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#### Review article

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### New developments on vermifiltration as a bio-ecological wastewater treatment technology: Mechanism, application, performance, modelling, optimization, and sustainability

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#### ABSTRACT

The review discusses the advancements in vermifiltration research over the last decade, focusing on pollution removal mechanisms, system performance, the fate of filter components, and byproducts. Vermifiltration has demonstrated remarkable capabilities, particularly in treating highly contaminated wastewater with Chemical Oxygen Demand (COD) levels exceeding 92,000 mg/L and Biochemical Oxygen Demand (BOD<sub>5</sub>) levels over 25,000 mg/L, achieving removal rates of approximately 89% and 91%, respectively. Importantly, vermifiltration maintains its effectiveness even with fluctuating organic loads at the inlet, thanks to optimization of parameters like Hydraulic Loading Rate, biodegradable organic strength, earthworm density and active layer depth. Clogging issues can be minimized through parameters optimization. The review also highlights vermifiltrations' potential in co-treating the organic fraction of municipal solid waste while significantly reducing heavy metal concentrations, including Cd, Ni, Pb, Cu, Cr, and Zn, during the treatment process. Earthworms play a pivotal role in the removal of various components, with impressive removal percentages, such as 75% for Total Organic Carbon (TOC), 86% for Total COD, 87% for BOD5, 59% for ammonia nitrogen, and 99.9% for coliforms. Furthermore, vermifiltration-treated effluents can be readily utilized in agriculture, with the added benefit of producing vermicompost, a nutrient-rich biofertilizer. The technology contributes to environmental sustainability, as it helps reduce greenhouse gas emissions (GHG), thanks to earthworm activity creating an aerobic environment, minimizing GHG production compared to other wastewater treatment methods. In terms of pollutant degradation modeling, the Stover-Kincannon model outperforms the first-order and Grau second-order models, with higher regression coefficients ( $R^2 = 0.9961$  for COD and  $R^2 = 0.9353$  for TN). Overall, vermifiltration emerges as an effective and sustainable wastewater treatment solution, capable of handling challenging wastewater sources, while also producing valuable by-products and minimizing environmental impacts.

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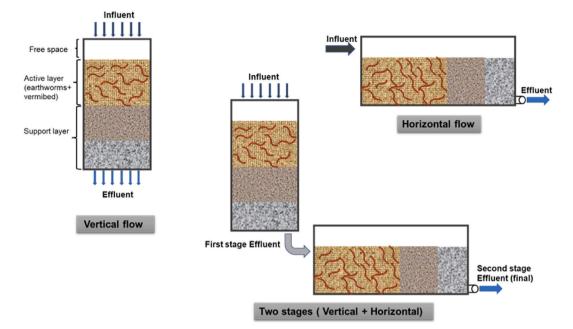


Fig. 1. Schematic diagrams of different types of vermifilter configurations.

#### 1. Introduction

To address the water scarcity, sanitation, health, and environmental problems created by poor wastewater management, wastewater treatment is considered as a possible alternative. Wastewater is also a source of various valuable nutrients such as nitrogen and phosphorus that, if properly treated, can be a potential resource in agriculture [1]. Intensive wastewater treatment systems, such as activated sludge can be solutions, but small communities cannot access them due to high installation and operation costs [2]. It is assumed that by adapting decentralized wastewater treatment technologies, reliable and effective long-term solutions can be introduced [1].

Over the years, studies have shown that vermifiltration, also called lombrifiltration can be used for domestic and industrial wastewater treatment. It is a good alternative to treatment systems such as activated sludge, especially in terms of energy, operating cost and non-waste generation [3]. During the vermifiltration process, earthworms are used to increase and diversify the microbial communities living in soil-based biofilters [4]. Earthworms have been around for about 600 million years and have adapted to toxicity. They can help clean wastewater by devouring microorganisms [5]. Earthworm-based technologies are self-promoting, self-improving, and self-reinforcing, requiring very little energy and producing no waste. In addition, they are simple to design, use and to maintain. Vermifiltration is a technology that is known to be self-sustaining, natural, and environmentally friendly with an integrated mechanism to remove not only organic matter but also nutrients and pathogens [6]. It can also be used in the treatment and reduction of excess sludge. This technology is easily adaptable in developing countries due to its simplicity and the fact that it treats water to acceptable standards [7]. This review will focus on the recent developments made in the technology.

The main objective of the review is to highlight the latest research in the field regarding the pollution removal mechanism, performance, fate of filter components including earthworms, and system by-products. The manuscript is particular in that it highlights the research methods applied by the different authors to study the vermifiltration mechanism. It also presents the challenges and bottlenecks in the field. Recent studies have addressed the modeling and optimization of filter design and operating parameters. Particular attention is also paid to these optimization solutions, to allow a better choice of the different components of the system, in the implementation. In addition, this paper aims also to highlight vermifiltrations' sustainability features and future research considerations to improve the performance of the vermifiltration system.

#### 2. General information on vermifiltration: definition and mechanism

#### 2.1. Definition and types of vermifilters

Vermifiltration is a wastewater filtration process in which epigeic earthworms interact with microorganisms for the removal of wastewater pollution. It encompasses all forms of treatment, including primary (removal of sand, silt, etc.), secondary (biological degradation), and tertiary (removal of pathogens) in one unit [8]. The wastewater treatment system that is used for vermifiltration is the vermifilter (VF). It includes an active zone in which earthworms live, and a filter bed in which microorganisms grow [9]. It is also called microbial-earthworm ecofilter [10]. Different filters configurations have been implemented in recent years. They are vertical



Fig. 2. Synergy between earthworms and microorganisms in the degradation of OM.

subsurface flow vermifilters (VVFs) horizontal subsurface flow vermifilters (HVFs) and two-stage vermifilters (VVFs + HVFs) (Fig. 1).

#### 2.1.1. Vertical subsurface flow vermifilters

The bed materials are stacked in vertical layers, and more often coarse gravel is placed in the lowest layer as a support [11–13]. The wastewater flows vertically through the filter material and the treated effluent is received at the filter outlet. According to Ilyas and Van Hullebusch [14], the distribution of wastewater over the filter surface is better, allowing a higher oxygenation capacity. This favors the development of aerobic bacteria responsible for the degradation of pollutants. The high oxygenation also allows the complete degradation of organic matter (OM) in the wastewater. Carbon dioxide (CO<sub>2</sub>) is thus released as the only gaseous emission. Environmental sustainability is therefore ensured by negotiating the emissions of greenhouse gases such as methane (CH<sub>4</sub>) and nitrous oxide (N<sub>2</sub>O) [15].

#### 2.1.2. Horizontal subsurface flow and two stages flow vermifilters

In contrast to the VVFs, in the HVFs, the bed materials are stacked horizontally and the gravel layer is located right at the outlet [16]. Here, the wastewater flows horizontally through the different layers of the filter and the treated effluent is received at the outlet. The coexistence of aerobic and anaerobic conditions is higher in this configuration. Due to the combined effect of horizontal subsurface flow and earthworms, nitrification is facilitated in the upper layer due to earthworm activities and denitrification in the anoxic lower layer [17]. According to Singh et al. [16], one advantage of horizontal flows is that they allow a longer contact time compared to deep beds. In the two-stages flow vermifilter, the water is filtered in two stages as the name suggests [18,19]. Generally, the first stage consists of a VVF. The treated effluent from the VVF is reintroduced as influent into the HVF and the final effluent from the system is received at the outlet of the HVF (Fig. 1). The advantages of both VVF and HVF configurations are thus combined here. In addition, the path length of the wastewater through the treatment system is higher, increasing the contact time between earthworms, microorganisms, and pollutants, which increases the treatment efficacy.

To conduct a comprehensive comparative analysis, it is necessary to further explore several factors. These include scalability, costeffectiveness, maintenance requirements, and potential challenges associated with each vermifiltration configuration. Additionally, it is important to evaluate the effectiveness of each configuration in removing specific types of pollutants and assess their suitability for different wastewater treatment scenarios. By considering these aspects, a more thorough and well-rounded comparison can be made between the vermifiltration configurations.

#### 2.2. Mechanism of vermifiltration

#### 2.2.1. Symbiosis earthworms - microorganisms in the degradation of pollutants

According to several studies, earthworms and microorganisms work synergistically to remove pollution from wastewater [20,21]. In that synergy, the microorganisms biochemically degrade the matter while the earthworms by muscular actions also allow a degradation but especially a homogenization of the matter. Through the investigation of bacterial and protein characteristics, it has been demonstrated by Arora et al. [22] that the feeding of earthworms and the interactions between earthworms and micro-organisms are responsible for the removal of pollutants during the vermifiltration process. Indeed, the digestion of organic matter (OM) in the digestive tract of earthworms is carried out within the framework of a mutualism existing between the earthworm and the micro-organisms ingested in the environment. In the digestive tract, the earthworm secretes water and mucus which will reactivate the microorganisms in the environment. Under the action of this « priming effect », the stimulated microorganisms will decompose and mineralize a part of the OM (Fig. 2). The earthworms will then be able to assimilate some of the carbon and nutrients released by the microbial activity. The passage through the gastrointestinal tract of earthworms has a qualitative and quantitative influence on the microbial community of the biofilm in the vermifilter. It is important to understand the synergistic relationship between earthworms and microorganisms for the degradation of pollutants and to highlight the respective actions of each entity on the outcome. The study conducted by Arora et al. [22], reveals that only 40% of the bacterial species that were investigated in the system did not originate from

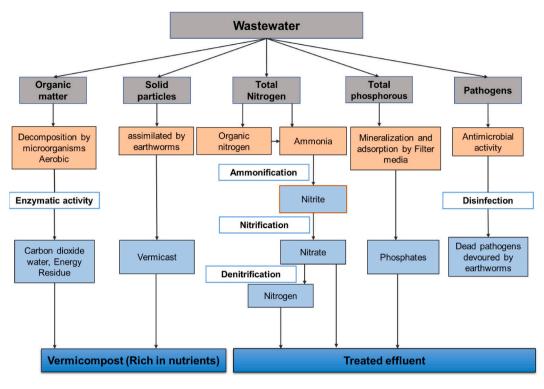


Fig. 3. Schematic diagram of the vermifiltration mechanism.

the worm gut, so less than half. Adugna et al. [23] noted a five-fold increase in the microbial population of the vermifilter compared to the control filter, without the earthworms. It was also observed that there was a more extensive variety within the population present in the vermifilter [24]. The higher the density of earthworms in the system, the greater the diversity of microorganisms [25].

#### 2.2.2. The different stages of degradation are the following

First, the larger organic and inorganic solid particles are broken down by earthworms into finer particles. In this way, the surface area of the particles increases, thus facilitating microbial degradation [26,27]. The finer particles are trapped within the pores of the vermifilter bed media and subsequently undergo degradation by microorganisms [21,23]. Beyond his physical function, the bed media serves as an essential component by offering a habitat conducive to the growth of beneficial microorganisms. Additionally, the media is effective in adsorbing specific contaminants, including heavy metals and pathogens.

Nutrients undergo a kind of recycling and are added to the vermicompost and treated effluent which are the by-products of the vermifiltration mechanism (Fig. 3). The processes of ammonification, nitrification, and denitrification have been listed in the literature as the origin of nitrogen elimination in the vermifilter [26,28,29]. One part of the organic phosphorous is mineralized to inorganic phosphate. The remaining portion is adsorbed by the filtering media.

#### 2.2.3. Role of earthworms in degrading pollutants

The significant role of earthworms in the decomposition of pollutants has been well-established over the years. Chao et al. [30] conducted a comprehensive meta-analysis to assess the extent of earthworms' impact on the breakdown of organic pollutants. The findings from this analysis reveal a statistically significant average effect size, indicating a 128.5% increase in organic pollutant degradation (p < 0.05). The increase in the concentration of dissolved organic matter resulting from earthworm treatment, promotes the proliferation of soil bacteria involved in decomposition [31]. Furthermore, earthworms, by their burrowing activity create galleries i.e., many small holes in the system, thus promoting the development of aerobic bacteria for wastewater treatment [32–34]. The intestinal system of earthworms plays a crucial role in the processes of nutrient solubilization, bioremediation, and the strengthening of advantageous microbial populations. Analysis of microbial diversity reveals the presence of various microorganisms in vermicast, including Azotobacter, Rhizobium, Nitrobacter, nitrogen-fixing bacteria in the system shows a higher concentration of aerobic bacteria than facultative and anaerobic ones [36]. In addition, Arora et al. [32] were able to detect 26 species of bacteria in the VF with *Bacillus* as the dominant genus, compared to only 11 in the filter without earthworms. It is known that *Bacillus* produces metabolites that have antimicrobial and cytotoxic or antifungal properties. Moreover, typical biofilm macrographs and micrographs showed that the feeding and dejections of the earthworms actually changed the bacterial population in the VF. The use of Automatic Testing Bacteriology (ATB) expression and sequencing data also reveals that the presence of earthworms leads to an increase in microbial

diversity and metabolic activities in the biofilm [37]. This observation underscores that the presence of earthworms significantly amplifies the bacterial population within the system. The research of Wang et al. [38] on the biofilms revealed a significantly higher pore number in the VF than in a filter without earthworms or non-vermifilter (NVF). The total organic carbon (TOC) was also higher in the VF. Dehydrogenase activity (DHA) and adenosine triphosphate (ATP) are better in the VF than in the NVF. This activity is improved by more than 16%. The burrowing activities of the earthworms can also increase parameters including the hydraulic conductivity of the system, the specific surface of the substrate, the nutrient exchange rate and the aeration of the system [39]. Another role of earthworms in the vermifilter is the consumption of the bacterial population that competes for substrates with the microorganisms responsible for the degradation of pollutants. Earthworms secrete a viscous fluid that traps these harmful bacteria. The microbes left behind by the earthworms, which are not harmful, also compete with the pathogens for nutrients [6]. It has also been shown that earthworms accumulate heavy metals in their gut, leading to a substantial decrease in pollution levels in the final treatment products [40].

#### 3. Benefits and limitations of vermifiltration

#### 3.1. Benefits

The benefits of vermifiltration are numerous, including.

- Low methane emissions: From the study of Dore et al. [11] it was observed that in the course of the vermifiltration process, methane emissions were significantly diminished in comparison to those from an anaerobic lagoon, showing a remarkable reduction ranging from 97% to 99%. The methane emissions are minimal at about 0.7kgCH<sub>4</sub>/m<sup>2</sup>/year.
- Odor-free process: lombrifiltration systems operate in a predominantly aerobic state, meaning an oxygen-rich environment for earthworms and micro-organisms. This state favors the decomposition of organic matter without generating foul-smelling anaerobic by-products. The system also ensures a continuous flow, avoiding water stagnation which can lead to the build-up of odorous gases [41].
- Little or no sludge production: vermifiltration systems have the advantage of not producing sludge requiring treatment [41,42]. Sludge yield is generally less than 0.2 kg of suspended solids per kg of chemical oxygen demand (COD) [43].
- High performance in pollutant removal: vermifiltration excels in organic matter removal from wastewater due to the earthworms' ability in breaking down and digest organic matter. The treated effluent from the VF can easily be reused in many industrial plants for production processes and various secondary uses. Oils and fats are also removed at a rate of approximately 84–89% and surfactants at a rate of 95–99% [44].
- Low Operating Costs: vermifiltration approach proves cost-effective by taking advantage of natural processes and eliminating the need for substantial energy consumption or expensive equipment. Unlike conventional wastewater treatment methods, vermifiltration is less energy-intensive, making it a sustainable choice [45]. It is possible to have a cost reduction of about 60–70% compared to other systems [46].
- Adaptability and environmentally friendly: vermifiltration is an environmentally-friendly method that promotes the recycling of organic matter while reducing the environmental impact associated with wastewater disposal. Also, vermifiltration can be used flexibly in a variety of environments, including rural areas and decentralized wastewater treatment systems.

#### 3.2. Limitations

- Limited performance in the removal of certain pollutants: for the optimal removal of nitrates and phosphorus, it is necessary to combine other processes or techniques with vermifiltration [12]. Nie et al. [47] also concluded that the VF has a poor performance in the removal of these two elements. For pathogen removal also, additional disinfection steps may be required for safe reuse of wastewater. Furthermore, the concentrations of heavy metals in VF effluent often do not meet acceptable thresholds, especially for industrial waters [48].
- Environmental sensitivity: The effectiveness of vermifiltration is sensitive to environmental conditions, including temperature and humidity levels, which can have an impact on earthworm activity. Temperature in particular, is a critical factor, as earthworms thrive in a specific range, generally between 15 °C and 25 °C, as pointed out by Edwards and Arancon [49]. Earthworms cannot survive at temperatures below 10 °C or above 35 °C, making vermifiltration unviable in such extreme temperature conditions. Some toxic constituents present in the wastewaters to be treated could also be fatal to earthworms.
- Clogging problems: Over time, vermifiltration systems can become clogged with solids and organic matter. Clogging can be triggered by a range of factors including the hydraulic loading rate, precipitation on the bed, salinity, exposure to sunlight, and the characteristics of the filter bed. Clogging can diminish the effectiveness of the vermifiltration system and escalate maintenance expenses [13,50,51].
- Capacity limitations: Lombrifiltration may not be well-suited for handling large volumes of wastewater, making it more appropriate for small-scale applications, as highlighted by Suhaib and Bhunia [51].

Different types of wastewaters treated by vermifiltration.

Type of wastewater	Place of experiment	Influent COD (mg/ L)	COD Removal (%)	Influent BOD (mg/ L)	BOD Removal (%)	References
University campus	India	325-400	92	180-250	98	[22]
Cattle feedlot	USA	300-550	61.4-69.1	-	-	[57]
Clinical Laboratory	India	390-420	75–80	200-250	80-85	[26]
Distillery	Trichy, Inde	54400	90	18100	95	[58]
Domestic greywater	Burkina Faso	1075-3520	83	800-2100	97.6	[23]
Urban (Sewage and rainwater)	-	$450\pm10$	87.6	$210\pm10$	91.3	[59]
Swine wastewater	Portugal	1997	-	149	83	[12]
Slaughterhouse wastewater, synthetic	USA	2100-2400	70–85	-	-	[57]
Synthetic brewery wastewater	-	2250-11250	92–96	-	-	[34]
Side-stream of dairy wastewater	USA	2100-3400	$45\pm4.1$	-	-	[13]
Textile dye effluent	India	5800	85-89	1933	76–80	[60]
Urban sewage	Spain	$706\pm407$	$88\pm7$	$392\pm262$	$85\pm19$	[2]
Domestic septic tank sewage	Zimbabwe	-	_	49.9	68–97	[61]
Hospital wastewater	Iran	227-461	75	145-300	93	[55]
Municipal wastewater	China	240-320	$83.5\pm2.1$	120-200	$81.3 \pm 2.9$	[38]
Dairy wastewater	India	2560	67	_	_	[62]
Domestic wastewater		$92.2\pm18$	67.6	$39.1\pm10.2$	78	[43]
Synthetic Dairy Wastewater	India	$1734 \pm 110$	80.7	$1103.6\pm82$	88.4	[63]
Sewage sludge	China		$53.01 \pm 10.53$	9900-20000	$61.06 \pm 13.87$	[64]
Organized industrial zone			80			[48]

#### 4. Different factors to consider in the implementation of a vermifilter

#### 4.1. Wastewater type and organic loading rate

In the last few years, vermifiltration has been used in the treatment of different types of wastewaters and not only for domestic wastewater as in its early stage of testing. Water from dairies, palm oil processing plants [24], the textile industry [52], piggeries [53], from feedlots, to hospital water, and several other sources (listed in Table 1) have been treated. This showcases the versatility and potential of vermifiltration as a viable option for handling different types of wastewaters. Moreover, the organic loading rate (OLR) admissible by the system, ranges from low levels of the order of 100-200 mg/L of chemical oxygen demand (COD) to higher levels (Table 1). Vermifiltration can be used as a treatment process for highly polluted waters. Manyuchi et al. [54] have worked on distillery waters with COD values of more than 92,000 mg/L and biochemical oxygen demand (BOD) values of more than 25,000 mg/L, with removal rates of about 89% and 91% respectively. Other authors have also had similar results (Table 1). Despite the fluctuation of the organic load at the inlet, the VF can work over long periods [55]. However, wastewaters can have varying characteristics that can affect VF performances. For example, industrial wastewater may contain toxic chemicals that can inhibit earthworm activity, reducing treatment efficiency [48]. Earthworms are well-known for their ability to withstand various types of contaminants, including herbicides, heavy metals, and organic pollutants that often originate from industrial, factory, and hospital wastewater as well as sewage sludge [56]. They possess the capacity to decrease the concentrations and toxicity of wastewater contaminants by storing them in their body tissues [41]. However, when wastewater containing these contaminants is applied, it can result in the accumulation of these substances within the earthworm's body, which in turn could induce physiological and biochemical harm. This can result in growth abnormalities, enzyme inhibition, and toxicity, which may extend to genetic levels [56].

#### 4.2. Hydraulic loading rate (HLR) and hydraulic retention time (HRT)

The hydraulic loading rate (HLR) is the amount of water passing through the filter per unit area and per unit time. It is one of the most important parameters in water filtration. It has been observed that the treatment performance and the clogging of the VF are closely dependent on the administered HLR. The natural activities of earthworms, such as feeding and burrowing, help improve the porosity of the filter bed. However, when the HLR is increased, earthworm activity tends to decrease, which can result in the accumulation of clogging materials in the initial section of the filter bed, leading to severe clogging problems [50,51]. According to Suhaib and Bhunia [65], the instability of earthworm casts within the filter bed at higher HLRs can further reduce the beds' permeability. Moreover, the studies conducted by the same authors [66] revealed that a filter bed at an HLR of  $2.34 \text{ m}^3/\text{m}^2/\text{day}$  causes an increase in organic and suspended solid loads on the filter bed compared to a (lower) HLR of  $0.66 \text{ m}^3/\text{m}^2/\text{day}$ , resulting in significant accumulation of clogging materials like biofilm biomass and suspended solids at the inlet end of the filter bed.

On the other hand, Samal et al. [65] concluded that the performance of the VF decreased with increasing HLR. A too-high HLR decreases the contact time of the wastewater with the filter media, the hydraulic retention time (HRT). The HRT represents the duration of the interaction between the wastewater and the active layer of the VF. It is mainly dependent on the HLR, the porosity of the bedding materials, and the volume of the filter profile. It is a very important parameter because it represents the time that the worms and microorganisms will spend in contact with the wastewater. Indeed, they need enough time to be able to degrade and

HLR an HRT applied in vermifiltration and their impact on removal performance.

Wastewater	HLR (m <sup>3</sup> /	HRT	Removal per	formance (	%)					Reference
	m²/day)	(Hours)	COD	BOD	TSS	TDS	TN	TP	NO <sup>3-</sup>	
University campus	1	4–6	92	98	90				-50	[22]
Cattle feedlot parc wastewater	0.5		61.4–69.1	-	-	-	34.4–38.8	48.0–54.0	-	[57]
Clinical Laboratory	1	7–8	78-85	-	-	-	-	-	-	[26]
Urban (Sewage and rainwater)	0.89	6	87.6	91.3	98.4	-	-	_	-	[59]
Synthetic Dairy wastewater	0.6	10	83.2	-	-	-	-	-	-	[68]
Side-stream of dairy wastewater	0.48	4	$45\pm4.1$		$68 \pm 10$		$\textbf{77} \pm \textbf{8.4}$	$48\pm 6$	74 ±9.5	[13]
Textile dye effluent		8	85-89	76-80	73–77	71–76	-	-	-	[60]
Hospital wastewater	1	-	75	93	89	-	-	-	-	[55]
University campus wastewater	2.5	-	-	88	78	75	-	-	-	[69]
Municipal wastewater	2	-	$83.5\pm2.1$	$\begin{array}{c} 81.3 \pm \\ 2.9 \end{array}$	$\begin{array}{c} 93.7 \pm \\ 2.6 \end{array}$	$\begin{array}{c} 32.4 \pm \\ 10.6 \end{array}$	-	$\begin{array}{c} \textbf{38.6} \pm \\ \textbf{3,6} \end{array}$	$\begin{array}{c} 55.6 \\ \pm 11.6 \end{array}$	[38]
Synthetic wastewater	1	7.8	74	92	-	-	-	-	-	[32]
Synthetic domestic wastewater	0.2	-	$\begin{array}{c} \textbf{76.6} \pm \\ \textbf{5.18} \end{array}$		-	-	$63.8 \pm 2.75$	$\begin{array}{c} 81.0 \\ \pm 2.25 \end{array}$	-	[37]
Domestic wastewater	4.2	-	67.6	78	-	89	-	-	-	[43]
Synthetic Dairy Wastewater	0.6	-	80.7	88.4	-	-	61.7	77.8	-	[63]
Domestic greywater	0.16		83	97.6	99.4			31.3	62.2	[23]

#### Table 3

Species used and inoculation rate of earthworms in vermifilter.

Species	Inoculation rate	Wastewater type	COD removal perfor	References		
			Influent(mg/L)	Removal (%)		
Eisenia fetida	10000 worms/m <sup>3</sup>	University campus	325-400	92	[22]	
Lumbricus terestris	1000 worms/m <sup>3</sup>	Cattle feedlot	-	61.4-69.1	[57]	
Eisenia fetida	$1000 \text{ worms/m}^3$	Clinical Laboratory	_	78-85	[26]	
Eisenia fetida	20g/L	Urban (Sewage and rainwater)	-	87.6	[59]	
Eisenia fetida	10–12 g/L	Swine wastewater	1997	83	[12]	
Eisenia fetida	$10,000 \text{ worms/m}^3$	Synthetic Dairy wastewater	$1758.6\pm144$	83.2	[68]	
Eudrilus eugeniae	1000-1500worms/m <sup>3</sup>	Textile dye effluent	5800	85-89	[60]	
Eisenia fetida	$15000 \text{ worms/m}^3$	Sewage	$706\pm407$	$88\pm7$	[2]	
Eisenia fetida	$10000 \text{ worms/m}^3$	Hospital wastewater		75	[55]	
Eisenia fetida	32 g/L	Municipal wastewater	300	$83.5\pm2.1$	[38]	
Eisenia fetida	$10000 \text{ worms/m}^3$	Synthetic wastewater	$456\pm32$	74	[32]	
Eisenia fetida	8 g/L	Domestic wastewater	92.2	67.6	[43]	
Eisenia fetida	$10,000/\text{worms/m}^3$	Synthetic sewage		73.9	[8]	
Eisenia fetida	40 g/L	Piggery wastewater	$2960 \pm 88.8$	99.2	[53]	
Eisenia fetida	$10,000 \text{ worms/m}^3$	Synthetic dairy Wastewater	$1734 \pm 110$	80.7	[63]	

stabilize solids, organic matters (OM), and nutrients. According to Rajpal et al. [67], a sufficient HRT allows the rapid mineralization of nitrogen into nitrate. It is therefore preferable that the HRT is as long as possible, which would mean reducing the HLR as much as possible. However, a too-low HLR means that a large quantity of wastewater will not be treated at the same time. This represents a loss of time, space and therefore an increase in the treatment cost.

It is thus essential to have an optimal HRT/HLR ratio to allow an efficient and effective treatment of the wastewater in the VF with a good balance between performance and cost. Singh et al. [16] concluded to an optimal HLR of  $1.84 \text{ m}^3/\text{m}^2/\text{day}$  for brewery wastewater treatment and Adugna et al. [23] to a  $64L/\text{m}^2/\text{day}$  HLR for domestic greywater. Various HLR and HRT with associated performances are presented in Table 2. Their impact on the removal of different wastewater pollution parameters can be noted.

#### 4.3. Earthworms' species and density of application

The earthworms used in vermifiltration are epigeic. Also called surface-dwelling earthworms, they live in the upper layer of the soil and deposit wormcasts on the surface. Their involvement in the breakdown of organic matter is widely recognized as crucial. Experiments have been conducted so far on the species including *Eisenia fetida*, *Lumbricus rubellus*, *Lumbricus terestris*, *Eudrilus eugeniae*, *Perionyx sansibaricus*, *Eisenia Hortensis*, and *Eisenia andrei*. In recent years the species that have been used the most are *Eisenia fetida* and *Eudrilus Eugeniae* as shown in Table 3. *Eisenia fetida* is however the most used because it can withstand a wide range of temperatures

Wastewater type HLR (m <sup>3</sup> / m <sup>2</sup> /day)	HLR (m <sup>3</sup> /	HRT	Bedding material	0	Removal p	erformance	e (%)					References
	(Hours)	(cm)	COD	BOD	TSS	TN	TP	NO3-	Nh4+ –N			
University campus wastewater	1	4–6	Cow dung + vermigratings	20	92	98	90			-50		[22]
Clinical Laboratory	1	7–8	Cow dung + vermigratings	30	78-85							[26]
Distillery		8-10	Garden soil	7	90	95	80					[58]
Domestic greywater	0.16		Sawdust	40	83	97.6	99.4		31.3	62.2	75	[23]
Urban (Sewage and rainwater)	0.89	6	Vermicompost	16	87.6	91.3	98.4				76.5	[59]
		6	Sawdust	16	79.7	90.5	98.4				63.4	
Domestic sewage	2		Lombricompost	20	83					60		[80]
Commercial dairy		4	Woodchips	30				84+-8%				[11]
Dairy wastewater, synthetic	0.6		1:3 mixture Garden soil and vermicompost	30	83.2			57.3				[68]
Side-stream of dairy wastewater	0.48	4	Wood shaving and chips	20	45+-4.1		68+- 10	77+-8.4	48+-6	74+- 9.5		[13]
Synthetic Dairy wastewater	0.3		vermicompost and garden soil on the ratio of 1:3by volume	30	83.2			57.3				[65]
Textile dye effluent		8	garden soil	7	85-89	76–80	73–77					[60]
hospital wastewater	1		garden soil	30	75	93	89					[55]
Synthetic wastewater	1.5		River bed material + vermicompost	15	72.3	81.2	75					[33]
•			Wood coal + vermicompost		64.6	74.5	64					
			Glass balls + vermicompost		61.5	72.7	59					
			Mud balls + vermicompost		59.8	70.9	55					
Synthetic domestic	0.2		Artificial soil $+$ padding soil $+$ rice straws	35	76.6+-			63.8+-	81.0+-			[81]
wastewater					5.18			2.75	2.25			
Synthetic sewage	1.3		Gravel + mature vermicompost	30	73.9	84.8						[8]
Synthetic Dairy	0.6		1:3 mixture of vermicompost and garden	25	80.7	88.4		61.7	77.8			[63]
Wastewater			soil									

## Table 4Impact of bedding material and vermifilter bed height on treatment.

[38,61,70]. It also has a high reproduction rate and an optimum operating temperature of 25–27 °C [71]. It is a specie of epigeic earthworms that contains digestive enzymes such as alkaline, protease, phosphates, and cellulose. They possess an exceptional microflora in their intestine that allows the development of a very varied microbial community in their gut. They use their body as a filter, like other species used [61]. The effects of inoculation rate of earthworms have also been investigated in the context of wastewater treatment by vermifiltration. It was shown that the variation of the inoculation rate affects the treatment [72]. In general, the more earthworms there are, the higher the rate of water pollution removal. This is perfectly justified, considering the role of earthworms in the VF that was presented above. The more earthworms there are, the more the burrowing activity, allowing the aeration of the system to increase, and also the microbial activity to be enhanced. Earthworm stocking density also regulates microbial community structure and fatty acid profiles during vermicomposting of lignocellulosic waste [73]. The higher application rate would therefore allow for the treatment of more highly loaded wastewater (Table 3). However, as with the HLR, it is important to maintain an optimal rate.

#### 4.4. Vermibed media and height of active vermibed

The bedding materials and especially the active layer are very important parameters to consider when implementing a vermifiltration system. Indeed, the time that the wastewater will spend in the filter closely depends on these parameters, given the different porosities, permeability, and other characteristics of the filter materials. Singh and Kaur [74] have shown that the growth, reproduction, and performance of earthworms in the degradation of OM depend on the type of active layer. One of the most frequent signs of the discomfort of earthworms in an environment is that they try to escape and or die [75]. Some materials of the active vermibed can even harm earthworms' bodies. The type of filter material also has a great impact on the structuring of the microbial community responsible for the decomposition of OM [33]. The rate of clogging of the filtration system is also closely related to the porosity and permeability of the filter material. They control hydraulic conductivity. Low hydraulic conductivity leads to rapid clogging [76]. Materials with good porosity also provide a larger surface area for biofilm development and wastewater treatment [77]. The porous nature of the bed media permits aeration, creating a controlled environment conducive to microbial activity. Most of the active vermibed materials used in recent years are listed in Table 4. It can be seen that vermicompost is the most used and its purification performance is superior to that of sawdust. In addition, according to the research of Kumar et al. [33], river bed material has been found to be better than wood coal, glass balls, and mud balls both in terms of pollution removal performance (Table 4), and increase in earthworm biomass. The COD removal rate for example is 72.3% for river bed material versus 64.6%; 61.5%; and 59.8% for wood coal, glass ball and mud balls respectively.

As mentioned above, the earthworms used for vermifiltration are epigeic and they generally live and develop in the first centimeters of the soil, between 10 and 15 cm. It is therefore essential to consider this information in the design of the VF. Table 4 shows that in most cases, the depth of the active layer is between 10 and 30 cm. The research of Nie et al. [47] on rural synthetic wastewater, showed that the majority of pollution removal takes place in the first 40 cm of the filter bed. An anaerobic zone can develop in the lower zones of the filter bed if the depth of the bed is too great. This will have a negative impact on earthworms and biofilm microorganisms [78]. Wang et al. [79] showed that variation in VF height had a significant effect on COD and total phosphorus (TP) removal rates, earthworm population, and *actinomycete* numbers, but had no effect on total nitrogen (TN) and ammonia nitrogen (NH<sub>3</sub>–N) removal rates, and on bacterial and *fungi* numbers.

#### 4.5. Planted vermifilters

Macrophyte VFs are a very promising technique to remove organic and mineral matter from wastewater. In the literature, it is reported that the average removal rates of planted VFs are better than those of simple VFs. A theory put forward by Singh et al. [57], is that the root exudates from the plants in the planted VF make the microbial activity more intense by promoting the growth of microorganisms. In addition, the increase in aeration due to the roots would also increase the population of microorganisms [82]. Huang et al. [83] showed a significantly higher growth rate of earthworms in the planted VFs compared to the ones without plants. The increase in the number of earthworms was also significantly higher in the planted VFs, by about 76.5%. For the growth of their biomass, the plants would absorb the phosphates as nutrients. With the plants in the VF, there is also a decrease in clogging due to the fact that the roots of the plants by their growth, create cracks in the filter bed [28]. Plants also enhance the denitrification potential of the system [80]. The most widely used macrophyte species according to the literature is *Canna indica*. It is a specie of herbaceous flowering plants in the *Cannaceae* family. It is sometimes called *Indian canna*. Compared to other macrophytes such as *Saccharum spontaneum* and *Typha angustifolia* the percentage of COD removal is 3.9 % higher than the former, 7.3% higher than the latter, and 13.4% higher than the VF without plants [34].

#### 4.6. Temperature

Earthworms are sensitive to temperature fluctuations. Fecundity, cocoon production, and maturation of earthworms are some parameters that may be affected by temperature [49]. Therefore, the temperature has a significant impact on the performance of the VF. specifically on the removal of total ammonia nitrogen (TAN) and COD [13]. The effect of temperature was also noted on the removal of NH<sub>3</sub>–N by Wang et al. [10]. Indeed, the diversity and composition of the NH3–N oxidizing *Betaproteobacteria* community in the different layers of the VF over the year were probably influenced by temperature changes. Arora and Kazmi [71], also revealed that variations in ambient temperature had a significant effect on the reduction of COD, biochemical oxygen demand (BOD), and

#### pathogens.

#### 5. Modeling and optimization of the vermifiltration process

Various authors have worked on the modeling and optimization of the vermifiltration process. Mathematical models of vermifiltration can be used not only to design and optimize the operation of the system but also to predict its behavior and to control it [84]. Numerous authors used the response surface methodology (RSM) and the Box-Behnken design (BBD) to evaluate both the individual effect of the different parameters and their simultaneous interaction on the system performance [16,19,84–86]. Samal and Dash [84] worked to optimize biochemical oxygen demand (BOD) removal in the treatment process of a synthetic dairy wastewater. Hydraulic loading rate (HLR), biodegradable organic strength (BOS) and depth of the active layer (ALD) of the Vermifilter unit were considered as the main parameters influencing the VF performance. According to the developed model, the optimal conditions to achieve maximum BOD removal for the designed system were at an influent concentration of 1701.00 mg/L, HLR of  $0.39 \text{ m}^3/\text{m}^2/\text{day}$ , and ALD of 34.40 cm. The experimental BOD removal of 86.45% was obtained against a predicted value of 87.08% under optimal conditions. Singh et al. [16] sought to optimize the chemical oxygen demand (COD) removal from a synthetic brewery wastewater, with HLR and BOS, as the influencing parameters, but this time earthworm density (EWD) instead of ALD. The optimal conditions to achieve maximum COD removal for the designed system were at an influent concentration of 3542 mg/L. HLR of  $1.84 \text{ m}^3/\text{m}^2/\text{day}$ , and EWD of 9661 earthworms/m<sup>3</sup>. Under the optimal conditions, a COD removal of 94.99% was obtained against the predicted value of 95.85%. Model verification on real brewery wastewater also showed minimal error from the predicted COD removal. The COD removal index (CRI) a mathematical tool has also been developed by Singh et al. [85], to predict the organic removal performance of vermifilter. The authors defined the BOS, the EWD but the HRT this time as the influencing parameters. The  $R^2$  value of the plot between the calculated value of the CRI and the obtained removal was found to be 0.8, showing that the tool can be well used to predict the removal of organic matter in a VF. Samal et al. [19] optimized the removal of COD and TN from a synthetic dairy wastewater with HRT, BOS, and ALD as influencing factors. They obtained maximum removal percentages of 83.2% for DOC and 57.3% for TN. Concerning the kinetic model, the Stover-Kincannon model was found to be the most suitable for the degradation of pollutants by vermifiltration than the first-order model and the Grau second-order model with the best regression coefficient both for COD.  $R^2 = 0.9961$  and for TN,  $R^2 = 0.9353$  against  $COD R^2 = 0.5212$  for the first-order model and  $R^2$  for COD and TN, 0.9817 and 0.8395, respectively for the Grau second-order model. It has been shown by Singh et al. [50] that the clogging of the VFs can also be minimized by optimizing the HLR, EWD, and COD concentration still by the RSM method. They were able to find that for brewery wastewater, the minimum clogging is obtained with HLR of 1.84, EWD of 9475 earthworms/m<sup>3</sup>, and 3701 mg/L of COD, with insignificant errors. Mathematical models and response surface methodology (RSM) are thus, important to better understand the behavior of the vermifiltration system. They aid understanding and enable accurate predictions, facilitating more efficient system design and operation. Research highlights the adaptability of vermifiltration to various wastewater treatment scenarios. Performance can be optimized by fine-tuning variables such as HLR, BOS, EWD and ALD. By optimizing these parameters, specifically HLR, EWD and COD, clogging problems can also be minimized. CRI has proved invaluable in assessing the organic removal efficiency of vermifiltration systems. This index can be useful for monitoring and improving system efficiency over time.

#### 6. Fate of wastewater parameters and performance of vermifilter

#### 6.1. pH

Various studies have shown that the VF acts as a buffer. One passes respectively from acidic pH to neutral pH and also from basic pH to neutral pH. The vermifilter would thus allow neutralization of the pH of the effluents [12,26]. In the experiments conducted by Arora et al. [26] concerning the application of vermifiltration for treating clinical wastewater, the initial influent pH levels exhibited considerable variability within the range of 4.2–9.3. However, at the vermifilter outlet, the pH levels stabilized and remained relatively constant, falling within the range of 6.7–7. In their study, Ghasemi et al. [80] noticed an increase in pH at the beginning of the treatment and stabilization at a neutral pH until the end of the treatment. Thus, one can go from pH 8.5 to about pH 7 [22], and from pH 4 to pH 7 [54]. This tendency to neutralization and stabilization of pH can be explained by the elimination of bio-contaminants contained in the influents, and the decrease of biochemical oxygen demand (BOD) and chemical oxygen demand (COD) concentrations also [54]. It could also be attributed to the aerobic degradation of organic matter generating carbonic acid [29]. Another explanation is that *VTpase*, a protein upregulated by earthworms in response to stressful wastewater conditions, may be responsible for stabilizing pH.

#### 6.2. Electrical conductivity (EC)

No significant difference was observed between the electrical conductivity (EC) of the effluent and the influent in most of the studies. But in the work of Kannadasan et al. [58], a significant difference was noted between the EC of distillery effluents treated by a NVF and those treated by vermifiltration. The EC values of the simple filter effluent were in the range of  $2.03 \pm 0.04$  mS/cm for the 50% diluted effluent while that of the vermifiltration treated effluent were in the range of  $5.41 \pm 0.01$  mS/cm. The increase in EC in the vermifilter effluent could be due to the release of minerals from the earthworm gut. Other authors also noticed an increase in EC [12].

#### 6.3. Dissolved oxygen (DO)

Authors have reported on the behavior of DO during the treatment by vermifiltration. A clear increase of this parameter is noted at the outlet of the VF [26,69]. However, on closer observation, Rustum [62] observed that the DO of the water is reduced at the beginning of the treatment, but as the treatment progresses, the levels begin to increase until the end. According to him, this phenomenon is attributed to the initial consumption of DO by microorganisms during the initial stages of treatment, which is essential for breaking down the organic matter. With the aeration of the system during the treatment, this DO is replaced by oxygen from the atmosphere. At this point, the dissolved oxygen concentration starts to increase [62].

#### 6.4. Organics and solids removal

The different mechanisms that occur during the degradation of organic matter during wastewater treatment are: dehydrogenation, oxidation, stabilization of OM, and transformation of unsaturated structures into saturated structures [87].

The degradation and stabilization of the OM in the vermifilter is possible by the enzymatic activity of the micro-organisms. It allows the decomposition of starch, proteins, and cellulose. More specifically, it would be the activity of cellulase, amylase, and protease [32]. The high COD removal rates due to the enzymatic activity of the microorganisms would not have been possible without the presence of earthworms [32,69]. According to Wang et al. [10], the diversity of bacteria in the VF could influence COD removal. The high COD removal rates is also attributed to the symbiotic action of earthworms and microorganisms. The different burrowing actions of the earthworms keep the environment aerated and allow the oxidative work of the aerobic bacteria [16]. A correlation has been observed between COD removal in the VF and the activity of antioxidase enzymes and oxygen reactive species in the earthworm tissues and also the length of the earthworm burrow [88].

Microorganisms are responsible for the degradation of OM and dissolved, suspended, organic and inorganic solids in the VF. The degradation of the solid matter is done by catabolic activity. However, to facilitate the removal of total suspended solids (TSS) and total dissolved solids (TDS), the earthworms perform grinding, dispersal, ingestion, and digestion activities of these TSS and TDS [20]. The earthworms then release finer particles to be trapped in the pores of the VF for direct removal from the wastewater. Quantitative analysis using  $\delta$ 15 N showed that earthworm feeding and earthworm-microorganism interaction were responsible for approximately 21% and 79%, respectively, of the highly volatile suspended solids removal [89].

#### 6.5. Nutrient removal

Nitrogen compounds are among the most present elements in wastewater. They are also the main cause of water eutrophication. As mentioned earlier ammonification, nitrification, and denitrification are the processes through which nitrogen is eliminated from wastewater. Further analysis of the transformation of nitrogen speciation by the  $\delta 15$ N–NO<sup>3-</sup> isotope dilution method also confirmed the occurrence of nitrification and denitrification processes [90]. First, the organic nitrogen is converted to ammonium. Then ammonium is converted to nitrates, and nitrites during nitrification. The heterotrophic bacteria that enhance the mineralization of nitrogen come largely from the gut of earthworms [59]. Finally, as a result of the last reaction, denitrification, nitrates and nitrites are converted into nitrogen gas by the action of denitrifying bacteria [11]. Comamonadaceae, from the Betaproteobacteria family, were identified as involved in the denitrification process. Analysis of 16S rRNA gene profiles in influent and effluent from the VF concluded that these bacteria were increased during vermifiltration [91]. The VF is mainly dominated by proteobacteria. More precisely the y-proteobacteria followed by the Acidobacteria, the Bacteroidetes, and the planctomycetes [43]. Besides these types of bacteria, another community in a lower number but functional has been detected from the study. These are Gemmatimonadetes, Verrucomicrobia, Actinobacteria, and Chloroflexi. It is found that the dominant bacteria at more than 76-92% of the microbial community of the earthworm gut are the gammaproteobacteria [89]. It is also reported in the literature that, the low ammonium content of treated effluent may be due to the rapid mineralization of nitrogen. An increase in nitrification genes has also been noticed by Lai et al. [91] suggesting that nitrogen removal is due to ammonium conversion. The results of Kannadasan et al. [58] show that the residues from the treatment of distillery water diluted to 50% by vermifiltration are 1.81% richer in nitrogen than the water treated with the NVF. According to the authors, the dead earthworm tissues are a cause of the increase in Total Nitrogen (TN). Nitrification is accelerated with oxygen and therefore denitrification is slowed down, so the denitrifying bacteria are anaerobic. To have a favorable performance for nitrate removal, the conditions must be modified by creating anoxic conditions for the microorganisms to limit the penetration of air in the lower parts of the system and by forcing the microorganisms in these sections to use nitrate instead of oxygen [29,44].

The phosphate contained in wastewater is rapidly mineralized to inorganic phosphate  $PO_4^{3-}$ . This would be due to the enzymatic activities [92]. For this purpose, Lourenço and Nunes [59] observed that the removal efficiencies of TP in wastewater were negative. The leaching of vermicasts along the filter to the outlet can also be an explanation for the increase of TP in the effluents treated by vermifiltration, these being rich in nutrients including phosphorus [69]. The mineralization process is more pronounced in VFs than in NVFs. According to Kannadasan et al. [60], Total Calcium and Total Magnesium concentrations almost quadrupled from VF effluent to NVF effluent. Values go from 1.64 to 1.05 respectively to 4.1 and 4.92.

#### 6.6. Pathogens removal

The pathogens would undergo the effect of antibiotic and toxic secretions of the earthworms - the coelomic liquid - and of the microflora of the system [21,61]. This would therefore lead to an inhibition of the activity and growth of these pathogens. The mucus

shed by the earthworm sticks to the surface of the bedding material and traps microbes that may be harmful in nature by competing with favorable microbes. The trapped pathogens are then killed being unable to move and also due to the lack of food and oxygen nearby [93]. This would be a possible reason for the elimination of pathogens in the VF. River bed materials and mud balls have been reported to be very suitable filter media for pathogen removal [94]. They also observed that there is a large diversity of microor-ganisms that have the ability to prevent the growth of other pathogens. The research of Arora et al. [8] also showed antibacterial activity of the microorganisms isolated from the VF against *gram-positive Staphylococcus aureus (ATCC 29213)* and *gram-negative E. coli (ATCC 25922)*, further confirming the pathogen elimination mechanism.

#### 6.7. Heavy metals and antibiotics remediation

The bioavailability of some heavy metals such as Cd, Ni, Pb, Cu, Cr, and Zn decreases significantly during vermifiltration [60]. Heavy metals are accumulated in the organisms of earthworms [40,95]. *Metallothioneins* are proteins in the gut of worms that have the ability to bind heavy metals and make them biologically inactive [41,68]. It is more precisely the *chloragogens* that accumulate these metals. Vermicast also offers good adsorption sites for these metals and chemical pollutants in wastewater [81]. The vermifilter is also capable of reducing the ecotoxicity of certain antibiotics. Even if this alone cannot be considered as a full-fledged treatment of hospital wastewater, because of its high concentration of various antibiotics, it can nevertheless be a tertiary or refining treatment [3].

#### 6.8. Stabilization of sewage sludge

VFs allow for the stabilization of sewage sludge. Research showed that VF has clear advantages over NVFs in the stabilization of sewage sludge [96]. Xu et al. [97] reported an average decrease of 0.05 in the settled sludge volume (SSV)/SSV ratio, along with an enhanced SSV reduction efficiency of 13.86% as compared to reduction efficiencies of 14.5% [98] and 14.7% [99]. The Volatile Suspended Solids (VSS)/Suspended solids (SS) ratio can reach  $0.65 \pm 0.02$ , and a VSS reduction of  $48.09 \pm 2.21\%$  in the VF, which is consistent with the 40% sludge stabilization level required for anaerobic and aerobic digestion. These results suggest a close relationship between stabilization performance and earthworms. Chen et al. [100] also found that the addition of earthworms to a sludge treatment system improved sludge stabilization remarkably with a decrease in the volatile solids VS/TS ratio from 49% to 18% in the accumulated sludge. A vermireactor was also employed for the co-treatment of organic fraction of municipal solid waste (OFMSW) and sewage. The presence of earthworms in the treatment process resulted in the removal of various components, with removal percentages reaching up to 75% (TOC), 86% (Total COD), 87% (BOD5), 59% (ammonia nitrogen), and 99.9% (coliforms) [101]. These findings suggest that earthworms are also well-suited for co-treating OFMSW and municipal sewage on-site.

#### 7. Fate of the VF system

#### 7.1. The vermibed

Clogging is a recurrent problem in deep filtration, especially in wastewater filtration due to high pollution loads. It results from a decrease in the porosity of the filtering materials, due to the accumulation of organic and inorganic solid matter in the pores of the filtering materials [23]. Comparative studies of vermifilters (VFs) with non-vermifilters (NVFs) filters have shown that VFs are slower to clog. The parameters often used to judge clogging in the filter are hydraulic conductivity and Headlosses. Singh et al. [29] showed a decrease in hydraulic conductivity of 10.5 m/day for the NVFs and a decrease of 4.7 m/day for the VF, for the same period of operation. Also, an increase of 0.41 cm of the headloss was observed in the VF against an increase of 0.97 cm in the NVF, representing more than half of the precedent value. According to these results, the presence of earthworms in the filter delays the clogging even if it is not completely eliminated. Adugna et al. [23] confirm these results. The burrowing, tunnelling and ingestion activities of earthworms are responsible for this decrease in clogging. The strong aeration of the filter bed due to these activities leads to the destruction of the solids contained in the pores so that the VF can work smoothly and uninterruptedly for a long time. Furthermore, in a normal biological treatment system, suspended solids accumulate on top of the filter. With time this forms sludge which is responsible for clogging, as it chokes the system. In the VF, these suspended solids are permanently consumed by the earthworms and rejected in the form of vermicompost [102]. The system can operate for 12 months without clogging, despite high concentrations of pollutants Adugna et al. [23].

#### 7.2. Fate of the microorganism's community

The Shannon index (H), which expresses the diversity of a studied population, has been determined in various studies to judge the diversity of the microbial population in the VF. It appears that compared to a NVF this index is always higher. Xu et al. [27] obtained an H = 2.58 for the VF and H = 1.99 for the NVF. They were also able to detect the presence of *Actinobacteria* and *Acidobacteria phyla* only in VF and not in NVF. Other authors like Zhao et al. [98] found respectively HVF = 3.77 and HNVF = 3.49, i.e. 16% higher for the VF.

#### 7.3. Fate of earthworms

Research on the earthworm population and their enzymatic activity in the VF area indicates that: during the first week, which often represents the acclimatization phase to their new environment, signs of discomfort are noticed in the earthworm community. Weight

Fate of earthworms during vermifiltration.

Start of the experiment	End of the experiment	Duration of the experiment (days)	Reference
Number of earthworms			
200	214	77	[57]
200	206	77	[57]
480	670	133	[55]
800	950	120	[32]
800	1000	91	[6]
Density of earthworms biomass (g,	/L)		
39.05	47.64	77	[57]
32	43	60	[104]
50	86.5		[33]
50	73.5		[33]
50	64.5		[33]
50	65		[33]
Percentage of increase (%)			
28.3–31.5		100	[65]
19.1–26.2		100	[65]
20.29–27.82			[84]
11.4			[69]
26-32.4			[19]

loss, swelling of the clitellum area, curling up and some deaths can be noticed [103]. But after this acclimatization phase, the earthworms start to show signs of comfort and well-being. They grow in number and stature as they go along. Table 5 shows the variation in the density of earthworms in the filter during the treatment. Indeed, earthworms develop defense strategies against the hostile environment that wastewater often offers with several types of waste, organic matter, nutrients, heavy metals, etc. Some differential proteins such as *V*-ATPase, actin, and tubulin are up-regulated and contribute to cell motility and stress response [20].

#### 8. By-products of vermifiltration

#### 8.1. Treated effluent

Given the current challenges in accessing and managing conventional sanitation, it is essential to explore low-cost technologies and systems that provide a closed loop between sanitation and agriculture [105]. Several studies have concluded that vermifiltration treated effluents can easily be used without restriction in agriculture [69,106]. According to the results of Manyuchi et al. [54], distillery effluent treated by vermifiltration can be reused for crop irrigation and thus reduce the demand on other water sources which are sometimes insufficient. An important indicator in water that can be used to judge the usefulness for agriculture is the DO. The high amounts of DO in treated effluent are very promising for reuse in agriculture because they reduce the septicity of the water [16,18, 107]. The amounts of DO in the study of Arora et al. [22] increased from 0 to 0.35 mg/L in the influent to 3 and 5 mg/L in the effluent. Also, the mineralization of phosphate and nitrogen from wastewater with *polysaccharides* and proteins, secreted by earthworms, during vermifiltration produces bioavailable compounds for plants. The research of Arora et al. [22] also showed removal rates of total coliforms (TC), faecal coliforms (FC), and faecal sludge (FS) pathogens of more than 99% making the waters suitable for irrigation according to the 1989 World Health Organism (WHO) standards. There are essential plant nutrients, which make these vermifilter (VF) effluents highly nutritious for plants and potentially useable for crop irrigation. They are very rich in NPK (Nitrogen Phosphorus and Potassium). Having been tested for the irrigation of onions (*Allium cepa*), the effluents have demonstrated their performance for the increased germination of roots without any malformation or chromosomal anomaly [41].

#### 8.2. Vermicompost

Vermicompost or lombricompost, a biofertilizer recognized for its high properties, rich in NPK, is a by-product of vermifiltration [54]. The earthworm gut acts as a bioreactor and can ingest solid and liquid organic waste from wastewater and expel them as vermicompost [55]. During the vermifiltration process the earthworms consume the organic matter contained in the wastewater and release castings [18]. These vermicastings are full of many enzymes and microorganisms good for soil fertility. The vermifilter would recover 20% of the initial nitrogen beneficial to the crops [11]. It provides plants with macro and micronutrients [108]. Medina-Sauza et al. [109] supported the idea that the work of microorganisms with earthworms leads to a privileged selection of certain bacteria in the vermifilter. Bacterial species contained in the earthworms are mostly gram-negative and those present in the filter media are gram-positive. This confirms that the earthworms also purify the filter medium making it suitable for use as an organic fertilizer. Some *Pseudomonas* sp. are found to be plant growth promoters and are used as biological control and bioremediation agents and *Klebsiella* is said to fix nitrogen in the soil. Thus, their presence indicates an enriched quality of the compost formed as a result of the treatment. Also, the growth rate of earthworms in the VF is a good indicator of the ecology of the vermifilter.

#### 9. Sustainability of vermifiltration

To be identified as sustainable, a technology must be so on the three main dimensions including environmental, social and economic. The technology must be environmentally sustainable, economically viable and socially acceptable. Without addressing these key aspects of sustainability, a treatment system will fail during its operational phase [110,111].

#### 9.1. Environmental sustainability

The various aspects to consider for the environmental sustainability assessment of a technique include.

- Life Cycle Assessment (LCA): LCA involves evaluating the environmental impacts of a product, process, or system throughout its entire life cycle. This assessment takes into account all stages, from raw material extraction and manufacturing to use, maintenance, and disposal or recycling. Regarding vermifiltration, it has been demonstrated that vermifiltration technology has a minimal dependence on fossil fuels, thereby contributing to the conservation of non-renewable natural resources. Researches of Passteni et al. [112] and Lourenco and Nunes [113] on this matter has shown significant variations in fossil fuel consumption between different wastewater treatment technologies. These variations include quantities of up to 0.04 kg/kg of BOD<sub>5</sub> removed for Activated Sludge Process (ASP), up to 0.03 kg/kg of BOD5 removed for Aerated Lagoons (ALs), and 0 kg/kg of BOD5 removed for Vermifiltration. Vermifiltration also does not require the use of fossil fuels during the construction phase, whereas ASP consumes slightly over 0.6 kg of fossil fuels per equivalent inhabitant during construction. In the operational phase, electricity consumption is 1,160,000 MJ per equivalent inhabitant for ASP compared to only 4520 MJ for VF. In general, the use of vermifilters in domestic wastewater treatment is associated with relatively low greenhouse gas (GHG) emissions. This low emission can be partly attributed to the activity of earthworms, which promote an aerobic environment, thereby reducing the production of gases such as methane (CH4) and nitrous oxide ( $N_2O$ ), both of which are considered potentially harmful GHGs. In comparison, other wastewater treatment technologies, such as anaerobic lagoons, macrophyte ponds, upflow Anaerobic Sludge Blanket Reactors (UASBR), and ASP, are often associated with significant GHG emissions, especially when their operation depends on the use of fossil fuels for electricity generation [114]. Furthermore, as mentioned earlier, VF generates very little sludge, with only 0.08 kg of suspended solids (SS) per kg of COD removed, primarily in the form of vermicompost [43]. In comparison, conventional processes can produce up to 100 g of sludge per cubic meter of treated domestic wastewater. Vermicompost has highly advantageous characteristics for agriculture [54].
- Life Cycle Impact Assessment (LCIA): LCIA aims to assess the potential environmental impacts of a product, process, or system on the environment throughout its life cycle. Research has shown that VF is the most environmentally friendly method, with a reduced environmental footprint compared to other technologies. For example, VF has a climate change impact (CC) of 6.7 m<sup>3</sup>/(kg CO2 eq./kg COD removed) and an eutrophication impact (EUT) of 10,984.1 m<sup>3</sup>/(kg P eq./kg COD removed), while ASP has a climate change impact of 3.8 m<sup>3</sup>/(kg CO2 eq./kg COD removed) and an eutrophication impact of 10,518.5 m<sup>3</sup>/(kg P eq./kg COD removed). ALs also show higher environmental impacts than VF. Therefore, VF is the best option in terms of environmental sustainability for domestic wastewater treatment [112].
- Preserving the quality of water, air, soil and ecosystems: inadequate treatment of domestic wastewater can harm water quality and the aquatic ecosystem due to the presence of pollutants such as pathogens, heavy metals, and emerging contaminants. However, as demonstrated earlier, VF has the ability to effectively remove them, thus preserving water quality and the balance of the aquatic ecosystem. Furthermore, the production of wastewater treatment sludge in other methods can pose environmental problems, such as greenhouse gas (GHG) emissions during their treatment. In contrast, VF uses earthworms to digest the sludge, producing nutrient-rich vermicasts that are beneficial for soils and terrestrial ecosystems. Therefore, VF has minimal impact on air quality. In comparison, other wastewater treatment methods, such as activated sludge, have shown higher environmental impacts, including significant GHG emissions. In summary, VF proves to be an environmentally friendly option for domestic wastewater treatment, with measurable benefits in terms of water quality, GHG emissions, and soil and ecosystem benefits [11,67].
- Reuse of treated effluent: the reuse of treated effluent depends on its level of contamination, with indicators such as Dissolved Oxygen (DO), organic matter (BOD and COD), pathogens, NH4+-N, and NO3–N. The effluent from domestic wastewater VF is clear, odorless, rich in DO, and has a neutral pH [115]. Therefore, it can be effectively used for various non-potable purposes such as soil irrigation, toilet flushing, cooling tower makeup in industries, and agricultural applications

#### 9.2. Economic affordability

The economic feasibility assessment of a technology considers investment costs, operational and maintenance costs, treatment efficiency, and waste management.

• Investment costs (ICs): ICs encompass expenses related to land acquisition, raw materials, energy consumed during construction, transportation of raw materials, and the installation of various equipment. According to a study, the land area required for Small Infiltration at Reduced Flow (SRI), Constructed Wetlands (CWs), and Activated Sludge Process (ASP) in the treatment of domestic wastewater for small communities of 120, 120, and 500 inhabitants, respectively, was significantly reduced when these processes were replaced by vermifiltration (VF) technology. VF requires a much smaller land area. Regarding raw materials, materials including sawdust, wood shavings, vermicompost, and others used in VF are generally locally available at a low cost, or even obtained as waste from other activities. They are, therefore, very cost-effective. Furthermore, acquiring earthworms at a very low

Overview of both capital costs and operational costs for each wastewater.

Wastewater Treatment Method	Capital Costs (€ per User)	Operational Cost ( $\ell$ /year or $\ell$ /m <sup>3</sup> /year)	References
Vermifilter (VF)	100 to 150	$0.05 \ \epsilon/m^3/year$	[116,117]
Conventional Sewage Treatment Plant	Up to 3,750,530	Varies (e.g., up to 3,750,530 \$ in rural settings)	[117]
Septic Tank with Percolation Area	Approximately 1132	14 €/user/year	[118]
Membrane Bioreactor (MBR)	1800 to 2000	50-70 €/user/year	[118]
Moving Bed Biofilm Reactor (MBBR)	Approximately 1500	20-30 €/user/year	[118]
Sequential Batch Reactor (SBR)	620 to 900	4-7 €/user/year	[118]
Conventional Sewage Treatment	Up to 3,750,530	Varies (e.g., up to 3,750,530 \$ in rural settings)	[117]

cost is considered a one-time investment, as earthworms can be reused in new vermifilters. Compared to conventional methods, VF technology is decentralized and requires less space, making it suitable for use near the source of domestic wastewater. This reduces costs associated with transporting wastewater to treatment sites [113]. Furthermore, VF technology does not require heavy equipment, which contributes to its cost-effectiveness. Studies have shown that the capital costs of VF technology are significantly lower than those of other on-site domestic wastewater treatment systems. For example, the capital costs of systems such as septic tanks with percolation areas, membrane bioreactors (MBR), moving bed biofilm reactors (MBBR), sequential batch reactors (SBR), and constructed wetlands (CWs) were significantly higher than those of VF technology [116,117].

- Operating and Maintenance Costs (OMCs): OMCs of wastewater treatment methods encompass energy consumption, bed material renewal (especially for VF), equipment replacement or repair, skilled manpower, sludge management, and chemical requirements. Longevity also impacts costs. As demonstrated above, VF technology consume significantly less energy during the operational phase, mainly due to earthworm-driven natural aeration. VF's simplicity eliminates the need for skilled manpower, further reducing operational costs [115]. Maintenance expenses are also lower compared to conventional WWTPs. VF technology boasts longevity, lasting up to 3–8 months for low-strength domestic sewage chemical requirements are lower, reducing costs compared to other methods [112]. Table 6 provides a comprehensive overview of both capital costs and operational costs for each wastewater treatment method.
- **Treatment efficiency and by-products:** VF has demonstrated high effectiveness in removing various pollutants from domestic wastewater, including organics, nutrients, and pathogens [6,33]. Studies have indicated that VF-treated effluent meets stringent surface water discharge standards, particularly concerning pathogens [6]. As discussed above, VF also generates beneficial by-products. The vermicompost can contribute to a reduction in food production costs and enhance food safety by reducing the risk of chemical contamination [119]. Vermicompost offers additional advantages such as improved soil moisture retention, higher agricultural yields, and efficient resource utilization [119].

In summary, VF emerges as an economically viable and sustainable option for domestic wastewater treatment, delivering substantial environmental benefits while positively impacting agriculture and food security.

#### 9.3. Social acceptability

The acceptance of a specific wastewater treatment technology within society hinges on several key factors: safeguarding public health, engaging the public and fostering community development, and considering aesthetic aspects.

- Safeguarding of public health: given that Vermifiltration (VF) technology has the potential to substantially remove various pollutants, including organic matter, nutrients, and pathogens, waterborne disease outbreaks would decrease, thereby reducing the risk of human toxicity [6]. Furthermore, Kumar et al. [92] postulated that the concentration of NO<sub>3</sub>–N in the effluent obtained from domestic wastewater VF was <45 mg/L. Therefore, discharging the effluent from VF into surface water bodies would not cause blue baby syndrome [17]. Moreover, as previously mentioned, as an organic fertilizer, the application of vermicompost enables the production of healthy, chemical-free food, thus reducing the risk of harmful impacts on humans from food consumption. Since VF technology is capable of reducing contaminant levels below permissible limits, it can be stated that the application of VF technology promotes public health protection.
- Economic development of the local community: given that vermifiltration is a decentralized technology, it can be effectively used to treat domestic wastewater generated by individual households or small communities. Due to the ease of VF construction and operation, residents can decide how to construct and use the system for their economic growth. In other words, VF ensures public involvement, which is not possible with centralized wastewater treatment methods. Vermicompost can be sold in the market as biofertilizer. Devkota et al. [120] reported that vermicompost was sold in the market at a price of 25 rupees/kg, with a net profit of 9.32 rupees/kg. Additionally, earthworms can also be sold to various farms as raw material. Thus, the VF process also helps boost the economy, attracting rural residents to the VF process. Since it is a decentralized method, all community members can also benefit from the VF facility. Moreover, the treated effluent can be used for various non-potable purposes by the beneficiaries. It also promotes social resilience and stability through the wise use of resources. Therefore, through the implementation of VF for domestic wastewater treatment, all community members will be able to prosper through appropriate natural resource-based development.

Vermifiltrations' sustainability features.

DIMENSIONS	FEATURES	REFERENCES
ENVIRONMENTAL	No consumption of any fossil fuel during all the stage of the vermifiltration process.	[112]
	Protection the air quality, especially while treating the domestic sewage.	[39]
	Nutrient recovery - organic fertilizer.	[54]
	Reusability of treated effluent.	[16,18,107]
	Preservation of aquatic ecosystem.	[6]
	No chemical toxicity on the soil-based organisms.	[115]
	Air pollution - can potentially cut down the risk of GHG emissions and thereby minimizing the GWP to a great extent.	[112,113]
	No odor emission.	[41]
	No sludge production.	[41,42]
ECONOMICAL	Does not necessitate the installation of the heavy-duty instruments, cost-effective alternative.	[113]
	No requirements of external aerators-energy efficiency.	[18]
	Efficient removal of pollutants, including organics, nutrients, and pathogens from various types of water.	[32,69].
	Value-added by-products linked to circular bio-economy.	[1]
SOCIAL	Public Health – drastic reduction of outbreak of the water-borne diseases, human toxicity risk reduction.	[6,33]
	Public involvement and community.	[119]
	Development - growth of local socio-economy.	
	Cultural acceptance – no odor, no pungent small, no sludge, clear effluent.	[18,41]

• Aesthetic aspects: VF is socially accepted due to its positive impact on environmental aesthetics and the absence of unpleasant odors. It is particularly suitable for small communities as it preserves aesthetics and avoids sludge production. Its effluent is clear and does not alter the color of the water, maintaining aesthetic value [41].

The vermifiltration sustainability features are summarized in Table 7. It can be concluded that vermifiltration is a sustainable alternative for the treatment of domestic and industrial wastewater. Almost all the criteria regarding the different dimensions are satisfied.

#### 10. Conclusion and future perspectives

From the research results of the last ten years on vermifiltration (VF), it can be concluded that the vermifilter can be considered as a good alternative for wastewater treatment and reuse. It appears from the literature that the VF has a very high potential as a suitable treatment technology for wastewater from various sources, especially for countries facing severe challenges including insufficient investment costs and skilled labor. Furthermore, it has been demonstrated to be a sustainable technology. The analysis of the removal performance of vermifiltration showed its ability to remove various pollutants from the water, with by-products such as treated wastewater and vermicompost. Additionally, it demonstrates the potential for optimizing the operational parameters of the vermifilter to achieve peak performance. However, VF still has some limitations in a non-negligible number of aspects, due to the increasingly strict water quality standards for wastewater treatment and reuse.

The areas of research to be explored are the following.

- Additional research is needed to evaluate long-term removal performance at the field scale, outside the laboratory. These studies will help to judge the removal performance of vermifilters in real cases and the sustainability regarding the treatment efficiency.
- In addition, vermifilters should be installed in communities and households to evaluate the robustness and performance of the system and to study people's perceptions of the system. Vulgarization methods to disseminate information should be adopted to encourage the use of vermifilters by individuals or communities who need them as a sanitation tool.
- So far, very few studies have been conducted in sub-Saharan Africa, which faces great sanitation challenges. The technology, given its advantages, should be more widely used in this part of the world.
- To improve the design of the vermifilters for optimal performance, it is necessary to perform fundamental mathematical modelling of the system, taking into account the different dynamics: dynamics of filtering materials, dynamics of microorganisms, and dynamics of earthworms. This modelling would also integrate a larger number of influencing parameters and would allow a more thorough understanding of the system.

#### Data availability

The data that support the findings of this study are available on request from the corresponding author.

#### **CRediT** authorship contribution statement

Sidesse S. Y. Saapi: Writing – original draft, Methodology, Investigation, Formal analysis, Conceptualization. Harinaivo A. Andrianisa: Writing – review & editing, Supervision, Methodology, Conceptualization. Malicki Zorom: Writing – review & editing, Validation, Supervision, Methodology. Lawani A. Mounirou: Writing – review & editing, Supervision, Methodology, Formal analysis.

#### Hemez Ange Aurélien Kouassi: Conceptualization. Mahugnon Samuel Ahossouhe: Writing - review & editing.

#### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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