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Biochemical profiling of *Albizia procera* seeds during germination

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Abstract
This study investigates the biochemical changes in total lipid, soluble sugar, and reducing sugar contents during early seed germination of *Albizia procera* (white siris), a multipurpose tree species in the Fabaceae family. Seeds were collected from three individual trees and monitored from day 1 to day 15 of germination under controlled laboratory conditions. Biochemical analyses revealed a consistent and progressive decline in total lipid content across all three trees, indicating active mobilization of lipid reserves to support seedling development. In contrast, soluble sugar and reducing sugar levels fluctuated considerably throughout the germination period, with no uniform pattern of increase or decrease. The magnitude and dynamics of these changes varied among the individual trees, highlighting inherent tree-to-tree variation in seed biochemical composition. These findings provide valuable insights into reserve mobilization and metabolic processes during early germination of A. procera and underscore the importance of genetic variation among seed sources. Further research on associated enzymatic activities is recommended to clarify the mechanisms underlying these biochemical changes.

Keywords: *Albizia procera*, seed germination, biochemical profiling, lipid mobilization, soluble sugars, reducing sugars

Introduction

Albizia procera (Roxb.) Benth., commonly known as white siris, safed siris or karai, is an important multipurpose tree species belonging to the family Fabaceae. The species is widely distributed in tropical and subtropical regions and is valued for its timber, fuelwood, fodder, shade and ecological importance.

Seed germination is a metabolically active process during which stored reserves are mobilized to support embryo growth and seedling establishment. Lipids, soluble sugars and reducing sugars constitute important biochemical components associated with energy metabolism during early seed development. Changes in these biochemical constituents can therefore provide useful information about the metabolic activities occurring during germination.

Previous work on *A. procera* has demonstrated variation in seed germination and seed characteristics among mother trees, indicating the importance of tree-to-tree variation in seed quality (Gera *et al*., 2001) [3]. Studies have also shown that pre-sowing treatments can influence germination and seedling development in *A. procera* (Tiwari & Dhuria, 2018; Jaliya *et al*., 2024) [4, 9]. Seed dormancy in the species has been associated with restrictions to water uptake and seed-coat characteristics (Sajeevukumar *et al*., 1995) [6].

The mobilization of stored biochemical reserves is particularly important during the early stages of germination. Lipid reserves may be utilized as an energy source, whereas soluble and reducing sugars may fluctuate according to the balance between reserve mobilization, respiration, biosynthesis and transport to developing tissues.

The present study was therefore undertaken to investigate changes in total lipid, soluble sugar and reducing sugar contents during the early stages of seed germination of *Albizia procera*. The study followed biochemical changes from the first to the fifteenth day of germination in seeds collected from three individual trees. The same general experimental objective and observation period are reported in the published study by Sett (2016) [7].

Materials and Methods

1. Experimental Site

The study was conducted using seeds collected from *Albizia procera* trees growing at the Tropical Forest Research Institute (TFRI), Jabalpur, Madhya Pradesh, India.

Three individual trees were selected for the experiment. Seeds collected from each tree were treated separately to examine variation in biochemical composition during germination.

2. Plant Material

Healthy and mature seeds of *Albizia procera* were collected from three selected trees. The seeds were carefully examined, and damaged or unhealthy seeds were removed before experimentation.

The seeds were washed thoroughly with running tap water. Surface sterilization was subsequently carried out according to the experimental protocol, followed by washing with sterile water.

3. Seed Germination

The seeds were soaked in water for 24 h and subsequently placed in sterile Petri plates containing moistened filter paper.

The Petri plates were maintained under controlled germination conditions. Germinating seeds were collected at different intervals, namely 1, 3, 6, 9, 12 and 15 days after germination.

4. Experimental Design

Seeds collected from three trees were designated as Tree 1, Tree 2 and Tree 3. Germination was carried out separately for each tree.

Samples were collected on the 1st, 3rd, 6th, 9th, 12th and 15th days of germination. Three replicates were maintained for biochemical analysis.
The experimental arrangement was designed to determine changes in lipid, soluble sugar, and reducing sugar contents during progressive germination.

5. Estimation of Total Soluble Sugar

Total soluble sugar was estimated using the phenol–sulfuric acid method of Dubois *et al*. (1956) [2].
A known quantity of germinating seed material was extracted using 80% ethanol. The extract was centrifuged, and the supernatant was collected. The extraction was repeated, and the supernatants were pooled.
For estimation, a known aliquot of the extract was taken, and the volume was adjusted with distilled water. Phenol solution and concentrated sulfuric acid were added, and the contents were mixed thoroughly. The reaction mixture was heated, and absorbance was measured spectrophotometrically.
D-glucose was used as the standard, and the soluble sugar concentration was calculated from the standard curve.

6. Estimation of Reducing Sugar

Reducing sugar was estimated using the Nelson–Somogyi method (Nelson, 1944; Somogyi, 1952) [5, 8].
A known aliquot of the sample extract was taken and treated with Somogyi's copper reagent. The mixture was heated in a boiling-water bath and subsequently cooled. Nelson's arsenomolybdate reagent was added, and the volume was adjusted with distilled water.
Absorbance was measured at the appropriate wavelength using a spectrophotometer/colorimeter. D-glucose was used as the standard for calculating reducing sugar concentration.

7. Estimation of Total Lipid

Total lipid was estimated according to the method described by Becker *et al*. (1978) [1].
A known quantity of germinating seed material was homogenized with a chloroform–methanol mixture. The homogenate was allowed to stand and subsequently centrifuged. The organic phase containing extracted lipid was collected and evaporated.
The remaining lipid fraction was weighed and the lipid content was expressed in terms of the sample weight.

Results

1. Selection of Experimental Trees

Three *Albizia procera* trees were selected for the present investigation. The geographical coordinates and elevation of the selected trees are presented in Table 1.

Table 1: Geographic location of the three selected *Albizia procera* trees

Tree	Latitude (N)	Longitude (E)	Elevation (ft)
Tree 1	23°05′58.27″	79°59′21.80″	1353
Tree 2	23°05′58.57″	79°59′22.26″	1353
Tree 3	23°05′49.65″	79°59′17.65″	1355

2. Overall Biochemical Changes Tree 1

Table 2: Changes in total lipid, reducing sugar and soluble sugar during germination of *Albizia procera* — Tree 1

Germination day	Total lipid	Total reducing sugar	Total soluble sugar
0	0.110 ± 0.010	1.824 ± 36.73	0.007 ± 11.73
3	0.040 ± 0.020	10.29 ± 50.85	19.90 ± 41.41
6	0.021 ± 0.002	46.35 ± 44.14	6.844 ± 27.03
9	0.011 ± 0.008	50.65 ± 63.88	0.077 ± 11.78
12	0.004 ± 0.003	18.37 ± 74.71	41.58 ± 39.89
15	0.004 ± 0.005	0.127 ± 16.03	0.721 ± 35.85

Tree 1 showed a marked and continuous decline in total lipid content throughout germination, decreasing from 0.110 on day 0 to 0.004 on day 15. The most pronounced reduction occurred during the first six days, followed by a gradual decline thereafter. This indicates rapid mobilization of stored lipid reserves during the early stages of seedling development.
Reducing sugar content increased substantially from 1.824 on day 0 to 50.65 on day 9, followed by a sharp decline to 0.127 on day 15. Soluble sugar content also showed considerable fluctuations, increasing from 0.007 initially to 19.90 on day 3, declining during subsequent stages, and reaching 41.58 on day 12. These fluctuations suggest active carbohydrate mobilization and utilization during germination.
Overall, Tree 1 exhibited the strongest depletion of lipid reserves, accompanied by pronounced fluctuations in both reducing and soluble sugars.

3. Overall Biochemical Changes Tree 2

Table 3: Changes in total lipid, reducing sugar and soluble sugar during germination of *Albizia procera* — Tree 2

Germination day	Total lipid	Total reducing sugar	Total soluble sugar
0	0.030 ± 0.01	13.96 ± 45.06	0.007 ± 11.80
3	0.028 ± 0.02	22.01 ± 76.17	26.96 ± 77.63
6	0.020 ± 0.02	1.074 ± 31.89	4.447 ± 14.90
9	0.012 ± 0.40	35.58 ± 39.03	52.41 ± 50.93
12	0.010 ± 0.06	18.95 ± 78.33	29.281 ± 65.05
15	0.004 ± 0.02	31.51 ± 33.26	27.71 ± 59.92

In Tree 2, total lipid content declined progressively from 0.030 on day 0 to 0.004 on day 15. The decline was relatively gradual compared with Tree 1, indicating continuous utilization of stored lipid reserves throughout germination.

Reducing sugar content fluctuated considerably, increasing from 13.96 on day 0 to 22.01 on day 3, declining sharply on day 6, and subsequently increasing to 35.58 on day 9. It then decreased at day 12 before increasing again to 31.51 on day 15.

Soluble sugar concentration increased from 0.007 on day 0 to 26.96 on day 3 and subsequently reached a maximum of 52.41 on day 9, followed by a decline to 27.71 on day 15. This pattern suggests active carbohydrate conversion and utilization during seedling establishment.

Overall, Tree 2 showed continuous lipid depletion with substantial fluctuations in carbohydrate reserves, particularly soluble sugars.

4. Overall Biochemical Changes Tree 3

Table 4: Changes in total lipid, reducing sugar and soluble sugar during germination of *Albizia procera* — Tree 3

Germination day	Total lipid	Total reducing sugar	Total soluble sugar
0	0.051 ± 0.002	55.92 ± 52.19	8.966 ± 45.37
3	0.033 ± 0.224	30.35 ± 68.62	20.27 ± 77.15
6	0.030 ± 0.021	18.92 ± 80.94	30.78 ± 76.14
9	0.025 ± 0.001	0.084 ± 16.45	21.53 ± 48.18
12	0.024 ± 0.002	8.768 ± 34.28	25.30 ± 46.22
15	0.020 ± 0.021	9.213 ± 17.87	32.89 ± 35.42

Tree 3 displayed a comparatively slower decline in total lipid content, from 0.051 on day 0 to 0.020 on day 15. Unlike Trees 1 and 2, a relatively larger proportion of the initial lipid reserve remained at the end of the germination period, suggesting slower lipid mobilization.

Reducing sugar content decreased progressively from 55.92 on day 0 to 0.084 on day 9, followed by slight increases at days 12 and 15. This pattern indicates substantial utilization of reducing sugars during the early stages of germination.

Soluble sugar content, in contrast, generally increased from 8.966 on day 0 to 32.89 on day 15, although intermediate fluctuations occurred. The increase in soluble sugars toward later germination stages may reflect continued carbohydrate mobilization and accumulation of soluble metabolites in developing seedlings.

Overall, Tree 3 showed slower lipid depletion and a different carbohydrate pattern compared with Trees 1 and 2.

Discussion

The present study demonstrated clear biochemical changes during seed germination of *Albizia procera*. Total lipid content progressively declined in all three trees, indicating mobilization of stored lipids for energy and carbon supply during seedling development, consistent with Sett (2016) [7]. However, the magnitude of lipid decline varied among trees, suggesting differences in seed biochemical composition, as reported by Gera *et al*. (2001) [3].

Soluble and reducing sugars showed considerable fluctuations rather than a consistent trend, probably reflecting the dynamic balance among reserve mobilization, respiration, biosynthesis, and sugar transport during germination. Overall, the findings agree with Sett (2016) [7], who reported decreasing lipid content but fluctuating soluble and reducing sugar levels during germination of *A. procera*.

Conclusion

The study revealed significant biochemical changes during early seed germination of *Albizia procera*. Total lipid content progressively decreased with germination, indicating the utilization of stored lipid reserves. In contrast, soluble and reducing sugars fluctuated without a consistent trend. Variations among the three trees suggest differences in biochemical characteristics among seed sources. These findings provide useful insights into reserve mobilization during early seedling development. Further studies on germination-associated, carbohydrate- and lipid-metabolizing enzymes are recommended to elucidate the mechanisms underlying reserve utilization.

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