PATNA WOMEN'S COLLEGE 1940

IRIS
Journal for Young Scientists
ISSN 2278 – 618X **(Print)**
ISSN 2278 – 6384 **(Online)**

http://www.patnawomenscollege.in/journal

The effect of different growth media on seed germination, phytochemical constituents and nutritional elements present in *Spinacia oleracea*

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Received : November 2014
Accepted : March 2015
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Abstract : *The present study was carried out to determine the effect on the nutritional and phytochemical content of Spinach plant grown in various organic and inorganic media mixed with soil.Out of 50 seeds that were sown in soil supplemented with organic and inorganic media , 48 seeds germinated in the soil mixed with coir and vermiculite media and 41 seeds germinated in garden soil. After 7 days and 14 days the size of the seedlings were observed as 7.8 cm and 11 cm respectively ,which were found to be maximum in green waste with vermiculite media because green waste offered a large surface area for microbial decomposition during composting. Addition of inorganic materials increased the available water holding capacity and reduced excessive concentrations of soluble salts. Vermiculite which is 2:1 clay has a high cation exchange capacity. They are weathered micas in which the potassium ions between the molecular sheets are replaced by magnesium and iron ions. UV-VIS spectrophotometeric estimation of protein in aqueous extract of leaf was found to be maximum (800 µg/ ml) in green waste and vermiculite and minimum (385 µg/ ml) in garden soil. Similarly Concentration of Iron was maximum (8.02 × 10 $^{-5}$ mol L^{-1}) in the leaf extract of spinach grown in green waste and vermiculite media and minimum (4.02 × 10 $^{-5}$ mol L^{-1}) in garden soil. Concentration of vitamin C was highest in green waste (850 µg/ml) and lowest in coir and vermiculite (180 µg/ ml) even though coir contains good amount of cellulose and lignin and it had good water retention capacity and aeration. This quality of coir helped the roots to grow faster.The results of the work suggest that the yield and nutritional value of the plant increased when the soil was supplemented with organic and in organic media.*

Key Words : *Spinacia oleracea, Vermiculite, Coir, Green waste, Spectrophotometer.*

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Introduction:

Spinacia oleracea is a good source of iron and claims a special place among vegetables in terms of its phytonutrient contents. So, the cultivation and consumption of this green leafy vegetable has often been suggested by using supplemented media for

better growth for the plant (Bharadwaj 2011). The supplemented media can include organic materials such as peat , compost ,tree bark ,coconut coir or inorganic materials such as peat and perlite ,coir and clay ,peat and compost (Nair et al., 2011) Supplementation with different supplements in the soil have different effect such as better aeration , better water holding capacity ,better spacing of the seed (Alan et al., 1994 ; Bilderback et al . , 2005). Keeping in view the influence of media in seed germination and seedling growth the present investigation was carried out to study the effect of different media viz. coir, coir with vermiculite, green waste, green waste mixed with vermiculite and garden soil are used in the seed germination, seedling growth and vigour of spinach seedlings .The results obtained has been thus described .

Materials and Methods:

The experiment was carried out in the laboratory of Botany Department, Patna Women's College. All the required growth media were purchased from Bihar Scientific Research store, Patna.

Preparation of clay pots with media: Fifteen clay pots with 200gm of garden soil were randomly designed in 3 replicates to prepare five different growth media. 100gm each of coir and green waste was mixed with the garden soil separately in 3 pots each. Three pots each of 25gm Coir + 100gm Vermiculite mixture and 25gm Green waste+ 100gmVermiculite were taken in the ratio of 1:4 to carry out the work. (Nair et al., 2011) Fifty seeds were sown in each pot. At regular interval 500ml of water was added to the pots.

Phytochemical screening of the sample: To detect the chemical composition of the extract qualitatively, preliminary phytochemical tests were conducted according to the standard protocol (Chaturvedi et al., 2013). Fresh leaves of *Spinacia oleracea* were collected from the pots after 14 days. 100 grams of fresh leaves of the plant from each pot were taken and washed thoroughly in running tap water to clean the adhering unwanted things. It was then crushed properly in a mortar pastel, mixed with 100 ml of distilled water (w/v). Filterate obtained was used as crude extract.

Estimation of protein by UV - VIS spectrophotometer (Lowry method): Ten ml acetate buffer (0.2M Acetic acid + 0.2M Sodium acetate) was added to 1ml of leaf extract and centrifuged at 2500 rpm for 10 minutes .The whole mass of the separated supernatant was collected with a pipette.To 1 ml of supernatant, 5 ml of alkaline solution (Alkaline Sodium carbonate 2% +1% Copper sulphate+ 2% Sodium potassium tartarate) was added. It was left for 10 minutes. 0.5 ml Folin reagent was added to the mixture.The absorbance was taken at 750 nm.

Estimation of Iron by UV–vis spectrophotometer (Sandell, 1994): Five gm. of leaf sample from each pot were weighed, crushed and filtered. To the filtrate (plant extract) 10 mol L^{-1} hydrochloric acid was added and stirred for 5 minutes .Then 5 ml. of distilled water was added. 10 ml of 1 mol L^{-1} ammonium thiocyanate solution was added to each of the sample. The solution was mixed by swirling. A stable red colour appeared. The absorbance was taken at 490 nm.

Estimation of vitamin C by UV- vis spectrophotometer (Alam, 1996) : From each pot 5gm of the leaf sample was collected, crushed and homogenized with about 25 ml acetic acid solution (10%Acetic acid). The solution was filtered and the clear filtrate was collected for the determination of vitamin C in the samples.

To the filterate sample solutions 1 ml of bromine water was added until the solution became coloured.Then1 ml of thiourea solution was added to it to remove the excess bromine and thus the clear solution was obtained. 2, 4-Dinitrophenyl hydrazine solution was added and the absorbance was taken at 280 nm.

Results and Discussion:

A good growing medium provided sufficient anchorage and support to the plants, served as a reservoir for nutrients and water. Water allowed oxygen diffusion to the roots and permitted gaseous exchange between the roots and atmosphere (Abad et al., 2002). As suggested by Wilson et al., (2001) media composition used influenced the quality of seedling. The results of our work also suggest that different growth media have significant effect on growth pattern and phytonutrients of spinach leaves.

Table 1. Effect of different soil media on seed germination, and seedling height

S. No.	Type of growth media	No. of seeds germinated in 7 days	Size of seedlings on After 7 days	Size of seedlings after 14 days
1.	Garden soil	41	4 cm	7 cm
2.	Coir	45	5 cm	8.5 cm
3.	Coir + vermiculite	48	5.5cm	9.5 cm
4.	Green waste	44	6.5 cm	10.5 cm
5.	Green waste + vermiculite	46	7.5 cm	11 cm

Table 1 shows the growth pattern of *Spinacia oleracea* influenced by different growth media .The result from our research revealed that the yield of the vegetable tend to be higher for the plant which were grown in growth media as compared to those grown in the normal garden soil. Seeds which were sown in Coir+ Vermiculite medium showed maximum viability. Number of seeds germinated were about 48 out of 50 in this medium . But we have found that the size of seedings were largest in Green waste + Vermiculite and smallest in garden soil. The size of seedlings in Coir+Vermiculite occupied third position .Similar result of cucumber plants grown in vermiculite has also been reported by Sawan et al., (1999) .Least viability of seeds was observed in garden soil.

Spinach is rich source of vitamins, minerals like iron calcium and a number of antioxident components like polyphenol, flavonoids, caotenoids which are shown to posses anti-inflammatory effect , anti-mutagenic potential. This study also reveals presence of these compound in the leaf extract of spinach.

Table 2. Phytochemical screening of aqueous extract of *Spinacia oleracea* leaves

S. No.	Phytochemicals	Teste performed	Observations	Inferance
1.	Flavonoid	Shinoda test	Red crimson colour	+
2.	Phenol	Ferric chloride test	Red colour	+
3.	Protein	Biurete test	Violet colour	+
4.	Glycosides	Libermann's test	No change in colour	–
5.	Saponins	Froth test	No stable foam	–
6.	Carbohydraye	Molish test	Purple ring formed	+
7.	Alkaloid	Mayer's test	Precipitate formed	+
8.	Vitamin C	Indophenol test	Colourless solution	+
9.	Tanin	Leadacetate test	Green precipitate formed	+
10.	Iron	Ammonium thiocynate test	Blood red colour	+
11.	Calcium	Ammonium oxalate test	White precipitate	+
12.	Phosphorus	Ammonium molybdate test	Bright yellow preciitate	+
13.	Magnesium	Ammonium hydroxide test	Geletinous white precipitate	+
14.	Manganese	Potassium permanganate test	Dark brown precipitate	+
15.	Zinc	Potassium ferrocynide test	Cream precipitate	+

Key: + (present) ; – (absent)

Table 2 shows different phytocompounds present in *Spinacia oleracea* which indicates that it claim a special place among vegetables in terms of its phytonutrient contents . Chaturvedi et al., (2013) in his work has also suggested presence of these phytocompounds in his work.

Proteins are polymer of aminoacids .It is vital for proper growth and development . Protein content of spinach varies depending on the type of spinach. Concentration of protein contents in spinach also varies with different supplemented media used.

Table 3. Estimation of protein in different plant sample UV-vis spectrophotometer

S. No.	Sample from different media	Absorbance at 750 nm	Concentration (µg/ml)
1.	Garden soil	0.685 nm	385 µg/ml
2.	Coir	0.814 nm	410 µg/ml
3.	Coir+ vermiculite	0.934 nm	630 µg/ml
4.	Green waste	0.794 nm	490 µg/ml
5.	Green waste + vermiculite	1.004 nm	800 µg/ml

The results shown in Table 3 indicate that the protein concentration was highest in greenwaste and vermiculite media and lowest in the normal garden soil. It was due to high nitrogen contents in green waste and proper aeration and water holding capacity of vermiculite .

Spinach along with other green leafy vegetables is rich in iron. Spinach contains iron absorption - inhibiting substances including high levels of oxalate, which can bind to the iron to form ferrous oxalate and render much of the iron in spinach unusable by the body.

Table 4. Estimation of iron in different plant sample by UV-vis spectrophotometer

S. No.	Sample from different media	Absorbance at 750 nm	Concentration (mol L^{-1})
1.	Garden soil	0.716 nm	4.02×10^{-5} mol L^{-1}
2.	Coir	0.778 nm	7.09×10^{-5} mol L^{-1}
3.	Coir+ vermiculite	0.933 nm	630×10^{-5} mol L^{-1}
4.	Green waste	0.516 nm	490×10^{-5} mol L^{-1}
5.	Green waste+vermiculite	1.097 nm	800×10^{-5} mol L^{-1}

The results shown in Table 4 indicate that the iron concentration was highest in green waste and vermiculite media and lowest in normal garden soil.

Fresh spinach has long been known to contain relatively large amounts of vitamin C Vitamin C is a water soluble vitamin C is instrumental in neutralizing the free radicals, which are harmful to the body. Concentration of Vitamin C varied with different media used to propagate the plant.

Table 5. Estimation of Vitamin C content in different plant sample by UV-vis spectrophotometer

S. No.	Sample from different media	Absorbance at 280 nm	Concentration (mol L^{-1})
1.	Garden soil	3.863 nm	4.02×10^{-5} mol L^{-1}
2.	Coir	3.876 nm	7.09×10^{-5} mol L^{-1}
3.	Coir+ vermiculite	3.796 nm	630×10^{-5} mol L^{-1}
4.	Green waste	nm	490×10^{-5} mol L^{-1}
5.	Green waste+vermiculite	1.097 nm	800×10^{-5} mol L^{-1}

The results shown in Table 5 indicate that the Vitamin C concentration was highest in green waste and lowest in Coir and Vermiculite media .So Vitamin C concentration was highest in organic medium. This finding is similar to the findings of Bangerth (1976). It has been suggested that greater concentration of Vitamin C enhances nutritional value of the fruits and vegetables.

Conclusion :

The findings from the research work revealed that the growth of the plant was higher in green waste and vermiculite medium than the rest of the media due to high nitrogen contents in greenwaste and proper aeration and water holding capacity of vermiculite. The contents of phytocompounds like protein, Iron, Vitamin was also higher in the plant which was grown in green waste and vermiculite medium. Growth in plants was also good in coir and vermiculite medium but was less than green waste and vermiculite medium. Hence it is concluded that Green waste and vermiculite media are best for the propagation of *Spinacia oleracea.* Coir and vermiculite medium occupied the second position

in enhancing the growth of the plant. The research also revealed that the primary and secondary metabolites and minerals contents varied in different plant samples which were grown in different media. The present work helped us to conclude that the best media for the propagation of *Spinacia oleracea* is green waste with vermiculite mixed with soil.

Acknowledgements:

We are grateful to Dr. Sister Doris D'Souza A.C., former Principal , Patna Women's college (PWC) and Research committee for providing facilities facilities and financial support . We thank Prof. Sheila Bedi, Head Department of Botany PWC for taking keen interest in our research work .

References:

Abad M , Noguera P , Puchades R , Maquieira A , Noguera (2002). Physiochemical and chemical properties of some coconut dust for use as a peat substitute for containerized ornamental plants. *Biores*. *Technaoro*. 82 : 241-245.

Alan R., Zuladir A. , Padem H (1994) . The influence of growing media on growth , yield and quality of tomato grown under green house condition. *Acts Horticulture*. 37 : 309 - 312 .

Bangerth F. (1976). Relationship between the Ca content and Ca treatment and the ascorbic acid content of apple , pear ,tomato fruits. *Qualitus Plantarum* 26 : 341 - 348 .

Bharadwaj R.L. (2014). Effect of growing media on seed germination and seedling growth of papaya *CV . 'Red lady'*. 8 (4) : 178-184.

Bilderback TE , Warren SL , Owen Jr. JP (2005) . Healthy substrates need physicals tool. *Hort . Technology*. 15 : 747 - 751.

Chaturvedi N., Sharma P. and Agarwal H. (2013). Comparative Nutritional and phytochemical analysis of spinach cultivars : *Basella alba* and *Spinacia Oleracea*. *International journal of Research in pharmaceutical and Biomedical Sciences,* 4(2):674-679.

Nair A., Ngouajio M., Biernbaumj (2011). Alfalfa based Organic amendment in peat compost growing medium for tomato transplant production . *Hort . Science* 46 : 253 - 259 .

Sandell, E.B. (1994). Calorimetric determination of metals , *Inter science : NY*: 271 - 273 .

Sawan D.H., Eissor A.M., Abou Hadid A.F. (1999). The effect of different growing media on cercumbu seeding production, fruit yield and quality under green house condition. *Acta Horticultarae*, 491:369-376.

Wilson SB , Stoffela PJ , Graetz DA (2001) . Use of compost as a media amendment for containerized production of two subtropical perennials. *J . Environ . Hortic* . , 19 : 37 - 42 .