed deterioration during storage under accelerated ageing conditions. Biosyst. Eng. 89 (4), 425–433.
Kumar, M., Singh, B., Ahuja, S., Dahuja, A., Anand, A., 2015. Gamma radiation and magnetic field mediated delay in effect of accelerated ageing of soybean. J. Food Sci. Technol. 52, 4785–4796.
Kumar, M.V., Eevera, T., Ramesh, D., Masilamani, P., 2019. Seed deterioration in long lived sesame seeds under accelerated ageing conditions. J. Pharmacogn. Phytochem. 8 (3), 706–709.
Langa, S., Magwaza, L.S., Mditshwa, A., Tesfay, S.Z., 2024a. Seed dormancy and germination responses of cannabis landraces to various pre-treatments. South Afr. J. Bot. 165, 91–100. https://doi.org/10.1016/j.sajb.2023.12.021.
Langa, S., Magwaza, L.S., Mditshwa, A., Tesfay, S.Z., 2024b. Temperature effects on seed germination and seedling biochemical profile of cannabis landraces. Int. J. Plant Biol. 15, 1032–1053. https://doi.org/10.3390/ijpb15040073.
Lerin, J., Sousa, K.R., Vieira, H.D., Silveira, V., Santa-Catarina, C., 2021. Physiological and proteomic insights related to the loss of seed viability in *Cariniana legalis* (*Lecythidaceae*). Theoret. Exp. Plant Physiol. 33 (2), 173–186.
Mandizvo, T., Odindo, A.O., 2019. Seed coat structural and imbibitional characteristics of dark and light coloured Bambara groundnut (*Vigna subterranea* L.) landraces. Heliyon 5 (2), e01249.
Min, T.G., Kang, W.S., 2011. Simple, quick and nondestructive method for *Brassicaceae* seed viability measurement with single seed base using resazurin. Hortic. Environ. Biotechnol. 52 (3), 240–245.
Mohammadi, H., Soltani, A., Sadeghipour, H.R., Zeinali, E., 2011. Effects of seed aging on subsequent seed reserve utilization and seedling growth in soybean. Int. J. Plant Prod. 5, 1.
Nazari, R., Parsa, S., Tavakkol Afshari, R., Mahmoodi, S., Seyyedi, S.M., 2020. Salicylic acid priming before and after accelerated aging process increases seedling vigor in aged soybean seed. J. Crop Improv. 34 (2), 218–237.
Peltzer, K., Ramlagan, S., 2007. Cannabis use trends in South Africa. S. Afr. J. Psychiatr. 13 (4), 126–131.
Prudente, D.O., Paiva, R., 2018. Seed dormancy and germination: physiological considerations. J. Cell and Dev. Biol. 2 (1), 2.
Rastegar, Z., Sedghi, M., Khomari, S., 2011. Effects of accelerated aging on soybean seed germination indexes at laboratory conditions. Not. Sci. Biol. 3 (3), 126–129.
Smychniuk, A.A., Calvi, G.P., Ferraz, I.D.K., 2020. Moist heat overcomes physical dormancy at the seed coat lens in *Schizolobium parahyba* var. *amazonicum*. Floresta e Ambiente 27 (1), e20190093. https://doi.org/10.1590/2179-8087.009319.
Singh, R.K., Verma, S.S., Kumar, R., 2017. Prediction of seedling establishment in coriander (*Coriandrum sativum* L.) based on laboratory tests: prediction of seedling establishment in coriander. J. AgriSearch 4 (4), 242–246.
Sobarzo-Bernal, O., Gómez-Merino, F.C., Alcántar-González, G., Saucedo-Veloz, C., Trejo-Téllez, L.I., 2021. Biostimulant effects of cerium on seed germination and initial growth of tomato seedlings. Agronomy 11 (8), 1525.
Suriyong, S., Krittigamas, N., Pinmanee, S., Punyalue, A., Vearasilp, S., 2015. Influence of storage conditions on change of hemp seed quality. Agric. Agric. Sci. Procedia 5, 170–176.
Tabatabaei, S.A., 2015. The changes of germination characteristics and enzyme activity of barley seeds under accelerated ageing. Cercetări Agronomice în Moldova 48, 61–67.
Visscher, A.M., Yeo, M., Barreiro, P.G., Stuppy, W., Frances, A.L., Di Sacco, A., Seal, C.E., Pritchard, H.W., 2018. Dry heat exposure increases hydrogen peroxide levels and breaks physiological seed coat-imposed dormancy in *Mesembryanthemum crystallinum* (*Aizoaceae*) seeds. Environ. Exp. Bot. 155, 272–280.
Zheng, Y.L., Ma, H.C., 2014. Effects of seed aging on seed germination and seed reserve utilization in mumian. HortTechnology (Alex., Va.) 24 (4), 471–474.

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