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**Research Article**

# Amylase activity and carbohydrate accumulation in seeds of *Afzelia africana* Sm. and *Gambeya albida* (G. Don) Aubrév. & Pellegr. during desiccation

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## Abstract

**Background:** Desiccation is a critical factor influencing seed viability and storage potential. Among the physiological changes accompanying seed drying are alterations in carbohydrate reserves and the activities of carbohydrate metabolizing enzymes.

**Aims:** This study investigated changes in reducing sugar and starch content, as well as the activities of α-amylase, β-amylase, and total amylases, in the seeds of two tropical timber species, *Afzelia africana* Sm. and *Gambeya albida* (G. Don) Aubrév. & Pellegr., during progressive desiccation.

**Methods:** Mature seeds were harvested directly from parent trees and desiccated under ambient conditions for 0, 3, 7, 10, 13, 15, 22, and 35 days. Germination capacity, carbohydrate levels, and amylolytic activities were assessed at each desiccation interval.

**Results:** Germination in *A. africana* increased gradually with desiccation, whereas seeds of *G. albida* exhibited high initial germination that declined significantly ($p \leq 0.05$) following moisture loss. Reducing sugar levels were consistently higher in *G. albida* than in *A. africana*, while starch content increased in *A. africana* but declined in *G. albida* during later stages of desiccation. Activities of α-amylase, β-amylase and total amylases increased during early desiccation (0–7 days) in both species, followed by a significant ($p \leq 0.05$) decline, with *A. africana* exhibiting higher overall amylolytic activity. These results indicate contrasting carbohydrate metabolic responses to desiccation, consistent with orthodoxlike behavior in *A. africana* and recalcitrant behavior in *G. albida*.

## Özet

**Dayanak:** Kurutma, tohumların canlılığını ve depolama potansiyelini etkileyen kritik bir faktördür. Tohum kurutulmasıyla birlikte meydana gelen fizyolojik değişiklikler arasında karbonhidrat rezervlerinde ve karbonhidrat metabolize eden enzimlerin aktivitelerinde meydana gelen değişiklikler bulunmaktadır.

**Amaçlar:** Bu çalışmada, iki tropikal ağaç türü olan *Afzelia africana* Sm. ve *Gambeya albida* (G. Don) Aubrév. & Pellegr. türlerinin tohumlarında, aşamalı kurutma sırasında indirgen şeker ve nişasta içeriğindeki değişiklikler ile α-amilaz, β-amilaz ve toplam amilazların aktiviteleri incelenmiştir.

**Yöntemler:** Olgun tohumlar doğrudan ana ağaçlardan hasat edilmiş ve ortam koşullarında 0, 3, 7, 10, 13, 15, 22 ve 35 gün boyunca kurutulmuştur. Her kurutma aralığında çimlenme kapasitesi, karbonhidrat seviyeleri ve amilolitik aktiviteler değerlendirilmiştir.

**Bulgular:** *A. africana*'da çimlenme kuruma ile birlikte kademeli olarak artarken, *G. albida* tohumları yüksek bir başlangıç çimlenme oranı sergilemiş, ancak nem kaybının ardından önemli ölçüde ($p \leq$ 0,05) azalmıştır. Azaltıcı şeker seviyeleri *G. albida*'da *A. africana*'ya göre sürekli olarak daha yüksekken, nişasta içeriği *A. africana*'da artmış, ancak *G. albida*'da kurumanın ilerleyen aşamalarında azalmıştır. α-amilaz, β-amilaz ve toplam amilaz aktiviteleri her iki türde de erken kurutma aşamasında (0–7 gün) arttı, ardından önemli ($p \leq 0,05$) bir düşüş gerçekleşti, *A. africana* ise daha yüksek toplam amilolitik aktivite gösterdi. Bu sonuçlar, kurutmaya karşı zıt karbonhidrat metabolik tepkileri olduğunu göstermektedir ve bu,

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**Conclusion:** The findings provide physiological insight relevant to seed storage and reforestation strategies for tropical forest species.

*A. africana*'nın ortodoks benzeri davranışıyla ve *G. albida*'nın dirençli davranışıyla tutarlıdır.

**Sonuç:** Bulgular, tropikal orman türleri için tohum depolama ve yeniden ağaçlandırma stratejileri ile ilgili fizyolojik bilgiler sağlamaktadır.

***Keywords:*** Seed desiccation tolerance, carbohydrate metabolism, germination response, seed storage behavior

## Introduction

*Afzelia africana* Sm. and *Gambeya albida* (G. Don) Aubrév. & Pellegr. are tropical rainforest timber species. *A. africana*, commonly referred to as African mahogany, is a large deciduous woody species threatened by overexploitation and poor natural regeneration in its native habitats (Padonou et al., 2013). It is a leguminous tree of the family Fabaceae and subfamily Caesalpiniaceae, occurring in both humid and dry forest ecosystems (Umedum et al., 2014). Every part of the plant is valuable. The species produces high quality timber, which is termite resistant but somewhat difficult to work; the seeds contain approximately 31% fat and have potential as a source of oil for both domestic and industrial applications. The seeds are also ground into flour for various culinary uses. The leaves serve as forage and food, while decoctions made from the flowers and roots are traditionally used to treat various ailments. Despite its usefulness, this plant is under threat due to overexploitation, urban expansion, and the degradation of natural forests. It is classified as *Vulnerable* by the International Union for Conservation of Nature (2004). *G. albida*, commonly known as white star apple, is a forest fruit tree belonging to the family Sapotaceae. It possesses significant economic value, particularly due to findings that jams made from its pulp could rival commercial raspberry jams and jellies. Additionally, the seed oil has various applications (Amusa et al., 2003). The fruit is also a rich source of natural antioxidants, contributing to health by combating oxidative stress related diseases (Burits & Bucar, 2000). The timber of *G. albida* is very dense, moderately strong and fairly durable (Etukudo, 2003). The seeds are edible and used locally in soup thickening. The leaves are an excellent fodder and fruit pods are good fuel wood.

Seeds are essential components of plant production systems, serving both direct planting needs and, seedling production and propagation. While certain seeds can be stored without losing viability, others are highly desiccation sensitive and have limited storage potential, restricting their usage in production systems (Hay & Probert, 2013). In the case of *A. africana* and *G. albida*, that both inhabit tropical forests and produces fruits shortly before the onset of the dry season, responses to seed desiccation is important because natural seedling regeneration depends on the moisture content (MC) of the forest floor.

Although some studies have reported that seeds of *A. africana* exhibit desiccation sensitivity under specific conditions (Adelani et al., 2017), available evidence on its seed storage behavior remains inconclusive. Consequently, the present study adopts a physiological perspective, examining MC and germination relationships alongside metabolic responses during progressive drying, rather than assuming a predefined storage classification. In contrast, seeds of *G. albida* have consistently been described as desiccation sensitive, with marked declines in germination observed as seed MC decreases (Adelani et al., 2017). While the general processes regulating responses to desiccation in tropical forest species have been described, the mechanisms governing seed desiccation responses in tropical forest species have not been well documented (Daws et al., 2011). Current studies on other recalcitrant taxa show that desiccation interferes with energy metabolism, osmotic balance, leading to a breakdown of regulatory proteins (Berjak & Pammenter, 2013), while orthodox seeds accumulate protective soluble sugars and carbohydrates to maintain cellular structure (Farrant & Moore, 2011). These varying responses reveal the specific biochemical responses to desiccation and the need for a direct physiological assessment rather than generalizing unrelated taxa (Berjak & Pammenter, 2013).

Although there are suggestions that both *A. africana* and *G. albida* are sensitive to desiccation, there is no published study that has examined how carbohydrate metabolizing enzymes, in particular amylase and reserve carbohydrates (sugar and starch), change during drying in these species. This is a critical gap in knowledge because carbohydrate metabolism plays a major role in the acquisition or loss of desiccation tolerance (Berjak & Pammenter, 2013). Also, without species specific biochemical data, the physiological basis of desiccation sensitivity in these economically and ecologically important species remains unclear. Therefore, this study addresses this gap by investigating amylase activity and carbohydrate accumulation during progressive desiccation of seeds of both species. The research objectives are to quantify changes in MC during specific desiccation intervals; determine the amylase activity in response to desiccation; assess the changes in reducing sugar and starch accumulation during desiccation; and relate these changes to potential desiccation tolerance or sensitivity in both species.

## Materials and Methods

### Study Area/Experimental Site

The study was conducted in Calabar, the capital city of Cross River State, Nigeria. Laboratory analyses were performed at the Graduate Research Laboratory, Department of Plant and Ecological Studies, Faculty of Biological Sciences, University of Calabar.

### Seed Preparation/Desiccation

Mature fruits of *A. africana* and *G. albida*, were harvested directly from the tree stands within Calabar metropolis. The fruits were de-pulped to obtain their seeds. The seeds were desiccated under ambient conditions on a laboratory bench for 0, 3, 7, 10, 13, 15, 22,

and 35 days; with average daytime temperatures of approximately 28 to 32 °C, relative humidity ranging between 60 to 75%, and natural sunlight exposure filtered through the screen house. On each of these sampling dates, twenty seeds of each species were randomly picked and weighed to obtain their fresh weight. Seeds were oven dried at 103 ± 2 °C until constant weight, following standard seed moisture determination protocols (Association of Official Analytical Chemists [AOAC], 2006). The fresh and dry weights obtained were used to calculate the percentage MC of the seed lot. MC was calculated adopting Somrug et al. (2024) as

$$MC\ (\%) = \frac{\text{fresh weight} - \text{dry weight}}{\text{fresh weight}} \times 100$$

### Desiccation and Germination Tests

Forty seeds were sown in germination trays at each desiccation level to investigate their germination potentials. The seeds were steeped in water for 12 h before sowing. An equal mixture of smooth sea sand and sawdust was used as the growth medium; 2 kg of the planting medium was weighed into germination trays and laid out in a complete randomized design, replicated three times. The germination of the planted seeds was observed daily, and germination counts were recorded daily. The emergence of radicle through the seed coat was the criterion for germination. The germination records were used in computing the percentage maximum germination ($G_{max}$) after Ngele et al. (2024) as follows:

$$G_{max}\ (\%) = \frac{\text{number of germinated seeds}}{\text{total number of sown seeds}} \times 100$$

### Determination of Reducing Sugars and Starch

The seeds employed for sugar and starch extraction were dried at 60 °C for 24 hours. Sugar and starch concentration in seeds on collection and following desiccation was assayed using the dinitrosalicylic acid (DNS) method (AOAC, 2006) with 1% glucose solution as standard.

### Calibration Curve for Sugar

Serial dilutions of the 1% glucose solution were prepared to obtain glucose concentrations of 0, 2, 4, and 6 mg/mL in labeled test tubes. To each of the test tubes were added 1 mL DNS reagent and 2 mL of 1.5% NaOH, and brought to boil for five minutes. The resultant solutions were allowed to cool and the absorbance readings determined spectrophotometrically at 540 nm wavelength. The absorbance readings obtained were plotted against sugar concentration to obtain a calibration curve.

### Determination of Reducing Sugars

Reducing sugars were extracted using 80% ethanol. Precisely 0.2 g of dried cotyledonary tissue (dried at 60 °C) from seeds of each species was ground in 5 mL of 80% ethanol. The homogenate was centrifuged at 4000 rpm for 5 minutes. In a test, 0.5 mL of the resulting sugar extract was mixed with 1 mL of DNS reagent and 2 mL of 1.5% NaOH. The mixture was then heated in a water bath for 5 minutes. Reducing sugar content (µmol $g^{-1}$ dry weight) was estimated spectrophotometrically at 540 nm, and concentrations were determined using a standard calibration curve prepared from 1% glucose solution.

### Starch Determination

Starch present in the insoluble residue following sugar extraction was solubilized with 1 M NaOH and subsequently neutralized with 1 M acetic acid. The starch content (µmol $g^{-1}$ dry weight) was then determined by acid hydrolysis using 0.5 mL of 1 M HCl, followed by the colorimetric estimation of the resulting reducing sugars.

### Determination of Alpha, Beta and Total Amylolytic Activities in Seeds

### Enzyme Extraction

The extraction buffer was composed of 50 mM mixed phosphate buffer (prepared from monobasic potassium phosphate [$KH_2PO_4$], and dibasic potassium phosphate [$K_2HPO_4$] salts), containing 1% (w/v) polyvinyl polypyrrolidone (PVPP), a phenolic binding agent. The pH was adjusted to 7.0 at 30 °C (Ngele et al., 2024). To prepare the extraction buffer, 2.613 g of $K_2HPO_4$ was dissolved in 300 mL of distilled water, and 2.042 g of $KH_2PO_4$ was dissolved in another 300 mL of distilled water. The two solutions were then combined and 6 g of PVPP was added. The buffer was stored refrigerated until used. For enzyme extraction, 0.5 g of cotyledonary seed tissue from each species at specified desiccation intervals was homogenized in 5 mL of cold extraction buffer. The homogenate was first for 5 minutes, and the resulting filtrate was centrifuged again under the same conditions. The final supernatant was kept on ice and served as the crude enzyme extract.

### Enzyme Assay

Amylase activity was assessed by measuring the amount of reducing sugars released from soluble starch and expressed as µmol of glucose equivalents $g^{-1}$ dry weight produced in 10 minutes at 25 °C, using 600 µL of crude enzyme extract, following the procedure described by Nkang (2002).

To determine α-amylase activity, β-amylase was selectively inactivated by heating the crude enzyme extract at 70 °C for 30 minutes, a temperature previously reported to preferentially inactivate β-amylase while retaining α-amylase activity in plant tissues (Nkang, 2002). A reaction mixture containing 0.1 mL of 1% (w/v) soluble starch and 600 µL of the heat-treated enzyme extract was incubated at 25 °C for 10 minutes. The reaction was terminated by adding 1 mL of DNS reagent followed by 2 mL of 1.5% sodium hydroxide, and the mixture was boiled for 2 minutes. Absorbance was measured at 540 nm, and α-amylase activity was quantified using a glucose standard curve and expressed on a dry weight basis (µmol reducing sugar released $g^{-1}$ dry weight $min^{-1}$).

For β-amylase activity, α-amylase was selectively inhibited by incubating the crude enzyme extract with 25 mM ethylenediaminetetraacetic acid (EDTA) at 25 °C for 30 min, as EDTA chelates divalent cations required for α-amylase activity, thereby allowing preferential estimation of β-amylase (Nkang,

2002). A reaction mixture comprising 0.1 mL of 1% soluble starch and 600 µL of the treated enzyme preparation was incubated at 25°C for 10 min. The reaction was stopped by adding 1 mL of DNS reagent and 2 mL of 1.5% sodium hydroxide, followed by boiling for 2 min. The absorbance was read at 540 nm, and β-amylase activity was expressed as µmol reducing sugar released $g^{-1}$ dry weight $min^{-1}$.

Total amylase activity was assayed using untreated enzyme extract. A reaction mixture containing 2 mL of assay buffer, 600 µL of enzyme extract, and 0.1 mL of 1% soluble starch was incubated at 25 °C for 10 minutes. The reaction was terminated as described above, and absorbance was recorded at 540 nm. Total amylase activity was calculated from the glucose standard curve and expressed on a dry weight basis (µmol reducing sugar released $g^{-1}$ dry weight $min^{-1}$).

All enzyme assays were conducted in triplicate. While no separate inhibition controls were included, the use of heat treatment and EDTA for selective estimation of α- and β-amylase followed previously validated protocols, and the approach provides comparative rather than absolute measures of enzyme activity during desiccation.

### Statistical Analysis

All experiments were conducted using a completely randomized design. Data obtained from MC, germination percentage, carbohydrate concentrations, and amylase activities were based on three independent replicates and are presented as mean ± standard error of the mean. Statistical analyses were performed using the Statistical Package for Social Sciences for Windows, version 20.1.

Differences among desiccation intervals and between species were evaluated using one way analysis of variance (ANOVA). When ANOVA indicated significant treatment effects, mean separation was performed using Duncan's Multiple Range Test (DMRT) at a significance level of $p \leq 0.05$. DMRT was selected because of its sensitivity in detecting treatment wise differences across multiple desiccation intervals, allowing effective discrimination of gradual physiological changes associated with progressive drying. This approach is commonly applied in plant physiological and seed biology studies where treatments represent ordered stress gradients rather than independent categorical factors.

## Results

### MC and Germinability of *A. africana* and *G. albida* Seeds After Desiccation

The MC levels in seeds of *A. africana* and *G. albida* on collection and following desiccation are presented in Table 1. Seeds of *G. albida* had relatively higher MC on collection (40.04%) compared to *A. africana* (13.81%). MC declined significantly ($p \leq 0.05$) throughout the desiccation period in seeds of *G. albida*. In *A. africana*, MC showed a gradual decline but remained relatively stable during the later desiccation stages (days 15–35). Germination capacity gradually improved with desiccation, reaching its maximum at 15 days (90.00%) when MC was 4.86%.

However, a significant decline in $G_{max}$ ($p \leq 0.05$) was observed at 22 and 35 days of desiccation, coinciding with a further reduction in MC to approximately 4%. In *G. albida* $G_{max}$ was significantly higher ($p \leq 0.05$) on collection (76.67%) but declined gradually with desiccation. No germination occurred after 13 days at MC below 26.7%.

### Reducing Sugar Content in Seeds of *A. africana* and *G. albida* During Desiccation

The reducing sugar content in seeds of *A. africana* was lower than that of *G. albida* across the desiccation period (Figure 1). A significant increase ($p \leq 0.05$) in reducing sugars was observed in *A. africana*, peaking at 7 days (3.69 µmol $g^{-1}$ dry weight), after which sugar levels declined.

Overall, *G. albida* seeds maintained consistently higher reducing sugar levels, indicating species specific differences in carbohydrate behavior. Reducing sugar content increased significantly ($p \leq 0.05$) from 0 to 3 days in *G. albida*, with the highest levels at 3 days. Although sugar levels declined afterward, there was a secondary

**Table 1.** MC and $G_{max}$ of *A. africana* and *G. albida* seeds during desiccation.

| Desiccation period (days) | *A. africana* | | *G. albida* | |
|---|---|---|---|---|
| | MC (%) | $G_{max}$ (%) | MC (%) | $G_{max}$ (%) |
| 0 | $13.81^{a} \pm 0.62$ | $66.28^{bcd} \pm 0.23$ | $40.04^{a} \pm 0.34$ | $76.67^{a} \pm 3.33$ |
| 3 | $8.36^{b} \pm 0.24$ | $71.67^{abcd} \pm 4.41$ | $37.23^{b} \pm 0.18$ | $56.67^{b} \pm 3.33$ |
| 7 | $6.76^{c} \pm 0.08$ | $79.67^{ab} \pm 5.78$ | $34.77^{c} \pm 0.12$ | $15.00^{c} \pm 2.89$ |
| 10 | $6.36^{c} \pm 0.13$ | $77.67^{abc} \pm 8.88$ | $32.11^{d} \pm 0.19$ | $11.67^{c} \pm 3.33$ |
| 13 | $5.34^{d} \pm 0.15$ | $85.00^{ab} \pm 2.89$ | $27.63^{e} \pm 0.14$ | $0.00^{d} \pm 0.00$ |
| 15 | $4.86^{de} \pm 0.06$ | $90.00^{a} \pm 5.77$ | $25.56^{f} \pm 0.30$ | $0.00^{d} \pm 0.00$ |
| 22 | $4.23^{e} \pm 0.05$ | $55.00^{d} \pm 8.66$ | $10.31^{g} \pm 0.30$ | $0.00^{d} \pm 0.00$ |
| 35 | $4.03^{e} \pm 0.37$ | $60.00^{cd} \pm 5.77$ | $9.02^{h} \pm 0.07$ | $0.00^{d} \pm 0.00$ |

Values represent the mean ± standard error of the mean of three replicates. Means within each column for a given species followed by different superscript letters are significantly different at $p \leq 0.05$ according to DMRT. Means sharing at least one common letter are not significantly different.
*A. africana* = *Afzelia africana*; DMRT = Duncan's Multiple Range Test; *G. albida* = *Gambeya albida*; $G_{max}$ = maximum germination; MC = moisture content.

significant increase ($p \leq 0.05$) at 35 days (3.16 µmol $g^{-1}$ dry weight) suggesting a possible delayed metabolic adjustment to prolonged desiccation.

### Starch Content in Seeds of *A. africana* and *G. albida* During Desiccation

Starch content in *A. africana* increased markedly ($p \leq 0.05$) during early desiccation, with maximum concentration at 3 days (5.37 µmol $g^{-1}$ dry weight), followed by a significant decrease ($p \leq 0.05$) between days 7–13 (Figure 2). Starch content then increased steadily ($p \leq 0.05$) from days 15–35. In *G. albida*, starch content increased slightly up to 3 days but not significantly, followed by a significant decline ($p \leq 0.05$) after that. This suggests distinct starch metabolism patterns between both species during desiccation.

### Influence of Desiccation on Activities of Amylases in Seeds of *A. africana* and *G. albida*

Alpha-amylase activity in *A. africana* increased significantly ($p \leq 0.05$) from 0 to 3 days of desiccation (Figure 3). Activity then declined significantly ($p \leq 0.05$) between days 7–13, followed by a subsequent increase. In *G. albida*, alpha amylase activity increased significantly during the initial desiccation phase, peaking at 7 days, and subsequently declined. The delayed peak in *G. albida* (7 days) compared with *A. africana* (3 days) shows differences in the timing of enzyme responses in the two species.

Beta amylase activity in seeds of *A. africana* during desiccation followed a similar trend as that of alpha amylase, with significantly ($p \leq 0.05$) higher activity on the 3rd day of desiccation (Figure 4). Similarly, seeds of *G. albida* also demonstrated a significant ($p \leq .05$) increase in beta amylase activity with a peak at the 7th day of desiccation, thereafter activity declined.

Total amylase activity in *A. africana* increased significantly ($p \leq 0.05$), with the highest activity observed at 3 days (21.39 µmol reducing sugar released $g^{-1}$ dry weight $min^{-1}$) (Figure 5). Activity decreased thereafter but showed slight significant increases ($p \leq 0.05$) between days 15–35. In *G. albida*, total amylase activity increased significantly at 0, 3, and 7 days having values of (2.40 µmol reducing sugar released $g^{-1}$ dry weight $min^{-1}$, 9.47 µmol reducing sugar released $g^{-1}$ dry weight $min^{-1}$ and 12.72 µmol reducing sugar released $g^{-1}$ dry weight $min^{-1}$ respectively), followed by a significant decline. Generally, *A. africana* exhibited an earlier enzymatic peak, whereas *G. albida* showed delayed and brief increases (Table 2).

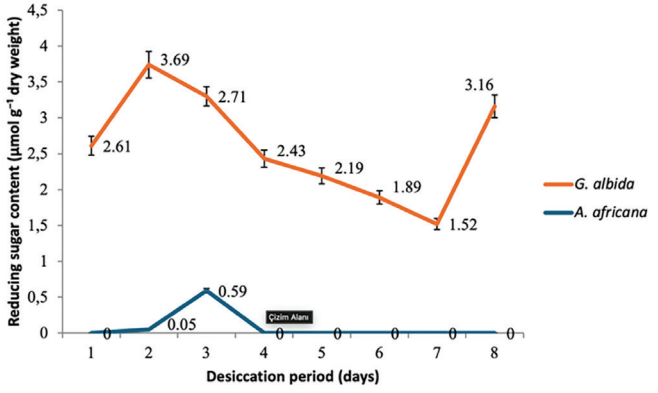


**Figure 1.** Content of reducing sugar (µmol $g^{-1}$ dry weight) in seeds of *A. africana* and *G. albida* during desiccation under ambient conditions; error bars indicate variability at $p \leq 0.05$.
*A. africana = Afzelia africana; G. albida = Gambeya albida.*

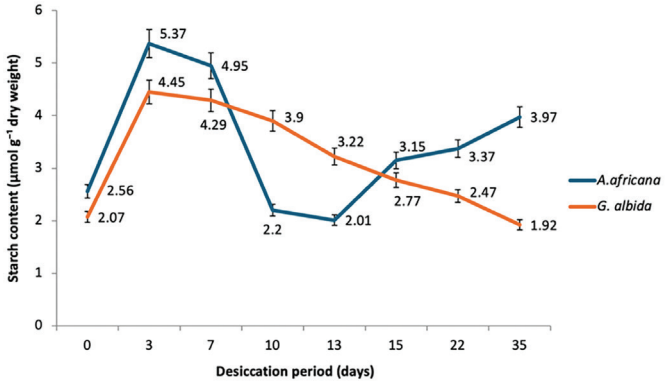


**Figure 2.** Starch content (µmol $g^{-1}$ dry weight) in seeds of *A. africana* and *G. albida* following desiccation; error bars indicate variability at $p \leq 0.05$.
*A. africana = Afzelia africana; G. albida = Gambeya albida.*

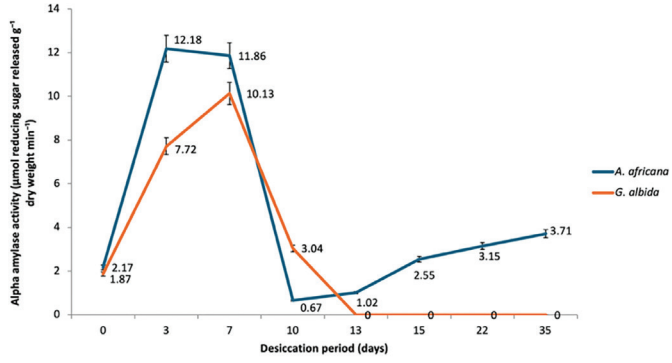


**Figure 3.** Alpha amylase activity (µmol reducing sugar released g-1 dry weight min-1) in seeds of *A. africana* and *G. albida* following desiccation; error bars indicate variability at $p \leq 0.05$.
*A. africana = Afzelia africana; G. albida = Gambeya albida.*

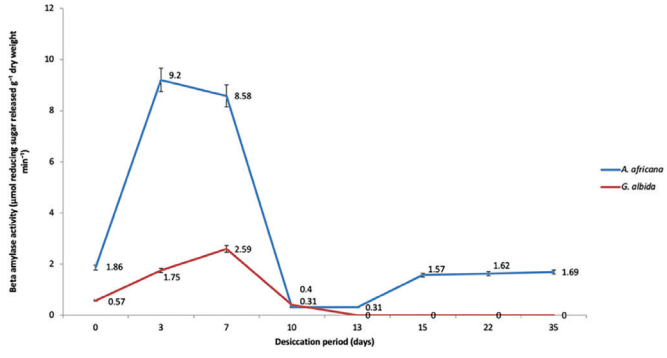


**Figure 4.** Beta amylase activity (µmol reducing sugar released $g^{-1}$ dry weight $min^{-1}$) in seeds of *A. africana* and *G. albida* following desiccation; error bars indicate variability at $p \leq 0.05$.
*A. africana = Afzelia africana; G. albida = Gambeya albida*

## Discussion

*A. africana* seeds exhibited significantly lower MC at collection (13.81%) compared with *G. albida* seeds (40.04%), indicating clear differences in seed water status at maturity. Such contrasts are commonly associated with differences in seed storage behavior. Orthodox seeds typically undergo maturation drying during late development, acquiring tolerance to substantial moisture loss, whereas recalcitrant seeds retain high MC and are sensitive to desiccation (Berjak & Pammenter, 2013; Maia et al., 2011). In the present study, the ability of *A. africana* seeds to retain high germination capacity at low moisture levels supports an orthodox-like response under the conditions tested, whereas the rapid loss of viability in *G. albida* during drying is consistent with recalcitrant behavior.

Reducing sugar content in *A. africana* seeds was relatively low both at collection and following desiccation, whereas *G. albida* maintained consistently higher levels. Previous studies have associated high concentrations of reducing sugars, particularly monosaccharides, with increased desiccation sensitivity, whereas oligosaccharides are more often linked to cellular stabilization during dehydration (Prajith et al., 2014; Whittaker et al., 2004). However, since only reducing sugars were quantified in this study, interpretations are restricted to observed changes in reducing sugar pools rather than specific protective sugars. The higher reducing sugar levels in *G. albida* may therefore reflect limited metabolic regulation during drying rather than an adaptive protective response. Although oligosaccharides such as raffinose have been implicated in desiccation tolerance in other species, these compounds were not directly measured in the present study and are therefore not considered further here.

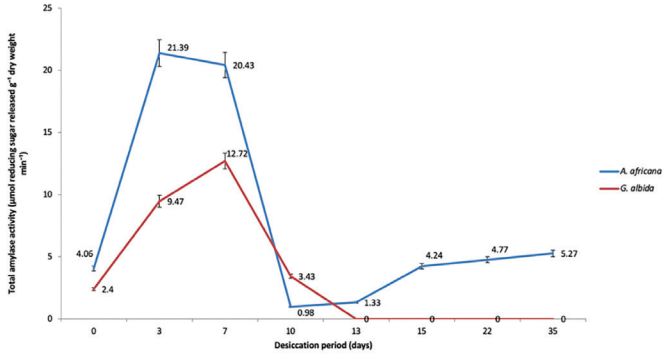


**Figure 5.** Total amylase activity (µmol reducing sugar released $g^{-1}$ dry weight $min^{-1}$) in seeds of *A. africana* and *G. albida* following desiccation; error bars indicate variability at $p \leq 0.05$.
*A. africana = Afzelia africana; G. albida = Gambeya albida.*

The starch metabolism also differed markedly between the two species. Starch content increased during desiccation in *A. africana* but declined in *G. albida*, indicating contrasting patterns of carbohydrate reserve regulation. The accumulation of starch in *A. africana* during drying may reflect a capacity to maintain reserve stability under reduced water availability, whereas starch depletion in *G. albida* coincided with declining germination capacity. Activities of α-, β-, and total amylases increased during early desiccation (0–7 days) in both species, followed by a decline with prolonged drying. *A. africana* exhibited higher and more sustained amylolytic activity, suggesting greater metabolic resilience during dehydration. Similar temporal modulation of amylase activity in orthodox seeds has been reported under varying intensities of desiccation (Lee & Tan, 2020). In contrast, the sharp reduction in amylase activity observed in *G. albida* during the later stages of desiccation may indicate the loss of metabolic integrity, a feature commonly reported in recalcitrant tropical seeds (Nguyen et al., 2023). Although reducing sugars interact with oxidative metabolism in plants, we did not assess reactive oxygen species levels, antioxidant enzymes, or related redox pathways. Consequently, no conclusions are drawn regarding the regulation of oxidative stress during seed desiccation. Such interactions remain important avenues for future investigation.

Overall, this study distinguishes contrasting carbohydrate reserve dynamics and amylolytic responses associated with desiccation tolerance and sensitivity in two tropical tree species. The results provide physiological evidence supporting the suitability of *A. africana* seeds for conventional drying and storage, while highlighting the vulnerability of *G. albida* seeds to moisture loss. These findings are relevant for seed conservation and restoration programs that require speciesspecific handling protocols (Food and Agriculture Organization, 2021).

Although this study offers useful insights, it is limited by the absence of molecular or biochemical markers beyond carbohydrate

**Table 2.** Summary of key physiological responses during desiccation in *A. africana* and *G. albida.*

| Parameter | *A. africana* | *G. albida* |
|---|---|---|
| MC trend | Gradual decline; stabilizes after day 15 | Rapid decline; no germination after day 13 |
| Peak germination | 15 days (90%) | Day 0 (76.7%), then declines to 0% |
| Peak reducing sugars | Day 7 | Day 3; secondary rise at day 35 |
| Peak starch content | Day 3 | No significant early rise; declines after day 3 |
| Peak α-amylase | Day 3 | Day 7 |
| Peak β-amylase | Day 3 | Day 7 |
| Peak total amylase | Day 3 | Days 0–7 |

*A. africana = Afzelia africana; G. albida = Gambeya albida*; MC = moisture content.

metabolism and the restricted number of species examined. Future studies integrating analyses of specific sugar profiles, enzyme regulation at the molecular level, and controlled environmental conditions would further refine the understanding of desiccation responses in tropical forest seeds.

## Conclusion

Based on the findings, it is suggested that *A. africana* seeds may be suitable for conventional drying and storage under conditions similar to those tested in this study, indicating potential for *ex situ* conservation. In contrast, *G. albida* seeds exhibited characteristics consistent with desiccation sensitivity and may therefore require alternative management approaches, such as short term hydrated storage, rapid propagation, or further evaluation for specialized preservation techniques. Future research should incorporate longer term storage trials under controlled environmental conditions to validate storage performance, as well as molecular or ecological investigations to better understand the mechanisms underlying the observed physiological responses.

**Ethics**

**Ethics Committee Approval:** This study was conducted in accordance with national and international ethical guidelines for research involving plants. The plant materials (*Afzelia africana* Sm. and Chrysophyllum albidum Linn) were collected from non-protected areas with due consideration for environmental sustainability and without causing harm to endangered or protected species. No human participants or animals were involved in the research; therefore, formal ethical approval was not required. All experimental procedures complied with relevant institutional, national, and international laws and conventions.

**Data Sharing Statement:** All data are available within the study.

**Footnotes**

**Authorship Contributions:** Conceptualization: A.E.N.; Design/methodology: B.A.N.; Execution/investigation: B.A.N. and A.E.N.; Resources/materials: A.E.; Data analysis/interpretation: A.E.; Writing – original draft: B.A.N.; Writing – review & editing/critical revision: A.E.N.

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