SINGULAR PROJECT

DEVELOPMENT AND DIVERSIFICATION PROJECT
FROM USEFUL NATIVE AQUATIC PLANT

Introduction

Within the Singular Project framework, a list of requirements for new potential species in relation to bio filtration, mechanics and landscape capacity was made. From here, Viver Tres Turons, the Barcelona Botanical Garden team and the other participants of the project, selected different native species with potential for functioning in natural purification systems. The management team of the URL decided to use the 12 short channels to the mentioned project.

The URL (Urban River Lab) is an experimental place located at the Montornès del Vallès WWTP (Barcelona) where a multidisciplinary team formed by members of the Center for Advanced Studies of Blanes (CEAB-CSIC), University of Barcelona (UB), Besòs-Tordera Consortium (CBT) and Naturalea, are conducting different research projects that evaluate the effects of WWTP effluents on river systems. (www.urbanriverlab.com)

It was decided to use three channels to each specie.

Sparganium erectum and *Apium nodiflorum* work as the species that have been used so far in the URL and therefore are planted in a substrate of gravel, which allows us to already have a control channel (only gravel) and to be able to compare it with other species.

Potamogeton pectinatus has another function, so in addition to the channels used to study this specie, a channel without plants has been recreated in three channels that will serve as a control treatment. These channels consist of a bed with 5 cm of gravel and mud and the rest with a sheet of water about 25 cm deep.

Background

The studies realized in this laboratory in 2016 show that the presence of helophytes (reed, lily...) creates favorable conditions for the microorganisms that are associated with the rhizosphere, which substantially increase the retention capacity of solutes and therefore improve the quality of wastewater or effluent from sewage treatment plants. Specifically, before Singulars Project the following species were studied:

- *Thypa angustifolia*
- *Lysimachia vulgaris*
- *Iris pseudacorus*
- *Scirpus lacustris*
- *Phragmites australis*

They were planted on a 20-40 cm gravel substrate such as those used in natural purification systems and with sub-surface flow.

The experimental design consisted of three channels of each species and 3 channels with the same substrate but without plants (control), in order to evaluate the effect of the presence of plants and the possible differences between the plant species.

Material and method in the Singular Project

A total of 7 samples were performed for this project over 6 months of experiment (August 19-January 20). At the beginning of the experiment it was sampled more frequently (biweekly) and the rest monthly.

At the inputs and outputs of each channel, pH, conductivity and dissolved oxygen were measured and water samples were taken to determine the concentrations of ammonium, nitrates, nitrites, phosphorus, DOC.

<u>Variables to study:</u>

1. Nutrients Retention: Water samples were taken at the inlet and outlet of each channel to measure nutrient retention capacity (ammonium, nitrate, nitrate and phosphate) and organic matter (DOC).
2. Measurement of the evapotranspiration rates in the canals: the flow of water at the inlet and outlet of each channel was measured with a bucket, a stopwatch and a balance to determine the % of water loss.
3. Measurement of water residence times: Water residence time was calculated using bromide (Na Br) additions.
4. Measurements of biomass and elemental content (CNP) over time: the biomass of each plant species was measured at the beginning, middle and end of the study period and plant tissue was stored to measure the content of C, N and P.
5. Metabolism measurements: In the channels with surface flow, the oxygen concentration was measured over time with O_2 probes..

<u>Investigation team of URL that has taken part in the project:</u>

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Plantation of *Sparganium erectum* and *Apium nodiflorum:*

Preparation of the control channels for the submerged macrophytes:

Plantation of *Potamogeton pectinatus:*

Channels with Apium and Sparganium once they have grown:

Plants at the end of the experiment (December 19).

Results

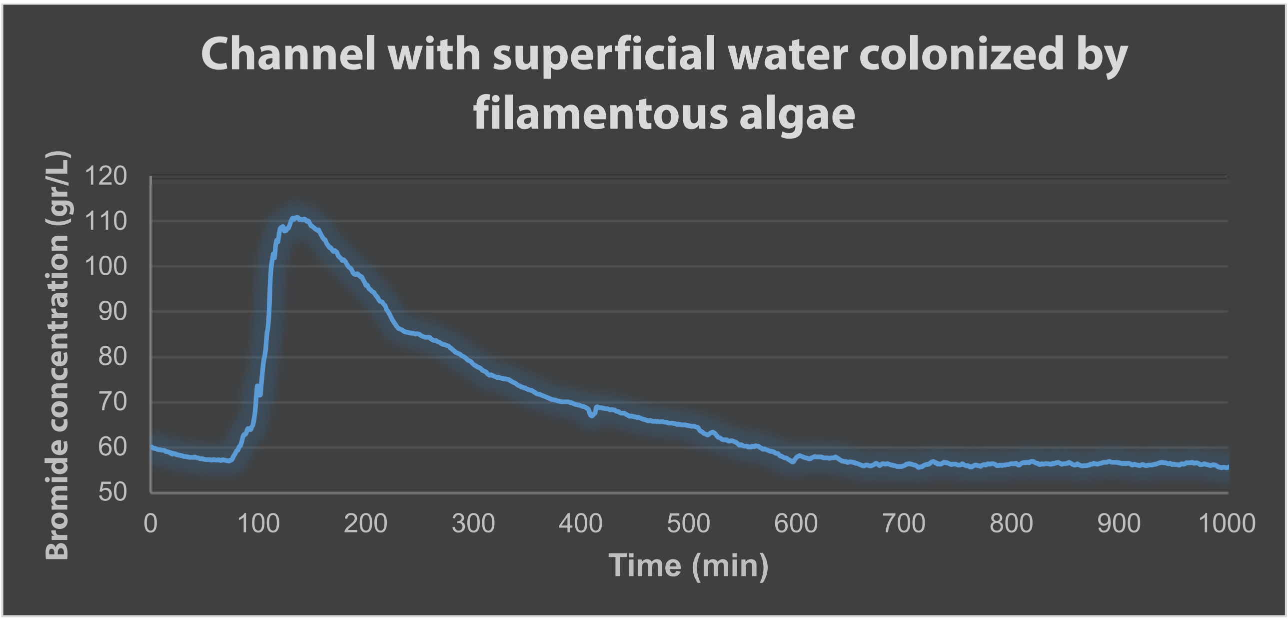
Evapotranspiration rates:

Treatment	Evapotranspiration rates (%)
Control	2,7
Potamogeton pectinatum	2,6
Apium nodiflorum	1,5
Sparganium erectum	3,3

Summary table of the rates of evapotranspiration in the different treatments after two months of the planting. For each treatment, the average of the 3 channels is presented. To calculate the evapotranspiration rates, the flow of water at the inlet and outlet of each channel is measured and the percentage of variation of the flow at the outlet with respect to the inlet is calculated.

Time of residence of the water

Example of bromide vs. concentration curve time to measure the "residence time" of the water within each channel. The peak of the curve indicates the time required for the water entering the channel to move to the end of it. We call it "nominal travel time". This curve corresponds to Channel 1 (Control channel full of filamentous algae) 1 month after planting.

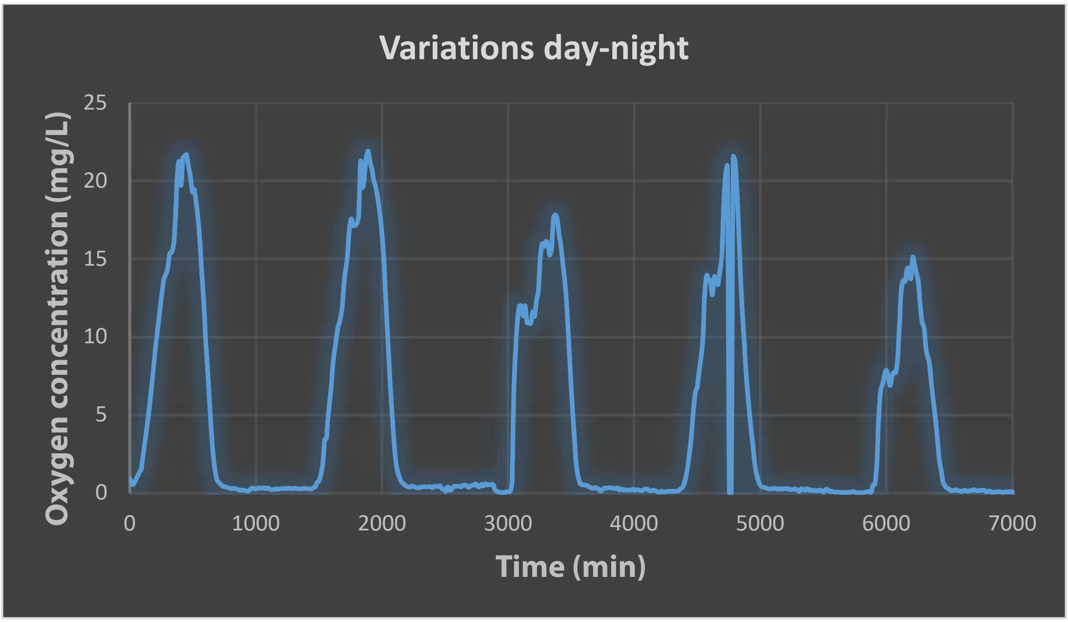


Treatment	Residence time of the water (min)
Control	137
Potamogeton pectinatum	207
Apium nodiflorum	274
Sparganium erectum	180

Summary table of the residence times of the water in the different treatments after one month of planting. For each treatment, the average of the 3 channels is presented.

Daily changes in oxygen in the channels

Channel with *Potamogeton pectinatus* colonized by filamentous algae



Firsts results of nutrient retentions

Summary

Channel	Treatment	SRP	NO_2	NH_4	NO_3	DOC
Ch 1	Control	-3,10	-250,25	2281,00	1620,58	-60,00
Ch 2	Potamogeton	-1,03	-0,70	57,43	170,18	643,33
Ch 3	Sparganium	-9,29	216,17	1334,38	8,40	1260,00
Ch 4	Apium	1,03	126,06	1794,73	-225,51	292,00
Ch 5	Control	7,23	-310,48	2109,88	-317,02	-233,33
Ch 6	Potamogeton	-24,78	140,07	3382,63	2271,89	686,67
Ch 7	Sparganium	-7,74	117,66	2252,28	773,64	335,33
Ch 8	Apium	-8,26	-28,01	1216,49	483,93	539,67
Ch 9	Control	-3,10	-77,04	640,11	-229,71	-183,33
Ch 10	Potamogeton	19,62	-136,80	398,73	-877,29	473,33
Ch 11	Sparganium	19,62	146,60	1992,69	-514,51	1083,67
Ch 12	Apium	15,49	52,29	1755,98	-1104,66	360,67

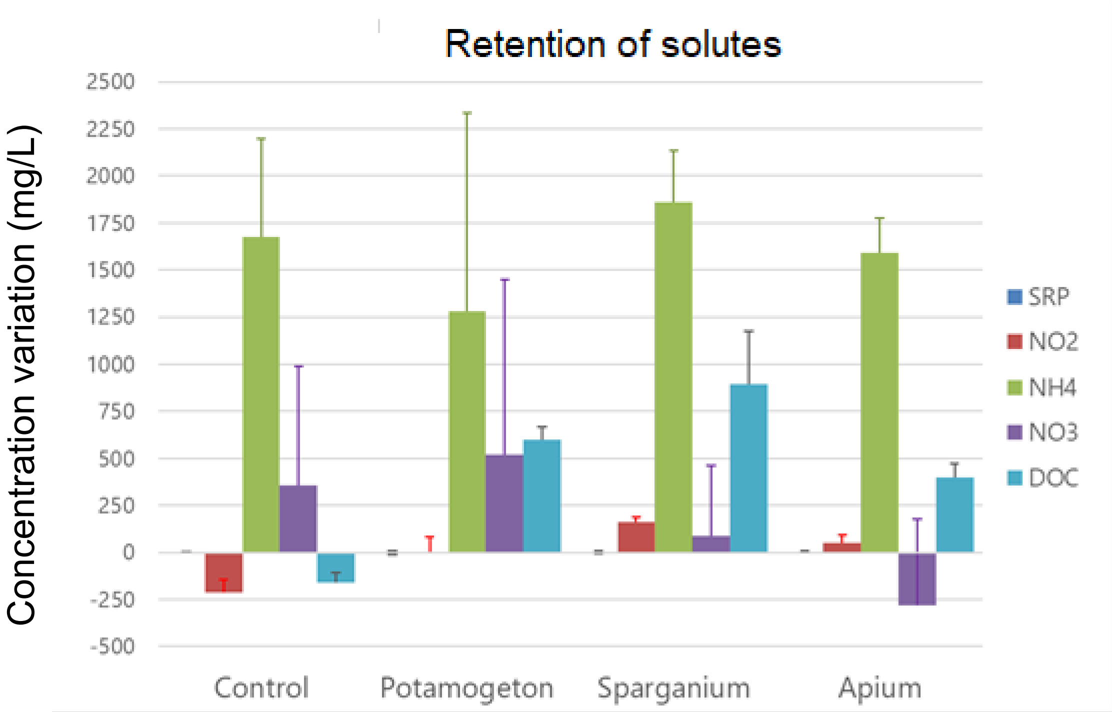
Average

Treatment	SRP	NO_2	NH_4	NO_3	DOC
Control	0,34	-212,59	1677,00	357,95	-158,89
Potamogeton	-2,06	0,86	1279,59	521,60	601,11
Sparganium	0,86	160,14	1859,78	89,18	893,00
Apium	2,75	50,11	1589,06	-282,08	397,44

Standard error

Treatment	SRP	NO_2	NH_4	NO_3	DOC
Control	3,44	69,97	520,79	631,82	51,51
Potamogeton	12,83	79,93	1056,12	925,91	65,10
Sparganium	9,39	29,23	273,18	374,04	283,44
Apium	6,91	44,49	186,63	459,46	73,82

Average retention of the different solutes: phosphorus (srp), nitrite (NO2), ammonium (NH4), nitrate (NO3) and dissolved organic carbon (DOC) for each of the plant species considered: *Potamogeton pectinatus, Sparganium erectum and Apium nodiflorum*. No plants were placed in the control canals, however, at the time of sampling they were colonized by filamentous algae and water lentil. The concentration variation in each channel was calculated as the concentration at the inlet minus the concentration at the outlet. Positive values indicate a decrease in concentration at the outlet of the channel relative to the inlet (net retention of solute in the channel), and negative values indicate an increase in concentration throughout the channel (net release of the solute in the channel). Each plant species was planted in 3 different channels (3 replicates per species) and in each channel 3 inlet and outlet samples (3 replicates per channel) were taken. The mean values and the variation (standard error) per sampling are shown for each plant and solute species. Data corresponding to the sampling made on September 27, 2019.



Conclusions

1. In conditions of well-oxygenated water, ammonium is transformed into nitrates and nitrites by nitrification.
2. Anoxia conditions should favor the removal of N through denitrification.
3. During the day, there is primary production that causes the increasing of oxygen concentration, oxidative conditions and this causes an increase in pH. At basic pH, the P tends to precipitate and ammonia passes into ammonia which is lost in the atmosphere (ammonification).
4. During the night there is shortness of breath, low oxygen concentration, reduced conditions and low pH causing resuspension of P.
5. *Apium nodiflorum* reduces evapotranspiration rates, which is of great interest in the Mediterranean region and in the context of the climate crisis because allow a higher rate of water reuse. This is also due to the high residence time of the water, which in principle can favor purification processes. Another important aspect of this species is that it stays green and active all year.
6. *Sparganium erectum* is the most efficient of the species worked on to retain nutrients (especially ammonium) and dissolved organic matter.

Preliminary results of this experiment suggest that *Sparganium* is a specie with potential for use in natural purification systems along the lines of common helophytes such as reed and yellow lily. In very warm places, *Apium nodiflorum* can also be a very interesting specie. In the case of *Potamogeton pectinatus* due to the fact that the load of the waters to be treated has many nutrients, they generate biofilm which affects its capacity and therefore it is not a specie indicated in waste water purification, although it can be very useful in bathing waters because it has a good efficiency without so much substrate.