Fecal pollution indicators removal by a vertical Multi-Soil-Layering system in domestic wastewater in Morocco

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Abstract. The aim of this study is to evaluate the ability of the vertical Multi-Soil-Layering filter (V-MSL) to remove indicators of fecal pollution from domestic wastewater under Moroccan conditions. To do this, a V-MSL filter measuring 20 cm deep, 60 cm wide, 78 cm high was installed near the guardian's house of the Razi’s middle school in Meknes to treat domestic wastewater. Three hydraulic loads (250 L/m²/day, 350 L/m²/day and 500 L/m²/day) were tested. This filter showed an average reduction of (97.5 ± 0.3)%, (97.7 ± 0.4)% and (96.0 ± 0.7)%, for total coliforms, fecal coliforms and fecal streptococci, respectively. The performance of the filter was not affected a lot when changing the hydraulic head from 250 L/m²/day to 500 L/m²/day. The bacterial load concentrations at the filter outlet are slightly higher than the WHO recommended standard (1000 CFU / 100mL) for reuse of wastewater in irrigation. To remedy this situation, we recommend the adoption of the principle of treatment of excreta at the source by the use of dry toilets and the treatment of gray water only by the V-MSL filter.

1 Introduction

In rural areas of Morocco, the population generally uses traditional autonomous drainage systems to get rid of black water and practices the direct discharge of gray water into nature without treatment [1]. With the increase in the supply of drinking water in recent years (97%), the amount of wastewater generated is very large. In addition, open defecation remains the only way for the population without toilets [1-2]. These practices still present a major risk for public health and the environment; exposing many people to infection and dangerous diseases [3-5]. Indeed, human excrement can contain pathogens such as bacteria and parasites capable of causing diarrheal diseases (cholera, typhoid, etc.) [6].

The treatment of wastewater in these areas requires particular attention in the choice of purification technique to be used [7]. In these areas, wastewater treatment system strategies are needed which are environmentally, socially and economically sustainable [8-9]. Currently, a sustainable approach that holds interesting potential for decentralized sewage treatment is multi-soil-layering (MSL) system [10]. The first MSL system was developed in Japan (1990), to treat wastewater [11-12]. This system was also tested in China, Thailand and the USA, for the treatment of domestic wastewater [13], polluted river water [14], and leachate [15] etc. The MSL system is typically composed of soil mixture blocks (SMB) arranged in a brick-like pattern and surrounded by water-permeable particles, such as zeolite and gravel [10]. This system is economic because it can be developed mainly from local resources such as soil, sawdust, iron, charcoal, gravel and alternative materials and required only a small land area [9,16-19]. According to Guan et al., [20], this new system was less prone to clogging supporting a higher hydraulic load rate (HLR). Some authors suggested that effective contact between the wastewater and SMB increased the MSL performance [14, 21].

As a promising treatment technology and a cost-effective solution for sustainable water management, it is essential to evaluate its performance in Moroccan conditions. To do this, a pilot-scale of vertical Multi-Soil-Layering system (V-MSL) was designed to treat domestic wastewater next to a single house. The aim of this study is to evaluate the ability of the V-MSL system to remove the fecal pollution indicators from domestic wastewater under three hydraulic loading rates (250 L/m²/day, 350 L/m²/day and 500 L/m²/day) in Moroccan conditions.

2 Materiel and methods

2.1 Description of treatment system

The pilot-scale system used in this study is a vertical Multi-Soil-Layering filter (V-MSL) measuring 20 cm deep, 60 cm wide, 78 cm high with a feeding tank (100L) used to store prescreened wastewater from the...
inlet of the household wastewater using submersible pump. This is installed next to the guardian’s house at Razi’s middle school (Meknes, Morocco). Three hydraulic loads were tested (HLR1: 250 L/m²/day, HLR2: 350 L/m²/day and HLR3: 500 L/m²/day). The characteristics of this system are illustrated in figure 1. The choice of materials used in our MSL system, is mainly related to their availability and cost in Morocco. Indeed, the permeable layer (PL) is composed by the gravel with a diameter between 3 to 5 mm. Their role is to improve the water distribution and reduce the risk of clogging. While the SMBs measuring 20 cm (L) x 16.5 cm (W) x 4 cm (H), are composed by 60% of soil, 10% of charcoal, 10% of iron sawdust and 20% of wood sawdust.

The filling of the filter is done in the following order: the first layer (20 cm thick) is composed by the pebbles followed by the first permeable layer (5 cm thick of gravel) followed by the first layer of bricks (SMBs); the alternation of the permeable layers and the brick layers succeed one another until the sixth permeable layer. The bricks are arranged horizontally with a space of 5 cm between them.

2.2 Analysis of fecal indicator bacteria in influent and effluent

After the stabilization period (three months), samples of raw and treated water were collected at the same time each month (from June 2017 until April 2018) to perform bacteriological analyzes. These samples were collected in sterile 500 mL glass vials and stored in a refrigerated cooler (± 4 °C). These analyzes were carried out in the BETA laboratory of Dr. BENGOUMI Driss in Meknes. The following table summarizes the various bacteriological parameters analyzed as well as the method undertaken (Table 1).

| Parameter          | Technique                                      | Reference     |
|--------------------|------------------------------------------------|---------------|
| Total coliforms (TC) | Count by membrane filtration (0.45 μm) on Tergitol agar with TTC | ISO 9308-1:2014 [22] |
| Fecal coliforms (FC) | Count by membrane filtration (0.45 μm) on Tergitol agar with TTC | ISO 9308-1:2014 [22] |
| Fecal streptococci (FS) | Count by membrane filtration (0.45μm) on Slanetz and Bartly agar | ISO 7899-2:2000 [23] |

Table 1. Bacteriological parameters and methods of analysis

The statistical data treatment was carried out by software: Excel 2010 and SPSS 20. Analysis of variance (ANOVA) was used to evaluate the significant difference between the raw water samples and the treated water as well as between the different hydraulic loading rates used.

3 Results and discussions

3.1 Concentrations of the fecal pollution indicators

The mean, maximum and minimum concentrations of the fecal pollution coliforms in domestic wastewater to be treated were respectively, 

\[(1.31 ± 0.15) \times 10^6\ CFU/100mL, \ 1.53 \times 10^6 \ CFU/100mL \text{ and } 1.03 \times 10^6 \ CFU/100mL\] for total coliforms, 

\[(1.05 ± 0.19) \times 10^6 \ CFU/100mL, \ 1.33 \times 10^6 \ CFU/100mL \text{ and } 6.80 \times 10^5 \ CFU/100mL\] for fecal coliforms and 

\[(1.62 ± 0.41) \times 10^5 \ CFU/100mL, \ 2.53 \times 10^5 \ CFU/100mL \text{ and } 1.17 \times 10^5 \ CFU/100mL\] for fecal streptococci (Table 2).

| Parameter | Mean | SD | Max. | Min. |
|-----------|------|----|------|------|
| TC        | 1.31 x10⁶ | 1.55 x10³ | 1.53 x10⁶ | 1.03 x10⁶ |
| V-MSL     | 3.20 x10⁴ | 1.40 x10¹ | 3.42 x10⁴ | 2.93 x10⁴ |
| Removal % | 97.5 | 0.3 | 97.9 | 96.9 |
| FC        | 1.05 x10⁶ | 1.94 x10³ | 1.33 x10⁶ | 6.80 x10⁵ |
| V-MSL     | 2.36 x10⁴ | 2.08 x10¹ | 2.67 x10⁴ | 1.95 x10⁴ |
| Removal % | 97.7 | 0.4 | 98.2 | 96.6 |
| FS        | 1.62 x10⁸ | 4.11 x10⁴ | 2.53 x10⁸ | 1.17 x10⁸ |
| V-MSL     | 6.27 x10³ | 1.00 x10¹ | 8.40 x10³ | 4.94 x10³ |
| Removal % | 96.0 | 0.7 | 97.02 | 94.9 |

(TC: total coliforms; FC: fecal coliforms; FS: fecal streptococci; SD: Standard Deviation)

Fig. 1. Schematic representation of Vertical Multi-Soil-Layering (V-MSL)
The variability of bacterial concentration during the study period is mainly linked to the quality of the organic load of the wastewater to be treated and the hydraulic loading rates applied (Fig. 2).

After treatment, the average concentrations of these fecal indicators were \((3.20 \pm 0.14) \times 10^4\) CFU / 100 mL, \((2.36 \pm 0.21) \times 10^4\) CFU / 100 mL and \((6.27 \pm 1.00) \times 10^3\) CFU / 100 mL for total coliforms, fecal coliforms and fecal streptococci, respectively (Table 2 and Fig. 2). The V-MSL filter ensured an average reduction of \((97.5 \pm 0.3)\%\), \((97.7 \pm 0.4)\%\) and \((96.0 \pm 0.7)\%\), for TC, FC and FS, respectively (Table 2).

The ANOVA test shows that there is a very highly significant difference between raw water and treated water regarding the average bacterial loads of TC, FS and FC (Table 4).

Table 3. Analysis of variance (ANOVA) of bacterial concentrations between inflow and outflow

| Parameter | Inflow (I) | Outflow (J) | Difference (I-J) | P≤0.001 |
|-----------|------------|-------------|------------------|---------|
| TC        | DWW        | V-MSL       | 1 274 460.8***   | .000    |
| FS        | DWW        | V-MSL       | 155 541.5***     | .000    |
| FC        | DWW        | V-MSL       | 1 029 429.3***   | .000    |

*** Signification at p<0.001

DWW: domestic wastewater

I: concentration of parameter in inflow

J: concentration of parameter in outflow

3.2 Reduction of fecal pollution indicators depending on the hydraulic load used and the season

The average removal rates of fecal pollution during the study period under the three hydraulic loads tested (HLR1, HLR2 and HLR3) were respectively 97.40\%, 97.51\% and 97.67\% (for the TC), 97.54\%, 97.76\% and 97.80\% (for FC) and 96.57\%, 96.13\% and 95.32\% (for FS) (Fig. 2). Despite the differences in the reduction rates reported between the hydraulic loads applied to the filter, the ANOVA test did not show any significant difference between these rates for the TC (p>0.21), FS (p>0.45) and FC (p>0.24). The average removal rates of fecal pollution according to the seasons (summer to spring) were 97.4\%, 97.5\%, 97.6\% and 97.7\%, respectively, for TC; 97.2\%, 97.8\%, 97.8\% and 98.0\% for FC and 96.6\%, 96.1\%, 95.8\% and 95.5\% for FS (Fig. 2).

Despite the differences in reduction rates reported between seasons, the ANOVA test did not show any significant difference between these rates for TC (p>0.5), FS (p>0.7)and FC (p>0.1); however, this test revealed a significant difference between summer and spring for FC.

In general, the V-MSL filter has shown significant efficiency in eliminating fecal pollution indicators and it has also shown some stability in terms of performance during all seasons.

Fig. 21. Reduction of fecal pollution indicators depending on the hydraulic load and season
The high removal rates recorded by our V-MSL system can be explained by the high dissolved oxygen concentrations within the filter during the study period, as well as the water temperature which was relatively optimal throughout the study period (especially in summer and fall).

The mean elimination of fecal pollution indicators ranged from 1.24 ± 0.18 to 1.64 ± 0.07 log units (Table 4). Indeed, the total coliforms were reduced by 1.61 ± 0.05 log units, 1.64 ± 0.07 log units for fecal coliforms, and 1.40 ± 0.08 log units for fecal streptococci. These results are better than those reported by El Hamouri et al. [24] on 1.40 ± 0.08 log units for fecal streptococci. These results are also better than those reported by Eturki et al., [25] with their infiltration and percolation horizontally constructed wetland, to those obtained by Eturki et al., [25] with their infiltration and percolation horizontally constructed wetland, to those obtained by Morató et al. [28] who obtained removal rates in the order of 4.13 and 4.47 log units for the reduction of coliforms. Despite the difference between the three hydraulic loading rates tested (97.7 ± 0.4) % and (96.0 ± 0.7) %, for TC, FC and FS, respectively. Generally, the abatement of these indicators decreased with increasing of hydraulic loading rates. On the other hand, the retention time of wastewater in the filtration system increases with the increase in the applied hydraulic loading rate, giving the system sufficient time to adsorb, react and remove organic pollutants and the bacterial load from the wastewater [9].

Filtration and adsorption could be the first bacteria elimination mechanisms in the MSL system followed by the other reduction process such as predation by other organisms (eg, protozoa, nematodes, etc.) and natural death. In fact, predation by the intervention of protozoa, nematodes and bacteriophages has been proven to play an essential role in the elimination of germs indicative of fecal pollution in vertical flow filters by several studies [35-39].

### Table 4. Reduction of fecal bacterial indicators expressed by log10 per 100 mL during the study period

|       | TC         | FS         | FC         |
|-------|------------|------------|------------|
| Summer|            |            |            |
| HLR1  | 1.50       | 1.50       | 1.47       |
| HLR2  | 1.57       | 1.53       | 1.61       |
| HLR3  | 1.68       | 1.38       | 1.58       |
| Mean+SD | 1.59±0.09  | 1.47±0.08  | 1.56±0.07  |
| Fall  |            |            |            |
| HLR1  | 1.54       | 1.48       | 1.63       |
| HLR2  | 1.62       | 1.44       | 1.67       |
| HLR3  | 1.62       | 1.32       | 1.69       |
| Mean+SD | 1.60±0.05  | 1.41±0.08  | 1.66±0.03  |
| Winter|            |            |            |
| HLR1  | 1.63       | 1.48       | 1.64       |
| HLR2  | 1.61       | 1.34       | 1.63       |
| HLR3  | 1.63       | 1.33       | 1.68       |
| Mean+SD | 1.62±0.01  | 1.38±0.08  | 1.65±0.03  |
| Spring|            |            |            |
| HLR1  | 1.69       | 1.40       | 1.74       |
| HLR2  | 1.61       | 1.36       | 1.69       |
| HLR3  | 1.60       | 1.29       | 1.58       |
| Mean+SD | 1.63±0.05  | 1.35±0.06  | 1.70±0.03  |

On the other hand, our results corroborate those of Morató et al. [28] who obtained removal rates in the order of 1.2-2.2 log units for total coliforms, 1.4-2.2 log units for fecal streptococci in their planted constructed wetland filters.

Almost similar results were obtained by Arias et al. [29] concerning the abatement of TC and FC in two-stage of vertical constructed wetland filters with pretreatment and post-treatment in a calcite filtration unit in Denmark. They reported that the first bed of their system had the highest efficiency with 1.5 log and 1.7 for TC and FC, respectively. However, the results reported by Latrach et al. [19] are clearly better than ours with their hybrid system combining MSL technology and sand filters, they obtained reduction rates of the order of 4.13 and 4.47 logarithmic units for the reduction of coliforms. Despite the performance given by their hybrid system, these authors reported that the sand filters were clogged after two months of operation with hydraulic loads of 100, 200 and 400 L/m²/day.

The moderate efficiency obtained by the present study could be due, on the one hand, to the high porosity of the gravel which is used as a permeable layer (diameter between 3 and 5 mm); on the other hand, to the hydraulic load used which is relatively high and to the short residence time of the water to be treated within our filter (approximately one hour). The same observation is reported by Stevik et al. [30] and Ausland et al. [31] who concluded that germ infiltration is influenced by the physical characteristics of the filter substrate and by the hydraulic loading rate used. In the same context, Bomo et al. [32] showed a significant improvement in the elimination of pathogenic bacteria in the case of fine sand (d10 = 0.25) rather than coarse (d10 = 0.86) filters. Additionally, Vacca et al. [33] concluded that the grain size of the filter substrate affects the elimination of bacteria. Torrens et al. [26] also confirmed that bacterial reduction in porous media is provided by filtration and adsorption of the bacterial load to the filter substrate. Along the same lines, Stevik et al. [30]; Garcia et al. [34] have shown that fine materials such as sand, clay and silt (very fine pores) act as a filtration system for bacteria. This could be explained by the hydraulic retention time which is high in filters with fine substrate compared to filters with coarse substrate.

Therefore, the hydraulic residence time appears to be a crucial parameter for the performance of the system. On the other hand, the retention time of wastewater in the filtration system increases with the decrease in the applied hydraulic loading rate, giving the system sufficient time to adsorb, react and remove organic pollutants and the bacterial load from the wastewater [9].

Filtration and adsorption could be the first bacteria elimination mechanisms in the MSL system followed by the other reduction process such as predation by other organisms (eg, protozoa, nematodes, etc.) and natural death. In fact, predation by the intervention of protozoa, nematodes and bacteriophages has been proven to play an essential role in the elimination of germs indicative of fecal pollution in vertical flow filters by several studies [35-39].

### 4 Conclusion

The V-MSL used in this study was able to achieve the reductions of the fecal pollution indicators: (97.5 ± 0.3) %, (97.7 ± 0.4) % and (96.0 ± 0.7) %, for TC, FC and FS, respectively. Generally, the abatement of these indicators decreased with increasing of hydraulic loading rates. However, the ANOVA test did not show any significant difference between the three hydraulic loading rates tested at p <0.05. On the other hand, despite the difference reduction rates reported between seasons, this test did not show any significant at p <0.05, except between summer and spring for FC.

The bacterial load concentrations at the filter outlet are slightly higher than the WHO recommended standard
(1000 CFU / 100mL) for reuse of wastewater in irrigation. To remedy this situation, we recommend the adoption of the principle of treatment of excreta at the source by the use of dry toilets and the treatment of gray water only by the V-MSL filter.

Acknowledgements. Our thanks to the principal and school safety agent (Razi, Meknes) for their assistance.

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