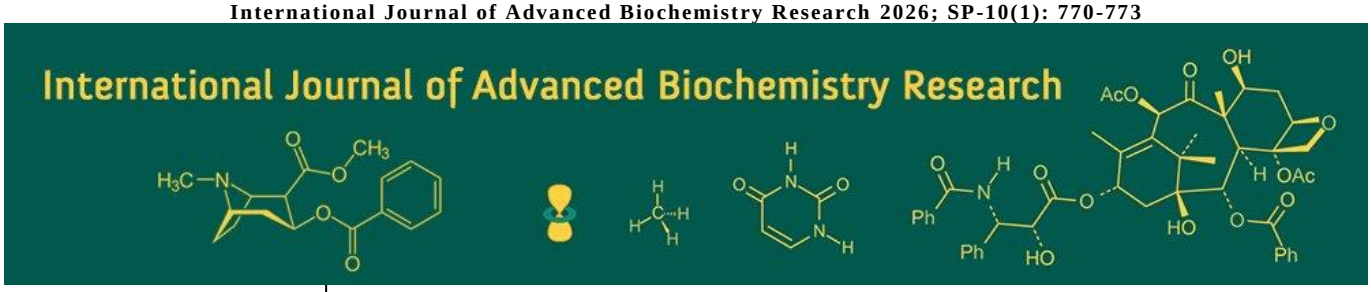

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Effect of age and size of tuberous roots on biochemical and mineral composition of swallow root (*Decalepis hamiltonii* Wight & Arn.)

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Abstract
The present investigation was conducted to evaluate the effect of age and size of tuberous roots on the biochemical and mineral composition of swallow root (*Decalepis hamiltonii* Wight & Arn.), an endangered medicinal plant endemic to southern India. Tuberous roots were collected from two-year-old and three-year-old plants and classified into large (>3 cm diameter) and small (<3 cm diameter) categories, forming four treatment combinations. Dried root powder from each treatment was analyzed for biochemical parameters, including moisture, total carbohydrates, protein, total ash and total phenolic content, as well as mineral composition comprising calcium, iron, potassium and sodium. Significant variations were observed among treatments for all parameters studied. Three-year-old large-sized tuberous roots recorded higher ash content (8.00%), phenolic content (2.75 g/100 g) and mineral composition, particularly calcium (1.10%), iron (564.60 mg/kg) and potassium (3.62%), indicating enhanced nutraceutical potential with increased physiological maturity. Younger and smaller roots exhibited lower moisture content, suggesting better shelf stability but comparatively reduced biochemical richness. Overall, the results demonstrated that root age and size play a crucial role in determining quality attributes of *D. hamiltonii*. Harvesting mature, large-sized tuberous roots is recommended for improved biochemical quality, enhanced mineral composition and value addition, while also supporting sustainable utilization of this threatened species.

Keywords: *Decalepis hamiltonii*, tuber age, root size, biochemical composition, mineral composition, nutraceutical quality

1. Introduction
India is one of the world's most biodiversity-rich countries, supporting nearly 8 per cent of global biodiversity within only 2.4 per cent of the global land area (Wahile and Mukherjee, 2006) [18]. Among its diverse medicinal plant resources, *Decalepis hamiltonii* Wight & Arn., commonly known as swallow root, is an endangered and endemic woody climber of southern India with considerable nutritional, medicinal and commercial importance. The species is widely distributed in the Deccan Peninsula and the Western Ghats and has been traditionally used as a health-promoting food and herbal medicine (Gamble and Fischer, 1957; Vedavathy, 2004) [3, 17].

The tuberous roots of *D. hamiltonii* are the economically important plant part and are valued for their unique aroma, nutritional content and bioactive constituents. Biochemically, the roots contain appreciable levels of carbohydrates, phenolic compounds, volatile oils and antioxidants, along with essential macro-and micro-minerals such as calcium, iron, potassium and sodium (Nagarajan *et al.*, 2001; Kumar *et al.*, 2015) [9, 6]. The volatile oil fraction, dominated by 2-hydroxy-4-methoxy benzaldehyde, contributes to the characteristic flavour and preservative properties of the roots, while the mineral content enhances their nutritional and therapeutic value. These biochemical and mineral composition attributes form the basis for their widespread use in traditional formulations, beverages such as "Nannari," and various value-added products (Vedavathy, 2004; Reddy and Murthy, 2013) [17, 12].

In perennial medicinal plants, biochemical and mineral composition is known to vary with plant age and organ size due to differences in physiological maturity, assimilate accumulation and tissue differentiation.

Tuberous roots harvested at different developmental stages may exhibit significant variation in nutrient concentration, fibre content and bioactive compounds. Younger roots often have higher moisture and lower storage reserves, whereas older and larger roots may show increased accumulation of carbohydrates and minerals but may also undergo lignification, potentially affecting quality and palatability.

In *D. hamiltonii*, harvesting practices are largely unstandardized and driven by market demand, with roots collected irrespective of age or size, leading to inconsistent nutritional quality and reduced product uniformity (Giridhar *et al.*, 2005; Ved and Goraya, 2017) [4, 16]. Moreover, indiscriminate harvesting has intensified pressure on natural populations, contributing to the endangered status of the species (Ravikumar and Ved, 2000; Ved *et al.*, 2015) [11, 15]. Despite the growing commercial importance of swallow root and its increasing use in value-added products, scientific information on the influence of tuber age and size on biochemical and mineral composition remains limited.

Understanding how age and size of tuberous roots affect their biochemical and nutritional quality is therefore essential for determining the optimum harvest stage, improving product quality and promoting sustainable utilization of this endangered medicinal species. In this context, the present investigation was undertaken to evaluate the effect of plant age and tuber size on the biochemical and mineral composition of *Decalepis hamiltonii* tuberous roots, with the aim of identifying suitable harvesting criteria for commercial exploitation and value addition.

2. Materials and Methods

2.1 Sample Collection and Experimental Design

Tuberous roots of *Decalepis hamiltonii* Wight & Arn. were collected from the College of Horticulture, Anantharajupeta, Dr. Y.S.R. Horticultural University, Andhra Pradesh, India. Roots were harvested from two different plant ages, namely two-year-old and three-year-old plants, to study the influence of age on biochemical and mineral composition. Immediately after harvest, the tuberous roots were washed thoroughly with tap water to remove adhering soil and debris. The roots were then categorized based on diameter into two size classes: large-sized roots (>3 cm diameter) and small-sized roots (<3 cm diameter). Based on plant age and root size, four treatment combinations were established. These treatment groups were used for biochemical and nutritional analysis.

T_1: Three-year-old plants with large-sized tuberous roots (>3 cm diameter)

T_2: Three-year-old plants with small-sized tuberous roots (<3 cm diameter)

T_3: Two-year-old plants with large-sized tuberous roots (>3 cm diameter)

T_4: Two-year-old plants with small-sized tuberous roots (<3 cm diameter)

2.2 Sample Preparation

Fresh tuberous roots were cut into small segments measuring approximately 3-5 cm in length. The cut pieces were sun-dried for 7-10 days until a constant weight was achieved. The dried roots were ground into a fine powder using a mechanical grinder and stored in airtight containers at room temperature until analysis. All biochemical and nutritional analyses were carried out using the dried root powder.

2.3 Biochemical analysis

2.3.1 Determination of Moisture Content

Moisture content of swallow root powder was determined using a Radwag moisture analyzer (Model: MAC 50, Poland). Approximately 2 g of the powdered sample was placed in the analyzer, and the moisture content was recorded once a constant reading was achieved. The results were expressed as percentage moisture.

2.3.2 Estimation of Total Carbohydrate Content

Total carbohydrate content was estimated using the Anthrone method as described by Sadasivam and Manickam (1997) [13]. A dried sample (100 mg) was hydrolysed with 5 ml of 2.5 N hydrochloric acid in a boiling water bath for 3 h. After cooling, the hydrolysate was neutralized with sodium carbonate until effervescence ceased and the volume was made up to 100 ml with distilled water. The solution was centrifuged and 0.2 ml of the supernatant was taken and diluted to 1 ml. Four millilitres of anthrone reagent were added, and the mixture was heated in a boiling water bath for 8 min, followed by rapid cooling. Absorbance was measured at 630 nm using a spectrophotometer. A standard curve was prepared using glucose, and carbohydrate content was expressed as g per 100 g of sample.

2.3.3 Estimation of Protein Content

Protein content was estimated by Lowry's method (Sadasivam and Manickam, 1997) [13]. A 500 mg sample of dried root powder was extracted using 5-10 ml of Tris buffer (tris hydroxymethyl aminomethane) and centrifuged. An aliquot of 0.1 ml of the supernatant was mixed with 5 ml of alkaline copper reagent and allowed to stand for 10 min. Subsequently, 0.5 ml of Folin-Ciocalteu reagent was added, and the mixture was incubated in the dark at room temperature for 30 min. Absorbance was measured at 660 nm. Protein content was calculated using a standard curve and expressed as g per 100 g of sample.

2.3.4 Determination of Total Ash Content

Total ash content was determined following AOAC (1980). Approximately 5 g of the sample was weighed into a pre-weighed crucible and charred over a low flame. The crucible was then placed in a muffle furnace at 600 °C for 4-5 h until white or greyish-white ash was obtained. The crucible was cooled in a desiccator and weighed. The process was repeated until a constant weight was achieved, and total ash content was expressed as a percentage.

2.3.5 Estimation of Total Phenolic Content

Total phenolic content was estimated using the Folin-Ciocalteu method (Sadasivam and Manickam, 1997) [13] with gallic acid as the standard. One gram of dried root powder was homogenized with 10 ml of 80 per cent ethanol and centrifuged at 10,000 rpm for 10 min. The residue was re-extracted with 80 per cent ethanol, and the supernatants were pooled. An aliquot of 2 ml of extract was taken and the volume was made up to 3 ml with distilled water. To this, 0.5 ml of Folin-Ciocalteu reagent was added and allowed to stand for 3 min, followed by the addition of 2 ml of 20 per cent sodium carbonate solution. The mixture was heated in a boiling water bath for 1 min, cooled, and absorbance was measured at 650 nm. Phenolic content was calculated using a gallic acid standard curve and expressed as g per 100 g of sample.

2.4 Mineral Analysis

2.4.1 Calcium and Iron

Calcium and iron contents were determined using an Inductively Coupled Plasma-Optical Emission Spectrophotometer (ICP-OES) following the method of Perkin and Elmer (1982). One gram of dried root powder was digested using a diacid mixture of nitric acid and perchloric acid (9:4). The digested sample was diluted to 100 ml with distilled water and directly analyzed. The results were expressed as percentage.

2.4.2 Potassium and Sodium

Potassium and sodium contents were estimated using a flame photometer as described by Jackson (1973). One gram of dried root powder was digested with a nitric acid and perchloric acid mixture (9:4) and diluted to 50 ml. Potassium and sodium concentrations were determined using KCl and NaCl standards, respectively, and the results were expressed as percentage.

2.5 Statistical analysis

The data recorded from the study was subjected to statistical analysis. The data on biochemical and mineral composition of root powder were subjected to completely randomized design (CRD) analysis using GRAPES software.

3. Results and discussion

3.1 Biochemical Parameters

The biochemical composition of *Decalepis hamiltonii* tuberous roots varied significantly with plant age and root size, reflecting differences in physiological maturity and metabolic activity. Moisture content ranged from 9.25 to 10.08 per cent, with the lowest value recorded in T_4 (two-year-old, small-sized roots) and the highest in T_1 (three-year-old, large-sized roots), indicating better shelf stability in younger and smaller roots. Total carbohydrate content showed considerable variation, with the highest value observed in T_3 (5.03 g/100 g), followed by T_1 and T_4, suggesting enhanced assimilate accumulation in large-sized roots during active growth phases. Protein content was highest in T_2 (1.07 g/100 g), possibly reflecting greater enzymatic and metabolic activity at intermediate stages of maturity. Total ash content, an indicator of total mineral matter, was significantly higher in T_1 (8.00%) compared to other treatments, highlighting increased nutrient accumulation with age. Similarly, phenolic content was highest in T_1 (2.75 g/100 g), indicating enhanced synthesis of secondary metabolites associated with plant defence and antioxidant capacity. Overall, three-year-old large-sized tuberous roots (T_1) exhibited superior biochemical and functional attributes, supporting their higher nutraceutical potential. These findings are consistent with earlier reports by Mahesh *et al.* (2025) [7], Samydurai and Thangapandian (2012) [14], Mohan and Kalidas (2010) [8] and Das *et al.* (2017) [2].

3.2 Mineral Composition

Mineral composition of *Decalepis hamiltonii* tuberous roots was significantly influenced by the age and size of the roots. Calcium content was highest in T_1 (1.10%), followed by T_2 (0.82%), while the lowest values were recorded in T_4 and T_3, indicating improved mineral accumulation in mature roots due to prolonged uptake and storage. Iron content exhibited marked variation among treatments, with T_1 recording the highest iron concentration (564.60 mg/kg), substantially exceeding the values observed in younger and smaller roots. Potassium content followed a similar trend, with the maximum level in T_1 (3.62%), followed by T_2, T_3 and T_4, reflecting its role in carbohydrate translocation and enzyme activation. Sodium content was also highest in T_1 (1.25%), which was statistically comparable to T_2, while lower levels were recorded in T_3 and T_4. The consistently higher mineral content in three-year-old, large-sized tuberous roots indicates greater physiological maturity and nutrient storage capacity. These results corroborate the findings of Mohan and Kalidas (2010) [8] and emphasize the advantage of harvesting mature roots for enhanced mineral quality and value addition.

Table 1: Biochemical parameters of *Decalepis* roots of different age and size

Treatments	Moisture (%)	Total carbohydrates (g/100 g)	Protein (g/100 g)	Total ash (%)	Phenol content (g/100 g)
T_1	10.08^a	4.87^b	0.97^b	8.00^a	2.75^a
T_2	9.54^b	4.24^c	1.07^a	6.78^b	1.69^b
T_3	9.47^b	5.03^a	0.89^c	6.19^c	1.61^b
T_4	9.25^c	4.80^b	0.93^{bc}	6.05^c	1.28^c
C.D. @ 5%	0.21	0.13	0.05	0.20	0.09
SE (m)	0.07	0.04	0.02	0.07	0.03
CV (%)	1.65	2.04	3.66	2.23	3.70

Superscripts with same alphabets in column represents no significant difference at the 5% level.

Table 2: Mineral composition of *Decalepis* roots at different age and size

Treatments	Calcium (%)	Iron (mg/kg)	Potassium (%)	Sodium (%)
T_1	1.10^a	564.60^a	3.62^a	1.25^a
T_2	0.82^b	333.70^b	2.94^b	1.14^a
T_3	0.60^b	317.34^b	2.43^{bc}	0.54^b
T_4	0.50^b	300.38^b	2.23^c	0.45^b
C.D. @ 5%	0.14	72.29	0.52	0.17
SE (m)	0.05	24.11	0.17	0.06
CV (%)	14.36	14.23	13.93	14.79

Superscripts with same alphabets in column represents no significant difference at the 5% level.

4. Conclusion

The present study demonstrated that both age and size of tuberous roots significantly influence the biochemical characteristics and mineral composition of *Decalepis hamiltonii*. Among the treatments evaluated, three-year-old, large-sized tuberous roots (>3 cm diameter) consistently exhibited superior quality attributes, including higher ash, phenolic content and enhanced accumulation of essential minerals such as calcium, iron, potassium and sodium. While younger and smaller roots showed comparatively lower moisture content, which may contribute to improved shelf stability, they were inferior in terms of overall biochemical and mineral richness.

The findings clearly indicate that physiological maturity plays a crucial role in determining the nutraceutical potential of swallow root. Harvesting mature, larger tuberous roots can therefore ensure improved biochemical quality, enhanced mineral composition and better suitability for processing and value addition. Establishing such standardized harvesting criteria is essential not only for improving product quality and market value but also for

promoting sustainable utilization and conservation of this endangered medicinal species. The results of this study provide a scientific basis for recommending optimum harvest maturity for *Decalepis hamiltonii* cultivation and commercial exploitation.

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