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# Potential of distillery effluents for safe water through vermifiltration

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### Abstract

Vermifiltration of wastewater using waste eater earthworms is a newly conceived novel technology. The present study evaluated BOD, COD and TS showing significant variation in decrease by 95%, 90% and 80% respectively through vermifiltration of distillery effluents. The nutrient contents (TN, TP, TK, TCa and TMg) in the vermicasts had increase (1.82 % in TN, 0.24% in TP, 2.15% in TK, 2.07% in TCa and 2.86 % in TMg) in the range of fold than the control level. The morphology of the control and experimental vermicast samples were analyzed with SEM and the image showed significant variation. The FT-IR spectrum analysis showed reduction of aliphatic/aromatic (C=C and OH) compounds in the vermicompost. Thus, the present study significantly highlights the vermi-filtration technology in treating distillery effluent.

Keywords: Earthworms, Distillery Effluents, Vermifiltration, HRT, Vermicast, FT-IR.

## INTRODUCTION

Pollution is a gift of rapid industrial revolution and excessive exploitation of natural resources. Use of water in India is spectacular as more water consuming industries like Distillery, Textiles, Dye, Diary, Sugar, Tannery, Paper, Soap and Brewery are housed along various cities and accumulate sludge on land wastewater into rivers. The above mentioned industrial waste (sludge) and wastewater should be converted into useful manner. To avoid pollution problem the sludge and wastewater must be recycled. Both in developed and developing countries in modern age the solid waste is increasing in bulk (Abdel-Shafy and Mansour, 2018).

The government of numerous countries have striven to find solutions to reduce this problem.

Distillery listed at the top of Red Category industry among the pollution industries as per Ministry of Environment and Forests (MoEF). The industry generates biomethananted spentwash of 8-9° brix is concentrated from approximately 320 M<sup>2</sup>/day to 51 M<sup>3</sup>/day (CPCB, 2018). According to Chowdhary *et al.* (2017), the distillery units in India increased up to 319 with their annual production of alcohol and waste water making a serious pollution threat. It is mandatory for distilleries to take appropriate measures to check the disposal of wastewater.

Vermifiltration (VF) has been studied extensively due to its effectiveness in removing pollutant form wastewater, and its positive effects on the environment (Aguilera, 2003). Organic wastewater treatment using vermifiltration is the safe and the best one. This technique is first advocated by the

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Kannadasan, N. *et al.* (2019). Potential of distillery effluents for safe water through vermifiltration. *Journal of Applied and Natural Science*, 11(4): 768 - 777 https://doi.org/ 10.31018/jans.v11i4.2112 late professor Jose Toha of Chile University in 1992 (Wang *et al.*, 2010). Sinha and his group investigated the potential of the VF system in treatment of dairy industry effluent (Sinha *et al.*, 2008). They reported that earthworms by biodegradation remove BOD over 98%, COD 80-90%, TDS by 90-92%. Ghatnekar *et al.* (2010) reported VF as an efficient system to remove COD and BOD load of wastewater generated from gelatin industry. The proposed research work aimed at appraising the conversion of distillery industrial effluents and sludge retreating wastewater into through vermifiltration us in the earthworm *Eudrilus eugeniae*, and their effect on physico-chemical parameter for the benefit of the environment.

## MATERIALS AND METHODS

**Collection and culturing of worms:** The earthworms *Eudrilus eugeniae*, used for the present study was purchased from the Periyar Research Organization for Bio-Technique and Ecosystem (PROBE), Periyar Maniyammai University, Vallam, Tanjore Dist, Tamil Nadu and cultured in cement tanks for further studies. The earthworms were reared in garden soil. The vermibed of dimension 4 (length) x 2 (breadth) x 4/4 feet (height) sufficient for 1Kg (1,000 to 1,500) worms with moisture 35 - 45% was made. Entry of predators into the vermibed is presented by using nylon net. Optimum moisture was maintained by pouring waster daily.

**Collection of samples:** Distillery Effluent (DE) samples were collected from Trichy Distilleries, Senthanirpuram, Tiruchirappalli District. Only these samples were used for the experiment after proper dilution. Physico-chemical parameter were analyzed in the distillery effluent at Environmental Biotechnology Laboratory, P.G. and Research Department of Zoology, Periyar E.V.R College, Tiruchirappalli.

**Non-vermifilter (NVF) and vermifilter (VF):** The size of the VF reactor was 36 cm (long) x 36 cm (wide) x 36 cm (height). It had an upper filtering unit and lower collection unit. Filtering units were filled with gravel, sand and garden soil. The bottom most layers were filled up to 7 cm with gravel aggregated of size 10 - 20 mm, followed by gravel by size 2 - 4 mm size up to 7 cm, sand of size 1 - 2 mm up to 7 cm and the top most layer with garden soil up to 7 cm. VF reactor is same as that of the NVF reactor except for the presence of earthworms.

**Experimental design and management:** Plastic drum of 10 L capacity were filled with 6 L of effluents (DE). These drums were kept on an elevated platform just near the VF unit. One end of the flexible rubber tube was fitted to the tap of the plastic drum and the other end was placed over the VF unit. The effluent distribution system consisted of simple 0.5 inch flexible rubber pipe with hole for

trickling effluent above the soil surface of vermibed. Effluents percolated down through various layers in the vermifilter bed and at the end were collected in a bottom chamber. When the experiment started, 1 kg (1,000 to 1,500 worms) earthworms were introduced in the soil bed. The hydraulic retention time in the vermifilter bed was kept uniformly for 8 - 10 hours in all experiments.

**Hydraulic retention time:** HRT, the time the wastewater remain in contact with the worm is on essential factor for wastewater treatment. HRT is essential and it depends on the flow rate of wastewater to the VF unit, the volume of soil profile and quality of soil used. High hydraulic loading rate varies among the nature of the soil according to Sinha *et.al.*, (2008), which greatly reduce the treatment efficiency.

**Analysis of physico-chemical parameters:** VF and NVF filtered water were collected at the collection unit and analyzed for physico-chemical parameters like pH, Electrical Conductivity, Biological Oxygen Demand, Chemical Oxygen Demand, Total Solids, Total Suspended Solids and Total Dissolved Solids. The physico-chemical parameters were analyzed using standard methods (APHA, 2012). All the samples were analyzed in triplicate and the results were averaged during a working condition.

Analysis of soil layer and vermicasts: Using horizon-wise collection method, soil sample were collected both in control and vermicast. Samples of topsoil upto7 cm were collected from sampling points at intervals of 7 days. Subsequently, earthworms were removed from the substrates. Total nitrogen was measured by micro Kjeldahl method (Jackson, 1975). Extractable phosphorous was determined (Olsen *et.al.*, 1954). Exchangeable elements (K, Ca and Mg) were determined using ammonium acetate extractable method (Simard, 1993) and the vermicasts samples were measured/estimated in the Non-vermifilter (NVF) and vermicast samples.

**Morphology of the vermicast:** Vermicasts were collected from vermibed of control and DE experimental samples (VF-75%, VF-50%, VF-25%) from different sampling points. Samples from the same depth were mixed to give one composite sample. Finally, from the samples casts were freeze-dried, sieved (<2 mm) and their surface structure was analyzed using a Scanning Electron Microscope (SEM).

**FT-IR Spectra analysis:** The Fourier transform infrared (FT-IR) spectra used to find out the major functional groups in the earthworm cast (He *et al.*, 2011 and Xing *et al.*, 2011).

**Statistical analysis:** The data entry and Analysis of Variance (ANOVA) were analyzed using SPSS version-16.0. One-way ANOVA was used to test differences in the related physico-parameters of effluents using of VF systems under similar influ-

ent conditions. Duncan's multiple range tests was used to further assess differences among VF systems that were significant in ANOVA. The statistical significance were at p < 0.05 probability levels.

# **RESULTS AND DISCUSSION**

**Physico-chemical characteristics:** The quality of DE in terms of physico-chemical characteristics are given in Tables 1 - 2. The pH value of raw DE was acidic in nature 4.5, the raw spent wash is acidic in nature and the pH values of distillery effluents range from 3.5 to 5.0. But when it was diluted with different dilution 25%, 50% and 75%.

The pH values of raw, diluted and filtered effluent from VF and NVF units were observed. The average pH of DE was  $5.56 \pm 0.02^{a}$  in 75%,  $5.54 \pm 0.01^{a}$  in 50% and  $5.54 \pm 0.02^{a}$  in 25%. The pH recorded after the treatment was  $6.12 \pm 0.02^{a}$  in 75%,  $7.07 \pm 0.01^{b}$  in 50%,  $7.36 \pm 0.01^{c}$  in 25% NVF and  $7.02 \pm 0.02^{a}$  in 75%,  $7.51 \pm 0.01^{b}$  in 50%,  $7.61 \pm 0.01^{c}$  in 25% VF unit. From ANOVA test, it showed significant difference in values of pH, when worm density is varied, but no significant difference when concentration is varied throughout the VF process (p < 0.05).

The result of the present study is in accordance

Table	1. Ph	vsico-chemical	characteristics	of the ra	aw and	diluted	distiller	/ effluent
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Paramotors	Raw Effluent -	Diluted Distillery Effluent			
Farameters		75%	50%	25%	
pН	4.5	5.56 ± 0.02 <sup>a</sup>	5.54 ± 0.01 <sup>a</sup>	5.54 ± 0.02 <sup>a</sup>	
EC (mS/cm)	18.86	15.47 ± 0.35 <sup>c</sup>	12.34 ± 0.03 <sup>b</sup>	$7.52 \pm 0.02^{a}$	
BOD <sub>5</sub> (mg/L)	32533.00	18100.00 ± 3.60 <sup>c</sup>	11700.00 ± 21.36 <sup>b</sup>	5863.67 ± 3.21 <sup>a</sup>	
COD (mg/L)	97600.00	$54400.00 \pm 50.00^{\circ}$	35200.00 ± 50.00 <sup>b</sup>	17600.00 ± 17.55 <sup>a</sup>	
TS (mg/L)	70000.00	57200.00 ± 200.00 <sup>c</sup>	35100.00 ± 100.00 <sup>b</sup>	23200.00 ± 251.66 <sup>a</sup>	
TSS (mg/L)	8000.00	7116.67 ± 104.08 <sup>c</sup>	5073.33 ± 87.36 <sup>b</sup>	4143.33 ± 128.97 <sup>a</sup>	
TDS (mg/L)	62000.00	50100.00 ± 104.08 <sup>c</sup>	32000.00 ± 25.16 <sup>b</sup>	19100.00 ± 138.92 <sup>a</sup>	

\*Results are the mean value in triplicates ± SD with significant difference at p < 0.05.

Table 2. Physico-chemical characteristics of the distillery effluent at different concentration.

Deremetere	Non-Vermifilter (NVF)			Vermifilter (VF)			
Farameters	75%	50%	25%	75%	50%	25%	
pН	$6.12 \pm 0.02^{a}$	7.07±0.01 <sup>b</sup>	7.36±0.01 <sup>c</sup>	7.02±0.02 <sup>a</sup>	7.51±0.01 <sup>b</sup>	7.61±0.01 <sup>c</sup>	
EC (mS/cm)	6.53 ± 0.01 <sup>c</sup>	5.41±0.01 <sup>b</sup>	4.73±0.03 <sup>a</sup>	3.95±0.03 <sup>c</sup>	2.03±0.04 <sup>b</sup>	1.74±0.01 <sup>a</sup>	
BOD <sub>5</sub> (mg/L)	3092.67 ± 2.51 <sup>c</sup>	2027.00±2.64 <sup>b</sup>	1066.00±4.00 <sup>a</sup>	929.33±27.68 <sup>c</sup>	527.67±25.42 <sup>b</sup>	425.00±3.60 <sup>a</sup>	
COD (mg/L)	9265.00 ± 15.00 <sup>c</sup>	6058.33±20.20 <sup>b</sup>	2809.33±9.01 <sup>a</sup>	3174.00±22.71°	1615.00±20.22 <sup>b</sup>	1250.67±25.71 <sup>a</sup>	
TS (mg/L)	23100.00±152.75 <sup>c</sup>	14100.00±76.37 <sup>b</sup>	11000.00±36.05 <sup>a</sup>	7016.67±20.81 <sup>c</sup>	6036.67±55.07 <sup>b</sup>	3016.67±15.27 <sup>a</sup>	
TSS (mg/L)	6080.00±75.49 <sup>c</sup>	4026.67±25.16 <sup>b</sup>	3011.67±10.40 <sup>a</sup>	3011.00±16.52 <sup>c</sup>	2254.33±437.92 <sup>b</sup>	1011.67±10.40 <sup>a</sup>	
TDS (mg/L)	17100.00±83.86 <sup>c</sup>	12100.00±51.31 <sup>b</sup>	8028.33±25.65 <sup>a</sup>	5005.67±5.13 <sup>c</sup>	4115.67±194.31 <sup>b</sup>	2005.00±5.00 <sup>a</sup>	

Results are the mean value in triplicates  $\pm$  SD with significant difference at p < 0.05.

Table 3. Level of macro and micronutrients in DE vermicast at different concentrations.					
S. N.	Parameters	Control	VF - 75%	VF - 50%	VF - 25%
1	Total Nitrogen (%)	1.03 ± 0.01 <sup>c</sup>	1.78 ± 0.01 <sup>b</sup>	1.82 ± 0.01 <sup>a</sup>	1.82 ± 0.01 <sup>a</sup>
2	Total Phosphorus (%)	0.16 ± 0.01 <sup>b</sup>	0.23 ± 0.01 <sup>a</sup>	0.23 ± 0.01 <sup>a</sup>	$0.24 \pm 0.00^{a}$
3	Total Potassium (%)	1.03 ± 0.01 <sup>c</sup>	2.14 ± 0.04 <sup>b</sup>	2.11 ± 0.00 <sup>c</sup>	2.15 ± 0.00 <sup>a</sup>
4	Total Calcium (%)	1.64 ± 0.01 <sup>d</sup>	2.02 ± 0.01 <sup>c</sup>	2.05 ± 0.01 <sup>b</sup>	$2.07 \pm 0.01^{a}$
5	Total Magnesium (%)	1.05 ± 0.01 <sup>d</sup>	2.25 ± 0.01 <sup>c</sup>	2.46 ± 0.01 <sup>b</sup>	2.86 ± 0.01 <sup>a</sup>

Results are the mean value in triplicates  $\pm$  SD with significant difference at p < 0.05.

Table 4. Assignment of typical infrared bands in FT-IR spectra of OM (NVF and VF).				
Wave Number (cm-1)	Vibration	Functional Group or Component	References	
3700 – 3400	OH stretch	Hydroxyl (Polysaccharide) & Amino Group	Simkovic <i>et al.</i> , 2008	
1649 – 1521	C=C stretch	Aromatic Group	Karthika <i>et al</i> ., 2015	
1490 – 1340	OH deformation, Symmet- ric COO– & OH stretch	Aliphatic, Phenols & Car- boxylic Groups	Hafidi <i>et al</i> ., 2005	
	Combination of C-O- C		Grube et al., 2006; Contreras-	
1095 – 1030	Stretching & O-H defor- mation	Polysaccharides	Ramos <i>et al.</i> , 2004; Ravindran <i>et al.</i> , 2008	
1030 – 790	Si-O-Si stretch	Clay minerals	Senthil kumar et al., 2013	
			Carrasquero-Duran and Flores,	
850 – 750	C - O stretch	Carbonate	2009; Contreras-Ramos et al.,	
		<b></b>	2004; Ravindran <i>et al.</i> , 2008	
475 – 460	SI-O stretch	Silica	Nayak and Singh, 2007	

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DE Non-Vermifilter sludge DE Vermifilter vermicast

Fig. 1. SEM images of after NVF Sludge and VF vermicast

with Azuar and Ibrahim (2012) and Garkal and Jadhao (2014) applied the VF technology in treating the palm oil mill effluent. Earthworm has an inbuilt pH buffering ability by increasing the pH, hence neutralizing the sewage wastewater pH (Manyuchi *et al.*, 2012). The pH value difference between control and VF could be related to earthworm mediated rapid mineralization of organic fractions of wastewater (Rajpal *et. al.*, 2012). The pH of effluent from all the VFs increased initially during the treatment and then settled to neutral range signifying the inherent capability of earthworms to act as buffering agent to neutralize (Arora *et al.*, 2014).

The Electrical Conductivity (EC) of DE showed significant changes after treatment through a filtration system in both NVF and VF processes. The EC of the raw and diluted (75%, 50% and 25%)

DE was 18.86 (mS/cm) and 15.47 ± 0.35<sup>c</sup> in 75%,  $12.34 \pm 0.03^{b}$  in 50%, 7.52  $\pm 0.02^{a}$  (mS/cm) in 25%. The EC of treated DE was 6.53 ± 0.01°, 5.41  $\pm$  0.01<sup>b</sup>, 4.73  $\pm$  0.03<sup>a</sup> (mS/cm) in NVF and 3.95  $\pm$  $0.03^{\circ}$  in 75%, 2.03 ± 0.04<sup>b</sup> in 50%, 1.74 ± 0.01<sup>a</sup> (mS/cm) in 25% VFs. The difference between NVF and VF reactor was statistically significant. The microbial activity of earthworms gut increase the EC of the wastewater by mieralization than control. Increased EC of substrate may be the result of release of minerals from the gut of earthworm (Kaur et al., 2010 and Suthar, 2010). Biochemical Oxygen Demand (BOD<sub>5</sub>) is the most important parameters used to determine the dearee of pollution of aquatic habit. Table 1 - 2 indicates the BOD of the raw DE as 32533.00 mg/L. The BOD of raw and diluted DE was 18100.00 ±  $3.60^{\circ}$  in 75%, 11700.00 ± 21.36<sup>b</sup> in 50% and 5863.67 ± 3.21<sup>a</sup> mg/L in 25%. The level of BOD removal from the DE in VF, was 929.33 ± 27.68<sup>c</sup> in 75%, 527.67 ± 25.42<sup>b</sup> in 50% and 425.00 ± 3.60<sup>a</sup> mg/L in 25%. Where as for NVF it was 3092.67 ± 2.51<sup>c</sup> in 75%, 2027.00 ± 2.64<sup>b</sup> in 50% and 1066.00

 $\pm$  4.00<sup>a</sup> mg/L in 25%. The average BOD removal from the DE by earthworm is over 95%, while that without earthworms is over 80% for NVF. This could be due to the symbiotic activity of earthworms and aerobic microbes. The decomposition of organic waste by the symbiotic activity of earth-







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Fig. 4. FT-IR spectra of the DE sample of vermicast produced by vermibed in NVF – 25%.





Fig. 6. FT-IR spectra of the DE sample of vermicast produced by vermibed in VF – 50%.

worm and aerobic microbes acts as biological catalysts to reduce the BOD of wastewater (Tomar and Suthar, 2011; Sinha *et al.*, 2008; Ghatnekar *et al.*, 2010 and Azuar and Ibrahim, 2012).

The values of Chemical Oxygen Demand (COD) in filterate from VF and NVF is illustrated in Table

1 and 2. The data showed that the COD of effluent was significantly low (p < 0.05) in VF and NVF units as compared to influent as mentioned in the Table. COD is an important indicator of chemical load of industrial and domestic wastewater. The COD of the raw DE was 97600.00 mg/L, diluted DE were 54400.00 ± 50.00<sup>c</sup> in 75%, 35200.00 ±

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Fig. 7. FT-IR spectra of the DE sample of vermicast produced by vermibed in VF – 25%.

50.00<sup>b</sup> in 50% and 17600.00  $\pm$  17.55<sup>a</sup> mg/L in 25%. COD removal of the treated DE in different dilution in VF was 3174.00  $\pm$  22.71<sup>c</sup> in 75%, 1615.00  $\pm$  20.22<sup>b</sup> in 50% and 1250.67  $\pm$  25.71<sup>a</sup> mg/L in 25%, while for NVF it was 9265.00  $\pm$  15.00<sup>c</sup> in 75%, 6058.33  $\pm$  20.20<sup>b</sup> in 50% and 2809.33  $\pm$  9.01<sup>a</sup> mg/L in 25%. In case of VF reactor, the average removal efficiency of COD was recorded as 90%, while in NVF the COD reduction was represented as 80%. The acceleration of microbial colonization in variables by assimilable carbon and other nutrients reduce 46% of volatile suspended solids and thereby reduce the COD by vermifilteration (Wang *et al.*, 2010; Zhao *et al.*, 2010; Aira *et al.*, 2007 and Zhao *et al.*, 2010).

Total Solids (TS) significantly reduced by VF (p <0.05) as shown in Table 1 and 2, the raw DE had 70000.00 mg/L. These values decrease after dilutions of DE. It was 57200.00 ± 200.00<sup>c</sup> in 75%, 35100.00 ± 100.00<sup>b</sup> in 50% and 23200.00 ± 251.66<sup>a</sup> mg/L in 25%. DE filtered from VF was 7016.67  $\pm$  20.81° in 75%, 6036.67  $\pm$  55.07° in 50% and 3016.67  $\pm$  15.27° mg/L in 25%, while in NVF it was 23100.00 ± 152.75<sup>c</sup> in 75%, 14100.00 ± 76.37<sup>b</sup> in 50% and 11000.00 ± 36.05<sup>a</sup> mg/L in 25%. The overall TS removal efficiency in both experimental plants were 80% in VF and 60% in NVF. Total Suspended Solids (TSS) and Total Dissolved Solids (TDS) showed a drastic reduction during NVF and VF (Table 1 and 2). The concentration of both TSS and TDS were reduced during VF significantly (p < 0.05).

The TSS and TDS were 8000.00 mg/L and 62000.00 mg/L in raw DE. The TSS and TDS of the diluted DE was 7116.67  $\pm$  104.08<sup>c</sup> in 75%, 5073.33  $\pm$  87.36<sup>b</sup> in 50% and 4143.33  $\pm$  128.97<sup>a</sup> mg/L in 25% and 50100.00  $\pm$  104.08<sup>c</sup> in 75%, 32000.00  $\pm$  25.16<sup>b</sup> in 50% and 19100.00  $\pm$  138.92<sup>a</sup> mg/L in 25%. The results of the resent study suggest the efficiency of vermifiltration pro-

cess in removing the total solid fraction of effluent. The TSS and TDS removal rate were high in VF unit ( $3011.00 \pm 16.52^{\circ}$  in 75%, 2254.33  $\pm 437.92^{b}$  in 50% and 1011.67  $\pm 10.40^{a}$  mg/L in 25% and 5005.67  $\pm 5.13^{\circ}$  in 75%, 4115.67  $\pm 194.31^{b}$  in 50% and 2005.00  $\pm 5.00^{a}$  mg/L in 25%) than NVF system ( $6080.00 \pm 75.49^{\circ}$  in 75%, 4026.67  $\pm 25.16^{b}$  in 50% and 3011.67  $\pm 10.40^{a}$  mg/L in 25% and 17100.00  $\pm 83.86^{\circ}$  in 75%, 12100.00  $\pm 51.31^{b}$  50%, 8028.33  $\pm 25.65^{a}$  mg/L in 25%). The VF unit significantly removed the TSS and TDS from the DE about 80% and 88%, while the NVF bed indicated 70% and 65% decrease. The reduction in total solid waste may be attributed to the earthworms biodegradation using gut microbes (Kumar *et al.*, 2014).

During VF process, a tortuous behavior for TSS concentration was also observed accordingly with Sharma *et al.*, (2014), Sharma and Kazmi, (2014). From literature available it has been evidenced that these organic and inorganic particles are further trapped in the voids of vermifilter and causes high removal efficiency of TSS and TDS from wastewater (Sinha *et al.*, 2008). According to Cooper *et al.* (1996) and Vymazal *et al.* (1998) the suspended solids that are not removed through pre-treatment system are effectively removed by filtration and settlement processes. The efficacy of VFs in TDS and TSS removal is also reported by earlier authors were comparable with the present results.

Effect of the physico-chemical characteristics of vermicast: The percentage of Total Nitrogen (TN) showed an increase in DE sludge treated with *Eudrilus eugeniae*  $1.78 \pm 0.01^{\text{b}}$  in 75%,  $1.82 \pm$  $0.01^{\text{a}}$  in 50%,  $1.82 \pm 0.01^{\text{a}}$  in 25% and  $1.03 \pm$  $0.01^{\text{c}}$  in control (Table-3). Significant variation (*p*<0.05) was observed in TN, as compared to control. The present data clearly suggested that nitrogen increase in vermicomposted material was directly related to the physico-chemical properties of the initial substrates as compared with the results of Battacharya *et al.* (2004). Bedding materials modify the physical structure of waste and also accelerate the waste mineralization rate in vermibeds. Other reports on vermicomposting (Suthar, 2008) have reported a higher N increase at the end. Decaying tissues of dead worms are also one of the factors for TN increase in significant amount.

The elevated level of Total Phosphorus (TP) content was recorded in DE sludge treated with earthworm  $0.23 \pm 0.01^{a}$  in 75%,  $0.23 \pm 0.01^{a}$  in 50%,  $0.24 \pm 0.00^{a}$  in 25% and  $0.16 \pm 0.01^{b}$  in control (Table 3). Recently Suthar (2009) reported higher content of available P in organic wastes due to concentration of sludge in bedding material as observed in the present result. Difference in TP content in all the reactors in all the sampling days were significant (p < 0.05) in the present study. The range of Total Potassium (TK) content was recorded in DE sludge treated with Eudrilus eugeniae 2.14  $\pm$  0.04<sup>b</sup> in 75%, 2.11  $\pm$  0.00<sup>c</sup> in 50%,  $2.15 \pm 0.00^{a}$  in 25% and 1.03  $\pm 0.01^{c}$  in control (Table-3). The results of this study agree with previous report of Suthar, (2009) that the vermicomposting process accelerates the microbial populations in waste and subsequently enriches the end product with most available forms of plant nutrients. Earthworms are very sensitive to ammonia and cannot survive in organic waste containing high level of cation (Dominguez, 2004).

The observed values of Total Calcium (TCa) and Total Magnesium (TMg) present in the control were found to be  $1.64 \pm 0.01^{d}$  and  $1.05 \pm 0.01^{d}$ . Analysis of total Ca and Mg present in the DE sludge treated with earthworms Eudrilus eugeniae was found to be  $2.02 \pm 0.01^{\circ}$  in 75%,  $2.05 \pm 0.01^{\circ}$ in 50%, 2.07  $\pm$  0.01<sup>a</sup> 25% and 2.25  $\pm$  0.01<sup>c</sup> in 75%, 2.46  $\pm$  0.01<sup>b</sup> in 50%, and 2.86  $\pm$  0.01<sup>a</sup> in 25% (Table-3). Ca and Mg increased significantly (p<0.05) from control to different concentration of VF treatments. The increase in Ca had a higher proportion of bedding materials. Earthworm drives the mineralization process efficiently which results in higher concentration of Mg in the vermicompost (Suthar, 2008; 2009 and Kannadasan et al., 2013).

**Morphology of vermicast:** Scanning Electron Microscope (SEM) was presented with the changes of physico-chemical structure of the cast in Vermifilter (VF) and Non-vermifilter (NVF). Most of the organic contaminants were removed by precipitation and adsorption in the voids of the soil or vermicast which served as wastewater filter media (Wang *et al.*, 2010). Typical SEM micrographs of VF and NVF samples, as well as the vermicast sample, are shown in Fig. 1. The NVF sample appeared to have a loose, flaky structure, while the vermicast sample exhibited a distinct physical

appearance characterized by a predominantly spherical cell-like structure, which was more compact than in the NVF (Zhao *et al.*, 2010).

The structure of VF sample was more compact than that of the NVF sample, but more loosely arranged than that noted in the vermicast and was looser than that of the control earthworm cast, implying that structure of VF and may be an important constituent of VF. These results confirm that earthworm casts are an important factor that results in higher activities of microbial enzymes in VF compared with NVF. This finding is important because the improved sludge settling characteristics notonly reduce the amount of sludge that needs to be disposed of, but also lead to a decrease in the associated sludge processing costs and environmental burden (Elissen *et al.*, 2006).

Evaluation of vermifiltration process by FT-IR spectroscopy: The type of chemical compound of a material confirms the presence of functional group through FT-IR analysis. The presence or absence of peaks for functional groups of metabolites indicates the degradation of stabilization of the biowaste during the bioconversion process. The infrared spectra were in accordance to the previous studies (Ellerbrock and Kaiser, 2005; Romero et al., 2007; Li et al., 2011). Moreover, the main absorption bands and corresponding assignments are listed in table-4. In general, the FT-IR spectra of DE vermicast from different vermifiltration stages were similar but varied significantly in the relative intensity of absorption bands. With increasing vermicast time, the main changes in FT-IR spectra were summarized as follows:

In the region from 3750 to 3400 cm<sup>-1</sup> absorbances are reported to correspond to hydroxyl (OH) stretching vibrations (polysaccharide) and amino groups (Simkovic *et al.*, 2008). Gupta and Garg (2010) have reported that the vermicomposting process reduced in band 3600 to 3100 cm<sup>-1</sup> region in FT-IR spectra of vermicomposted cow dung as compared to raw cow dung. Li *et al.* (2011) and Busato *et al.* (2012) have reported that vermicomposting process caused the disappearance of easily biodegradable compounds and enhanced the increase of aromatic compounds, which was confirmed by FT-IR analysis. Phenols and Carboxylic group are present (O–H stretching at 3500 to 3400 cm<sup>-1</sup>).

The FT-IR spectra of water extracts from the vermicast were similar but significant in the relative intensity of absorption bands. Decrease of the polysaccharides-like substance, protein like, aliphatic compounds and carboxylic groups were observed from the FT-IR after the vermifiltration treatment (Yang *et al.*, 2014). The peak value around 1649 to 1521 cm<sup>-1</sup> was found due to be the presence of C = C aromatic structure (Karthika *et al.*, 2015). The increase in the aromatic C and aliphatic C ratio is considered as an indicator of an increasing degree of organic matter humification in the natural condition of biodegradation (Senesi and Brunetti, 1996) which could be associated with the stability and maturity of compost in their transformation of highly humified substrate (Huag *et al.*, 2006).

The absorbance peak situated from 1490 to 1340 cm<sup>-1</sup> might be associated with the bending vibration of aliphatic, OH deformations, symmetric carboxyl (COO<sup>-</sup>) stretch, and stretching of phenolic OH as compared with the results of Hafidi et al., (2005). A slight broad band around 1095 to 1030 cm<sup>-1</sup> related to C–O–C groups in polysaccharides. The sharp band appearing at 850 to 750 cm<sup>-1</sup> was related to C-O stretching of carbonate and silica as observed from the results of Carrasquero-Duran and Flores (2009), Contreras-Ramos et al., (2004) and Ravindran et al., (2008). However, the intensity of the bands of 1033 cm<sup>-1</sup> and 468 cm<sup>-1</sup> slightly decreased after effluent treatment and the absorbance of Si-O stretching vibrations was characteristic for the 1033 cm<sup>-1</sup> region. Those of Si-O bending vibrations are from 475 to 460 cm<sup>-1</sup> (Nayak and Singh, 2007).

Furthermore, Fig. 2-7 shows that the organic matter content in the sand was less decomposed or accumulated in VF than in the NVF. The spectral information of these results by Galle *et al.*, (2004) reflects a critical change in the OM quality of the media before and after sewage treatment, which has been consistently observed in river sediments. Certain functional chemical attributes could evaluate the OM contents at the VF media. Thus, the functional group changes probably can explain why the NVF served as the main filter, as well as why NVF played an important role in transforming C cycle in the earthworm packing bed.

# Conclusion

Vermifiltration technology which uses epigeic earthworms as a means of aerobically treating wastewater is increasingly becoming an ecofriendly wastewater treatment technique. From the above study it is concluded that the polluted wastewater could be treated with an optimal dilution through vermifiltration. The water analyzed after the filtration showed decrease level in BOD, COD and TS by 95%, 90% and 80% respectively. The vermicast from vermifilter on analysis of parameters such as TN, TP, TK, TCa and TMg showed an increased level. The FTIR spectra of DE vermicast were similar but varied in the relative intensity of absorption bands. The increase in the aromatic carbon ratio is considered as an indicator of an increasing degree of organic matter humification, which the stability and maturity of compost their transformation highly humified substrate. Here in this study 25% dilution was found to be a suitable. Hence vermifiltration is a low cost, odourless and non-labour intensive method

of wastewater treatment. In addition, the refuge of such techniques might be easily used in agriculture and in environmental decontamination.

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