

SPONTANEOUS MACROFAUNA BIODIVERSITY IN VERTICAL CONSTRUCTED WETLANDS IN OCCITANIA

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ABSTRACT

Introduction

For several decades now, nature-based solutions such as constructed wetlands and artificial marshes are reputed to be robust solutions for wastewater treatment and recycling. All these solutions have in common an operation inspired by that of natural wetlands and the water quality regulation service they provide, in order to purify wastewater by recreating the natural functioning of an ecosystem as close as possible to that of a wetland (Vymazal, 2016; Vymazal et al., 2021). They are nature-based infrastructures that biomimic biogeochemical processes occurring in natural wetlands. The Vertical Constructed Wetlands (VCWs), called also Filtres Plantés Verticaux, invented by Käthe Seidel in Germany in the 1950s, quickly demonstrated the ability of these devices to treat different types of wastewater of domestic, agricultural and even industrial origin under certain conditions (Molle et al., 2004; Lombard-Latune et al., 2020). Used alone or in addition to secondary treatment, VCWs are now recognized in all countries as an effective, inexpensive and sustainable alternative to conventional treatment plants in specific cases of difficult access or low population concentration (Masi et al., 2018).

VCWs are basically built with layers of sands and gravels with a known granulometry to perform the hydraulicity. However, more than the space requirement per population equivalent, the main risk is the clogging effect due to the permanent input of organic matter from wastewaters. Because earthworms are known for their ability to reduce clogging through the bioturbation effect, these ecological engineers are increasingly seen as an asset for optimizing the water purification function of VCWs (Gerino et al., 2022). However, very few studies have focused on the macrofauna role in such systems (Zhang et al., 2020 ; Gilibert et al., 2022), missing a possible positive impact of VCWs on soil biodiversity management.

Materials and method

In this purpose, we studied spontaneous colonization by macrofauna in 5 VCWs built in the Lauragais Region, Occitania, between 2017 and 2020 (so 2 to 5 years old). In each VCW, 1.6 dm³ of soil was randomly cored

at 3 locations to a depth of 15 cm, in April and May 2022. Macrofauna specimens were recovered by sieving and scanning under a stereo microscope. Then they were identified down to family or order level, using identification keys (Loriot, 2004 ; Noël et Séchet, 2007 ; FCPN, 2018 ; Mignon et al., 2021 ; Fort, 2022).

At last, we discuss earthworm's behavior observed in the experimental VCW platform of Toulouse Campus (BioROC project, 2022-2025), inoculated at 500 g/m² of *Eisina* genus with 3 species *E. Faetida*, *E. Andreis* and *E. Hortensis*, based on the results from Gilibert et al. (2022). Four VCW have been set up with soilscan device : one buried soil imaging device was placed diagonally in the soil of four VCWs, from 5 to 30 cm depth as described in Belaud et al. (2024). The soilscan device consists in a desktop planar optical scanner (Epson V39 Perfection (Epson™, Japan)). The devices were waterproofed and connected to a computer for automatic image capture. The A4 image format (21 × 29.7 cm) was set with a resolution of 1200 dpi. The images were captured at 6-hour intervals (12 am, 6 am, 12 pm and 6 pm).

Results and discussion

The collected macrofauna belonged to 11 orders (Oligochaeta, Coleoptera, Hymenoptera, Diptera, Hemiptera, Mites, Chilopoda, Diplopoda, Arachnida, Isopoda and Gasteropoda), from 6 classes (Annelida, Insecta, Myriapoda, Chelicerata, Crustacea and Mollusca). These orders match with the natural soils and wetlands macrofauna (Plum, 2005). In details, sampled macrofauna was distributed as follows (Fig.1) : 62% Lumbricidae, 9% beetles including 3% larvae (mostly from the Staphyllinidae family), 7% Formicidae, 7% dipteran larvae (mostly from the Tipulidae family) and undetermined pupae, 5% myriapods (Lithobiomorphich chilopoda, Cryptopidae scolopendromorphs and Geophilomorphs, and a few diplopods), 5% isopods (from the Armadillidiidae and Trichoniscidae families and *Philoscia* genus), 3% mites, 1% hemipterans from the Lygaeidae family, 1% slugs and less than 1% arachnids (spiders and pseudoscorpions).

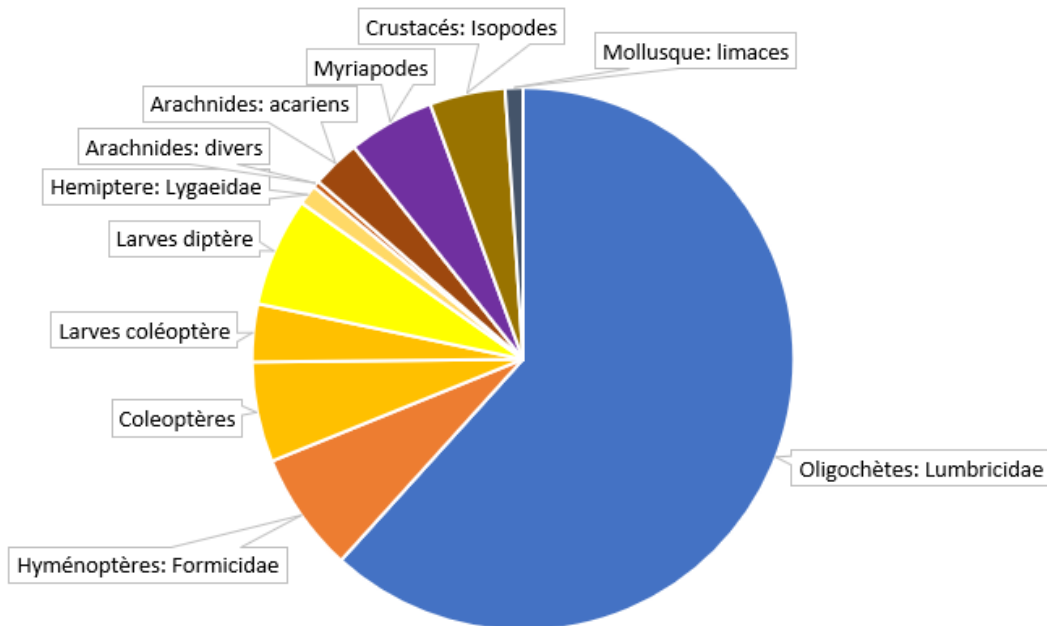


Figure 1: Macrofauna distribution per species of 5 VCWs built between 2017 and 2020 by Epurtek in the Lauragais Region (Occitania, South France) and sampled in April-May 2022.

This macrofauna distribution is part of the macrofauna of natural wetlands (Plum, 2005). A study carried out in constructed wetlands in Ivory Coast (Ouattara et al., 2009) also recorded the presence of Oligochaeta, Coleoptera, Hymenoptera, Diptera, Lepidoptera, Hemiptera, Diplopoda, Arachnida, Isopoda and Gasteropoda, Dermaptera and a proportion of annelids (annelida Oligochaeta) much more important (93%) than found in this study (62%). It's probably due because the age of the constructed wetlands. In Ouattara et al. (2009), the studied infrastructures are very young, only 150 days after the wastewater treatment, against several years in this study.

The greatest taxonomic richness (up to 22 taxa) was observed in the oldest VCW, and taxonomic richness decreased with VCW age. The total density of soil macrofauna in studied VCWs ranged between 15,000 and 39,600 ind/m³. Such results demonstrate that VCWs are rapidly spontaneously colonized by macrofauna. So VCWs can be considered such as living soil system, despite the initial construction with only sands and gravels.

Another important point is the large dominance of Lumbricidae (62%), that confirms the great adaptation capacity of earthworms to difficult environmental conditions linked to both heavy organic pollution and intermittent flooding due to regular inputs of domestic wastewaters by baches. Indeed, Gilibert et al. (2022) have demonstrated in mesocosms study that the optimal worms biomass density to avoid the clogging effect and to facilitate the sub-surface water flow was around 500 g/m². And in their study, it seems *Eisenia* genus would be the best adapted to VCW environmental condition. These 3 species are epigenic engineers that dig different galleries networks with geometries that varies according to soil depth, the deeper being *E. hortensis*. In the VCWs of Toulouse Campus, the soilscan devices give evidence of subsurface occurrence of worms, in the top 20-25 cm of sediment, even though the worms are able to burrow much deeper in experimental column.

The biomass in the VCWs of Toulouse Campus has more than doubled after 2 months of experiment. And the size of the worms has been changing during the experiment with a majority of adults that was introduced at the initial time, and a majority of juveniles that was found at the end of the experiment. These results indicate a capacity of resistance of this population to environmental perturbation (industrial or building renovation or pharmaceutical punctual waste input), probably also with possible “eggs” that permit to keep the population in dormancy during the perturbation and regeneration when the conditions return favorable.

At last, it's known that the microfauna diversity and activity is dependent on macrofauna diversity, particularly bioengineers such as earthworms (Lavelle, 1997 ; Gérino et al., 2022 ; Orange et al., 2022). Indeed macrofauna facilitates the biodegradation through two ways: by degrading complex biopolymers into easily biodegradable components for the microfauna, and by increasing aeration and water flows in the subsurface soil.

Conclusion

The first and main result of this study is to confirm VCWs can be considered as a reserve of soil biodiversity, due to the spontaneous colonization by invertebrates community where the oligochetes are the dominate species. Because of the rapid geographical expansion of VCWs in the world, they could balance the loss and degradation of natural wetlands. These results demonstrate the key role such an interstitial macrofauna community may play as main actors of the bioturbation processes known to strongly influence physical, chemical and biological properties on the surrounding substrate of the VCWs. In this way, the “in-filter” macrofauna community should be intricately involved in pollutants remediation in such systems, downward oxygen transfer into the soil and thus increasing the bioavailability of soil contaminants.

On the other hand, our results suggest a resilience capacity of the earthworms population in the VCWs that allow the maintaining of the VCWs functioning and performance over the time. We confirm the potentially importance of introducing earthworms into VCWs to improve their wastewater purification performance.

In addition, the demonstration of the involvement of this earthworms population in the long term performance of the VCWs should be the next demonstration to be made, together with identification of the perturbation threshold not to be exceeded to keep a safe environment for these organisms. More in-depth research on the effects of such macrofauna biodiversity on mesofaunal and microbial activities is now required to improve our knowledge on biodiversity use as a tool to transform and remove emerging pollutants, as it's currently tested on two experimental sites of the BioROc project : in the VCWs pilots built on the Toulouse Campus, and in the VCWs from the wastewater treatment plant of Murviel near Montpellier.

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