

Article

Sustainable Production of Reclaimed Water by Constructed Wetlands for Combined Irrigation and Microalgae Cultivation Applications

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Abstract: Considering the increasing pressure on freshwater resources due to the constant increase in water consumption and insufficient wastewater control and treatment, recovering wastewater is a path to overcoming water scarcity. The present work describes the potential of reusing treated wastewater (reclaimed water) for irrigation and production of microalgae biomass in an integrated way, through experimental evaluation of plant and microalgae growth, and creation of an application model. First, two parallel experiments were conducted to evaluate the use of reclaimed water produced by a constructed wetland filled with a mix of solid waste: the irrigation of a set of small pots filled with soil and planted with *Tagetes patula* L., and the cultivation of microalgae *Chlorella* sp. and a mixed microalgae population with predominant species of the genus *Scenedesmus* sp. in shaken flasks and tubular bubble column photobioreactors. Results indicated no negative effects of using the reclaimed water on the irrigated plants and in the cultivated microalgae. The growth indicators of plants irrigated with reclaimed water were not significantly different from plants irrigated with fertilized water. The growth indicators of the microalgae cultivated with reclaimed water are within the range of published data. Second, to apply the results to a case study, the seasonal variability of irrigation needs in an academic campus was used to propose a conceptual model for wastewater recovery. The simulation results of the model point to a positive combination of using reclaimed water for the irrigation of green spaces and microalgae production, supported by a water storage strategy. Water abstraction for irrigation purposes can be reduced by 89%, and 2074 kg dry weight microalgae biomass can be produced annually. Besides the need for future work to optimize the model and to add economical evaluation criteria, the model shows the potential to be applied to non-academic communities in the perspective of smarter and greener cities.

Keywords: microalgae production; irrigation; reclaimed water; treatment wetlands; wastewater reuse; water circularity; sustainability



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1. Introduction

The increasing demands for water resources for multiple purposes such as agriculture, industry, public water supply, recreational uses, and other applications are leading to water scarcity and the deterioration of freshwater quality [1]. The intensification of severe weather conditions, such as droughts [2], and urban development has also put a significant strain on freshwater supplies [3].

Water scarcity is one of the great challenges of the current century, a concern reflected in the United Nations' 6th Sustainable Development Goal: "Ensure the availability and sustainable management of water and sanitation for all" [4]. Water scarcity can be mitigated through various strategies and technologies, including more eco-efficient industrial production approaches to reduce water consumption and loss [5], as well as in water supply systems [6], urban users [7], and irrigation of crops or green spaces [8]. The production

of reclaimed water is another approach that is being given relevance, particularly at the European level [9,10].

The use of reclaimed urban and industrial water, which refers to water that has been used once, has subsequently undergone treatment processes, and is then intentionally used again, is an example of potential contribution to partial circularization [11]. Although reuse of all wastewater generated by domestic and industrial activities is unfeasible due to technical and economic barriers or even environmental issues, available treatment technologies can guarantee sufficient water quality parameters to provide its use for irrigation purposes [12].

Producing and using reclaimed water is aligned with both sustainable water management and circular economy, allowing [13] (i) reduction in freshwater consumption, which contributes to mitigating water scarcity; (ii) decrease of discharges in water resources, which reduces the risk of eutrophication of watercourses; and (iii) reduction of the consumption of nutrients and minerals, particularly of some already scarce, such as phosphorus.

Reclaimed water can be used in different ways such as for washing of industrial and urban places, firefighting, irrigation of crop fields and green spaces, and algae production, among other non-drinking purposes [14–18]. However, production of reclaimed water from wastewater requires more advanced technology than conventional primary and secondary operations typically implemented in industrial and urban wastewater treatment plants (WWTP), and costs may be a handicap particularly for low-developed countries and regions [19–21].

Constructed wetlands (CWs), which consist of a green and affordable technology for the treatment of wastewater, can be an alternative to costly conventional wastewater treatment technologies [22]. In addition to the recovery of water, CWs are effective to recover valuable nutrients, such as nitrogen and phosphorus compounds [13].

CWs consist of engineered systems based on the same processes that occur in natural wetlands, but especially improved by control over the hydraulic load and flow arrangement, and the design of its main components [23]: (i) a retention basin or structure; (ii) a granular filling media; and (iii) the planted vegetation (macrophyte plants). The granular filling media is a relevant component to the treatment efficiency of sub-superficial flow CWs, being the medium for plant root development, since the CWs vegetation contributes to wastewater treatment by assimilation of nutrients, by supporting the development of biofilms, and by oxygenation of the roots environment [24,25]. The filling medium may also contribute directly to wastewater treatment by physical and chemical processes, such as filtration, adsorption, precipitation, as well as by supporting the development of biofilms [26,27]. Usually, CWs are filled with sand, gravel, or similar materials, combined or not with soil [28]. Using solid waste as filling media may contribute to enhancing the sustainability of the CW technology and represent a circular economy of waste [29], but the ecotoxicity risk of using these solids should be addressed.

Using reclaimed water for irrigation purposes can be a straightforward application, considering that treated water may contain the required amounts of nutrients and micronutrients required for plant growth and does not contain harmful substances for the plants and the environment [16,30]. Using reclaimed water avoids or diminishes the requirements for artificial fertilizers, contributing to surface water and aquifer protection, which may be degraded by excessive use of crop fertilization [31].

Considering the recovery of nutrients from wastewater, reclaimed water can also be a potential growth medium for microalgae production. Microalgae may be used to produce a variety of valuable products, including food, feed, cosmetics, pharmaceuticals, bioplastics, and biofuel raw materials [32–35]. When downstream processing of the algal biomass is not economically feasible, the produced biomass can be used for composting [36,37]. However, its large-scale production still presents some technical constraints, such as the need for high amounts of water and the low concentrations of biomass obtained. To overcome

these issues, using wastewater or reclaimed water can be a contribution to increasing the eco-efficiency of the algal biomass production process [38–40].

This work describes the use of reclaimed water for irrigation of green spaces and the production of algae biomass, exploring the potential of CWs for tertiary wastewater treatment. This work also considers the responsibility of higher education institutions to promote actions under sustainable development goals, acting as examples for the academic and surrounding community. Moreover, academic campuses are examples of small cities, making it possible to implement solutions to promote smart city principles. Production and use of reclaimed water can be an essential pillar of smart communities, among other smart city foundations. Thus, the paper is organized considering the following two complementary works. First, it describes the experimental work carried out to evaluate the potential of using reclaimed water produced by a pilot-scale modular constructed wetland filled with solid waste to irrigate ornamental plants (French marigold), chosen as representative of green spaces plants, and two cultures of microalgae: one pure culture of commercial species (*Chlorella* sp.) and one mixed culture obtained from wastewater environments, with a predominance of *Scenedesmus* sp. Besides evaluating the reclaimed water potential to replace algae culture media and fertilized water, the experiments aim to evaluate the toxic effects of the eventual leaching of harmful compounds from the solid waste that fills the CWs. Second, a model is proposed for combining reclaimed water production by CWs simultaneously with green space irrigation and microalgae production, using the campus of the Polytechnic Institute of Tomar (IPT) as a case study, applying the results of the experimental work.

IPT is a higher education institution located in Central Portugal that offers postsecondary, bachelor's, and master's programs in social and management sciences, arts, and engineering. Besides, as this work results from a project-based learning approach, it intends to drive the development of green behavior among the enrolled students, which is the first step to spread environmental best practices by the community concerning water protection. Sustainable growth and related issues have been selected as the core content of IPT's mission, driving all academic initiatives and being the core for new curricular contents and research projects. As an example, the Valorbio project, with several research activities developed under project-based learning [41], and its ongoing continuation projects (SmarterCW and CW4GreenerCities) focus on sustainable development issues such as freshwater protection, solid waste and nutrient recovery, and renewable energy sources. In addition, those projects aim to improve the participation of students and involve the academic community through its connection to curricular programs and life on the campus.

2. Materials and Methods

The experimental work was carried out to evaluate the reusing of treated wastewater for irrigation of ornamental plants and the production of algae biomass. Urban-type wastewater after secondary pretreatment was subjected to tertiary treatment by a pilot-scale CW filled with a mix of solid waste from significant Portuguese industrial sectors, namely, fragmented residues from Moleanos limestone extraction and processing activities, coal slags from coal power plants, and cork granulates, a waste from cork-processing industries. The pilot-scale CW was developed under the Valorbio research project [41]. Details on the design and experimental setup of the modular-type pilot-scale CW, characteristics of the solid waste used as filling media, and the main results obtained are described elsewhere [42–45].

Two parallel experiments were conducted to evaluate the potential of using the reclaimed water produced by the pilot-scale CW: for the irrigation of a set of small pots filled with soil and planted with *Tagetes patula* L., and for the cultivation of microalgae *Chlorella* sp. and a mixed microalgae population isolated from the CWs itself, and therefore already adapted to wastewater, with predominant species of the genus *Scenedesmus* sp. The main goal was to evaluate harmful effects in plant and microalgae growth due to compounds eventually liberated into the water by the solid waste filling of the CWs. As

far as the authors are aware, there are no data available in the literature on the risk of inhibitory effects occurring from using reclaimed water produced by CW filled with a mixture of solid waste.

2.1. Analytical Methods

Wastewater and treated water samples were analyzed according to the standard analytical methods [46]. Temperature and pH were measured with an HI-98194 multiparameter meter and respective probes supplied by Hanna Instruments. The analysis of total nitrogen (TN), total phosphorous (TP), chemical oxygen demand (COD), and water color was carried out using an HI-83399 spectrophotometer and an HI-839800 reactor block, both supplied by Hanna Instruments. Standard procedures were used, as highlighted by the supplier. The 5-day biochemical oxygen demand (BOD₅) determination was carried out according to the standard methods for the examination of water and wastewater [46]. The counting of *Escherichia coli* bacteria was made according to the standard ISO 9308-1:2014. Total suspended solids (TSS) was determined by filtration of 100 mL of sampled water through glass fiber filters, according to the standard ISO 11923:1997. Metal ion content of reclaimed water was determined by a certified laboratory using inductively coupled plasma optical emission spectrometry (ICP-OES).

Microalgae biomass concentrations (dry weight) were measured from optical densities using a UV–vis spectrophotometer at 570 nm (CADAS 100 spectrometer; Hach, formerly DRLange, Düsseldorf, Germany) and previously achieved calibration curves for each alga.

Specific growth rate (day^{-1}) was calculated by linear regression of the exponential growth phase concentrations versus time, allowing the calculation of the doubling time ($\text{DT} = \mu / \ln 2$). Biomass productivity (mg/L/day) was estimated for the exponential growth phase ($\Delta Y / \Delta t$). Relative lipid content and biomass productivity were used to calculate the relative lipid productivity (mg/L/day). To estimate lipid content, Fourier transform infrared spectroscopy (FTIR) was conducted according to Liu and Bangert [47]. Relative lipid content was determined by calculating the ratio of the lipid 1740 (cm^{-1}) band to the amide II (1540 cm^{-1}) band.

The content in chlorophyll a and b and carotenoid of microalgae were determined by the method of methanol extraction described by Yu et al. [48]. First, 1.5 mL culture media was sampled into a 2 mL Eppendorf tube and centrifuged ($9000 \times g$, 10 min). Then, the supernatant was replaced by 1.5 mL of methanol. The tube was incubated for 24 h in the dark at 45 °C. After that, the solution was centrifuged ($9000 \times g$, 10 min) and the absorbance of the supernatant was measured at wavelengths of 470, 652.4, and 665.2 nm on a UV–vis spectrophotometer (CADAS 100 spectrometer; Hach, formerly DRLange, Düsseldorf, Germany). The photosynthetic pigment concentrations and carotenoids in mg/L were calculated from Equations (1) to (3). Total chlorophyll content was obtained adding results of Equations (1) and (2).

$$\text{Chlorophyll}_a = 16.72 \text{ Absorbance}_{665.2 \text{ nm}} - 9.16 \text{ Absorbance}_{652.4 \text{ nm}}, \quad (1)$$

$$\text{Chlorophyll}_b = 34.09 \text{ Absorbance}_{652.4 \text{ nm}} - 15.28 \text{ Absorbance}_{665.2 \text{ nm}}, \quad (2)$$

$$\text{Carotenoids} = (1000 \text{ Absorbance}_{470 \text{ nm}} - 1.63 \text{ Chlorophyll}_a - 104.9 \text{ Chlorophyll}_b) / 221. \quad (3)$$

The content in chlorophyll a and b of plants' leaves was determined by the method of acetone extraction described by Porra et al. [49]. First, approximately 50 mg of leaves were extracted with 1 mL 80% acetone and the photosynthetic pigments were determined by a spectrophotometer assay method. Absorbance was read at both 663.6 and 646.6 nm on a UV–vis spectrophotometer (CADAS 100 spectrometer; Hach, formerly DRLange, Düsseldorf, Germany). Quantification of the photosynthetic pigment content was performed using Equations (4) and (5), for chlorophyll concentrations in mg/L , and converted to fresh weight basis in $\mu\text{g/g}$.

$$\text{Chlorophyll}_a = 12.25 \text{ Absorbance}_{663.6 \text{ nm}} - 2.55 \text{ Absorbance}_{646.6 \text{ nm}}, \quad (4)$$

$$\text{Chlorophyll}_b = 20.31 \text{ Absorbance}_{646.6 \text{ nm}} - 4.91 \text{ Absorbance}_{663.6 \text{ nm}} \quad (5)$$

At least two replicates were made of all assays and measurements. Statistical tests were performed using IBM SPSS® software, version 24. T-test and post hoc tests were performed at a significance level of 95% ($p = 0.05$). The Solver functionality of the Microsoft Excel® 365 was used to numerically solve the case study model. Uncertainty intervals ($\pm$) were calculated with a 95% confidence level.

2.2. Wastewater Characteristics and Treatment Setup

The pilot-scale CW consists of a modular system based on a structure of wood supporting a geomembrane bag with 60 cm height $\times$ 120 cm width $\times$ 99 cm length. The CW is filled with four layers of solid waste, from bottom to top: 5 cm of fragmented residues of limestone rock; 10 cm of coal slag; 25 cm of cork granulates; and 10 cm of fragmented residues of limestone rock. This stratified layer combination of solid waste was optimized in previous works to benefit from the combined hydraulic properties and contributions to removing pollutants from wastewater [44,45].

The CW is continuously fed with secondary urban-type pretreated wastewater. The average flow rate of produced water is $40 \text{ L day}^{-1} \text{ m}^{-2}$. Table 1 shows the average composition of the wastewater.

Table 1. Characterization of the wastewater before the treatment by the pilot-scale constructed wetland.

Parameter ¹ $\rightarrow$	COD (mg/L)	BOD ₅ (mg/L)	TN (mg/L)	TP (mg/L)	TSS (mg/L)	NTU	pH
Result	269 ± 8	130 ± 40	72.9 ± 0.6	1.5 ± 0.7	39.0 ± 0.3	11.4 ± 0.7	8.2 ± 0.1

¹ Analytical parameter: COD = chemical oxygen demand; BOD₅ = biochemical oxygen demand, 5 days; TN = total nitrogen; TP = total phosphorous; TSS = total suspended solids; NTU = nephelometric turbidity units.

The reclaimed water was then analyzed to assess its quality for reuse and subsequent use in the irrigation experiment (Tables 2 and 3). The reclaimed water meets the requirements of class A recycled water (Portuguese Decree-Law. No. 119 of 2019; EU Regulation 2020/74 of the European Parliament and the Council of 25 May 2020 on minimum requirements for water reuse).

Table 2. Quality parameters of the reclaimed water after tertiary treatment by the pilot-scale constructed wetland.

Parameter ¹ $\rightarrow$	COD (mg/L)	BOD ₅ (mg/L)	TN (mg/L)	TP (mg/L)	TSS (mg/L)	NTU	pH	Fluoride (mg/L)	<i>E. coli</i> ³
Result	29	4.2	35	0.96	6.9	2.0	7.2	0.0	0
NQAR ²	n.r.	≤ 10	n.r.	n.r.	≤ 10	≤ 5	6–9	≤ 2.0	≤ 10

¹ Analytical parameter: COD = chemical oxygen demand; BOD₅ = biochemical oxygen demand, 5 days; TN = total nitrogen; TP = total phosphorous; TSS = total suspended solids; NTU = nephelometric turbidity units. ² Portuguese standards for A-type reclaimed water for irrigation purposes (n.r. = not required). ³ *Escherichia coli* colony forming units per 100 mL.

Table 3. Contents in heavy metals of the reclaimed water after tertiary treatment by the pilot-scale constructed wetland.

Parameter $\rightarrow$ (mg/L)	Al	B	Be	Co	Fe	Li	Mn	Mo	Se	V
Result	0.0	<0.01	<0.01	<0.02	0.25	0.0	0.19	0.0	<0.02	<0.02
NQAR ¹	≤ 5	²	≤ 0.1	≤ 0.05	≤ 2.0	≤ 2.5	≤ 0.2	≤ 0.01	≤ 0.02	≤ 0.1

¹ Portuguese standard for A-type reclaimed water for irrigation purposes. ² Depending on crop sensitivity.

2.3. Irrigation Experiments

Four pots with 10.8 cm height, 11.5 cm width, and 23.8 cm length (270 cm² of area) and filled with soil collected from the IPT's campus were planted with six shoots of *Tagetes patula* L. (usually known as French marigold) each. Two pots were regularly irrigated with tap water with dissolved fertilizer, as control sets, and two pots were irrigated with the reclaimed water. Plant growth indicators were evaluated at the end of the experiments, three months after planting.

The fertilized water was prepared with an NPK-type 12-4-6 liquid fertilizer containing secondary macronutrients and micronutrients (VITAL, Epagro, Portugal). The fertilized water content in TN, TP, and K was respectively 24.0, 1.75, and 4.98 mg/L. It also contained the secondary macronutrients Mg and SO₃ and the micronutrients, B, Cu, Fe, Mn, Mo, and Zn, being free of Cl.

Three months after planting, the plants were harvested and the following parameters were evaluated: the number of flowers and buds, length of shoots and roots, amount of plant biomass, and levels of chlorophyll a, b, and total chlorophyll in the leaves.

2.4. Microalgae Cultivations

The evaluation of algal cultivation in the treated wastewater was carried out in 250 mL shaken flasks and lab-scale bubble column photobioreactors (PBRs). The PBR consists of a vertical cylinder made of crystal-clear acrylic material, with a 1.4 L working volume. The reactors' dimensions are 495 mm height, 70 mm inner diameter, and 3 mm wall width. The PBR is closed on the top and bottom by two acrylic plates sealed with O-rings.

Two algae genera were used: *Chlorella* sp. obtained from the Coimbra Collection of algae (ACOI) and a mixed population of microalgae, with predominant species of the genus *Scenedesmus*, isolated from the CWs used for the wastewater treatment. The mixed population was examined morphologically using light microscopy. A water sample was collected from the CW, the sample was immediately inoculated and successively sub-cultivated in 100 mL conical flasks with BG11 medium [50], at room temperature (between 15 and 25 °C), and exposed to the natural light cycle for enrichment and selection of the autotrophic microorganisms. Samples of the subcultures were observed at an optical microscope, leading to the conclusion that the genus *Scenedesmus* represents the predominant microalgae [51].

The cultivations were conducted indoors for two weeks under a daily photoperiod of 12 h provided by tubular lamps of 20 W and 2000 lux. The temperature ranged from 15 to 25 °C.

The culture medium consisted of the reclaimed water with no addition of freshwater or other constituents such as nutrients, oligo-elements, or vitamins. The experiments were all conducted in duplicate.

Samples were collected daily and immediately analyzed for biomass concentration. The pigment and lipid content were determined at the end of the experiments.

3. Results and Discussion

3.1. Irrigation Results

It was found that the wastewater treated by CWs fulfills the water quality requirements to be reused for irrigation under the Portuguese Decree-Law. No. 119 of 2019, and was classified as class A water, which means it can be used to irrigate green spaces without restrictions.

French marigold plants were used here as an example of vegetation of irrigated gardens and green spaces. Table 4 presents the results obtained in the irrigation tests of the plants, where it can be observed that the plants irrigated with the recovered water show, after three months, a development slightly superior to those irrigated with fertilized water. Regarding the production of chlorophyll pigments, plants watered with fertilized water have a higher total chlorophyll content. However, none of the results obtained for the

plants irrigated with reclaimed water are statistically different from the results obtained for the plants irrigated with fertilized water.

Table 4. Growth indicators of French marigold plants irrigated with reclaimed water compared to results obtained for the control sets ¹.

Growth Indicator	Type of Irrigation Water	
	Reclaimed	Fertilized
Wet weight (g)	4 ± 1	4 ± 2
Plant height (cm)	11 ± 1	12 ± 1
Root length (cm)	9 ± 3	9 ± 2
Flowers per plant	1.4 ± 0.7	1.3 ± 0.6
Sprouts per plant	0.8 ± 0.5	0.8 ± 0.6
Chlorophyll a (µg/g)	1210 ± 130	1476 ± 107
Chlorophyll b (µg/g)	412 ± 70	526 ± 73

¹ All results for each growth indicator are not different under a statistical level of 0.05.

Besides demonstrating the potential to replace artificial fertilizers with reclaimed water, the experiments demonstrate that no ecotoxicity effects result from using solid waste as filler in the CWs. Using solid waste as CW filler is a significant improvement in enhancing the sustainability and reducing the environmental impact of this green wastewater treatment technology, but it may present risks of leaching harmful compounds to the treated water. The observed normal growth indicators of the plants' development under reclaimed water irrigation point to no harmful effects.

3.2. Microalgae Growth Results

Cultivation of two microalgae genera was carried out in two experimental scales: shaken flasks (SF) and bubble column PBR. Table 5 presents the algae productivity indicators obtained with reclaimed water produced by a solid waste-filled CW.

Table 5. Productivity indicators of the two microalgae genera cultivated in the reclaimed water ¹.

Productivity Indicator	Type of Microalgae and Reactor			
	<i>Chlorella</i> sp.		<i>Scenedesmus</i> sp.	
	SF	PBR	SF	PBR
Specific growth rate (day ⁻¹)	0.070 (a)	0.378 (b)	0.089 (a)	0.470 (b)
Final biomass concentration (kg/m ³)	0.14 (a)	0.37 (b)	0.38 (b)	0.64 (a)
Biomass productivity (mg _{dw} /L/day)	5.2 (a)	33.1 (b)	16.0 (c)	61.2 (d)
Doubling time (day)	9.96 (a)	1.83 (b)	7.77 (a)	1.48 (b)
Total chlorophyll (mg/L)	1.02 (a)	n.d.	3.83 (b)	4.31 (b)
Carotenoids (mg/L)	0.33 (a)	n.d.	1.26 (b)	1.55 (c)
Lipid productivity (mg/L/day)	0.70 (a)	n.d.	1.43 (b)	5.47 (c)

¹ SF = shaken flasks; PBR = bubble column photobioreactor. Values marked with the same letter in the same row are not significantly different under a statistical level of 0.05.

As expected, productivity results are better for cultivations in the PBR. However, the results obtained in both scales demonstrate an effective growth of both microalgae in the reclaimed water produced by the solid waste-filled CW. Like the experiments of growth of

the French marigold, the most significant result is the apparent absence of negative effects of using solid waste as CW filler.

Growth indicators are slightly better for the microalgae *Scenedesmus*, but not significantly different from the results obtained for the microalgae *Chlorella*. In addition to no observation of ecotoxicity effects, the results obtained are comparable or even better than the results available in the literature. Experiments of cultivation of *Scenedesmus* sp. in wastewater reported a biomass productivity from approximately 5 to 95 mg/L/day, a doubling time from 1.81 to 8.62 days, and a specific growth rate of 0.08 to 0.38 day⁻¹ depending on wastewater composition and initial cells concentration [52]. Additionally, for cultivations in wastewater, it is reported that the growth of *Chlorella vulgaris*, depending on the operative conditions, presents biomass productivities from approximately 10 to 27 mg/L/day and specific growth rates from 0.031 to 0.134 day⁻¹ (and doubling time from 5 to 22 days, estimated from the reported data) [53].

3.3. Simulation of Reclaimed Water Use for Green Space Irrigation and Microalgae Production

The assessment of using reclaimed water produced by constructed wetlands for irrigation and microalgae cultivation was carried out in Sections 3.1 and 3.2. The results obtained indicate that using solid waste as CW fillers does not hinder plant and microalgae growth. Thus, reclaimed water produced by CWs filled with a mixture of limestone waste, coal slags, and cork waste may be used for irrigation and microalgae cultivation. Using solid waste in wastewater treatment systems and further application of water for irrigation or algae biomass production may be a sustainable symbiosis of water and solid waste circularization.

Considering that the need for irrigation water depends on the weather conditions, reclaimed water availability may exceed irrigation demand in rainy and cold seasons. In this section, a conceptual model is proposed to combine irrigation with the use of reclaimed water for microalgae cultivation in the months where irrigation demand is lower or null. To set up the model, a simulation is carried out based on the academic campus of the Polytechnic Institute of Tomar (IPT), located in the city of Tomar (39°35′57.084″, −8°23′25.3932″).

IPT's academic community is composed of nearly 2500 students and 300 staff. The campus of Tomar contains the School of Technology and the School of Management. The campus area is approximately 99,460 m², 31,700 m² of which is green spaces (Figure 1). In addition to the buildings that hold the IPT's services, classrooms, amphitheaters, library, and laboratories, the campus has a canteen, one bar, and two residences for students.

Currently, greywater is not collected separately from blackwater, and all wastewater is discharged in the municipal sewage network. However, the implementation of a greywater collection network allowing the recovery of this kind of wastewater is under consideration. Besides that, the generated wastewater can be primarily treated in situ, before being recovered by the constructed wetland system. This alternative was considered as a basis for the case study.

IPT has an irrigation network based on water extracted from one of the two existing wells. The typical period of irrigation is between May and October, corresponding to the lower average precipitation (Figure 2).



Figure 1. Satellite photo of IPT's Tomar campus (Google Maps). The dotted green polygons are the areas available for wastewater treatment by constructed wetlands and the microalgae production plant. The blue polygon shows the lake reservoir near the campus library.

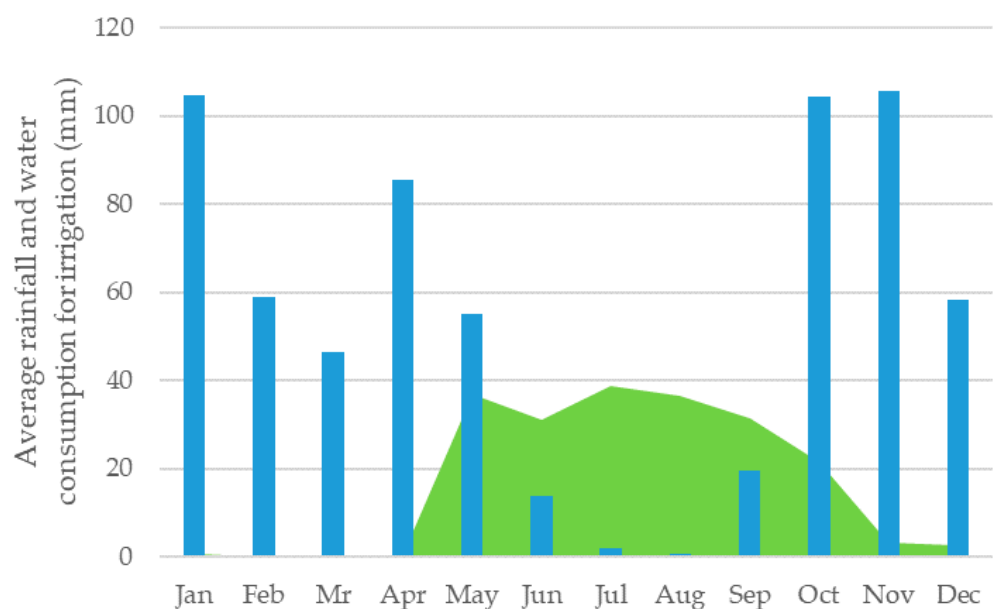


Figure 2. Typical monthly average rainfall (vertical bars, mm) and the average consumption of water for irrigation of the green spaces in the campus, in equivalent units (shaded area, mm). Precipitation data from SNIRH-APA datasets [54].

This work intends to simulate the total or partial replacement of the extracted underground water by wastewater generated in the campus after adequate treatment. Although the wastewater flowrate is not measured, it is estimated from the tap water consumption. Figure 3 shows the monthly average wastewater flowrate estimated from the tap water consumption, considering a shortage of 5%. Main flows originated in the two students'

residences. Lower flows were generated at the canteen and bar, both located in the same building. The remaining sources were from the campus laboratories and toilets.

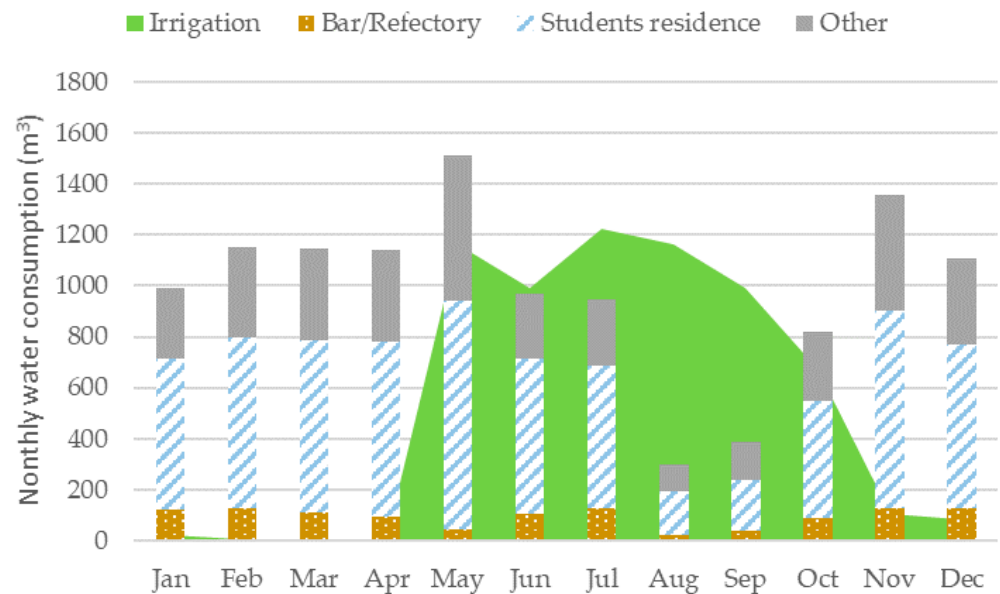


Figure 3. Typical monthly average of tap water consumption in the campus (stacked columns: figures corrected in 95% to consider water losses, m³) compared to the water consumed for irrigation (shaded area, m³).

Potentially available reclaimed water is less than the required irrigation water in the dry months because those months coincide with lower academic activity periods. August and September are the critical months. Thus, using treated wastewater for irrigation requires storing the wastewater or the reclaimed water during the months before the summer period.

Using the reclaimed water to produce algae biomass is an alternative way to valorize water and can be a solution to the periods with higher wastewater flowrates coincident with the periods of no need for irrigation. Production of microalgae in the cold and rainy months may correspond to lower productivity due to lower temperature and solar radiation. However, a temperature higher than 35 °C is lethal for some microalgae species [55]. In addition, the culture temperature in PBRs may reach values of 10 to 30 °C higher than the ambient temperature in the hot and sunny months [56].

Figure 4 shows the typical monthly averages of the maximum air temperature and the maximum solar radiation intensity in the region of Tomar, Portugal. Microalgae production in the period between May and September requires cooling of the PBRs, which may represent high production costs [57]. Thus, running the microalgae PBRs in the period from October to April may represent lower productivities but avoids PBR cooling devices.

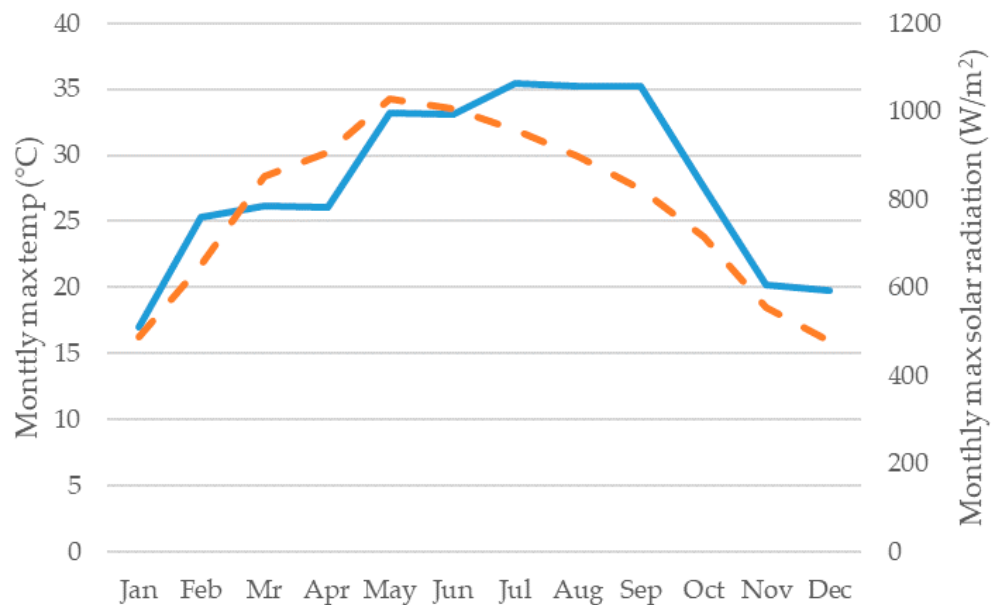


Figure 4. Monthly average maximum temperature (continuous line) and solar radiation (dotted line). Data from SNIRH-APA datasets [54].

A conceptual model for complementing the use of reclaimed water in irrigation of the campus with the production of microalgae is proposed in the scheme in Figure 5. The collected campus wastewater can be preliminarily treated in a compact wastewater treatment plant (WWTP) to be implemented, feeding a set of constructed wetland modules. Groundwater from the existent well can complement the wastewater flow into the constructed wetlands in the hot months to compensate for the lowering of wastewater flow. An already existent 450 m³ capacity cistern can be used as the main reservoir to store the treated wastewater in the months with higher reclaimed water production. In addition to the cistern, an artificial lake near the campus library can be used as a complementary reservoir. Considering an acceptable variation in the water depth of 0.50 m, this lake can store up to 600 m³ of water. To prevent risks for public health, this water should be disinfected. Water can be recirculated between the lake and the cistern as required to ensure a regular flow for irrigation or microalgae production. Water from the cistern can then feed the campus irrigation network in the dry months and feed the microalgae PBRs in the rainy months. Algae biomass separated from the water by sedimentation or filtration can be used for compost production, together with biodegradable waste from the bar and canteen, and then applied as green fertilizer on the campus. The discarded water can be disinfected and discharged in a watercourse near the campus or conducted to the current municipal sewage system, depending on the water quality. The required disinfection systems and all pumping drives can be powered by a renewable energy station combining photovoltaic collectors, batteries, and, eventually, a secondary power source such as a fuel cell or a biofuel-fueled generator.

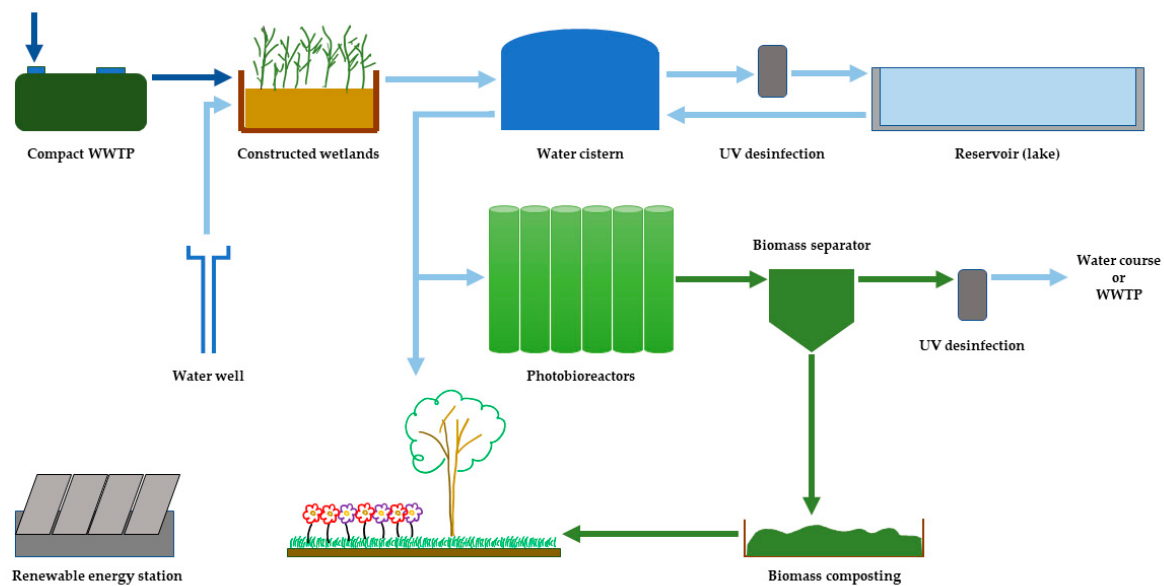


Figure 5. Scheme of the proposed solution to produce reclaimed water for irrigation with constructed wetlands and integration of microalgae production for water polishing and compost production.

The monthly need for water reserves can be estimated from Equation (6), where the subscript refers to the month and R is the reserve volume, G is the wastewater generated and treated, P is the precipitation, I is the irrigation consumption, A is the water feed to the microalgae PBRs, and W is the water extracted from the well, all in m^3 per month. The precipitation input is estimated from the average height in mm and the area of the lake (approximately 1200 m^2). The reserve volume is set to a minimum of 200 m^3 .

$$R_n = R_{n-1} + G_n + P_n - I_n - A_n + W_n \quad (6)$$

Four scenarios are evaluated: (1) maximum storage corresponding to cistern capacity only; (2) maximum storage corresponding to cistern capacity and half lake capacity; (3) maximum storage corresponding to cistern capacity and full lake capacity; and (4) additional extra storage to allow no use of groundwater extraction. Table 6 presents the results of the simulation, including an estimate for algae biomass production considering a productivity half the maximum value obtained experimentally in Section 3.2 for the *Scenedesmus* species, allowing for the consequences of non-optimal conditions in the cold periods. Water availability for microalgae production was set to ensure the limiting storage capacity is respected, considering microalgae production from November to April each year.

Table 6. Simulation of PBR plant capacity and productivity, and consumption of underground water, depending on the reservoir capacity.

Storage Resources	Storage Capacity (m^3)	Water Available for PBRs (m^3/year)	Biomass Production ($\text{kg}_{\text{dw}}/\text{year}$)	Water Extraction from the Well (m^3/year)	Water Saving (%)
Cistern	450	7380	2362	1276	80
Cistern + Lake 50%	750	6930	2218	976	85
Cistern + Lake 100%	1050	6480	2074	676	89
Plus additional storage ¹	1726	5467	1749	0	100

¹ Considering the cistern and lake maximum capacities.

Although using the available cistern and full lake capacities, the use of the well is necessary to ensure water requirements for irrigation in August and September (Figure 6). Including an extra storage capacity of 676 m³ can avoid the use of the well. However, for that scenario, the wastewater availability for the CWs is far below the monthly maximum of 1500 m³ in August and September. Using the well to partially feed the wetland beds in those hot and dry months may reduce the heat stress in the reed plants. This option corresponds to scenario (3). According to Table 6, using the available storage capacity of 1050 m³ allows avoidance of 89% of the freshwater consumption and ensures an estimated production of 2074 kg dry weight of microalgae biomass per year. Using reclaimed water for irrigation also can result in avoidance or a reduction in fertilizer consumption. Microalgae cultivation in reclaimed water can avoid or reduce the consumption of artificial growth media.

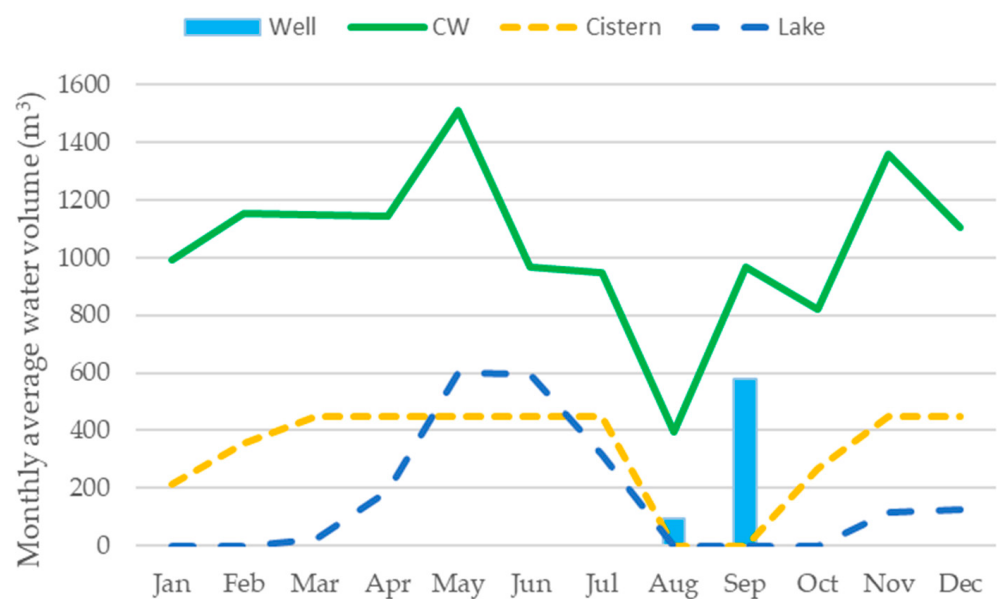


Figure 6. Simulation of the water balance in the campus framework for wastewater recovery for irrigation, considering the combined storage capacity of the water cistern (450 m³) and the campus lake (600 m³). The well water is necessary to overcome the storage shortage in August and September.

Maximum flowrate in the CW input stream is expected during May. Considering a maximum design flow rate of 0.052 m³/m²/day for stratified solid waste-filled CWs [45], it is required to set up a total area of wetland beds of 900 m². This area can be achieved using nine CW beds, each 5 m wide and 20 m long.

Future work will be carried out in the following directions:

- (i). Estimation of investment and operational costs to set up the proposed framework, namely, the CW beds, the PBR plant, the compact WWTP, the disinfection units, the renewable energy station unit, and the additional tubing and pumps;
- (ii). Estimation of financial benefits from avoiding the use of commercial fertilizers and compost for treatment of the green spaces and from avoiding the consumption of nutrients for the microalgae cultivation;
- (iii). Experimental work to optimize microalgae cultivation in PBRs placed outdoors;
- (iv). Evaluation of potential uses of the microalgae biomass for valuable compounds under the biorefinery concept;
- (v). Scale-up of the proposed water recovery model for application to different size communities under the smart cities concept;
- (vi). Spread of an educational message and green behavior motivation for the protection of water as a fundamental resource.

4. Conclusions

Lab-scale experiments were carried out to evaluate the potential to reuse wastewater treated by solid waste-filled constructed wetlands for irrigation of green spaces and cultivation of microalgae. Reuse of treated wastewater (also designed as reclaimed water) is a relevant route to mitigate water scarcity and to protect the aquatic environment. In addition to the reduction of freshwater consumption, and the decrease of pollution in water bodies, using reclaimed water is an application of the circular economy principles toward the reduction of the consumption of nutrients and minerals, some of which are already scarce, such as phosphorus.

It was found that the wastewater treated by the CW fulfills the water quality requirements to be reused for irrigation under the Portuguese Decree-Law. No. 119 of 2019, and was classified as class A water, which means that it can be used to irrigate green spaces without restrictions. CWs are a greener technology for wastewater treatment and reclaimed water production, particularly when CWs are designed using recovered solid waste as filling media.

The main result of the lab experiments, besides the confirmation of the potential of using the reclaimed water produced by the solid waste-filled CW as irrigation water and as microalgae cultivation media, is the observation of no apparent toxic effects on the plants tested (*Tagetes patula* L.), the microalga *Chlorella* sp., and the mixed culture whose predominant microalga is *Scenedesmus* sp. Growth indicators for the plants irrigated with reclaimed water were not significantly different than the results obtained with fertilized water. In the same way, the growth parameters obtained for the cultivation of microalgae are consistent with the values found in the literature.

Considering that the need for reclaimed water for irrigation is seasonal in most regions, a conceptual model to combine irrigation with microalgae production and water storage was proposed, taking into account the limited access to storage capacity. Moreover, the model considers the strategy to use the reclaimed water for microalgae production only in the cold periods, simultaneously avoiding the use of cooling devices required in hot seasons and freeing the reclaimed water for irrigation in the hot and dry periods. In this approach, microalgae production is considered a complementary valorization of the produced reclaimed water, diminishing the economic issues related to the high costs of the downstream processing of microalgae biomass.

The model was assessed through a simulation taking the campus of the Polytechnic Institute of Tomar as a case study. The simulation results show a synergetic effect of integrating reclaimed water production with irrigation, water storage, and microalgae production. Using reclaimed water may represent a reduction of 89% of the consumption of abstracted groundwater for irrigation of the campus green spaces. The reclaimed water produced in the cold and rainy months, when its use for irrigation is not required, allows the production of up to 2074 kg dry weight per year of microalgae biomass for composting purposes. Using reclaimed water for irrigation also avoids or reduces fertilizer consumption. Microalgae cultivation in reclaimed water avoids or reduces the consumption of artificial growth media.

The proposed approach contributes simultaneously with a model to reduce freshwater consumption and to disseminate among the students and the surrounding communities the principles of sustainability and the circular economy of water, and the need for freshwater protection.

Future work will be carried out to optimize the model, to validate the technical results, and to perform the required economical evaluation. In addition, alternative uses of the microalgae biomass can be evaluated under the principles of biorefinery, such as raw materials for obtaining high-value products.

Besides the technical results and future economic analysis, the work, which is developed under a project-based learning context, is contributing to enrolling students, the academic community, and the general society on the search for solutions that mitigate the increasing issue of freshwater scarcity.

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