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DAIRY WASTEWATER TREATMENT USING MEMBRANE FILTRATION SUPPORTED BY BIOLOGICAL PROCESSES

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Key words: wastewater treatment, dairy wastewater, membrane filtration, bioaugmentation, Microalgae.

Abstract: This paper presents efficient treatment methods for dairy wastewater using membrane techniques and applying the biological utilization of secondary wastes. The influence of ultrafiltration (UF) and both ultra- and nanofiltration (UF/NF) systems on the removal efficiency of the contaminants was determined. It has been found that the UF enables the removal of larger organic compounds, whose presence on the NF membrane surface would reduce its efficiency. The recovery of water from these processes is associated with the generation of retentate, which is difficult to treat. For the utilization of the retentate, biological methods based on bioaugmentation process and microalgal culture were used. The combination of both these methods contributed to the significant reduction in the content of nutrients in the regenerated water (the total nitrogen <math><1.0\text{ mg/dm}^3</math>, the total phosphorus <math><0.1\text{ mg/dm}^3</math>). Furthermore, it was possible to obtain a microalgae biomass, which becomes more commonly used, e.g., as renewable energy resources. These studies are part of the prospective trends in the development of the bioeconomy, especially in a closed circuit.

Oczyszczanie ścieków mleczarskich przy użyciu filtracji membranowej wspomaganej procesami biologicznymi

Słowa kluczowe: oczyszczanie ścieków, ścieki mleczarskie, filtracja membranowa, bioaugmentacja, mikroalgi.

Streszczenie: W pracy przedstawiono metody efektywnego oczyszczania ścieków mleczarskich z zastosowaniem technik membranowych i procesów biologicznych. Zbadano wpływ ultrafiltracji (UF) oraz zintegrowanych procesów ultra- i nanofiltracji (UF/NF) na skuteczność usuwania zanieczyszczeń. Stwierdzono, że UF umożliwia usuwanie większych cząstek organicznych, których obecność na powierzchni membrany NF pogorszyłaby jej wydajność. Odzyskiwanie wody z zastosowaniem tych procesów wiąże się z powstawaniem retentatu, który jest trudny do uzdatnienia. Do tego celu w pracy wykorzystano metody biologiczne, oparte na bioaugmentacji i hodowli mikroalg. Połączenie obu tych metod przyczyniło się do znacznego zmniejszenia zawartości związków biogenych w wodzie regenerowanej (azot ogólny <math><1,0\text{ mg/dm}^3</math>, fosfor ogólny <math><0,1\text{ mg/dm}^3</math>). Ponadto uzyskano biomasę alg, która może znaleźć zastosowanie m.in. jako odnawialny surowiec energetyczny. Podjęte badania wpisują się w perspektywiczne trendy rozwoju biogospodarki, w szczególności w obiegu zamkniętym.

Introduction

The food industry is one of the most important and fastest growing sectors of the economy. The share in the sales value of the food industry among industries in Poland is about 24%, and it is one of the highest in Europe [1]. The dairy industry, considered the largest source of food processing waste, is also one of the largest in the food industry, both in terms of total raw material and water consumption. It is estimated that 1 dm³ of processed milk consumes 1.44 dm³ of water, while the

production of cheese, butter or curd cheese is even more water-consuming (1.6–4.0 dm³ of water per 1 dm³ of milk), while milk powder requires yet more water (15–20 dm³ water per 1 dm³ of milk) [2]. Approximately 80–90% of used water becomes wastewater, which forces the needs for economical water management [3]. The effective use of water resources in production processes requires counteracting the wastage of water, which means the necessity to reduce the amount of industrial wastewater and to recover as much water as possible for reuse.

Modern technologies of wastewater treatment that permit closed water circuits involve processes of membrane filtration, which have already found numerous applications in the dairy industry [4–7]. Appropriately selected membrane techniques provide the opportunities to fractionate industrial wastewater mixtures into groups of approximately defined composition, thereby facilitating further use or utilization. In practice, membrane filtrations have some limitations. Thus, wastewater requires an appropriate pre-treatment step before it is directed into the filtration system. A no less important problem, requiring an urgent solution, is the utilization of retentate, which is a concentration of pollutants that did not pass through the filtration membrane. The separation of high molecular weight organic substances (fats, proteins, polysaccharides) from minerals would enable the reuse of retained compounds as components of culture media for the production of high value-added biotechnological products. One example assumes the use of food industry wastewater for fungal culture *Rhizopus oryzae* [8] or xanthan production [9]. Wastewater from the dairy industry, owing to the high water content and nutrients, is essential for microorganism growth including algae, which might be used for the production of bioenergy [10,11].

For the treatment of dairy wastewater, biological methods are also used, among which, due to its high efficiency and relatively low costs, the most common is activated sludge process [12]. This method uses activated sludge flocs that form as a result of combining heterotrophic bacteria with organic and inorganic particles. Enzymes produced by these bacteria are capable of decomposing macromolecules such as proteins, lipids, and fats into more assimilable forms, while inorganic compounds are assimilated immediately. As a result of the biochemical processes which occur, the pollutants are removed from the wastewater. The use of biological methods for the treatment of wastewater from the food industry has already been widely described in the literature [13–16]. Dairy wastewaters can be treated using systems of aerated lagoons, trickling filters, sequencing batch reactor (SBR), upflow anaerobic sludge blanket (UASB) reactors, and anaerobic filters. In some cases, anaerobic, aerobic, and anoxic series of processes are used [16]. Owing to the limitations of prolonging activated sludge processes, there is an increasing need to enhance these methods by commercially available bioactivators of selected strains of microorganisms, which are capable of oxidizing specific types of pollutants such as proteins, fats, and oils [17–20]. Bioaugmented inocula should meet specific requirements of ability to degrade pollutants and contaminants *in situ*, and after inoculation, to be durable and competitive. The result of bioaugmentation carried out by bacteria depends on the relationship between them and the

environment they occupy in terms of survival, activity, and migration [19]. Therefore, the production of such inocula involves the selection of bacteria from the same ecological niche.

An alternative treatment for wastewaters as well as secondary wastes generated during membrane filtration is to use them as media for the culture of single-celled algae [21]. Microalgae (among others, *Chlorella sp.*) are the most promising organisms due to their fast growth rate, low culturing requirements, high photosynthetic yield, and the ability to accumulate large amounts of lipids (some algae species contain even 75% of lipids in the dry residue [22]). Therefore, their valuable biomass is a renewable source of chemicals for many industrial applications [23, 24]. The ability to rapid reproduction has made them more desirable substrates than edible grains. In particular, microalgal lipids have become a promising energy source for biofuels, since the use of edible oil plants for this purpose had raised concerns about food safety [25, 26]. An industrial scale of algal lipids production is limited by high costs. Thus, different strategies are being developed to increase biomass productivity, especially desirable metabolites, reducing the costs of the culture medium, (through simplifying the way it is prepared, using low cost synthetic substitutes, and reducing energy consumption) [27].

The promising sources of nutrients for microalgae are municipal and industrial wastewater (dairy wastewater in particular), and effluents resulting from the biological decomposition of organic matter rich in easily absorbed nitrogen and phosphorus compounds [28]. Wastewater treated in anaerobic conditions is subjected to further technological processes until acceptable and safe levels of nutrients for the environment are achieved. According to the literature, the content of nutrients such as nitrogen and phosphorus in dairy wastewater is sufficient for effective algal biomass production [21]. Microalgae can be used in bioremediation of polluted waters, thus contributing to the protection of water reservoirs against the eutrophication [29,30]. The natural biosorption properties of algae can also be used to limit heavy metal ions from wastewaters and to recover valuable ones [31].

The aim of the study was to investigate the possibility of the effective treatment of dairy wastewater using membrane techniques along with the utilization of the secondary wastes. To achieve this goal, it was necessary to study the influence of the proposed membrane filtration processes on the efficiency of removal of pollutants from raw dairy wastewater and the use of biological methods, based on the bacterial and microalgal culture, in order to utilize the retentates. This proposed method of dairy wastewater treatment is a part of the strategy for sustainable development by saving water resources and minimizing environmental emissions resulting from wastewater and secondary wastes from industrial sources.

1. Materials and methods

1.1. Dairy wastewater

The object of the research was the raw dairy wastewater generated during the cleaning of the technological line in the production plant. The experiment included the analyses of raw wastewater, permeate, and retentate after

ultra- (UF) and nanofiltration (NF), as well as samples after biological wastewater treatment using bacterial and microalgal culture. Due to the presence of suspended solids, it was necessary to pre-treat the dairy wastewater before the membrane filtration processes. For this purpose, a polypropylene filter bag (removal rating 5 μm) was used. The values of the physico-chemical parameters of raw and filtered dairy wastewater are listed in Table 1.

Table 1. Physico-chemical properties of dairy wastewater

Parameter	Value	
	Raw wastewater	Filtered wastewater
Total nitrogen, mg/dm ³	51.7	40.5
Total phosphorus, mg/dm ³	9.7	8.7
Chemical oxygen demand, mg/dm ³	2 430	1 090
Volatile fatty acids, mg/dm ³	139	136
Total organic carbon, mg/dm ³	1 551	571
pH	8.9	8.7
Conductivity, $\mu\text{S}/\text{cm}$	840	933
Dry residue, mg/dm ³	1 980	1440
Turbidity, NTU	844	400
Total suspended solids, mg/dm ³	1 092	426
Sulphates, mg/dm ³	210	143
Calcium, mg/dm ³	22.8	23.0

1.2. Membrane filtration

The membrane processes (UF and NF) were carried out using laboratory scale set-up (Fig. 1) with detailed description given in the previous work [32]. The ultra- and nanofiltrations were performed under transmembrane pressure: 6 and 14 bar, respectively. The retentate flow rate was 0.23 m³/h for both processes. The values of transmembrane pressures were determined based on the previous studies on the dairy products and

dairy wastewater [33–35], and the retentate flow rate depended on the construction of the laboratory scale membrane set-up. During the membrane processes, the temperature in the feed tank was constant and equal to 25 \pm 1 $^{\circ}\text{C}$. The ultra- and nanofiltration were carried out in the batch mode. A permeate was collected in a separate tank and a retentate was recycled to the feed tank.

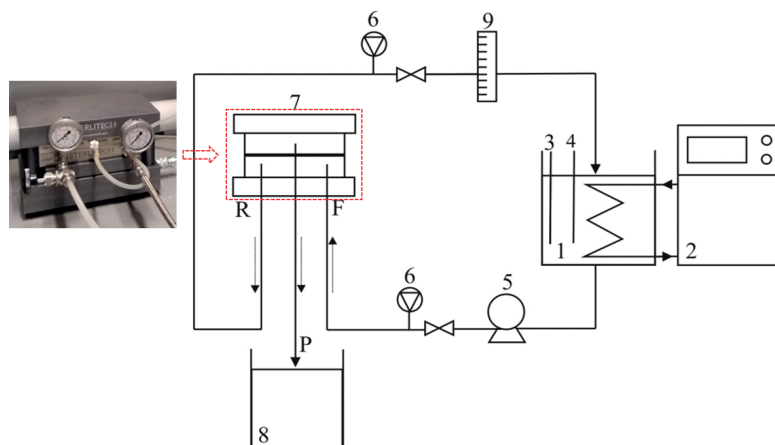


Fig. 1. Schematic diagram of the membrane system set-up: 1 – feed/retentate tank, 2 – thermostat, 3 – thermometer, 4 – pH-meter, 5 – pump, 6 – manometer, 7 – SEPA CF membrane cross-flow cell, 8 – permeate tank, 9 – flow-meter, P – permeate, F – feed, R – retentate

The flat sheet ultra- and nanofiltration membranes dedicated for protein-contaminated wastewaters were used in the experiments. The total active membrane

area was 0.0140 m². The characteristics of the membranes used in experiments were shown in the Table 2.

Table 2. Characteristics of the membranes used in experiments

Characteristics	Manufacturer's data	
	UF membrane	NF membrane
Manufacturer	Synder	GE Osmonics
Material	PVDF*	PPZ/PSU**
Cut-off, g/mol	50 000	150-300
pH of feed	3-10	2-11
Process temperature, °C	<60	<50

*PVDF – POLYVINYLIDENE FLUORIDE, **PPZ – POLY(PIPERAZINE-AMIDE), PSU – POLYSULFONE

1.3. Calculated parameters

The efficiency of the membrane processes of dairy wastewater were determined with permeate flux (J_p , $\text{dm}^3\text{m}^{-2}\text{h}^{-1}$):

$$J_p = V_p / (A \cdot t), \quad (1)$$

where

V_p – permeate volume, dm^3 ;

A – membrane area, m^2 ;

t – time needed to collect a defined volume of permeate, h.

The effectiveness of dairy wastewater treatment, using membrane filtration and biological methods, were evaluated based on weight-to-volume percentage reduction in solution contamination (R , % w/v):

$$R = (1 - C_1/C_2) \cdot 100, \quad (2)$$

where

C_1 – the concentration of the component in the solution after the treatment, mg/dm^3 ;

C_2 – the concentration of the component in the solution before the treatment, mg/dm^3 .

In the membrane terminology, R is called the retention.

1.4. Bacterial culture

The UF retentate was utilized using biological processes via an inoculum of bioaugmented bacteria. The experiments were conducted in a laboratory bioreactor with an active capacity of 1 dm^3 which were equipped with magnetic stirring elements and aeration system. The bacterial activators which were used (SK-BIOACTIV-05) in the study were comprised of the specialized, highly efficient aerobic, anaerobic, and facultative anaerobic microorganisms capable of degrading pollutants in dairy wastewaters. In addition to bioaugmented bacteria, the bio-product also contains specific micro- and macroelements (trace metals, vitamins, amino acids, and metabolic stimulants) necessary for their sustainable development. In order to maintain the environment suitable for the

microorganisms, the pH of the retentate was adjusted to 7.5 with 1M hydrochloric acid. The biological treatment of the UF retentate was carried out in a 24-hour bioreactor cycle at 20°C . The process duration depended on the dynamics of the stabilization of the organic and nutrients content and was 11 days.

1.5. Microalgal culture

The UF retentate and the same after the treatment with bacteria were used as culture media for growth of microalgae biomass. The selection of algae species for the experiments was mainly based on the ability of the species to adapt to the specific environmental conditions, resistance to the contaminants, and rapid cell growth. The most commonly found (in both freshwater and the marine environment) species of algae from the green algae group is *Chlorella vulgaris*.

The laboratory culture was inoculated with *Chlorella sp.* (inoculum volume was 20 cm^3), which was derived from our own culture, initiated by the inoculum obtained from Culture Collection of Baltic Algae (Institute of Oceanography of the University of Gdańsk). The experiments were performed in the Erlenmeyer flasks with a capacity of 1 dm^3 . The volume of each suspension was 700 cm^3 . The cellulose plugs provided free gas exchange between the culture medium and the environment. The source of the artificial light with the selected wavelength (characteristic for chlorophyll) was 30 W LED lamp (*Neonica Growy LED 118*) placed in a horizontal position within 20 cm of the flasks. The lighting conditions of the culture were changed periodically – photoperiod of 16h/8h (light/dark). The culture was conducted at room temperature ($25 \pm 1^\circ\text{C}$) with a forced stirring (160 rpm) using a *Heidolph Unimax 2010* orbital shaker. The appropriate circulation of the medium facilitated the maintenance of algal cells in a suspension and their flows to the best lit areas, the transport of CO_2 , as well as the distribution of nutrients.

The experiment was continued until the total phosphorus content in the culture medium decreased below $1 \text{ mg}/\text{dm}^3$. The UF retentate required 25 days, while UF retentate after biological treatment using bacteria needed 16 days.

1.6. Physico-chemical analysis

The total suspended solids (TSS) in water solutions (25-cm³ sample volume) was determined by a *HACH DR6000* UV-VIS Spectrophotometer (accuracy +/-1 nm, resolution +/-0.1 nm, optical path 1"). Furthermore, chemical oxygen demand (COD), total nitrogen bound (TNb), total phosphorus (TP), volatile fatty acids (VFAs), total organic carbon (TOC) and sulphates, using appropriate LCK cuvette tests, were identified in the samples. The instrument automatically averages 10 measurements for each sample and eliminates outliers. The samples of retentates after biological treatment (with both bacterial and algal culture) were taken every few days, and before the physico-chemical analysis, they were filtered with a paper filter (0.17 mm, 75 g/mm²), since a suspended biomass may be the reason of measurement errors.

Measurements of pH and conductivity were carried out using a *Mettler Toledo Seven Multi* pH/Conductivity Meter. The concentrations of calcium ions were determined by means of *Mettler Toledo* ion selective electrodes. The turbidity measurements were performed using a *HACH 2100Q is* Portable Turbidimeter.

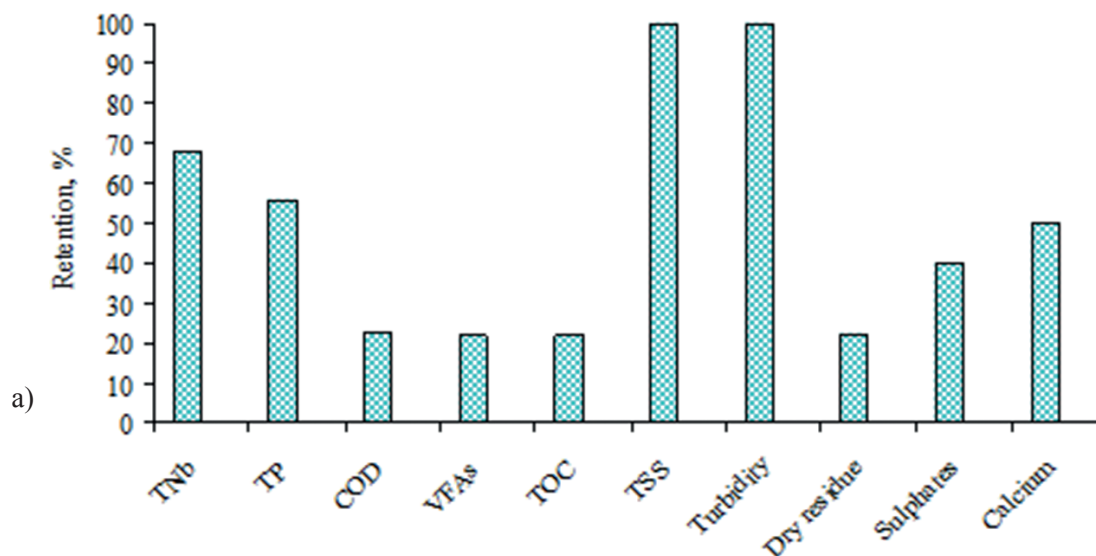
The dry residue content was determined by weight method. The samples (~5 cm³) were placed on aluminium disposable weighing pans inside a Radwag MAC 50/1 Moisture Analyser. During analysis, a standard drying profile was used (no change in weight of 0.001 g over 60 s at 105°C).

2. Results and discussion

2.1. Membrane filtration performance

In the first step of the experiments, the possibility of using UF for the treatment of dairy wastewater was

investigated (Fig. 2a). During the UF process, a permeate flux was reduced by 13% compared to the initial value of permeate flux 25 dm³/(m²·h). It was found that the UF membrane used was able to completely remove total solid suspensions in the dairy wastewater, and it also contributed to a significant reduction in the concentration of nutrients such as total nitrogen (68%) and total phosphorus (56%) (Fig. 2a). It was also found that the only 22% of organic compounds were retained in the UF (Fig. 2a). This suggests that the predominant organic substances in the wastewater constituted lactose, organic acids, and peptides, which, like mineral salts, penetrate the pores of the UF membrane. The retention of calcium ions at 50% (Fig. 2a) was likely due to the presence of casein micelles containing calcium phosphates. Nevertheless, the treatment of dairy wastewater in the UF was not efficient. Since the dairy wastewater primarily had contained organic compounds of low molecular weight, it was necessary to use a membrane with a much lower cut-off limit. In order to remove low molecular weight organic compounds from the dairy wastewater, the NF was used, whose separation capabilities have been widely described in the literature [33–36]. However, during the NF of dairy wastewater, a 60% decrease in permeate flux was observed compared to the initial value of 67 dm³/(m²·h). It was caused by the deposition of dairy components on the surface of the NF membrane. The dominant components of the filter cake on polymer membranes used for whey and milk filtration are proteins, but lactose and minerals have also been identified [37]. The same composition of the filter cake on the NF polymeric membranes during dairy wastewater treatment was most likely. For economic reasons, direct treatment of dairy wastewater in the NF is not highly recommended. Therefore, the dairy wastewater was subjected to treatment in an integrated system consisting of UF and NF membranes.



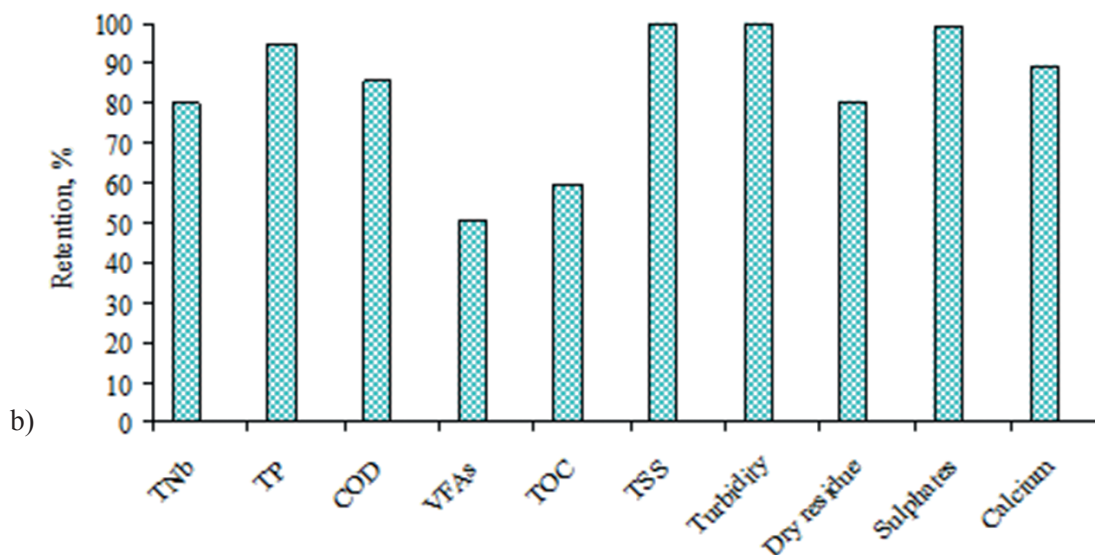


Fig. 2. Retention of individual components of dairy wastewater in the process of: (a) UF and (b) UF/NF

The ultrafiltration made it possible to remove the larger molecular organic compounds, which would reduce the efficiency of the process by adsorption on the surface of the NF membrane. In the case of the nanofiltration of dairy wastewater pre-treated via ultrafiltration, only a 4% decrease in permeate flux was noted compared to the initial value $117 \text{ dm}^3/(\text{m}^2 \cdot \text{h})$. It was found that the system consisting of UF and NF allowed a very high degree of the removal of organic matter from the dairy wastewater (Fig. 2b). The NF membrane also allowed a significant depletion of the sulphate and calcium ions concentrations (Fig. 2b).

Based on the obtained results, it has been found that the pre-filtration through $5 \mu\text{m}$ pores filter bag followed by integrated system of UF/NF is a rational way to treat dairy wastewater. These key steps enable the efficient regeneration of water from dairy wastewater. However, the recovery of water from dairy wastewater through membrane filtration is related to the waste stream in the form of a retentate, which is a concentrated mixture of pollutants retained by a membrane. In the system of UF/NF proposed for dairy wastewater treatment, there was a retentate generated in UF process, which was characterized by high molecular weight organic compounds and nutrients. The obtained results were compared to the acceptable parameters, which, according to the current regulations (in Poland and in the EU), should be met by wastewater discharged into water or soil. Significant exceeding of these parameters necessitates the need to develop methods of effective utilization of the retentate. The organic and inorganic compounds present in wastewater should not be discharged into water and soil without appropriate pre-treatment. Otherwise these might contribute to undesirable interferences in the aquatic ecosystems of living organisms as well as deterioration of water quality [33]. Therefore, wastewater with high nutrient

content is usually directed towards further technological processes (including biotechnological processes) aimed at achieving the levels of contaminants which are safe for the environment.

In the next step of the experiment, the possibility of utilization of the UF retentate was investigated using bacteria as well as algae for which wastewater could be a source of valuable nutrients.

2.2. Efficiency of bacterial treatment

The retentate which was a concentrate of pollutants retained by the UF membrane was subjected to bioaugmentation process with the aid of a bioactivator consisting of bacterial cells. The effectiveness of the method applied was evaluated based on the degree of removal of individual pollutants. It has been found that the bacterial process allowed over 90% removal of organic matter (COD) from the UF retentate (Table 3). Similar COD removal efficiencies have been obtained in another studies on dairy wastewater treatment using bacteria [12, 38]. The effectiveness of biological treatment method was lower for nutrients (nitrogen and phosphorus compounds) (Fig. 3). The content of TNb and TP was reduced by 63% and 50%, respectively.

Denitrification is a process of the reduction of inorganic nitrogen forms such as nitrates. This process requires the presence of readily biodegradable carbon compounds as electron donors for denitrifying bacteria and anoxic conditions [39]. Its efficiency is directly dependent on the ratio of COD to TNb [38, 40]. The effectiveness of nitrogen removal using denitrification bacteria increases with an increasing COD/TNb ratio and a minimum COD/TNb ratio of 10 is required for total nitrogen reduction in dairy wastewater [41]. Denitrifying bacteria belong to the group of facultative anaerobes. In the UF retentate,

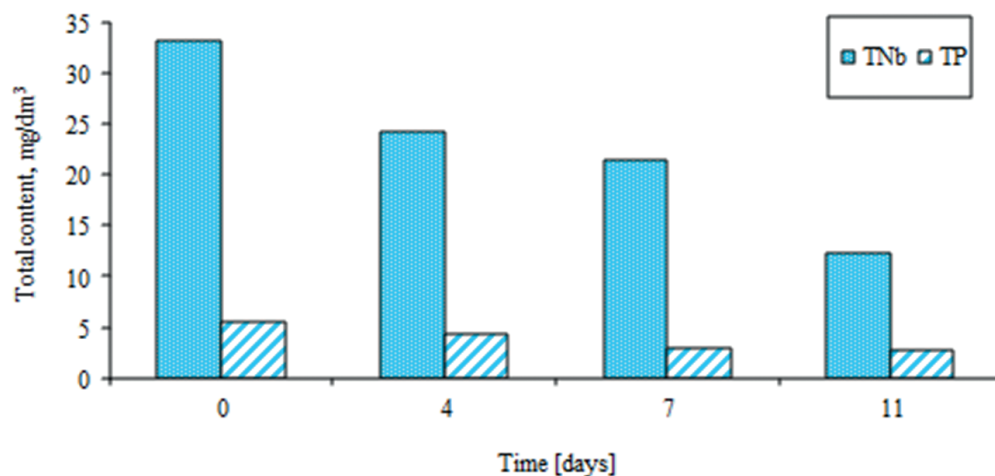
the initial COD/TNb ratio was ~35, and the total nitrogen was reduced by only 63%. The reason for relatively low reduction of total nitrogen at high COD/TNb ratio might be the poor or lack of the process of ammonification performed by the microbes,

which conduct mineralization to utilize the organic molecules of nitrogen as electron donors, acquiring energy and producing ammonium under both aerobic and anaerobic conditions. Thus, the subsequent nitrification and denitrification processes are limited.

Table 3. Physico-chemical properties of the UF retentate treated with bacteria

Parameter	Value
Total nitrogen, mg/dm ³	12.2
Total phosphorus, mg/dm ³	2.67
Chemical oxygen demand, mg/dm ³	74.6
pH	8.7
Total suspended solids, mg/dm ³	12
Turbidity, NTU	6.5
Conductivity, μ S/cm	1 574
Dry residue, mg/dm ³	1 060

Fig. 3. Changes in the total nitrogen (TNb) and phosphorus (TP) content in the UF retentate during bacterial treatment



The efficiency of phosphorus removal by the microbes is also the COD/TP ratio-dependent. The best

efficiency of biological dephosphorylation is observed at COD/TP >50 [42]. The ratio of organic matter to the TP in the retentate after UF was 213, which seems to be favourable to ensure the required depletion of phosphorus from wastewater. On the other hand, not only is the COD/TP ratio important for the reduction of total phosphorus, but parameters such as pH and the oxygen content are important [42]. There are also publications with data [43] indicating difficulties with the simultaneous removal of nitrogen and phosphorus in bioreactors. This is conditioned by the oxygen content in the bioreactor. The biological dephosphorylation process requires alternating anaerobic-aerobic and anaerobic-anoxic conditions to select and develop specific microorganisms that exhibit the ability to store greater amount of phosphorus within the cells than they require in normal physiological demands [44]. Most of the

plants removing phosphorus thus far utilize chemical precipitation using aluminium and lime [42]. In dairy wastewater, for this purpose, lanthanum-modified bentonites have been already tested [45]. Another types of bioreactor or modification of their operating times might also be helpful, taking into account both aerobic and anaerobic phases.

The physico-chemical parameters of the UF retentate treated by bacteria (Table 3) were compared to the values to be met by wastewater discharged into water or soil. It was found that bacterial treatment allowed the effective removal of pollutants. Most of the parameters are within the acceptable limits in Poland and other EU countries. However, the limit for the TP content, according to the EU Directive, was slightly exceeded. It is possible that the treatment using algae might be a suitable method for its complete removal.

2.3. Efficiency of the microalgae treatment

Both the UF retentate and the same after subsequent treatment with bacteria were proposed to be used as the media for microalgae cultivation. The control of the process primarily consisted in the study of changes in the total nutrients (nitrogen and phosphorus) content in culture media, which resulted from the metabolism of algae. Based on the studies, it was also possible to determine

the efficiency of microalgae biomass production. The cultivation was continued until the phosphorus content in the culture medium drastically decreased, since its deficiency is a limiting factor for the growth of algae [46].

Figure 4 shows the changes in the total nitrogen and phosphorus content in the culture medium during the experiments. The algal cultivation based on the UF retentate was inoculated with *Chlorella sp.* in an aqueous solution of BG-11 medium [47].

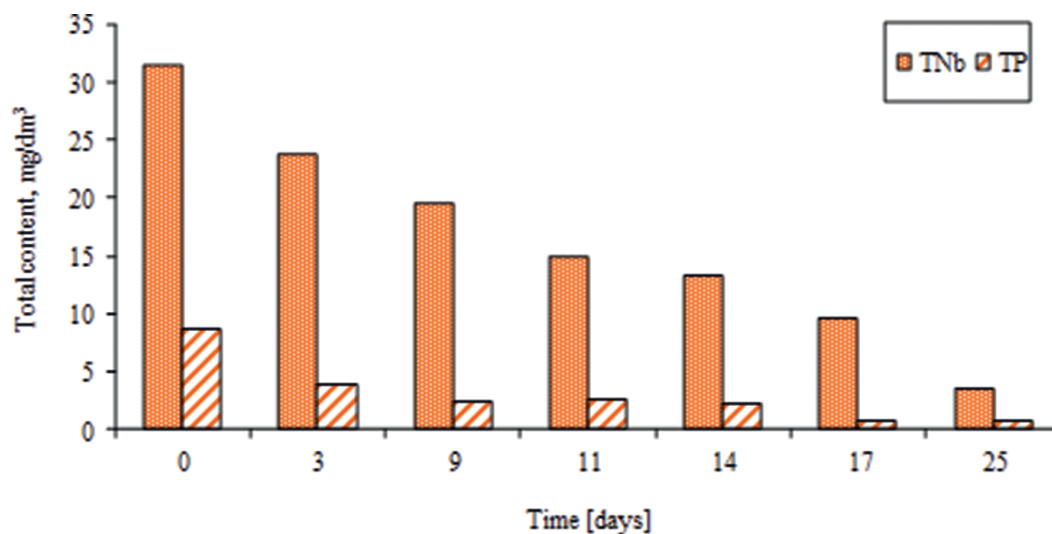


Fig. 4. Changes in the total nitrogen (TNb) and phosphorus (TP) content in the UF retentate used as a medium for the cultivation of microalgae

The total phosphorus content in the culture medium decreased by more than half within the first 3 days, and the total nitrogen decreased by more than half within 11 days. The initial intensive reduction in the phosphorus concentration (about 1.6 mg/dm³ per day), despite the small populations of growing algae, was probably due to over assimilation of the elements. It is believed that the phosphorus is stored in algae cells and used when its content in the medium becomes insufficient [48]. This also demonstrates that the adaptation of algae to the new environmental conditions was very rapid. An average daily assimilation of nutrients from the retentate within 25 days of cultivation was equal to 1.12 mg/dm³ and 0.32 mg/dm³ for nitrogen and phosphorus, respectively. From the beginning of the experiment until its completion, the total phosphorus content was reduced by ~92% (from 8.71 to 0.73 mg/dm³) and nitrogen by ~89% (from 31.5 to 3.45 mg/dm³). The final values obtained indicate that the removal of phosphorus and nitrogen from wastewater, using microalgae *Chlorella sp.*, was very efficient. The content of TP and TNb in the obtained solution after separation of algae biomass was lower than in the UF permeate (3.84 mg/dm³ and 13.0 mg/dm³, respectively) and in the UF/NF permeate (0.47 mg/dm³ and 8.1 mg/dm³, respectively).

Furthermore, it was also found that the final phosphorus and nitrogen content in the culture medium of algae was also lower compared to the retentate after bacterial treatment (TP – 2.7 mg/dm³, TNb – 12.2 mg/dm³). Therefore, an attempt to inoculate *Chlorella sp.* in this sample, which still contained nutrients, was made. The retentate treated with the bacteria did not exceed the standards for wastewater discharge into the environment. However, further declines in nitrogen and phosphorus content could result in the possibility of water reuse in an industrial plant, for example, as technical water for boiler or cooling systems. Fig. 5 shows the results of the experiment which took 16 days. Introducing the inoculum to the liquid after bacterial treatment resulted in initial increases in nitrogen (from 12.2 to 12.6 mg/dm³) and phosphorus content (from 2.7 to 3.4 mg/dm³) as well as pH (from 8.7 to 9.1). The nutrients remaining in the solution after bacterial treatment turned out to be the excellent sources for algae growth. As a result of the study, it was found that the microalgae absorbed almost all of the nitrogen and phosphorus from the medium, i.e. ~93% of TNb (final value 0.872 mg/dm³) and ~99% m/m of TP (final value 0.037 mg/dm³).

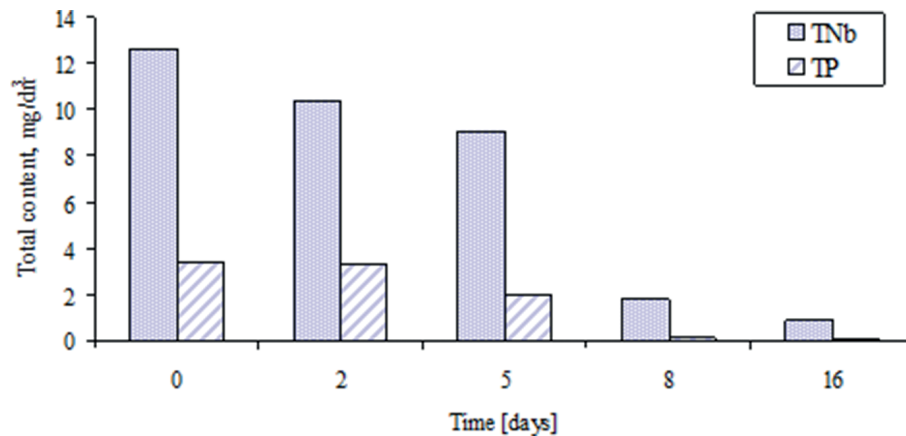


Fig. 5. Changes in the total nitrogen (TNb) and phosphorus (TP) content in the UF retentate, after bacterial treatment used as a medium for the cultivation of microalgae

Owing to the combination of two different biological processes (bioaugmentation with microalgal biomass production), the efficiency of the removal of nitrogen and phosphorus compounds from the retentate after ultrafiltration of dairy wastewater has considerably increased. Within several days of the experiment, after separation of algal biomass, the purified water met the environmental requirements for the discharge of wastewater into waters or soil within the content of these substances. Biomass, after dehydration and essential technological processing, can be a valuable raw material with high energy potential [49]. In the literature there are also examples of the use of microalgae capable of accumulating large quantities of lipids, among others, to remove specific impurities from wastewaters (e.g., antibiotics) during biomass production [50].

Conclusions

The obtained results have shown that an effective treatment of dairy wastewater can be successfully achieved using an integrated UF/NF system preceded by a pre-filter bag. However, the UF retentate is characterized by a high organic loading as well as high levels of nitrogen and phosphorus, which can cause environmental problems. The methods proposed for biological treatment, using bacterial and algal culture, effectively solved this problem. As a result of coupling these two methods, the concentration of TNb in purified water decreased $<1.0 \text{ mg/dm}^3$ and TP $<0.1 \text{ mg/dm}^3$. In addition, raw material (algae biomass) was obtained.

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