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INFLUENCE OF TUBER MATURITY AND SIZE ON THE MICROBIAL STABILITY OF SYRUP AND POWDER FROM SWALLOW ROOT (*DECALEPIS HAMILTONII* WIGHT & ARN)

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ABSTRACT

An investigation was conducted to evaluate the microbial stability of swallow root (*Decalepis hamiltonii*) syrup and powder prepared from different tuber maturity stages and sizes. Four treatment combinations were studied, namely T₁ – three-year-old large tubers (>3 cm diameter), T₂ – three-year-old small tubers (<3 cm diameter), T₃ – two-year-old large tubers (>3 cm diameter) and T₄ – two-year-old small tubers (<3 cm diameter). Both syrup and powder were assessed for bacterial and fungal load during storage. No bacterial or fungal growth was detected in either product up to 30 days. In syrup, microbial growth first appeared at 60 days, with T₁ (0.6×10^6 CFU/ml bacterial; 0.4×10^4 CFU/ml fungal) and T₂ (0.8×10^6 CFU/ml bacterial; 0.6×10^4 CFU/ml fungal) showing significantly lower counts compared to T₃ and T₄, which recorded higher microbial loads. By 90 days, microbial proliferation increased across treatments, although T₁ consistently maintained the most favourable microbial profile. Powder formulations demonstrated greater inherent stability, with no bacterial or fungal growth up to 60 days and only minimal counts at 90 days, particularly in T₃ and T₄. These findings suggest that syrup benefits from mature, large-sized tubers for improved microbial safety, while powder is inherently more stable, with younger roots offering a slight advantage. The results highlight the importance of tuber maturity, size and product form in determining the microbial quality and shelf life of swallow root value-added products.

Keywords : Swallow root (*Decalepis hamiltonii*), Tuber Age and size, Microbial count, Syrup and Powder.

Introduction

Decalepis hamiltonii Wight & Arn., widely known as “swallow root,” is a rare perennial climbing shrub belonging to the family Asclepiadaceae. It is an endemic species of India, confined mainly to the Deccan Plateau and the forested regions of the Western Ghats. Both areas globally recognized for their rich biological diversity and ecological importance. The plant is well integrated into the cultural and traditional knowledge systems of South India, where it is identified by a variety of local names: “Maredu kommulu,” “Nannari kommulu,” and “Maredu gaddalu” in Telugu; “Makaliber” in Kannada; “Magalikizhangu” in Tamil; and “swallow root” in

English (Vedavathy, 2004). In response to increasing demand, this species has been brought under organized cultivation in several pockets of Andhra Pradesh, Karnataka, and Tamil Nadu, particularly in tracts ranging between 300 and 1200 m above sea level (Sharma and Shahzad, 2014).

The commercial and therapeutic relevance of *D. hamiltonii* stems from its underground root system, which develops into fleshy, elongated tubers arranged in compact clusters. These roots, usually cylindrical and woody at the core, are distinctive for their strong vanilla-like fragrance, attributed mainly to the phenolic aldehyde 2-hydroxy-4-methoxy benzaldehyde. This compound constitutes nearly 96 per cent of the volatile

oil fraction, accounting for about 0.68 per cent of the dry root biomass (George *et al.*, 2004; Nagarajan *et al.*, 2001). The unique aroma and mild bitterness of the tubers contribute to both their medicinal value and their acceptability as a raw material in food and beverage formulations (Raju and Ramana, 2011).

From an ethnopharmacological perspective, the tubers of *D. hamiltonii* occupy an important place in Ayurvedic medicine and indigenous healing traditions. They have long been consumed either raw, chewed directly or processed into herbal decoctions and syrups (sharbat) that are widely believed to enhance digestion, stimulate appetite, relieve fatigue and promote general health (Vedavathy, 2004; Reddy and Murthy, 2013). Earlier ethnobotanical investigations have also documented their role in treating a range of ailments, including gastrointestinal disturbances, respiratory complaints, metabolic disorders and haematological imbalances (Kumuda *et al.*, 2011; Arutla *et al.*, 2012). Pharmacological studies further validate these uses, reporting antibacterial, antifungal, antioxidant, hepatoprotective and anti-ulcer activities among others (Harish *et al.*, 2005; Monika *et al.*, 2020; Devi and Latha, 2012).

In recent decades, the species has gained prominence in the nutraceutical, functional food and flavouring industries. Syrups and powders derived from the roots are increasingly marketed as refreshing beverages, natural flavouring agents and herbal health tonics. However, like many plant-derived products, their safety and quality are closely linked to their microbial status. Since the tubers are harvested from the soil, they are naturally exposed to a wide range of microorganisms during cultivation, harvesting, handling and processing. If not adequately managed, such microbial contamination may compromise both the shelf life and the consumer safety of value-added products.

An additional consideration is that tuber age and size may strongly influence not only the phytochemical profile but also the degree of microbial colonization. Mature tubers with larger biomass may accumulate different microbial populations compared to younger or smaller ones. Hence, it becomes essential to establish the relationship between tuber characteristics and microbial quality in processed forms such as syrup and powder. Despite the cultural and commercial importance of *D. hamiltonii*, only limited scientific attention has been directed toward understanding the microbial aspects of its processed derivatives.

Given this background, the present study was designed to investigate the bacterial and fungal load in

syrup and powder prepared from swallow root tubers of different maturity stages and sizes. The findings are expected to contribute to establishing appropriate harvesting and processing strategies that ensure microbiologically safe and consumer-acceptable products.

Materials and Methods

Materials

Tuberous roots of swallow root (*Decalepis hamiltonii* Wight & Arn.) were collected from the College of Horticulture, Anantharajupeta, Dr. YSR Horticultural University. Roots were harvested from plants at two maturity stages *viz.* two-year-old and three-year-old tubers. After harvest, the roots were thoroughly washed and sorted based on age and size. For size classification, roots with a diameter greater than 3 cm were categorized as large-sized, while those measuring less than 3 cm in diameter were grouped as small-sized. The experiment consisted of four treatments, distinguished by tuber age and size.

T₁ - Three-year-old & large sized tuberous roots (>3cm diameter)

T₂ - Three-year-old & small sized tuberous roots (<3cm diameter)

T₃ - Two-year-old & large sized tuberous roots (>3cm diameter)

T₄ - Two-year-old & small sized tuberous roots (<3cm diameter)

Syrup and powder were prepared using the four treatments in the Department of Post-Harvest Technology, College of Horticulture, Anantharajupeta. The procedure for the preparation of syrup and powder was carried out according to the method described by Mahesh *et al.* (2025). The products were stored under ambient room conditions and subjected to microbial analysis at 0, 30, 60 and 90 days of storage at Department of Plant Pathology, College of Horticulture, Anantharajupeta.

Microbial Analysis

Sample Preparation

Ten milliliters of syrup or the equivalent of 10 g of powder was homogenized with 90 ml of sterile distilled water in a conical flask. The mixture was shaken for 10 min to ensure uniform mixing. From this stock solution, serial tenfold dilutions were prepared up to 10⁻⁶ by transferring 1 ml of the suspension into sterile test tubes containing 9 ml of sterile distilled water.

Enumeration of Bacteria and Fungi

Bacterial counts were determined using Nutrient Agar (NA), whereas fungal counts were estimated using Potato Dextrose Agar (PDA). For bacterial analysis, the 10^{-6} dilution was used, while for fungal analysis the 10^{-4} dilution was selected. From each dilution, one milliliter of the sample was transferred into sterile petri dishes in duplicate, followed by the addition of sterilized molten agar medium using the pour plate method. After solidification, plates were incubated at 28 ± 1 °C for three to five days. The resulting colonies were enumerated using a digital colony counter and results were expressed as colony forming units (CFU) per milliliter of syrup or per gram of powder. The microbial load was calculated according to the method of Harrigan and Cance (1966) using the following formula:

$$\text{CFU/ml} = \frac{\text{Number of colonies} \times \text{Wt. of the sample}}{\text{Volume of culture plated (ml)} \times \text{Dilution factor}}$$

Result and Discussion

The bacterial and fungal counts of swallow root syrup and powder prepared under different treatments are presented in Table 1 and Table 2, respectively.

Bacterial count

No bacterial growth was observed in either the syrup or powder formulations prepared with different treatments, both at the initial stage and after 30 days of storage. In syrup samples, at 60 days after storage, bacterial growth was first observed, with the significant lowest count in T_1 (0.6×10^6 CFU/ml), followed by T_2 (0.8×10^6 CFU/ml). Significantly higher bacterial counts were recorded in T_3 (1.6×10^6 CFU/ml) and T_4 (1.8×10^6 CFU/ml). After 90 days of storage, bacterial populations increased further, with T_1 and T_2 showing comparatively lower counts (1.6×10^6 CFU/ml each), while T_3 and T_4 reached significantly higher values of 2.4×10^6 CFU/ml.

In powder samples, no bacterial colonies were detected during the initial 60 days of storage. At 90 days, bacterial growth was observed across treatments, although the levels remained relatively low. The lowest bacterial count was recorded in T_4 (0.6×10^6 CFU/g), followed by T_3 (0.8×10^6 CFU/g). Slightly higher counts were noted in T_1 and T_2 (1.0×10^6 CFU/g each). Statistical analysis revealed no significant differences among the treatments.

Fungal count

No fungal growth was observed in either the syrup or powder formulations prepared with different treatments, both at the initial stage and after 30 days of storage. In syrup samples, at 60 days after storage, fungal growth was first observed, with the significantly lowest count in T_1 (0.4×10^4 CFU/ml), followed by T_2 (0.6×10^4 CFU/ml). Significantly higher fungal loads were recorded in T_3 (1.2×10^4 CFU/ml) and T_4 (1.4×10^4 CFU/ml). After 90 days of storage, fungal proliferation increased across treatments, with T_1 and T_2 maintaining relatively lower counts (0.8×10^4 and 1.0×10^4 CFU/ml, respectively), while significantly higher counts were noted in T_3 (1.6×10^4 CFU/ml) and T_4 (1.8×10^4 CFU/ml).

In powder samples, no fungal colonies were detected during the initial 60 days of storage. At 90 days, fungal growth was observed across treatments, although the levels remained relatively low. The lowest fungal count was recorded in T_4 (0.6×10^4 CFU/g), followed by T_3 (0.8×10^4 CFU/g). Slightly higher counts were noted in T_1 and T_2 (1.0×10^4 CFU/g each). Statistical analysis revealed no significant differences among the treatments.

The absence of bacterial and fungal growth in syrup for up to 30 days across all treatments indicates that the initial processing and hygienic handling were effective in ensuring microbial safety. However, after 60 days of storage, both bacterial and fungal colonies began to appear. Treatments prepared from three-year-old, large-sized tuberous roots (T_1) consistently maintained the lowest microbial load, followed closely by T_2 , while two-year-old root samples (T_3 and T_4) supported significantly higher bacterial and fungal proliferation. This suggests that the maturity of the root contributes to microbial stability, likely due to higher concentrations of bioactive secondary metabolites such as phenolics, alkaloids, or saponins in older roots, which have been reported to exert antimicrobial properties.

At both 60 and 90 days, syrup from large-sized roots (T_1 and T_3) showed relatively lower microbial counts compared to small-sized root samples (T_2 and T_4). Larger tubers may possess higher dry matter and phytochemical levels, which not only reduce water activity but also provide natural resistance to microbial colonization. Hence, syrup prepared from three-year-old, large-sized roots (T_1) exhibited the most favourable microbial profile, while syrup from two-

year-old, small-sized roots (T_4) showed the highest bacterial and fungal loads after extended storage.

In contrast, powder formulations demonstrated greater inherent stability than syrup, with no detectable bacterial or fungal colonies up to 60 days of storage. Microbial growth was first observed only at 90 days and even then, the overall counts remained relatively low compared to syrup. Interestingly, powders prepared from two-year-old roots (T_3 and T_4) showed lower microbial counts compared to those from three-year-old roots (T_1 and T_2). This reversal of the trend observed in syrup may be attributed to the reduced moisture content and denser structure of younger roots, which limit microbial proliferation in dried products. Furthermore, only minor differences were observed between large and small-sized roots in powders, suggesting that once moisture is minimized, root size exerts little influence on microbial stability.

Overall, these findings emphasize that the microbial safety of swallow root products depends not only on product form but also on the physiological characteristics of the raw material. Syrup, as a liquid formulation, is more susceptible to microbial

proliferation and benefits from the use of older, larger roots that provide enhanced antimicrobial protection. Powder, on the other hand, is inherently more stable during storage, with younger roots offering a slight advantage in reducing microbial load. These observations underline the importance of carefully selecting both root age and size when designing value-added swallow root products to maximize shelf life and ensure microbiological quality.

Conclusion

The study revealed that syrup formulations prepared from three-year-old, large-sized tubers (T_1) of *Decalepis hamiltonii* exhibited superior microbial stability, with consistently lower bacterial and fungal counts during storage. Powder formulations were inherently more stable than syrup, showing no microbial growth up to 60 days and only minimal levels at 90 days, particularly in samples from younger roots (T_3 and T_4). These findings highlight the combined influence of product form and tuber characteristics on microbial safety, underscoring the suitability of mature roots for syrup and younger roots for powder to ensure extended shelf life.

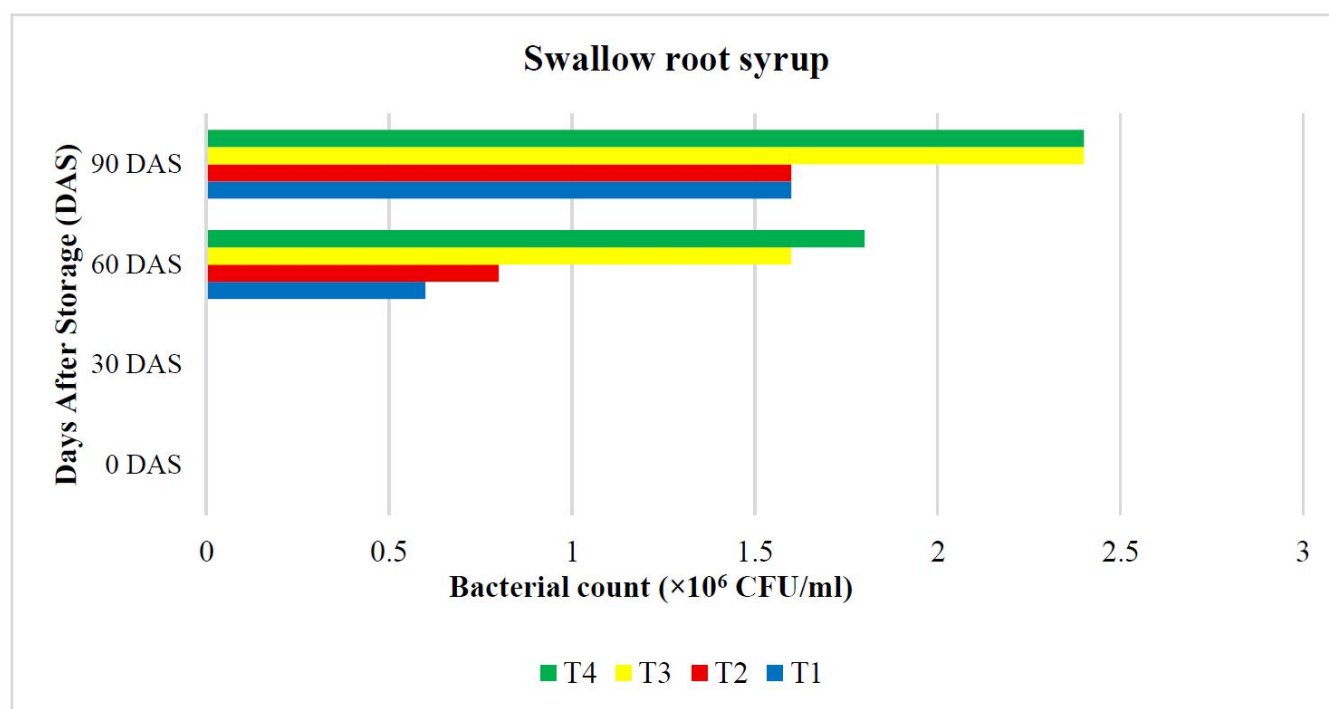


Fig. 1 : Bacterial count (CFU/ml) of swallow root syrup prepared with different age and size of the tuberous roots.

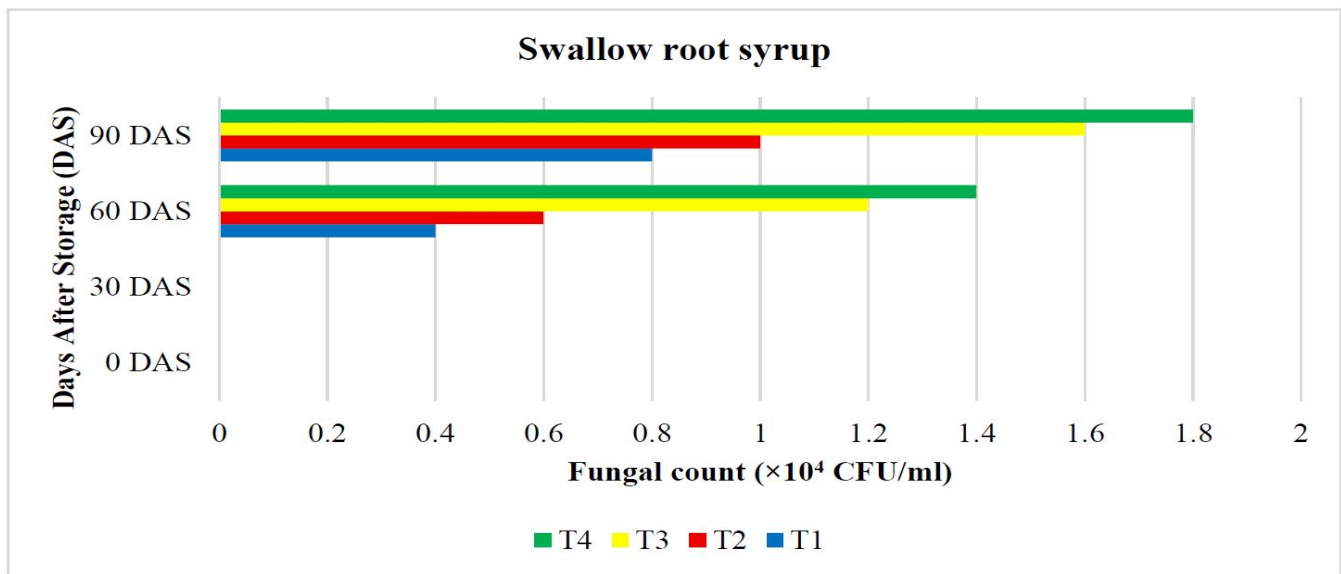


Fig. 2 : Fungal count (CFU/ml) of swallow root syrup prepared with different age and size of the tuberous roots.

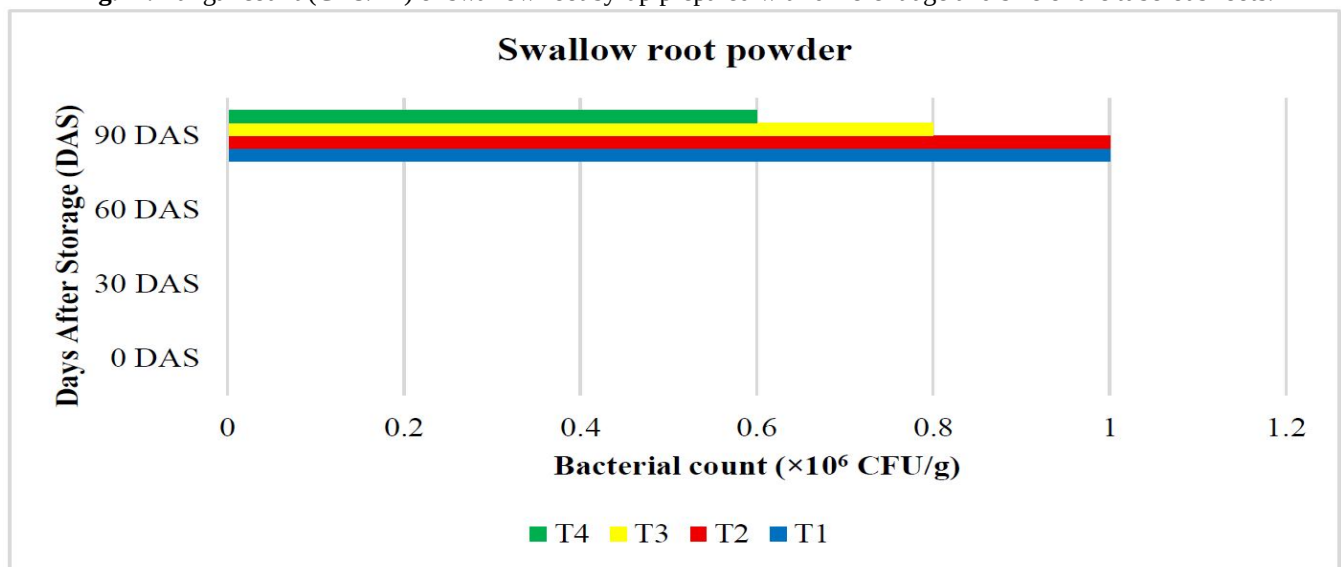


Fig. 3 : Bacterial count (CFU/g) of swallow root powder prepared with different age and size of the tuberous roots.

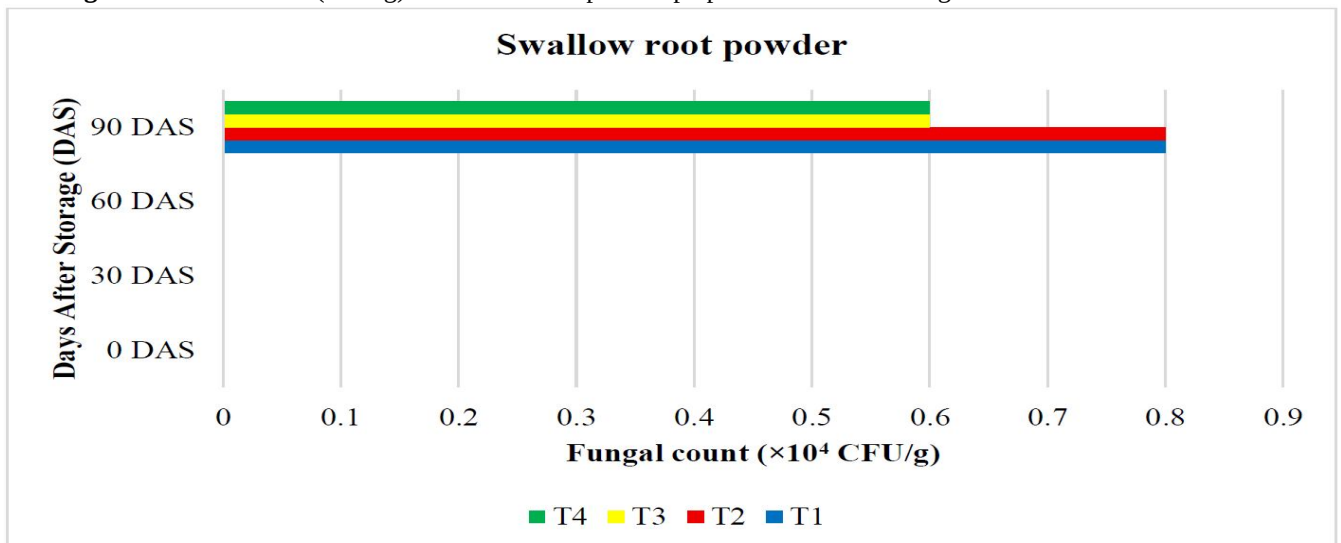


Fig. 4 : Fungal count (CFU/g) of swallow root powder prepared with different age and size of the tuberous roots.

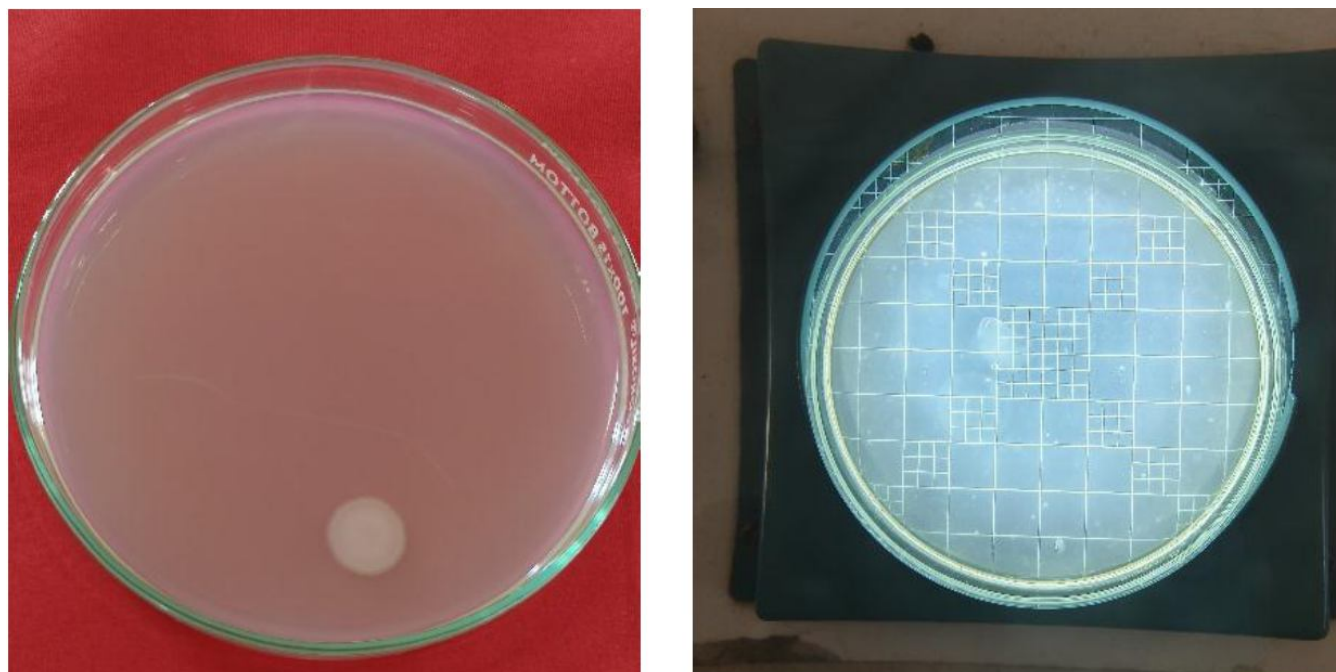


Fig. 5 : Quantitative study of fungal growth in swallow root syrup samples

Table 1: Microbial count (CFU/ml) of swallow root syrup prepared with different age and size of the tuberous roots.

Treatments	Microbial count (CFU/ml) (Bacteria)				Microbial count (CFU/ml) (Fungi)			
	0 DAS	30 DAS	60 DAS	90 DAS	0 DAS	30 DAS	60 DAS	90 DAS
T ₁	-	-	0.6×10^6	1.6×10^6	-	-	0.4×10^4	0.8×10^4
T ₂	-	-	0.8×10^6	1.6×10^6	-	-	0.6×10^4	1×10^4
T ₃	-	-	1.6×10^6	2.4×10^6	-	-	1.2×10^4	1.6×10^4
T ₄	-	-	1.8×10^6	2.4×10^6	-	-	1.4×10^4	1.8×10^4
C.D.@5%	-	-	0.67	0.73	-	-	0.70	0.56
SE (m)	-	-	0.22	0.24	-	-	0.23	0.19

T₁ – Syrup prepared by using three-year-old & large sized tuberous roots (>3cm diameter)

T₂ - Syrup prepared by using three-year-old & small sized tuberous roots (<3cm diameter)

T₃ - Syrup prepared by using two-year-old & large sized tuberous roots (>3cm diameter)

T₄ - Syrup prepared by using two-year-old & small sized tuberous roots (<3cm diameter)

Table 2: Microbial count (CFU/g) of swallow root powder prepared with different age and size of the tuberous roots.

Treatments	Microbial count (CFU/g) (Bacteria)				Microbial count (CFU/g) (Fungi)			
	0 DAS	30 DAS	60 DAS	90 DAS	0 DAS	30 DAS	60 DAS	90 DAS
T ₁	-	-	-	1×10^6	-	-	-	0.8×10^4
T ₂	-	-	-	1×10^6	-	-	-	0.8×10^4
T ₃	-	-	-	0.8×10^6	-	-	-	0.6×10^4
T ₄	-	-	-	0.6×10^6	-	-	-	0.6×10^4
C.D.@5%	-	-	-	-	-	-	-	-
SE (m)	-	-	-	0.16	-	-	-	0.22

T₁ – Powder prepared by using three-year-old & large sized tuberous roots (>3cm diameter)

T₂ - Powder prepared by using three-year-old & small sized tuberous roots (<3cm diameter)

T₃ - Powder prepared by using two-year-old & large sized tuberous roots (>3cm diameter)

T₄ - Powder prepared by using two-year-old & small sized tuberous roots (<3cm diameter)

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