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"HYDROPONIC PROPAGATION OF SELECTED MICRO GREENS AND THEIR ESTIMATION OF PROTEIN CONTENT"

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ABSTRACT:

The purpose of this study was to propagate micro greens using soil less culture or hydroponic deep flow culture and their protein estimation. We selected 9 varies seeds for micro green cultivation in four different solutions NPK, vermiwash, jeevamrutham they are and panchagavya. We observed that Vigna unguiculata in jeevamrutham showed highest growth rate on the seventh day (23.2±0.18cm) followed by panchagavya, vermiwash and NPK. No growth was seen in jeevamrutham of *Brassica nigra* and *Eleusine coracana*. We also conducted protein estimation for selected micro greens resulted that Vigna radiate showed high protein value of 0.86 in NPK. Finally, we observed that cultivation of micro greens using both organic and inorganic solutions in deep flow hydroponic technique (DFT) is an effective technique to provide high growth rate and protein content for the prevention of nutritional deficiency in human populations.

Keywords: Hydroponics, Deep Flow Technique, Micro greens, NPK and Estimation of proteins.

1. INTRODUCTION

Hydroponic or soil-less culture is a system or technology of growing plants in nutrients solutions that supply all nutrients elements required for successive plants growing with or without the use of an insert medium such as coir dust, gravels, rock wool, vermiculite, peat moss, coco peat and saw dust etc... to provide mechanical support. It is a more rational use of water resources, to provide better opportunities for a sustainable food supply in both developing and developed countries. It was practiced in Babylon, Amazon, Egypt, China and India (Besthorn, 2013). The term 'hydroponics' is derived from two greek words i.e. "Hydro" and "Ponos" means water and labour respectively (Beibel, 1960). The first modern use of hydroponics was done by W.F. Gericke from the University of California during the 1930's. In India, Hydroponics was introduced in year 1946 by an English scientist, W.J. ShaltoDuglas. He established a laboratory in Kalimpong area, West Bengal and had written a book on Hydroponics, named as 'Hydroponics- the Bengal System' (Pant et al., 2018). Various commercial and special crops can be grown using hydroponics including leafy vegetables, cucumber, tomato, pepper, strawberry and many more. Europe is considered the biggest market for hydroponics in which France, the Netherlands and Spain are the three top producers, followed by the United States of America and Asia-Pacific region (Prakash et al., 2020). Now a days number of hydroponicstechniques are available for the production of micro greens but deep flow technique or pipe system is a one of the circulatory system easy vertical farming save water, power, easy set up and reliable for to (https://puregreensaz.com/blog/benefits-of-deep-flow-technique-dft/).

Microgreens, also known as young vegetable greens, shoots and aromatic plants eat in the stage of cotyledon leaves or when they have developed their first pair of leaves from seedlings. They are a new type of colourful greens that were first well liked in high-end restaurants and markets and have become a new creativitytrend in the last years. Over the years, the interest for micro greens has been increasing and today there are over 80 species on the market (Xiao *et al.*, 2013). Micro greens are commonly three to ten centimetres in height they are picked at seven to fourteen days after germination and sold as whole plant moss. They are valued for their wide range of colours, strong flavours, shapes, textures and nutrient rich such as used as ingredients in soups, burgers, desserts, salads and sandwiches and also for seasoning of variety of dishes in restaurant's and also easy to grow, quick to harvest, rich nutrients help for different diseases (Kim *et al.*, 2009; Millard *et al.*, 2014; Murphy *et al.*, 2010 & Threadwell *et al.*, 2010).

2. MATERIALAND METHODS

Set up of Deep Flow Technique& Cultivation of selected Micro greens

Basic principles of hydroponic Deep Flow Technique (DFT) is a nutrient solution circulates continuously for 24 hours on a closed flow circuit. Plant nutrient solution in the tank is pumped by the water pump to the tub through a network of irrigation pipes planting, and then a solution of plant nutrients in planting tubs fed back into the tank. For Deep Flow Technique of hydroponic unit setup, we used some materials. These materials are divided in to physical, chemical and biological materials. Physical materials are including reservoir, PVC pipes, net pots, electrical circuit, timer, coco coir and coco peat. Chemical materials are used as some nutritional solutions. One is inorganic and another three are organic solutions. Inorganic solution is NPK and organic solutions are vermiwash, jeevamrutham and panchagavya. Biological samples are seeds they are *Brassica nigra* (L.) Czern., *Cicer arietinum* L., *Eleusine coracana* (L.) Gaertn, *Macrotylo mauniflorum* (L.) verdc, *Trigonella foenumgraecum* L., *Triticum aestivum* L., *Salvia hispanica* L., *Vigna radiata* (L.) Wikzek and

Vigna unguiculata (L.) walp. From different families were procured from local ritubazar market in Kurnool.

Initially we took 6 feet of an iron rack for supporting a hydroponic unit. Then for the PVC setup, the flg length had been made 2 inches pipe of diameter is cut into 40 inches length of 8 pieces. To this, reducers are attached at both the ends. Drill holes into these pipes with 1-2 inches distance to insert the net pots. Now to the reducers elbows are attached. The rows of 1st horizontal lines are connected to each other's with elbows, so that the inlet of water from the motor is supplied. Now, the others end of 1st horizontal line are connected to the successive horizontal lines with 30 cm PVC pipes. Likewise, the entire hydroponic setup is connected. A plastic drum of 5 liters capacity is taken as a reservoir in which nutrients used for DFT were dissolved in it. A small motor having capacity of 20 volts is placed in it for the hydroponic setup with the help of plastic pipe of 1m and operation with electricity. Finally, the last 2 rows of the horizontal pipe lines at one end are left free into the water reservoir. So, that the circulated water is drained/flows back. Now in the reservoir motor is immersed and power supply is given to it. Soak few seeds in water over night for swelling. We have to take netted plastic pots and used coco coir over the surrounding to avoid holes. Reservoir drum is 50 litters capacity filled with the nutritional solution of 40 litres. For 1liter water 1gm of NPK is used as a measurement. Next nutrition solution we used 10 ml of organic vermiwash, jeevamrutham and panchagavya was used as a measurement.

In hydroponics system, plants roots were suspended in nutrient-rich water so that they could grow without the use of any chemicals. The whole experiment was taken almost 3 months to complete. The growths of plants were measured for 7days. The height of plants was measured to indicate the difference in growth (Mariyappillai *et al.*, 2020 & https://howtostartfarmingbusinesshome.wordpress.com/2020/01/04/what-is-dft/).



Figure: 1 Tolls and nutrient solution for DFT system Figure: 2

Figure: 2 Deep flow technique unit

Estimation of total protein content

Selected micro greens were harvested from DFT unit and cleaned with tap water immediately for surface cleaning and removal of coco peat particles. After that individual samples were transferred to beakers containing distilled water. 1gm of micro green samples were measured and macerated using mortar pestle and phosphate buffer saline (PBS) was added to make up final volume of 20ml. Then extracted solution was centrifuged at 10000 rpm for 10 minutes. After centrifugation supernatant is transferred to 25 ml of conical flask. Standard curve is obtained by obtaining and plotting graph for absorbance value of Bovine serum albumin (BSA) as standard reagent. 1 ml of extracted supernatant of sample was added to reagent A with 48 ml of 2% sodium carbonate prepared in 0.1N sodium hydroxide, 1ml of 1% sodium potassium tartarate and 1ml of 0.5 % copper sulphate. Then incubate for 15 mintues 0.5 ml of

reagent B with water and Folin Cioclteau reagent in ratio of 1:1. After 30 minutes of incubation absorbance was measured at 660 nm for the estimation of protein in the selected micro greens (Ghaly and Alkoiak 2010; Nomura *et al.*, 2019 & Sarkar *et al.*, 2020). **Statistical Analysis**

Data are represented as mean values plus or minus standard deviation calculated from triplicate determinations.

3. RESULTS AND DISCUSSION

We have used two different nutrient solutions, first one is organic nutrient solution and the later, is inorganic nutrient solution. Inorganic solution is NPK and organic solutions are vermiwash, jeevamrutham and panchagavya for the cultivation of varies micro greens hydroponically using DFT method. The results are tabulated below.

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Hydroponic propagation of Cicer arietinum L.	Hydroponic propagation of Salvia hispanica L.	Hydroponic propagation of Vigna anguiculata (L.) Walp		
Hydroponic propagation of <i>Cicer arietinum</i> L. NPK Vermiwash Jeevamrutham Panchagavya	Hydroponic propagation of Subia hispanica L. NPK Verminash Jeevamrutham Panchagavya	Vigna unguiculata (L.) Walp		
Cicer arietinum L.	Salvia hispanica L.	Vigna unguiculata (L.) Wa lp		
Cicer arietinum L. NPK Vermivash Jeevamrutham Panchagavya Day-3 Day-6 Doctor Doctor Doctor	Sabia hispanica L. NPK Vernivash Jevanruthan Panchagaya	Figna anguiculata (L.) Walp NPK Verniwash Jeevamruthan Panchagayya		

Figure: 3 Hydroponic propagation of selected micro greens in DFT system

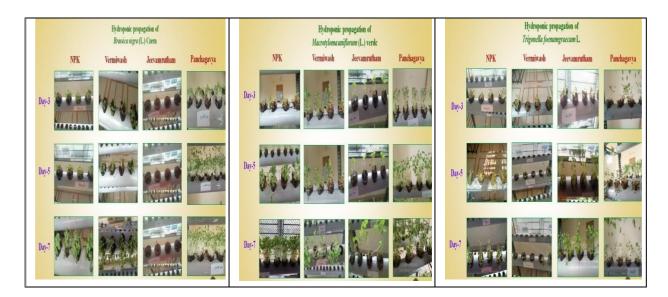


Table 1: Growth rates of micro greens in different nutrient solutions in day 3

Micro greens	Growth rates in Centimetres			
	NPK	Vermiwash	Jeevamrutham	Panchagavya
B. nigra	2.3±0.14	2.3±0.14	0±0.00	2.4±0.08
C. arietinum	1.3±0.20	0.6±0.14	1±0.10	1.3±0.31
E. coracana	1.8±0.12	1.8±0.12	0±0.00	1.5±0.08
M. uniflorum	3.7±0.15	3.3±0.11	4.5±0.33	4.3±0.20
S. hispanica	1.3±0.25	1.1±0.11	2±0.14	1.3±0.12
T. aestivum	2.4±0.25	3.2±0.14	3.1±0.22	2.9±0.15
T. foenumgraecum	1.5±0.08	1.2 ± 0.08	3.3±0.22	2.2±0.11
V. radiata	2.8±0.21	7±0.10	8.3±0.17	7.7±0.17
V. unguiculata	3.2±0.19	0.5±0.86	9.4±0.12	8±0.07

Note: Values are Mean \pm Standard deviation

From the above tabulated data, the growth of micro greens propagated through hydroponics with 4 different nutrient solutions of day-3 has shown that *B. nigra with* high growth of the 2.4 cm in Panchagavya and no growth in Jeevamrutham, *C. arietinum* with high growth of 1.3cm in NPK and Panchagavya with least growth of 0.6cm in Vermiwash, *E. coracana* with high growth of 1.8cm in NPK and Vermiwash and no growth in Jeevamrutham, *M. uniflorum* with high growth of 4.5cm in Jeevamrutham and with least growth of 3.3cm in Vermiwash, *S. hispanica* with high growth of 2cm in Jeevamrutham and least growth of 1.1cm in Vermiwash, *T. aestivum* with high growth of 3.2cm in Vermiwash and least growth of 2.4cm in NPK, *T. foenumgraecum* with high growth of 3.3cm in Jeevamrutham and least growth of 1.2cm in Vermiwash, *V. radiata* with high growth of 8.3cm in Jeevamrutham and least growth of 2.8cm in NPK and finally *V. unguiculata* with high growth of 9.4cm in Jeevamrutham and least growth of 0.5cm in Vermiwash.

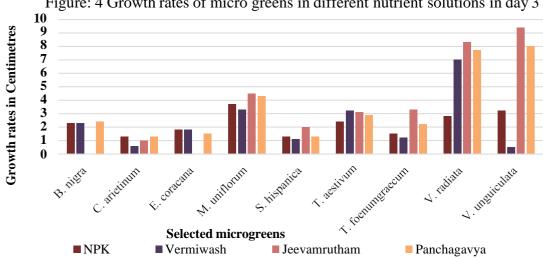


Figure: 4 Growth rates of micro greens in different nutrient solutions in day 3

Table 2: Growth rates of micro greens in different nutrient solution in day 5

Micro greens	Growth rates in Centimetres			
	NPK	Vermiwash	Jeevamrutham	Panchagavya
B. nigra	4.7±0.14	4.2±0.11	0±0.00	4.5±0.18
C. arietinum	2.2±0.21	3.2±0.46	5.6±0.25	4.5±0.05
E. coracana	4.4±0.54	2.2±0.72	0±0.00	4.2±0.20
M. uniflorum	6.8±0.07	6.5±0.22	7.5±0.35	5.4±0.14
S. hispanica	3.3±0.15	3.3±0.48	4.1±0.22	3.4±0.05
T. aestivum	8.8±0.11	9.3±0.15	9.4±0.07	12.4±0.36
T. foenumgraecum	2.3±0.19	3.2±0.14	5.6±0.18	5.4±0.11
V. radiata	9.5±0.16	9.1±0.18	9.4±0.07	12.5±0.18
V. unguiculata	10.9±0.11	4.8±5.31	19±0.18	16.9±0.12

Note: Values are Mean ± Standard deviation

From the above the data, the growth of micro greens propagated through hydroponics with 4 different nutrient solutions of day-5 has shown that B. nigra with high growth of the 4.7cm in NPK and no growth in Jeevamrutham, C. arietinum with high growth of 5.6cm in Jeevamrutham and with least growth of 2.2cm in Vermiwash, E. coracana with high growth of 4.4cm in NPK and no growth in Jeevamrutham, M. uniflorum with high growth of 7.5cm in Jeevamrutham and with least growth of 5.4cm in Panchagavya, S. hispanica with high growth of 4.1cm in Jeevamrutham and least growth of 3.3cm in NPK and Vermiwash, T. aestivum with high growth of 12.4cm in Panchagavya and least growth of 8.8cm in NPK, T. foenumgraecum with high growth of 5.6cm in Jeevamrutham and least growth of 2.3cm in NPK, V. radiata with high growth of 12.5cm in Panchagavya and least growth of 9.1cm in Vermiwash and finally V. unguiculata with high growth of 19cm in Jeevamrutham and least growth of 4.8cm in Vermiwash.

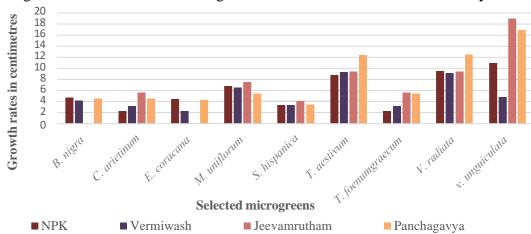


Figure: 5 Growth rates of micro greens in different nutrient solution in day 5

Table 3: Growth rates of micro green in different nutrient solution in day 7

Micro greens	Growth rates in Centimetres			
	NPK	Vermiwash	Jeevamrutham	Panchagavya
B. nigra	6.8 ± 0.08	6.5±0.11	0±0.00	8.7±0.20
C. arietinum	3.2±0.22	6.1±0.19	4.2±0.19	6.2±0.21
E. coracana	5.9±0.20	6.3±0.14	0±0.00	5±0.11
M. uniflorum	10.4±0.21	9.4±0.21	10.3±0.33	11.2±0.11
S. hispanica	3.8±0.16	4.2±0.61	4.2±0.19	4±0.11
T. aestivum	11.9±0.07	12.3±0.15	9.4±0.17	16.6±0.42
T. foenumgraecum	5.3±0.14	3.5±0.07	7.6±0.12	6.5±0.12
V. radiata	12.7±0.48	12.7±0.48	12.5±0.25	17.2±0.22
V. unguiculata	14±0.08	6.1±5.78	23.2±0.18	20.3±0.14

Note: Values are Mean ± Standard deviation

From the above the data, the growth of micro greens propagated through hydroponics with 4 different nutrient solutions of day-7 has shown that *B. nigra* with high growth of the 8.7cm in Panchagavya and no growth in Jeevamrutham, *C. arietinum* with high growth of 6.1cm in Vermiwash and with least growth of 3.2cm in NPK, *E. coracana* with high growth of 6.3cm in Vermiwash and no growth in Jeevamrutham, *M. uniflorum* with high growth of 11.2cm in Panchagavya and with least growth of 9.4cm in Vermiwash, *S. hispanica* with high growth of 4.2cm in Vermiwash & Jeevamrutham and least growth of 3.8cm in NPK, *T. aestivum* with high growth of 16.6cm in Panchagavya and least growth of 9.4cm in Jeevamrutham, *T. foenumgraecum* with high growth of 7.6cm in Jeevamrutham and least growth of 3.5cm in Vermiwash, *V. radiata* with high growth of 17.2cm in Panchagavya and least growth of 23.2cm in Jeevamrutham and finally *V. unguiculata* with high growth of 23.2cm in Jeevamrutham and least growth of 6.1cm in Panchagavya and least growth of 23.2cm in Jeevamrutham and least growth of 5.1cm in Panchagavya and least growth of 5.1cm in Panchagavya and least growth of 23.2cm in Jeevamrutham and least growth of 5.1cm in Panchagavya and least growth of 5.1cm in Jeevamrutham and least growth of 5.1cm in Panchagavya and least growth of 5.1cm in Jeevamrutham and least growth of 5.1cm in Panchagavya and least growth of 5.1cm in Jeevamrutham and Jeevamrutham and Jeevamrutham and Jeevamrutham and Jeeva

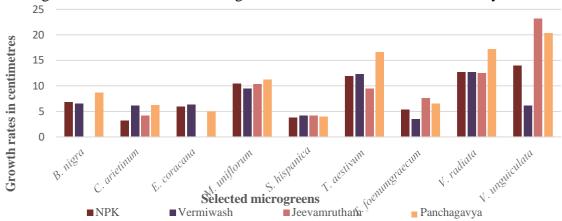
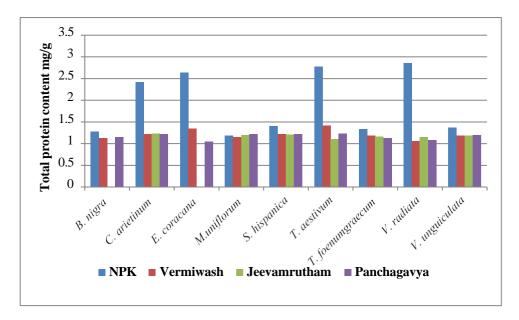


Figure: 6 Growth rates of micro green in different nutrient solution in day 7

Table4: Estimation of total	l protein content for selected micro gree	ns
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Micro greens	Total protein(mg/ 1g)			
	NPK	Vermiwash	Jeevamrutham	Panchagavya
B. nigra	1.27 ±0.56	1.13±0.94	-	1.15±0.64
C. arietinum	2.42±0.28	1.22±0.20	1.23±0.18	1.22±0.44
E. coracana	2.64±0.16	$1.34{\pm}1.22$	-	1.04±0.33
M.uniflorum	1.19±0.51	1.15±0.78	1.2±0.57	1.22±0.25
S. hispanica	1.40 ± 0.92	1.21±0.48	1.21 ± 1.06	1.22±0.73
T. aestivum	2.78±0.52	1.41±0.57	1.1±0.88	1.23±1.57
T. foenumgraecum	1.33 ± 1.28	1.18±0.92	1.16±0.54	1.13±0.48
V. radiata	2.86 ± 0.66	1.06 ± 0.98	1.15±0.58	1.08±0.66
V. unguiculata	1.37±0.27	1.18±0.47	1.19±0.56	1.20±0.74

The above tabulated data obtained after statistical analysis can be described for three different variables and total protein content of selected micro greens grown in 4 nutrients solutions for gram weight. *B. nigra* showed high protein value of 1.27 mg/g in NPK and low protein value of 1.13 mg/g in Vermiwash, *C. arietinum* high protein value of 2.42 mg/g in NPK and low protein value of 1.22 mg/g in Vermiwash and Panchagavya, *E. coracana* high protein value of 2.64 mg/g in NPK and low protein value of 1.04 mg/g in Panchagavya, *M. uniflorum* high protein value of 1.22 mg/g in Panchagavya and low protein value of 1.15 mg/g in Vermiwash, *S. hispanica* high protein value of 1.40 mg/g in NPK and low protein value of 1.21 mg/g in Vermiwash and Jeevamrutham, *T. aestivum* high protein value of 2.78 mg/g in NPK and low protein value in 1.10 mg/g in Jeevamrutham, *T. foenumgraecum* high protein value of 1.33 in NPK and low protein value in 1.13 mg/g in Panchagavya, *V. radiata* high protein value of 1.06 mg/g in Jeevamrutham and low protein value of 1.08 mg/g in Panchagavya and finally *V. unguiculata* high protein value of 1.37 mg/g in NPK and low protein value of 1.18 mg/g in Vermiwash.



Based on the above results we revealed that most of the micro greens with Panchagavya solution has shown faster growth rate as compared to Jeevamrutham, Vermiwash and NPK but high protein content was observed in NPK solution than other solutions. All micro greens in the DFT hydroponic system survived and germinated them only could die in the middle of experiment when there was shortage of nutrients in water system as well as oscillations in temperature. The crops grown in soil less culture is healthier and also more reliable than crops grown in soil. In fact, the hydroponic system requires more skills good knowledge of its principles to maintain the production and because this system depends on electricity, power outages can cause damage to the planted crops. In context to the cost, hydroponics required less money and investment as compared with soil based traditional farming regardless its saving in the run.

4. SUMMARY AND CONCLUSION

The final results showed that hydroponic DFT system has a good growth effect in the Panchagavya nutrient solution. Based on the above observations we concluded that Panchagavya solution has good growth with *V. unguiculata*. But we also observed comparatively and tested protein estimation of all samples we get good results in NPK solution of *V. radiata*. On the other hand, the planting system has no significant effect on the length of leaves. For future work, the experiment will be done on a large scale that will be helpful in exploration whether the hydroponic system will meet the demand of today and future market and sustained demand of agriculture system for livelihood, mankind and economy.

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