

Treatments of Wastewater in Rural Communities using a Pilot Plant Photobioreactor

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In order to avoid eutrophication phenomena of surface water-courses, the reduction of the nutrient content, such as nitrogen and phosphorus, becomes ever more pressing. Respect to traditional depuration systems, the phytodepuration by microalgae presents different advantages: the simultaneous reduction of nitrogen and phosphorus by photosynthesis with biomass production without external organic sources and the production of oxygenated effluents after microalgae treatment. Moreover, the produced biomass could be used as raw material for energy or chemical purposes and the depuration systems with microalgae require less surface area than traditional phytodepuration ones. In this work data recovered from phytodepuration of rural municipal waste by microalgae are presented. Microalgae were grown in a semi-pilot photobioreactor system with a total capacity of 0.95 m³, consisting of: a photobioreactor composed by 40 vertical plexiglass tubes fed in parallel, a recycling vessel with a volume of 500 L, a recycling centrifugal pump and a submerged centrifugal feed pump. The area occupied by the plant was 5.4 m². The photobioreactor was continuously fed with the final effluent of San Pietro Vara (Imperia, Italy) wastewater treatment plant and the effluent from the photobioreactor was piped in the no-longer used sludge draining bed, and from here in the feed sump. The microalgae growth has been identified by microscopic analysis. The predominant algal species were Chlorophyceae and Cyanophyceae, but the total biomass included also protozoa and small metazoan. Biomass concentration was recorded periodically. Two multiparametric probes were used for the determination of pH, OD, NO₃, NH₄⁺, Chlorophyll-a, temperature and turbidity, while COD, total N and P were measured traditionally. The main results obtained from the closed system have been average removal of 50, 60 and 50 % for COD, total N and total P respectively. Results indicated that, in general, the outlet of phytodepuration met the quality standards required by law (Annex 5 – Italian Law 152/06 according to the EC Dir. 91/271).

1. Introduction

As well-known, water resources and greenhouse gases, mainly CO₂, are a major challenge to worldwide environmental sustainability. Regarding the conservation of water resources, the need for treatment of wastewaters and their reuse or recycling becomes more and more pressing (Bixio et al., 2006).

About the water resources conservation, the wastewater treatment plants must have the ability to efficiently remove high concentrations of nutrients, namely nitrogen and phosphorous, in order to prevent eutrophication of waters. In fact, this phenomenon causes a deterioration, often irreversible, of water quality (Cai et al., 2013).

Currently there are several technologies based on biological or physical-chemical processes for nitrogen and phosphorus removal from wastewater but, these processes are highly energy-intensive and thus producing high amounts of greenhouses gases, large amounts of waste sludge and requiring high investment and management costs. Finally, all these processes are not able to simultaneously remove nitrogen and phosphorus from wastewater (Coast et al., 2011; Giuliano et al., 2018).

The natural processes of water treatment like phytodepuration overcome the drawbacks above mentioned, however, they need extended land surfaces and therefore are mainly applied for the treatment of wastewater related to small communities. Oversized, the treatment of wastewater with algae has the following long-term

environmental benefits such as the lower involved surfaces than the ones required in case of macrophytes and the increased efficiency of CO₂ photosynthetic conversion. Moreover, the effluent discharged into receiving water bodies is oxygenated and high value products can be extracted from produced algal biomass (Casazza et al., 2017).

In literature, different authors reported the potential of microalgae for the removal of nitrogen and phosphorus. Main mechanisms in algal nutrient removal from wastewater include uptake into the cell and stripping ammonia through elevated pH (Bich et al., 1999). The advantages of using algae for that purpose include: the low cost of the operation, the possibility of recycling assimilated nitrogen and phosphorus into algae biomass as a fertilizer avoiding a sludge handling problem, and the discharge of oxygenated effluent into the water body. The widely used microalgae cultures for nutrient removal are *Chlorella* sp. (Lee and Lee, 2001), *Scenedesmus* (Martinez et al., 2000), and *Spirulina* (Olguín et al., 2003). Nutrient removal capacities of *Nannochloris* (Jimenez-Perez et al., 2004), microalgae *Botryococcus braunii* (An et al., 2003), and cyanobacter *Phormidium* (Dumas et al., 1998) have also been investigated.

One of the well-known algae containing bioprocesses for wastewater treatment is high-rate algae ponds (HRAP) (Deviller et al., 2004). In the past years, corrugated raceways (Olguín et al., 2003) and closed photobioreactors (Dumas et al., 1998) have been developed for nutrient removal. The other open-air and closed bioprocesses used for commercial production of algae were reviewed by Borowitzka (1999). Immobilization techniques have been applied to algae to solve harvesting problems encountered in suspended growth systems.

The algal biomass is a bioresource of primary importance since it can be transformed into a wide range of valuable products: health care products, food additives, chemicals, third generation biofuels. For this reasons, the research aims at developing technologies that allow the intensive production of microalgae. The intensive cultivation of microalgae requires water, light, nitrogen and phosphorus salts, carbon dioxide. Therefore the environmental impact resulting from this activity are water pollution, air pollution due to the production of reagents and consequent energy consumption. The intensive production of microalgae may constitute an environmental problem that can be significantly reduced by using the nutrients contained in wastewater and, when possible, the CO₂ present in the flue gases from combustion processes or better the CO₂ produced by the processes of conversion of biogas from anaerobic digestion systems to biomethane (Casazza et al., 2016).

Based on such considerations, the civil wastewater, wastewater produced by the livestock and food industry appear suitable for the algal biomass production; therefore, the microphytodepuration technique turns out as a long-term sustainable solution.

This study presents a part of the results obtained during the three years LIFE project named ECOMAWARU (LIFE08 ENV IT 000390 Project). A semi pilot plant photobioreactor was used in order to treat the effluent of a wastewater treatment plant located in San Pietro Vara (La Spezia, Italy). The inlet and outlet photobioreactor waters were characterized in terms of total nitrogen, phosphorous and suspended solids concentration. The chemical oxygen demand, the pH and the temperature were also monitored over period of one year.

2. Materials and methods

2.1 San Pietro Vara municipal plant and semi pilot photobioreactor

The San Pietro Vara municipal plant was designed to treat an average flow rate of 48 cubic meters per day of wastewater (300 inhabitants equivalent (IE); 200 liters per day per IE, 0.80 sewage inflow coefficient).

Microalgae were grown in a semi-pilot system with a total capacity of 0.95 m³ (Figure 1 a,b), consisting of a photobioreactor composed by 40 vertical plexiglass tubes fed in parallel, a recycling vessel with a volume of 500 L, a recycling centrifugal pump and a submerged centrifugal feed pump. The area occupied by the plant was 5.4 m². The inlet and outlet waters and the inside condition of the photobioreactor were monitored for a period of one year (from September 2012 to August 2013). In this work, the photobioreactor was fed with the final effluent of the municipal wastewater treatment plant at a flow rate of 200 liters per hour, corresponding to 10 % of the wastewater entering the treatment plant and to a hydraulic retention time of about 5 hours. A greater retention time would have led to the installation of an additional module that was considered not appropriate in this case given the low concentrations of nutrients incoming.

2.2 Microalgal biomass

The microalgae used in the photobioreactor has been identified by microscopic analysis after a natural selection of microorganism proliferated in the studied rural wastewaters. The selected heterogeneous biomass included different species of the algal cells, protozoa and small metazoan. The predominant microalgal species were Chlorophyceae and Cyanophyceae.



Figure 1a,b: Semipilot plant photobioreactor installed at San Pietro Vara (La Spezia, Italy) wastewater treatment plant.

2.3 Analytical analysis

Photobioreactor biomass was characterised in terms of classes (optical microscope) and elemental composition (FLASH EA1112 CHNS-O Elemental Analyser, ThermoQuest), while the quality of phytodepuration plants (inlet and outlet waters) was determined weekly, monitoring the pH, the total nitrogen (N), the total phosphorous (P) and the total suspended solid concentrations and the chemical oxygen demand (COD), SST values by laboratory analysis after the weekly samples collection (Casazza and Rovatti, 2017). In situ analysis were performed using two multiparameter probes (6920 - V1 and 6920-V2, YSI) with telemetry and sensors for temperature, pH, conductivity, dissolved oxygen (measured electrochemically), nitrates, ammonia, chlorophyll-a, and an integrated telemetry unit with internal battery power and solar panel integrated (TUBE 300 ANT- Advance Network Telemetry Watec Technology) with reporting functions via sms, email, ftp with programmable alarms.

3. Results and discussions

3.1 Biomass characterization

During the experiment, the biomass inside the photobioreactor changed, not only in terms of concentration, but also in composition. In fact, during the first 8 months (from September to April) of growth, the predominant microalgal species were Chlorophyceae, mainly *Chlorella* sp. (Figure 2,a), while from March to the end of the study, the biomass was constituted by filamentous Cyanophyceae and *Scenedesmus* sp. (Figure 2 b,c).

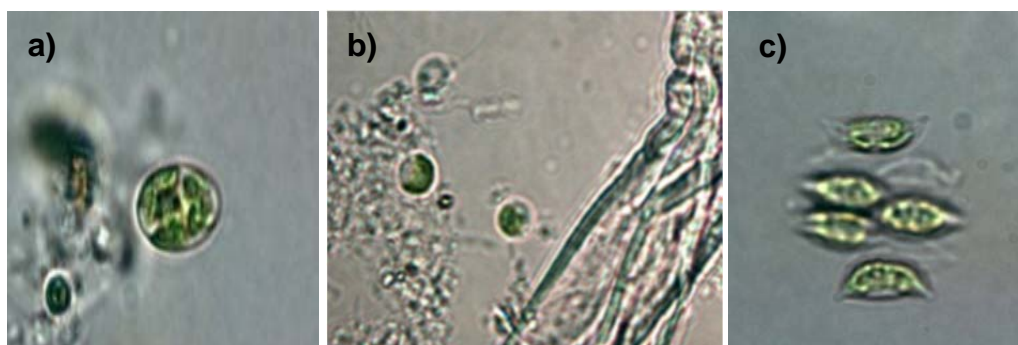


Figure 2a,b,c: Representative microscope images (100x) of biomass observed in outlet waters.

The elemental analysis gave an average biomass composition constituted by 45.2 ± 0.29 , 4.51 ± 0.10 , 4.58 ± 0.10 , 0.10 ± 0.02 , 2.43 ± 0.14 and 1.1 ± 0.05 , of carbon, hydrogen, nitrogen, sulphur, phosphorous and potassium, respectively.

3.2 Wastewater purification

In Table 2, the average values of inlet and outlet waters during the four seasons are reported. It is important to consider that the nutrient and organic pollution removal was influenced by meteorological factors (e.g. temperature, solar radiation) that, together with the concentration of nutrients, are crucial for algal growth.

Table 2: The average values of the photobioreactor system for the 4 seasons

	Autumn		Winter		Spring		Summer	
	Inlet	Outlet	Inlet	Outlet	Inlet	Outlet	Inlet	Outlet
pH	7.3	7.5	7.1	7.3	7.4	7.7	7.6	8.3
T (C°)	9	14	5	10	14	19	21	26
Total Nitrogen (mg/L)	3.68	1.64	2.63	1.34	3.85	0.97	6.15	0.61
Total Phosphorous (mg/L)	0.16	0.09	0.20	0.13	0.21	0.10	0.30	0.07
COD (mg/L)	20	16	12	13	24	15	31	16
TSS (mg/L)	15	14	11	12	37	51	53	81

In general, the pH values were constant during the period of analysis, while significant changes of N, P, COD and TSS concentrations in inlet waters were observed depending on the season.

In Figure 3 are reported the trends of total nitrogen and phosphorous concentrations, the chemical oxygen demand and the total suspended solid of the photobioreactor inlet and outlet waters.

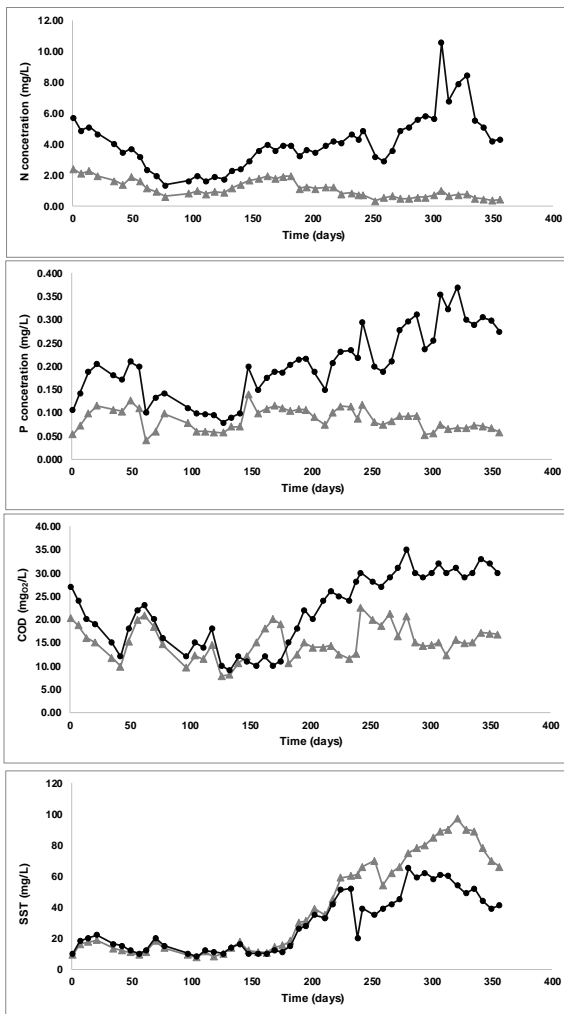


Figure 3: Trends of nitrogen (a), phosphorous (b), Chemical Oxygen Demand (c) and total suspended solids (d) contents in the photobioreactor inlet (●) and outlet (▲) waters.

As can be seen, even if the total nitrogen content in the inlet waters increased during the study, the nitrogen concentration in the outlet waters remained constant (Figure 3,a). This could be due to the increase of biomass concentration in the photobioreactor (Figure 3,d) and to the consequent increased request of nutrients. A similar consideration could also be done for total phosphorous.

In Table 3, an example of data collected during a day of analysis by the multiparameter probe is reported. During the day the temperature changed significantly, as well as NH_4^+ and NH_3 . It is interesting to see a change in chlorophyll content due to the periodicity in cellular Chl a accumulation driven by factors other than cell division, this phenomenon is indicated in different studies (Jørgensen, 1966; Owens et al., 1980).

Table 3: Data collected using the multiparametric probe during a day of analysis (September 23th 2013).

Time (hour)	T (°C)	Cond. (uS/cm)	pH	NH_4^+ (mg _N /L)	NH_3 (mg _N /L)	Chl-a (ug/L)
00:00	20.4	679.0	7.5	1791.3	0.023	32.8
01:30	19.6	700.0	7.5	1881.0	0.022	26.8
03:00	19.0	716.0	7.5	1961.3	0.022	21.8
04:30	18.6	729.7	7.5	2166.7	0.026	16.6
06:00	18.2	739.0	7.6	2394.3	0.030	13.9
07:30	17.8	744.7	7.6	2476.7	0.033	23.1
09:00	18.1	738.0	7.7	1714.3	0.032	34.4
10:30	20.6	749.0	7.7	1583.7	0.030	25.7
12:00	23.7	764.0	7.6	1657.7	0.036	18.2
13:30	26.8	776.0	7.6	1759.3	0.046	12.3
15:00	28.4	782.3	7.6	1741.7	0.051	19.2
16:30	27.5	785.0	7.6	1667.7	0.046	18.0
18:00	25.7	787.7	7.6	1547.0	0.037	16.6
19:30	24.1	785.7	7.6	1319.0	0.029	15.8
21:00	22.6	785.3	7.6	1085.7	0.022	15.4
22:30	21.5	783,5	7.7	1019.0	0.020	14.9

In Figure 4 the removal efficiencies in terms of nitrogen, phosphorous and COD operated by the photobioreactor plant were reported. As can be seen, the removal efficiency increased during the runs, reaching values of 90, 80 and 60 % during the last days of growth for total nitrogen and total phosphorous contents and COD, respectively. This could be due to a microalga adaptation to the medium used during the runs. Similar results of removal efficiency were reported by De Alva et al. (2019).

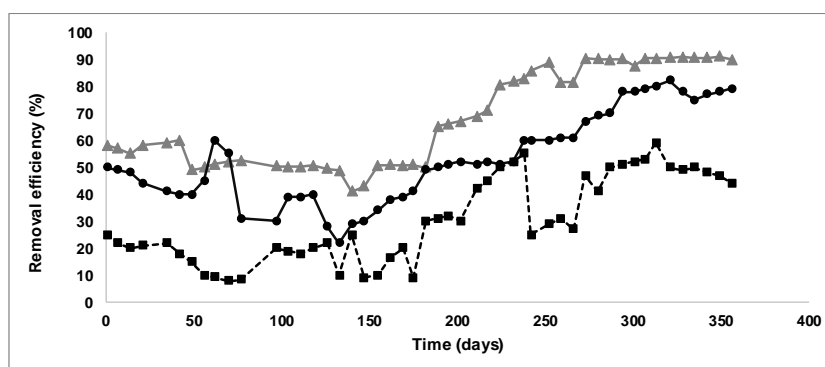


Figure 4: Removal efficiency (%) of Total Nitrogen (▲), Total phosphorous (●) and Chemical Oxygen Demand (■).

4. Conclusions

The wastewater after the microalgal treatment presented a low content in nitrogen, phosphorous a low chemical oxygen demand, resulting in a water with a less eutrophic potential.

The results of this work proved that the photobioreactor plant could be used with good results for the rural wastewater treatment. Moreover, the treated effluent can be used as irrigation waters according to the Italian Decree by Law 185/03 and the possible reuse of the algal biomass as fertilizer will be considered.

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