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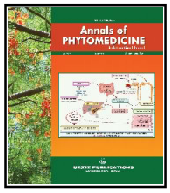

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Phenological and propagation studies of *Curcuma neilgherrensis* Wt., a wild medicinal plant

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Abstract

Phenological and propagation studies of *Curcuma neilgherrensis* Wt., a wild medicinal plant carried out to understand the differences in its growth and development under diverse environmental and growth conditions. The phytoconstituent curcumin is a major bioactive compound present in *Curcuma* species. Medicinal plant cultivation is an important factor to be implemented by the pharmaceutical industries for crude drug collection and to extract required phytoconstituents. It is also essential to know the suitable physical, chemical, and biotic factors affecting its growth and yield. These cultivation practices help to know the conservation measures of wild turmeric at different localities to maintain the germplasm for future generations. Hence, the present investigation aimed to study the pattern of variation in the growth, yield and quantity parameters of *C. neilgherrensis* under different soil conditions, *viz*., sand, red soil, red soil + sand, soil + organic matter. Under these conditions, phenology and propagation of *C. neilgherrensis* were recorded besides the soil physical and chemical testing. *C. neilgherrensis* propagation in different soils proved that growth under red soil and sand (1:1) combination was promising and yielded high quantities of rhizomes with secondary and tertiary roots following natural and wild conditions.

1. Introduction

Phenology is the study of the effects of seasonal and climatic changes on plant and animal life cycles. In general, these phenological changes such as flowering, leaf unfolding, insect emergence and bird, fish, and mammal migration are predominant during the spring season. For a better understanding of ecological adaptations, succession, and interactions of individual species and also for germplasm conservation phenological studies play a vital role (Stern and Roche, 1974; Thomson, 1978; Waser, 1979). These studies are also important for orchid industries, flower trading, and silvi culture (Ganapathya and Rangorajan, 1964; Kaul and Raina, 1980; Bisht, 1986; Navchoo and Kachroo, 1986; Beniwal, 1987; Ghate and Kumbhojkar, 1991; Sagreiya, 1992a; 1992b). Besides these, phenological studies are also useful for medicinal and aromatic plant cultivation, new varieties development and associated industries. There are many medicinal and aromatic plants, which are being cultivated for their socio-economic importance, one among which is wild *Curcuma*.

The genus *Curcuma* is recognized for numerous species of economic significance, beyond its use as a spice and in medicinal applications (Jyothi *et al*., 2003; Policegoudra and Aradhya, 2008). This genus contains many species among which *C. amada*, *C. aromatica*, *C. zedoaria*, *C. purpurascens*, *C. mangga*, *C. heyneana*, *C. xanthorrhiza*, *C. aeruginosa*, *C. phaeocaulis* and *C. petiolata* (Velayudhan *et al*., 1999) are industrially important. In India, turmeric production is nearly 80% of the world's total consumption and occupies approximately 6% of the total area dedicated to spices and condiments. Globally, turmeric production stands at around 11 lakh tonnes per annum. In the year 2020-2021, India exported 1.71 lakh tonnes of turmeric, an increase from the 1.37 lakh tonnes exported in the previous year. The major turmeric-producing states in India are Andhra Pradesh and Kerala, followed by Tamil Nadu, Orissa, Karnataka, West Bengal, and Gujarat. Indian Institute of Spice Research in Calicut is supporting strong research for cultivation in association with the state universities.

Wild *Curcuma* is known to possess tremendous therapeutic potencies such as antiulcer, antimicrobial, antiageing, antidiarrhoeal, antioxidant, antidiabetic, anti-Alzheimer and anti-inflammatory attributed to the presence of various secondary metabolites such as curcuminoids, volatile oils, turmerones and oleoresins (Rangachari, 1991; Pullaiah, 1997; Jadav, 2001; Yesodaram, 2007; Arinathan, 2007; Gantait *et al*., 2011; Samyudurai *et al*., 2012; Naikodi *et al*., 2012; Chaithra *et al*., 2022; Naikodi and Ansari, 2021b; Rasheed *et al*., 2012 and 2013; Venkatesham *et al*., 2021; Bamne *et al*., 2023; Rasheed *et al*., 2017). *C. neilgherrensis* produces starchy rhizomes which are used as a remedy for infections, antimicrobials, inflammations, diabetes, gastric and skin disorders by the local herbalists but have not been evaluated pharmacognostically (Chaithra *et al*., 2013). *C. neilgherrensis* rhizomes are also reported as efficient antimicrobials,

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anthelmintic, antidiarrhoeal and antiulcer agents due to the presence of major secondary metabolites (Yasodamma *et al*., 2013; Yasodamma *et al*., 2014; Chaithra *et al*., 2013; Chaithra *et al*., 2015).

Due to the high production of this crop, plantlets of high production and highly resistant varieties were produced using *in vitro* studies (Villamor, 2010). *Curcuma* production also increased *ex situ* by application of microbial bioinoculants under nursery conditions (Sumathi *et al*., 2011). Breeding among the intra-specific and inter-specific populations by the *in situ*/*in vivo* conservation improved the diversity of the species. Natural breeding processes in wild populations ensure gene flow, which is usually absent in cultivated populations. Hence, the wild populations need to be preferably conserved as gene pools, rather than used as raw material for consumption (Jadav, 2001). With this background, the present investigation aims to study the pattern of variation in the growth and yield and quantify the growth parameters of *C. neilgherrensis* under diverse growth conditions.

2. Materials and Methods

2.1 Rhizome and soil collection

The rhizomes of *Curcuma neilgherrensis* Wt. were collected between April and September 2012 from the hilly part of the Eastern Ghats region of the Tirumala hills. Taxonomic identification and authentication of the collected rhizomes were conducted by a taxonomist, who assigned Voucher Specimen Number DC 922. Soil samples were collected from the Tirupati, Talakona, Kadapa, and Nellore areas. A composite sample of an area is normally preferred and follows the same principle for the current study.

Collected soil samples were transported to the laboratory and tested for pH-reduced ionic states, redox potential, organic nitrogen, and phosphorous fractions with fresh soil samples. The remaining soil stored at a cool temperature after air dried used for the analysis of nitrate, nitrite, ammonium, nitrogen halides, amino acids, carbohydrates and volatile fats.

2.1.1. Water holding capacity (WHC)

Water holding capacity (WHC) was assessed by placing a filter paper, typically Whatman No. 1 or 44, with dimensions suitable for covering the entire perforated bottom of a brass box. The initial weight of the filter paper, denoted as W_1 was recorded. To this, dry soil (0.5 mm particle size) was added, until the box was full.

Water was added in a row to the soil until it was completely moist. The next day after 12-16 h record the weight (W_2). After that, the box was kept in the oven for about 24 h at 105^{o}C, following cooling in a desiccator, and weight was recorded (W_3). The weight of a single filter paper after saturation with water was recorded (W_4). And the percentage of water holding capacity was calculated as below:

% of water holding capacity =$W_2 - W_3 - W_4 / W_3 - W_1 \times 100$. ($W_1$= weight of brass box + filter paper; W_2 = weight of brass box + saturated soil; W_3 = weight of brass box + oven dry soil; W_4 = amount of water retained by the filter paper).

2.1.2 pH determination

For the determination of the pH of soil, in 1:5 ratio of soil suspension was used. pH was recorded using digital pH unfiltered soil suspension.

2.1.3 Alkalinity

Total alkalinity was assessed through direct titration of the soil solution using a strong acid, with methyl orange employed as an indicator. A 1:5 ratio soil suspension was prepared by combining 20 g of soil with 100 ml of aerated distilled water, which was then shaken for approximately one hour. The suspension was subsequently filtered through Whatman No. 50 filter paper using a Buchner funnel and vacuum pump. A 100 ml portion of the filtered solution was extracted and mixed with 2-3 drops of methyl orange indicator before titration with 0.1 N HCl. The endpoint of the titration was indicated by a color change to pink. Calculated the total alkalinity by using the formula meq/100 g = (ml × N) of HCl × 500/ml soil solution.

2.1.4 Organic matter

10 g of 0.5 mm particle-sized dried soil was weighed and transferred to a dried 500 ml conical flask to which 10 ml of 1 N $K_2Cr_2O_7$ and 20 ml concentrated sulphuric acid having silver sulphate dissolved in it was added and mixed using gentle swirling. After swirling the flask was allowed to settle for 30 min and diluted the contents by adding 200 ml of distilled water. To it, 10 ml of phosphoric acid and 1 ml of diphenylamine indicator were added and a change in color (bluish purple) was observed which was titrated with ferrous ammonium sulphate until the color changed to brilliant green. The calculations made with the formula = [V_1- V_2 /W × 0.003 × 100, *i.e.,* organic matter is % of carbon × 1.724]. (Whereas, V_1: volume of $K_2Cr_2O_7$ (10 ml); V_2: volume of ferrous ammonium sulphate; W: weight of the soil taken).

2.1.5 Carbonates

5 g of dried soil were taken into a 150 ml flask; added 100 ml of 1 N HCl was covered with a watch glass and allowed to settle for an hour with intermittent vigorous stirring. 20 ml of supernatant was pipetted out into a conical flask, added 6-8 drops of bromothymol blue indicator, and titrated with 1N NaOH till the colour changed to blue. Blank samples without soil were also titrated following the above steps. Carbonates were calculated as the % of $CaCO_3$= (B –T) × 5, where B refers to reading with blank and T refers to reading with soil.

2.1.6 Chlorides

1:5 suspension of water and soil were made and dissolved chlorides in the water were determined directly by titrating it with 0.02 N silver nitrate using 5% potassium chromate as an indicator. Percentage of chlorides was determined by the formula: [(ml × N) of $AgNO_3$ × 35.5/ ml of soil solution × 2] and converted the results for % of chlorides in mg/100 g by multiplying with 1000.

2.2 Propagation studies

Vegetative propagation was carried out under greenhouse conditions for one year, from August 2013 to August 2014. The experiment involved filling pots (each with an area of 0.25 m^2) with 10 kg of air-dried soil. In each pot, ten rhizomes were planted at a depth of 6 cm. A total of ten pots were utilized for each type of soil. Chemical fertilizers were intentionally omitted to isolate the effects of soil's

physical and chemical properties on turmeric growth and yield parameters. Adequate watering was provided daily to maintain optimal soil moisture levels for proper rhizome sprouting and plant development. After 90 days, once the main shoot had completed leaf formation and reached maturity, plant height and the number of tillers were recorded. Plants were harvested for rhizomes after the shoots had completely withered.

2.3 Phenological studies

The collected rhizomes are cultivated and maintained both in forest and greenhouse conditions in different soils along with (soil, sand, soil + sand) farm yard and recorded phenological observations, *viz*., all crop growth stages starting from rhizome sprouting till the plant maturity, *i.e*., plant height, leaf size, number of leaves, inflorescence development, number of flowers, day of flower bud initiation, longevity of flower, duration for fruit formation, seed development and dehiscence.

2.4 Biomass

At the end of the growing season, each plant was uprooted to evaluate the yield of both its aerial components (leaves, scapes, and flowers) and underground parts (rhizomes). The harvested plant parts underwent thorough washing with running water to remove soil particles. Subsequently, their fresh weights were measured before being sliced into small pieces and subjected to shade drying. Once fully dried + the dry weight of each part was recorded.

3. Results

In the current study, the phenological and propagation studies of *C. neilgherrensis* has been carried out. The results are depicted in Tables 1-5 and Figures 1-9.

3.1 Physicochemical analysis of soil samples

Eight types of soils were collected for the study and their physico-chemical parameters such as pH, chlorides, carbonates, carbonates, alkalinity, organic matter, water holding capacity were analyzed and shown in the Table 1.

3.2 Phenological characteristics of *C. neilgherrensis*

Phenological characteristics such as growth period, leaf size, duration of inflorescence development, flower/flowering and fruiting patterns of *C. neilgherrensis* was assessed and illustrated in Table 2 and Figures 1, 2 and 3.

3.3 Propagation studies-growth and development of *C. neilgherrensis* under different soil conditions

Propagation of *C. neilgherrensis* under different soil conditions was studied by evaluation the different growth and development parameters such as phenotypic characteristics of plant (plant height, leaf parameters, root and rhizome parameters), yield attributes of the rhizome and the biomass presented in Tables 3, 4, 5 and Figures 4a, 4b, 5, 6, 7, 8, 9.

Table 1: Physicochemical analysis of soil samples

S. No.	Soil samples	pH	% chlorides	% carbonates	Alkalinity (meq)	Organic matter (meq)	% Water holding capacity
1	Tirumala	6.0 ± 0.05	59.9 ± 0.02	36.8 ± 0.10	7.8 ± 0.01	4.1 ± 0.01	14.7 ± 0.01
2	Talakona	5.7 ± 0.10	51.2 ± 0.01	32.9 ± 0.75	6.0 ± 0.01	3.1 ± 0.01	11.9 ± 0.01
3	Kadapa	6.3 ± 0.10	60.0 ± 0.01	41.0 ± 0.03	6.9 ± 0.01	3.9 ± 0.01	15.9 ± 0.02
4	Nellore	5.9 ± 0.10	54.7 ± 0.02	43.2 ± 0.01	5.9 ± 0.01	3.03 ± 0.01	13.7 ± 0.01
5	Farmyard soil	5.6 ± 0.05	52.9 ± 0.07	34.3 ± 0.12	6.7 ± 0.01	3.2 ± 0.01	12.8 ± 0.01
6	Red soil	5.9 ± 0.15	54.9 ± 0.05	37.8 ± 0.03	6.9 ± 0.01	4.3 ± 0.01	13.9 ± 0.01
7	Sand	4.9 ± 0.20	49.3 ± 0.63	30.1 ± 0.01	4.5 ± 0.10	3.3 ± 0.05	10.0 ± 0.02
8	Red soil + sand	5.5 ± 0.11	52.6 ± 0.01	33.4 ± 0.01	5.2 ± 0.04	4.09 ± 0.02	11.2 ± 0.01

Table 2: Phenological studies under different conditions

S. No.	Parameters	Garden			Natural habitat	(Soil + Organic matter)
		Sand	Red soil	Red soil + Sand		
1	Growth period (days)	92-130	94-120	100-143	90-120	90-120
2	Leaf size L × W (cm)	20.3 × 4.2	64.3 × 8.4	60.0 × 7.5	58.0 × 8.0	48.0 × 4.8
3	Inflorescence development (days)	80-100	60-75	70-75	60-75	60-75
4	No. of flowers	2-4	2-4	2-4	4-6	2-4
5	Flowering period	August-October	August-October	August-October	August-October	August-October
6	Flower longevity (days)	1	2	2	3	2
7	Flower opening time (am)	10:20 to 11:00	9:10 to 9:20	9:40 to 10:10	9:10 to 9:20	9:10 to 9:20
8	Fruiting (days)	50-60	30-45	30-45	30-45	30-45

Table 3: Plant growth and development under different soil types and garden conditions (90 days)

Plant part		Soil types				
		Red	Sand	Red + Sand (1:1)	Forest soil	Farm yard
Plant height		157.4 ± 0.24	82.4 ± 0.08	140.2 ± 0.08	139.3 ± 0.16	149.3 ± 0.16
No. of leaves		14.6 ± 0.94	12.6 ± 0.47	6.3 ± 0.47	7.3 ± 0.47	21.0 ± 0.81
Leaf	L	64.7 ± 0.37	20.2 ± 0.12	60.3 ± 0.08	58.3 ± 0.08	48.3 ± 0.12
	W	8.4 ± 0.04	4.1 ± 0.08	7.5 ± 0.08	8.2 ± 0.16	4.7 ± 0.04
Scape	L	68.5 ± 0.08	64.2 ± 0.08	60.2 ± 0.12	71.3 ± 0.04	48.3 ± 0.47
	W	3.8 ± 0.04	3.0 ± 0.08	6.4 ± 0.08	3.5 ± 0.08	3.8 ± 0.04
Mother rhizome	L	9.2 ± 0.012	3.1 ± 0.08	9.1 ± 0.08	5.7 ± 0.04	9.3 ± 0.08
	W	4.6 ± 0.08	0.7 ± 0.04	5.3 ± 0.08	1.4 ± 0.04	6.8 ± 0.08
Primary root	PRN	120	82	21	08	35
	L	28.6 ± 0.047	34.2 ± 0.08	16.7 ± 0.12	8.1 ± 0.08	17.0 ± 0.81
	W	1.1 ± 0.08	1.0 ± 0.04	0.5 ± 0.08	4.3 ± 0.21	1.03 ± 0.12
Primary rhizome	PRhN	52	0	21	08	35
	L	7.2 ± 0.04	0	3.5 ± 0.08	5.8 ± 0.08	6.1 ± 0.08
	W	1.1 ± 0.08	0	0.5 ± 0.04	0.8 ± 0.04	3.1 ± 0.04
Secondary root	SRN	52	0	0	0	35
	L	29.0 ± 0.81	0	22.2 ± 0.12	8.2 ± 0.09	21.0 ± 0.81
	W	15.0 ± 0.08	0	0.8 ± 0.08	4.8 ± 0.08	1.5 ± 0.04
Secondary rhizome	SRhN	52	0	0	0	23
	L	6.7 ± 0.12	0	0	0	5.5 ± 0.04
	W	0.9 ± 0.08	0	0	0	2.5 ± 0.12
Tertiary root	TRN	52	0	0	0	12
	L	7.5 ± 0.08	0	0	0	2.3 ± 0.04
	W	2.3 ± 0.04	0	0	0	1.3 ± 0.04

L: Length; **W:** Width; **N:** Number; **PRN:** Primary root number; **PRhN:** Primary rhizome number; **SRN:** Secondary root number; **SRhN:** Secondary rhizome number; **TRN:** Tertiary rhizome number.

Table 4: Obtained yield of rhizome in different types of soil conditions

S. No.	Types of soil	Rhizomes/plant(g)
1	Red soil	1071.2 ± 0.57
2	Sand	50.1 ± 0.05
3	Red soil + Sand	281.2 ± 0.09
4	Forest soil	336.2 ± 0.20
5	Farmyard soil	922.2 ± 0.20

Table 5: Biomass (%) in different parts of the plant under different conditions

S. No.	Part	Red soil	Sand	Red soil + Sand	Forest soil	Farm yard soil
1.	Leaf	89.5 ± 0.10	79.7 ± 0.20	88.3 ± 0.20	78.3 ± 0.15	92.2 ± 0.20
2.	Scape	95.2 ± 0.20	83.4 ± 0.10	86.1 ± 0.15	72.6 ± 0.20	90.5 ± 0.20
3.	Mother rhizome	79.3 ± 0.20	79.8 ± 0.25	77.7 ± 0.20	84.6 ± 0.20	83.1 ± 0.15
4.	Primary rhizome	97.2 ± 0.20	0	94.4 ± 0.20	93.1 ± 0.20	97.9 ± 0.35
5.	Secondary rhizome	93.7 ± 0.10	0	0	0	93.3 ± 0.20
6.	Primary root	90.9 ± 0.25	94.2 ± 0.20	91.4 ± 0.20	76.9 ± 0.10	89.6 ± 0.30
7.	Secondary root	90.6 ± 0.20	0	87.0 ± 0.20	66.0 ± 0.20	86.2 ± 0.20
8.	Tertiary root	90.5 ± 0.15	0	0	0	86.0 ± 0.20
9.	Flowers	61.2 ± 0.20	48.7 ± 0.20	61.9 ± 0.10	73.3 ± 0.20	77.4 ± 0.20

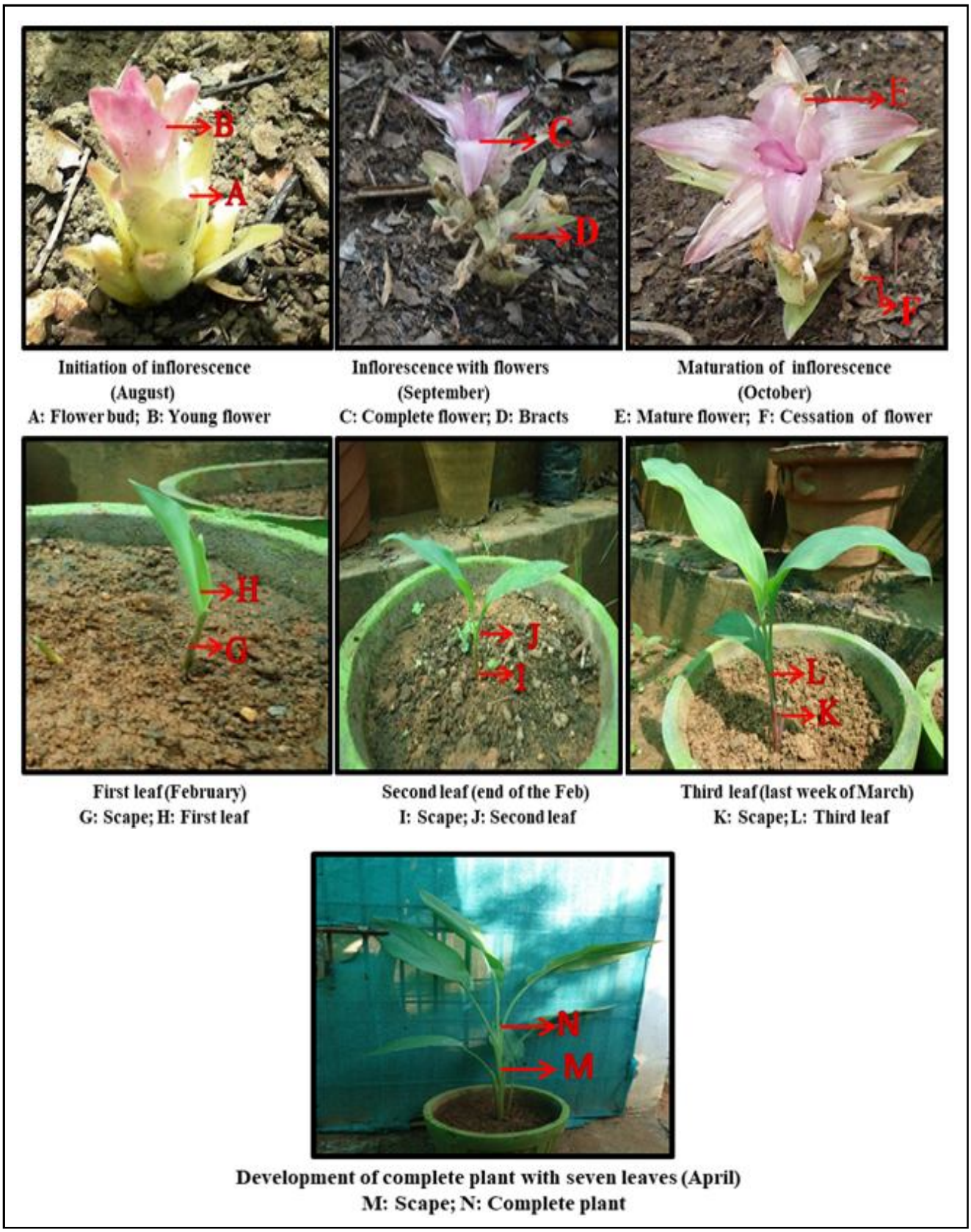


Figure 1: Phenological studies of *C. neilgherre nsis* (Garden soil).

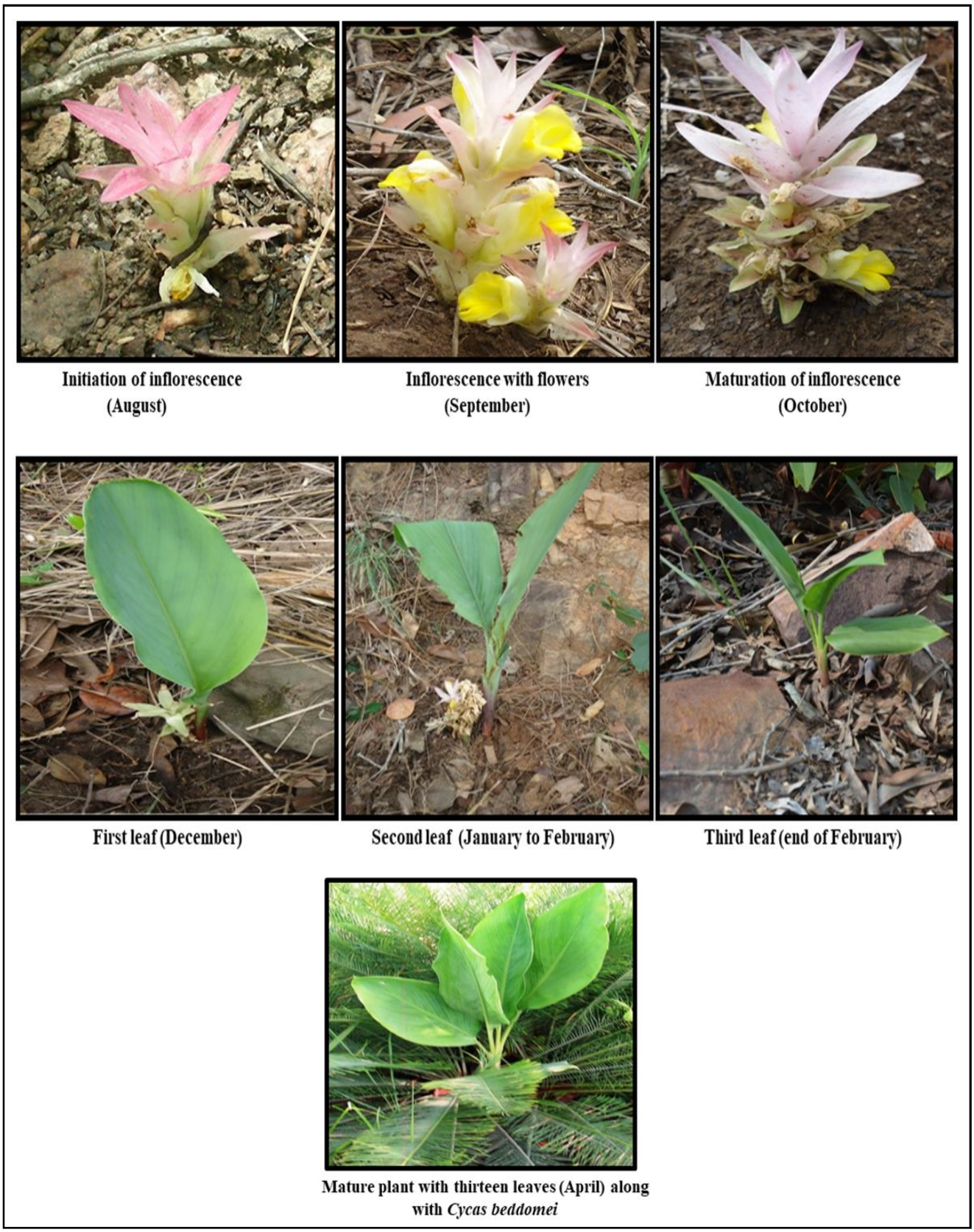


Figure 2: Phenological studies of *C. neilgherrensis* (Natural habitat).

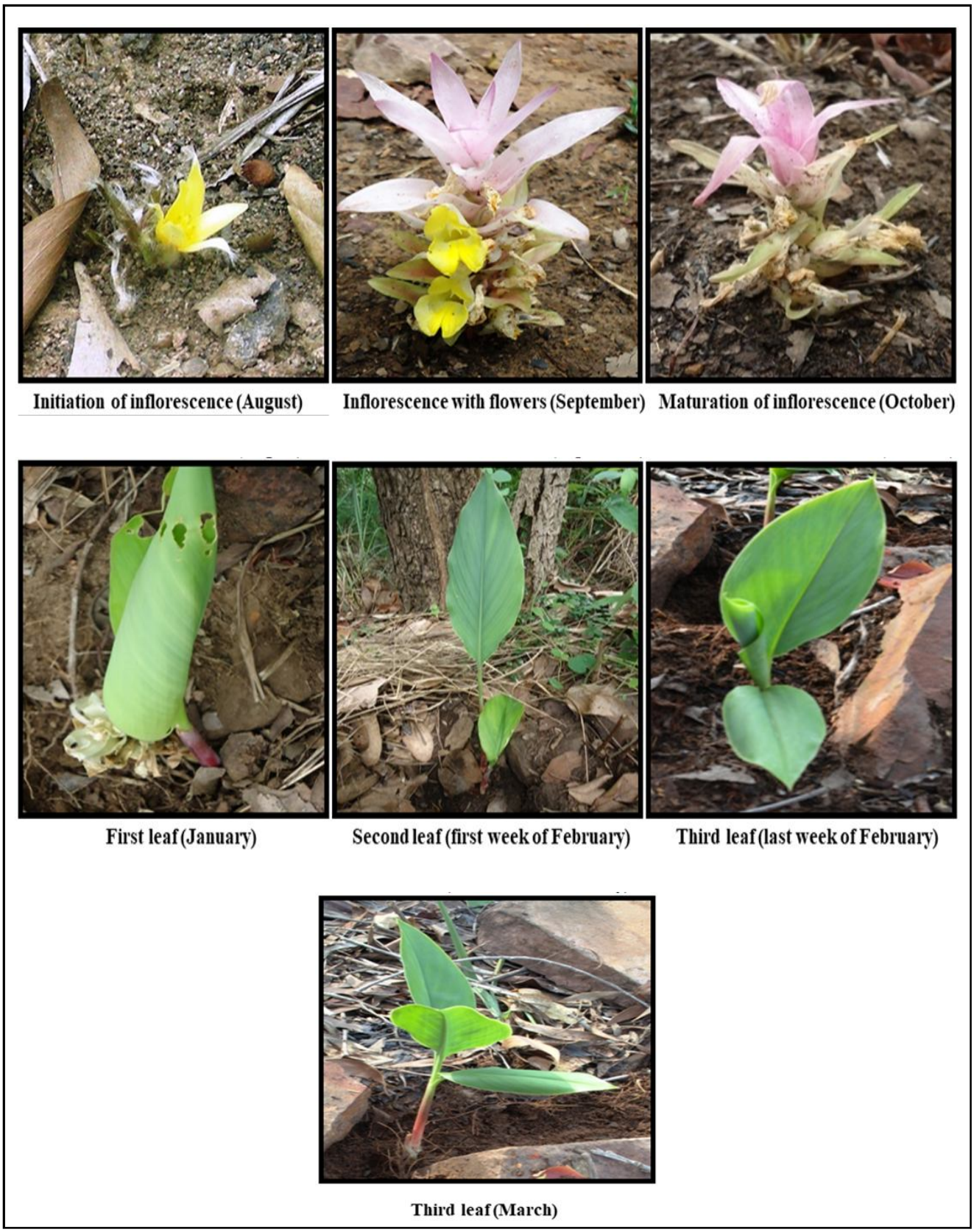


Figure 3: Phenological studies of *C. neilgherrensis* farm yard (soil + organic matter).

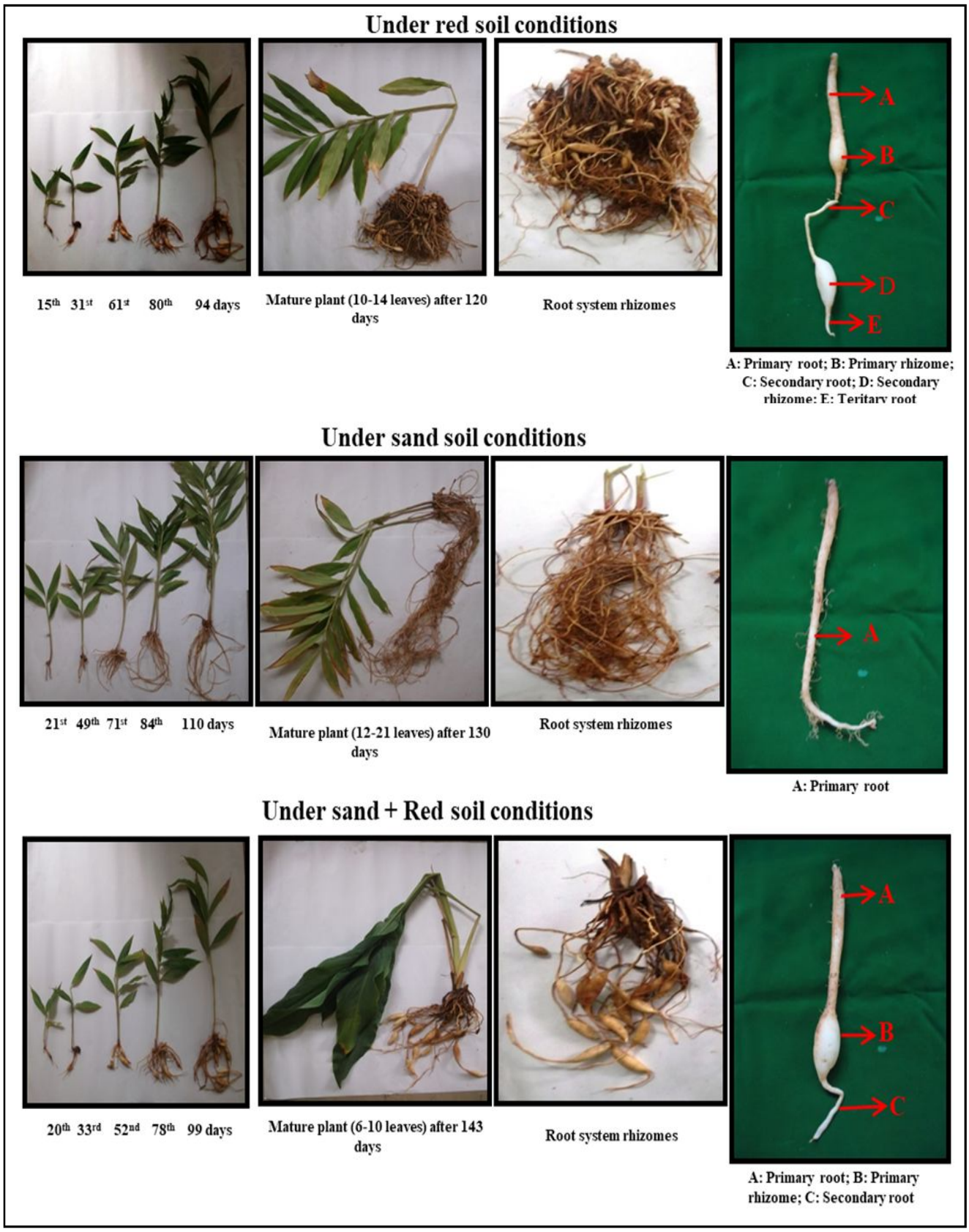


Figure 4a: Propagation stages under different soil conditions.

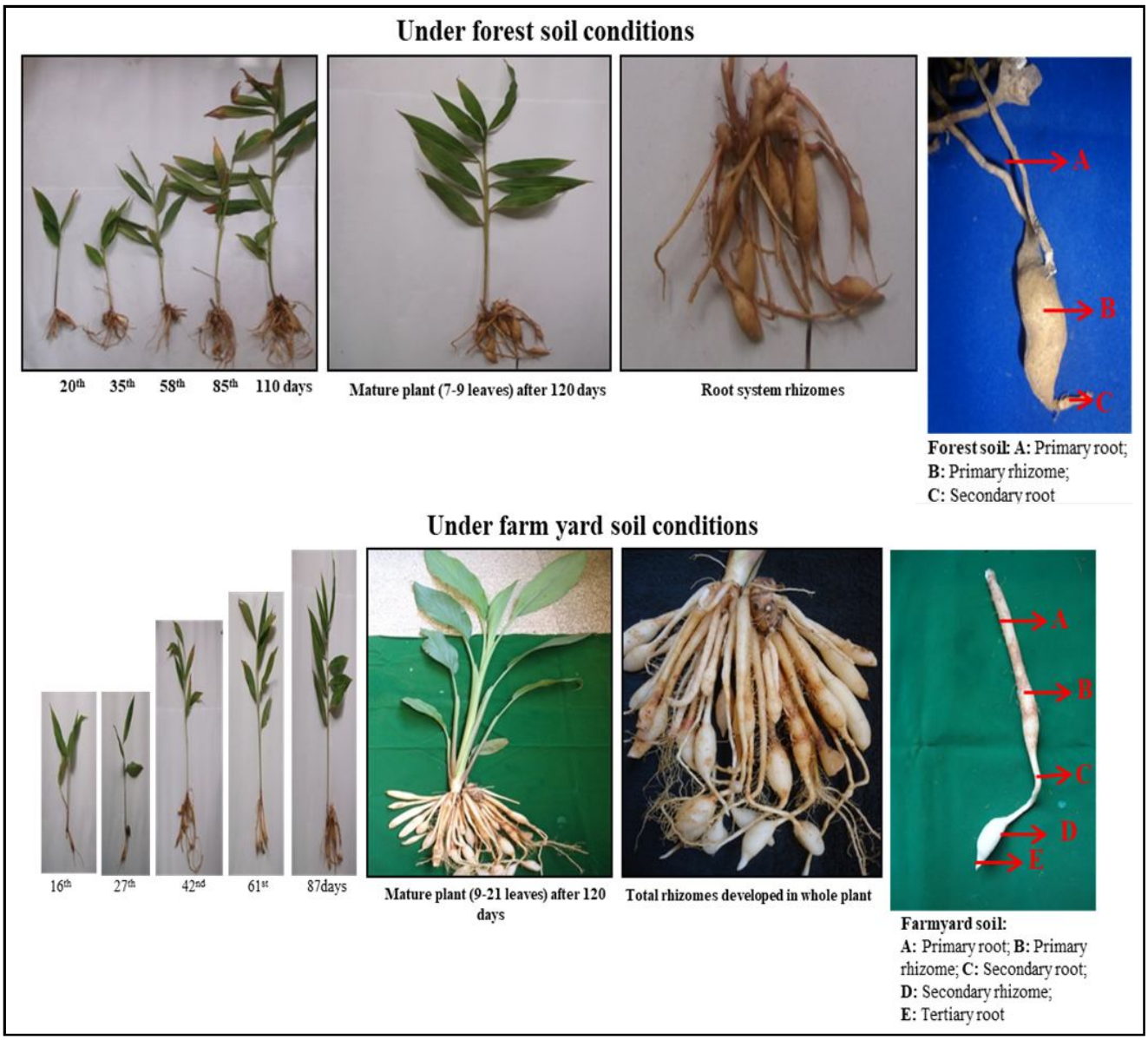
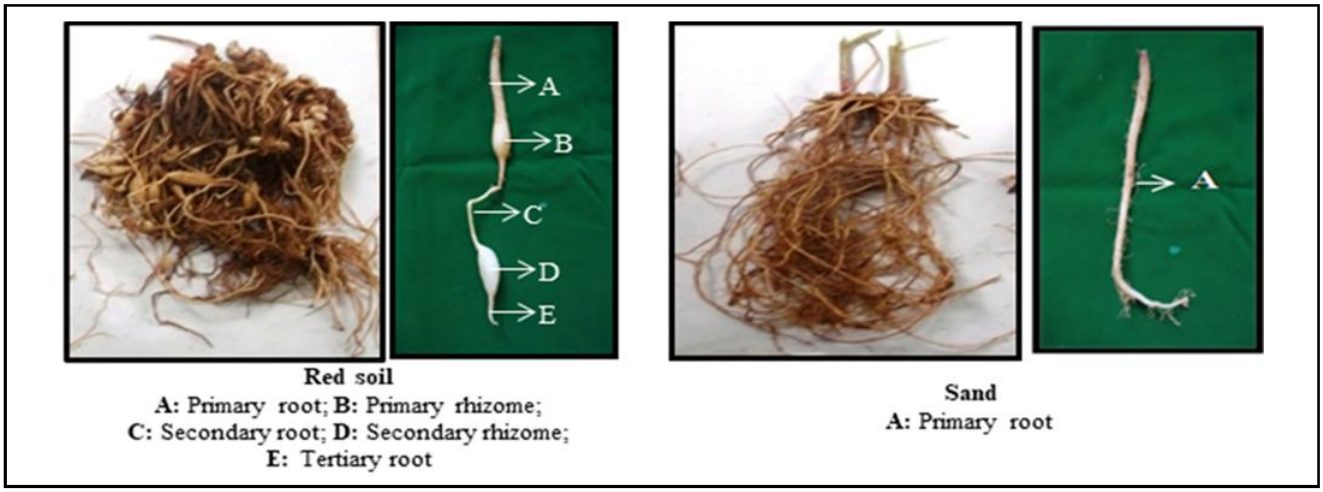


Figure 4b: Propagation stages under different soil conditions.



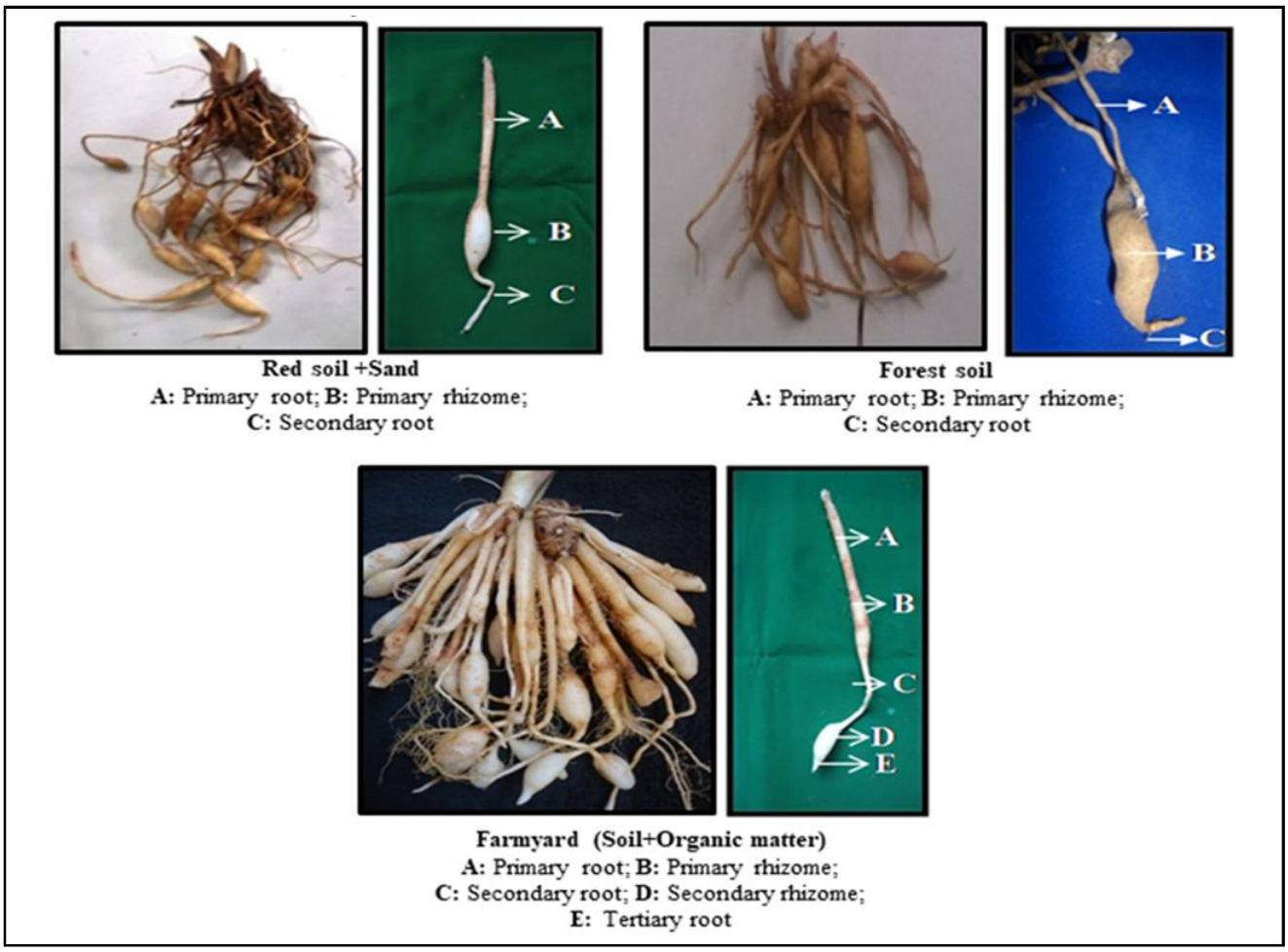
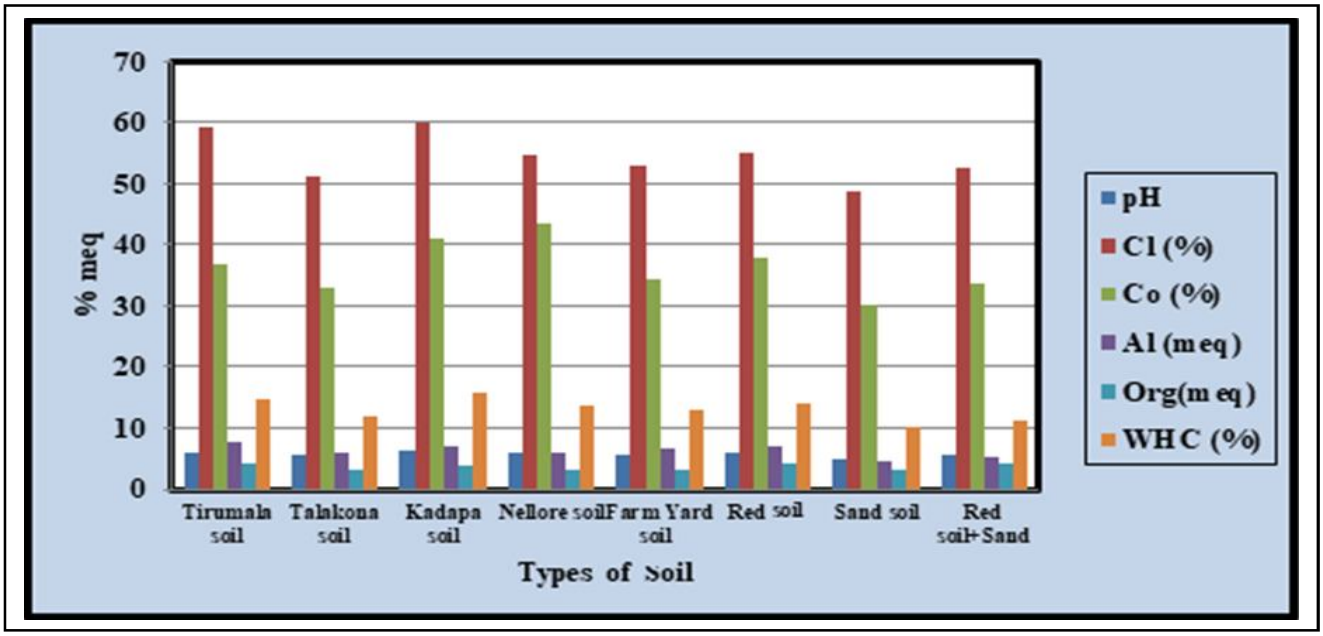
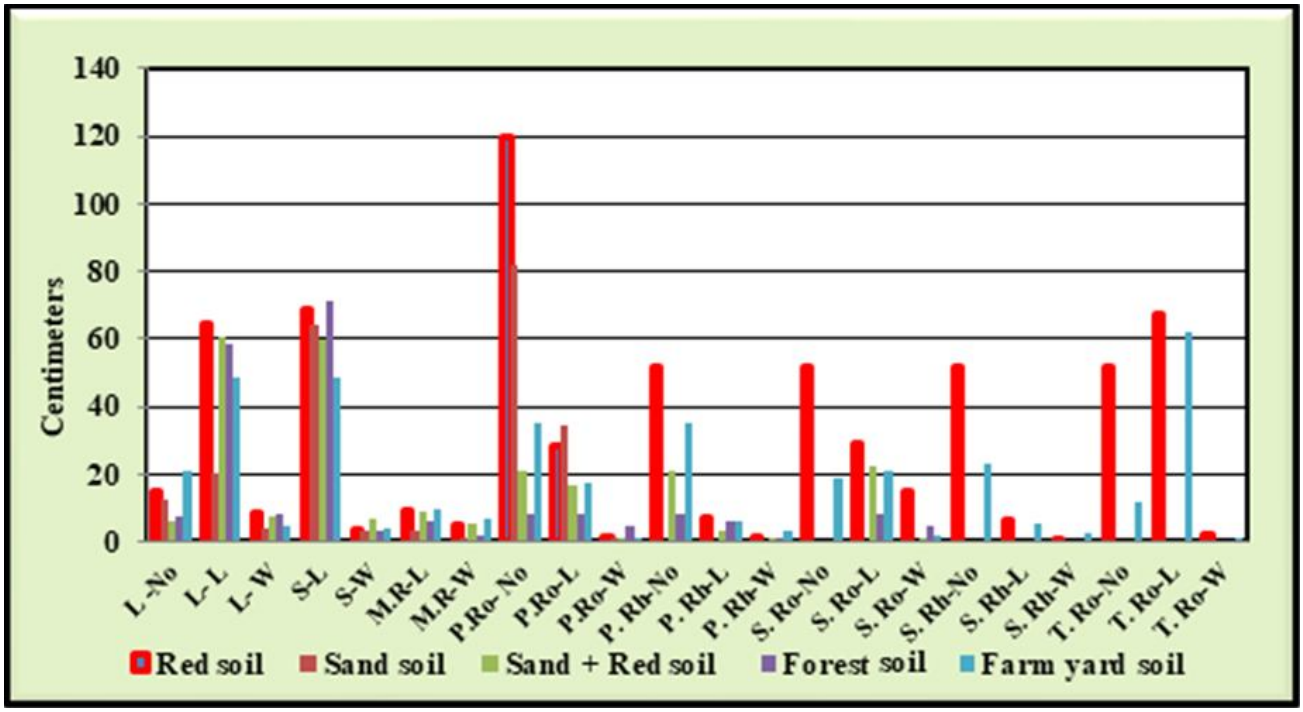


Figure 5: Photographs of obtained rhizomes and their yield.



Cl: Chlorides; **Co:** Carbonates; **Al:** Alkalinity; **Org:** Organic Matter; **WHC:** Water holding capacity

Figure 6: Physicochemical analysis of different soil samples.



L.No: Leaf Number; **L.L:** Leaf Length; **L.W:** Leaf Width; **S.L:** Scape Length; **S.W:** Scape Width; **M.R.L:** Mother Rhizome Length; **M.R.W:** Mother Rhizome Width; **P.Ro.No:** Primary Root Number; **P.Ro.L**: Primary Root Length; **P.Ro.W:** Primary Root Width; **P.Rh.No:** Primary Rhizome Number; **P.Rh.L:** Primary Rhizome Length; **P.Rh.W:** Primary Rhizome Width; **S.Ro.No:** Secondary Root Number; **S.Ro.L:** Secondary Root Length; **S.Ro.W:** Secondary Root Width; **S.Rh.No:** Secondary Rhizome Number; **S.Rh.L:** Secondary Rhizome Length; **S.Rh.W:** Secondary Rhizome Width; **T.Ro.No:** Tertiary Root Number; **T.Ro.L:** Tertiary Root Length; **T.Ro.W:** Tertiary Root Width.

Figure 7: Plant growth and development under garden conditions.

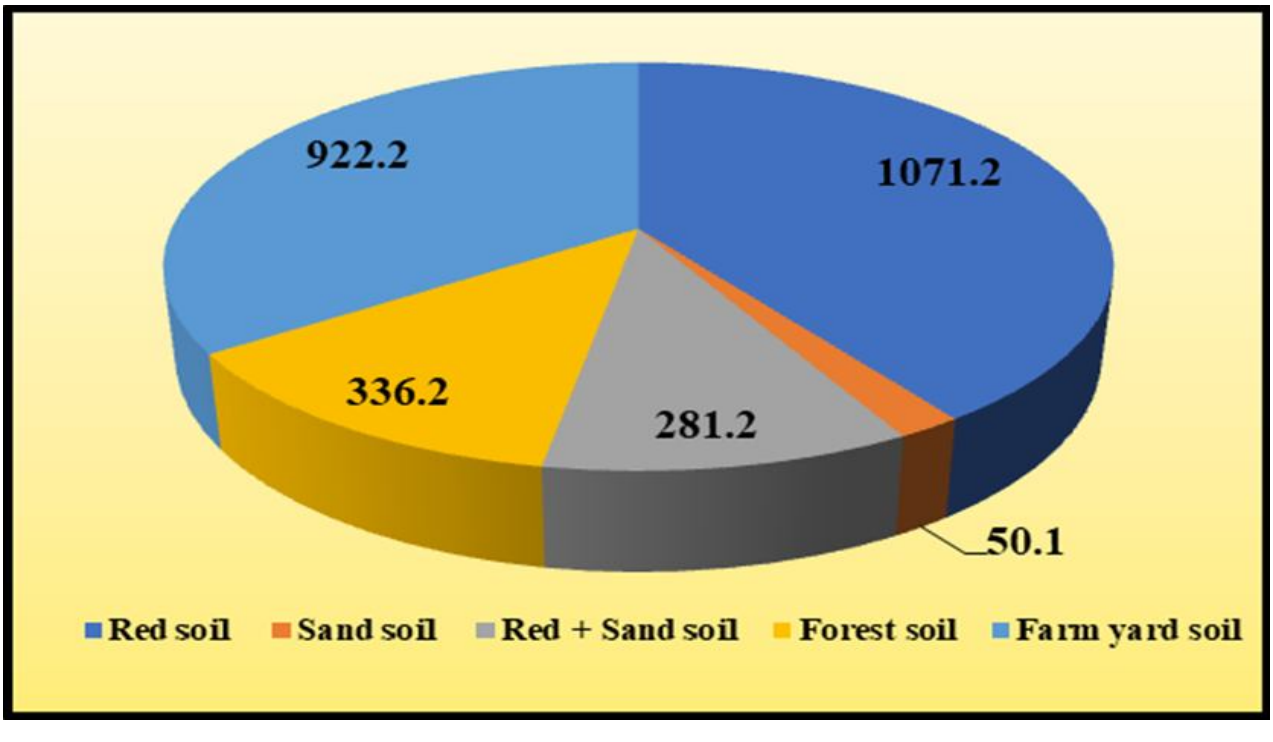


Figure 8: Graphical representation of rhizome yield.

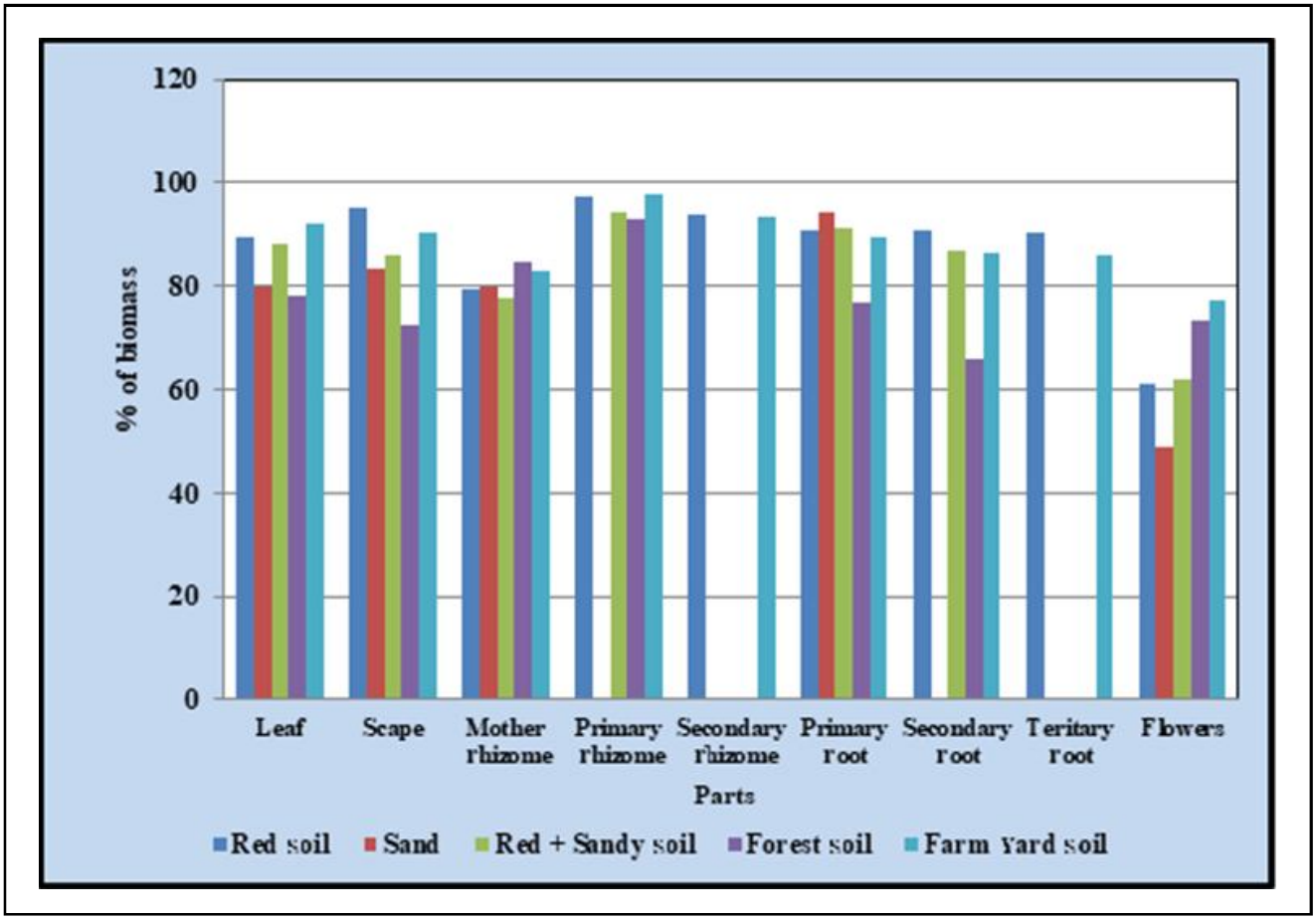


Figure 9: Graphical representation of the percentage of biomass obtained.

4. Discussion

In phenological studies of different soil samples (sand, red soil, red soil + sand, soil + organic matter) results are given below in Table 2 and Figures 1-3. Plant growth and development under different soil types and garden conditions in 90 days are presented in Table 3, Figures 4a, 4b and 7. Propagation stages under different soil conditions showed the following results in Table 4, Figures 5 and 8. Biomass (%) in different parts of the plant under different conditions showed the following results as presented in Table 5, Figure 9.

4.1 Physicochemical analysis of soil samples

The data infer that chloride content in the Kadapa sample was higher in the soil at 60.01% whereas the lowest was found in the Tirupati areas in the sand with 48.75%. The carbonate content was found to be highest in the soil from Nellore with 43.29% whereas, the lowest was in the Tirupati area sand with 30.12%. The alkalinity of Tirumala soils was 7.86 meq which was among the tested samples and the lowest was observed in the Tirupati soil at 4.56 meq. The red soil from Tirupati showed the highest content of organic matter at 4.34 meq, and the lowest content was found in the Nellore region having 3.03 meq. Further, the water holding capacity (soil moisture) of soil was found to be higher in Kadapa at 15.91% in comparison to sand with 10.02% which is found to be lowest in the regions of Tirupati. The results for the physicochemical parameters in different soil samples are presented in Table 1 and the graphical representation was shown in Figure 6.

4.2 Phenological characteristics of *C. neilgherrensis*

Critical observations were made about the phenological characteristics of *C. neilgherrensis* under garden conditions and natural habitats (Forest). Different soils like farmyard soil (with manure) and pure soils like red, red + sand, and sand were selected for the propagation studies under garden conditions (*ex situ*). Results are mentioned in Table 2 and Figures 1, 2, and 3. Initiation of the first leaf was observed after 3 days in all soils. The most suitable soil is red soil + sand (1:1) than its original habitat (Forest ecosystem). Mature plants persist up to 100-143 days in red soil + sand conditions; whereas in forest soil and farmyard soil, it is 90-120 days. Leaf average size is much smaller in sand with a length of 20.3 cm and width of 4.2 cm as compared to the red soil with a length of 64.3 cm and a width of 8.4 cm. Inflorescence developed during August to October is also varied interestingly which is noticed to be a longer period in the sand for 80-100 days following red soil + sand for 70-75 days and red soil, forest and farmyard soils up to 60-75 days. 4-6 number of flowers per inflorescence found in forest (Natural habitat) soil which is the highest number compared to 2-4 flowers in remaining all combinations of soils. Flower longevity was more in the forest soil which is for 3 days following 2 days in other soils and, 1 day in the sand condition which was the shortest among all. Opening of the flower was observed to be shortest in red soil, *i.e.*, 9:10-9:20, whereas, in farmyard soil, it was between 10:20-11:00 am, in the sand and red soil + sand observed between 9:40-10:10 am. Fruit development took 50 days in the sand

which was the longest period whereas, in the rest of all the soils, fruit development occurred in 30-45 days. Based on these results, red soil, and farm yard soils are observed to be the most suitable soils for better cultivation practices.

4.3 Propagation studies - growth and development of *C. neilgherrensis* under different soil conditions

Growth and development of *C. neilgherrensis* under different soil conditions, *i.e*., red soil, sand, red soil + sand (1:1), forest/natural/ habitat (clay loamy soil), and farmyard was recorded daily for 90-120 days throughout the growth period of the plant until its full maturity which is presented in Table 3, Figures 4a, 4b and 7. Growth and development studies were initiated with the collection of mature rhizomes. After the collection of mature rhizomes, a few rhizomes were kept open under normal conditions and dried after 10-12 days to test the viability of the rhizomes. The viability of rhizomes persisted for a maximum of one week under natural conditions and up to 90-120 days kept under controlled temperatures (cold storage 10-18^{0}C). Few of the mature rhizomes of 6-8 cm diameter from the natural source, were sowed in different soils. The initiation of the leaf was observed after 6-7 days with a regular sprinkling of water. Growth rates were recorded as the height of the scape, total number of leaves, leaf and scape length and width, rhizome and adventitious root size (L × W) after 90 days also recorded and observed the growth of conspicuous primary and secondary rhizomes with interesting finger-like secondary and tertiary roots.

Under natural (*in situ*) conditions flowering stage of *C. neilgherrensis* was observed between August and October after the cessation of vegetative growth. Leaf and scape development was observed between December to March. During the flowering period, mother rhizomes and roots were reduced in size and shrank without primary, secondary and tertiary roots and rhizomes. Vegetative growth started in October and persisted up to December and full growth was reached by March and April with a maximum of 13 leaves. The rhizomes were collected before the leaf shed stage and also observed full growth of the rhizome and roots along the primary rhizome.

Under garden conditions (*ex situ*) growth dynamics were observed in the selected 5 soils after 90 days, *i.e.* from the end of April to July and the flowering period persisted till the end of August. The total height of the scape was highest in red soil, *i.e*., 157.4 cm followed by farm yard soil (149.3 cm), red soil + sand (140.2 cm), forest soil (139.3 cm), and sand (82.4 cm). The highest number of leaves per plant observed was 21 in the farm yard soils and the lowest number of leaves, *i.e*., 6 leaves observed in red soil + sand. Based on the leaf characteristics, plants grown under red soil showed the highest leaf size with a length of 64.7 cm and 8.4 cm in width, whereas the leaf size was lowest in sand with a length of 20.2 cm and width of 4.1 cm. The scape was also observed to be highest in forest soil with a length of 71.3 cm and 3.5 cm in width and lowest in farm yard soil with 48.3 cm in length and 3.8 cm in width.

Mother rhizome was observed to be highest in size in farmyard soil measuring 9.3 centimeters in length and 6.8 centimeters in width followed by red soil 9.2 centimeters in length and 4.6 centimeters in width and lowest size was observed under sand conditions with 3.1 cm length and 0.7 cm width. Primary roots were observed to be highest in red soil nearly about 120 per plant and least in forest soil having only 8 roots. Primary rhizomes developed in all the soil conditions except in sand, 52 rhizomes formed in red soil followed by 35 rhizomes in farm yard soil, 21 rhizomes in red soil + sand, and 8 rhizomes in forest soil. Further, the growth observations extended to the secondary roots. Secondary rhizomes and tertiary roots in red soil were 52 each, whereas in the farmyard 35 secondary roots and 23 secondary rhizomes with 12 tertiary roots were observed. Primary, secondary rhizome, and secondary, tertiary roots were not found in sand, but it was noticed that the well-developed primary root with a 28.6 cm length and 1.1 cm width; secondary root showed a 29.0 cm length and 1.5 cm width was present in sand soils. In red soil, the primary rhizome was found to be 7.2 centimeters in length and 1.1 centimeters in width; the secondary rhizome was observed to be 6.7 cm in length with 0.9 cm in width and the tertiary root showed a measurement of 7.5 cm in length and 2.3 cm in width. Whereas secondary roots in farm yard soils measured about 2.3 cm in length and 1.3 cm in width. The results confer that the red soil is more suitable for the propagation and cultivation of *C. neilgherrensis* compared to all the other soil conditions.

Rhizome yield observations made in 5 selected soils state that red soil is highly suitable for rhizome growth and development and resulted in a high yield of rhizomes, *i.e*., 1071.2 g per plant followed by the farm yard soil (922.2 g), forest soil (Tirumala) (336.2 g), red soil + sand (281.2 g) and sand soil which yielded only 50.1 g having only a mother rhizome and primary roots (Table 4, Figures 5 and 8).

Fresh weight of *C. neilgherrensis* leaf, scape, mother, primary and secondary rhizomes and primary, secondary, and tertiary roots were recorded. Dry weight and the percentage of biomass were observed as the highest (97.2%) in the primary rhizome; followed by the secondary rhizome (93.7%); mother rhizome (79.3) and flower (61.2%). An average of all roots amount showed 90.9%; scape 95.2% and leaf 89.5%. There was not much water loss of *C. neilgherrensis* plant observed in the red soil plants resulting in higher biomass after the red soils, biomass content was higher in farm yard soils followed by red soil + sand, forest soil, and sand. Although, the plant biomass was least in the sand conditions, mother rhizome and primary root biomass were highest at 79.8% and 94.2%, respectively, in the sand (Table 5 and Figure 9).

According to a study (Sajitha *et al*., 2014), the plant height of *C. amada* and *C. aromatica* after 180 days noticed as 83.25 and 78.75 cm. Leaf number had showed a decreased trend. The rhizome yield and dry recovery increased by the age as 398.5 and 274.0 g after 180 days of planting. Maximum essential oil yield was noticed after 90 DAP, *viz*., 4.42% and 6.98%; Curcumin yield was 0.047% and 0.06%, respectively, in *C. amada* and *C. aromatica* (Sajitha *et al*., 2014).

When the *C. neilgherrensis* rhizome yield, biomass and growth rates from the current study are compared with the other *Curcuma* species, it was observed that the results of our study were on par with the published data. In current study, the plant height was 82.4-157.4 cm whereas other species height was evaluated for *C. zedoaria* was of 135-150 cm in height, *C. malabarica* 100-120 cm of height; *C. raktakanta* and *C. sylvatica* height as 100-125 cm. However, *C. longa* height was 73-79 cm which was very shorter when compared to *C. neilgherrensis*. Rhizome yield (fresh weight) (1.00-1.20 kg/ plant) was on par with all *Curcuma* species but it was three times higher than *C. longa* (0.435 kg/plant); however, the rhizome dry matter was observed to be 55-60% higher for *C. neilgherrensis* than the other *Curcuma* species and it was observed to be 14.31% higher than in *C. longa* as accordingly reported in the Annual Report 1999-2000 of Central Crop Research, Tropical Minor Tuber Crops.

5. Conclusion

The cultivation and propagation studies of *C. neilgherrensis* complete the total life cycle within a month, especially by the end of August to April. Initiation of the inflorescence to the maturation of inflorescence was observed from August to October. From the first leaf to the complete plant was developed from February to April. Rhizomes start sprouting at high temperatures during the pre-monsoon showers. Variations in flowering time and pollinator activity hold ecological significance, potentially leading to differences in pollination efficiency and consequently diverse reproductive outcomes across populations. There is an immense need to conserve the rare medicinal plant *C. neilgherrensis* both *in vivo* and *in vitro* propagation methods for future generations.

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Conflict of interest

The authors declare no conflicts of interest relevant to this article.

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