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SYNERGISTIC EFFECT OF VERMICOMPOST ASSOCIATED WITH COWDUNG ON THE SEEDLING GERMINATION OF "PHAESEOLUS RADIATUS" (GREEN GRAM)

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ABSTRACT

Earthworms are often referred to as farmer's friend and nature's ploughman. Studies on degradation of organic wastes by earthworms are one of the recent developments in biological sciences. Vermicomposting, method of converting wastes into useful products through action of earthworms, has gained awareness in India, to reduce the soild waste pollution and sustaining our environment. The major role of earthworms in the soil is decomposition of organic materials, developing soil structure and altering physico-chemical properties of soil. Earthworms speed up the composting process and transform wastes into nutrient-rich castings. The worm 'casting' are a good fertilizer additive for agricultural crops. Vermicompost is a finely divided peat-like material with good structure, porosity, aeration and moisture holding capacity. The earthworm, *Eudrilus eugeniae*, a tropical species commonly called African night crawler, is large in size, grows rapidly, breeds fast and is capable of decomposing large quantities of organic materials into usable vermicompost **[1, 2].** The present study was undertaken to convert the locally available cowdung and testing the efficiency of such vermicompost on seed germination and yield of plant "*Phaseolus radiatus*".

Key words: Vermicompost, *Phaseolus radiates,* Seedling, germinatiom.

INTRODUCTION

Earthworms are often referred to as farmer's friend and nature's ploughman. Studies on degradation of organic wastes by earthworms are one of the recent developments in biological sciences. Vermicomposting, method of converting wastes into useful products through action of

earthworms, has gained awareness in India, to reduce the soild waste pollution and sustaining our environment. The major role of earthworms in the soil is decomposition of organic materials, developing soil structure and altering physico-chemical properties of soil. Organic matter decomposition, nutrient cycling, soil structure and plant productivity have been studied by several authors **[3, 4, 5]**. The worm castings are good fertilizers additive for agricultural crops.

Degradation of organic wastes by earthworm is known as vermicomposting it is a safe and ecofriendly technique for quick disposal of organic wastes **[6]**. Organic wastes comprises of crop residues, plant wastes, fruit peels, kitchen and house hold refuges, animal excreate and waste from agricultural based industries. Annual production of organic wastes in india is about 3000 million tones **[7]**. Earthworm can consume practically all kinds of organic wastes, consume two to five times of its body weight and after using 5-10% of the feed stock for its growth, excrete mucous coated undigested matter as worm casts **[8]**. The use of vermicompost, as a source of organic manure in supplementing chemical fertilizer is becoming popular among the farmers of the country. Increase in crop yield, soil nutrient status and nutrient uptake is due to application of vermicompost. Composting is considered to be the best alternative method to treat food wastes, since it provides an agricultural amendment capable of mitigating and offsetting the serious deficit of organic matter experienced by many agricultural soils.

The cities and towns generate tones of vegetables residues which is a reverse of pathogens to cause epidemic **[9]**. The composting technique will help to reduce the volume of social wastes and converts it into usable manure. The adoption of mechanized farming in many parts of the country has resulted in leaving crop residues in field after harvesting. These crops are burnt in the field. The burning of agricultural wastes and dung cake produced higher quantities of carbon monoxide and total suspended particles as smoke and creating greater environmental pollution. Worked on the solid wastes and opined that the resources can be recycled by composting. Vermin-biotechnology is an aspect of biotechnology that studies worms as versatile bioreactors. The conversion of any organic waste in the form of vermicomposting is the major role played by the earthworms in nature. Earthworms degrade all types of organic wastes such as agricultural wastes, animal wastes, weeds, forest litter and also industrial wastes. Now days,

vermicomposting technology is very useful in the agricultural field. The major role of earthworm in the soil is the decomposition of organic materials, developing soil structure and altering the physico-chemical properties of soil **[3, 5]**. Worms have been associated with soils and they modify soil structure and fertility, improve plant growth and are important in sustaining productivity **[10]**. The earthworm has always interested mankind because of its action as tiller, aerator, crusher, composer and moisture-builder and above all as intimate friend and benefactor of the farmers **[11, 12]**.

Earthworms speed up the composting process and transform wastes into nutrient-rich castings. The worm 'casting' are a good fertilizer additive for agricultural crops. Vermicompost is a finely divided peat-like material with good structure, porosity, aeration and moisture holding capacity. The role of the earthworm is enhancing soil fertility was, known even to ancient farmers. But with the advent of modern agricultural practices, during the last decades, and use of chemical fertilizers, their significance has faded. In recent years farmers are once again realizing the value of these highly beneficial animals and are making all possible efforts to promote vermiculture. The beneficial effect of earthworms in increasing soil fertility was documented since the time of **[13]**.

From research carried out recently on the effects of earthworms on waste materials it's clear that the worms especially perionyx excavatus found quicker with in 1-.2.5 months in the residues of crop hardweeds. And home refuges. Vermicompost is a complex bioferilizer and is not desirable to compare its status as a mere supplier of NPK fertilizers. Vermicompost is richer than other types of compost. The earthworm, *Eudrilus eugeniae*, a tropical species commonly called African night crawler, is large in size, grows rapidly, breeds fast and is capable of decomposing large quantities of organic materials into usable vermicompost **[1, 2]**. The present study was undertaken to convert the locally available cowdung and testing the efficiency of such vermicompost on seed germination and yield of plant "*Phaseolus radiatus*".

MATERIALS AND METHODS

Sampling

"Eudrilus eugeniae" is a common earthworm, most widely used in vermincomposting. The worms that feed actively assimilate only 5-10 % and the rest is excreted as loosed granular mounds of vermicasting on the surface, generally away from the food source is about 2-10 days.

These have to be brushed aside and collected into separate trays.

The casting, thus collected has to be let over night in conical helps for the worms to move to the bottom. The tops of the cones which are free of worms are then collected and lightly air dried. The dried vermicasting are sieved through 3 mm mesh to separate cocoons and young ones from the vermicastings. The sieved castings are now ready for use as vermicompost. It was collected from the vermiculture department, Sri Paramakalyani centre for environmental studies, Manonmaniam Sundaranar University, Alwarkurichi.

Pot Culture

Eight pots of equal size were choosen for the analysis, done in green gram plant (Phaseolus radiatus). The cowdung compost prepared were applied to the each pot in the concentration of 1:1:1, 1:1:2, 1:1:3, 1:1:4, 1:1:5, 1:1:6, each pot have 15 plants. The control contains 2kg of red soil and 2 kg of sand soil alone. The compost added in grams to the pots are given in series ie, 70, 105, 140,175,210, 245 gm. The eight pots contains chemical fertilizers of about 35gm of DAP.

Chlorophylls in Leaf Development

During the development of leaf from germination through maturation as well as in light and dark, interesting changes takes place in the levels of their pigments, particularly in chlorophyll a and chlorophyll b.

Catalase Test

Sodium perborate serves as the substrate for catalase. Sodium perborate in aqueous solution is added to catalase. Hydrogen peroxide present in the solution is acted upon by catalase. The remaining hydrogen peroxide is determined by titration with potassium permanganate in the presence of sulphuric acid.

Controls were conducted along with the experimental by adding the enzymes at the end of the incubation period endpoint – appearance of pale pink colour.

Assay of Peroxidase

Peroxidase activity is measured by aminoanti pyrine phenol assay. The basic principle of this reaction is a coupling reaction. H2O2 rapidly reacts with phenol and 4-amino antipyrine (eldonar) in the presence of peroxidase to produce a quinoamine chromogen, which is intensely coloured with a maximum absorbance at 510nm.

$$H_2O_2 + \text{Phenol} + \text{4-aminoantipyrine} \leftrightarrow \text{quinoamine} + 2H_20_2.$$

Hydrogen Peroxide

The activity of peroxides was calculated from the absorbance change

$$\Delta A = A1 \text{ min} - A0 \text{ min where}$$

A = over all absorbance change

A1 min = Absorbance at 510 nm after 1 minute

A0min = Absorbance at 510 nm after 0 minute

Enzyme activity is expressed as units per mg.

Units Mg = Δ A/min/6.58 X mg of sample

Enzyme extraction

Weighed 100mg of fresh sample (Germinated seed and grinded well with 5ml of ice cold buffer. Then centrifuged (or) filtered and collected the supernatant. Used the supernatant as the enzyme source.

Estimation of Ascorbic Acid By 2,6 Dichlorphenol Indophenol Method

Ascorbic acid reduces the 2,6 dichloro phenol indophenol dye to a colourless leuco-base. Ascorbic acid gets oxidized to dehydroascorbic acid. Though the dye is blue coloured Compound the endpoint is the appearance of pink colour. The dye is pink coloured in acid medium. Oxalic acid is used as the titrating medium.

Assay of Protease

The enzyme protease hydrolyse the peptide bonds of proteins, liberating amino acids. Owing to its atmospheric character an aminoacid cannot be directly titrated with alkali. When formaldehyde is added to the aminoacids, and the aminoacid formal complex is formed. This complex is acidic because the basic character of the aminogroup is suppressed and hence it can be titrated with alkali, using phenolphthalein as indicator this method is called "Sorenson's formal titration".

Assay of Glycine

5g of glycine in 50ml of distilled water.

Enzyme extraction

100mg of fresh sample was weighed (leaf & germinated seed) and grinded well with 5ml of ice cold buffer. Then centrifuged (or) filtered and supernatant was collected as the enzyme solution.

Estimation of Phosphorus by fiske and Subbarrow method

When the given solution is treated with ammonium molybdate, phospho molybdic acid is formed, this phosphor molybdic acid is reduced by the addition of 1, 2, 4 amino napthol sulponic acid (ANSA) to produce molybdenum blue (a blue colour solution) is a measure of inorganic phosphate buffer.

Estimation of total free amino acids

Ninhydrin, a powerful oxidizing agent, decarboxylates the alpha amino acids and yields an intensely coloured bluish purple prduct which is measured colorimetrically at 570nm.

Ninhydrin+ alpha-amino acids ⟶ Hydrindantin+CO2+NH3+Decarboxylated aminoacid.

Hydrindantin+Ninhydrin+Ammonia ⟶ Purple coloured product + water.

Estimation of protein by biuret method

Biuret method based on the fact that Co-NH group of protein forms a purple colour complex with copper complex with copper ions in an alkaline medium.

Stock standard BSA (Bovine Serum Albumin)

Weigh accurately 400mg of BSA and dissolve in distilled water and make up to 100ml in a standard flask. The concentration of standard was 4mg/ml.

Estimation of protein

Pipette out 0.5, 1.0, 1.5, 2.0, 2.5 ml of standard into a series of test tubes, and pipette out 0.5 ml of sample extract in other two tubes. Make up the volume to 5ml with distilled water. A tube with 5ml of water serves as blank. Add 3ml of biuret reagent to each tube including blank mix well and keep at 37 C for 10 minutes. Purple colour was developed. The Optical density for each tube was measured at 540nm using a reagent blank. The standard graph was drawn by taking concentration on x-axis and optical density on y-axis. From the standard graph we can calculate the amount of protein in given sample.

RESULT AND DISCUSSION

The physico-chemical analysis of compost – cowdung was analysed using standard method & recommended by Agricultural Health centre. The compost and soil ratio (1:1:1) upto (1:1:6) was prepared for germination study. Similar pure soil sample (1:1) was used as control. biochemical paramets and growth rate of *phaseolus radiatus* was observed in both the compost and control. The growth rate & biochemical activity was recorded on 15^{th} day after germination in the yield. The data were collected for following parameters.

Root Length

The root length of vermicompost and control was noted, when compared to control the vermicompost has higher range of growth of 12.9cm. The control has only 9.0cm. Increased measure of root length fixes and absorbs more minerals and growth factors from the soil.

Shoot length

The shoot length of control and vermicompost was measured. It is clear that the shoot length was much more than control. The average shoot length of vermicompost was 12.8cm where the control has 10. 5 cm, this shows the increasing growth rate of vermicompost than control.

Internode. The distance between two leaf nodes was internode. The comparative data in **Table 1-4** below shows higher in vermicompost than the control. The average means of vermicompost was 2.0cm, the control has 1.2cm.

Table 1: Germination Efficiency of Green Gram

S. No.	Sample	No of seed sown	No of seeds germinated
1	(1:1)Control	15	8
2	(1:1:1)	15	13
3	(1:1:2)	15	13
4	(1:1:3)	15	14
5	(1:1:4)	15	14
6	(1:1:5)	15	15
7	(1:1:6)	15	15
8	Chemical Fertilizer (1:1:1)	15	2

Table 2: Growth Rate of *Phaseolus Radiatus* – 15th Day

S. No.	Sample	Root length	Shoot Length	Internode
1	Control (1:1)	9.0	10.5	1.2
2	(1:1:1)	10.5	11.0	1.7
3	(1:1:2)	11.5	13.0	2.0
4	(1:1:3)	13.0	13.0	2.0
5	(1:1:4)	13.5	13.2	2.0
6	(1:1:5)	14.0	13.5	2.2
7	(1:1:6)	14.9	14.0	3.0

Table 3: Leaf Area in Green Gram – 15th Day

S. No.	Samples	Length (cm)	Breadh (cm)
1	Control (1:1)	5.4	4.5
2	(1:1:1)	5.6	3.9
3	(1:1:2)	6.0	4.2
4	(1:1:3)	6.1	4.5
5	(1:1:4)	6.5	4.8
6	(1:1:5)	6.5	5.0
7	(1:1:6)	6.7	5.2

Table 4: Bio-Chemical Parameters Result

S. No.	Biochemical parameters	Control seed	Vermicomposted seed
1	Protein	332.0	356.66
2	Aminoacids	1.422	2.301
3	Phosphorous	2.9025	4.4310
	Anti Oxidants		
4	Ascorbic acid	0.4347	1.377
5	Peroxides	-	-
6	Catalase	+	+
7	Enzyme Protease	+	+
8	Chlorophyll	-	-

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