D8. Functional tailored prototype of Watersave+ at Barcelona Zoo

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Content

1. Watersave+ pilot description

Before the construction of the Watersave+ pilot treatment system, the land was prepared. For this reason, all existing vegetation was removed, necessary earthworks were carried out to create the proposed platforms, and the entire surface was leveled and compacted. It was very important for these systems that the soil where all the elements would be placed was stable, as there was water circulation between the systems. The earthworks were performed manually, trying to fit the structures as much as possible into the existing space. The Watersave+ pilot system comprises four vertical subsurface flow (VSSF) treatment wetlands and two horizontal subsurface flow (HSSF) treatment wetlands (Figure 1) built with stainless steel. Additionally, a designated sampling point is established at the outflow of each system to facilitate monitoring and analysis. *Phragmites australis* reeds were planted with a density of 10 units per square meter. Annex 1 provides photographic documentation of the Watersave+ pilot.

Due to the design of the Watersave+ pilot system, a 1m³ reservoir tank was installed to supply the pilots. An agitator was incorporated into the tank to ensure uniform wastewater homogenization. The inflow wastewater is transported from each animal installation to the pilot. The Watersave+ pilot operates through a remotely controlled automatic feeding program, optimizing efficiency and consistency.

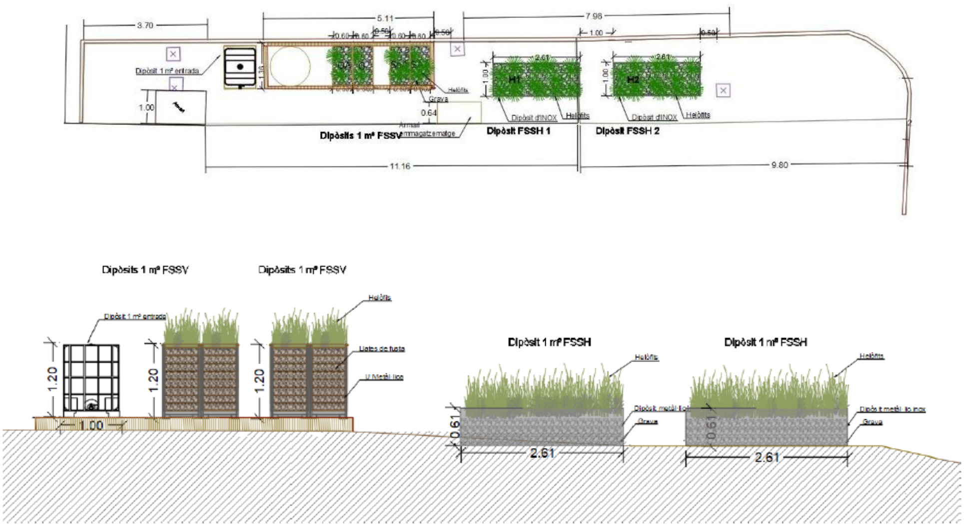


Figure 1. Watersave+ pilot scheme

The differences between the VSSF treatment wetlands were based on the quality of the supply wastewater (Figure 2). Two systems, filled with gravel (V1 and V2), were designed to treat wastewater with higher organic and suspended solids loads. One of these gravel-filled systems was equipped with an artificial aeration system (V1), while the other one did not have an aeration system (V2). The other two VSSF treatment wetlands, filled with sand (V3 and V4), were intended for wastewater with a low organic and suspended solids load, differing only in their system height: V3 has a depth of 1.20 m and V4 has a depth of 0.75 m.

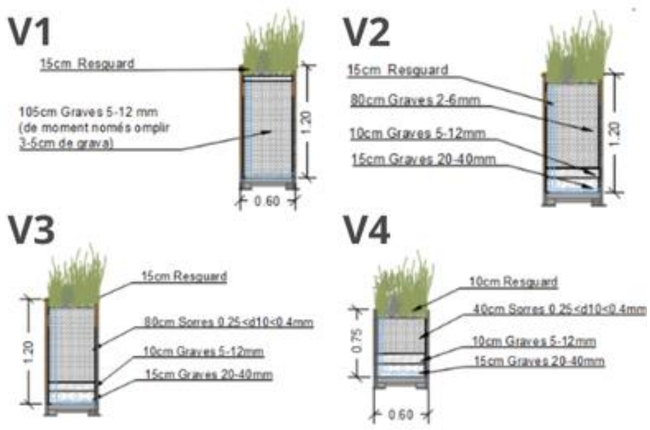


Figure 2. Vertical subsurface flow treatment wetland description.

The horizontal subsurface flow (HSSF) systems were divided into two types based on their filling materials. H1 consisted of an HSSF system filled with gravel, while H2 featured an HSSF system filled with a combination of different organic matter (OM) sources (compost and woodchips) and sand (Figure 3).

Figure 3. Horizontal subsurface flow treatment wetland description.

The Watersave+ pilot system is monitored and automate feeding with the Hydrawise program (Figure 4). The system is monitored and controlled via a smartphone, tablet, or computer, allowing for remote adjustments to optimize water usage and maintain consistent operation.

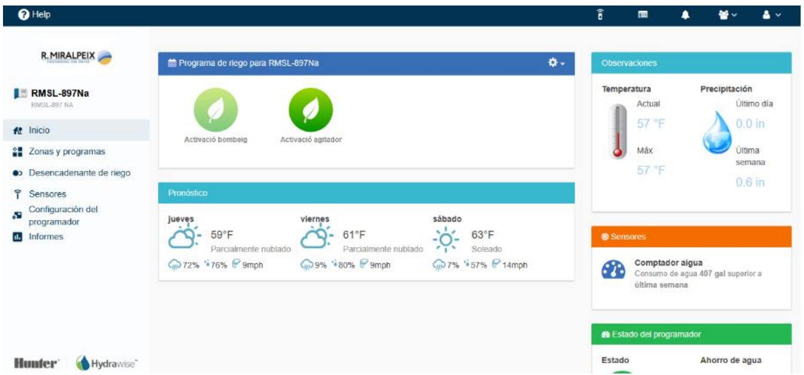


Figure 4. User interface of Hydrawise program.

2. Experimental design

Different treatment lines will be implemented based on the characteristics of the wastewater (Table 1). For wastewater with high Biological Oxygen Demand (BOD_5) and Suspended Solids (SS), the VSSF filled with gravel (V1 and V2) will be operated in parallel to analyse the impact of aeration on wastewater treatment. Alternatively, for wastewater with low BOD_5 and SS, the VSSF filled with sand (V3 and V4) will be operated in parallel to assess the effect of prototype size differences. Furthermore, the various treatment lines will be implemented during different seasons to analyse the effect of temperature on the depuration efficiency of the Watersave+ pilot.

2.1 Wastewater quality from animal facilities

The following animals facilities were selected for the study: Pygmy Hippopotamus, Crocodylus, Tapir, Chilean Flamingo, ornamental Fish pond and Spider Monkey. The selection was based on the quantity and quality of wastewater used.

V1 and V2 will be used to treat wastewater called 'Quality 1', while V3 and V4 will be used to treat wastewater called 'Quality 2'.

- Quality 1 (High Pollution Load): This category includes wastewater from the facilities of the Pygmy Hippopotamus, Spider Monkey, and Chilean Flamingo. Based on the wastewater characterization conducted in 2024, the average values for Quality 1 wastewater are 132 mg/L of BOD_5 and 290 mg/L of SS.
- Quality 2 (Low Pollution Load): This category includes wastewater from the Tapir, Crocodylus, and Ornamental Fishpond facilities. The results of the wastewater characterization for Quality 2 show an average of 57.5 mg/L of BOD_5 and 8 mg/L of SS.

The wastewater from each animal facility will be treated individually using the different treatment lines.

2.2 Watersave+ pilot management

Several factors will be considered in the pilot management, including the eight treatment lines (four for each type of wastewater), feeding strategy, hydraulic load, and retention time. These variables will be carefully monitored and adjusted to optimize the performance of the system, ensuring efficient treatment and addressing the specific needs of each wastewater type. The management approach will also consider the interactions between these factors to achieve the best possible outcomes for water quality and system sustainability.

2.2.1. Treatment lines

The treatment lines were designed to address the two types of wastewaters (Quality 1 and Quality 2), with four distinct treatments tested for each wastewater type. Each treatment line consists of two stages: the first stage using the VSSF, followed by the second stage through the HSSF. Table 1 presents an overview of the characteristics of the treatment lines. Each treatment line will be operated for a duration of 21 days to assess performance under controlled conditions. Additionally, the systems will be thoroughly drained between different wastewater types to prevent cross-contamination.

Table1. Treatment line description for Watersave+ pilot management.

Nº treatment line	Type of wastewater	1st stage	2nd stage
A	Quality 1 (High pollution load)	V1	H1
B		V2	H2
C		V1	H2
D		V2	H1
E	Quality 2 (Low pollution load)	V3	H1
F		V4	H2
G		V3	H2
H		V4	H1

2.2.2. Hydraulic load and feeding strategy.

At the beginning of the implementation, the hydraulic load (HL) will be set at 20 cm/day/m and gradually increased to 60 cm/day/m for each VFFS. The increase or decrease of the HL will be changed depending on the efficiency; reduced if signs of clogging or low performance are detected or increased if the performance is exceptionally high. The pilot will be fed discontinuously, the flow tested will be 10 L/min in each VFFS and the number of batches depend on the HL. Table 2 describes the feeding strategy of each HL.

Table 2. Hydraulic load and feeding strategy.

HL (m/day)	Q (m^3/day)	Number of batches per day	Feeding time (min)	Time between batches (min)
0.20	0.144	15	1	96
0.60	0.432	45	3	32

2.2.3. Hydraulic retention time

Hydraulic monitoring was conducted by measuring the volume of water flowing into and out of each system in March 2025. Regular measurements will help identify variations in hydraulic behavior, allowing for adjustments to improve overall treatment effectiveness. Table 3 describe the theoretical hydraulic retention time using the size of the systems, the porosity of the filling material and the flow rate.

Table 3. Theoretical hydraulic retention time in each system.

Wetland	Porosity	Area (m^2)	Effective depth (m)	Q (m^3/day)	THRT (day)
V1	0.40	0.72	1.05	0.144	2.10
V2	0.30	0.72	1.05	0.144	1.57
V3	0.33	0.72	0.80	0.144	1.32
V4	0.33	0.45	0.40	0.144	0.41
H1	0.33	2.61	0.50	0.144	2.99
H2	0.45	2.61	0.50	0.144	4.07

Additionally, to evaluate the hydraulic retention time (HRT) in each step of each treatment line, a tracer experiment will be conducted using Amino G, a fluorescent dye commonly used for hydraulic studies. The experiment aims to determine the wastewater flow behavior and assess retention efficiency. The experiment will be carried out in the following steps:

1. *Tracer selection and preparation:* a known concentration of Amino G will be diluted in a precise volume of wastewater to create the tracer solution.

2. *Tracer injection:* The tracer solution will be introduced as a pulse injection at the inflow of each treatment line. The injection will be performed at a single time point to track its movement through the system.

3. *Sampling strategy:* Wastewater samples will be collected at different points, including inflow of the VSSF, outflow of the VSSF (before entering the HSSF), and final outflow of the HSSF. Frequent samples will be taken over time to capture the tracer's movement and concentration changes.

4. *Measurement and data collection:* The concentration of Amino G will be measured using a fluorometer, which detects fluorescence intensity. Measurements will be recorded at regular intervals until the tracer is no longer detected in the outflow.

5. *Data analysis:* A breakthrough curve (tracer concentration vs. time) will be plotted for each step. Finally, the mean HRT will be calculated based on the time required for the tracer to appear and reach peak concentration at the outflow.

2.2.4. Experimental schedule

Following the initial hydraulic test conducted in March 2025, the schedule for the first experimental test using wastewater from animal installations was developed (Table 4). This schedule outlines the procedures and timeline for testing the system with this specific wastewater source, allowing for the assessment of its performance under real-world conditions. The experimental test aims to evaluate the system's response to the characteristics of animal installations wastewater, including any potential variations in flow, treatment efficiency, and system behavior.

Table 4. Experimental test schedule for Watersave+ pilot implementation.

Date	Activity	Description
March 01 - 31	Initial hydraulic test and preparation phase	Executed the initial hydraulic test to assess the system's flow characteristics and performance, while setting up and calibrating the system for the subsequent experimental test
April 01 – 21	Test 1: treatment line A and B	Wastewater treatment for the pygmy hippopotamus will be conducted through lines A and B, operating in parallel, with a hydraulic loading rate (HLR) of 0.20 m/day.
April 29 – May 19	Test 2: treatment line C and D	Wastewater treatment for the pygmy hippopotamus will be conducted through lines C and D, operating in parallel, with a HLR of 0.20 m/day.
May 20 – June 09	Test 3: treatment line E and F	Wastewater treatment for the tapir will be conducted through lines E and F, operating in parallel, with a HLR of 0.20 m/day.
June 10 - 30	Test 4: treatment line G and H	Wastewater treatment for the tapir will be conducted through lines G and H, operating in parallel, with a HLR of 0.20 m/day.
July 01 - 21	Test 5: treatment line A and B	Wastewater treatment for the Chilean flamingo will be conducted through lines A and B, operating in parallel, with a HLR of 0.20 m/day.
July 22 – August 11	Test 6: treatment line C and D	Wastewater treatment for the Chilean flamingo will be conducted through lines C and D, operating in parallel, with a HLR of 0.20 m/day.
September 09 - 29	Test 7: treatment line E and F	Wastewater treatment for the Crocodylus will be conducted through lines E and F, operating in parallel, with a HLR of 0.20 m/day.
September 30 – October 20	Test 8: treatment line G and H	Wastewater treatment for the Crocodylus will be conducted through lines G and H, operating in parallel, with a HLR of 0.20 m/day.

Each test implemented have a total of 30 samples:

- Inflow: 6
- Outflow V1 or V3: 6
- Outflow V2 or V3: 6
- Outflow H1: 6
- Outflow H2: 6

The schedule will be adjusted or modified based on the system's performance and efficiency results.

3. Water quality monitoring

Different parameters will be monitored to evaluate the efficiency of each wetland, with a focus on analyzing the inflow and outflow of each system. Table 5 provides an overview of the microbiological and physicochemical parameters that will be used in the water quality monitoring process. These parameters are essential for evaluating the effectiveness of the treatment systems and ensuring that water quality standards are met throughout the testing period.

Table 5. Microbiological and physico-chemical parameters for general monitoring.

Microbiological parameters for general monitoring	Methodology	Interest
E. coli	ISO 9308	Indicator of faecal contamination
Clostridium perfringens	ISO 14189	Indicator of faecal contamination, giardia cysts and cryptosporidium oocysts
Somatic coliphages	ISO 10705	Indicator of virus
Enterococci	ISO 7899	Indicator of faecal contamination
Parasites	Microscopic and molecular analysis	

Physico-chemical parameters for general monitoring		
pH*	Electrical conductivity*	TSS**
COD***	BOD5**	TP****
NH_4^+- N****	NO_2^- - N****	NO_3^- - N****
TN***		

*pH meter and electrical conductivity meter Crison
**Standard methods
***kit
****Ionic chromatography

Parameters for general monitoring will be assessed twice a week, while others will be analyzed once a week (see Table 6). In addition to these basic parameters listed previously, targeted analyses will be conducted to assess the system's effectiveness in removing emerging contaminants of concern.

Table 6. Monitoring parameters analysed and their analysis frequency.

Frequency of analysis	Parameter	Laboratory
Twice per week	*E.coli*	UB
	Total coliforms	UB
	Enterobacteria	UB
	Clostridium perfringens	UB
	Coliphages	UB

	SST	UB
	COD	UdG
	TN	UdG
	NH_4^-	UdG
	NO_2	UdG
	NO_3^-	UdG
	PO_4	UdG
Once per week	Parasites	UB
	BOD_5	UB

4. Further actions

To further enhance the treatment systems, several actions are planned based on the results from the initial tests. These next steps will focus on assessing how temperature fluctuations affect the performance of the systems. The upcoming tests will replicate the treatment lines under controlled conditions, with adjustments to key variables to evaluate the systems' efficiency and adaptability. These actions are essential for optimizing system performance and ensuring it can handle varying environmental conditions effectively.

Based on the results of the various tests and the efficiency of each treatment line, additional tests will be conducted using both wastewater quality types (Quality 1 and Quality 2), with an increase in the hydraulic loading (HL) rate. The objective of this modification is to determine the maximum level of efficiency that can be reached by each system under varying conditions.

The following objectives include replicating the same treatment lines with identical characteristics in January and February 2026 to evaluate the impact of temperature variations on the systems' performance and operation. This will help assess how temperature influences the efficiency and behavior of the treatment processes under different climatic conditions.

Annex 1. Photographic documentation of Watersave+ pilot

- Construction phase

HSSF treatment wetlands

VSSF treatment wetlands

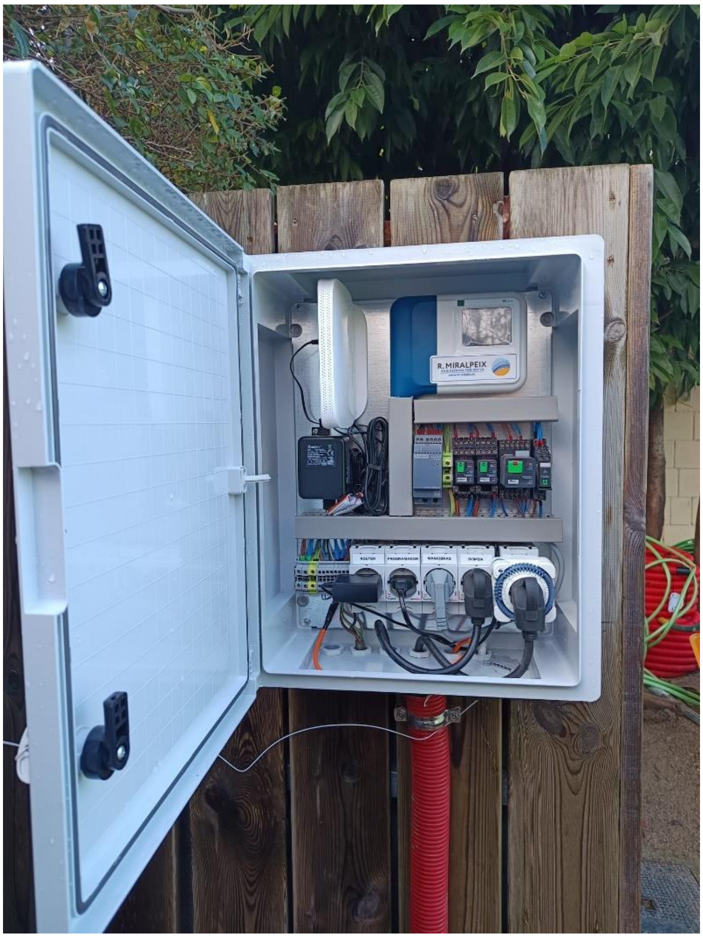
Aeration system

Implementation phase

Reservoir tank with agitator

VSSF treatment wetlands

HSSF treatment wetlands



Control system

Aeration system implementation