Bull. Environ. Contam. Toxicol. (1999) 62:483-489

Bulletin of Environmental Contamination and Toxicology

Algae Growth Potential Measurement in Distillery Wastes

L. Travieso, F. Benitez, R. Dupeyrón

Department of Environmental Pollution Studies (DECA), National Center for Scientific Research (CNIC), Post Office Box 6990, La Habana, Cuba

Received: 8 April 1998/Accepted: 29 January 1999

The trophic status of waters can be evaluated using algae techniques. Algae Growth Potential (AGP) determination is routinely used (Lukavsky 1986; Leon and Travieso 1992; Richmond 1996). Theoretically, the trophic level of a water indicates the relationship between the quantity of organic matter entering the system per unit of time (Hall and Rao 1994; Hall 1996). Algae Growth Potential bioassay is an standard, reproducible and understandable method. Also, it constitutes an economic assay to determine the potential of water bodies, natural waters and wastewaters, to support or inhibit the microalgae growth (De Pawn and De la Noüe 1983; De la Noüe and De Pawn 1988; Vonshak et al. 1996; Vonshak et al 1996a). Algae Growth Potential determination is based on the relation of a maximun biomass yield concerning the biologically used nutrients for microalgae growth. In a water body, nutrients could be consumed, partially or totally, depending on the nutritional status of the water.

This study evaluated the effectiveness of different pretreatment processes of distillery wastewater to be tested as a culture media for microalgae. Algae Growth Potential was determined for two different culture media of microalgae: distillery wastewater pretreated by anaerobic processes and distillery wastewater pretreated by a combined anaerobic-aerobic system.

MATERIALS AND METHODS

For the development of this work a strain of ***Chlorella vulgaris SR/2*** was used. This strain was provided by the Microalgae Collection of CNIC (Valiente and Travieso 1994). The strain was maintained in a modified Zarrouk culture media (MZ) and using artificial illumination with lamps of 40 Watts.m^{-2}. The strain of ***Chlorella vulgaris*** used in the experiments was adapted to strong organic loading variations, and, to light intensity and temperature fluctuations.

The experiments for AGP determinations were carried out in cylinders of 2 liters of effective volume. In both reactors, the filtered and saturated air was supply. Light cycles were 8/16 light/dark hours. The illumination was provided by lamps of 40 watts.m-2 (Travieso and Benitez 1990).

Correspondence to: L. Travieso

For the First experiments, distillery wastewater was previously treated by a combined system anaerobic-aerobic, consisting on an anaerobic filter and a trickling filter (AGPA). For the second experience, distillery wastewater was previously treated by anaerobic filter (AGPAN).

The analysis of chemical oxygen demand (COD), total solids (TS), chlorophyll a (Ca) and Total chlorophyll (Ct) were done following the Standard methods for the examination of waters and wastewaters (APHA 1993). The samples were taken daily for the analysis.

Four runs of each experiment were done with different initial COD. For AGPA samples, the COD values ranged from 400 - 2100 $mg.L^{-1}$. For AGPAN samples COD ranged 3000 - 7100 $mg.L^{-1}$.

Biokinetic parameters were determined for both culture media (AGPA and AGPAN). Growth rate (μ) and saturation constants (Ks) were calculated by the Michaelis Menten method (Oh-Hama and Miyachi 1988).

RESULTS AND DISCUSSION

Tables 1 and 2 show the values obtained for the different parameters.

From these tables it is possible to establish that for AGPA experiments the microalgae development was very satisfactory in all cases (different initial COD). When combined process was used, the united action of aerobic and anaerobic microorganisms could help the formation of non-soluble substances, removing them from the culture media. When only anaerobic treatment was used the results obtained are not the same (León and Travieso 1996).

For AGPAN culture media, at the beginning of the experiment the behavior was satisfactory, but after 10 days, the values of Chlorophyll diminish progressively. This could happen because of the toxic elements presence in the distillery wastewater after the anaerobic treatment

Table 3 shows the values of mass potential calculated for all the experiments. In both cases, from the theoretical point of view, distillery wastewater biologically pretreated could be used as culture media of microalgae. The values of the mass potential are over 1000 corresponding with hypertrophic level classification

Figure 1, shows the behavior of microalgae development for the different initial COD using both culture media (AGP and AGPAN).

In Table 4, biokinetic parameters were determined showing that μmax for AGPA culture media is higher than μmax for AGPAN culture media, corresponding this results with the microalgae development in both media.

Table 1. Behavior of the different parameters during the experimental time for the experiments using distillery waste water pre-treated by a combined system anaerobic -- aerobic (AGPA).

Time (d)	Chemical Oxygen Demand (COD) ($mg.L^{-1}$)											
	456			624			1654			2050		
	Ca	Ct	ST	Ca	Ct	ST	Ca	Ct	ST	Ca	Ct	ST
0	0	0	562	0	0	818	0	0	2270	0	0	1687
3	1.0	1.1	562	0.2	0.5	971	1.5	1.8	2290	1.0	1.6	1700
6	1.1	1.2	818	0.5	1.5	969	1.4	2.1	2290	1.2	2.0	1800
9	1.1	1.8	1125	1.2	1.8	820	1.8	2.5	2290	1.2	2.3	1800
12	1.0	1.8	1687	1.3	2.2	1120	1.9	2.7	2295	2.1	2.9	2250
15	2.0	2.3	2250	1.5	2.3	1126	2.8	2.9	4500	2.0	2.9	1925
18	2.0	2.8	816	1.6	1.9	2250	2.5	3.0	5630	2.8	3.2	1940
21	2.9	2.5	1680	2.0	2.1	3375	2.3	3.8	7895	2.8	3.8	1690
24	2.9	3.1	2240	2.1	2.2	3350	2.2	4.1	7895	2.8	4.1	1690
27	2.9	3.1	2210	3.0	3.9	4500	2.2	4.5	7900	2.8	4.1	1690
30	2.9	3.1	2550	3.1	4.0	5480	2.2	4.5	8094	2.8	4.0	1700

Ca = chlorophyll a ($mg.L^{-1}$) Ct = total chlorophyll ($mg.L^{-1}$) ST = total solids ($mg.L^{-1}$)

Table 2. Behavior of the different parameters during the experimental time for the experiments using distillery waste water pre-treated by anaerobic system (AGPAN).

Time (d)	Chemical Oxygen Demand (COD) ($mg.L^{-1}$)											
	3088			4312			5782			7069		
	Ca	Ct	ST	Ca	Ct	ST	Ca	Ct	ST	Ca	Ct	ST
0	0	0	3333	0	0	10832	0	0	2499	0	0	4999
3	0.3	1.0	2499	0.2	1.8	10900	1.7	1.8	3333	0.7	0.7	7800
6	0.4	0.9	1666	0.5	2.2	11000	0.8	1.9	4999	1.0	1.6	4999
9	0.6	1.2	1590	0.5	1.7	10050	0.7	1.9	5220	1.3	1.4	5900
12	0.65	1.1	3200	0.6	1.2	9999	0.6	2.0	6666	1.1	1.8	4730
15	0.65	0.8	2510	0.7	1.1	8900	0.6	1.4	5665	1.2	1.7	3800
18	0.35	0.7	3300	0.67	0.9	8332	0.5	1.0	5000	0.9	1.5	3333
21	0.3	0.7	3420	0.5	0.7	7200	0.4	0.7	7800	0.9	1.4	2500
24	0.5	0.6	4612	0.55	0.6	6666	0.45	0.7	8300	0.7	1.4	2480
27	0.4	0.5	5024	0.5	0.6	6325	0.38	0.7	11000	0.7	1.3	1820
30	0.35	0.6	5200	0.4	0.6	5810	0.30	0.7	9780	0.7	1.0	1666

Ca= chlorophyll a ($mg.L^{-1}$) Ct = total chlorophyll ($mg.L^{-1}$) ST = total solids ($mg.L^{-1}$)

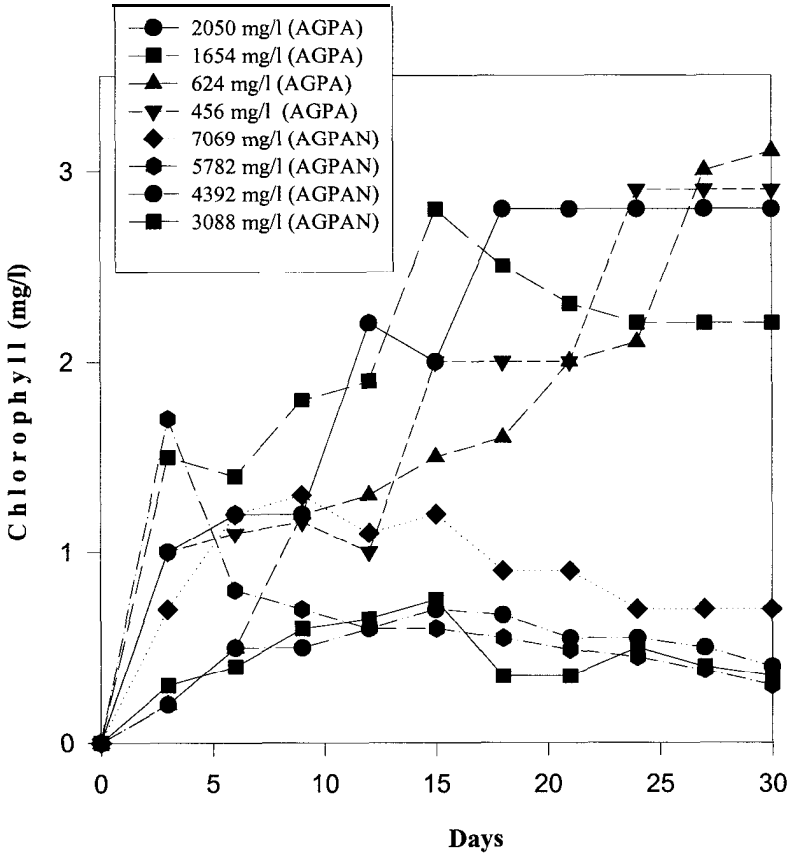

Figure 1 . Chlorophyll variations for different initial COD values

Table 3. Algae growth potential (AGP) during the experimental time

Experiments	Sample	Initial COPD ($mg.L^{-1}$)	AGP
1	AGPA	1654	8094
2	AGPA	2050	1700
3	AGPA	450	2550
4	AGPA	624	5480
5	AGPAN	5782	9780
6	AGPAN	7069	1666
7	AGPAN	4312	5810
8	AGPAN	3088	5200

Table 4. Biokinetic parameters for microalgae culture on AGPA and AGPAN culture media.

Parameter	AGPA				AGPAN			
COD ($mg.L^{-1}$)	2024	1654	624	465	7069	5782	4312	3088
$\mu(d^{-1})$	0.18	0.15	0.11.	0.10	0.02	0.18	-	0.15
$1/\mu$ (d)	5.55	6.66	9.09	10.0	50.0	55.5	-	66.5
1/COD ($L.g^{-1}$)	0.0005	0.0006	0.001	0.002	0.0014	0.0002	0.0002	0.003
μ_{max} (d^{-1})	4.05				0.69			
Ks ($mg.L^{-1}$)	0.143				0.20			

The results presented indicate that biologically treated distillery wastewater provided satisfactory conditions for microalgae growth. In both experiments (AGPA and AGPAN) hypertrophic level values were confirmed by algal bioassays.

The anaerobically treated distillery wastewater gives the necessary factors for the development of microalgae. Also, this behavior is present when a combined pretreatment (anaerobic-aerobic) was used.

In AGPA experiments microalgae growth shown normal development. In the case of APAN experiments at ten days of culture, a decay of the culture occurs. It is possible that microbial populations present in the growth media were producing a toxic substances or perhaps some protozoan forms were using the algae as a source of nutrient. The influence of the toxic substances and the degradation of them, must be studied to explain the behavior of distillery wastewater biologically treated as culture media of microalgae, taking into account the influence of the microalgae metabolism. The biokinetic parameters showed that for AGPA media μmax is

higher than μmax for AGPAN media (0.20 > 0.143)

Acknowledgments. We thank the Alexander von Humboldt Foundation for its support in this research work.

REFERENCES

American Public Health Association, APHA (1993). Standard methods for the examination of waters and wastewaters Ed. 17, American Public Health Association, Washington D.C.

De la Noüe J, de Pawn N (1988). The potential of microalgal biotechnology: a review of production and uses of microalgae. Biotechnol Adv 6: 725-770.

De Pawn N, De la Noüe J (1983). Microalgae waste water treatment system: Potentials and limits. In Phytodepuration and the employment of the biomass produced, Ed. E. Ghetti Centro Produz. Animali, Reggio Emilia, Italy, 211-287.

Hall DO , Rao KK (1994) . Photosynthesis, studies in ecology (5th Edition) Chapters 1-2. Cambridge University Press.

Hall DO (1986). The production of biomass: a challenge to our society. In: Richmond A., Handbook of microalgal mass culture CRC Press Inc 1-2, Cambridge University Press.

Leon M, Travieso L (1992). Determinación del Potencial de Crecimiento de Microalgas en Residual de Destilería. Revista CNIC. Ciencias Biológicas 23: 12-18.

Lukavsky J (1986). Metabolic activity and cell structure of immobilized algal cells. In, Organically Polluted Waters. Limnologica (Berlin) 17: 2, 355-363.

Oh-Hama T, Miyachi S (1988). Chlorella. In: Borowitzka and Borowitzka Eds.. Microalgae Biotechnology. Cambridge University Press. Cambridge U.K., 3-26.

Richmond A (1986) Microalgae of economic potential. In: Richmond A (Eds.) Handbook of Microalgal Mass Culture CRC Press Inc 1-2, Cambridge University Press.

Travieso L, Benítez F (1990). Influencia de la iluminación natural sobre las microalgas imnovilizadas. XII Scientific Seminar CNIC, La Habana, Cuba, Nov. 1990.

Valiente V, Travieso L (1994). Microalgae Cultures Catalogue. DECA-CNIC. La Habana, Cuba

Vonshak A, Kancha Raksa N, Bunnang B, Tanticharoen M (1996a). Light acclimation and photoinhibition in three Spirulina platensis (cyanobacteria) isolates. Appl Phycol 8: 35-40.

Vonshak A, Kancha Raksa N, Bunnang B, Tanticharoen M (1996b). Role of light and photosynthesis on the acclimation process of the cyanobacterium *Spirulina platensis* to salinity stress. Appl Phycol 8:119-121