Environment Protection Engineering

Vol. 15 1989 No. 1–2

JOANNA KARPIŃSKA-SMULIKOWSKA*, JANINA MOSKAL*,
MARIA PAWLACZYK-SZPILOWA*

CULTURE OF ALGAE ON DOMESTIC SEWAGE UNDER NATURAL CLIMATIC CONDITIONS

Algae were cultured for 1 year on domestic sewage. It was proved that their effective cultures could be conducted in spring, summer, and autumn which was supported by distinct increment of algal dry weight and protein content. Increment in COD values evidenced an effective utilization of some components of domestic sewage, and thus its purification.

1. INTRODUCTION

Intense culture of algae evokes a wide interest due to high possibilities of their biomass utilization in various branches of economy.

Algae contain many valuable components, so they may be used as a fertilizer improving the structure of soil by increasing the content of humus compounds [7]. In fish farming algae may be used either directly as fodder for plant-feeding fish or indirectly as feed for protozoa serving as food for fish [7]. They have also found application as fodder [10] or premixes [7] for other animals [8] because of high contents of nutritive substances [8], vitamins (B, C, D, E, K), and microelements (B, Co, Cr, J, Mo, Ni, Zn) [7]. In some countries, e.g., in Japan, China, Corea, New Zealand, Polynesia, Australia, South America, and Nigeria, some species of algae are used as food products [3].

Culture of algae on some kinds of wastewater [11] is of interest because of its role in utilization of waste substances and treatment of wastewater, as well as in neutralization of eutrophication of receivers. Our investigations performed under laboratory conditions have confirmed such a possibility [6].

Cultures of algae under natural climatic conditions were initiated in different countries in the fifties, e.g., algal farms in Trebon (Czechoslovakia) [1], Dortmund (West Germany), and Tokio [9]. Algae in pilot-scale cultures belong to the genera *Scenedesmus acutus*, *Chlorella vulgaris*, and *Spirulina maxima* [1].

* Institute of Environment Protection Engineering, Technical University of Wrocław, pl. Grunwaldzki 9, 50-377 Wrocław, Poland.

The present work is an attempt to conduct algal cultures on domestic sewage in different seasons of the year under natural atmospheric conditions.

2. MATERIAL AND METHODS

The strain *Chlorella* sp. 394 used for the culture was selected by us from 21 strains because of its high protein content and fast development [6] which guarantee the possibility of obtaining substantial amounts of biomass in short time. The experiments were conducted for 30 days under continuous culture conditions. 0.75 dm^3 of domestic sewage filtrated per 24 h through a filter paper was proportioned to 2 dm^3 culture chambers. Domestic sewage was taken from the intermediate pumping station "Szczytniki" in Wrocław. The culture was conducted under natural climatic conditions, in winter, spring, summer, and autumn. At the same time the control culture was conducted on the Uspienski medium [4].

The development of algae was estimated on the basis of dry weight and the number of cells determined in the Fuchs–Rosenthal chamber and calculated for 1 cm^3 of the culture. In the obtained biomass, the total protein content was determined by the Larry method [5].

Chemical oxygen demand of domestic sewage flowing into the culture chamber and in the effluent was determined by dichromate method and used to estimate the efficiency of treatment [2].

3. RESULTS OF INVESTIGATIONS

Culture of algae on domestic sewage was conducted in four seasons of the year. Selected climatic and metheorological parameters dominating in the separate periods of investigations are given in tab. 1.

Spring series of investigations was conducted from late April to early May. In this period, minimal temperature amounted to 272.9 K (–0.1°C) and the maximal one equalled 279.2 K (+24.2°C), while the mean daily temperature ranged from 276.5 K (+3.5°C) to 288.8 K (+15.8°C). The length of day varied from 13 h 22 min. to 15 h 22 min.

During 720 h of spring investigations, 4 days were totally sunless and the sum of sunny hours amounted to 136 h 30 min. Results obtained for this series are presented in tabs. 2 and 3. The data indicate a constant increment in the number of algal cells. The development of algae on domestic sewage was more intensive than that on the Uspienski medium. In the last day of the experiment, the number of algal cells in experimental chambers was 124% with respect to that in the control chamber.

In both experimental and control cultures, the dry weight increased with time, being however higher in experimental reactors. In the last day of investigations, dry weight of algae grown on domestic sewage amounted to 139.4% with respect to that in the control culture. Total protein content in algae grown on domestic sewage, determined in 30th day of investigations, was 302.4% of that found in the control culture. The values of COD given in tab. 3 indicate a high variation in chemical composition of domestic sewage used as culture medium. That is why, each time the COD reduction (in percent) of a treated sewage was calculated with respect to the sewage being proportioned. For the particular series, these values are also presented in tab. 3.

Table 1

Climatic and meteorological data for spring, summer, autum, and winter series of experiments

Season of year	Length of day h	Number of sunless days	Insolation h/day	Total number of sunny hours in series h	Sunny hours %	Mean daily temperatures K (°C)	Range: Minimal daily temperatures K (°C)	Range: Maximal daily temperaures K (°C)
Spring	13 h 22 min 15 h 22 min	4	11 h 10 min	136.3	18.9	276.5 (+3.5)– 288.8 (+15.8)	272.9 (–0.1)– 287.1 (+14.1)	280.3 (+7.3)– 279.2 (+24.2)
Summer	16 h 39 min 13 h 4 min	5	15 h 20 min	171.57	23.8	285.4 (+12.4)– 296.5 (+23.5)	281.1 (+8.1)– 288.9 (+15.9)	283.3 (+10.3)– 303.6 (+30.6)
Autumn	11 h 28 min 9 h 43 min	8	9 h	105.6	14.7	277.0 (+4)– 290.9 (+17.9)	273.5 (+0.5)– 286.1 (+13.1)	279.7 (+6.7)– 298.2 (+25.2)
Winter	9 h 55 min 10 h 14 min	2	8 h 4 min	18.1	12.6	264.0 (–9)– 271.8 (–1.2)	254.9 (–18.1)– 270.1 (–2.9)	268.1 (–4.9)– 272.8 (–0.2)

Table 2

Development of *Chlorella* 394 population in spring, summer, and autum

Time of culture days	Season of year	Cell numbers · $10^6/cm^3$			Dry weight of algae mg/dm^3			Total protein mg/dm^3		
		Experimental chamber I (domestic sewage)	Control chamber II (medium)	I : II %	Experimental chamber I (domestic sewage)	Control chamber II (medium)	I : II %	Experimental chamber I (domestic sewage)	Control chamber II (medium)	I : II %
1	2	3	4	5	6	7	8	9	10	11
0	Spring	13.528	13.528	100.0	0.30	0.30	100.0	48.0	48.0	100.0
1		13.325	13.156	101.3	0.25	0.29	86.2	32.0	49.0	65.0
5		13.784	13.562	101.6	0.38	0.51	74.5	167.2	60.2	278.7
9		14.418	13.806	104.4	0.54	0.57	94.7	264.6	76.4	246.3
13		14.625	14.050	104.1	0.86	0.44	195.6	283.8	73.0	388.8
16		15.412	14.125	109.1	1.09	0.62	175.8	305.2	89.9	339.5
19		17.650	14.875	118.6	1.09	1.04	103.5	311.7	134.1	232.4
23		18.471	15.659	117.9	1.22	1.05	114.7	319.6	134.4	237.8
27		20.525	16.362	125.4	1.41	1.11	127.2	410.3	142.1	288.7
30		21.190	17.068	124.0	1.51	1.08	139.4	440.9	145.8	302.4
0	Summer	14.130	14.130	100.0	0.25	0.25	100.0	46.7	46.7	100.0
1		13.287	13.312	99.8	0.20	0.18	90.0	32.2	55.0	58.5
4		13.675	12.525	109.2	1.34	0.75	178.5	278.7	78.0	357.3
7		14.375	14.250	100.9	1.12	0.58	191.9	280.0	51.6	542.6
11		15.416	13.362	115.4	1.13	0.62	179.7	282.5	63.9	442.1
13		16.132	14.725	109.5	1.24	0.64	190.2	322.4	70.0	460.6
17		17.800	15.509	114.8	1.38	0.67	202.6	303.6	65.6	462.8
20		18.941	16.128	117.4	1.45	0.70	207.0	304.5	57.4	530.5
25		20.775	18.369	113.1	1.41	0.97	144.7	267.9	106.7	251.0
28		23.600	19.041	123.9	1.46	0.84	170.8	262.8	84.0	312.8
30		25.144	20.750	121.2	1.42	0.88	161.4	255.6	105.6	242.0

1	2	3	4	5	6	7	8	9	10	11
0	Autumn	14.200	14.200	100.0	0.23	0.23	100.0	46.4	46.4	100.0
1		14.500	13.150	110.3	0.28	0.26	107.7	50.4	48.4	104.1
3		15.494	12.813	120.9	0.36	0.34	104.7	54.0	51.0	105.9
8		14.875	13.481	110.3	0.43	0.41	104.1	81.7	69.7	117.2
11		11.494	10.555	108.0	0.60	0.46	130.1	132.0	82.8	159.4
15		11.469	10.075	113.0	1.04	0.68	152.9	291.2	142.8	203.9
18		13.155	12.244	107.4	1.24	0.68	181.0	359.6	102.0	352.5
22		13.850	12.587	110.0	1.39	0.83	166.8	389.2	116.2	334.9
25		12.900	12.218	105.6	2.09	1.06	196.7	627.0	159.0	394.3
30		13.050	12.700	102.7	2.14	1.58	135.5	642.0	252.8	253.9

Table 3

Variations in COD of *Chlorella* 394 culture in spring, summer, and autumn

Season of year	Time of culture days	COD mg O_2/dm^3		Reduction of COD	
		Raw domestic sewage	Treated domestic sewage	mg O_2/dm^3	%
Spring	0	408.0	–	–	0
	1	408.0	226.0	182.0	44.6
	5	408.0	105.0	213.0	52.2
	9	446.0	135.9	310.1	69.7
	13	446.0	139.0	307.0	71.1
	16	326.0	81.3	244.7	75.1
	19	233.8	33.6	200.0	85.5
	23	233.8	44.4	189.4	81.0
	27	281.0	30.3	250.7	89.2
	30	248.0	20.3	227.7	92.5
Summer	0	284.0	–	–	0
	1	384.0	205.4	179.0	46.5
	4	302.0	97.2	204.8	67.8
	7	302.0	70.3	231.7	60.0
	11	350.0	52.5	297.5	75.0
	13	231.0	64.7	166.3	72.0
	17	216.0	72.0	144.0	66.7
	20	282.0	74.2	207.8	73.7
	25	282.0	68.3	213.7	75.8
	28	316.0	84.1	231.9	73.4
	30	316.0	68.9	247.1	78.2
Autumn	0	244.0	–	–	0
	1	244.0	124.4	119.6	49.0
	3	244.0	115.0	129.0	52.9
	8	209.0	107.9	101.0	48.3
	11	209.0	106.9	102.1	54.5
	15	217.8	91.3	126.5	58.1
	18	217.8	91.0	126.8	58.2
	22	276.4	82.0	194.4	70.9
	25	276.4	100.0	176.4	63.8
	30	208.2	82.6	125.6	60.5

In spring, the COD values of the proportioned domestic sewage ranged from 233.8 to 446 mg O_2/dm^3. By comparing the COD values of the proportioned and treated domestic sewage, it could be stated that large amounts of the sewage components have been utilized by algae. The COD reduction in the last day of experiment was as high as 92.55%.

The second (summer) series of experiments lasted from mid June to mid July. The highest temperatures recorded in this series ranged from 283.3 K (+10.3°C) to 303.6 K (+30.6°C), whereas mean daily temperatures varied from 285.4 K (+12.4°C) to 296.5 K (+23.5°C), and the lowest daily temperatures ranged between 281.1 K (+8.1°C) and 288.9 K (+15.9°C). The sum of sunny hours during 720 h investigations amounted to 171 h 57 min. In this series, the increment of algae grown on domestic sewage was much higher than that in control culture.

The number of algal cells determined in experimental reactors was higher than that in control chambers. In the last day of investigations, the number of cells in experimental chamber amounted to 121.2% with respect to that determined in control chamber. Like in spring series, the increments in dry weight and total protein of algae grown in experimental cultures were much higher. In 30th day of investigations, dry weight and total protein of algae in experimental reactors determined with respect to those in control culture amounted to 161.4% and 243%, respectively.

The COD values of proportioned and treated sewage were also subject to high variations, ranging from 216 to 384 mg O_2/dm^3 for the former, and from 52.5 to 205.4 mg O_2/dm^3 for the latter. COD reduction varied from 46.5% to 78.2%.

Autumn series of experiments was conducted from October to November. During this period mean daily temperatures ranged from 277.0 K (+4°C) to 290.9 K (+17.9°C). The lowest daily temperature amounted to 273.5 K (+0.5°C), while the highest one was equal to 298.2 K (+25.2°C). In this series, the sum of sunny hours was 105 h 6 min., 8 days were totally sunless.

In this series of investigations, the numbers of algal cells both in experimental and control cultures decreased with respect to $14.2 \cdot 10^6$ cells/cm^3 at the beginning of this experiment. In the course of the investigations, the minimal number of cells in experimental and control chambers amounted to about $11.5 \cdot 10^6/cm^3$ and about $10^6/cm^3$, respectively, thus the rate of cell divisions significantly decreased. In the last day of experiment, the number of cells in experimental reactors determined with respect to control samples was the lowest one being scarcely 102.7%.

Dry weights of algae grown in experimental and control chambers increased with time. In the 30th day of experiment, dry weight of algae in experimental chambers, determined with respect to that in control samples, amounted to 135.5%. The same trend was observed while determining the total protein, which in algae grown on domestic sewage, determined with respect to that in control reactor, was equal to 253.9%.

The COD values of proportioned sewage ranged from 208.2 to 276.4 mg O_2/dm^3. Culture of algae on domestic sewage led to the reduction of pollutants, manifested by the reduction of COD value. COD values of treated sewage varied within 82.0 and 124.4 mg O_2/dm^3. In this series of experiments, COD reduction ranged from 48.3% to 70.9%.

Low temperatures recorded in winter made our investigations impossible. Minimum daily temperature ranged from 254.9 K (−18.1°C) to 270.1 K (−2.9°C), and mean daily temperature varied within 264 K (−9°C) and 271.8 K (−1.2°C).

4. DISCUSSION OF RESULTS

The obtained results show that the cultures of algae can be conducted in spring, summer, and autumn.

The development of algae in these seasons of the year was determined basing on the cell number, dry weight, and total protein content. While comparing the obtained data, it has been stated that the intensity of algal growth in domestic sewage was much higher than that in control culture. This proves that the sewage being investigated contains some components advantageous for the development of algal population.

In spring and summer series, the increments in biomass and total protein of algae were accompanied with the increment in cell numbers. Only in autumn the stated drop of the cell number with respect to the inoculum was associated with distinct increments in dry weight and total protein. This phenomenon was stated both in experimental and control chambers. Dry weight in experimental and control chambers ranged from 0.23 to 2.14 mg/dm^3 and from 0.23 to 1.58 mg/dm^3, respectively, whereas total protein, equal initially to 46.4 mg/dm^3, increased in these chambers to 542 mg/dm^3 and 252.8 mg/dm^3, respectively.

It seems possible that the decrease in the number of algal cells observed in autumn was related to a greater number of sunless days (8 sunless days recorded in this time). This hypothesis is supported by the fact that in spring the numbers of algal cells increased distinctly (from $13.528 \cdot 10^6$ to $21.190 \cdot 10^6/\mathrm{cm}^3$) though thermal conditions were similar to those in autumn, but the number of sunless days was two times smaller (4 sunless days). Thus, climatic conditions and varying composition of sewage could result in the decreasing rate of cell divisions with a simultaneous increment in biomass.

Hence, the sizes of newly formed cells had to be larger and this fact was confirmed by microscopic observations.

Increments in biomass and total protein content associated with a reduced value of COD prove that some sewage components are utilized by algae. In all investigation periods, COD values of the proportioned domestic sewage used as culture medium varied, giving the evidence to the variable chemical composition of this sewage. The highest COD removal was stated in spring and amounted to 4.6%–92.5%. In summer, it varied from 46.5% to 78.2%, and in autumn, from 48.3% to 70.9%.

The obtained results indicate that in autumn the amount of high-protein biomass obtained in experimental reactor was the highest, the effects of sewage treatment being somewhat worse, in particular, if compared with those obtained in spring.

Summing up, it should be stated that algal culture on domestic sewage (used as a culture medium) can be conducted in natural climatic conditions. Such a culture may be conducted during the whole year, if the culture chambers are placed in rcoms illuminated periodically.

REFERENCES

[1] BOGEN H. J., *Biotechnika – osiągnięcia i perspektywy*, Warszawa 1987.

[2] HERMANOWICZ W., DOŻAŃSKA W., DOJLIDO J., KOZIOROWSKI B., *Fizyczno-chemiczne badania wody i ścieków*, Arkady, Warszawa 1976.

[3] ILCZUK Z., *W poszukiwaniu nowych źródeł białka*, R XXIV, No. 3 (1968), pp. 142–149.

[4] JANKOWSKI A., *Badania nad selekcją glonów dla potrzeb kultur masowych*, Instytut Zootechniki, Kraków 1964.
[5] MEJBAUM-KATZENELLENBOGEN W., MOCHNACKA J., *Kurs praktyczny z biochemii*, PWN, Warszawa 1964.
[6] MOSKAL J., PAWLACZYK-SZPILOWA M., DZIENDZIEL E., *Ocena rozwoju hodowli wybranych szczepów glonów na ściekach bytowo-gospodarczych*, (in press).
[7] PODBIELKOWSKI Z., *Glony*, WSiP, Warszawa 1978.
[8] PROŃCZUK A., ROSZKOWSKI W., *Nowe produkty białkowe – charakterystyka produkcji i wartości odżywczych*, No. 4 (1977), pp. 159–170.
[9] TAMY'a H., *Masowa uprawa glonów. Życie i człowiek*. Tom – *Roślina*, PWN, Warszawa 1965.
[10] WIERNY A., *Próby wykorzystania alg do żywienia zwierząt*, Przegląd hodowlany, No. 10 (1966), pp. 16–20.
[11] WRÓBEL S., *Eliminacja mineralnych związków azotu ze ścieków przy udziale roślin niższych*, Wiadomości ekologiczne, Vol. XVII (1972), pp. 29–35.

HODOWLA GLONÓW NA ŚCIEKACH KOMUNALNYCH W NATURALNYCH WARUNKACH KLIMATYCZNYCH

W ściekach bytowo-gospodarczych hodowano glony przez okres 1 roku. Wykazano, iż możliwe jest prowadzenie wydajnej hodowli przez znaczną część roku, o czym świadczy przyrost suchej masy i białka glonowego. Obniżenie chemicznego zapotrzebowania tlenu wskazuje, iż glony efektywnie obniżają stopień zanieczyszczenia ścieków bytowo-gospodarczych.

РАЗВЕДЕНИЕ ВОДОРОСЛЕЙ НА КОММУНАЛЬНЫХ СТОЧНЫХ ВОДАХ В НАТУРАЛЬНЫХ КЛИМАТИЧЕСКИХ УСЛОВИЯХ

В бытово-хозяйственных сточных водах разводили водоросли в течение 1 года. Обнаружили, что возможно ведение эффективного разведения в течение значительной части года, о чем свидетельствует привес сухой массы и белков. Понижение ХПК показывает, что водоросли эффективно понижают степень загрязнения бытово-хозяйственных сточных вод.