ISSN PRINT 2319 1775 Online 2320 7876

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Total Chlorophyll Determination in Leafy Vegetables Cultivated in Hydroponics and Soil

Labya Prabhas[1], Dr. Amia Ekka[2]

[1]Assistant Professor, School of Studies in Life Science,
Pt. Ravishankar Shukla University, Raipur, Chhattisgarh, India.
[2]Professor, School of Studies in Life Science, Pt. Ravishankar Shukla University, Raipur, Chhattisgarh, India.

ABSTRACT:

There are many factors that can be used to describe the growth outline of the plant. Qualitative and quantitative estimation of phyto-chemical composition of the plant's can directly reflects the growth pattern. These may also reflects the nutraceutical values of the plant for human consumption. Selected plant species are leafy vegetables and popular among the people of central India. Cultivation of selected plant's species is carried out in two different ways. Traditional method of plant cultivation includes soil based cultivation and other is hydroponic technique. In hydroponics, there is no need of soil, liquid media remains in direct contact with the seed and root of the plant. Hydroponically grown *S. oleracea* L (1.447 mg/g) was recorded with highest amount of chlorophyll, followed by *M. arvensis* (1.338 mg/g), *C. sativum* (1.162 mg/g), *T. F. graceum* L. (1.097 mg/g), *C. olitorius* L. (1.060 mg/g), *A. viridis* (0.917 mg/g) and *C. arietinum* (0.643 mg/g). On the other hand total chlorophyll content in soil cultivated plants was found highest in *M. arvensis* (1.206) followed by *S. oleracea* L. (1.085), *C. sativum* (1.046 mg/g), *T. F. graceum* L. (0.906) ,*C olitorius* L. (0.859 mg/g), *C. arietinum (*836 mg/g) and *A.viridis* (0.794). This study may reveal the compatibility and acceptance of hydroponics for plant cultivation. Chlorophyll content was consistently high in most of the experimental plants cultivated in hydroponic system as compared to soil cultivated plants.

Keyword: Phyto-chemical, neutraceutical, hydroponics, total chrolophyll, protein content, carbohydrate, recognize, suitable.

INTRODUCTION:

Plant is composed of various type of light capturing pigment like chrolophyll, carotenoid and much other type of pigments. Chlorophyll is an important fraction of photosynthetic machinery. Amount of chlorophyll directly represent the number of chloroplast in plant cell. Richness in chlorophyll molecules is responsible for capturing sunlight and conversion into sugar compound. Hence, optimum rate of production of sugar inside plant cell mainly depends upon amount chlorophyll molecule. If optimum sugar is produced and stored by plant then this will result imitate optimum growth of plant too. It means chlorophyll is an important tool which is directly associated with growth of the plant. Chlorophyll is found in

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the chloroplast and morphologically we can observe mainly in the green part of the leaves. Some amount of chlorophyll is also present in stem, flowers and roots also. There are seven general types of chlorophyll found in nature; chlorophyll a, chlorophyll b, chlorophyll c, chlorophyll d, chlorophyll e and chlorophyll f. But chlorophyll and a (composed of side chain with $–CH_3$ gp) and b (composed of side chain with –CHO gp) are most important part of photosyst em and they are essentially required for photosynthesis in green plants.

In this study we can observe the effect of changing plant cultivation method over chlorophyll synthesis. Here hydroponic technique is used for cultivating plant species of edible varieties. This may also result into compatibility check of hydroponics for selected plant cultivation. Higher productivity along with healthy plant body may reflect higher probability of acceptance of hydroponics. On the other hand if any alteration in anatomy and morphology of plant body occurs due to change in cultivation technique this would also indicate the rejection or acceptance of the technique applied. Increase in number of chlorophyll or no change in chlorophyll with some other advantages will allow for the acceptance of hydroponic technique. Selected species of the plants are highly popular among the people of state Chhattisgarh in central India and all neighboring states like Orrisa, Maharashtra, and Madhya Pradesh (MP) for their daily consumption. *Amaranthus viridis,* (Chaulai), *Trigonella foenum graceum L.*,(Methi), *Chorchorus olitorius L.,* (Chech), *Coriandrum sativum* (Coriander), *Mentha arvensis* (Mint), *Cicer arietinum* (Gram), *Spinacia oleracea* L (Palak). These selected green leafy vegetable plant species are also holding therapeutical and ethano-botanical values along with nutritional importance.

Cultivation of common but popular edible green leafy plants through novel technique without altering anatomy and morphology may be of interesting concern and challenging. Studying structure, shape, size and weight of morphological and anatomical parts of the plant body at various stages of plant growth may disclose similarity and dissimilarity between soil and hydroponically cultivated plants. Here we can test the total chlorophyll content of mature and edible part of the plant at adult stage. Adult stage means that leafy vegetable plant is ready for consumption as nutritional source. Food security is a major concern for global community. There is a strong need of modification in old traditional agricultural methods by scientific but simple approaches. Increasing global population, climate change and decreasing agriculture land are some an indicative of threat to food security for future generation. To overcome the problem there is a requirement of collaborative effort from all part of human society including researchers, farmers, educationist and local people also.

METHDOLOGY:

A small scale setup for compatibility check for selected plant species (seven in no.) cultivation in hydroponics has been maintained in glasshouse with modern temperature and humidity controlling devices. Another setup was established in open environment suitable for agriculture. Healthy seeds from the same batch was selected and used for sowing in both

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hydroponics (indoor) and soil based (outdoor) method. The temperature was maintained up to 25-30 ± 5 °C. Humidity was around 80 % ± 10 throughout the period inside the glasshouse. In soil based cultivation temperature was recorded up to 30-35 ± 5 °C in day time and it was recorded 20-25 ± 5 °C at night. Some of the selected plants are seasonally available hence this study was carried out in winter season (November-December). This period is most preferred time interval by agriculturists and plant cultivators of leafy vegetable type in Raipur, Chhattisgarh (Central India). Availability of all selected species in local market area was recorded.

DMSO method

Dimethyl sulfoxide is a colourless liquid used for paper making and other commercial purposes. Leaves were collected from healthy plants and veins were removed with the help of sterilized surgical blades. After surface sterilization tissue samples of 1gm leaf was taken and cut into small pieces and collected in separate test tube/petri dish. All samples with 10 ml solvent were incubated in water for 1 hr at 60 °C ± 5 °C. A colorless solution was found after completing the process. All samples were filtered after proper cooling till emergence of room temperature. Absorption was measured at 663 nm (A_{665}) and 645 nm (A_{648}). DMSO solvent was used as blank. UV-VIS Spectrophotometer of SHIMADZU (UV-1900) was used for determination of absorbance. (Manolopoulou *et al*., 2016)

Calculation is done according to following equation – (Kumari *et al*., 2018)

Chlorophyll a (mg/g) = 12.7 x A_{663} – 2.69 A_{645}

Chlorophyll b (mg/mg) = 22.9 x A_{645} – 4.68 A_{663}

Total chlorophyll (mg/g) = 20.2 A_{645} + 8.02 A_{663}

Where: A_{663} is absorption value at 663 nm

A_{645} is absorption value at 645 nm

Morphological Observation:

Morphological studies and analysis was done after regular interval of 24 hr. Appearance of new parts and noted. It was confirmed by measurement of change in weight using weighing balance (SCALETEC. Model CWS-203L) and altered physiology of sown seed in both soil and hydroponics is minutely observed. Initiation of seedling was defined by appearance of small curved structure and rupture of seed coat. Leaf initiation was observed after the growth of longitudinal stem. Leaves were collected in the initial stage of the harvesting time for chlorophyll estimation.

ISSN PRINT 2319 1775 Online 2320 7876

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RESULT AND DISCUSSION:

Table 1: Showing spectrophotometric estimation of total chlorophyll determination in leafy plants cultivated in hydroponic system.

Scientific name of the selected plant species.	Common Name	Wavelengths		Chlorophyll A (mg/g)	Chlorophyll B (mg/g)	Total Chlorophyll (mg/g)
		663 nm	645 nm			
Amaranthus viridis	Chaulai Bhaji	0.492	0.198	0.720	0.196	0.917
Cicer arietinum	Chana Bhaji (Gram)	0.509	0.212	0.339	0.304	0.643
Chorchorus olitorius L.	Chech Bhaji	0.522	0.318	0.577	0.483	1.060
Coriandrum sativum	Dhaniya (Coriander)	0.712	0.293	0.825	0.337	1.162
Mentha arvensis	Pudina (Mint)	0.811	0.341	0.938	0.401	1.338
***Spinacia oleracea* L.**	Palak (Spinach)	0.818	0.392	0.933	0.514	1.447
Trigonella foenum graceum L.	Methi Bhaji (Fenugreek)	0.588	0.310	0.663	0.434	1.097

Table 2: Showing spectrophotometric estimation of total chlorophyll determination in leafy plants cultivated in Soil.

Scientific name of the selected plant species.	Common Name	Wavelengths		Chlorophyll A (mg/g)	Chlorophyll B (mg/g)	Total Chlorophyll (mg/g)
		663 nm	645 nm			
Amaranthus viridis	Chaulai Bhaji	0.492	0.198	0.571	0.223	0.794
Cicer arietinum	Chana Bhaji (Gram)	0.509	0.212	0.589	0.247	0.836
Chorchorus olitorius L.	Chech Bhaji	0.522	0.218	0.604	0.254	0.859
Coriandrum sativum	Dhaniya (Coriander)	0.552	0.299	0.620	0.426	1.046
Mentha arvensis	Pudina (Mint)	0.721	0.311	0.832	0.374	1.206
***Spinacia oleracea* L.**	Palak (Spinach)	0.618	0.292	0.706	0.379	1.085
Trigonella foenum graceum L.	Methi Bhaji (Fenugreek)	0.532	0.238	0.611	0.296	0.906

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Hydroponically grown *S. oleracea* L. (1.447 mg/g) was recorded with highest amount of chlorophyll, followed by *M. arvensis* (1.338 mg/g), *C. sativum* (1.162 mg/g), *T. F. graceum* L (1.097),*C. olitorius* L. (1.060), *A. viridis* (0.917), and *C. arietinum* (0.643) (Table 1). On the other hand total chlorophyll content in soil cultivated leafy plant's species was found highest in *M. arvensis* (1.206) followed by *S. oleracea* L. (1.085), *C. sativum* (1.046 mg/g), *T. F. graceum* L. (0.906), *C. olitorius* L. (0.859 mg/g), *C. arietinum* (0.836 mg/g) and *A. viridis* (0.794) (Table 2). This study may reveal the compatibility and acceptance of hydroponics for plant cultivation. Chlorophyll content was consistently high in most of the experimental plants cultivated in hydroponic system as compared to soil cultivated plants (Table 1and Table 2). One plant species of *C. arietinum* was an exception among all other species after applying similar conditions in both techniques.

Table 3: Showing morphological observation in selected leafy plants.

Scientific name of the selected plant species.	**Seed initiation (days after sowing seed)**	**Leaf initiation time (days after sowing seed)**	**Seed initiation (days after sowing seed)**	**Leaf initiation time (days after sowing seed)**	**Time to obtain young leaves after sowing (days)**
	Hydroponics		**Soil Cultivation**		**Soil& Hydroponic**
Amaranthus Viridis	**6**	**15**	**9**	**27**	**35**
Cicer arietinum	**4**	**8**	**14**	**16**	**30**
Chorchorus olitorius L.	**2**	**3**	**4**	**5**	**15**
Coriandrum sativum	**17**	**20**	**20**	**22**	**32**
Mentha arvensis	**11**	**15**	**15**	**22**	**30**
***Spinacia oleracea* L.**	**6**	**9**	**10**	**13**	**20**
Trigonella foenum graceum L.	**2**	**5**	**4**	**7**	**20**

Morphological evidences reveal that there is a significant difference between hydroponic and soil cultivated plants. In all seven experimental models i.e. *A. viridis*, *C. arietinum*, *C. oliotorius* L., *C. sativum*, *M. arvensis*, *S. oleracea* L. and *T.F. graceum* L. seed initiation and leaf initiation time was rapid in hydroponics (Table 3). While on the other hand seed initiation and leaf initiation time was found under standard harvesting time as reported earlier in various studies and literature reviewed. Rapid growth time and short life cycle is indicating high compatibility of hydroponic towards cultivation of leafy vegetables. A apparent effect of

ISSN PRINT 2319 1775 Online 2320 7876

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higher amount of water availability and nutrient supports rapid seed initiation and early leaf initiation in almost all the cases. Hence, resulting into short life cycle and reduced harvesting time of the plant. In terms of economical and neutraceutical values hydroponic system was significantly effective and compatible with selected leafy plant varieties. Still there is need to put more effort by researchers. Many other phyto-chemicals estimation like carotenoids, protein, carbohydrates etc. is also important to ensure the efficacy of the applied method for plant cultivation.

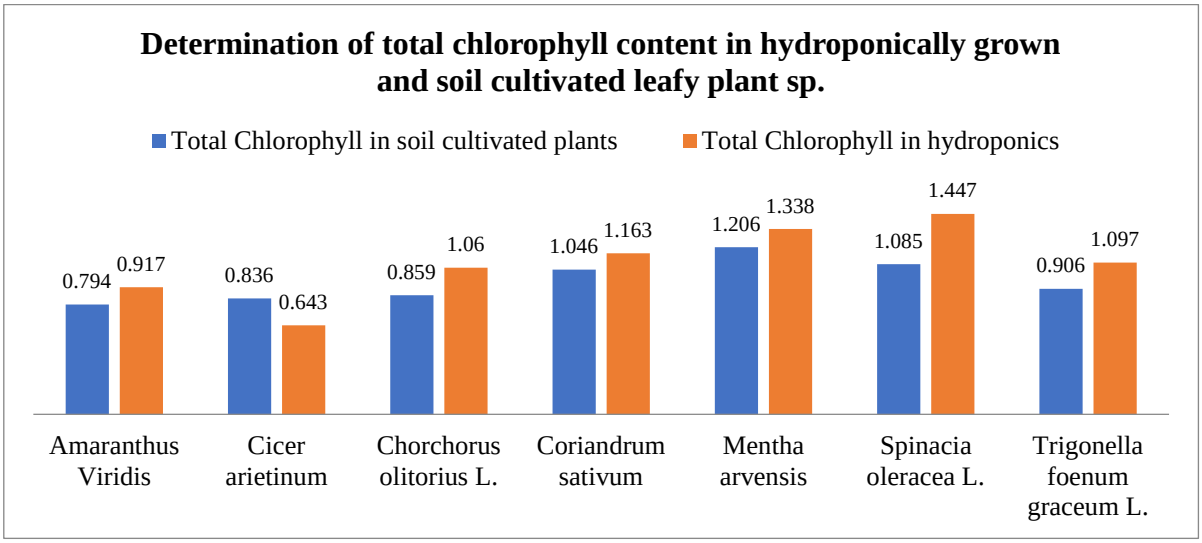

Fig. 1: Comparative analysis of total chlorophyll content in hydroponically grown and soil cultivated leafy plant's species.

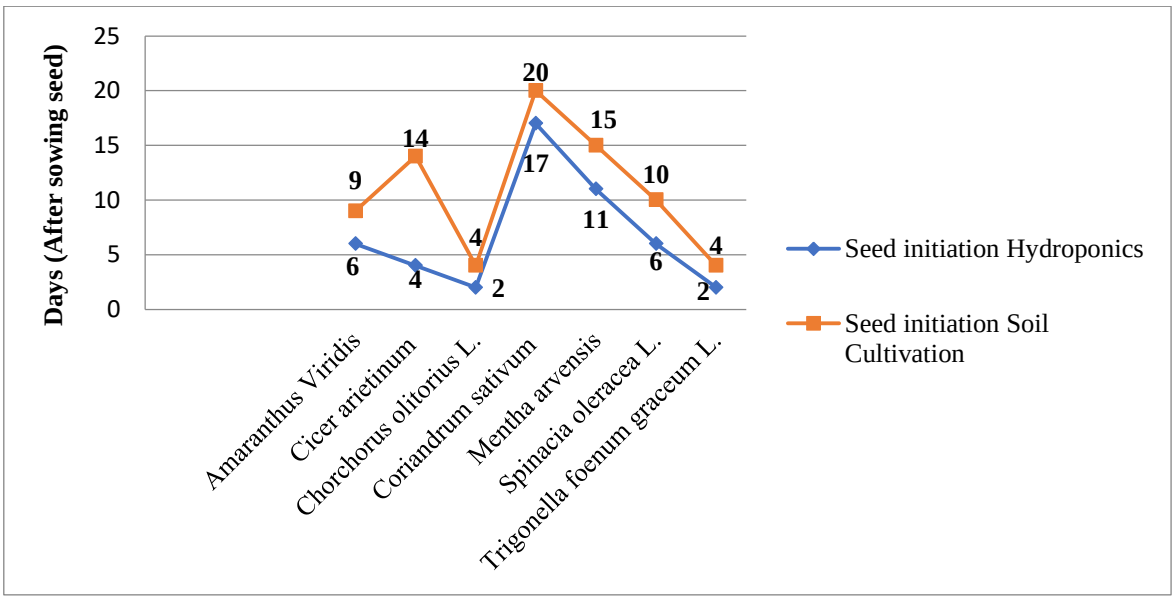

Fig. 2: Comparative analysis of seed initiation time in hydroponically grown and soil cultivated leafy plant's species.

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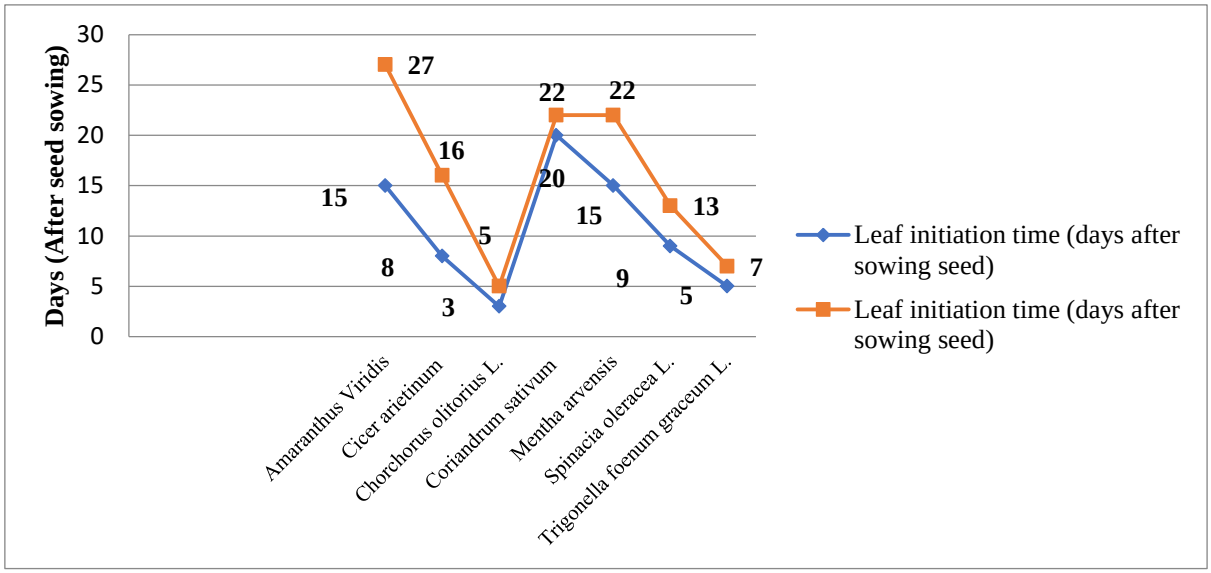

Fig. 3: Comparative analysis of leaf initiation time in hydroponically grown and soil cultivated leafy plant's species.

ACKNOWLEDGEMENT:

I am deeply indebted to Dr. Megha Agrawal, Assistant professor, Gurukul Mahila Mahavidyalaya, Raipur (India) for his valuable suggestions and encouragement during this project. I am also very thankful to Mr. Suresh sonkar and all technical staff of SoS in life Science, Pt. RSU, Raipur (Chhattisgarh - India) for their continuous support also. I would also like to take this opportunity to acknowledge everyone who has helped us directly or indirectly in every stage of this experimental study.

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