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

ON THE POSSIBILITY OF OBTAINING ALGAL PROTEIN IN THE PROCESS OF DOMESTIC SEWAGE TREATMENT

The purpose of the investigation was to answer the question whether the components of domestic sludge can be used for culture of algae. It could be expected that such a culture, because of high dynamics of its development and protein content, will yield a remarkable amount of valuable biomass. Control culture was conducted at the same time on mineral Uspienski medium. In culture of *Chlorella* 394 conducted under constant laboratory conditions, the number of algal cells, biomass and protein content increased. The amount of protein obtained from 1 dm^3 of domestic sewage was equal to 342.9 mg. At the same time the concentration of pollutants in the effluent was reduced. The culture of this strain on mineral medium gave much worse results, the increments in the number of cells, biomass and protein content being distinctly lower.

1. INTRODUCTION

Intense search for new unconventional sources of protein, which could be used as a component of fodder, has been conducted all over the world since a number of years. The attention was paid to algae because of high dynamics of their development as well as high concentrations of proteins and other valuable components [9].

Strains of algae of the genera *Chlorella* and *Scenedesmus* appeared to be particularly useful, since their biomass is composed of about 50% of protein [12] with high content of exogeneous amino acids [6]. These algae may also be rich sources of vitamins A, B_1, B_2, B_6, B_{12}, C, K, nicotinic acid, folic acid, and biotin [8], [9].

A direct usage of some species of algae as feed encounters, however, serious difficulties. They are chiefly due to the fact that thick cell walls composed of cellulose cannot be digested by animals other than ruminants. The latter have at their disposal cellulolytic enzymes which make possible the decomposition of algal cell walls. Hence, only in such a case algal protein might be used as a component of fodder.

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

Algae, in which metabolic process runs correctly, produce some substances of antibiotic and toxic nature. Purine bases of nucleic acids, high amounts of which are contained in algae, are in metabolic processes transformed into uric acid which can cause some disturbances in animal organisms. That is why algae should not be used as a basic protein feed, but as its supplement. There are, however, some species of algae containing smaller amounts of nucleic acids, e.g., *Spirulina maxima* [2] from which protein feeds produced on industrial scale [9].

Another property of algae is their ability to biocumulate many contaminants, such as metals and aromatic hydrocarbons [1], [6], [7] harmful to both men and animals.

Algal cultures on domestic sewage, which is a waste material, make it possible to obtain cheap and valuable algal biomass, contributing at the same time to the reduction of pollutants in water environment.

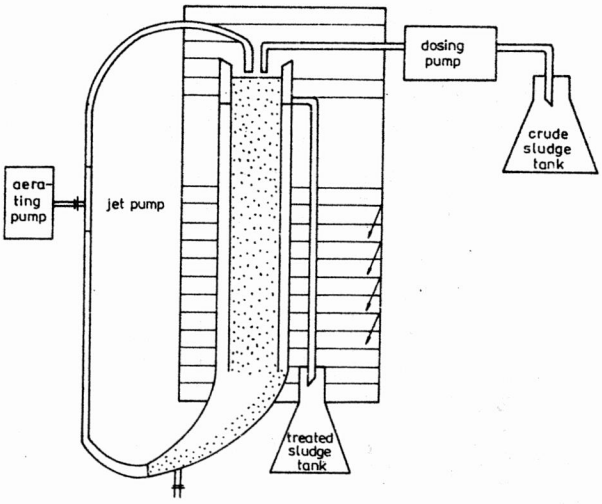
The purpose of the present paper was to study the possibility of algal cultures (*Chlorella*, strain 394) on nutrients contained in domestic sewage.

2. MATERIAL AND METHODS

The investigations were performed on algae of the genus *Chlorella*, strain 394. As it follows from our investigations [3], its culture on domestic sewage gave a remarkable increment of biomass with the highest protein content. The strain 394, obtained from the Institute of Experimental Zootechnique at Zator near Cracow, was isolated from among other 21 strains subject to investigations. This strain, after having been adapted to domestic sewage for 14 days, was used as experimental culture. Domestic sewage came from the intermediate sewage pomping station "Szczytniki" in Wrocław. The same strain of algae growing on the Uspienski mineral medium [11] was a control culture. The culture of algae was conducted for 29 days in 3 replications.

The culture of algae by a continuous method was conducted in 2 dm^3 glass reactors, consisting of 2 glass pipes 68 cm long, of which one was placed into the second one. Crude domestic sewage (0.75 dm^3/24 h) was dosed to the internal pipe and after its sedimentation clarified in the external pipe. Scheme of the experimental set-up is presented in figure. Domestic sewage was fed by means of metering pump of the type 309, produced by UNIPAN and aerated by the pump manufactured in Veb Elmet Hettstedt. Laboratory tests were performed in air-conditioned room at 293 ± 2 K (20 ± 2°C) at fluorescent light of 4500 lux. The number of algal cells was determined in Fuchs–Rosenthal chamber. The obtained results were calculated for 1 dm^3 of algal culture. In order to determine dry weight of algae, 10 cm^3 of the culture was subject to centrifugation. Biomass was then washed with distilled water and centrifuged again. Supernatant was rejected and the sediment suspended in 10 cm^3 of distilled water and dried at 378 K (105°C) until the weight became constant. Dry weight was calculated for 1 cm^3 of the culture.

Total protein content was determined in biomass of algae after 10 cm^3 dose of their culture was centrifuged. Algal biomass after supernatant removal was washed with physiological solution, treated with 10 cm^3 of 3% sodium dodecyl sulphate and put into a freezer at 259 K (–14°C) for 2 h. Thereupon, the samples were defrosted and centrifuged; total protein was determined in supernatant on spectrophotometer (C. Zeiss, Jena) at the wavelengths $\lambda = 260$ nm and $\lambda = 280$ nm. The content of total protein was read out from the nomogram [10]. Chemical oxygen demand in the flowing-in and -out wastewater was determined by dichromate method [4].



Scheme of experimental set-up

3. RESULTS

Synthetic ability of algae grown on nutrient compounds present in domestic sludge was investigated under laboratory conditions.

The obtained results are presented in tab. 1. As it may be easily seen, the growth of algae on domestic sewage was much more intensive than on the Uspienski medium. This fact is proved by both the number of cells and dry weight of algae falling to 1 dm^3. The data concerning protein content in the examined material are of a particular interest. Biomass of algae grown on domestic sewage was characterized by distinctly higher content of proteins than that of algae culture conducted on mineral medium. The number of algal cells in experimental chamber increased gradually up to the 29th day of experiment. In the last few days, this number slightly dropped. In control chamber the number of cells initially increased, but after few days it remained almost unchanged (16.8–18.2 cm^3/cm^3). In the last day of investigations, the number of cells amounted to 134% of that in control chamber, assumed to be 100%. Dry weight of algae increased both in experimental and control chambers. In the 28th day of experiment, the amounts of biomass in experimental and control chambers increased 7 and 5 times, respectively.

The total protein content (calculated for 1 mg of dry weight) in biomass of algae grown on domestic sewage increased distinctly in the first period, later on it remained almost unchanged, whereas in control chamber it slightly varied during the whole time of experiment. In the last day of investigations, the amount of protein in biomass of algae on domestic sewage was two times higher than that determined in the inoculant.

The measurements of COD showed that algae grown on domestic sewage contributed to a remarkable reduction of pollutant concentrations in the effluent. COD removal in the treated domestic sludge with respect to that in the crude one ranged from 35 to about 65%.

Table 1

Development of *Chlorella* 394 algae and the COD values

Time of culture days	Cell number mln/cm^3 of culture			Dry weight of algae mg/cm^3			Total protein mg/mg of dry weight			COD mg O_2/dm^3		
	Experimental set-up I	Control set-up II	Control to experimental set-up ratio %	Experimental set-up I	Control set-up II	Control to experimental set-up ratio %	Experimental set-up I	Control set-up II	Control to experimental set-up ratio %	Crude sludge	Treated sludge	COD removal %
0	14.6	14.6	100	0.22	0.22	100	0.160	0.160	100	282.2	0	–
3	15.2	13.7	110.9	0.30	0.24	125.0	0.212	0.110	192.7	282.2	98.26	65.2
8	17.5	17.4	100.6	0.62	0.58	106.9	0.305	0.115	265.2	208.0	134.93	35.1
11	18.2	16.8	108.3	0.98	0.78	125.6	0.331	0.118	280.5	208.0	106.67	48.7
15	21.5	17.5	122.8	1.24	0.91	136.3	0.308	0.132	233.3	271.6	133.08	50.7
18	22.7	18.2	124.7	1.22	0.97	125.8	0.358	0.166	215.7	271.6	113.33	58.3
22	23.0	17.3	131.4	1.36	0.95	143.1	0.330	0.162	203.7	206.4	129.00	38.9
25	24.3	17.2	141.3	1.49	1.07	139.2	0.315	0.150	210.0	206.4	104.64	49.3
29	23.3	17.4	133.9	1.48	1.13	131.0	0.336	0.158	212.6	206.4	97.98	41.1

4. FINAL REMARKS

From the analysis of the obtained results it follows that the growth of algae of genus *Chlorella*, strain 394, on domestic sewage is more intensive than that on mineral Uspienski medium. This observation is verified by comparison of all results obtained for experimental and control chambers.

Protein contents in dry weight of algae present in chambers and in the treated effluents, determined during the period of investigations, were used to compute the amounts of protein obtained from the algal cultures on crude domestic sewage and on mineral medium. The obtained values are presented in tab. 2.

Table 2

Protein content in experimental and control set-ups during the culture *Chlorella* 394

Time of culture days	Control set-up mg of protein		Experimental set-up mg of protein	
	In chamber $V = 2$ dm^3	In effluent $V = 0.75$ dm^3	In chamber $V = 2$ dm^3	In effluent $V = 0.75$ dm^3
0	70.4	0	70.4	0
1	64.5	24.2	89.3	33.6
2	55.5	20.8	108.3	40.6
3	52.8	19.8	127.2	44.7
4	68.9	25.8	177.4	66.5
5	85.1	31.9	227.6	85.3
6	101.0	37.9	277.8	104.2
7	117.2	43.9	328.0	123.0
8	133.4	50.0	378.2	141.8
9	150.3	56.4	468.4	175.6
10	167.2	62.7	558.6	209.5
11	184.1	69.1	648.8	243.3
12	198.1	74.3	677.5	254.1
13	212.1	79.5	706.2	264.1
14	226.1	84.8	734.9	275.6
15	240.2	90.1	763.8	286.4
16	267.5	100.3	800.4	300.1
17	294.8	110.5	837.0	313.9
18	322.0	120.7	873.5	327.6
19	318.5	119.4	879.5	329.8
20	315.0	118.1	886.2	332.3
21	311.5	116.8	892.8	334.8
22	307.8	115.4	897.6	336.3
23	312.2	117.0	910.0	341.2
24	316.6	118.5	923.7	346.4
25	321.0	120.4	938.7	352.0
26	330.0	123.7	952.7	357.3
27	339.0	127.1	966.7	362.5
28	348.0	130.5	980.7	367.8
29	357.1	133.9	994.6	373.0

Sum of protein in chamber and effluent, 2827.1 mg.
Protein from 1 dm^3 sludge, 119.3 mg

Sum of protein in chamber and effluent, 8144 mg.
Protein from 1 dm^3 sludge, 119.3 mg

As it may be seen, the culture of algae conducted for 29 days gave 8144 mg of protein from 23.75 dm^3 of domestic sewage, while the algae grown on the Uspienski medium gave only 2827.1 mg of protein. Hence, it follows that 1 dm^3 of domestic sewage yielded 342.3 mg of protein, whereas the mineral medium gave only 119 mg. The results obtained by us prove that remarkable amounts of protein can be obtained from cultures of algae on domestic sewage. This statement was the basis for further investigations, the purpose of which was to examine the possibility of algal cultures under natural climatic conditions in four characteristic seasons of the year.

5. CONCLUSIONS

1. The investigations have shown that the culture of *Chlorella*, strain 394, can be conducted on mechanically treated domestic sewage, in constant laboratory conditions.

2. The culture of *Chlorella* 394 on domestic sewage yielded 342.9 mg of protein (counted for 1 dm^3 of sewage).

3. *Chlorella* 394 grown on domestic sludge contributed to the reduction of COD from 35.1% to 62.5%.

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BADANIE MOŻLIWOŚCI UZYSKANIA BIAŁKA GLONOWEGO W PROCESIE OCZYSZCZANIA ŚCIEKÓW BYTOWO-GOSPODARCZYCH

Podjęto badania nad możliwością wykorzystania składników ścieków bytowo-gospodarczych do hodowli glonów. Można było przypuszczać, iż hodowla glonów na ściekach, z powodu dużej dynamiki rozwoju i wysokiej zawartości białka, pozwoli uzyskać znaczną ilość cennej biomasy. Równolegle prowadzono bada-

nia kontrolne, w których glony hodowano na mineralnym podłożu Uspieńskiego. W hodowli glonów *Chlorella* 394 w stałych warunkach laboratoryjnych stwierdzono przyrost liczby komórek glonów, biomasy oraz przyrost zawartości białka. Z 1 dcm^3 ścieków bytowo-gospodarczych uzyskano 342.9 mg białka. Równocześnie stwierdzono obniżenie stężenia zanieczyszczeń w odpływających ściekach. Znacznie gorszy przyrost biomasy, liczby komórek i zawartości białka uzyskano prowadząc hodowlę glonów na mineralnym podłożu.

ВОЗМОЖНОСТИ ПОЛУЧЕНИЯ БЕЛКОВ ВОДОРОСЛЕЙ В ПРОЦЕССЕ ОЧИСТКИ БЫТОВО-ХОЗЯЙСТВЕННЫХ СТОЧНЫХ ВОД

Исследована возможность использования составных элементов бытово-хозяйственных сточных вод для разведения водорослей. Можно было судить, что разведение водорослей на сточных водах позволит получить большое количество биомассы. Параллельно вели контрольные исследования, в которых водоросли разводили на минеральном основании Успенского. В разведении водорослей *Chlorella* 394 в постоянных лабораторных условиях установили прирост числа клеточек водорослей, биомассы и содержания белков. Из 1 $дм^3$ бытово-хозяйственных сточных вод получили 342,9 мг белков. Одновременно установили понижение концентрации загрязнений в стекающих сточных водах. Значительно худший прирост биомассы, числа клеточек и содержания белков получили в разведении водорослей на минеральном основании.