STUDIES ON FACTORS AFFECTING VERMIFILTRATION FOR WASTEWATER TREATMENT

Ph.D. THESIS

by

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INDIAN INSTITUTE OF TECHNOLOGY ROORKEE
ROORKEE - 247 667 (INDIA)
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A THESIS

Submitted in partial fulfilment of the
requirements for the award of the degree
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CANDIDATE'S DECLARATION

I hereby certify that the work which is being presented in the thesis entitled "**Studies on Factors Affecting Vermifiltration for Wastewater Treatment"** in partial fulfilment of the requirement for the award of the Degree of Doctor of Philosophy and submitted in the Department of Civil Engineering of the Indian Institute of Technology Roorkee, Roorkee is an authentic record of my own work carried out during a period from July, 2010 to March, 2015 under the supervision of Dr. K.S. Hari Prasad, Professor, Department of Civil Engineering, Indian Institute of Technology Roorkee, Roorkee, India.

The matter presented in this thesis has not been submitted by me for the award of any other degree of this or any other Institute.

(TARUN KUMAR)

This is to certify that the above statement made by the candidate is correct to the best of my knowledge.

(K.S. Hari Prasad)
Supervisor

Date: March, 2015

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ABSTRACT

Pollutants removal through ecological system is regarded as the most viable means of decentralized wastewater treatment, as even at present there is a large gap between generation and treatment of wastewater which emphasizes the use of such technologies that could overcome these issues. The treatment systems that require relatively low costs, energy, and maintenance are preferable for the treatment of domestic wastewater. Hence, this work is an attempt to arrive at a consensus on treatment of these pollutants in a single arrangement using vermifiltration process. It comprise mainly of storage tank, wastewater distributer and the treatment unit.

The major gaps identified in order to pursue the experimental work were emphasized on optimization of factors affecting vermifiltration process, as it depends on Earthworm's population (Stocking Density), Hydraulic Loading Rate (HLR) and Hydraulic Retention Time (HRT). In addition to this for evaluating the potential impacts of physical characteristics on the overall performance of vermifilter different kinds of material such as river bed material, wood coal, glass ball and mud ball as a media have been tried.

Several laboratory experiments were conducted to evaluate the effect of stocking density (varied from 5000 to 30000 worms/cum of vermifilter bed), filter media and hydraulic loading rate (varied from 0.5 to 2.5 $m^3m^{-2}d^{-1}$) on vermifiltration process. Four types of media were used for experimentation, i.e. (a) River bed material, (b) Wood coal, (c) Glass balls and (d) Mud balls. Besides, the vermifilter was evaluated against varying degree of organic shock loads 675-1410 mg/L of COD. The main conclusions inferred from the study are presented below.

In Phase-I effect of earthworm's stocking density on vermifiltration was evaluated.

The stocking density of 10000 worms/cum of vermifilter bed was found to be relatively better as compared with other stocking densities like 5000, 15000, 20000, 25000 and 30000 worms/cum, to achieve the requisite quality of effluent. The results showed that effluent quality of vermifilter with 10,000 W/cum exhibited highest percentage removal of BOD (90%), COD (68%) and total suspended solids (70%). The average reduction of the population of indicator organisms i.e. TC, FC, FS and *E. coli* was observed as 3.61±0.90, 3.14±0.67, 2.73±0.37 and 2.27±0.28 log unit, respectively to the levels considered acceptable for either recreation or irrigation. The growth pattern of earthworm (*E. fetida)* showed a maximum individual biomass (121.4%) and growth rate (0.94 g wt. worm^{-1} day^{-1}) in earthworm's stocking density of 10000 worms/cum.

Phase-II emphasized on identification of suitable media as a vermifilter bed. Vermifilter was evaluated using different material as a media like river bed material, wood coal, glass balls and mud balls. River bed material was found to be relatively better as a media in vermifilter for better growth of earthworm biomass and achieve the requisite quality of effluent. The results demonstrated that effluent quality of vermifilter having river bed material showed highest percentage removal of BOD (78%), COD (71%) and total suspended solids (73%). The average reduction of the population of indicator organisms i.e. TC, FC, FS and *E. coli* was observed as 3.6±0.90, 3.4±0.67, 2.5±0.51 and 3.32±0.62 log unit, respectively. In reactor VFR maximum earthworm biomass was observed with 73% increment.

Phase-III is concerned with the effect of hydraulic loading rates on the performance of vermifilter. When the comparison was carried out among different HLRs like 0.5, 1.0, 1.5, 2 and 2.5 $m^3m^{-2}d^{-1}$ the optimum hydraulic loading rate was observed to be vary in the range of 1-1.5 $m^3m^{-2}d^{-1}$. Vermifiltration system represented perfect efficacy when it was compared with conventional geofilter with better quality of effluent. It observed to be having higher hydraulic conductivity as compared to geofilter. At HLR of 1.0 $m^3m^{-2}d^{-1}$ the removal of various pollutant i.e. BOD, COD and TSS was observed as 86.9, 79.6 and 68.9%, respectively. While at HLR of 1.5 $m^3m^{-2}d^{-1}$ it was resulted as 85.5, 73.9 and 76.1%, respectively. During study, the augmented earthworm biomass increased significantly with increase in HLR up to 1.0 m^3m^{-2} d^{-1}, above which the biomass growth gets reduced. Comparative study on hydraulic aspects of vermifilter and geofilter at different hydraulic loading rates indicated that hydraulic conductivity decreased from its initial value of 0.005 cm/sec as taken when the vermicompost was placed in both the reactors. However, it was more than that a conventional geofilter due to channelization in vermifilter through earthworms.

Phase-IV confined with development and design of vermifilter on the basis of outcomes from Phase I, II and III, for effective treatment of domestic wastewater. During this phase a comparison was also carried out between vermifilter and geofilter. The results demontrated that in vermifilter removed a considerable amount of BOD, COD, TSS and indicator organisms as compared to the conventional geofilter. In vermifilter the average COD removal efficiency was observed as 76% while in geofilter it was found as 63%. Similarly, about 85% average BOD removal efficiency was exhibited in vermifilter while in geofilter it was observed to be 71%. The results of three month study revealed that the presence of earthworms in vermifilter could efficiently remove TC, FC, FS and *E. coli* and the effluent concentration were observed as $2.89\times10^2 \pm 1.14\times10^2$, $1.38\times10^2 \pm 1.11\times10^2$, $1.86\times10^1 \pm 0.56\times10^1$ and $9.26\times10^1 \pm 0.85\times10^1$, respectively. While in geofilter it was observed as $5.30\times10^3 \pm 0.91\times10^3$, $4.30\times10^3 \pm 2.32\times10^3$, $7.31\times10^2 \pm 1.76\times10^2$ and $1.90\times10^3 \pm 9.48\times10^2$, respectively. This implies that vermifilter is able to bring indicator organisms to levels considered safe for irrigation as compared to conventional geofilter. When organic shock loads on vermifilter was applied by increasing the influent COD concentration from 1.5 to 3 times of normal values, the system recovered quickly. It proves its resilience power against organic shock loads, as no significant changes was noted on the performance during vermifiltration process. Results obtained in this study indicated that vermifilter employing *Eisenia Fetida* help to guarantee the organics removal efficiency and stability of vermifilter if subjected to organic shock loads. This study also promotes an understanding of organic matter removal in the system and experimental results can be used for estimating treatment efficiency of full-scale reactors under similar operational conditions. Kinetic study revealed that the biological degradation of organics follows first-order kinetics with respect to initial sCOD concentration during vermifiltration process. The treated effluent and final vermicompost both were found to be rich in nitrate and phosphate which can be used for agriculture purpose.

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ABBREVIATIONS

ALP	Alkaline phosphatase
BOD	Biochemical oxygen demand
BF	Biofilter
COD	Chemical oxygen demand
CR	Control reactor (geofilter)
CRR	Control reactor having river bed material
DO	Dissolved oxygen
EC	Electrical conductivity
EDCs	Endocrine disrupting chemicals
E. andrei	*Eisenia andrei*
E. coli	*Escherichia coli*
E. eugeniae	*Eudrilus eugeniae*
E. Fetida	*Eisenia fetida*
FC	Fecal coliform
FS	*Fecal streptococci*
HLRs	Hydraulic loading rates
HRT	Hydraulic retention time
OLR	Organic loading rate
PSS	Pseudo steady-state condition

sBOD	Soluble biochemical oxygen demand
sCOD	Soluble chemical oxygen demand
SOD	Superoxide dismutase
STP	Sewage treatment plant
TC	Total coliform
TN	Total nitrogen
TOC	Total organic carbon
TP	Total phosphate
TS	Total solids
TSS	Total suspended solids
VF	Vermifilter
VFC	Vermifilter having wood coal media
VFG	Vermifilter having glass ball media
VFM	Vermifilter having mud ball media
VFR	Vermifilter having river bed material

INTRODUCTION

1.1 General

Due to the increasing population and scarcity of treatment area, high cost of wastewater collection and its treatment is not permitting the conventional sewage treatment plant (STP) everywhere. If enhanced physico-chemical treatment at the initial stage is effective, then the organic load on any subsequent biological treatment phase will be considerably reduced (Bhuptawat et al., 2007; Marques et al., 2011; George et al., 2013). Worldwide, it is estimated that globally one person in five, i.e., about 1.1 billion people, lack access to safe water. With respect to adequate sanitation, the outlook is even more discouraging: about 2.6 billion people, almost half of the global population, are without access to proper sanitation (Torrijos et al., 2001; Ali et al., 2009). Hence, cost effective decentralized and eco-friendly treatments are needed which have the potential to overcome such issues. Besides, selection of appropriate wastewater treatment technology that enables sustainable development exhibits a challenge to national, regional and local policy makers. A decision support tool for wastewater treatment technology selection is needed in developing countries because there is no such tool available at present that is suitable for use in the context of a growing economy and increasing burdens on existing environmental resources. The need for such type of tool in developing countries is prominent because there is a large gap between wastewater generation and available treatment capacity (Arceivala and Asolekar, 2007; Patel et al., 2014). Study conducted by Kalbar et al., (2012) demonstrated that the technology selection in India is mainly skewed towards a certain number of criteria such as a compliance with stipulated regulatory standards and the technology cost. Many developing countries including India, cannot afford the construction of STPs, and hence, there is a growing need for developing some ecologically safe and economically viable onsite small-scale wastewater treatment technologies (Sinha et al., 2007; Kalbar et al. 2013; Kumar et al., 2014). At this crucial juncture, some ecologically engineered tools can address issues related with safe and cost-effective wastewater treatments technologies. Vermifiltration is one such an alternative treatment method wherein organically polluted wastewater can be treated using earthworms. Vermifiltration can be very useful for small communities, colonies and villages (Sinha et al., 2012). There is an increasingly important issue of a new category of contaminants that have been wildly distributed in the environment (Yan et al., 2009) and vermifiltration can play a good role in removing these contaminants (Sinha et al., 2008). In India, most of the population is using septic tanks for wastewater disposal which exhibits a largest volumetric source of contaminant that contribute to the ground-water zone (Ojha et al., 2011). Hence vermifiltration can be a good alternative in such area as treated wastewater can be directly useful for irrigation or sewage farming while the end product found on top layer can be useful as manure as it is nutrient rich with high nitrogen and phosphorus content (Hu et al., 2005; Gupta et al., 2007). Intensification of soil processes and aeration by the earthworms enable the soil stabilization and filtration system to become effective and smaller in size (Sinha et al., 2008). During vermifiltration, the suspended solids are trapped on top of the vermifilter and processed by earthworms and fed to the soil microbes for immobilization. Experiences have shown that the geo-microbial system gets ‘choked’ after sometimes due to slow deposition of wastewater solids as ‘sludge’ and becomes un-operational whereas, the vermifiltration system with earthworms continues to operate. Hence, no sludge formation during the vermifiltration process which requires additional expenditure on landfill disposal as sludge arising from wastewater treatment has become a critical environmental issue due to growing water demands and more stringent water quality standards (Kumar and Alappat, 2004; Lebigue et al., 2010).

1.2 Need of the Study

During vermifiltration, the earthworms play the critical role in wastewater purification. Hence, the earthworm’s number, population density (biomass) in soil, maturity and their health are important factors that affect the quality of treated wastewater. The population in a typical vermifilter may range from several hundreds to thousands (Sinha et al., 2008). However, the study on different earthworm stocking densities (Number or weight) is still lacking in understanding the effect of changes in treatment efficacy with the change in earthworm population. Earthworms have also been shown to bioaccumulate high concentrations of toxic chemicals including heavy metals and also the ‘endocrine disrupting chemicals’ (EDCs) from sewage without affecting their physiology and in particular when the metals are mostly non-bioavailable (Singh et al., 2013; Rajagopal et al., 2013). EDC are compounds that alter the normal functioning of the endocrine system of both wild life and humans. Numerous pesticides (organochlorides, organophosphates, carbamates) have been developed and used extensively worldwide and are rich source of EDC. However, ample literature is available in which various researchers have evidenced its treatment (Lambert et al., 1997; Kumar and Philip, 2006; Kubsad et al., 2005).

Various researchers have investigated the potential of vermitechnology for onsite wastewater treatment including mix and combination of different media for evaluating the potential impacts of physical characteristics on the overall vermifilter performance. In this direction, vermifiltration have been tried with different kinds of material as a media; like ceramsite, quartz sand and converter slag–coal cinder etc. Xing et al. (2011) studied the effect on earthworm's health and change in final characteristics of treated sludge from vermifiltration process using ceramsite and quartz sand material. In their investigation ceramsite material was found to be relatively better media for the process with better characteristics of sewage sludge and health of earthworms. Wang et al. (2010) studied the applicability of converted slag–coal cinder in vermifiltration process. They observed the removal of nutrients like ammonia nitrogen, phosphorous and organic contaminants in the treated wastewater. Despite recent advancement in the last few years, vermifiltration process still is in its infancy and needs a better media that could bridge different issues like pollutant removal, nutrient behavior and earthworm biomass (Health of earthworms).

The hydraulic loading rate also plays an important role on the quality of effluent. Higher hydraulic loading rate reduces the quality of treated water as the earthworms and microorganisms do not get sufficient time for digestion of organic matter present in wastewater (Li et al., 2009). Hence it is also one of the prime factors during the treatment of wastewater by vermifiltration. Most of the researchers have not studied in detail the effect of different hydraulic loading rate on vermifiltration (Kumar et al., 2014).

Generally, biological treatment systems are effective in the lower concentration range and relatively cheaper as compared to other traditional technologies (Ravi et al., 2013). But, the treatment systems should be able to deal with the changes in the composition of wastewater and the daily fluctuations in quality. Reactor stability against shock loads is an another important design characteristic as wastewater treatment plants are designed based on average flow rates and average biochemical oxygen demand loadings, with little or no recognition of peak conditions. The stability assessment is necessary to provide valuable information for the evaluation of plant performance and to offer a guide to process design (Tandukar et al., 2006; Seetha et al., 2010). There are no such studies available in literature on the effect of shock loads on vermifiltration process. So an attempt had been made with vermifiltration process to address such type of issues. Beside these factors, generally for representation of the removal of organic matter by earthworms, the kinetic model based on first order reaction is being used (Stein et al., 2006). The first order reaction model predicts an exponential decay of the concentration which attains zero value asymptotically. Physical and biological mechanisms are lumped into a single reaction coefficient (K), and effluent concentration is considered as a function of the product of reaction coefficient and hydraulic retention time. First-order kinetics is considered to be the basic model for organic matter removal in wetlands, being widely employed in USA, Australia and European countries (Mitchell and McNevin, 2001). In the present study the performances of zero order and second order kinetics were also investigated.

During vermifiltration, earthworms increase the hydraulic conductivity of filter media and natural aeration by granulating the clay particles (Sinha et al., 2008). But as such no data are available that could prove this statement in literature regarding this parameter. Hence an attempt has been made by comparing the vermifiltration process against geofilter (control reactor).

1.3 Objectives of the Present Study

The present study is being taken up by keeping in mind the identified gaps as discussed above. The specific objectives of the present investigation are as follows;

1. Effect of stocking density during vermifiltration process and its optimization.
2. Assess the appropriateness of various filter media for the vermi treatment of wastewater.
3. Performance evaluation of vermifilter at different hydraulic loading rates.
4. A comparative study on the hydraulic aspects of the vermifilter and geofilter.
5. Design of vermifilter for the treatment of domestic wastewater.
6. Evaluation of the concentration of nutrient and pathogen variation during the process and the final effluent.
7. Effect of organic shock load during vermifiltration process.
8. Comparison of different kinetic models of vermifiltration process.

1.4 Thesis Organization

The system proposed in this research is a low cost ecological wastewater unit i.e. vermifilter for decentralized communities. The reader had a brief overview of the important consideration before selecting optimum conditions and type of materials required for these kinds of systems. Following the intensive literature review and design of vermifilter unit, system was assessed under different operational conditions and stresses which can occur if applied for actual field applications. Along the performance analysis, a comparative account is also made between vermifilter and geofilter. The material presented here also allows readers to design an appropriate vermifilter using locally available materials.

A brief layout of the thesis is discussed below:

Chapter 1 presents the introduction and objectives of the present study. In the second Chapter a comprehensive literature has been carried out that gives a tentative idea about the present study. In the third chapter a detailed description about methodology and experimental plan is given with an ease to understand the description of the study that how the data was collected/ measured and finally how the analysis was done. Chapter 4 presents a detailed analysis of the effect of stocking density (Earthworm population) on number or weight basis on the performance of vermifilter. In Chapter 5, the effect of different filter media for the vermi treatment of wastewater has been covered. Chapter 6 presents a detailed study on the effect of hydraulic loading rates on the performance of vermifilter. In addition to this, a comparative study on the hydraulic aspects of vermifilter and geofilter is also given. In Chapter 7, performance of designed vermifilter at optimum conditions is covered in which the designed vermifilter was evaluated against organic shock load. In addition to this, a kinetic study for designed vermifilter unit is also accomplished. Chapter 8 presents the important conclusions drawn from the study.

LITERATURE REVIEW

2.1 General

Nowadays rural and urban areas demand decentralized wastewater treatment to solve the increasing water crises and overburden on exhausting natural resources. At present, the decentralized wastewater treatment has become a need for rural and urban sewage treatment. Directly or indirectly it may be beneficial for rural environment. The conventional treatment technologies for wastewater are costly and not energy efficient. On the other side, the infrastructural development in the building sector at fast rate makes the existing wastewater treatment facilities inadequate to cater to the growing needs. Sewage treatment facilities have to be linked with the colonization, so as to provide the onsite wastewater treatment facilities. At present, there is an abundance of ecological and decentralized wastewater treatment technologies exist such as constructed wetland (Babatunde et al., 2008; Zhang et al., 2009), stabilization ponds (Garcia et al., 2000; Heubeck et al., 2007), and land treatment (Li et al., 2005). These technologies have good treatment efficiencies, but they are restrictively applied on wastewater treatment due to their large occupied area. The treatment systems that require relatively low costs, energy, and maintenance are preferable for the treatment of wastewater. Therefore, energy-saving decentralized wastewater treatment technologies should be given importance with feasibility for utilization like sewage farming or recreation purposes. Vermifiltration is one of the practical alternatives for wastewater treatment and it is the part of vermitechnology (Kumar et al., 2014). In this Chapter, the efficiency of vermifiltration for wastewater treatment is studied in detail and the following section presents a comprehensive literature review on vermifiltration.

2.2 Wastewater Treatment Technologies

Rapid growth in population, urbanization, industrialization and demand of energy has drawn attention of many researchers towards the scarcity of water. Globally, billions of people are suffering due to inappropriate sanitation and unavailability of useable water. Due to lack of appropriate wastewater treatment and disposal systems in small communities, communities are suffering from inadequate hygienic efficiency and horrendous environmental losses also. Since 1970s World Bank is also making efforts to develop economic and environment friendly treatment methodologies for small communities in developing countries. Also these decentralized plants consume less energy as compared to large plants (Orth, 2007; Ødegaard et al., 1997; Watanabe et al., 1997). That is why in developing countries like India, wastewater engineers are showing serious concern for developing alternative wastewater treatment strategies for small communities than larger communities. Today various technologies have been successfully implemented for the treatment of wastewater generated in small communities. However, very few literature is available regarding the full scale application of these technologies (Ho, 2005; Orth, 2007; Boller, 1997). In India, many of the environmental engineers believe that decentralized wastewater treatment systems can be a permanent solution for the wastewater management in rural and urban areas as well as at isolated locations (Singh et al., 2014). India already has a considerable experience with such decentralized systems and currently hundreds of decentralized wastewater treatment plants are in working conditions based on different technologies. However, most of them are not functioning efficiently. In addition to this, currently there is no consolidated literature is available on the performance of existing decentralized wastewater treatment plants. Now a days, wastewater treatment using vermitechnology has been effectively applied for small communities. A brief description of vermitechnology is outlined below.

2.3 Vermitechnology

Vermitechnology is made up of two components, the vermiculture and vermicomposting and is a simple natural process of degradation of waste, in which certain species of earthworms are used to enhance the process of waste conversion and produce a better end product (Nayak et al., 2013; Sonowal, 2014). Status report on solid waste management by vermicomposting in India prepared by Vasudevan et al., (2001) indicated that the vermicomposting is a suitable technology for solid organic waste management for both rural and urban communities. Santra and Bhowmik, (2001) found that vermicomposting is attaining a special significance for the abatement of pollution hazards created by large amount of organic wastes in India and also reduces the demand for chemical fertilizers. Dominguez et al., (2000) studied the effect of different residual bulking agent (paper, card board, grass clippings, saw dust) in mixture with sewage sludge (1:1) on the growth and reproduction of *E. andrei* and concluded that maximum weight and highest growth were attained in the mixture with food waste. Thimmaiah, (2001) utilized *E. fetida* species of earthworms for vermicomposting of vegetable waste of a town by earthworm *E. eugeniae* and found that the final compost promoted seed germination in Phaseolus radiates (Green gram) and dolichos biflorus (horse gram) crops to a great extent. Werner and Cuevas,

(1996) reported that Cuba has more than 170 vermicomposting centers that are engaged in producing earthworm casting for the use of organic fertilizers in tobacco farming.

Vermicomposting involves the combined action of earthworms and microorganisms considerably improving the decomposition and stabilization of sludge. During vermicomposting process, earthworms act as mechanical blenders and by comminuting the organic matter and modify its physical and chemical status, gradually reducing its C:N ratio, increasing the surface area exposed to microorganisms and making it much more favorable for microbial activity and further decomposition (Dominguez et al., 2000; Ghatnekar et al., 2010).

2.4 Role of Vermifiltration in Decentralized Wastewater Treatment

Vermifiltration is the process in which earthworms and microorganisms simultaneously work to treat the waste available in wastewater. Vermifilter (Lumbrifiltration) was first advocated by the late Professor Jose Toha at the University of Chile in 1992 (Bouché and Qiu, 1998; Aguilera, 2003; Li et al., 2008), which is a low-cost sustainable technology over conventional systems with immense potential for decentralization in rural areas (Taylor et al., 2003; Sinha et al., 2008). It was firstly used to process organically polluted water using earthworms (Li et al., 2008). Introduction of earthworm was a considerable innovation to conventional biofilter of wastewater treatment and it had created a new method of biological reaction through extending food chains, conserving energy and transferring mass from the biofilm to the earthworm. Vermifiltration require relatively low costs, energy, and maintenance as compared to other conventional treatment technologies.

2.5 Vermifiltration

Fig. 2.1 shows the schematic diagram of a laboratory scale vermifilter. It comprise mainly of storage tank, peristaltic pump, wastewater distributer and the vermifilter unit. Storage tank are used to collect the required amount of wastewater to treat. For regulating the discharge rate a peristaltic pump are provided which pumps the wastewater at certain flow. Wastewater distributer trickles the wastewater and allows it uniformly on the surface of vermifilter bed. The vermifilter bed is the layer in which earthworms are present. The earthworms have tendency to ingest the waste on top side. So, mostly this part is responsible for treatment. Vermifiltration unit is composed of different types of layer, in which first layer, from top, is called vermifilter bed and rest of the layer comprised of media having different sizes. The bottom most layer is taken as supporting layer. The top most layer (vermifilter bed) acts as a wastewater treatment unit. It composed of some detritus material or cotton soil.

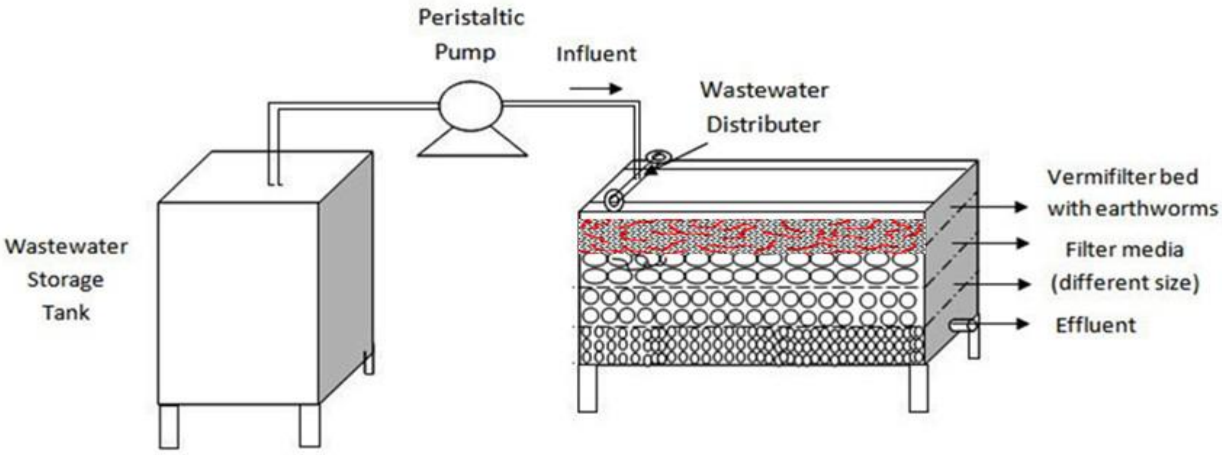

Fig. 2.1: Schematic diagram of a laboratory scale vermifilter

2.5.1 Factors Affecting Vermifiltration

Three important factors that need attention in vermifiltration are;

- (i) Earthworm population (Stocking Density)
- (ii) Hydraulic Loading Rate (HLR)
- (iii) Hydraulic Retention Time (HRT)

2.5.2 Earthworm Population

Earthworm participation enhances natural biodegradation and decomposition of organic waste. As the worms double their population every 60-70 days, the process becomes faster with time (Sinha et al., 2010). The higher population of earthworms gives a better quality of effluent. In literature it has been investigated that most of the earthworms consume half of their body weight of organic matter in the waste in a day. *Eisenia fetida* can consume organic matter at the rate equal to their body weight every day (Visvanathan et al., 2005).

2.5.3 Hydraulic Loading Rate (HLR)

The HLR is the rate of flow of wastewater per unit surface area of vermifilter bed. The HLR plays an important role in regulation of quality of effluent. Higher hydraulic loading rate reduce the quality of treated water as the earthworms and microorganisms do not get sufficient time for digestion of organic matter present in wastewater (Li et al., 2009). So, it is one of the prime factors for the treatment of wastewater during vermifiltration process.

HLR is usually computed as

$$HLR = \frac{V_{wastewater}}{A \times t} \tag{2.1}$$

Where

HLR = Hydraulic loading rate
$V_{wastewater}$ = Volume of wastewater
A = Surface area of vermifilter bed
t = Time

2.5.4 *Hydraulic Retention Time (HRT)*

While treating wastewater containing predominant solids or going for combined treatment of both solid and liquid waste, hydraulic retention time (HRT) should also be given due importance. If hydraulic retention time is more, the earthworms and microorganisms get sufficient time to digest the organic matter available in wastewater and from solid waste.

HRT is usually computed as

$$HRT = \frac{\phi V}{Q} \tag{2.2}$$

Where

HRT = Hydraulic retention time
ϕ = Porosity
V= Volume of vermifilter bed
Q = Flow rate

2.6 Other Factors

The other factors which affect the efficiency of vermifiltration are following as:

(i) *pH of wastewater* (ii) *Toxicity of ammonia/ammonium* (iii) *High Salt Concentration* (iv) *Selected Media*

2.6.1 *pH of Wastewater*

The pH of influent often varies considerably with household product use. The ingredients in many of the products contain caustic soda and alkaline salts, which can raise pH levels above 11 (Patterson, 2001). It creates certain kind of toxicity in vermifiltration units as the earthworm species involved in the system are very sensitive to environmental changes (Sinha et al., 2012). In addition pH of wastewater plays an important role for the proper growth of earthworm and microorganisms. Edwards, (1988) found that *Eisenia fetida* and *Eisenia andrei* dependent on pH levels between pH 5 and 9. Various studies have been carried out to assess the likelihood of biological inhibition and disruption from pH to earthworm species in vermifiltration and the system has showed the evidence of inbuilt pH buffering capacity (Hughes et al., 2007; Sinha et al., 2008). Hughes et al., (2007) observed that vermicompost and manure media has relatively high buffering capacity for pH. In their study it was found that the earthworm species can survive between pH level of 6.2 and 9.7 and at higher pH levels the survival of juveniles is impaired due to their inability of regulating the soluble salt uptake system.

2.6.2 Toxicity of Ammonia/ammonium

Wastewater ammonia has three principal sources i.e. household cleaners and disinfectants, food wastes and urine. The toxicity of ammonia from domestic influent to vermifiltration systems is largely unknown but may pose problems to domestic systems. The key species in vermifiltration systems, the worm species *Eisenia fetida*, is susceptible to ammonia levels present in manure (poultry) around 500 mg/kg (Edwards and Arancon, 2004; Hughes et al., 2008). This species is responsible for converting the organic waste in the vermifiltration process into a suitable matrix filter; hence, if toxic shock occurs the filter may clog due to the formation of an impermeable sludge barrier. Hughes et al., (2008) studied the assessment of the toxicity of ammonia/ammonium on mortality of worms within the vermifiltration process. In results vermifiltration system found very low toxicity to ammonium with ammonium chloride having an ammonium of 1.49 g/kg while ammonium sulfate did not show an effect on mortality at 2 g/kg ammonium. However, the system caused some mortality to the worms when it was subjected to ammonia hydroxide which somewhat increases the pH.

2.6.3 High Salt Concentration

High use of sodium in household products led to a high concentration in household wastewater and in the wastewater stream, soluble sodium is likely present as sodium chloride (NaCl) (Patterson, 2001). The worms present in a vermifiltration wastewater treatment system are likely to be sensitive to high concentrations of sodium salts, as they are terrestrial organisms adapted to relatively low salt concentrations (Volkov et al., 2003). Hughes et al., (2009) studied the assessment of toxicological risks from sodium accumulation in a vermifiltration process. In their study it was found that sodium chloride

(NaCl) is the more toxic component than the other common sodium salts found in wastewater to the worms. In the study, it has been investigated that NaCl is more toxic than Na_2SO_4 to the worms. The most susceptible life stage to NaCl was juvenile production. Edwards and Arancon, (2004) has also evidenced same kind of experience. Volkov et al., (2003) studied that many worm species accumulate chloride and other salts to stabilize internal and external osmotic pressures. It is unlikely that sulfate or carbonate would be accumulated to the same degree as chloride, as they are less soluble than chloride. The mean numbers of cocoons is more sensitive endpoint when compared to adult survival against salts concentration.

2.6.4 Selected Media

In vermifiltration process choice of media plays an important role which governs the treatment efficiency because it is directly related with earthworm's activity. It has been noted that in the conventional filters, particle size ratio (d_{60}/d_{10}) directly influences the microbial activity and flow rates of the filter. However, filter media plays an extraordinary role in the vermifiltration since changes of the exotic environment play a crucial role on the structure and function of the earthworm's body wall, which is closely relative with earthworm's activity and respiratory metabolism (Xing et al., 2011). Various researchers have tried to work with different kinds of material as a media for individual wastewater and sludge treatment and their combined treatment (synchronous treatment) also. Yet Ceramsite, quartz sand and converter slag–coal cinder are the media that has been used by various researchers. Each media has their individual objectives. For example Xing et al., (2010) was concerned about the health effect on earthworms and change in final characteristics of treated resulting sludge from vermifiltration process. During this study ceramsite and quartz sand material were used and ceramsite material was found better suitable media relatively for the process with better characteristics of sewage sludge. The health of earthworm was found much better than quartz sand due to less abrasion on earthworm's body wall with ceramsite. In another study, conducted by Wang et al., (2010), converter slag–coal cinder was found most suitable. The study was concerned with removal of nutrients like ammonia nitrogen and phosphorous and organic contaminants also.

2.7 Summarized Experience with Vermifiltration Process

Hughes et al., (2007) studied the effect of pH in vermifiltration process. The likelihood of biological inhibition and disruption against different levels of pH in vermifiltration process were evaluated and in their results, during the study, the vermifiltration process found an inbuilt buffering capacity for pH. During the study it was investigated that the earthworm species can survive at pH levels between 6.2 and 9.7.

Sinha et al., (2007) investigated the removal of high BOD and COD of primary liquid waste products from dairy industry through vermifiltration process. A considerable amount of total dissolved solids (TDS) and total suspended solids (TSS) removal were also evidenced in their study.

Li et al., (2008) reported a 30% increase in earthworm population in 4 weeks with 50% reduction in ammonia emission during vermifiltration of diluted swine manure. Higher water, carbon, and total nitrogen gaseous losses were also observed when the vermifiltration was compared with conventional breeding on a slatted floor. In various regions with intensive livestock production, excessive releases of liquid and solid manure lead to water, soil, and air pollution. In their study vermifiltration was found as a pollution transfer free process with higher efficacy of treatment of diluted swine manure in an integrated manner.

Vermfiltration indicated the synchronous sewage and sludge treatment also. During a study, Jian et al., (2008) experienced a decentralized and on-site option for rural settlements wastewater with the adoption of vermifiltration system. Simultaneously, the excellent reduction and stabilization of the sludge produced were also obtained during the study period. Same kind of results has been experienced by Sinha et al., (2008) in study of sewage treatment by vermifiltration with synchronous treatment of sludge by earthworms. In their study it is also mentioned that earthworms increase the hydraulic conductivity and natural aeration by granulating the clay particles. Earthworms grind the silt and sand particles which results an increase in total specific surface area and it enhances the ability to ‘adsorb’ the organic and inorganic compounds from the wastewater. In vermifiltration the suspended solids are trapped on the top layer and processed by earthworms which fed to the soil microbes immobilized in the vermifilter.

Hughes et al., (2008) studied the assessment of vermifiltration process against the toxicity of ammonia/ammonium to key earthworm species *Eisenia fetida*. During the study a solid phase tests showed relatively low toxicity to ammonium with ammonium chloride having LC50 for ammonium of 1.49 g/kg. Ammonium sulfate did not show an effect on mortality at 2 g/kg ammonium. The full-scale tests indicated that ammonia based cleaning agents increase the pH and ammonia concentration in wastewater when it interacted with ammonium hydroxide. Nevertheless, the effects on the system were quite minimal with no significant difference in terms of worm survival found between treatments.

Hughes et al., (2009) studied the risk of sodium toxicity from bed accumulation to key species in vermifiltration process. The study was undertaken for the assessment of toxicological risks from sodium accumulation during the process. It was observed that earthworm have an ability to detoxify the NaCl concentration. However, the reproduction was impaired when they exposed with moderate concentration of NaCl for a long period of time.

Khwairakpam and Bhargava, (2009) studied vermitechnology for sewage sludge recycling. Three different earthworm species *Eisenia fetida*, *Eudrilus eugeniae* and *Perionyx excavatus* in individual and combinations were utilized to compare the suitability of worm species for composting of sewage sludge as well as the quality of the end product. It was observed that the sewage sludge without blending can be directly converted into good quality fertilizer in stabilized form. The study also inferred that the application of sewage sludge in the agricultural fields after vermicomposting does not have any adverse effect as the heavy metals (Cu, Mn, Pb and Zn) observed to be within the permissible limits.

Liu et al., (2009) studied the application of ceramsite media based vermifilter for the treatment of domestic wastewater. During the study, vermifilter exhibited the efficient removal of basic physico-chemical parameter like COD, NH_4^+-N, suspended solids. Besides this, vermifiltration system showed the presence of biofilm, zoogloea, rotifer and vorticella when a microscopic observation was carried out during the study period.

Li et al., (2009) studied the vermifiltration process at field scale level and compared its performance with conventional activated sludge process. During the study vermifiltration process was found to have same efficiency as activated sludge process. An interrelationship between HRT, HLR and solid content were also observed during study. The study emphasized on increasing efficiency with lesser hydraulic loading rate which increased the quality of treated water as the earthworms and microorganisms gets sufficient time for retrieval of organic matter available in wastewater. If the wastewater contains sufficient amount of solid content then hydraulic retention time (HRT) should be given more importance.

The study conducted by Hendrickx et al., (2009) investigated the treatment of partial digested waste sludge through vermifiltration process using aquatic worm (*Lumbriculus* *variegatus*). The study was concerned with sludge reduction efficiency and it was correlated with various phenomenon like dissolved oxygen concentration, ammonia toxicity, temperature and light exposure. Dissolved oxygen concentration affects both consumption rate and digestion efficiency of sludge. During the study period higher sludge consumption rate was observed when DO concentration was kept around 8.1 mg/L with a TSS reduction of 36%. A four time lower sludge consumption rate was observed when DO concentration decreased to 2.1 mg/L. However during this, the TSS concentration in effluent resulted in 70% reduction. Besides, lower sludge consumption rate was also observed through worms at higher ammonia concentration in water. The study evidenced that an unionised ammonia is emitted in water compartment which creates certain kind of toxicity through earthworms. This effect reflects a pH dependency on availability of various forms of nitrogen in the system. The rate of ammonia nitrogen emission was 0.02 mg N per mg of TSS with the consumption of sludge through earthworms. An optimum sludge consumption rate was observed when the process was correlated with another phenomenon that is temperature. In the study, significant effect was found with change in temperature and sludge consumption through earthworms including sludge digestion efficiency. An optimum temperature around 15°C was found most suitable for better consumption of sludge and its digestion. Sludge digestion efficiency got reduced when the temperature gradually increased from 10 to 20°C.The temperature below 25°C ensured for earthworm survival. In study no light effect were observed in the system whether the process is running in light or dark. Various researchers have investigated the temperature effect on the performance wastewater treatment systems (Sundaresan and Philip, 2008; Yang et al., 2009).

Wang et al., (2010) investigated the domestic wastewater treatment performance through vermifilter enhancement by a converter slag-coal cinder filter. A significant removal of total chemical oxygen demand (TCOD), BOD, ammonia nitrogen (NH_4^+-N) and phosphorus were observed. During the study, it was also observed that vermifiltration process is effective for insoluble organic matter and suspended solid removal. The converter slag-coal cinder filter played an important role in phosphorus removal.

Xing et al., (2010) investigated the vermifiltration process having media of quartz sands and ceramsite for the treatment of domestic wastewater. Vermifiltration exhibited better performance achievement with greater removal of basic physico-chemical parameters like BOD, COD, suspended solids and total nitrogen. In addition vermifiltration process was evaluated against varying rate of hydraulic loading rate. It was observed that an increase in hydraulic loading rate led the process to a decrease in treatment efficiency and adult earthworm abundance. The activities of protease, alkaline phosphatase (ALP), and cellulase in earthworm body diminished with increase in hydraulic loading rate. Besides, superoxide dismutase (SOD) and catalase (CAT) increased. It was also found that larger earthworms play an important role as compared to smaller ones. The process exhibited the presence of various enzymes that are responsible for the treatment.

Zhao et al., (2010) studied the performance of a conventional biofilter (BF) and a vermifilter containing the earthworm, *Eisenia fetida*. Vermifilter (VF) for the treatment of domestic wastewater sludge was compared with the earthworm–microorganism interaction mechanisms involved in sludge stabilization. The results revealed that the presence of earthworms in the VF led to significant stabilization of the sludge by enhancing the reduction in volatile suspended solids (VSS). The study indicated that earthworms in the VF were capable of transforming insoluble organic materials to a soluble form and then selectively digesting the sludge particles of 10–200 mm to finer particles of 0–2 mm, which led to the further degradation of organic materials through microorganisms.

Xing et al., (2011) explored the suitability of filter media in vermifiltration process by comparing the process in reference of sludge yield, characteristics of by-product and abrasions on earthworm’s body wall. During the study two different types of material as a media like ceramsite and quartz sand were evaluated. Ceramsite was found relatively better as compared to quartz sand with lesser production of sludge yield. In ceramsite media the final by-product was more digested as compared to quartz sand as the earthworm activity was more due to smoother surface of ceramsite with less abrasion on earthworm’s body wall.

Study conducted by Luth et al., (2011) focused on emissions of NH_3, N_2O, CH_4 and CO_2 during vermifiltration of pig slurry. There was a decrease in emissions of ammonia and nitrous oxide and a sink of methane in treatment with earthworms during the process. Therefore, in vermifiltration earthworm abundance can be used as a bioindicator to certify low energy input, and low greenhouse gas and ammonia output.

Wang et al., (2011) studied the enhancement of rural domestic sewage treatment performance, assessment of microbial community diversity and structure using tower vermifiltration. During the study a composite system of tower vermifilter planted with *Penstemon campanulatus* was evaluated. Tower vermifiltration results an increasing order of dissolved oxygen in effluent which is responsible in reduction of ammonium and phosphorus concentration in effluent due to anaerobic and aerobic circumstances. Adsorption and precipitation for filter materials during the longer running distance, increasing nitrification ability between each stage and the functioning of Al^-, Fe^- and Ca-oxides also plays an important role for the reduction of ammonium and phosphorus concentration in effluent. In addition the presence of earthworms intensified the bacterial diversity in soils. The changes in microbial community diversity in different paddings and stages likely contributed to the introduction of earthworms, media properties and nutrient variations.

Wang et al., (2011) studied the mechanisms and performance of a microbial-earthworm ecofilter for removing organic matter and nitrogen from synthetic domestic wastewater. The working model was a combination of soil with sawdust possessed at higher porosity and specific surface area. Microporous structure together with wormcast provided in the system results in efficient removal of COD and NH_3-N. It was evidenced that the nitrogen variation depends on various properties like soil properties, earthworm's activities and wormcast characteristics. The system was found very efficient when depth wise study was carried out. A positive correlation was observed between Shannon biodiversity index for ammonia oxidizing bacteria (AOB) and decreasing NH_3-N concentration which indicates the dominancy of soil microbes. Soil microbes play an important role in removing NH_3-N and nitrogen conversion. During the study it was observed that some physical, chemical, and biological processes and synergistic effects of earthworms and microorganisms, including the adsorption of small particle organisms, colloid organisms and organic molecules, as well as the oxidation-reduction of organic matter and activity of earthworms are helpful for COD reduction. Earthworm layer with added sawdust provide higher porosity and surface area which is beneficial for removing most of the organic contaminants by precipitation and adsorption in the voids of the soil. During the process micrographs of wormcast, within the system, exhibits micro-pores on surface and abundant cylindrical organic matter acts as a filtering medium for the removal of organic matter available in the influent. The system comprised the largest component of the N-species when it was studied depth wise. It was observed that the majority of nitrogen present as NH_3-N in wastewater was removed mainly through soil and sand parts of the reactor due to rapid adsorption by the biomass and filters. The ratio of N_{org}/TN decreased slightly with soil depth, although the majority of N in the form of N_{org} was observed in wormcast during the study. Earthworm activity was found to be a major determinant of N_{org} in soil, as they promote nitrogen mineralization. The increased NH_3-N in the top soil may be due to adsorption or absorption of NH_4^+ ions from influent on to the mineral or organic fraction of the soil, as ammonium ions are relatively inert to cation exchange. Earthworm activity and the production of wormcast oxygenate the influent and this oxygenation facilitates the nitrification of ammonia by microbes. The addition of sawdust plays an important role as it acts as a biosorbent through complexation with NH_4^+ ions. Earthworms mediate the conversion of organic nitrogen to inorganic nitrogen and finally nitrification promotes the formation of nitrate and resulted into increased concentration of NO_3-N with depth. Besides, soil particles have the capacity to retain pollutants and capture NO_3-N from wastewater. Increased NO_3-N concentration with depth represents downward transport with outflow.

An another phenomenon was also evaluated during the study period in which effect of ammonia oxidizing bacteria (AOB) and nitrospira community structure and bacterial diversity were observed. The results revealed that soil is the most suitable substrate for nitrifying bacterial communities in the system and earthworm activities had no significant effect on the community structure of nitrobacteria at different soil depths. Some groups like indigenous ammonium oxidizers attach more strongly to clay particles. Earthworms maintain the balance of nitrifiers with the help of aerobic and anaerobic microflora in their guts and their burrowing action improve aerobic conditions in the soil body, thereby creating a favored microenvironment for aerobic nitrobacteria.

Yin et al., (2011) studied the effect of filter bed temperature on municipal wastewater treatment efficiency during vermifiltration process. The results indicated that the relation of filter bed temperature and organics removal follows a binomial distribution and that on removal of NH_4^+-N is in accordance with the Sigmoidal equation. No obvious effect was observed on the removal of TN and TP with change in temperature. The study indicated that the vermifilter having quartz sand media gives better performance at filter bed temperature in the range of 15-24°C while for media having ceramisite it was observed as 18-25°C. In addition ceramisite grain was found with certain kind of buffering capacity with the variations of the filter bed temperature. The buffering capacity has better resistance to the adverse impact of vermifiltration under lower and higher temperature conditions than that of the quartz sand. In addition ceramisite grain based vermifilter were observed more resistant to shocking temperature conditions.

Tomer and Suthar, (2011) demonstrated the urban wastewater treatment using vermi-biofiltration system. A small-scale vermi-biofiltration reactor was constructed using vertical subsurface-flow constructed wetlands (VSFCWs). Local earthworm species *Perionyx sansibaricus* was used in the system. The coco-grass: Cyprus rotundus at density 0.14 plants/in.^{2} was used to construct VSFCW. Vermi-biofiltration was found more efficient in removing TSS, TDS, COD, NO_3^-, PO_4^{3-} when it was compared with control system (devoid of earthworms).

From literature it is clear that the vermifiltration is a suitable technique for highly efficient treatment of wastewater. This is a good alternative for decentralized onsite wastewater treatment. On the basis of literature review, it is observed that vermifiltration is still at its infancy and an appropriate design methodology is yet to be conceived. Further, the following gaps are identified:

1. Effect of stocking density during vermifiltration process and its optimization.
2. Assess the appropriateness of various filter media for the vermi treatment of wastewater.
3. Performance evaluation of vermifilter at different hydraulic loading rates.
4. A comparative study on the hydraulic aspects of the vermifilter and geofilter.
5. Design of vermifilter for the treatment of domestic wastewater.
6. Evaluation of the concentration of nutrient and pathogen variation during the process and the final effluent.
7. Effect of organic shock load during vermifiltration process.
8. Comparison of different kinetic models of vermifiltration process.

The present study is undertaken to address these issues. The experimental set-up, description of experimental runs and the analysis of results are explained in detail in later chapters.

METHODOLOGY AND EXPERIMENTAL PLAN

3.1 General

The present chapter discusses the experimental plan adopted for conducting the experiments on vermifiltration. These experiments involve studying the effect of earthworm population, different media, hydraulic loading rate and organic shock load on the performance of vermifilter. The experimental procedure, scheme of reactor operations and related methodology during different phases are described here in order to accomplish the stipulated objectives.

3.2 Experimental Design

In order to accomplish the stipulated objectives mentioned in Chapter 1, the study was carried out in four different phases. A flow chart of the experimental plan is shown in Fig. 3.1. The experiments are conducted in a vermifilter built in Environmental Engineering Laboratory of Indian Institute of Technology Roorkee, Roorkee (India).

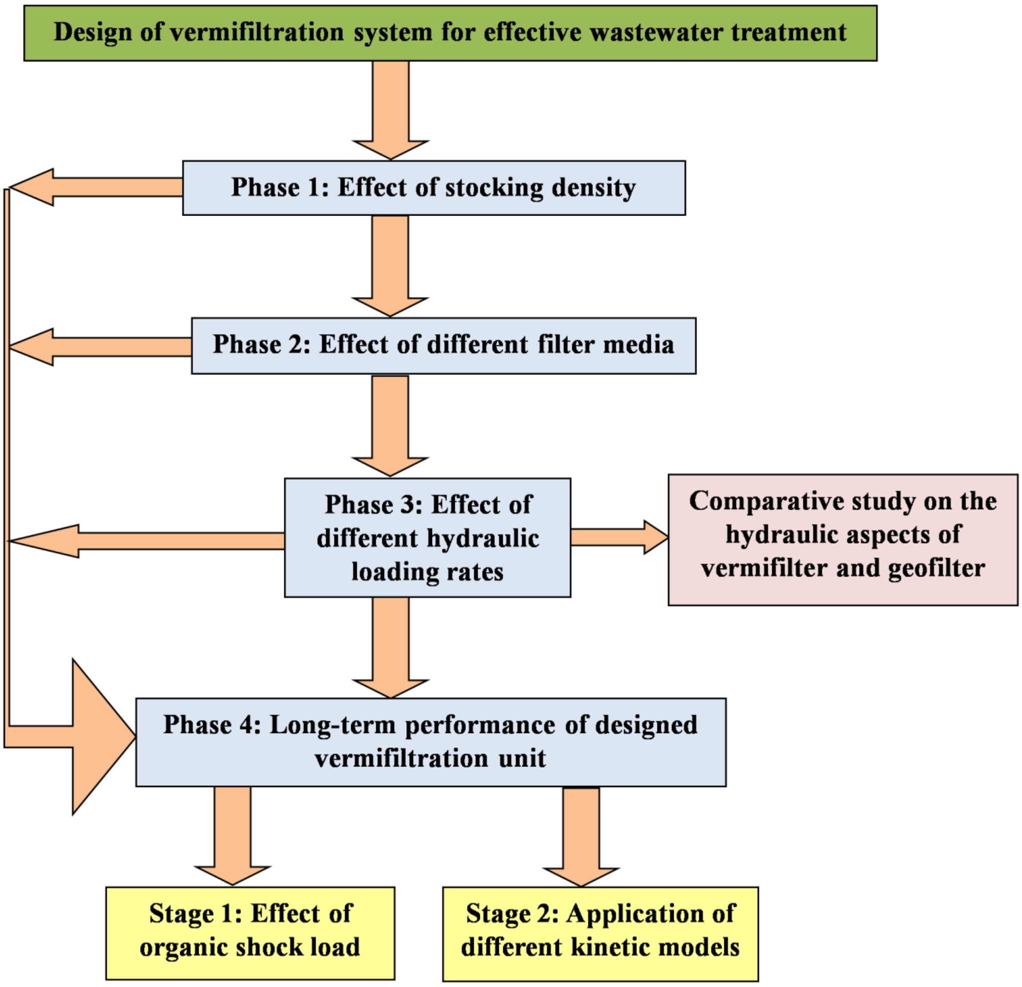

Fig. 3.1: Experimental design of the research work

3.3 Description of Vermifilter Reactor

Fig. 3.2 shows the schematic diagram of a laboratory scale vermifilter. It comprise mainly of storage tank, peristaltic pump, wastewater distributer and the vermifilter unit. Storage tank are used to collect the required amount of wastewater to treat. For regulating the discharge rate a peristaltic pump are provided which pumps the wastewater at certain flow. Wastewater distributer trickle the wastewater and allow it with uniform distribution on the surface of vermifilter bed. The vermifilter bed is the layer in which earthworms are present. The earthworms have tendency to ingest the waste on top side. So, mostly this part is responsible for treatment. Vermifiltration unit is composed of different types of layer, in which first layer, from top, is called vermifilter bed and rest of the layer comprised of media having different sizes. The bottom most layers are taken as supporting layer. The top most layer (vermifilter bed) acts as a wastewater treatment unit. It composed of some detritus material or cotton soil.

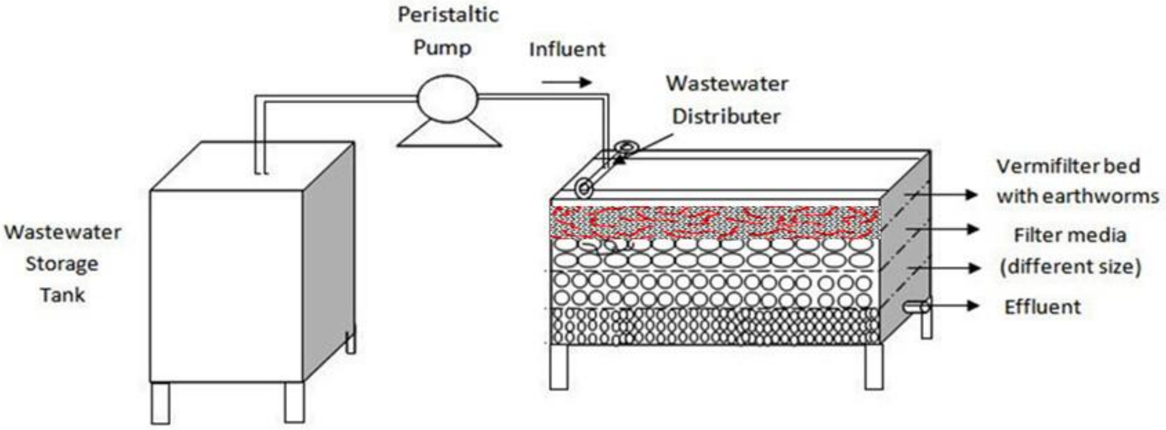

Fig. 3.2: Schematic diagram of a laboratory scale vermifilter

3.4 Wastewater Preparation

The wastewater was prepared in the laboratory by dissolving molasses, urea and KH_2PO_4 to give the ratio of COD/N/P as 300/30/1 (Seetha et al., 2010; Thalla et al., 2012) such that it simulates actual domestic wastewater of medium strength (Tchobanoglous et al., 2003). The influent had COD 480 ± 21 mg/L, BOD 330 ± 15 mg/L, BOD/COD ratio 0.69 ± 0.02, sBOD 296 ± 19 mg/L, sCOD 419 ± 25 mg/L, dissolved oxygen (DO) 3.2 ± 1.1 mg/L, total solids (TS) 817 ± 151 mg/L, TSS 230 ± 50 mg/L, NH_4^+-N 48.5 ± 11.4 mg/L, TP 5.2 ± 1.6 mg/L and pH of 7.3 ± 0.06. For initializing the growth of microbial population synthetic sewage was seeded with 1% domestic wastewater to mimic the actual sewage and prepared in 100 L bucket as shown in Fig. 3.3.

Fig. 3.3: Synthetic wastewater preparation

The microbial quality of the synthetic domestic wastewater was quite consistent with an average concentration of total coliform (TC), fecal coliform (FC), *fecal streptococci* (FS) and *Escherichia coli* (*E. coli)*, observed as $3.16 \times 10^5 \pm 1.58 \times 10^4$, $7.94 \times 10^4 \pm 1.26 \times 10^3$, $7.94 \times 10^3 \pm 1.58 \times 10^3$ MPN/100 mL and $1.26 \times 10^4 \pm 6.31 \times 10^3$ CFU/100mL respectively, throughout the study period. All the parameters were analysed according to the Standard Methods for the examination of water and wastewater (APHA, 2005).

3.5 Experimental Protocol and Reactor Setup

Based on the experimental design as illustrated in Fig. 3.1, the study was carried out in four different phases namely (i) Effect of stocking density, (ii) Effect of media used in vermifilter bed, (iii) Effect of hydraulic loading rate, (iv) Effect of organic shock loads on vermifiltration. The experimental setup, influent wastewater characteristics and all the phases is explained in detail in the following sections.

3.5.1 Phase-I: Effect of Stocking Density during Vermifiltration Process

This section discusses the effect of stocking density on the performance of vermifilter. The reactor consisted of plastic container having cross sectional dimension of 250 mm x 200 mm and a depth of 300 mm. The top layer consisted of 100 mm thick matured vermicompost (worm-bed). Second, third and fourth layers were of material of size 6 to 8 mm (50 mm thick), 2 to 4 mm (50 mm thick) and 10 to 12.5 mm (50 mm thick), respectively. The pictorial view of vermifilter is illustrated in Fig. 3.4.

To collect the wastewater a 350 liter storage tank was kept at certain height from the reactors that acted as an overhead tank. In addition to this, for maintaining hydraulic loading rate at constant level a 30 liter constant head tank was employed. For uniform distribution of wastewater a perforated plates with 1.5 mm diameter hole fixed at the top of the reactor for uniform distribution of wastewater over the reactor. It acted as a distributer on the top side of vermifilter bed for uniform distribution.

Fig. 3.4: Lab scale vermifilter having different earthworm stocking densities

Optimum stocking density was identified on the basis of quality of effluent and final weight of augmented earthworm biomass.

The lab-scale study was concerned with the following aspects:

- Performance evaluation of different vermifilters having different stocking densities based on the quality of effluent in terms of BOD and COD removal.
- Removal efficiency of microbiological indicators i.e. total coliform (TC), fecal coliform (FC), *fecal streptococci* (FS), *Escherichia coli* (*E. coli*) in vermifilter having different earthworm stocking density.
- Identification of relatively better stocking density.

In the first phase, the vermifilter bed consisted of river bed material and the active layer vermifiltration unit was inoculated with different number of earthworms with mature vermicompost (worm gratings). Different earthworm stocking densities namely VF1, VF2, VF3, VF4, VF5 and VF6 with corresponding stocking densities such as 5000, 10000, 15000, 20000, 25000 and 30000 worms/cum of vermifilter bed, respectively were used in vermifiltration process. The detailed analysis related with effect of stocking density on the performance of vermifilter is explained in Chapter 4.

3.5.2 Phase-II: Effect of Filter Media on the Performance of Vermifilter

This section discusses the evaluation of vermifiltration process using different materials as filter media by taking into account earthworm biomass and quality of effluent, so that a better media can be used as an alternative option according to local availability. For this purpose four different types of materials; river bed material (VFR), wood coal (VFC), glass ball (VFG) and mud balls (VFM) with vermicompost were used.

The lab scale study carried out on different vermifilters was concerned with the following aspects:

- Identification of relatively better media.
- Performance evaluation of different vermifilters operated using different filter media based on the quality of effluent in terms of BOD and COD removal.
- Removal efficiency of microbiological indicators i.e. total coliform (TC), fecal coliform (FC), *fecal streptococci* (FS), *Escherichia coli* (*E. coli*) in vermifilter having different filter media.

In vermifilter, only second layer from top was changed with different media (river bed material, Wood coal, glass balls and mud ball) in all reactors which act as a vermifilter bed and plays a major role during treatment process. The complete arrangement of reactor was taken same as discussed in Section 3.5.1. The wastewater was applied from the top of the reactor with the help of a peristaltic pump. Each of vermifilter was inoculated with 150 earthworms (*Eiseinia fetida*) based on the optimum stocking density as discussed in Section 3.5.1. A constant hydraulic loading rate of wastewater, around 1.5 $m^3m^{-2}d^{-1}$, was maintained in all reactors during study period.

For evaluating the effect of filter media on the efficiency of vermifilter, four different types of material were used as a vermifilter bed material i.e. river bed material, Wood coal, glass balls and mud ball as shown in Fig. 3.5 (a) to 3.5 (d).

Fig. 3.5 (a): River bed material

Fig. 3.5 (b): Wood coal material

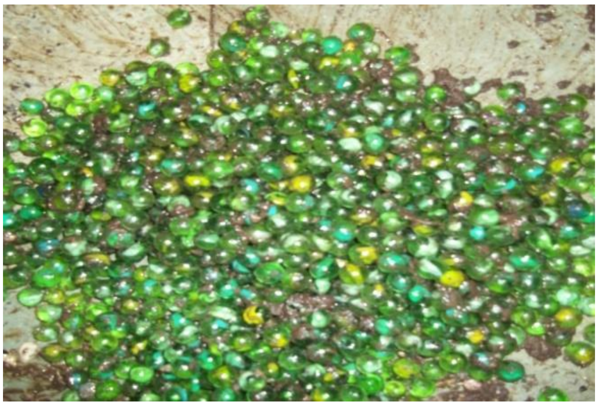

Fig. 3.5 (c): Glass balls media

Fig. 3.5 (d): Mud balls media

The specification and arrangement of different layer is presented in Table 3.1.

Table 3.1: Description of the filter bed layers for vermifilter

Layers from top	Description of layers	Description of Material in vermifilter	Size of material	Depth (mm)
Layer 1	Active layer	Matured vermicompost	600-800 µm	100
Layer 2	Active layer	Media (River bed material/ Wood coal/ Glass balls/ Mud balls)	6-8 mm	50
Layer 3	Third layer	Sand	2-4 mm	50
Layer 4	Supporting layer	Large gravel	10-12 mm	50

The porosity of different media is given in Table 3.2. The wastewater was applied from the top side of reactors under gravity.

Table 3.2: Porosity of different media used in study

S. No.	Media	Size (mm)	Porosity (%)
1	River bed material	6-8	35
2	Wood coal	6-8	45
3	Glass balls	6-8	40
4	Mud balls	6-8	43

The physical properties (specific surface area and porosity) of media also play an important role that affects the treatment performance of the reactor (Young and Dahab, 1983; Tay and Show, 1998). Media with relatively higher physical properties i.e. higher surface area or low porosity facilitate to greater biomass accumulation and attains higher treatment efficiency as compared to the performance of media with higher porosity or low specific surface area. The detailed analysis related with effect of filter media on the performance of vermifilter is explained in Chapter 5.

3.5.3 Phase-III: Performance Evaluation of Vermifilter at Different Hydraulic Loading Rates (HLRs)

The present section discusses the effect of hydraulic loading rates (HLRs) on the performance of vermifilter.

The lab scale study was concerned with the following aspects:

- Performance evaluation of different vermifilters operated at different HLRs based on the quality of effluent in terms of BOD and COD removal.
- Identification of relatively better HLR.
- Comparative Studies on the hydraulic aspects of the vermifilter and geofilter.

The vermifilter was run at five different hydraulic loading rates using river bed material. Each of the vermifilter was inoculated with 150 earthworms (*Eiseinia fetida*) based on the optimum stocking density as evidenced in Section 3.5.1. The reactors were run with wastewater at different hydraulic loading rates to find out the optimum rate. In addition, the performance of vermifilter is also compared with a geofilter (devoid of earthworm; CRR). Both vermifilter and geofilter were fed daily at five different HLR of 0.5, 1.0, 1.5, 2.0 and 2.5 $m^3m^{-2}d^{-1}$ respectively for 90 days.

To investigate the hydraulic aspects of the vermifilter the hydraulic conductivity pattern were measured at different HLRs and a comparison was made between the performance of vermifilter and geofilter.

- *Measurement of hydraulic conductivity (K)*

The measurement of hydraulic conductivity was carried out using constant head permeability test according to ASTM D2434 (ASTM 2006). A typical set-up of constant head permeameter is shown in Fig. 3.6. It is based on the measurement of the quantity of water that flows under a given hydraulic gradient through a sample of known length and cross-sectional area in a given time. During measurement the water was allowed to flow through the initial and final vermicompost sample from a reservoir designed to keep the water level constant by overflow. The flow of water was allowed to continue till steady states of flow get established, as evidenced by constant level in the manometer tube. Then the discharge was measured.

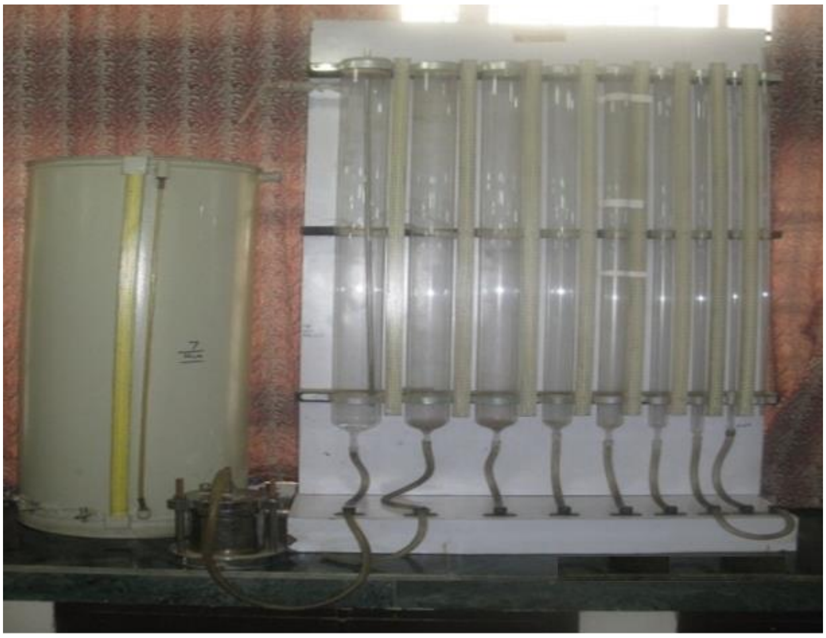

Fig. 3.6: Constant head permeameter

After this the value of K was determined using the equation

$$q = K.i.A \tag{3.1}$$

$$K = \frac{q}{iA} = \frac{QL}{Aht} cm/\sec \tag{3.2}$$

Where

K = Coefficient of permeability (cm/sec)

q = Discharge (cm^3/sec)

Q = Volume of water collected (cm^3) in time t (sec)

A = Cross-sectional area of sample (cm^2)

h = Difference in manometer level (cm)

L = Distance between manometer taping points (cm)

The detailed analysis related with effect of hydraulic loading rates on the performance of vermifilter is explained in Chapter 6.

3.5.4 Phase-IV: Performance Evaluation of Designed Vermifilter for Effective Wastewater Treatment

To study the effect of stocking density, type of filter media and HLR on the performance of vermifilter a methodology is presented for the design of vermifilter for domestic wastewater treatment. This design methodology involved the following aspects:

a. Comparative study of vermifilter (VFR) and geofilter (Control reactor; CRR).

b. Variation in concentration of nutrients and pathogens during the process and the effluent.
c. Effect of organic shock load during vermifiltration process.
d. Kinetics in vermifiltration process.

A pilot scale vermifilter reactor made up of perspex sheet, was set up as shown in Fig. 3.7.

Fig. 3.7: Pilot scale vermifilter and geofilter

It consisted of filter bed material, wastewater storage tank, wastewater distribution system and effluent collection system. The reactor was 800 mm long and 800 mm wide with a depth of 800 mm, and 650 mm of packed bedding of river bed materials. An empty space or free board of around 150mm is kept at the top for aeration purpose.

Table 3.3: Description of the filter bed layers for vermifilter

Layers from top	Description of layers	Description of Material in vermifilter	Size of material	Depth (mm)
Layer 1	Active layer	Matured vermicompost	600-800 µm	200
Layer 2	Active layer	River bed material	6-8 mm	150
Layer 3	Third layer	Sand	2-4 mm	150
Layer 4	Supporting layer	Large gravel	10-12 mm	150
Empty space	-	-	-	150

The reactor had a partition wall that divides the reactor it in two parts, one part acted as a vermifilter reactor and other was used as geofilter reactor. Hence, each reactor had a dimension of 800 mm long and 400 mm wide with a depth of 800 mm. The vermifilter consisted of 4 layers. The description of filter bed layers is illustrated in Table 3.3. Vermifilter was inoculated with 800 numbers (weighed about 450 g) of earthworm species *E. fetida* based on optimum stocking density and hydraulic loading rate as evidenced in Section 3.5.1 & 3.5.3. A centrifugal pump was installed to collect and transfer the wastewater to the overhead tank. Besides, a constant head tank was also provided that had a provision to maintain constant flow under gravity so that unnecessary energy cost can be reduced. Overall, the wastewater fed from storage tank to constant head tank and finally to treatment unit and it passed through different layers in sequence by gravity flow. Prior to the experimental analysis a 20 day acclimatization period were provided to the reactors. The reactor was started up in June 2013 and operated for a period of 90 days. During study the atmospheric temperature varied in the range of 32-38 ºC.

- *Effect of Organic Shock Load*

The system was continuously fed with synthetic wastewater at hydraulic loading rate of 1.3 $m^3m^{-2}d^{-1}$. It corresponded to a hydraulic retention time (HRT) of 3 h till the system achieved steady-state condition.

Pseudo steady-state condition (PSS) was assumed to be achieved when the variation in effluent COD concentration was found to be insignificant. After this, the effect of organic shock load on the performance and stability of the system was evaluated to identify its feasibility in actual onsite conditions. For this, a transient organic shock load conditions were created by varying the influent concentration (COD_{Inf}) in the range 675-1410 mg/L at constant HRT over a period of time until the reactor had reached steady state. This comes with influent COD concentration from 1.5 to 3 times of normal feed concentration. Each shock load was applied for a period of 3 h (3 h HRT). Normal loading conditions were resumed at the end of shock load.

Substrate loading rates: The substrate loading rates were determined by following equation

$$L = HLR \times C_{sub} / (1000 mg/L \times 1 m^3/kg) \tag{3.3}$$

Where, L is the substrate loading rates ($kg/m^2/d$), HLR the hydraulic loading rate ($m^3/m^2/d$) and C_{sub} is the substrate concentration (mg COD/L). The detailed analysis related with the performance of designed vermifilter is explained in Chapter 7 which deals with effect of organic shock load also.

3.6 Analysis and Measurements

The samples taken out during experimentation process were analysed and the detailed analysis (for both solid and liquid sample) was performed in accordance to the following section.

3.6.1 Analysis of Liquid Samples

All liquid samples were centrifuged and filtered through 0.45 µm filter for the analysis of pH, EC, TP, NH_4-N, NO_3-N, BOD and COD and measured according to Standard Methods for the examination of water and wastewater (APHA, 2005). COD digester (Model AL 38SC, Aqualytic, USA) and UV-Vis spectrophotometer (Model DR 5000 Hach, USA) were used for COD analysis. All soluble contents were determined by filtering the samples through 0.45-micron Whatman filter.

Total nitrogen (TN) was measured as sum of all forms of nitrogen (subject to presence) in a sample as N, where total Kjeldahl nitrogen (TKN) is the sum of NH_4^+-N and organic-N, and organic-N was measured as the difference of TKN and NH_4^+-N.

$$TN = NH_4^+\text{-N} + NO_3^-\text{-N} + \text{Organic-N} \tag{3.4}$$

The concentration of TC, FC and FS were enumerated by Multiple Tube Fermentation Technique (APHA, 2005) while *E. coli* was enumerated by serial dilutions of the sample plated on MacConkey Agar medium and incubated in an inverted position for 24-48 h at 37°C.

3.6.2 Analysis of Solid Samples

The solid samples collected free of earthworms, hatchlings and cocoons were referred to as wet samples and the samples after the oven dried, grounded and sieved were referred to as dry samples. Each dry sample was analyzed in triplicates for the following parameters.

- *pH and electrical conductivity (EC)*

5 g of the sub-sample was mixed well in 50 mL distilled water and pH was measured using a digital pH meter with a glass electrode, preiously calibrated and corrected for temperature (BIS, 1982). The above mixture was filtered by Whatman filter paper No. 42 and EC was measured using a conductivity meter.

- *Total Organic Carbon (TOC) and Ash Content*

About 0.25 g of sub-sample was used for determination of TOC by Shimadzu (TOC-V_{CSN}) Solid Sample Module (SSM-5000A). Ash content was measured by the ignition method (550ºC for 2 h in muffle furnace) (BIS, 1982).

- *Total nitrogen (TN), Ammonia nitrogen (NH_4-N) and Nitrate-nitrogen (NO_3-N)*

100 mL sample was prepared for analyzing TN using Kjeldahl method and HACH TKN kit by acid digestion of 0.5 g of sub-samples in 4 mL of conc. H_2SO_4 in the presence of 10-15 mL of 30% H_2O_2. KCl (30 mL of 2 M KCl) extraction of 3 g of each sub-sample was used for analysis of NH_4^+-N and NO_3-N using Standard methods (Tiquia and Tam, 2000; APHA, 2005).

3.6.3 Enumeration of Earthworm Biomass

After the completion of vermifiltration process earthworm biomass was measured. The earthworms were separated from the compost by light separation method. In this method the unsorted finished compost from vermifilter bed was placed under a light bulb for few minutes. The earthworms moved downwards, away from the light towards the bottom. When the entire earthworms move away at the bottom, the upper compost, free of earthworms was separated and later on sorted the cocoons, were sorted out from it by hand sorting method using magnifying glass if required (Fig. 3.8).

Fig. 3.8: Enumeration of Earthworm Biomass

3.7 Instruments and Equipment Used

All the instruments and equipment required for measurements of various physico-chemical and microbial parameters are detailed in Tables 3.4 to 3.6.

Table 3.4: Instruments used for measurement of various physico-chemical parameters

Parameters	Instruments/ Equipment	Model/Manufacturer/ Specifications
pH	Digital pH meter	Toshniwal Instrument Manufacturing Pvt. Ltd., India
EC	Conductivity meter	Hach Model CDC 401
Turbidity	Nephelometer	HACH 2100 AN Turbidimeter
COD	COD digester	Model AL 38SC, Aqualytic, Germany
BOD	BOD incubator	Digital TempCon DTC-201
TOC	TOC analyzer	TOC-$V_{CSN,}$ Shimadzu, Solid sample module, SSM-5000A
Solids	Oven , Muffle furnace	NSW, India
NH_4^+-N, NO_3-N	UV-VIS Spectrophotometer	DR/4000, HACH, USA

Table 3.5: Instruments used for measurement of various microbial parameters

Parameters	Instruments/ Equipment	Model/Manufacturer/ Specifications
Coliform (Total and Fecal coliform)	Incubators and Laminar flow supply hood	TempStar
Fecal Streptococci	Test tubes and Petriplates	Borosil, India
E. coli	Test tubes and Petriplates	Borosil, India

Table 3.6: Other instruments used for different processes

Instruments/ Equipment	Purpose	Model/Manufacturer/ Specifications
Peristaltic pump	Feeding to reactors	PP20EX, Miclins, Chennai, India
Micro Pipettes	Volume measurements	Qualigens Ltd
Weighing Balance	Weighing	MettlerTolido, AG 285
Centrifuge	Centrifuge	Research Centrifuge, REMI, India
Glassware	Analysis and storage	Borosil, India

EFFECT OF EARTHWORM'S STOCKING DENSITY ON VERMIFILTRATION

4.1 General

Earthworms play an important role in wastewater purification in vermifiltration process. Earlier, few studies on vermifiltration have been conducted in treating and stabilizing municipal as well as industrial (paper mill, dairy and textile industry) effluent and sludge which showed a perfect efficacy of treatment with high removal rates of COD, BOD and SS (Sinha et al., 2008; Kumar et al., 2014; Rajpal et al., 2014). These studies have been conducted employing different earthworm species such as Tiger Worm *(Eisenia fetida),* Red Tiger Worm *(E. andrei),* Indian Blue Worm *(Perionyx excavatus),* African Night Crawler *(Eudrilus euginae)* and the Red Worm *(Lumbricus rubellus)* for vermi treatment of organic waste (Tomar and Suthar, 2011). As earthworms play a critical role in wastewater treatment and purification, their stocking density (numbers) and population density (biomass) are important factors that affect the vermifiltration. The earthworm population may range from several hundreds to few thousands (Sinha et al., 2008). However, the study on earthworm's stocking density (Number or weight of earthworm per unit volume of vermifilter bed) on vermifiltration is still lacking in understanding the effect of changes in treatment efficacy with the change in earthworm population. Thus, the present Chapter is aimed on following aspects;

(i) To evaluate the performance of vermifiltration process at different stocking densities of earthworms to optimize it.
(ii) Removal efficiency of microbiological indicators i.e. total coliform (TC), fecal coliform (FC), *fecal streptococci* (FS), *Escherichia coli* (*E. coli*) in vermifilter having different earthworm stocking densities.
(iii) To establish a design criterion in vermifiltration process through which, one can decide the stocking density requirement with organic load to be applied on vermifilter for giving a better quality of the effluent.

For this purpose, a pilot scale vermifilter (VF) made of plastic container was set up and consisting of five parts: filter bed, filter material, earthworms, water distributor and drain system (as discussed in Section 3.5.1). The reactor consisted of plastic container having cross sectional dimension of 250 mm x 200 mm and a depth of 300 mm. The top layer consisted of 100 mm thick matured vermicompost (worm-bed). Second, third and fourth layers were of material of size 6 to 8 mm (50 mm thick), 2 to 4 mm (50 mm thick) and 10 to 12.5 mm (50 mm thick), respectively. Different sets of these reactors VF1, VF2, VF3, VF4, VF5 and VF6 were used with different stocking densities such as 5000, 10000, 15000, 20000, 25000 and 30000 worms/cum of vermifilter bed (as discussed in Section 3.5.1) and were allowed to acclimatize for about three week before the study. With high reproductive capability and good applicability, *Eisenia fetida* was chosen for use in the vermifilter (Yang et al., 2008). The reactors were started up in January 2012 and operated for a period of 90 days. During study the atmospheric temperature varied in the range of 24-29ºC.

4.2 Results and Discussion

The reactors VF1 to VF5 were operated simultaneously for 90 days duration. The characteristics of the influent wastewater are given in Section 3.4. The removal of BOD, COD, TSS, TC, FC, FS and *E. coli* from each of the vermifilter is measured and the detailed analysis is performed in the following section.

4.2.1 BOD Removal

The residual BOD in all reactors VF1, VF2, VF3, VF4, VF5 and VF6 having different earthworm stocking densities of 5000, 10000, 15000, 20000, 25000 and 30000 is presented in Table 4.1. During vermifiltration process a considerable amount of BOD removal was observed as compared to influent BOD concentration. From Table 4.1 it is clear that the reactor VF2 resulted in relatively higher removal as compared to the other reactors throughout the study period. In reactor VF1 the average BOD concentration was found as 32 mg/L, while the other reactors i.e. VF1, VF3, VF4, VF5 and VF6 the average concentration of BOD represented as 79, 48, 70, 59 and 36 mg/L.

Table 4.1: Comparison of BOD removal at different earthworm stocking densities

S. No.	Time (Days)	Influent (mg/L)	Effluent BOD concentration (mg/L)					
			VF1	VF2	VF3	VF4	VF5	VF6
1	1	350	76	18	121	68	64	20
2	7	346	86	50	90	68	36	42
3	13	341	152	30	48	144	60	56
4	19	336	92	40	40	84	32	56
5	25	331	50	28	26	52	34	44
6	31	323	68	38	56	42	30	64
7	37	321	46	24	42	60	64	28
8	43	317	44	26	29	38	54	12
9	49	314	122	30	46	36	58	18
10	55	311	190	40	32	114	42	29
11	61	307	74	34	28	190	90	32
12	67	339	50	30	58	66	44	38
13	73	334	50	32	40	42	44	36
14	79	329	66	29	35	22	84	34
15	85	319	60	35	35	58	120	33
16	90	300	30	29	38	42	92	31
Average		326	79	32	48	70	59	36

The average BOD removal from all the reactors is shown in Fig. 4.1. From Fig. 4.1 it can be concluded that the average removal of BOD, in all reactors, varied between 75-90% and the maximum of 90% removal was observed in reactor VF2, having a stocking density of 10000 W/cum. This could be due to the higher population of earthworm which plays an important role in degradation of organic matter available in wastewater. In reactor VF1 the BOD removal was 75%. The BOD removal efficiency in reactor VF3, VF4, VF5 and VF6 was observed by 85, 78, 81 and 89%, respectively.

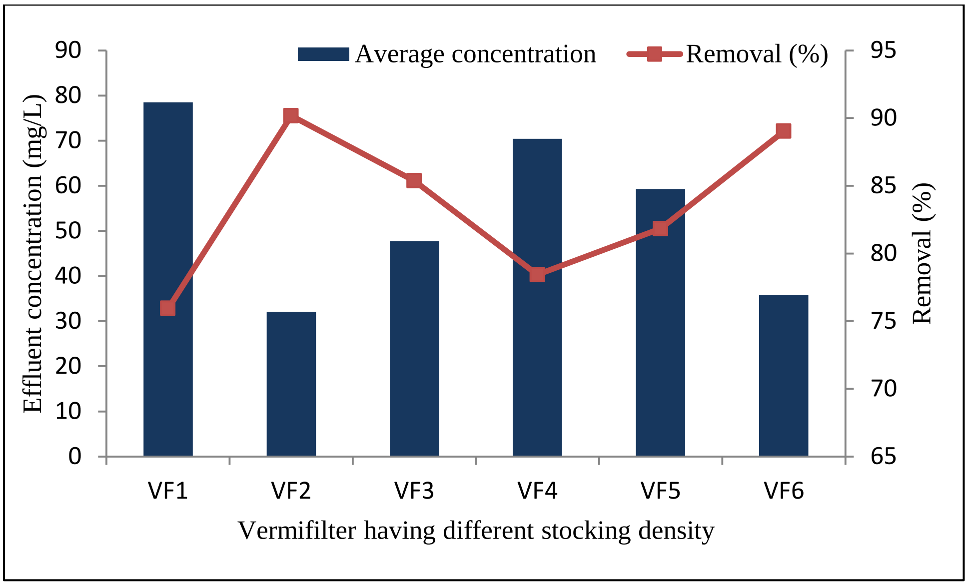

Fig. 4.1: Variation of average residual BOD at different earthworm stocking densities

The reduction in removal efficiency at higher stocking density attributed to the slowing down in earthworm activity due to scarcity of food which is also evident by less increase in earthworm biomass (as shown in Table 4.5) at higher stocking density at the end of the run.

4.2.2 COD Removal

Table 4.2 illustrates the COD in all reactors having different earthworm stocking densities. From Table 4.2 it is clear that the COD in all vermifilters having different earthworm stocking densities was observed to be significantly low as compared to influent through out of study period. The reactors VF1 and VF2 represented almost equal and relatively higher COD removal as compared to the other reactors throughout the study period. In reactor VF1 the average COD concentration was found as 145 mg/L, while in other reactor VF1 experienced as 147 mg/L. In other reactors i.e. VF3, VF4, VF5 and VF6 the average concentration of COD represented as 158, 165, 174 and 165 mg/L, respectively.

Table 4.2: Residual COD at different earthworm stocking densities

S. No.	Time (Days)	Influent (mg/L)	Effluent COD concentration (mg/L)					
			VF1	VF2	VF3	VF4	VF5	VF6
1	1	553	247.63	166.01	227.12	207.63	194.76	140.76
2	7	465.47	135.84	154.53	219.37	130.18	159.49	183.35
3	13	430.83	178.42	126.87	174.06	218.48	183.28	181.03
4	19	441.7	147.92	135.34	148.99	128.42	139.02	131.52
5	25	441.08	146.15	101.23	147.23	127.92	141.41	185
6	31	483.4	132.61	147.99	152.74	132.61	132.27	183.74
7	37	410.34	126.26	157.99	148.07	166.26	194.5	178.74
8	43	480.96	106.39	163.51	132.48	110.39	166.78	150.29
9	49	493.29	158.48	128.18	173	186.15	180.38	193.46
10	55	443.34	240.18	155.72	134.88	155.84	161.36	163.65
11	61	450.74	129.62	144.6	137.21	199.62	200.34	143.9
12	67	548.55	119.36	155.8	161.75	179.36	142.5	172.4
13	73	402.1	135.14	135.4	143.67	105.14	172.64	149.67
14	79	455	129.56	147.8	155	199.56	196.1	155.82
15	85	504.91	102.78	161.9	147	182.78	224.78	186.56
16	90	407	116.1	130.7	128.92	211.1	191.05	133.56
Average		463	147	145	158	165	174	165

Fig. 4.2 shows the comparison of average COD removal at different earthworm stocking densities. Overall, the average COD removal in all reactors varied in the range of 62-68%. In reactors VF1 and VF2 almost equal and maximum removal of COD was observed as 68%.

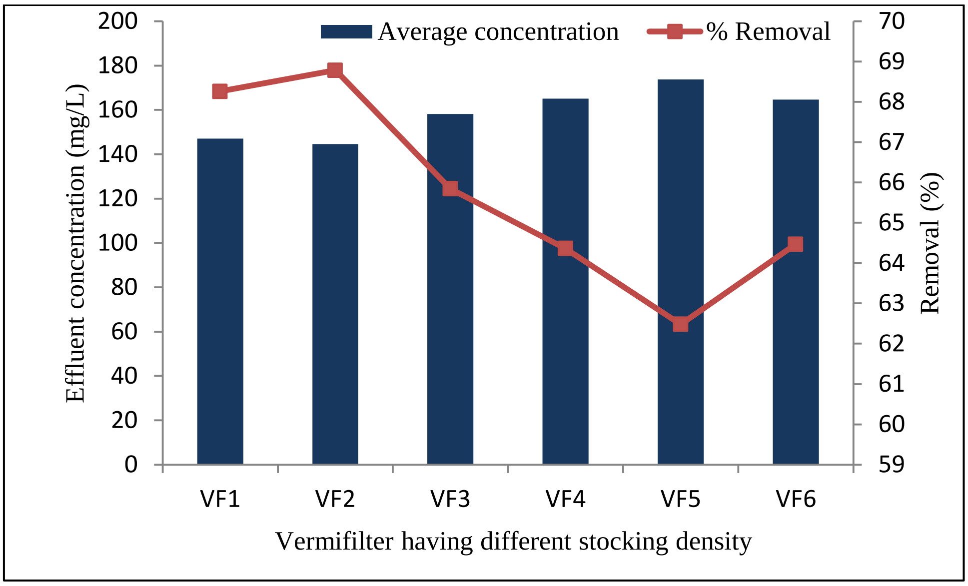

Fig. 4.2: Variation of COD at different earthworm stocking densities

The reactor VF2 represents maximum COD removal as 68% while the reactor VF5 represented minimum COD removal efficiency which is observed as 62%. This decline in COD as compared to influent attributed to the ingestion, grinding and digestion of organic matter with the help of aerobic and anaerobic microflora in the gut of earthworms. The synergistic effects of earthworms and microorganisms, including adsorption of small particle organisms, colloid organisms and organic molecules as well as the oxidation-reduction of organic matter and the activity of earthworms makes vermifilter efficient for removing BOD and COD. In reactors VF3, VF4 and VF6 no significant change in COD removal was observed as the removal efficiency was almost equal. The removal of BOD was more as compared to COD as the earthworms are more efficient to digest the biodegradable material which is used as an energy source by earthworms and microorganisms and converting into CO_2 (Rajpal et al., 2011; Kumar et al., 2014). The other researchers have also evidenced the same kind of scenario (Sinha et al., 2008; Kumar et al., 2015).

4.2.3 TSS Removal at Different Stocking Densities

Table 4.3 presents the TSS concentration pattern in all the reactors having different earthworm stocking densities with time. The TSS concentration in all vermifilters having different earthworm stocking densities was observed to be significantly low as compared to influent through out of study period. In reactor VF2 higher removal of TSS concentration was observed and the average concentration was observed as 72 mg/L. In other reactors, an increasing pattern was observed as compared to VF2.

Table 4.3: TSS concentration at different earthworm stocking densities

S. No.	Time (Days)	Influent (mg/L)	Effluent TSS concentration (mg/L)					
			VF1	VF2	VF3	VF4	VF5	VF6
1	1	236	146	127	161	142	136	137
2	7	227	124	110	141	110	72	112
3	13	230	111	55	110	131	113	114
4	19	243	85	59	103	118	75	72
5	25	220	79	71	122	135	117	133
6	31	219	148	51	89	127	137	56
7	37	238	50	78	135	138	113	104
8	43	298	46	96	157	91	94	91
9	49	294	106	101	112	96	109	76
10	55	271	116	62	92	117	91	98
11	61	254	129	47	131	111	144	73
12	67	231	108	85	121	99	55	102
13	73	242	59	46	82	81	108	85
14	79	222	143	57	102	64	121	95
15	85	217	133	37	92	78	145	47
16	90	238	102	64	104	68	126	139
Average		243	105	72	116	107	110	96

The average TSS concentration in all the reactors i.e. VF1, VF2, VF3, VF4, VF5 and VF6 is presented in Fig. 4.3. From Fig. 4.3 it can be seen that the reactor VF2 exhibited maximum TSS removal efficiency which is observed as 70% while in reactor VF3 represented minimum removal efficiency and represented as 52%.

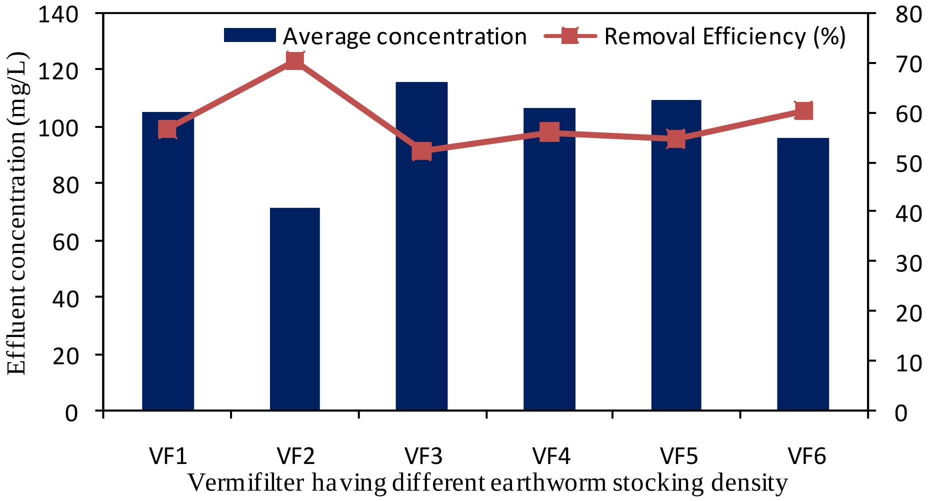

Fig. 4.3: Variation of TSS at different earthworm stocking densities

This could be due to the trapping of suspended solids on the top of the vermifilter and processed by earthworms (Komarowski, 2001). Trapped suspended solids further degraded through geo-microbial process. The dissolved and suspended organic and inorganic solids are trapped by adsorption and stabilized through complex biodegradation processes that take place in the vermifilter bed inhabited by earthworm and the aerobic microorganisms. In reactor VF1 an inappropriate quantity of earthworm may responsible for increased TSS concentration. However, the other reactors like VF3, VF4, VF5 and VF6 has represented an increasing pattern which may be due to the lack of earthworm activity at higher stocking densities as less increase in earthworm biomass (as shown in Table 4.5) observed at higher stocking densities resulting into poor performance of vermifiltration process.

From the above results and discussion it is clear that the reactor VF2 having earthworm stocking density of 10000 W/cum is relatively better for the removal of BOD, COD and TSS.

4.3 Removal of Indicator Organisms

Table 4.4 presents the microbial quality of effluent from all the vermifilters having different earthworm stocking densities. In Table 4.4, the removal of TC, FC, FS and *E. coli* in each of the vermifilters i.e. VF1, VF2, VF3, VF4, VF5 and VF6 is shown together. The coliform count can be used as a part of a measure of the overall sanitary quality of soils and water environments. Generally, high coliform counts indicate low sanitary quality. Elevated counts could indicate a potential health hazard in which pathogens or toxins may be present. The average value of TC in the influent concentrations was found to be 3.16×10^5 MPN/ 100mL.

Table 4.4: Microbial quality of effluent from vermifilter having different earthworm stocking densities (Mean ± Standard Deviation)

Parameter	TC (MPN/100mL)		FC (MPN/100mL)		FS (MPN/100mL)		*E. coli* (CFU/100mL)	
	Mean	Removal (log unit)	Mean	Removal (log unit)	Mean	Removal (log unit)	Mean	Removal (log unit)
Influent	$3.16\times10^5\pm1.58\times10^4$	-	$7.94\times10^4\pm1.26\times10^3$	-	$7.94\times10^3\pm1.58\times10^3$	-	$1.26\times10^4\pm6.31\times10^3$	-
VF1	$6.9\times10^3\pm1.16\times10^3$	1.84±0.35	$5.90\times10^3\pm2.32\times10^3$	1.33±0.31	$6.90\times10^2\pm4.76\times10^2$	1.23±0.16	$5.90\times10^2\pm9.48\times10^1$	1.42±0.14
VF2	$2.90\times10^2\pm1.24\times10^2$	3.61±0.90	$1.40\times10^2\pm1.23\times10^2$	3.14±0.67	$1.90\times10^1\pm0.68\times10^1$	2.73±0.37	$9.40\times10^1\pm0.95\times10^1$	2.27±0.28
VF3	$7.94\times10^2\pm1.32\times10^2$	3.12±0.34	$4.80\times10^2\pm2.58\times10^2$	2.75±0.32	$4.32\times10^2\pm1.26\times10^2$	1.95±0.51	$1.95\times10^2\pm1.79\times10^2$	1.32±0.62
VF4	$2.30\times10^3\pm2.13\times10^3$	2.18±0.29	$1.70\times10^3\pm1.89\times10^2$	1.77±0.18	$2.30\times10^2\pm1.31\times10^2$	1.74±0.26	$3.70\times10^2\pm2.83\times10^1$	1.28±0.17
VF5	$2.60\times10^3\pm1.58\times10^2$	2.11±0.41	$2.10\times10^3\pm1.16\times10^3$	1.32±0.22	$2.60\times10^2\pm1.84\times10^2$	1.39±0.41	$3.10\times10^2\pm2.64\times10^2$	1.16±0.11
VF6	$5.10\times10^3\pm0.96\times10^3$	1.83±0.19	$4.20\times10^3\pm2.58\times10^3$	1.19±0.47	$5.10\times10^2\pm3.29\times10^2$	1.29±0.09	$1.20\times10^3\pm4.73\times10^2$	1.11±0.09

Fig. 4.4 shows the average removal of TC at different earthworm stocking densities during vermifiltration process. It was observed that the reactors VF1 and VF6 represented minimum TC reduction and observed as 1.84±0.35 and 1.83±0.19 log reduction, respectively while in rector VF2, 3.61±0.90 log reductions was observed and exhibited relatively maximum reduction. Consequently with the increase in stocking density, the considerable reduction of coliform was observed. This may be due to release of antibiotics by earthworms which are responsible to kill the coliform microorganisms (Sinha et al., 2009). Previous studies have shown that vermicomposting and vermistabilization can reduce pathogens to safe levels (Rodriguez-Canche et al., 2010) through the enzymatic action in intestines of earthworms. In addition to this, the beneficial microorganism associated with the earthworms competes with the pathogenic microorganisms for limited nutrients in the system (Kumar et al., 2015). Results showed that VF2 with 10,000 W/cum are sufficient to bring coliforms to considerably safe levels (log 3) as recommended by WHO, 1989 for irrigation or recreation purposes.

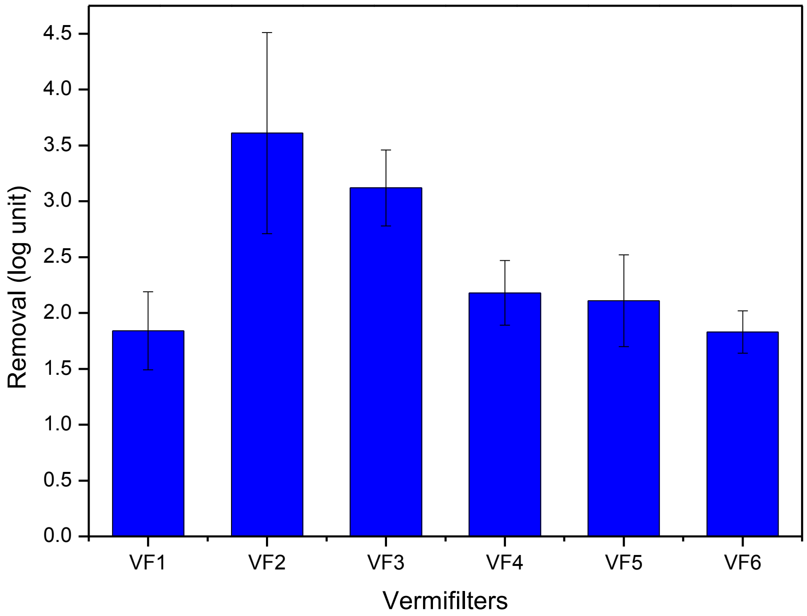

Fig. 4.4: Variation of TC in vermifilter having different earthworm stocking densities

4.3.1 Removal of Fecal coliform (FC) and Fecal Streptococci (FS)

A fecal coliform, common indicator of warm-blooded fecal matter includes many pathogenic forms such as *E. coli*, which indicates the presence of fecal contamination. Use of an indicator such as coliforms, as opposed to the actual disease-causing organisms, is advantageous as the indicators generally occur at higher frequencies than the pathogens and are simpler and safer to detect (Hassen et al., 2001).

The log removal profile for FC with different earthworm stocking densities is shown in Fig. 4.5. The average number of FC was 7.94×10^4 MPN/100mL in the influent. As the number of earthworm increases in different vermifilters, the average log removal initially increases from 1.33 ± 0.31 to 3.14 ± 0.67 log unit, which may be attributed to the antimicrobial activity, associated with the micro-flora inside vermifilter and earthworm enzymatic activity. However at 25,000 and 30,000 stocking densities the log removal profile shows a relatively decreasing trend and found as 1.32 ± 0.22 and 1.19 ± 0.47 log unit, respectively. Overall the reactor VF2 represented maximum FC reduction and observed as 3.14 log unit.

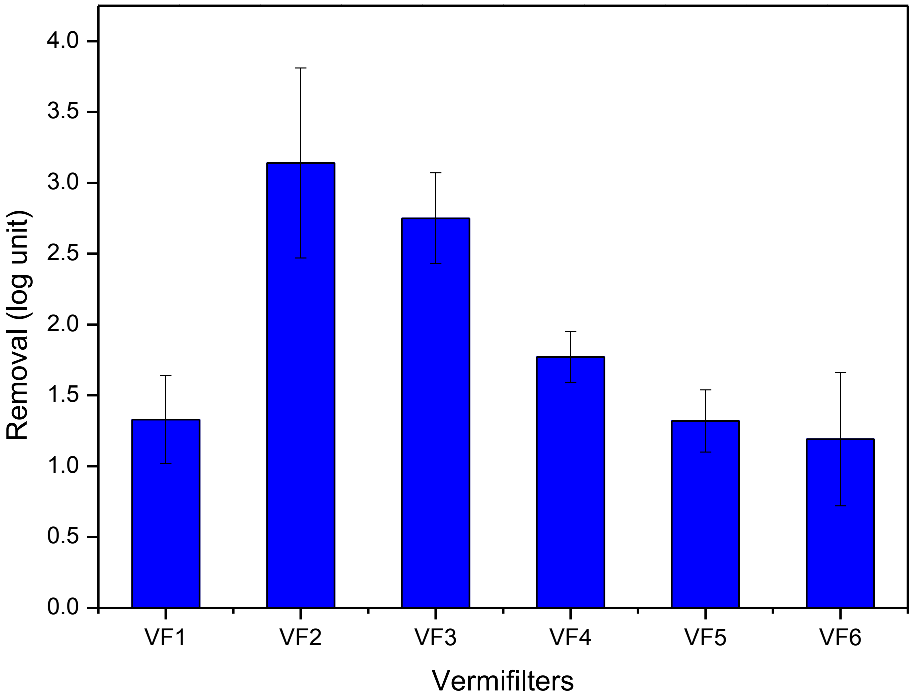

Fig. 4.5: Variation of FC in vermifilter having different earthworm stocking densities

Similar trend was observed for *fecal streptococci* which are commonly considered as the best indicators of fecal pollution. The presence of *fecal streptococci* is evidence of fecal contamination. They are more resistant to different environmental factors than coliforms and are represented by *Streptococcus faecalis, S. faecium and S. bovis.* Fig. 4.6 shows the *fecal streptococci* (FS) removal pattern at different earthworm stocking densities. A considerable reduction of FS, around 2.73 ± 0.37 log units, was observed at 10,000 earthworm stocking density from its initial influent concentration (as shown in Table 4.4). This may be attributed to the slow action of earthworms with microorganisms at higher stocking density due to scarcity of food. This implies that VF2 with 10,000 W/cum are able to bring fecal indicator organisms to levels considered safe for irrigation.

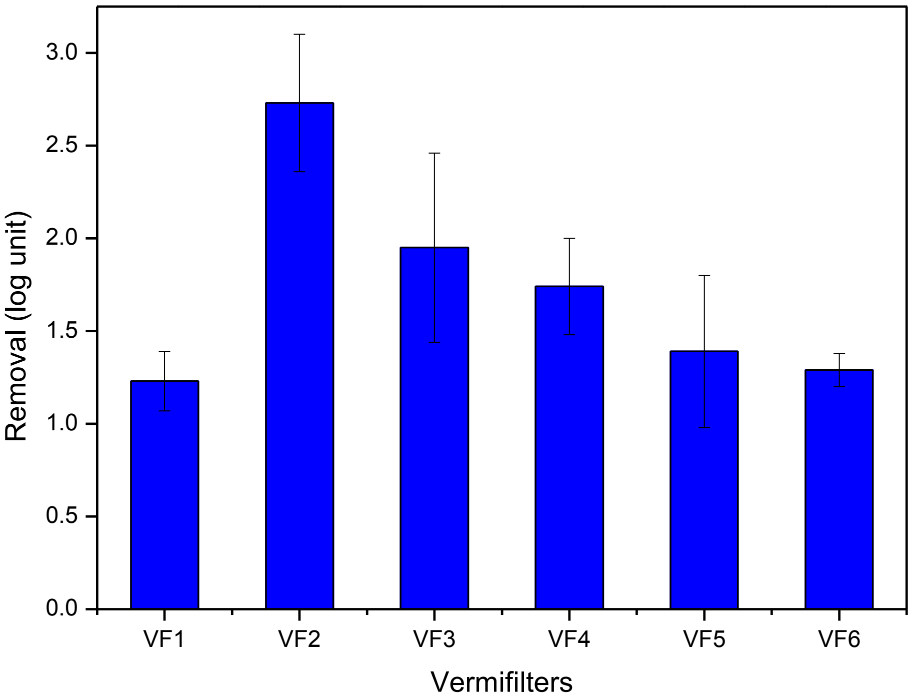

Fig. 4.6: Variation of FS in vermifilter having different earthworm stocking densities

4.3.2 Removal of E. coli

The log removal profile for *E. coli* with different earthworm stocking densities is shown in Fig. 4.7. Among the group of indicator organisms, special concern was given to *E. coli* which commonly presents in the intestinal tract of warm blooded animals and directly related with the risk of public health.

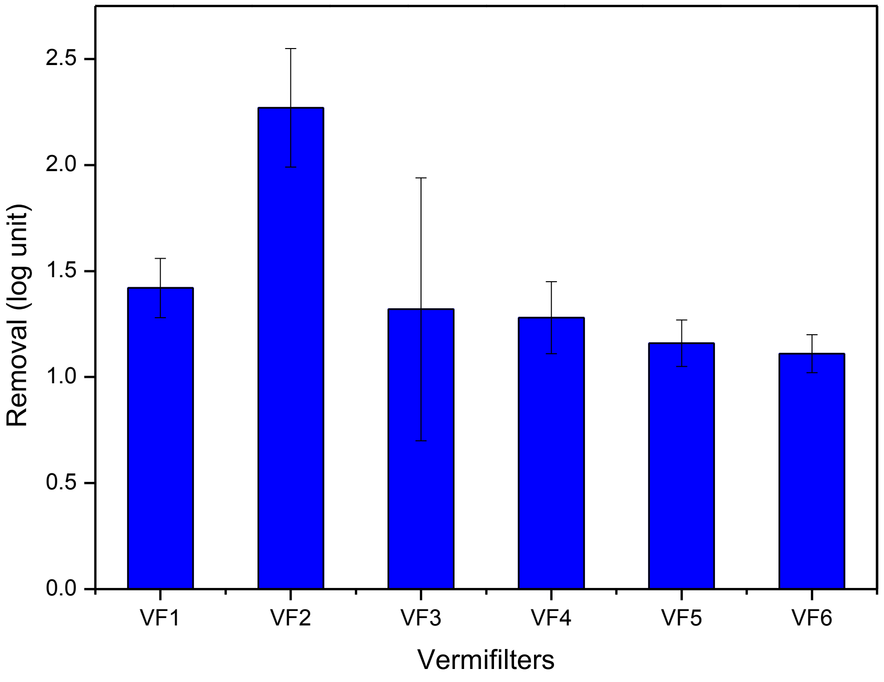

Fig. 4.7: Variation of *E. coli* in vermifilter having different earthworm stocking densities

Presence of *E. coli* in water sample is an indication of fecal pollution and potential existence of pathogenic bacteria. The average value of *E. coli* in effluent varied from 5.90×10^2 to 1.20×10^3 CFU/100mL during vermifiltration process having different stocking densities. However, VF2 reactor registered maximum reduction (2.27 ± 0.28 log) in comparison to other systems as VF1 (1.42 ± 0.14 log), VF3 (1.32 ± 0.62 log), VF4 (1.28 ± 0.17 log), VF5 (1.16 ± 0.11log) and VF6 (1.11 ± 0.09 log). The reduction of pathogens in vermifiltration processes mainly due to the action of intestinal enzymes secreted in earthworm's body wall (Khwairakpum and Bhargava, 2009). The microbial quality results indicate the potential of vermifilters for significant reduction of faecal count to less than permissible limit 1000 MPN/100mL as prescribed by WHO guidelines for unrestricted wastewater irrigation (WHO, 1989).

4.4 Earthworm Biomass Growth and Reproduction during Vermifiltration

Biomass study is very important parameter to evaluate feasibility of the VF system. Growth of earthworms (initial and final) studied for six different earthworm stocking densities and it is presented in Table 4.5. No mortality was observed in any VF and the earthworm number increased exponentially in VF1, VF2, VF3, VF4, VF5 and VF6 by 104.44, 80.89, 8.57, 16.22, 8.14 and 2.29%, respectively at 74g/day OLR on final day.

Table 4.5: Earthworm Biomass growth and reproduction from vermifiltration on T_{90} days

Stocking density (worms/cum)	Earthworm	Mean weight of EWs in gm		Live biomass	Number of EWs		Live Numbers	Cocoons ($worm^{-1} day^{-1}$)	Juvenile hatched ($worm^{-1} day^{-1}$)	Maximum Growth rate (gm wt. $worm^{-1} day^{-1}$)
		Initial	Final	% change	Initial	Final	% change			
5000	*E. Fetida*	56	124	121.43	180	368	104.44	0.52	N	0.76
10000	*E. Fetida*	70	155	121.43	225	407	80.89	0.51	N	0.94
15000	*E. Fetida*	120	174	45.00	350	380	8.57	0.46	N	0.60
20000	*E. Fetida*	160	192	20.00	450	523	16.22	0.48	N	0.36
25000	*E. Fetida*	175	193	10.30	565	611	8.14	0.47	N	0.2
30000	*E. Fetida*	210	224	6.70	675	695	2.96	0.48	N	0.16

Fig. 4.8 presents the growth of earthworm in vermifilter having different earthworm stocking densities at a fixed amount of organic loading rate. The distribution of constant amount of organic material for every individual and per gram of earthworm reduced as stocking density increased. From Fig. 4.8 it is clear that the accessible amount of food material directly influences the growth pattern of earthworms. *E. fetida* which showed a maximum individual biomass (121.4%) and growth rate (0.94 g wt. worm^{-1} day^{-1}) in VF2 under stocking density of 10000 worm/cum, while cocoon production rate was maximum (0.52 cocoon worm^{-1}day^{-1}) in VF6 under SD 30000 worm/cum of vermifilter bed at the end of experiment. Besides, only 94.64% and 15.24% increase in worm biomass was observed in VF1 and VF3, respectively. On the other hand, biomass reduction was observed as stocking density increased in case of VF4, VF5 and VF6 with 20000, 25000 and 30000 worm/cum at same organic loading rate (OLR). This loss in biomass can be attributed to deficiency of organic matter present at same OLR for higher stocking densities. Several numbers of juveniles were also observed in all earthworm stocking densities.

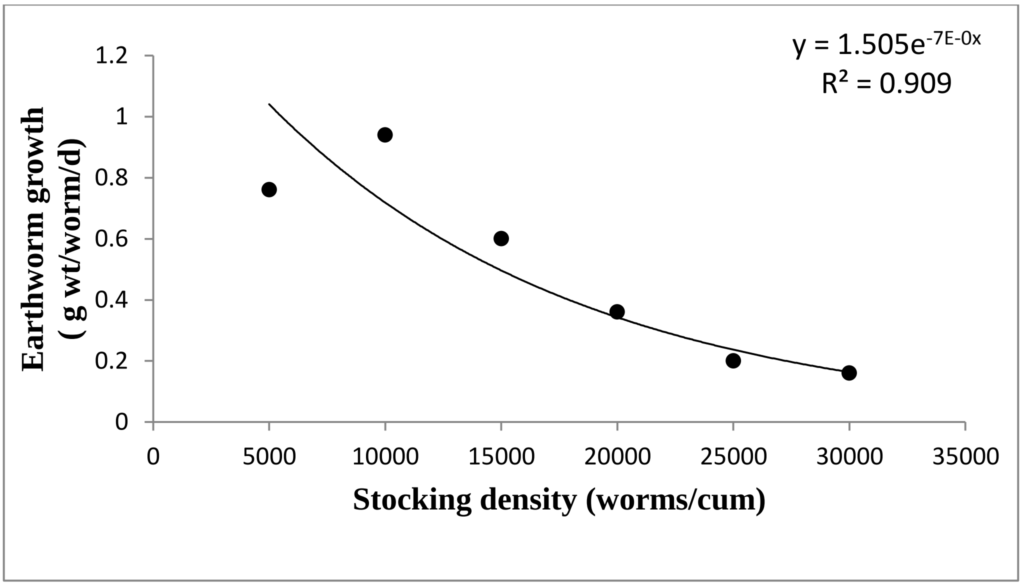

Fig. 4.8: Interrelationship between growth of earthworm and earthworms stocking densities

SUITABILITY OF DIFFERENT MATERIALS AS VERMIFILTER BED

5.1 General

The present chapter deals with identification of suitable media as a vermifilter bed for its better performance. For this purpose four different types of material i.e. river bed material (VFR), wood coal (VFC), glass ball (VFG) and mud balls (VFM) with vermicompost were used as media throughout the study as reported in Section 3.5.2. The performance evaluation was carried out on the basis of pollutant removal efficiency, earthworm biomass. There have been several attempts to mix and combination of different materials for evaluating the potential impacts of physical characteristics of the media on the overall vermifilter performance (Wang et al., 2010; Xing et al., 2010). In this direction various researchers have tried to work with different kinds of material as a media; like ceramsite, quartz sand and converter slag–coal cinder etc. The study related with these media had their individual objectives. Study conducted by Xing et al., (2010) demonstrated the health effect on earthworms and change in final characteristics of treated sludge from vermifiltration process. During this study ceramsite and quartz sand material were used and ceramsite material was found relatively better media for the process with better characteristics of sewage sludge. The health of earthworm was found better with ceramsite material in vermifiltration as compared to quartz sand due to less abrasion on earthworm’s body wall with ceramsite. In another study, conducted by Wang et al., (2010), converter slag–coal cinder was found most suitable. However, the study was concerned with removal of nutrients like ammonia nitrogen and phosphorous and organic contaminants. Thus, the present chapter is aimed on following aspects;

(i) Identification of relatively better media.

(ii) Performance evaluation of different vermifilters operated using different filter media based on the quality of effluent in terms of BOD, COD and TSS removal.

(iii) Removal efficiency of microbiological indicators i.e. total coliform (TC), fecal coliform (FC), *fecal streptococci* (FS), *Escherichia coli* (*E. coli*) in vermifilter having different filter media.

5.2 Physico-Chemical Changes during Vermifiltration and Performance of Vermifilter having Different Media Used in Vermifilter Bed

The reactors VFR, VFC, VFG and VFM were operated simultaneously for 90 days duration. The characteristics of the influent wastewater are given in Section 3.4. The removal of

BOD, COD, TSS, TC, FC, FS and *E. coli* from each of the vermifilter is measured and the detailed analysis is performed in the following section.

5.2.1 Removal of BOD

Table 5.1 presents the BOD variation in vermifilter having different media with time. During study relatively higher removal was observed in reactor having river bed material as compared to the other reactors as can be seen from Table 5.1.

Table 5.1: BOD variation in vermifilter having different media with different time (Mean± Standard Deviation, number of replicates=3)

S. No.	Time (Days)	Influent (mg/l)	Effluent BOD concentration (mg/L)			
			VFR	VFC	VFG	VFM
1	1	350±16	75±5	66±9	72±8	94±6
2	7	346±21	100±9	58±6	110±6	132±13
3	13	341±19	109±11	42±5	52±6	88±8
4	19	336±13	45±6	62±5	96±12	56±6
5	25	331±22	52±8	90±8	84±9	92±9
6	31	323±19	103±16	94±11	86±6	112±15
7	37	321±17	80±8	87±11	106±11	131±11
8	43	317±11	86±6	114±13	118±16	122±9
9	49	314±10	79±15	102±10	108±8	70±6
10	55	311±21	66±9	113±14	114±8	102±11
11	61	307±10	59±6	119±18	110±6	78±6
12	67	339±12	88±10	112±6	150±17	84±5
13	73	334±9	46±7	66±9	84±9	96±7
14	79	329±14	44±6	77±11	72±5	54±8
15	90	300±10	51±9	83±6	76±8	60±7

Overall, in reactor VFR the average value of BOD was observed as 72.2 mg/L while in reactor VFC it resulted 85.7 mg/L. Similarly, in reactor VFG the average value of BOD found as 95.9 mg/L while the reactor VFM exhibited as 91.4 mg/L throughout the study period.

Fig. 5.1 also shows the BOD removal pattern in vermifilter having different media. A sharp decreasing scenario was found for BOD in reactor VFR as compared to the other media (as shown in Table 5.1 also) and about 78% average removal was experienced. Besides, in reactor VFC, the average removal was observed as 74% and almost same average BOD removal efficiency was exhibited through vermifilter that contained glass balls and mud balls as a media but these media experienced lower earthworm biomass as mentioned in Table 5.6 and the removal was found as 71 and 70%, respectively. In literature it has been evidenced that during vermifiltration, removal of BOD may be attributed to symbiotic activity of earthworms and aerobic microbes which accelerate and enhance the decomposition of organic matter (Loehr et al., 1988; Kumar et al., 2014; Arora et al., 2014) and shows the efficacy of vermifiltration process having river bed material as a media.

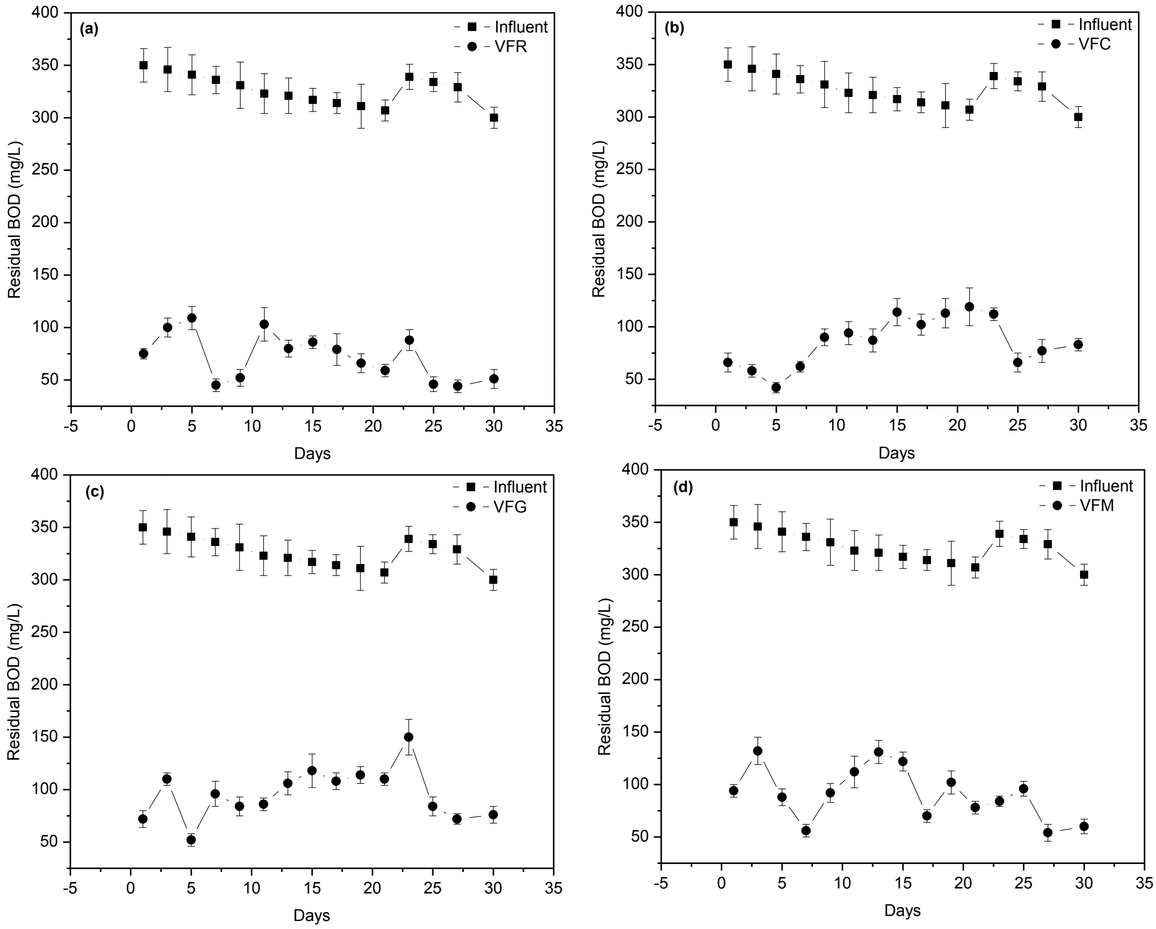

Fig. 5.1: BOD removal pattern in reactor VFR, VFC, VFG and VFM

5.2.2 Removal of COD

Table 5.2 presents the COD variation in vermifilter having different media with different time. Overall, in reactor VFR the average value of COD was observed as 133.3 mg/L while in reactor VFC it resulted 170 mg/L. Similarly, in reactor VFG the average value of COD found as 184.8 mg/L while the reactor VFM exhibited as 193.4 mg/L throughout the study period.

Table 5.2: COD variation in vermifilter having different media with time (Mean± Standard Deviation, number of replicates=3)

S. No.	Time (Days)	Influent (mg/l)	Effluent COD concentration (mg/L)			
			VFR	VFC	VFG	VFM
1	1	553±32	115.49±19	162.98±12	164.16±17	152.24±19
2	7	548.55±27	167.3±22	121.08±16	198.2±15	220.98±27
3	13	504.91±16	171.67±16	96.35±9	90.34±14	223.67±19
4	19	483.4±13	108.07±11	169.24±7	197.21±9	168.83±11
5	25	465.47±12	98.37±10	156.16±11	153.63±9	147.94±17
6	31	450.74±17	173.35±17	233.69±17	168.35±6	216.42±15
7	37	443.34±23	161.48±14	241.25±21	203.99±18	272.11±18
8	43	441.7±19	166.34±16	188.59±13	200.4±21	266.52±11
9	49	441.08±21	157.84±11	163.11±10	202.84±13	180.64±10
10	55	430.83±25	145.82±11	189.28±14	198.44±8	207.72±14
11	61	410.34±10	108.7±9	206.09±18	207.09±11	193.65±16
12	67	493.29±21	134.44±12	188.42±14	270.83±17	133.84±9
13	73	480.96±18	104.34±7	124.99±9	191.49±9	222.58±17
14	79	455±21	84.27±6	162.49±11	168.37±8	139.73±8
15	90	402.1±10	102.41±9	146.37±6	156.38±8	153.5±10

The variations of COD in vermifilter having different media are presented in Fig. 5.2. From Fig. 5.2, it can also be seen that the COD of effluent was significantly low in all reactors as compared to influent concentration. The reactor VFR exhibited relatively higher removal as compared to the other reactors.

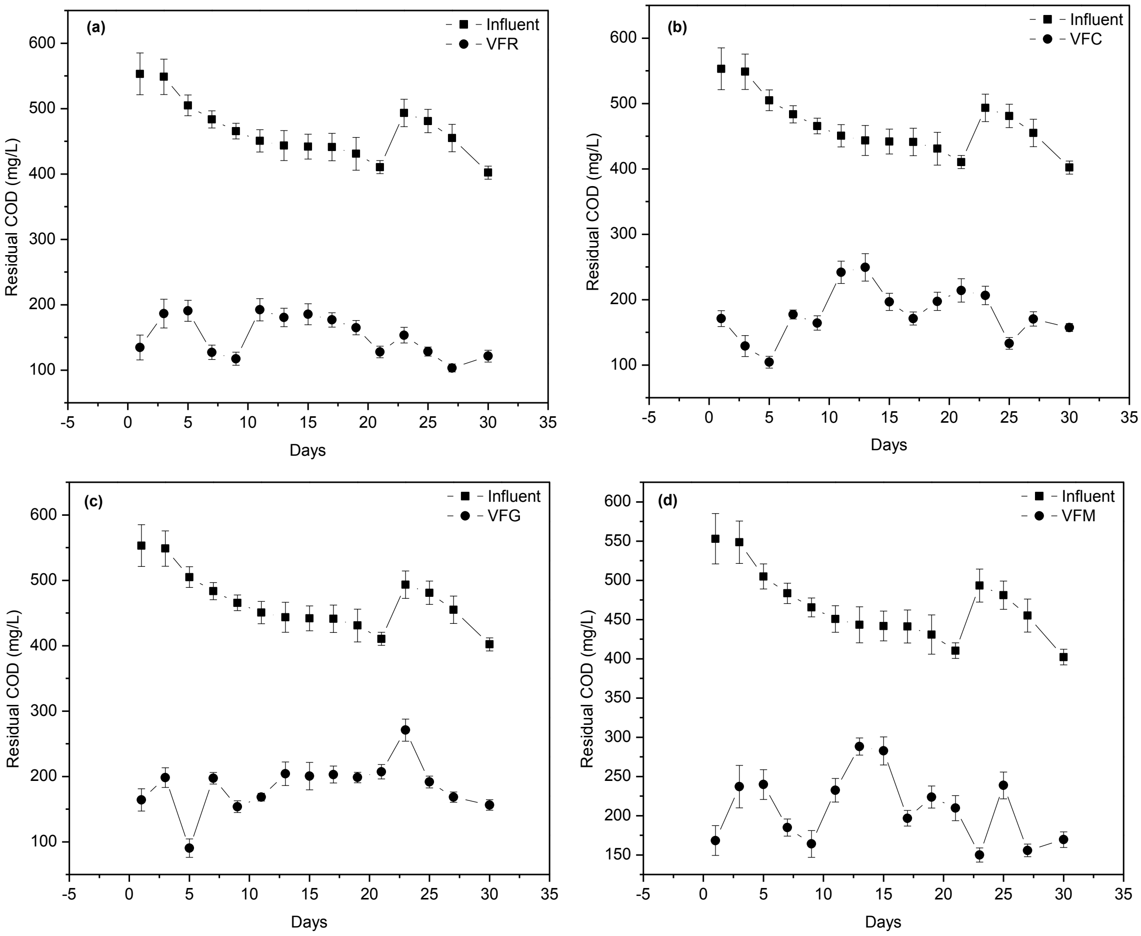

Fig. 5.2 COD removal in reactor VFR, VFC, VFG and VFM

This may be due to higher activity of earthworm in VFR as indicated in Table 5.6. COD removal can be correlated with earthworm activity. Higher activity of earthworm results in higher COD removal. About 71% average removal was observed in VFR while VFC exhibited 64% removal. For reactor VFG and VFM the removal was observed as 60 and 59% respectively. In addition to this, a physical property (specific surface area and porosity) of media also plays an important role, which is one of the factors that affect the treatment performance of the reactor (Young and Dahab, 1983; Tay and Show, 1998; Surampalli et al., 2007). Media with relatively higher surface area or low porosity facilitate greater biomass accumulation and attains higher treatment efficiency. Since, the river bed material has higher surface area and low porosity (as mentioned in Table 3.2 in Chapter 3), the reactor VFR exhibited higher removal efficiency.

5.2.3 Removal of TSS

Table 5.3 presents the TSS variation in vermifilters having different media with time. Overall, in reactor VFR the average value of TSS concentration was observed as 62 mg/L while in reactor VFC it resulted 89 mg/L. Similarly, in reactor VFG the average value of TSS concentration found as 103 mg/L while the reactor VFM exhibited as 111 mg/L throughout the study period.

Table 5.3: TSS variation in vermifilter having different media with time (Mean± Standard Deviation, number of replicates=3)

S. No.	Time (Days)	Influent (mg/l)	Effluent TSS concentration (mg/L)			
			VFR	VFC	VFG	VFM
1	1	235.8±26	49±10	79±8	85±16	67±14
2	7	237.75±21	101±13	67.14±8	115±15	153±21
3	13	230.4±18	102±9	50±13	57±18	166±21
4	19	243±23	43±11	81±19	114.9±19	77±16
5	25	220.4±22	37±9	74.63±16	59±15	65±15
6	31	219.4±29	117±9	109±18	94±18	132±18
7	37	237.8±26	75±8	175±10	125.18±21	169±22
8	43	298.4±25	91±13	96±15	117.15±18	168±27
9	49	294±25	63±11	79.31±15	123±17	90±16
10	55	171±20	53±16	105±10	117±15	123±22
11	61	153.6±19	45±16	105±17	128±21	118±20
12	67	230.35±25	50±10	93±20	133.71±18	51±15
13	73	242.95±19	39±8	73±21	109±14	159±21
14	79	220.35±20	29±10	75±21	102.21±11	62±15
15	90	219.35±20	38±9	73±18	66.57±15	70±17

Fig. 5.3 presents the TSS concentration pattern during entire study with time. The TSS concentration was reduced during vermifiltration in all media significantly as compared to influent. The maximum TSS removal about 73% in reactor VFR was observed while for VFC it followed as 61%. Besides, in reactor VFC the removal of TSS was observed as 55% and the reactor VFM this removal experienced as 52%. During vermifiltration, the removal of TSS could be attributed to the ingestion of organic and inorganic solid particles in wastewater through earthworm which excrete them as finer particles. In literature 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 from wastewater (Sinha et al., 2008). In VFR this phenomenon was dominated due to relatively higher activity of earthworm experienced during study (Table 5.6).

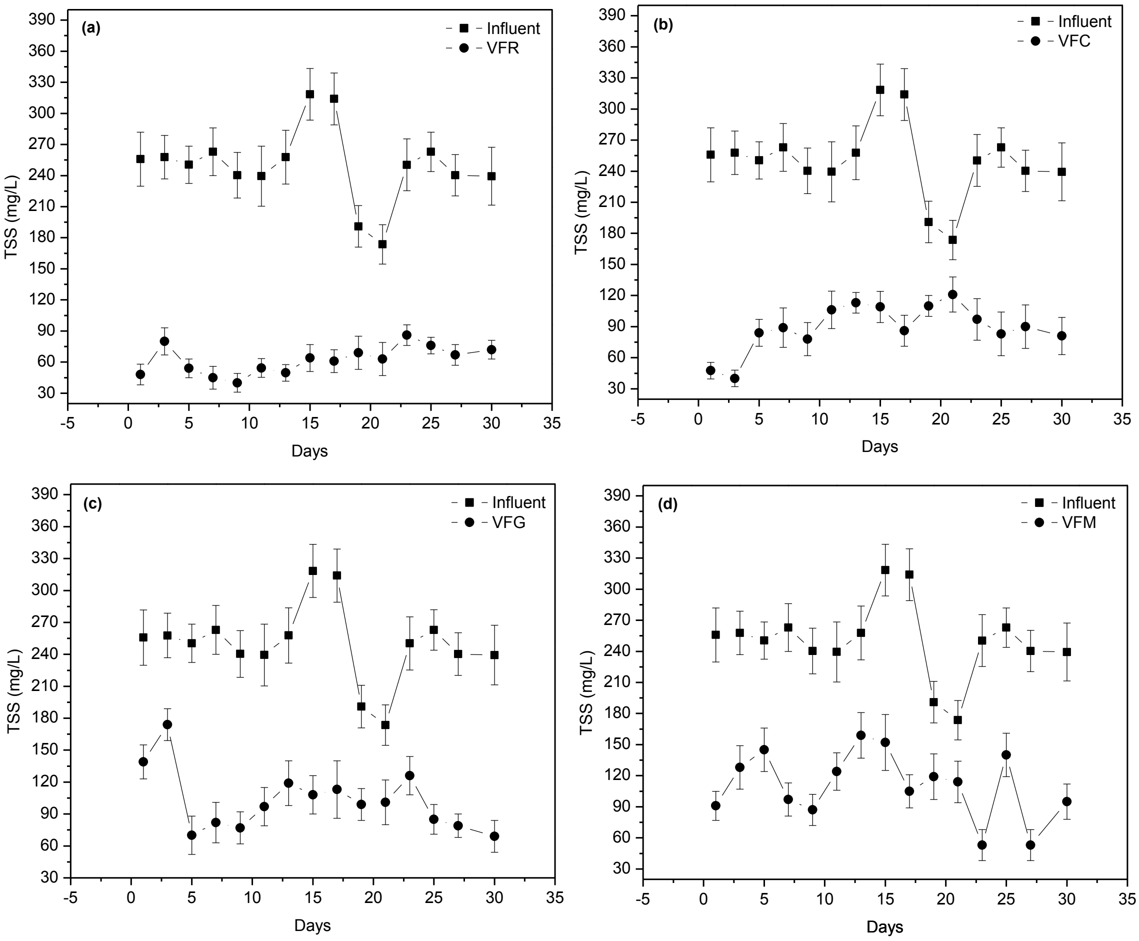

Fig. 5.3: TSS pattern in reactor VFR, VFC, VFG and VFM

Various physical, chemical and biological reactions take place in vermifiltration process including the adsorption of molecules and ions, oxidation–reduction of organic matter, the behavior of earthworms and their synergetic effects with microorganisms (Bouché and Soto, 2004). During vermifiltration process, a tortuous behavior for TSS concentration was also observed. This unfavorable performance might be due to turbulence, which led to washing out of the influent solids and settled biomass (Sharma et al., 2014). The stimulatory effect of earthworms could also be a reason behind this tortuous behavior in which mucus and cast are produced. The mucus is a source of easily assimilable carbon for microorganisms, while casts are often enriched with available forms of C, N and P (Aira et al., 2007). So due to leaching of vermicast this type of behavior has been observed. Further, the adsorption of the impurities in the wastewater will not fully adsorb on the sand and gravel particles and they will be washed away from the reactor (Manyuchi et al., 2013).

5.2.4 Removal of Indicator Organisms

According to WHO (2002), the presence of indicator organisms often used to provide total spectrum of overall pathogenicity of the sample. The average value of microbial quality of effluent coming out from vermifilters is presented in Table 5.4. In all the reactors, significant reductions of indicator organisms were observed. However, in VFR reactor, maximum reduction of TC, FC and FS has been evidenced and found as 3.6 ± 0.90, 3.4 ± 0.67 and 2.5 ± 0.51 log unit, respectively. On the contrary, for VFC reactor, the reduction of TC, FC and FS were found as 3.1 ± 0.65, 2.9 ± 0.88 and 2.0 ± 0.79 log unit, respectively. Similarly, in reactor VFG, the reductions were recorded as 2.8 ± 1.11, 2.6 ± 0.45 and 1.7 ± 0.92 log unit, respectively. In case of VFM reactor, it was reduced to 2.1 ± 0.83, 2.6 ± 1.05 and 1.7 ± 0.72 log unit, respectively.

Among the group of indicator organisms, the special concern was given to *E. coli* which commonly present in intestinal tract of warm blooded animals and directly related with the risk of public health. The presence of *E. coli* in water sample is an indication of fecal pollution and the potential existence of pathogenic bacteria. The average value of *E. coli* in effluent was observed as $1.95\times10^2 \pm 4.79\times10^1$ CFU/100mL during vermifiltration process having different media. However, VFR reactor registered maximum reduction (3.32 ± 0.62 log) in comparison to other systems as VFC (2.82 ± 0.81 log), VFG (2.52 ± 0.38 log) and VFM (2.52 ± 0.92 log). The reduction of pathogens in vermifiltration processes is mainly due to the action of intestinal enzymes secreted in earthworm's body wall (Khwairakpam and Bhargava, 2009). The gizzard and intestine available in earthworms work as a "bioreactor". Besides, the earthworms secrete enzymes proteases, lipases, amylases, cellulases and chitinases in their gizzard and intestine which bring about rapid biochemical conversion of the cellulosic and the proteinaceous materials in the waste organics. They ingest the food materials, cull the harmful microorganisms and deposit them mixed with minerals and beneficial microbes as “vermicast” in the soil (Sinha et al., 2010). In literature, it is also reported that the earthworms release coelomic fluids that have anti-bacterial properties and destroy all pathogens in the waste biomass. They produce antibiotics and kill the pathogenic organisms in the waste and inhabit it virtually sterile (Pierre et al., 1982). The microbial quality results indicate the potential of vermifilters for significant reduction of faecal count to less than permissible limit 1000 MPN/100mL as prescribed by WHO guidelines for unrestricted wastewater irrigation (WHO, 1989). Hence, the effluent from vermifilter particularly with VFR indicates safer as well as hygienic from agricultural point of view.

Table 5.4: Microbial quality of effluent from vermifilter having different media (Mean± Standard Deviation, number of replicates=3)

Parameter	TC (MPN/100ml)		FC (MPN/100ml)		FS (MPN/100ml)		E. Coli (CFU/100ml)	
	Mean	Removal	Mean	Removal	Mean	Removal	Mean	Removal
Influent	$3.16\times10^{5}\pm1.58\times10^{4}$	-	$7.94\times10^{4}\pm1.26\times10^{3}$	-	$7.94\times10^{3}\pm1.58\times10^{3}$	-	$1.26\times10^{4}\pm6.31\times10^{3}$	-
VFR	$7.94\times10^{2}\pm1.26\times10^{2}$	3.6±0.90	$4.8\times10^{2}\pm1.58\times10^{2}$	3.4±0.67	$4.32\times10^{2}\pm1.26\times10^{2}$	2.5±0.51	$1.95\times10^{2}\pm4.79\times10^{1}$	3.32±0.62
VFC	$1.26\times10^{3}\pm6.31\times10^{2}$	3.1±0.65	$7.6\times10^{2}\pm1.26\times10^{2}$	2.9±0.88	$6.85\times10^{2}\pm9.77\times10^{1}$	2.0±0.79	$5.52\times10^{2}\pm1.26\times10^{2}$	2.82±0.81
VFG	$2.0\times10^{3}\pm1.17\times10^{3}$	2.8±1.11	$1.21\times10^{3}\pm7.94\times10^{2}$	2.6±0.45	$1.08\times10^{3}\pm7.76\times10^{2}$	1.7±0.92	$8.75\times10^{2}\pm3.98\times10^{2}$	2.52±0.38
VFM	$1.58\times10^{3}\pm7.94\times10^{2}$	2.1±0.83	$9.57\times10^{2}\pm3.98\times10^{2}$	2.6±1.05	$8.62\times10^{2}\pm5.01\times10^{2}$	1.7±0.72	$6.95\times10^{2}\pm2.0\times10^{2}$	2.52±0.92

5.2.5 Variation of Other Parameters

In vermifiltration, major portion of nitrogen present as NH_4^+-N in wastewater removed mainly through soil and sand parts of the reactor due to rapid adsorption through biomass and filters (Wang et al., 2011). However, reactor VFR exhibited maximum removal about 69%, due to higher activity of earthworms. While in VFM minimum removal was observed as 41%. In reactor VFR and VFC no significant difference (as shown in Table 5.5) was observed and same kind of scenario was followed between VFG and VFM. This removed NH_4^+-N through rapid adsorption subsequently converted into nitrate form through biological nitrification (Kadam et al., 2009). Li et al., 2008 evidenced that the mechanism behind nitrogen removal might be that earthworm and microorganism bodies consume some nitrogen. Remaining part of nitrogen removed due to formations of N_2, NH_3 and NO_x through nitrification, de-nitrification and ammonification. Bajsa et al., (2003) has reported that earthworms secrete polysaccharides, proteins, and other nitrogenous compounds. They mineralize the nitrogen in the sewage to make it available to plants as nutrients. Similarly Wang et al., (2011) has also investigated that oxygen is available in abundance through the burrowing action of earthworms which favours a microenvironment for aerobic nitrobacteria.

The total phosphate (TP) concentration in effluent was observed to be significantly increased in all reactors as compared to influent concentration. However, among these reactors no significant difference was observed for TP (as shown in Table 5.5). In all reactors it varied in the range of 5.1-8.1 mg/L. In vermifiltration process increased TP concentration attributed to the enzymatic and microbial action of earthworms. Activities of earthworm and associated microbes in vermifilter bed promote rapid phosphate mineralization in the system causing increased concentration of TP in the effluent (Hait and Tare, 2011; Kumar et al., 2014).

The DO concentration in all reactors varied in the range of 5.4 to 6.2 mg/L as indicated in Table 5.5. However, no significant difference was observed among these reactors regarding this parameter. The increased DO concentration may be attributed to aerobic conditions created by earthworms in vermifilter bed and waste materials, through their burrowing actions, inhibiting the action of anaerobic microorganisms (Sinha et al., 2008). When earthworms continuously moved in the system it results in channeling of the system through their burrowing action and favoured an aerobic environment.

No significant change in pH was observed in all the reactors as compared to influent and earthworms tend to neutralize it. The pH in all the reactors varied in the range of 7.1 to 7.4 as indicated in Table 5.5. However, no significant difference was observed among these reactors regarding pH. Various studies have been carried out to assess the likelihood of biological inhibition and disruption from pH to earthworm species in vermifiltration and the system exhibited the evidence of inbuilt pH buffering capacity (Hughes et al., 2007; Sinha et al., 2008).

Table 5.5: Other parameters during vermifiltration having different media (Mean± Standard Deviation, number of replicates=3)

S. No.	Parameters	Influent	Concentration of effluent in vermifilter having different media			
			VFR	VFC	VFG	VFM
1	NH_4^+-N (mg/L)	48.5±11.4	11.8±3.7	12.4±2.1	20.2±4.4	22.5±7.3
2	NO_3-N (mg/L)	1.5±0.2	31.2±5.9	22.5±6.6	23.7±4.3	19.1±4.5
3	TP (mg/L)	5.2±1.6	18.1±4.6	16.6±3.3	13.3±2.1	13.8±3.2
4	DO (mg/L)	3.2±1.1	6.2±1.4	5.4±0.9	5.9±1.6	6.0±1.2
5	pH	7.3±0.6	7.1±0.3	7.3±0.6	7.4±0.6	7.2±0.6

5.3 Earthworm Growth and Reproduction

Biomass study is very important parameter for evaluating feasibility of vermifiltration system and its long term performance. Growth of earthworms studied for all different media (initial and final) is depicted in Table 5.6. In vermifilter having media river bed material, wood coal and glass ball a significant difference was observed. In reactor VFR maximum earthworm biomass was observed with 73% increment while in VFM, it was observed as 30%. So, river bed material exhibited a better alternative option, as a media comparatively that have already been used in literature.

Table 5.6: Earthworm biomass in different reactor (Mean± Standard Deviation, number of replicates=3)

S. No.	Vermifilter	Earthworm biomass	
		Initial weight of earthworm (gm)	Final weight of earthworm (gm)
1	VFR	50±4.7	86.5±5.3
2	VFC	50±4.7	73.5±3.6
3	VFG	50±4.7	64.5±4.1
4	VFM	50±4.7	65.0±4.4

5.4 Quality of Vermicompost on Top Layer of Vermifilter Bed

The initial and final characteristic of vermicompost in different reactor is presented in Table 5.7. In all the reactors, the final end product found as vermicompost from the top layer was dark brown (towards blackish) in colour. The C/N ratio of end product revealed stability of waste that was deposited during vermifiltration process. The data represented an increasing concentration pattern of TOC in all reactors and it was observed to be varying in the range of 318-347 g/Kg. In VFR and VFC reactor, no significant difference was observed and the TOC concentrations were recorded as 318±10 and 313±13 g/Kg, respectively. Thus, VFR reactor represented minimum TOC concentration. The increased concentration of TOC in vermifilters as compared to initial concentration of vermicompost could be attributed to the deposition of biodegradable part of synthetic wastewater on top layer of vermifilters (Kumar et al., 2014). Despite this, C/N ratio in end products were reduced and varied in the range of 8.6-12.8. This could be attributed to the increased concentration of TN in all reactors and its concentration varied in the range of 26.6-37.1 g/Kg. In VFR reactor, the maximum concentration TN was identified and recorded as 37.1±5 g/Kg. The higher TP concentration in vermicompost at the end of the run proves suitability of all media which provides favourable environment for the growth of earthworms. No obvious difference was observed regarding pH, C/N ratio, TP and ash content. The reactor VFR represented relatively higher concentration of TN while between VFC and VFG, no significant changes were recorded.

Table 5.7: Characteristics of vermicompost from vermifilter having different media (Mean± Standard Deviation, number of replicates=3)

S. No.	Parameters	Initial characteristics of vermicompost	Vermifilter			
			VFR	VFC	VFG	VFM
1	pH	7.3±0.2	7.6±0.4	7.2±0.5	7.0±0.1	7.2±0.4
2	C/N ratio	11.1±0.2	8.6±2.1	11.5±1.6	12.3±1.2	12.8±1.7
3	TOC (g Kg^{-1})	280±3.8	318±10	330±13	347±11	340±19
4	TP (g Kg^{-1})	23±1.6	29.3±3.6	26.4±2.4	26±3.7	25.3±1.2
5	TN (g Kg^{-1})	25.2±0.2	37.1±5.0	28.8±3.2	28.2±2.3	26.6±2.9
6	Ash content (%)	50.1±2.3	54.4±3.2	51±1.4	51.6±2.8	52±1.6

Overall, river bed material was found to be suitable media as vermifilter bed for efficient treatment of wastewater as the earthworm biomass was observed much higher as compared to other studied media like wood coal, glass balls and mud balls for requisite quality of effluent. It can be concluded that river bed material can be used as an alternative as a vermifilter bed for decentralized onsite wastewater treatment.

EFFECT OF HYDRAULIC LOADING RATES ON THE PERFORMANCE OF VERMIFILTER

6.1 General

The hydraulic loading rate plays an important role in regulation of quality of effluent. Higher hydraulic loading rate reduces the quality of treated water as the earthworms and microorganisms do not get sufficient time for digestion of organic matter present in wastewater (Li et al., 2009). So, it is one of the prime factors for the treatment of wastewater during vermifiltration process. Most of the researchers have not studied in detail the effect of different hydraulic loading rate on vermifiltration.

During vermifiltration earthworms increase the hydraulic conductivity of filter media and natural aeration by granulating the clay particles (Sinha et al., 2008). But as such no data are available that could prove this statement in literature regarding this parameter. Hence, an attempt has been made by comparing the vermifiltration process against geofilter (control reactor). Thus, the present chapter discusses the effect of hydraulic loading rates (HLRs) on the performance of vermifilter.

The lab scale study was concerned with the following aspects:

- Performance evaluation of different vermifilters operated at different HLRs.
- Identification of relatively better HLR.
- Comparative study on the hydraulic aspects of the vermifilter and geofilter.

For this purpose, a pilot scale vermifilter (VFR) made of plastic container was set up and consisting of five parts: filter bed, filter material, earthworms, water distributor and drain system (as discussed in Section 3.5.1). The reactor consisted of plastic container having cross sectional dimension of 250 mm x 200 mm and a depth of 300 mm. The top layer consisted of 100 mm thick matured vermicompost (worm-bed). Second, third and fourth layers were of river bed material (as evidenced relatively better media in Chapter 5) of size 6 to 8 mm (50 mm thick), 2 to 4 mm (50 mm thick) and 10 to 12.5 mm (50 mm thick), respectively. The reactors were run with wastewater at different hydraulic loading rates to find out the optimum rate. In addition, the performance of vermifilter (VFR) is also compared with a geofilter (devoid of earthworms; CRR). Both vermifilter and geofilter were fed daily at five different HLRs of 0.5, 1.0, 1.5, 2.0 and 2.5 $m^3m^{-2}d^{-1}$, respectively for 90 days. In addition, to investigate the hydraulic aspects of the vermifilter the hydraulic conductivity of filters were measured at different HLRs and a comparison was carried out between the performance of vermifilter and geofilter.

6.2 Physico-Chemical Changes during Vermifiltration and Performance Evaluation of Vermifilter and Geofilter at Different HLRs

The reactors VFR and CRR were operated simultaneously for 90 days duration at different HLRs i.e. 0.5, 1.0, 1.5, 2.0 and 2.5 $m^3m^{-2}d^{-1}$, respectively. The characteristics of the influent wastewater are given in Section 3.4. The removal of BOD, COD and TSS from each of the vermifilter and geofilter is measured and the detailed analysis is performed in the following section.

6.2.1 Removal of BOD

Table 6.1 presents the BOD variation in vermifilter (VFR) and geofilter (CRR) at different HLRs with time. During study relatively higher removal was observed in reactor VFR as compared to CRR at all HLRs. Besides, higher removal of BOD was observed at HLR of 1.0 and 1.5 $m^3m^{-2}d^{-1}$ as compared to the other HLRs as can be seen from Table 6.1. Overall, in reactor VFR the average value of BOD, at HLR of 1.0 and 1.5 $m^3m^{-2}d^{-1}$, was observed as 43.1and 47.7 mg/L, respectively. Similarly, in reactor CRR the average value of BOD was found as 78.9 and 84.4 mg/L at these HLRs throughout the study period.

The average BOD removal from both the reactors is shown in Fig. 6.1. From Fig. 6.1 it can be concluded that the average removal of BOD, in VFR, varied between 70.2-86.9% and the maximum of 86.9% removal was observed at HLR of 1.0 $m^3m^{-2}d^{-1}$. This could be due to the higher population of earthworm which plays an important role in degradation of organic matter available in wastewater. In reactor CRR relatively less BOD removal was observed as compared to the reactor VFR and it varied between 70-75%.

Table 6.1: Comparison of BOD removal at different HLRs between Vermifilter (VFR) and Geofilter (CRR)

S. No.	Time (Days)	Influent (mg/L)	Effluent BOD concentration at different HLRs (mg/L)									
			0.5 $m^3/m^2/d$		1.0 $m^3/m^2/d$		1.5 $m^3/m^2/d$		2.0 $m^3/m^2/d$		2.5 $m^3/m^2/d$	
			VFR	CRR	VFR	CRR	VFR	CRR	VFR	CRR	VFR	CRR
1	1	350	91	102	98	141	112	161	123	173	129	171
2	7	346	96	105	74	91	48	74	102	159	114	118
3	13	341	59	93	36	97	61	116	76	139	107	115
4	19	336	52	79	39	94	81	98	92	115	89	97
5	25	331	61	62	38	61	57	76	71	124	63	89
6	31	323	66	86	34	69	44	68	66	70	44	86
7	37	321	61	69	62	81	34	71	53	96	91	96
8	43	317	56	59	39	52	23	76	60	59	71	88
9	49	314	45	81	28	61	20	59	66	65	31	76
10	55	311	66	73	42	56	26	69	38	92	45	92
11	61	307	92	69	33	69	47	94	38	71	54	81
12	67	339	83	131	24	64	52	70	51	61	49	93
13	73	334	86	89	29	89	34	71	50	56	49	87
14	90	329	69	49	28	79	29	78	61	63	32	63
Average		328.5	70.2	81.9	43.1	78.9	47.7	84.4	67.6	95.9	69.1	96.6

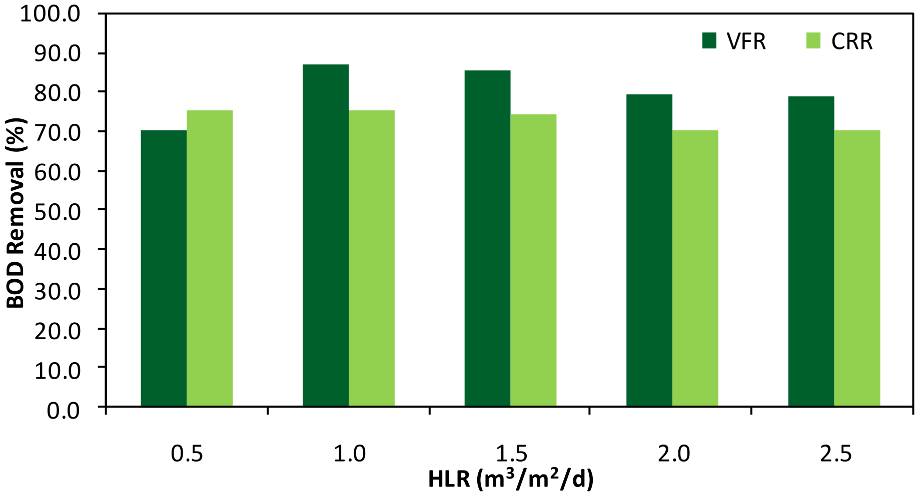

Fig. 6.1: Average BOD removal at different HLRs in VFR and CRR

At lower and higher HLRs from 1.0 and 1.5 $m^3m^{-2}d^{-1}$ the removal was not effective. At lower HLR of 0.5 $m^3m^{-2}d^{-1}$, the lower removal efficiency could be due to lack of availability of food to the earthworm which diminish the earthworm growth and finally affect the degradation process. Similarly, at higher HLRs the lower efficiency could be related to increased humidity and scouring of vermifilter which is not beneficial for earthworm growth and the performance of vermifiltration process (Xing et al., 2010). The HLR plays an important role in regulation of quality of effluent. Higher hydraulic loading rate reduces the quality of treated water as the earthworms and microorganisms do not get sufficient time for digestion of organic matter present in wastewater (Li et al., 2009). So, it is one of the prime factors for the treatment of wastewater during vermifiltration process. In literature also it has been evidenced that during the vermifiltration process micrographs of wormcast within the system exhibits micro-pores on surface and abundant cylindrical organic matter acts as a filtering medium for the removal of organic matter available in influent (Wang et al., 2011). In addition to this, the activities of protease, alkaline phosphatase (ALP) and cellulase in earthworm body diminished with increase in hydraulic loading rate and finally affect the performance of vermifilter (Xing et al., 2010).

6.2.2 Removal of COD

Table 6.2 presents the COD variation in vermifilter (VFR) and geofilter (CRR) at different HLRs with time. During study relatively higher removal was observed in reactor VFR as compared to CRR at all HLRs. Besides, higher removal of COD was observed at HLR of 1.0 and 1.5 $m^3m^{-2}d^{-1}$ as compared to the other HLRs as can be seen from Table 6.2. Overall, in reactor VFR the average value of COD, at HLR of 1.0 and 1.5 $m^3m^{-2}d^{-1}$, was observed as

96.7 and 123.8 mg/L, respectively. Similarly, in reactor CRR the average value of COD was found as 143.6 and 154.4 mg/L at these HLRs throughout the study period.

The average COD removal from all the reactors is shown in Fig. 6.2. From Fig. 6.2 it can be concluded that the average removal of COD, in VFR, varied between 69.4-79.6% and the maximum of 79.6% removal was observed at HLR of 1.5 $m^3m^{-2}d^{-1}$. This could be due to the higher population of earthworm which plays an important role in degradation of organic matter available in wastewater. In reactor CRR relatively less COD removal was observed as compared to the reactor VFR and it varied between 70-75% as only geomicrobial process works in such type of systems.

Table 6.2: Comparison of COD removal at different HLRs between Vermifilter (VFR) and Geofilter (CRR)

S. No.	Time (Days)	Influent (mg/L)	Effluent COD concentration at different HLRs (mg/L)									
			0.5 $m^3/m^2/d$		1.0 $m^3/m^2/d$		1.5 $m^3/m^2/d$		2.0 $m^3/m^2/d$		2.5 $m^3/m^2/d$	
			VFR	CRR	VFR	CRR	VFR	CRR	VFR	CRR	VFR	CRR
1	1	490	146.7	194.2	186.0	229.0	196.0	256.3	209.0	280.9	223.0	257.9
2	7	505	165.0	164.6	150.2	153.8	99.0	122.1	191.1	267.7	209.9	199.4
3	13	500	102.0	150.0	77.7	162.0	122.1	194.0	148.8	238.0	190.0	196.0
4	19	492	88.3	155.1	86.8	161.6	164.0	168.2	183.0	196.7	187.1	176.4
5	25	485	106.0	107.5	81.7	104.0	137.2	133.0	145.7	236.3	138.5	162.7
6	31	478	143.0	146.6	79.3	123.7	120.7	125.4	140.3	135.7	89.0	161.9
7	37	466	129.0	141.1	127.0	146.0	138.0	130.8	130.8	191.6	191.0	183.0
8	43	463	116.0	116.7	94.2	99.0	104.5	150.8	135.7	119.6	123.6	161.4
9	49	457	99.0	160.5	82.3	119.0	69.0	118.8	142.5	132.0	64.7	145.0
10	55	452	126.0	123.8	102.5	113.0	94.9	145.8	109.7	179.3	106.2	173.6
11	61	448	171.0	121.1	70.0	137.0	127.2	195.4	101.1	148.7	144.1	156.6
12	67	443	151.0	206.0	61.1	129.0	131.8	131.2	127.0	121.3	139.0	170.8
13	73	490	162.0	149.8	69.4	177.8	113.7	131.6	91.9	114.0	142.8	164.1
14	90	483	128.3	96.0	85.9	156.0	115.0	159.0	121.3	121.1	82.6	125.9
Average		475.1	131.0	145.2	96.7	143.6	123.8	154.4	141.3	177.4	145.1	173.9

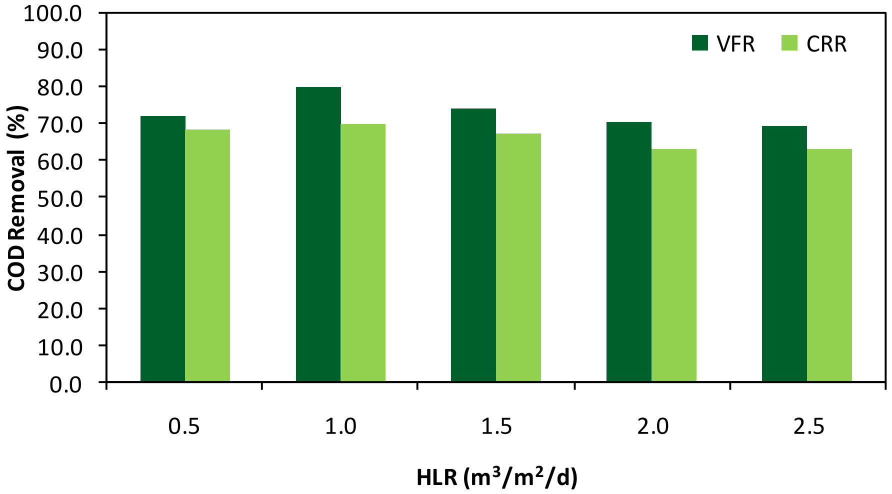

Fig. 6.2: Average COD removal at different HLRs in VFR and CRR

During vermifiltration, some physical, chemical and biological processes and synergistic effects of earthworms and microorganisms, including the adsorption of small particle organisms, colloid organisms and organic molecules, as well as the oxidation-reduction of organic matter and activity of earthworms are helpful for COD reduction. Further, Microporous structure together with wormcast provided in the system results in efficient removal of COD also (Wang et al., 2011).

6.2.3 Removal of TSS

Table 6.3 presents the TSS variation in vermifilter (VFR) and geofilter (CRR) at different HLRs with time. During study relatively higher removal was observed in reactor VFR as compared to CRR at all HLRs. Besides, higher removal of TSS was observed to be varying between 1.0 and 1.5 $m^3m^{-2}d^{-1}$, as it can be seen from Table 6.3 also. Overall, in reactor VFR the average value of TSS, at HLR of 1.0 and 1.5 $m^3m^{-2}d^{-1}$, was observed as 76.1 and 58.5 mg/L, respectively. Similarly, in reactor CRR the average value of TSS was found as 136.6 and 151.4 mg/L at these HLRs throughout the study period.

The average TSS removal from all the reactors is shown in Fig. 6.3. From Fig. 6.3 it can be concluded that the average removal of TSS, in VFR, varied between 62.6-76.1% and the maximum of 76.1% removal was observed at HLR of 1.5 $m^3m^{-2}d^{-1}$. This could be due to the higher population of earthworm which plays an important role in degradation of organic matter available in wastewater. In reactor CRR relatively less TSS removal was observed as compared to the reactor VFR and it varied between 37-47% as only geomicrobial process works in such type of systems.

Table 6.3: Comparison of TSS removal at different HLRs between Vermifilter (VFR) and Geofilter (CRR)

S. No.	Time (Days)	Influent (mg/L)	Effluent TSS concentration at different HLRs (mg/L)									
			0.5 $m^3/m^2/d$		1.0 $m^3/m^2/d$		1.5 $m^3/m^2/d$		2.0 $m^3/m^2/d$		2.5 $m^3/m^2/d$	
			VFR	CRR	VFR	CRR	VFR	CRR	VFR	CRR	VFR	CRR
1	1	236	142	129	152	170	132	189	163	203	168	198
2	7	227	120	158	144	151	22	115	117	196	122	176
3	13	230	50	142	48	161	62	173	83	189	100	164
4	19	243	48	159	72	159	115	171	95	174	96	151
5	25	220	50	129	65	97	101	157	77	184	82	125
6	31	219	80	115	50	124	26	123	71	129	70	121
7	37	238	78	110	137	149	102	129	65	170	110	156
8	43	298	54	101	93	77	22	164	66	117	77	121
9	49	294	50	154	51	112	16	112	72	123	63	116
10	55	271	57	114	108	106	17	161	58	165	71	142
11	61	254	135	109	31	148	67	176	56	140	90	119
12	67	231	86	163	13	136	90	138	64	119	81	139
13	73	242	88	136	30	167	23	143	56	109	82	128
14	90	222	58	89	72	156	24	168	64	126	69	112
Average		244.6	78.3	129.1	76.1	136.6	58.5	151.4	79.1	153.1	92	140.6

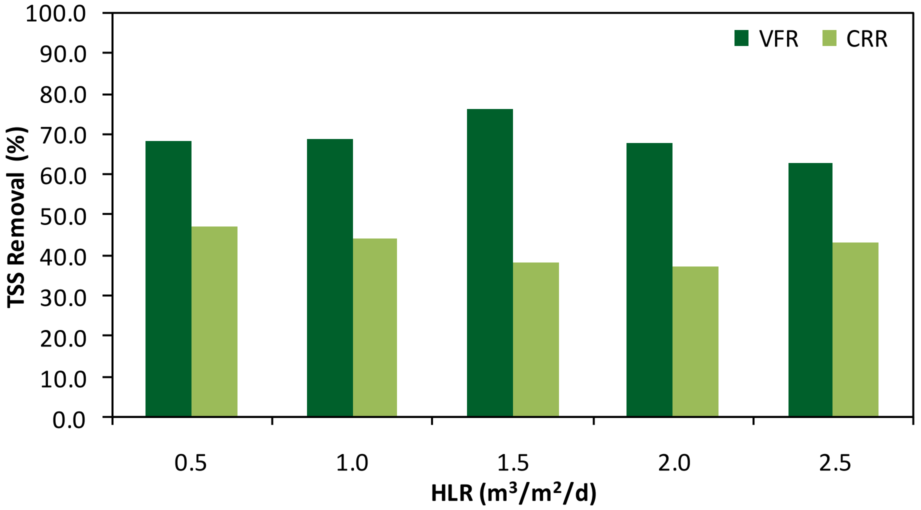

Fig. 6.3: Average TSS removal at different HLRs in VFR and CRR

The removal of TSS could be due to various physical, chemical and biological reactions during vermifiltration process including the adsorption of molecules and ions, oxidation-reduction of organic matter, the behaviour of earthworms and their synergetic effects with microorganisms (Bouché and Soto, 2004). At HLR 1.0 $m^3m^{-2}d^{-1}$, vermifilter showed highest efficacy as compared to geofilter. Besides, When the HLR increased upto 2.5 $m^3m^{-2}d^{-1}$ no significant reduction was observed for TSS as compared to the other HLRs and could be attributed to the lack of availability of time to degrade the solids during vermifiltration process but removal was more than the geofilter. The study conducted by Sinha et al., (2008) demonstrated that during vermifiltration the organic and inorganic solid particles available in wastewater are ingested through earthworms which excreted as finer particles. These finer particles are further trapped in the voids of vermifilter and causes high removal efficiency of TSS.

6.3 Growth of Earthworm Biomass at Different HLRs

The changes in earthworm's biomass in vermifiltration process at different HLRs are illustrated in Fig. 6.4. At the end of the run, the earthworm biomass increased significantly with increase in HLR up to 1.0 m^3m^{-2} d^{-1}, above which the biomass gets reduced. This could be due to the high flow rate and high moisture content affecting the performance of earthworms in the vermifilter bed (Kumar et al., 2014). The increase in weight of earthworm biomass during the vermifiltration period varied between 9.8 and 24.7%.

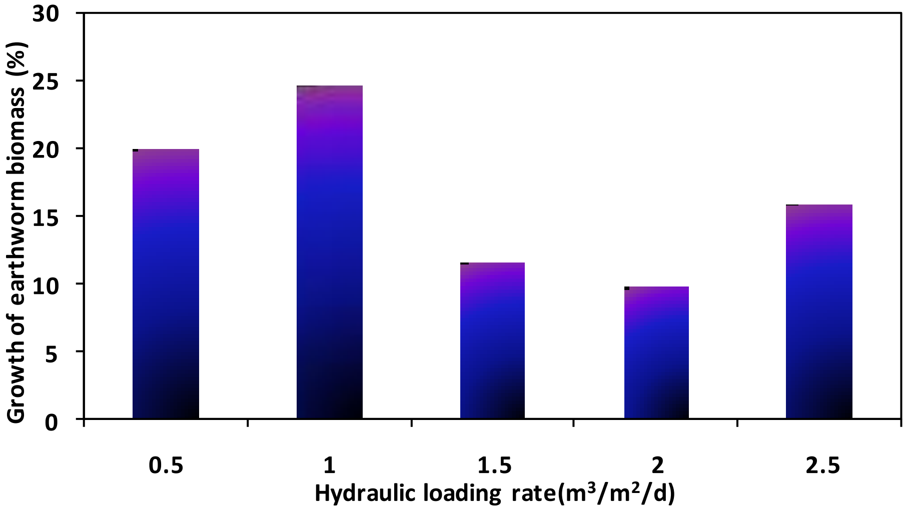

Fig. 6.4: Earthworm biomass growth at different HLRs

Overall, maximum growth of the earthworm biomass was observed at HLR 1.0 $m^3m^{-2}d^{-1}$, which proves the better performance of river bed material as a filtering material (as evidenced in Chapter 5 also) in vermifiltration process and existence of relatively batter HLR between 1.0 and 1.5 $m^3m^{-2}d^{-1}$.

6.4 Comparison of Hydraulic Aspects of Vermifilter and Geofilter

The measurement of hydraulic conductivity was carried out using constant head permeability test according to ASTM D2434 (ASTM 2006). A typical set-up of constant head permeameter is shown in Fig. 3.6 as discussed in Section 3.5.3. It is based on the measurement of the quantity of water that flows under a given hydraulic gradient through a sample of known length and cross-sectional area in a given time. During measurement the water was allowed to flow through the initial and final vermicompost sample from a reservoir designed to keep the water level constant by overflow. The flow of water was allowed to continue till steady state of flow gets established, as evidenced by constant level in the manometer tube. Then the discharge was measured.

Table 6.4 shows the hydraulic conductivity pattern at different HLRs in VFR and CRR. From Table 6.4 it can be seen that a decreasing pattern was reported in both the cases of VFR and CRR as HLR was increased. It was observed as 0.0048, 0.0048, 0.0047, 0.0044 and 0.004 cm/sec for VFR while in CRR it was depicted as 0.0039, 0.003, 0.002, 0.0016 and 0.0012 cm/sec at different HLRs i.e. 0.5, 1.5, 2.0 and 2.5 $m^3m^{-2}d^{-1}$, respectively.

Table 6.4: Hydraulic conductivity pattern at different HLRs and comparison with geofilter

Reactor	Hydraulic Conductivity at different HLRs, (cm/sec)					
	Initial	0.5 $m^3/m^2/d$	1.0 $m^3/m^2/d$	1.5 $m^3/m^2/d$	2.0 $m^3/m^2/d$	2.5 $m^3/m^2/d$
VFR	0.005	0.0048	0.0048	0.0047	0.0044	0.004
CRR	0.005	0.0039	0.003	0.002	0.0016	0.0012

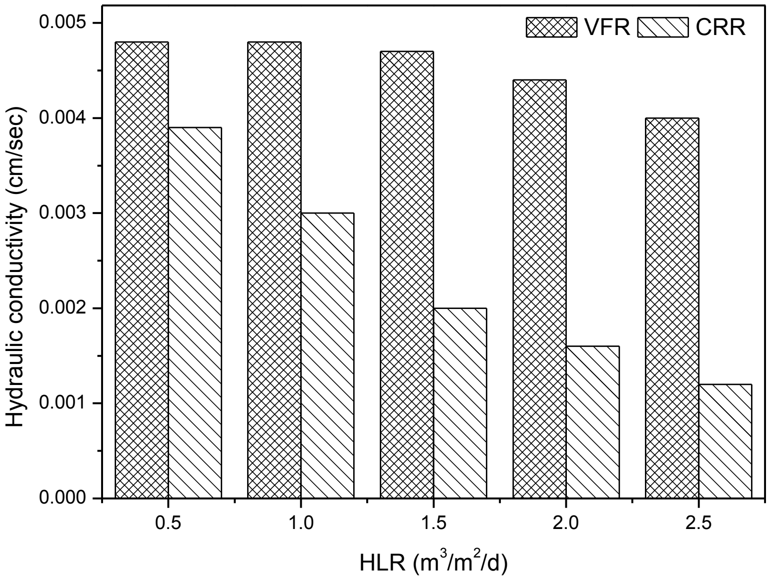

Fig. 6.5: Hydraulic conductivity pattern at different HLRs

Fig. 6.5 shows the variation of hydraulic conductivity in VFR and CRR at different hydraulic loading rates which indicates that hydraulic conductivity decreased from its initial value of 0.005 cm/sec, as taken when the vermicompost was placed in both the reactors. However, it was more than that a conventional geofilter due to channelization in vermifilter through earthworms. Earthworms ingested each and every thing which comes along their way. Further, they ingest bigger particles and secrete them in finer particles which fill the voids of the VFR. This is the basic reason due to which hydraulic conductivity decreased from its initial value of 0.005 cm/sec. On the other hand, in CRR due to the absence of earthworms, no channelization is possible. As a result, the hydraulic conductivity will be less than the VFR.

DEVELOPMENT AND DESIGN OF VERMIFILTER FOR THE TREATMENT OF DOMESTIC WASTEWATER

7.1 General

Wastewater characteristics usually fluctuate due to variation in population and operational problems (Orhon et al., 1999; Hammer and Hammer, 2008). This change in characteristics generally referred as hydraulic and shock loads, may be of qualitative and/or quantitative nature (i.e. change in composition, flow rate and concentration etc.). It is important to determine the stability of the treatment system against shock loads conditions, i.e., how a perturbation may destabilize or affect the treatment efficiency of a system (Xing et al., 1997; El Farhan and Shieh, 1999; Leitao et al., 2006). These shock loads can manifest themselves in two ways: either as transient (for few hours) or long term (for few days or a week). To investigate, whether sewage treatment plants can cope up with shock loads or not, stability assessment can be done under short-term hydraulic and organic shock loads (Tandukar et al., 2006; Gopala Krishna et al., 2008; Seetha et al., 2010).

Over the years a number of workers have looked at the effect of organic shock loads on various wastewater treatment systems. Recently ecological wastewater treatment processes are being designed as decentralized wastewater management. One of the bottlenecks is the lack of knowledge about the performance of these ecological systems under severe environmental and operational variations, such as vermifilter (Mahmood et al., 2013). Vermifiltration has been studied extensively due to its effectiveness for removing pollutants in wastewater and its positive effects on the environment. Sinha et al., (2008) reported that vermifiltration with earthworms increase the effectiveness of treatment process, whereas the system without earthworms showed poor performance. Moreover, several proposed solutions for the treatment of diffuse sources of domestic wastewater have been applied to onsite treatment in spacious rural areas including constructed wetlands, soil infiltration trenches, vegetation-based wastewater treatment, and vermifiltration (Cuyk et al., 2001; Ham et al., 2007; Kaoru et al., 2010; Sinha et al., 2008; Sharma and Kazmi, 2014; Singh et al., 2014). Among these technologies, vermifiltration, a process that separates wastewater solids by allowing it to be gravity-fed over the filtration material, is the most promising economical method for treating point and diffuse sources of domestic wastewater. Despite wide application of vermifiltration, till to date, no study is available regarding performance of vermifilter under shock load conditions.

Besides, degradation kinetics of organic matter was determined using traditional zero order, first-order, and second order models applied for organic matter removal in wetlands in USA, Australia and European countries (Stein et al., 2006; Mitchell and McNevin, 2001). Within this context, the experimental protocol of this work was planned, attempting to assess the effect of organic shock loads on efficiency of pilot scale vermifilter treating synthetic wastewater and in this way contribute to a better insight into the behavior of vermifilter with the view for potential full scale application. Bearing in mind such consideration, the main objective of present study was to gain insight about the performance of vermifilter, in particular, to evaluate the system behavior when exposed to increased organic shock load condition. In addition to this an attempt has also been made by taking COD removal kinetics in consideration for stability assessment. Results presented as a part of this study are unique as it provide the first known pilot scale study of vermifiltration reactor treating domestic municipal wastewater under shock loads conditions. Thus, the present chapter is aimed on following aspects:

(i) Comparative study of vermifilter (VFR) and geofilter (Control reactor: CRR).
(ii) Evaluation of the nutrients and pathogen variation during the process and the final effluent.
(iii) Effect of organic shock load during vermifiltration.
(iv) Kinetics in vermifiltration process.

For this purpose, a pilot scale reactor made up of perspex sheet, was set up (as discussed in Chapter 3 in Section 3.5.4). It consisted of filter bed material, wastewater storage tank, wastewater distribution system and effluent collection system. The reactor was 800 mm long and 800 mm wide with a depth of 800 mm, and 650 mm of packed bedding of river bed materials (as it is found relatively better media in Chapter 5). The reactor had a partition wall that divides it in two parts, one part acted as a vermifilter reactor and other was used as geofilter reactor. Hence, each reactor had a dimension of 800 mm long and 400 mm wide with a depth of 800 mm. An empty space or free board of around 150 mm was kept at the top for aeration purpose. The filter bed consisted of 4 layers. The description of filter bed layers is discussed in Table 3.3 as mentioned in Section 3.5.4. Vermifilter was inoculated with 800 numbers (weighed 450 g) of earthworm species *Eisenia fetida* based on 10,000 worms/cum stocking density (as evidenced relatively better stocking density in Chapter 4). A centrifugal pump was installed to collect and transfer the wastewater to the overhead tank. Besides, a constant head tank was also provided that had a provision to maintain a constant flow under gravity so that unnecessary energy cost can be reduced. Overall, the wastewater fed from storage tank to constant head tank and finally to the treatment unit and it passed through different layers in sequence by gravity flow. The systems was continuously fed with synthetic wastewater at hydraulic loading rate of 1.3 $m^{-3}m^{-2}d^{-1}$ (as evidenced in Chapter 6 that observed to be exist between 1.0 and 1.5 $m^{-3}m^{-2}d^{-1}$). Prior to analysis a 20 days acclimatization period were provided to the reactors. During study the atmospheric temperature varied in the range of 20-28 ºC.

7.2 Results and Discussion

The reactor was started up in September 2013 and operated for a period of 90 days. The removal of BOD, COD, TSS, TC, FC, FS and *E. coli* from the designed vermifilter is measured and its performance against geofilter is explained in following sections. In addition to this an attempt has also been made in which the designed vermifilter deals with effect of organic shock load also.

7.2.1 BOD and COD Removal

Fig. 7.1 and 7.2 show the pollutograph of BOD and COD for vermifilter and geofilter. In VFR the average COD removal efficiency was observed as 76% while in CRR it was found as 63%. Similarly, about 85% average BOD removal efficiency was exhibited in VFR while in CRR it was observed to be 71%.

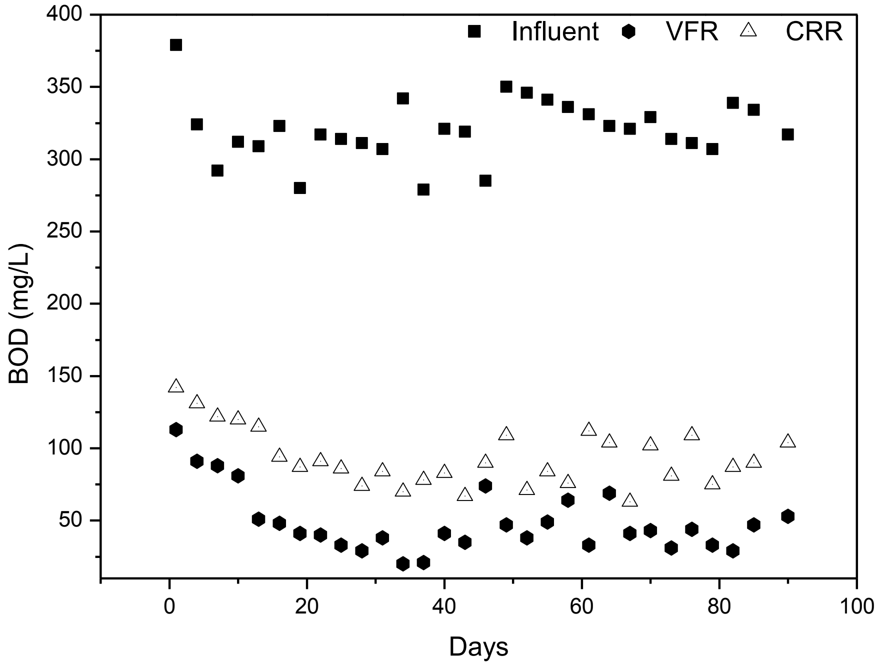

Fig. 7.1: Comparison BOD removal in VFR and CRR

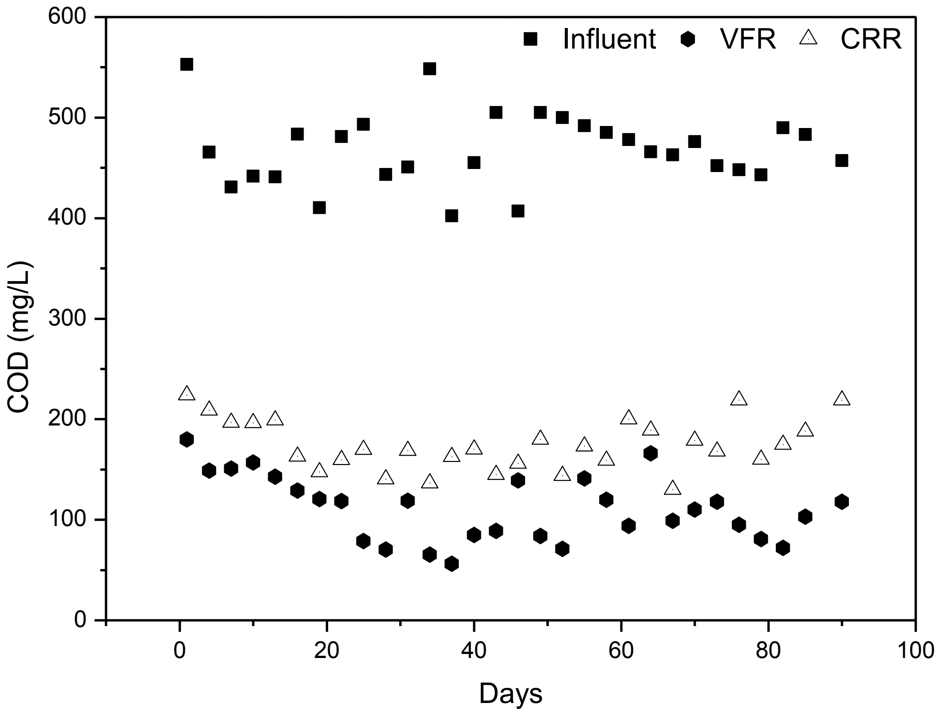

Fig. 7.2: Comparison COD removal in VFR and CRR

A sharp decreasing pattern was exhibited for BOD and COD removal in reactors, VFR and CRR upto 40 days. Between 40 to 80 days, a tortuous behavior BOD and COD was observed in both the cases for VFR and CRR. This unfavorable performance might be due to turbulence, which led to washing out of the influent solids and settled biomass as evidenced in Section 7.2.2.

7.2.2 TDS and TSS Removal

Fig. 7.3 and 7.4 shows the TDS and TSS pattern with time. The concentration of both TSS and TDS were reduced during vermifiltration significantly. For VFR maximum removal of TDS about 54% was observed while for CRR, it followed as 36%. Besides, maximum TSS removal for VFR reactor was observed by 75% while in VFM reactor, minimum removal was recorded as 55% at the end of the run. In VFR the removal of TSS was observed to be high as compared to the removal of TSS. In literature also, it has been evidenced that vermifiltration is effective for removing insoluble organic matter and suspended solid (Wang et al., 2010). During vermifiltration, TSS and TDS removal could be attributed to the ingestion of organic and inorganic solid particles in wastewater through earthworm which excrete them as finer particles. In literature, 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). The various physical, chemical and biological reactions take place during vermifiltration process including the adsorption of molecules and ions, oxidation–reduction of organic matter, the behavior of earth- worms and their synergetic effects with microorganisms (Bouché and Soto, 2004).

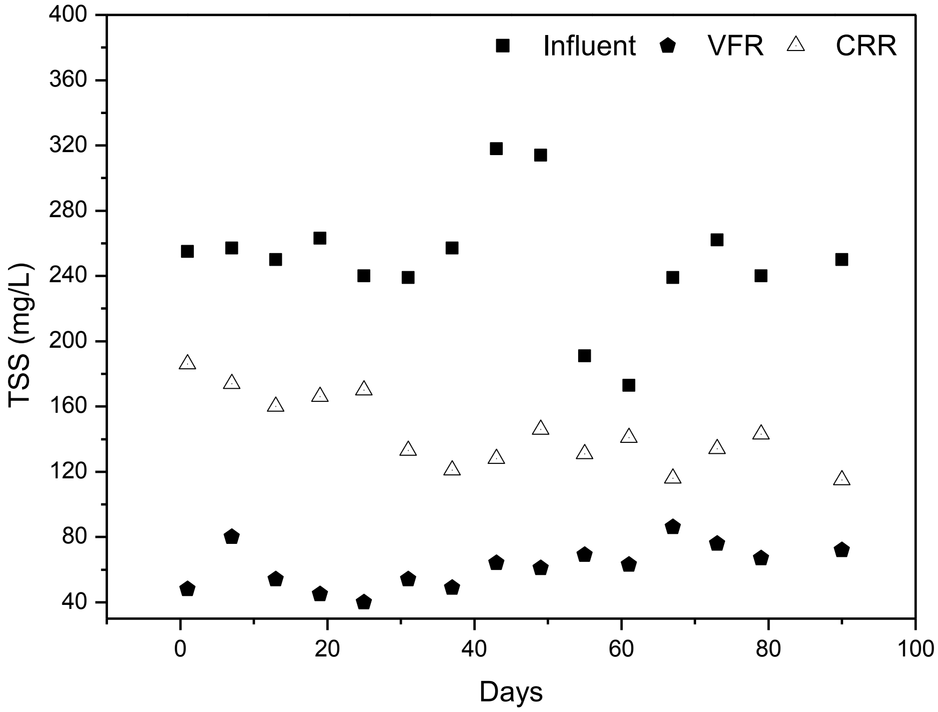

Fig. 7.3: Comparison TSS removal in VFR and CRR

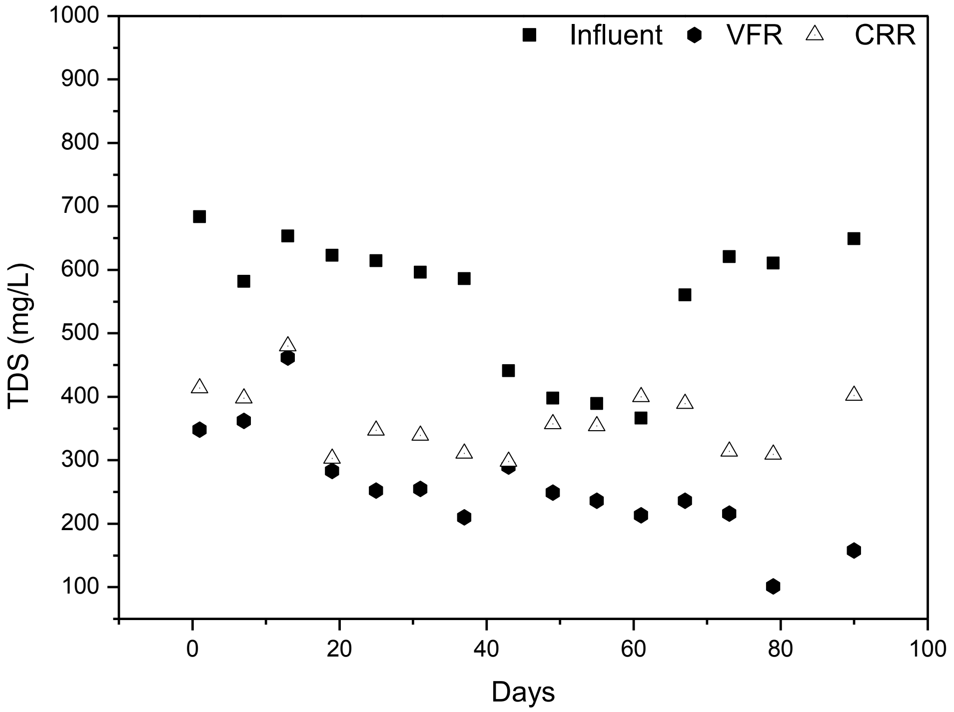

Fig. 7.4: Comparison TSS removal in VFR and CRR

During vermifiltration process, a tortuous behavior for TSS concentration was also observed. This unfavorable performance might be due to turbulence, which led to washing out of the influent solids and settled biomass (Sharma et al., 2014). However in geofilter this phenomenon was also experienced but it was lesser as compared to vermifilter. The stimulatory effect of earthworms could also be a reason behind this tortuous behavior in which mucus and cast are produced. The mucus is a source of easily assimilable carbon for microorganisms, while casts are often enriched with available forms of C, N and P (Aira et al., 2007). Hence, due to leaching of vermicast this type of behavior has been experienced. Further, the adsorption of the impurities in the wastewater will not fully adsorb on the sand and gravel particles and they will be washed away from the reactor (Manyuchi et al., 2013).

The DO concentration in vermifilter varied in the range of 5.3 to 6.8 mg/L while in geofilter it was observed to be varying in the range of 1.3 to 3.5 mg/L as compared to the influent DO concentration of 1.8-4.6 mg/L. The increased DO concentration may be attributed to aerobic conditions created by earthworms in vermifilter bed and waste materials, through their burrowing actions, inhibiting the action of anaerobic microorganisms (Sinha et al., 2008). When earthworms continuously moved in the system it results in channeling of the system through their burrowing action and favoured an aerobic environment.

7.2.3 Removal of Indicator Organisms

Table 7.1 shows the overall performance of vermifilter and geofilter related with removal of indicator organisms. The results of three month study revealed that the presence of earthworms in VFR could efficiently remove TC, FC, FS and *E. coli* and the effluent concentration were observed as $2.89\times10^2 \pm 1.14\times10^2$, $1.38\times10^2 \pm 1.11\times10^2$, $1.86\times10^1 \pm 0.56\times10^1$ and $9.26\times10^1 \pm 0.85\times10^1$, respectively. While in geofilter it was observed as $5.30\times10^3 \pm 0.91\times10^3$, $4.30\times10^3 \pm 2.32\times10^3$, $7.31\times10^2 \pm 1.76\times10^2$ and $1.90\times10^3 \pm 9.48\times10^2$, respectively. This implies that VFR is able to bring indicator organisms to levels considered safe for irrigation as compared to conventional geofilter.

Table 7.1: Microbial quality of effluent from vermifilter and geofilter (mean ± Standard Deviation)

Parameter	TC (MPN/100mL)		FC (MPN/100mL)		FS (MPN/100mL)		*E. coli* (CFU/100mL)	
	Mean	Removal (log unit)	Mean	Removal (log unit)	Mean	Removal (log unit)	Mean	Removal (log unit)
Influent	$3.16\times10^{5}\pm1.58\times10^{4}$	-	$7.94\times10^{4}\pm1.26\times10^{3}$	-	$7.94\times10^{3}\pm1.58\times10^{3}$	-	$1.26\times10^{4}\pm6.31\times10^{3}$	-
VFR	$2.89\times10^{2}\pm1.14\times10^{2}$	3.68±0.86	$1.38\times10^{2}\pm1.11\times10^{2}$	3.1±0.56	$1.86\times10^{1}\pm0.56\times10^{1}$	2.76±0.47	$9.26\times10^{1}\pm0.85\times10^{1}$	2.17±0.31
CRR	$5.30\times10^{3}\pm0.91\times10^{3}$	1.24±0.35	$4.30\times10^{3}\pm2.32\times10^{3}$	1.18±0.31	$7.31\times10^{2}\pm1.76\times10^{2}$	1.01±0.09	$1.90\times10^{3}\pm9.48\times10^{2}$	1.12±0.14

7.3 Effect of Organic Shock Load during Vermifiltration Process

The system was continuously fed with synthetic wastewater at hydraulic loading rate of 1.3 $m^{-3}m^{-2}d^{-1}$ (as evidenced in Chapter 6 that observed to be exist between 1.0 and 1.5 $m^{-3}m^{-2}$ d^{-1}). It corresponded to a hydraulic retention time (HRT) of 3 h till the system achieved steady-state condition. Pseudo steady-state condition (PSS) was assumed to be achieved when the variation in effluent COD concentration was found to be insignificant as depicted in Table 7.2.

Table 7.2: Reactor performance at PSS

Parameter	Influent	Vermifilter effluent
COD (mg/L)	400-600	98-169
NH_4^+-N (mg/L)	29.9-47.1	3-8
NO_3-N (mg/L)	0	21-43
pH	7.2-7.6	7.3-7.8
COD removal efficiency (%)	-	63-76
Nitrification efficiency (%)	-	70-91

After this, the effect of organic shock load on the performance and stability of the system was evaluated to identify its feasibility in actual treatment conditions. For this, a transient organic shock load conditions created by varying the influent concentration (COD_{Inf}) in the range 675-1410 mg/L at constant HRT over a period of time until the reactor had reached steady state. The influent COD concentration during shock loads was taken 1.5 to 3 times of normal feed concentration as represented in Table 7.3.

Table 7.3: Influent COD and OLR at various shock loads

S. No.	Influent COD (mg/L)	OLR (kg COD/m^2/d)	COD_{max} Removal (%)	Time to regain PSS condition
1	675	0.88	66	2hr 15 min
2	799	1.04	71	5 hr 45 min
3	1084	1.41	67	8 hr 45 min
4	1410	1.83	68	12 hr

Each shock load was applied for a period of 1 HRT. Normal loading conditions were resumed at the end of each shock load run.

Substrate loading rates: The substrate loading rates were determined by following equation

$$L = HLR \times C_{sub} / (1000 mg / L \times 1 m^3 / kg) \tag{7.1}$$

Where, L is the substrate loading rates (kg/m^2/d), HLR the hydraulic loading rate (m^3/m^2/d) and C_{sub} is the substrate concentration (mg COD/L).

7.3.1 Performance of Pilot-Scale Vermifilter against Different Short-Term Organic Shock Loads

At PSS, COD removal efficiency was observed in the range of 63-76% as discussed in Section 7.2.1. It may be attributed to the symbiotic activity of earthworms and aerobic microbes which accelerate and enhance the decomposition of organic matter (Loehr et al., 1988; Kumar et al., 2014) and shows the efficacy of vermifiltration process. Fig. 7.5 shows the influent and effluent COD variation under various shock loads in vermifilter. In all the cases, the effluent COD concentration from vermifilter increased in duration of organic shock loads. During different shock loadings of 675, 799, 1084 and 1410 mg COD/L the performance of vermifilter in terms of COD removal efficiency observed as 66, 71, 67, and 68 %. This unfavorable performance might be due to high impact loading, which led to stress increase on earthworm species (Sharma et al., 2014). However, the vermifilter demonstrated appreciable tolerance against organic shock loads with the pollutant removal efficiency dropping down slightly. The system exhibited appreciable stability because the configuration was strong enough for minimizing the effect of organic shock load. The resilience displayed by the system could be attributed to the availability of earthworms that make it capable to overcome the effect of high concentration of pollutant available in wastewater. Visvanathan et al., (2005) has also reported that *Eisenia fetida* can consume organic matter at the rate equal to their body weight every day.

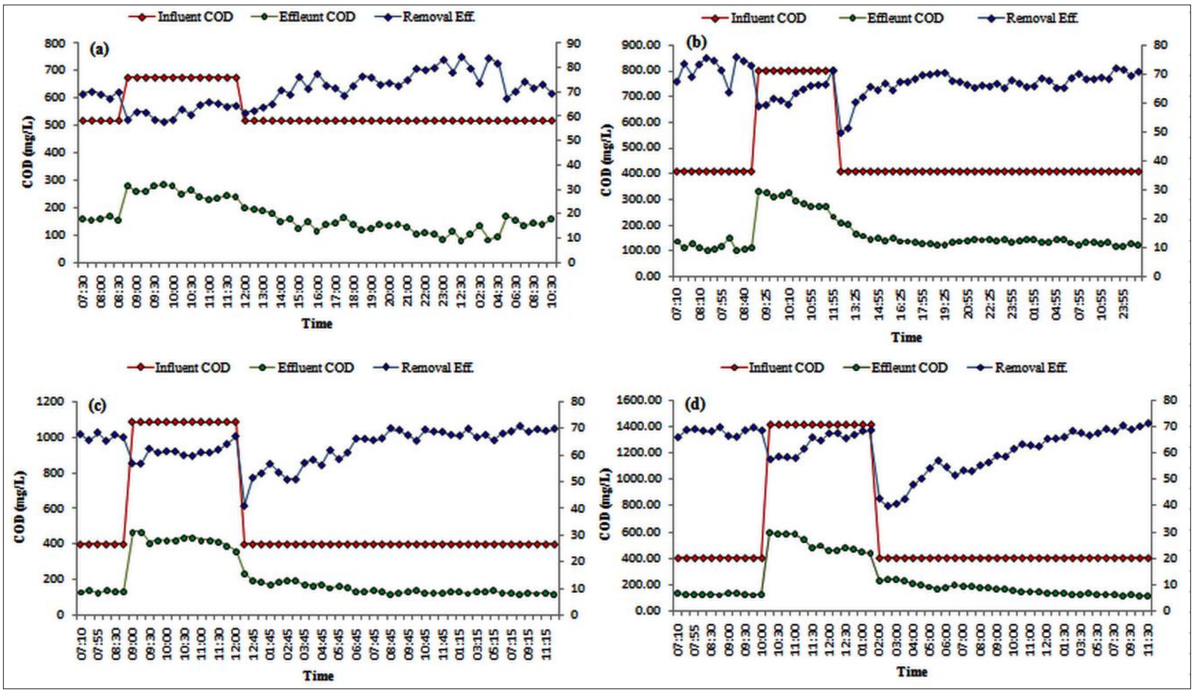

Fig. 7.5: Variation of COD in effluent of vermifilter at different organic shock loads (a) 675,(b) 799, (c) 1084 and (d) 1410 mg COD/L.

In vermifilter, the nitrate nitrogen concentration was observed as 19-32 mg/L at PSS condition. The nitrification efficiency through vermifiltration process exhibited as 66-78%. This can be attributed to mineralization of ammonia nitrogen into nitrate form. Bajsa et al., (2003) has reported that earthworms secrete polysaccharides, proteins and other nitrogenous compounds. In addition to this, the earthworms mineralize the nitrogen in the sewage to make it available to plants as nutrients. Recently, Wang et al., (2011) has also investigated that oxygen is available in abundance through the burrowing action of earthworms which favors a microenvironment for aerobic nitrobacteria. The ammonia nitrogen is removed through rapid adsorption by media and subsequently it is converted from ammonia nitrogen into nitrate form through biological nitrification (Kadam et al., 2009). During shock loading conditions nitrate nitrogen concentrations were slightly changed and quantified as 16-26.8 mg/L.

In vermifilter, the increased concentrations of total phosphate were observed and quantified as 10.6-12.3 mg/L from its initial influent concentration 5.6-6.8 mg/L. This augmentation is attributed to the enzymatic and microbial action of earthworms. Activities of earthworm and associated microbes in vermifilter bed promote rapid phosphate mineralization in the system causes increased concentration of phosphate in the effluent (Kumar et al., 2014). Study conducted by Lee, (1992) evidenced that when organic matter pass through the gut of earthworm then some amount of phosphorus is being converted with more availability to the plants. The researcher have also demonstrated that the release of phosphorus in available form is performed partly by earthworm gut phosphatases and further release of P might be attributed to the P-solubilizing microorganisms present in worm casts (Suthar and Singh, 2008; Kumar et al., 2015). During shock loading condition phosphate mineralization identified as slightly lesser and TP concentration remained in the range of 8-10.4 mg/L.

7.4 Kinetics of Vermifilter

Kinetic study for removal of COD in vermifilter has been performed at fixed influent concentration i.e. 500 mg/L and at fixed HRT of 3 h. Data obtained from experimental evaluation, as illustrated in Table 7.4, were fitted with various traditional zero order, first-order, and second order models applied for organic matter removal.

Table 7.4: Effluent COD at different depth

Time (min)	Depth (mm)	Effluent COD at different depth (C_0)
0	200	500
32	150	340
78	150	199
137	150	175
180	150	95

Different plots of kinetic studies for COD removal are shown in Fig. 7.6. From the results, it could be interpreted that the biological degradation of organics follows first-order kinetics (Fig. 7.6) with respect to initial COD concentration.

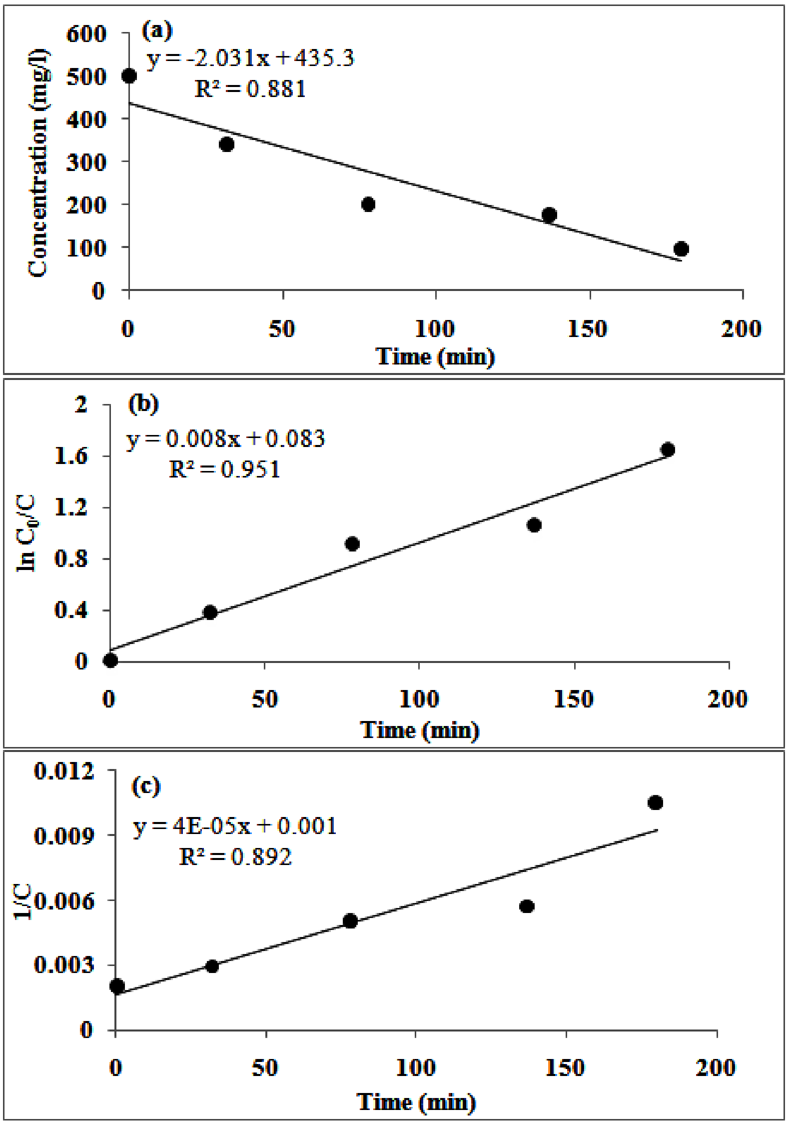

Fig. 7.6: Kinetics for COD removal a) zero order, b) first order, c) second order (Influent COD = 500 mg/L; HRT = 3 h)

The rate constant (k) for different orders of the COD removal was calculated from the slopes of the linear plots and presented in Table 7.5.

Table 7.5: Kinetic models used for COD removal in vermifiltration process

Kinetic model	Equation for linear fit*	Plot	Rate constant (k)	Correlation coefficient (R^2)
Zero order	$C = C_0 - kt$	C vs t	2.0316	0.881
First-order	$\ln C = \ln C_0 - kt$	$\ln (C_0/C)$ vs t	0.0084	0.951
Second-order	$(1/C) = (1/C_0) + kt$	(1/C) vs t	0.00004	0.892

***C_0= conc. at zero time; C = conc. at time t; k = rate constant; t = time**

The R^2, which is an index of the goodness - of - fit, was found highest for the first order model with a value of 0.951. From the correlation coefficient, it is assumed that the COD removal follows first-order kinetics better compared to others under the described experimental conditions (Singh et al., 2013; Singh et al., 2014).

7.5 Quality of Vermicompost

The initial and final characteristic of vermicompost in vermifilter is illustrated in Table 7.6. In vermifilter the end product as vermicompost, collected from the top layer of vermifilter, was dark brown in color and C/N ratio quantified as 9.47-10.5 which revealed stability of waste that deposited after vermifiltration process. However, the value of TOC slightly increased due to deposition of organic matter on the top surface which finally augmented the ash content and identified as 53-56% from its initial value of 49-51%. The TN and TP increased in final vermicompost found on top layer and observed as 30-32.2 and 28-30 g/Kg at the end of the run due to the activity of earthworm. The value of pH in final vermicompost varied in the range 7.4-7.8 and considered as almost in neutral condition.

Table 7.6: Characteristics of vermicompost in vermifilter

Parameter	Initial characteristics of vermicompost	Final characteristics of vermicompost
pH	7.3-7.6	7.4-7.8
C/N ratio	10.57-10.76	9.47-10.5
TOC (g/Kg)	282-296	294-305
Total nitrogen (g/Kg)	26.2-28	30-32.2
Total Phosphate (g/Kg)	23.4-25	28-30
Ash content (%)	49-51	53-56

7.6 Earthworm’s Growth and Reproduction

During vermifiltration process, the earthworm biomass plays an important role for evaluating its long term performance. At the end of the experiment, the number of earthworms was found as 986 from its initial value of 800 (about 450 gm), while the weight of earthworms was observed as 549 gm that results about 23.2% increase in earthworm biomass on number basis while on weight basis it could get as 22%. This may be attributed to continuous substrate (organic matter) availability to the earthworms and better environmental conditions for their propagation inside vermifilter.

The results indicates that the designed system is highly resilient to the organic shock loads up to thrice the average organic loading, as there was no significant change noted on the performance of the system during these shock loading conditions.

The results of the COD kinetics suggested compliance of the well-known first order model with organic matter degradation in vermifilter.

The results obtained in this study indicated that vermifilter employing *Eisenia fetida* help to guarantee the organics removal efficiency and stability of vermifilter, subjected to organic shock loads. This study also promotes an understanding of organic matter removal in the system and experimental results can be used for estimating treatment efficiency of full-scale reactors under similar operational conditions.

CONCLUSIONS AND RECOMMENDATIONS

8.1 General

In the present study several laboratory experiments were conducted to evaluate the effect of stocking density (varied from 5000 to 30000 worms/cum of vermifilter bed), filter media and hydraulic loading rate (varied from 0.5 to 2.5 $m^3m^{-2}d^{-1}$) on vermifiltration process. Four types of media were used for experimentation, i.e., (a) River bed material, (b) Wood coal, (c) Glass balls and (d) Mud balls. Besides, the vermifilter was evaluated against varying degree of organic shock loads 675-1410 mg/L of COD. The main conclusions inferred from the study are presented below.

8.2 Effect of earthworm's stocking density on vermifiltration

- The stocking density of 10000 worms/cum of vermifilter bed was found to be relatively better when compared with other stocking densities of 5000, 15000, 20000, 25000 and 30000 worms/cum, to achieve the requisite quality of effluent.
- The results showed that effluent quality of vermifilter with 10,000 W/cum showed highest percentage removal of BOD (90%), COD (68%), TSS (70%). The average reduction of the population of indicator organisms i.e. TC, FC, FS and *E. coli* was observed as 3.61 ± 0.90, 3.14 ± 0.67, 2.73 ± 0.37 and 2.27 ± 0.28 log unit, respectively to the levels considered acceptable for either recreation or irrigation.
- The growth pattern of earthworm species, *E. fetida*, showed a maximum individual biomass (121.4%) and growth rate (0.94 g wt. $worm^{-1}$ day^{-1}) in earthworm's stocking density of 10000 worms/cum.

8.3 Suitability of different materials as vermifilter bed

- Vermifilter was evaluated using different material as a media like river bed material, wood coal, glass balls and mud balls. The river bed material was found to be relatively better as a media in vermifilter for better growth of earthworm biomass and to achieve the requisite quality of effluent.
- The results demonstrated that effluent quality of vermifilter having river bed material showed highest percentage removal of BOD (78%), COD (71%) and TSS (73%).

- The average reduction of the population of indicator organisms i.e. TC, FC, FS and *E. coli* was observed as 3.6 ± 0.90, 3.4 ± 0.67, 2.5 ± 0.51and 3.32 ± 0.62 log unit, respectively.
- In reactor VFR, having river bed material, maximum earthworm biomass was observed with 73% increment.

8.4 Effect of hydraulic loading rate on the performance of vermifilter

- When the comparison was carried out among different HLRs like 0.5, 1.0, 1.5, 2 and 2.5 $m^3m^{-2}d^{-1}$ the optimum hydraulic loading rate was observed to vary in the range of 1-1.5 $m^3m^{-2}d^{-1}$.
- Vermifiltration system represented better efficacy when it was compared with conventional geofilter with better quality of effluent. It was observed to be having higher hydraulic conductivity as compared to geofilter.
- At HLR of 1.0 $m^3m^{-2}d^{-1}$, the percentage removal of various pollutants i.e. BOD, COD and TSS were observed as 86.9, 79.6 and 68.9%, respectively. At HLR of 1.5 $m^3m^{-2}d^{-1}$ it was resulted as 85.5, 73.9 and 76.1%, respectively.
- It was observed that earthworm biomass increases upto HLR of 1.0 m^3m^{-2} d^{-1} and decreases at higher HLRs.
- Comparative study on hydraulic aspects of vermifilter and geofilter at different hydraulic loading rates indicated that hydraulic conductivity decreased from its initial value of 0.005 cm/sec, as taken when the vermicompost was placed in both the reactors. However, it was more than that a conventional geofilter due to channelization in vermifilter through earthworms.

8.5 Development and design of vermifilter for the treatment of domestic wastewater

- A comparison was carried out between vermifilter and geofilter that was operated on the basis of outcomes coming out from above results. The results demontrated that vermifilter removes a considerable amount of BOD, COD, TSS and indicator organisms as compared to conventional geofilter.
- In vermifilter the average COD removal efficiency was observed as 76% while in geofilter it was found as 63%. Similarly, about 85% average BOD removal efficiency was exhibited in vermifilter while in geofilter it was observed to be 71%.

- The results of three month study revealed that the presence of earthworms in vermifilter could efficiently remove TC, FC, FS and *E. coli* and the effluent concentration were observed as $2.89\times10^2 \pm 1.14\times10^2$, $1.38\times10^2 \pm 1.11\times10^2$, $1.86\times10^1 \pm 0.56\times10^1$ and $9.26\times10^1 \pm 0.85\times10^1$, respectively. While in geofilter it was observed as $5.30\times10^3 \pm 0.91\times10^3$, $4.30\times10^3 \pm 2.32\times10^3$, $7.31\times10^2 \pm 1.76\times10^2$ and $1.90\times10^3 \pm 9.48\times10^2$, respectively. This implies that vermifilter is able to bring indicator organisms to levels considered to be safe for irrigation as compared to conventional geofilter.
- When organic shock loads on vermifilter was applied by increasing the influent COD concentration from 1.5 to 3 times of normal values, the system recovered quickly. It proves its resilience power against organic shock loads, as no significant changes was noted on the performance during vermifiltration process.
- Results obtained in this study indicated that vermifilter employing *Eisenia Fetida* help to guarantee the organics removal efficiency and stability of vermifilter if subjected to organic shock loads. This study also promotes an understanding of organic matter removal in the system and experimental results can be used for estimating treatment efficiency of full-scale reactors under similar operational conditions.
- Kinetic study revealed that the biological degradation of organics follows first-order kinetics with respect to initial sCOD concentration during vermifiltration process.
- The treated effluent and final vermicompost both were found to be rich in nitrate and phosphate which can be used for agriculture purpose.

List of Related Publications

1. **Kumar, T.,** Bhargava, R., Prasad, K.S.H., Pruthi, V. (2015). Evaluation of vermifiltration process using natural ingredients for effective wastewater treatment. Ecol. Eng. 75, 370-377.
2. **Kumar, T.,** Rajpal, A., Bhargava, R., Prasad, K.S.H. (2014). Performance evaluation of vermifilter at different hydraulic loading rate using river bed material. Ecol. Eng. 62, 77-82.
3. **Kumar, T.**, Rajpal A., Arora, S., Bhargava R., Prasad, K.S.H., “A comparative study on vermifiltration using epigeic earthworm *Eisenia fetida* and *Eudrilus eugeniae*”. Desalination and Water Treatment. (Accepted for Publication)
   DOI: 10.1080/19443994.2015.1010230
4. Arora, S., Rajpal A., **Kumar, T.**, Bhargava R., Kazmi, A.A., 2014 “Pathogen removal during wastewater treatment by vermifiltration”. Environ. Technol. 35(19). 2493-2499.
5. Bhargava, R., Verma, J., Hari Prasad, K.S., **Kumar, T.**, (2012), “Decentralized waste water treatment by vermifiltration using river bed material”, International conference of sustainable built environment, Sri Lanka. (Published)
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