

## Bioconversion of Vegetable and Flower Wastes into Compost by *Eudrilus euginae* Treated with Food Colouring Agent Tartrazine

Kalaivanan, K., Durairaj, S. and Selladurai, G.

P.G. and Research Department of Zoology, Arignar Anna Govt. Arts College, Cheyya r604 407,  
Tiruvannamalai District, TamilNadu.

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**Abstract:** Waste materials in recent years have been increasing day by day and it is a major threat to the environmental issue. To solve the problem in one way, the waste materials are utilized in a fruitful way by using *Eudrilus euginae*. The vegetable and flower waste were subjected to vermicompost separately and along with tartrazine treatment. In these treatments, the vermicompost materials were analysed the micro and macro nutrients to understand the impact of tartrazine on the bioconversion of waste into compost. The Nitrogen level was increased in the flower waste compost by 28.5% whereas it was decreased in the vegetable waste by 28.6%. Similarly, the decreasing trend of Phosphorus was observed in both the compost. Besides Nitrogen and Phosphorus, the Potassium showed significant increase in all the treatments. However, insignificant changes of Zinc, Manganese and Copper were observed in the compost of flower and vegetable waste. In the case of Ferrous, significant increase was observed. The physical properties such as the Electrical Conductivity (EC) and pH of the vermicompost showed almost equal to the control. On the Basis of observation, the nutrients of the compost of waste have been stabilized by the earthworm, *E. euginae*.

**Keywords:** *Eudrilus euginae*, Tartrazine, Vermicompost, Vegetable and Flower waste.

**Corresponding Author:** [gselladurai7@gmail.com](mailto:gselladurai7@gmail.com)

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### I. Introduction

A rapid increasing population and high rate of industrialization have increased the problem of solid waste management [1]. The large amount of solid waste and sludge produced by anthropogenic sources is becoming a serious problem [2]. Proper waste management is very crucial and has become the major challenge in many countries [3]. Vermicomposting has become an appropriate alternative for the safe hygienic and cost effective disposal of wastes. Earthworm is an eco-biotechnological process that transforms energy rich and complex organic substances into stabilized vermicompost [4]. Recycling of wastes using earthworm has become an important component of sustainable agriculture which has a multidirectional impact in terms of safe disposal of wastes preventing environmental pollution besides yielding nutrient rich material [5]. The environmentally acceptable vermicomposting technology using earthworm can well adopt for converting waste into wealth [6].

The earthworms have been fortunate to degrade wastes and turn them into a fertilizer which is popularly termed as vermicomposting; thus acquired product is recognized as vermicompost. It is the resourceful technique to renovate the biological wastes (human discarded, kitchen surplus, animal left-over etc.) into a biological fertilizer. Edwards and Burrows [7] reported that vermicompost has an abundant fine structure it is highly saturated with nutrients that are freely available for plants intake. Even vermicompost is rich in both the bacterial components [8] and fungal profusion [9] than the outmoded muck. Earthworms are used for refining and preserving soil fertility, change of organic waste into a useful fertilizer, for livestock [10] then used as bait for fish. Vermicomposting is a mesophilic process led to an everlasting generation of wastes, carried out by earthworms. Disposal and environmental organic waste followed by excretion of friendly management of these wastes are castings through the worm's metabolic becoming a serious global problem.

### II. Materials And Methods

#### Collection of Earthworms

The earthworm *Eudrilus euginae*, were collected from Chithathur, Thiruvannamalai Dt, Tamil Nadu. The collection of earthworm was brought to laboratory for acclimatization. The present study was carried out between June - August 2016 in the Zoology laboratory. The selected Flower and Vegetable waste were collected from Vembakkam market, and mixed thoroughly with 2 kg of soil. This mixture was placed in the form of heaps under shady place. Watering was done regularly twice in a day in order to maintain the optimum temperature and moisture. This set up was maintained for 30 days. In another pot the same mixture was taken with tartrazine

(a food colour chemical) then released earthworm. Suitable control was maintained. Similarly flower waste was also taken separately in another pot. The waste mixtures were allowed to pass through earthworm guts for vermicomposting. The moisture content of the organic substrates in each pot was maintained between 60% and 65% throughout the study period by sprinkling water after every 24 hours. The experiment was conducted by randomized design with three replications. In the vermicompost materials the experimental set up was maintained till 25 days. The compost material was selected for the analysis to micro and macro nutrient such as nitrogen (N), total phosphorus (P), total potassium (K) Zinc (Zn), Manganese (Man), Iron (Fe), and Copper (Cu).

### **Histological Studies**

The histology of anterior, clitellar, middle and posterior regions of earthworm was studied by adopting the routine paraffin method. The control and experimental animals were blotted free of mucus, washed thoroughly in physiological saline, cut into pieces of clitellar region of desired size and fixed in Boeings fluid fixative immediately after autopsy. Fixation was carried out at room temperature for 24hr, after which the tissues were transferred to 70% alcohol. Several changes of 70% alcohol were given until the yellow colour disappeared from the tissues. The tissues were then dehydrated by passing through ascending grade of alcohol, cleared in xylene, infiltrated with molten paraffin, and finally embedded in paraffin wax. Tissue section of 5- $\mu$ m thick of cross sections were obtained using a rotary microtome. The section, thus obtained, were stained in harries haematoxylin and eosin, dehydrated using alcohol, cleared in xylene and mounted using dihydroxy phthalate xylol (DPX).

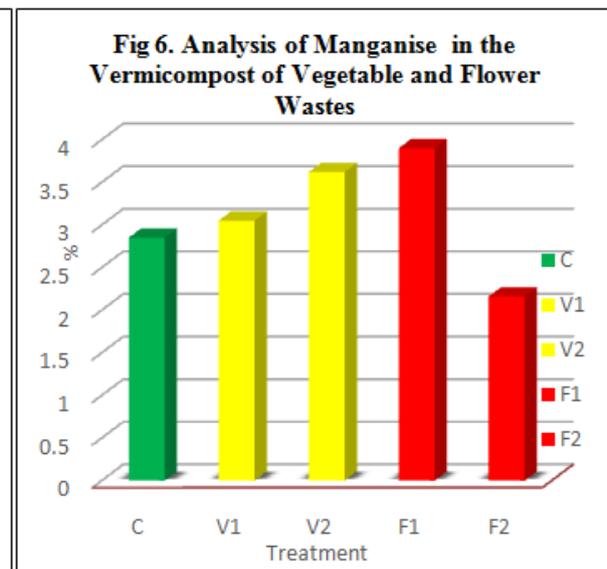
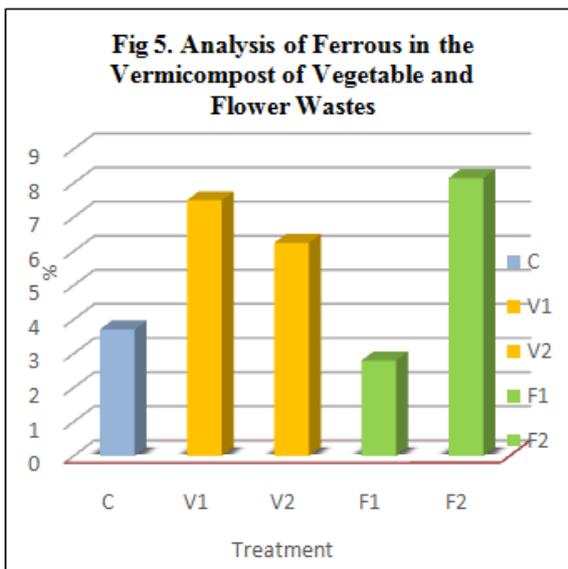
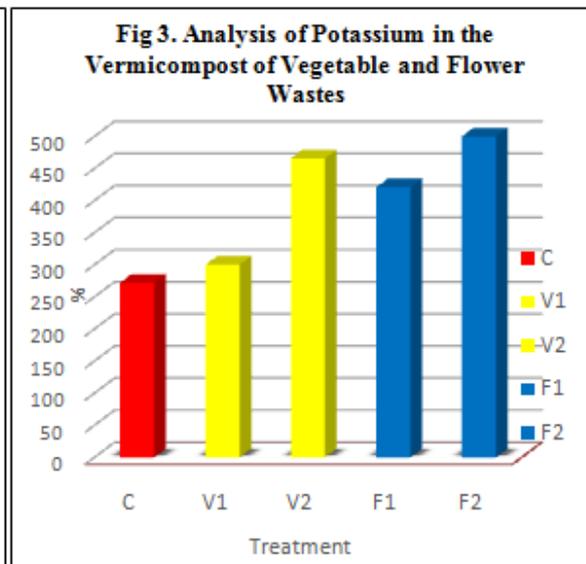
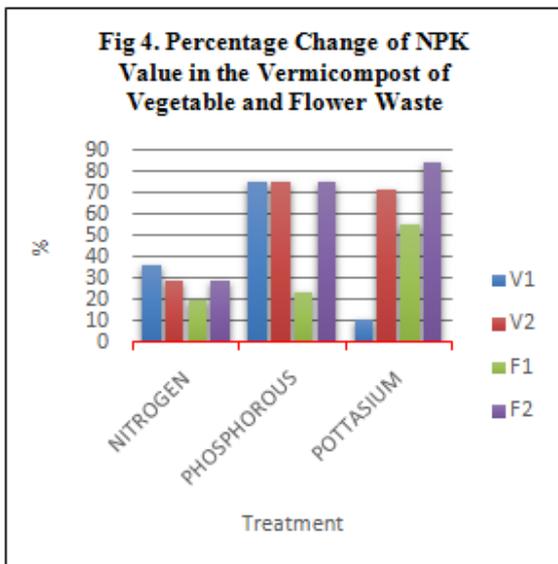
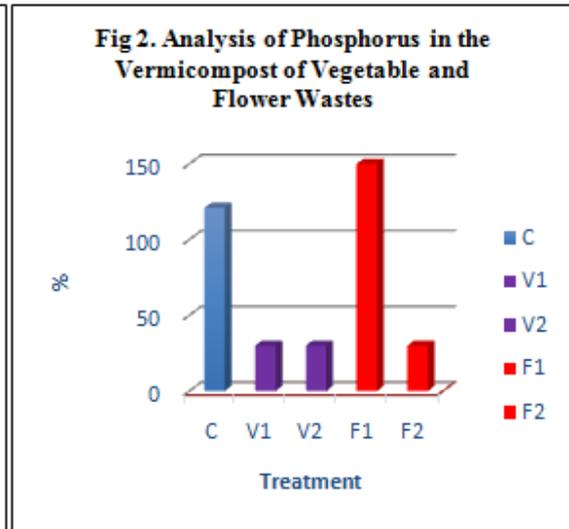
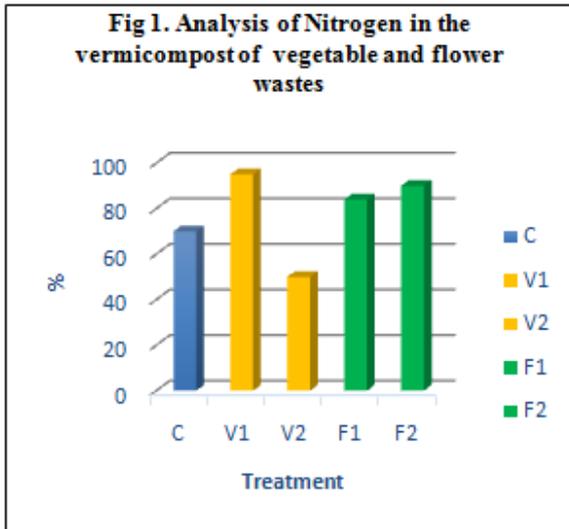
### **III. Results**

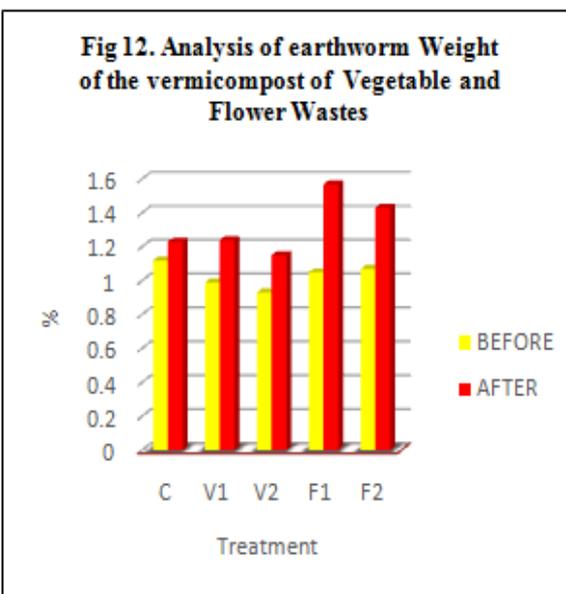
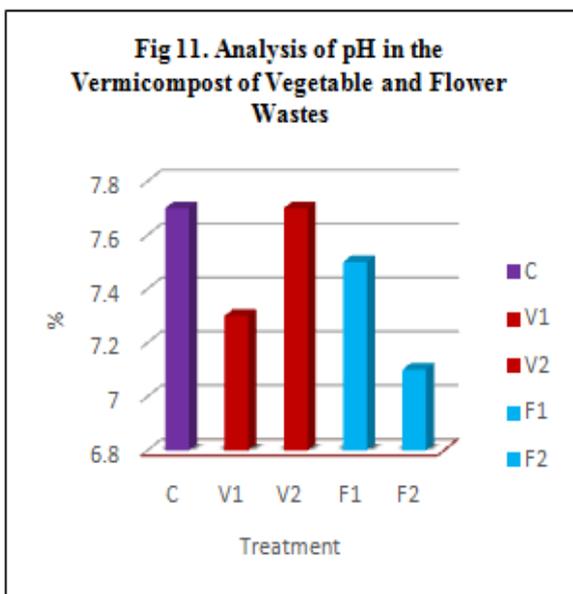
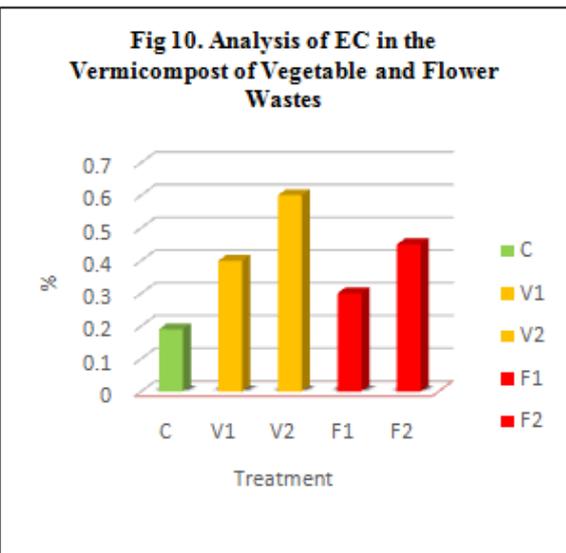
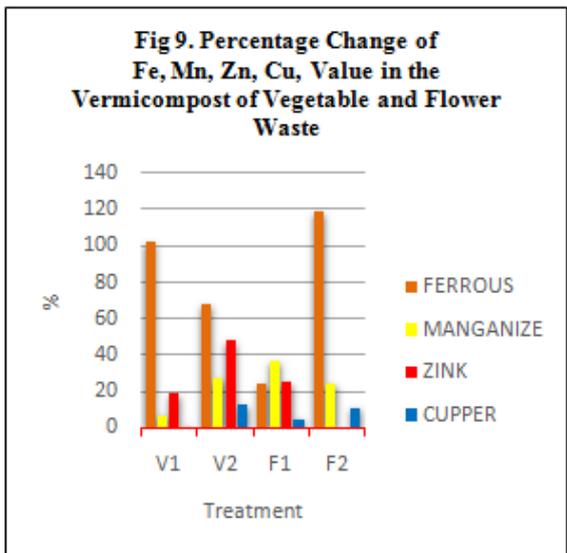
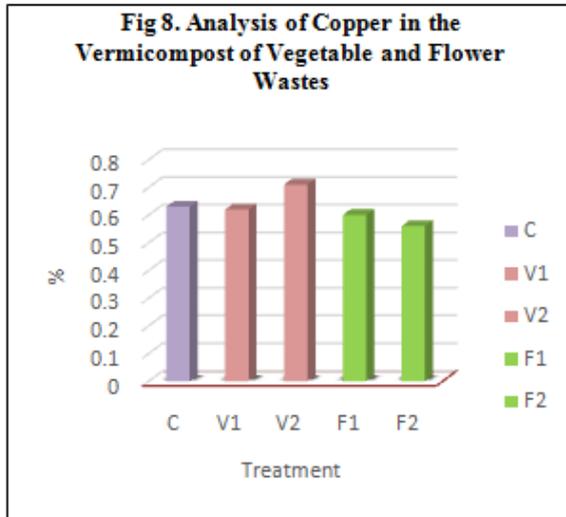
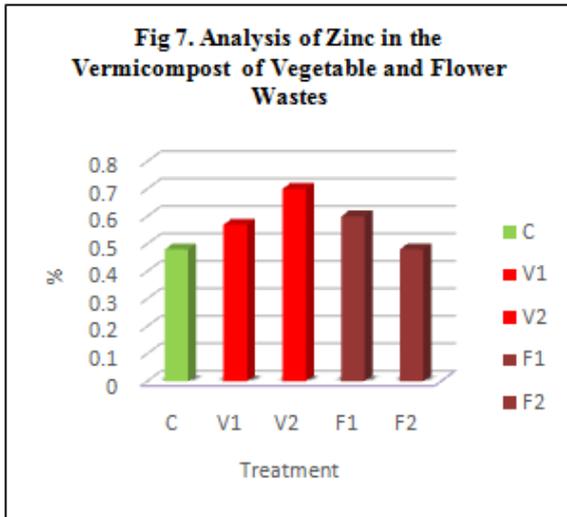
The vegetable and flower waste of different proportions were mixed with vermibed and seen the efficiency of the earthworm in the conversion of vegetable and flower waste into minerals on 25 day. Along with this, tartrazine was treated with the vegetable and flower waste as an experiment. The experimental setup was arranged which as follows; control, tartrazine with vegetable and flower waste of known weights taken separately. The weight of vegetable and flower waste was 100 gms. Mean number of days required for the biotransformation of waste into vermicompost was 25 days. The nutrient status of vermicomposting depends on the type of waste material processed by earthworms. An increase in the values was observed in the following parameters such as total Nitrogen (N), Total Phosphorous (P), and Total Potassium (K), in the different treatments of vegetable and flower waste when compared to control, however the significant decrease was observed in tartrazine treated vegetable and flower wastes.

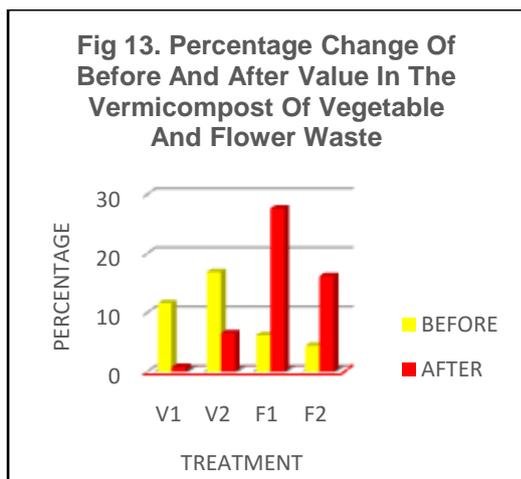
### **Physico-Chemical properties**

The cow dung used as the inoculants in the vermicomposting process enhanced the quality of feeding resource attracting the earthworms and accelerated the breakdown of wastes resulting in the reduction of increasing certain nutrients. The analysis of Nitrogen (N), Phosphorus (P), Potassium (K), Iron (Fe), Manganese (Mn), Zinc (Zn), Copper (Cu), Electrical Conductivity (EC), and pH were analyzed on 25 days in the control, tartrazine treated vegetable and flower waste. In the control, the N value was 70% and this level decrease in the tartrazine treated where as in the other treatment, the level of N was increased gradually. All the values were statistically significant at  $P < 0.01$ . The maximum of 35% increase of nitrogen was noticed in the 100 gms vegetable waste then 9% increase of nitrogen was noticed in the 100 gms flower waste. Equally in the tartrazine treatment also noticed the increased level of nitrogen. All the values were statistically significant  $p < 0.01$ . The maximum of 35% in the tartrazine treated also noticed, the level of nitrogen was decreased to 28% on 25<sup>th</sup> day, indicated the decreased of N level. The study indicated that the optimum level of mineralization takes atleast 25 days. Similarly, P was also decrease only in the treatment where the level was 75% and showed insignificant at  $P < 0.05$  (Fig.1 to 13).

The vermicompost was dark brown in colour and homogenous after 45 days of earthworm's activity. The changes in worm biomass for all the treatments over the experimental period were observed. At the end of the 25<sup>th</sup> day, the earthworm biomass had increased in all the treatments. A decrease in the value of N was observed in the vegetable treated with tartrazine following the parameters. Nitrogen when compared to control. In the flower waste treated tartrazine, this is increased 28 %. Similarly phosphorous was also decreased in vegetable with tartrazine treatment where as P revealed significant correlation in 100 gm treatment vegetable and flower waste. In contrast to NPK showed in significant when compared all other treatments. The analysis of Fe, Mn, Zn, Cu were also increased same level in the treatment with 100 gms of vegetable wastes. Similarly the same trend was also observed in the flower waste. Similar by Fe, Mn, Zn, and Cu were also showed significance of 100gm treatments. On 25 days analysis, Fe showed increase in all the treatments when compared to control. Mn also indicated the slight increased in all treatments on 25day. Similarly Zn and Cu were also increased significantly in the 100 gms of vegetable waste treatments at  $p < 0.05$  level (Fig. 1 to 13).







C=Control

V1=Vegetable Waste (Control)

V2=Vegetable Waste (Tartrazien Treated)

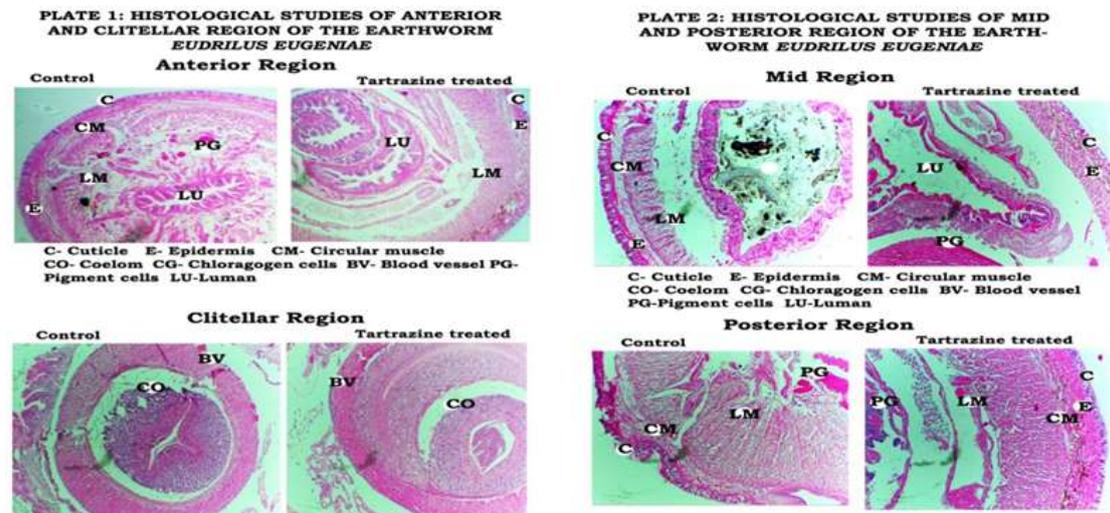
F1=Flower Waste (Control)

F2=Flower Waste (Tartrazien Treated)

Physico-chemical properties of the soil mixed with different vegetable and flower waste were analysed on 25 days along with control and experiment. The EC was increased from 100gm treatment including tartrazine treated vegetable wastes. However the was two increase in to flower waste. The increase in all the treatments were statistically significant at  $P < 0.01$  level (Fig. 1 to 13). Among the different treatments on 25 days, pH value was normal and no change was observed in the tartrazine treatments and indicated statistically insignificance. In this study, a reduction in the pH was recorded at the end of the experiment. The variability in pH could be due to the production of organic acids during organic waste decomposition. During their study on the vermicomposting some organic residues may be produced that could have lowered pH level at the end of experiment. The analysis on 25 days showed reduction of pH value in all the treatments, and showed in significance. The bioconversion of these waste will be completed in about 25-35 days or even a lesser than this time limit. Figure (Fig-13) showed the weight gain of earthworm, in all the treatments the little increase in weight gain showed statistically insignificance.

#### **Histological Studies of Earthworm *Eudrilus euginae***

The cross section of Fore gut (anterior and clitellar region), Mid gut (middle region), Hind gut (posterior region) of control, tartrazine treated and vegetable and flower waste treatments of higher proportion were taken to study the anatomical features of *Eudrilus euginae*. In the control animal, the epidermis of the *Eudrilus euginae* consists of an epidermal epithelium and an overlying fibrous cuticle. Below the epidermis, circular and longitudinal muscles were intact and from the body wall the coelom was clearly seen. Along with this, luman, blood vessels, chloragogen cells were seen clearly. Below the epidermis, the pigment cell was numerous. The tartrazine treated section of clitellar region showed less change when compared to control. The size of the blood vessel was reduced. The cuticle, epidermis and circular muscles were not changed in the experiment.



however, the coelom was enlarged. The supposed to be the pigment cells were reduced to a greater extent. Similarly in the tartrazine treated clitellar region showed very little changes in the luman, blood vessels and pigment cells. The intestinal epithelial lining, the lumen of the intestine are also damaged. Greater degree of injury to the dorsal and ventral blood vessel is observed. Ventral nerve cord is damaged beyond recognition. In the tartrazine treated and vegetable and flower waste treated sections were less affected. Inter vascular space was much reduced. Majority of the anatomical details were intact except few changes like the size of the luman, number of the pigment cells and the size of the coelom. Only in certain regions the thickness of the body wall was reduced. The vegetable and flower waste and tartrazine were not much impact on the anatomy of the *Eudrilus eugeniae*.

#### IV. Discussion

Food requirements play an important role in the growth and reproduction of earthworm population which depends on both physical and chemical properties of the soil, such as soil temperature, moisture, pH, salts, aeration and texture, as well as available food. Maximum growth of *Eudrilus eugeniae* grown in 100% cow dung. Further, they have observed that earthworms grew and reproduced favourable up to 1:1 press mud and cow dung feed composition. The present finding is in good accords with those of Sangwan, et al [11] that have registered increase in N and P in vermicompost employing *E. eugeniae* and have suggested that vermicomposting could be an alternative technology for the management of pH into useful fertilizer material, if mixed at maximum 50% with cow dung. Micronutrients like Zn content elevated, whereas Fe and Mn declined effect in flower waste compost. No considerable change in the Cu content of the vermicompost was observed in both vegetable and flower waste compost. On the other hand, during the composting process, micronutrients like Zn and Cu elevated. There was no drastic difference in the manganese content before and after composting. In contraction to the present observation, Kitturmth et al [12] have recorded Cu content (78.32 ppm) with vermicomposting 100% bagasse. Increase in the Zn content of the vermicomposted bagasse gains support from the observation of [12] that has also registered maximum Zn content of 245-61 ppm in bagasse (100%) vermicompost.

During the composting period nitrogen, phosphorous and potassium contents were measured. Generally, vermicompost was found to be rich in nitrogen and phosphorous and had good structure, low conductivity, high humic acid contents as well as good stability and maturity. Similarly, we also observed high content of nitrogen and phosphorous with good texture in the vermicompost. Nitrogen and phosphorous contents were dramatically increased in the vermicompost due to the impact of earthworms and the presence of cow dung. Recently, Abram and Ross [13] reported that nitrogen and potassium was enriched in my cost raw and cow dung based vermicompost compared to controls without earthworms. Jayakumar [14] Conducted an experiment in vermicomposting using turkey litter and cow dung mixtures (1:1) and showed a substantial variation in the nitrogen, phosphorous and potassium contents compared to the control compost. According to Atiyeh *et al.*, [15] earthworms play a major role in nitrogen transformations in manure, by enhancing nitrogen mineralization. The main aim of the present investigation was to know the extent to which flower waste is vermicomposted in order to maximize the potentials of the processes.

Earthworms can boost the nitrogen levels of the substrate during digestion in their gut adding their nitrogenous excretory products, mucus, body fluid, enzymes, and even through the decaying dead tissues of worms in vermicomposting subsystem [16]. The vermicompost prepared by the earthworm species showed a substantial difference in total N content ( $P < 0.01$ ), which could be attributed directly to the species feeding

preference of individual earthworm species and indirectly to mutualistic relationship between ingested microorganisms and intestinal mucus [17]. The total P was higher in the vermicompost harvested at the end of the experiment compared to that of the initial substrate. The enhanced P level in flower waste vermicompost suggests phosphorous mineralization during the process. The worms during vermicomposting converted the insoluble P into soluble forms with the help of solubilizing microorganisms through phosphatases present in the gut, making it more available to plants [17]; [18]; [19]. This was buttressed by increased trend of EC showing enhancement of exchangeable soluble salts in vermicompost. Vermicomposting proved to be an efficient process for recovering higher K from organic waste [17]; [20]. The present findings corroborated to those of [21] who demonstrated that higher K concentration in the end product prepared from sewage sludge.

The increase in K of the vermicompost in relation to that of the simple compost and substrate was probably because of physical decomposition of organic matter of waste due to biological grinding during passage through the gut, coupled with enzymatic activity in worm's gut, which may have caused its increase. [22] The microorganisms present in the worm's gut probably converted insoluble K into the soluble form by producing microbial enzymes [23]. Earthworms are sensitive to pH, thus pH of soil or waste is sometimes a factor that limits the distribution, numbers and species of earthworms. Little information is available on the effect of substrate pH during vermicomposting. The increased trend of pH in the vermicompost and increase in mineral constituents of waste have been observed. The increased EC during the period of the composting and vermicomposting processes is in consistence with that of earlier workers [23]; [24] which was probably due to the degradation of organic matter releasing minerals such as exchangeable Mg, K, and P in the available forms, that is, in the form of ions in the vermicompost [25]; [26]. Along with this study, a little increase of body weight was observed. Probably the increase may be due to good nutritional source of vegetable and flower waste. The indigenous species, *Eudrilus euginae*, exhibited better growth and reproduction performance is observed. The growth parameters of earthworm species cultured in Vegetable and flower waste showed that the weight increased by 27.5% in *E. euginae*, in Vegetable waste, while it increased by 49.5% in *E. euginae*, grown in flower waste(FW), whereas the in the tartrazine treated the weight was increased slightly. In the flower waste, the weight gain was increased by 49.5 %. However, the weight gain was much reduced by 33.64 in the tartrazine treated flower waste compost. The worms when introduced into wastes showed an increased growth rate and reproduction activities.

## V. Conclusion

In the present study a high degree of organic matter stabilization has been achieved in vermicompost preparation. On the basis of observation, the maturity and organic matter stabilization is found to be higher with vermicompost prepared from vegetable and flower wastes using *Eudrilus euginae*. This study demonstrates the role of earthworms and microbial population which could facilitate rapid decomposition and the rate of mineralization of wastes. The analysis clearly confirms the higher degree of degradation. The mineralization also showed high degree in the compost material. The potential of the species is observed in the analysis of micro and macro nutrients. Similarly, the physical properties also showed promising results in the present work. In the modern context, the formation of wastes increased at large level which can be utilized in a fruitful way by using earthworm for the conversions of varieties of waste into compost material. The present set of investigation is aimed in this direction and the results are encouraging. Researchers reported that the passage of organics through the earthworm's gut significantly alters the physical structure of the material. Large particles are broken down into numerous smaller particles, with a resultant enormous increase in surface area. Soil pollution bio-indicators are essential to establish environmental standards. Earthworms and hypogenous micro arthropods were chosen to test ecological risks derived from soil amendment with tartrazine. The objective of culturing earthworms in tartrazine revealed that the earthworm could live in the vegetable and flower waste. In the present investigation the *Eudrilus euginae*, was subjected to various concentrations of vegetable and flower waste. It has been observed that the waste is toxic to earthworms observed only minor damages. Earthworm can be used to dispose of all sorts of vegetable and flower wastes. The processing of waste by earthworm would help to reduce a major environmental problem, the mounting solid wastes.

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