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The role of UV-irradiation pretreatment on the degradation of 2,4-dichlorophenoxyacetic acid in water

O. Tchaikovskaya[a]*, I. Sokolova[a], G.V. Mayer[a], E. Karetnikova[b], E. Lipatnikova[a], S. Kuzmina[a] and D. Volostnov[c]

ABSTRACT: The degradation of 2,4-dichlorophenoxyacetic acid (2,4-D) in water by the combination process of UV-irradiation, humic acids and activated sludge treatment has been studied. The photoreaction rate of all irradiated samples was lowest for the sample irradiated at 308 nm (the XeCl excilamp) in the absence and in the presence of humic acids, and highest for the sample irradiated at 222 nm (the KrCl excilamp). Photolysis of 2,4-D has been shown to enhance the subsequent microbial degradation. Copyright © 2010 John Wiley & Sons, Ltd.

Keywords: UV-irradiation; 2,4-dichlorophenoxyacetic acid; activated sludge; photobiodegradation; humic acids

Introduction

The development and use of herbicides have played an important role in the increase of agricultural productivity. On a world level, herbicide production accounts for 46% of the total pesticide production. Among the herbicides, acids are the most used in the USA and Europe. Acidic herbicides are widely used for the control of broad-leaved weeds and other vegetation. They are relatively inexpensive and very potent even at low concentrations. The majority of herbicides are directly applied to soil or sprayed over crop fields, and as consequence of large production and high stability, they are released directly to the environment (1). The presence of the chlorine groups causes such compounds to be more resistant to biodegradation than the unsubstituted analogs. At present, a large number of studies concern a promising new technology for treatment of polluted water (2).

Wastewater treatment generally consists of a primary, secondary and sometimes an advanced treatment stage, with different biological, physical and chemical processes for each stage of treatment. Activated sludge treatment includes two major steps: degradation of the pollution in the aeration basin followed by the separation of sludge and treated water by setting in a clarifier (3).

Humic acids (HAs) are omnipresent in the natural environment and play a number of important roles, including controlling the pH balance, governing the mobility of contaminants through absorption, aggregation and sedimentation, and chelating metals. The photochemical reactivity of HAs in the environment has been investigated previously (4–8), and numerous articles have suggested that HAs act as sensitizers or precursors for the production of reactive intermediates, including $^1O_2$, superoxide anion and/or hydrogen peroxide in oxygenated natural water. Humic acids encompass very diverse structures containing chromophores that act as photosensitizers upon electronic excitation, either directly or from the excited states (8). HAs are able to degrade organic chemicals in water.

In this study the effect of UV-irradiation on 2,4-dichlorophenoxyacetic acid (2,4-D) utilization with the activated sludge in the presence and absence of HAs is reported. Also, the toxicity of 2,4-D solutions was assessed using the bioluminescence assay, which is based on lyophilized luminous bacteria *Photobacterium phosphoreum*.

Experimental

The stock solution of 2,4-D was prepared by dissolving accurately weighed amounts of analytical-grade 2,4-D (chemical purity 98%, purchased from Aldrich Chemical Co.) in distilled water with an ultrasonic stirrer. Then the stock solution was diluted. For the studies with the activated sludge, 100 mL of 2,4-D solution ($C = 1 \times 10^{-4}$ to 1×10^{-3} M) was pre-irradiated.

Sample of the wastewater sludge was collected in the aeration basin from a treatment plant about 5 km from Tomsk Town (Siberia, Russia), containing wastewater and activated sludge in the ratio 2:1. The sample sludge was collected in a dry autumn period. Cultivation was conducted using 250 mL conical flasks

* Correspondence to: O. Tchaikovskaya, Laboratory of Photophysics and Photochemistry of Molecules, Department of Physics, Tomsk State University, 36, Lenin Ave, 634050, Tomsk, Russia. E-mail: tchon@phys.tsu.ru

[a] Tomsk State University, 36, Lenine Ave, Tomsk, 634050, Russia

[b] Institute of Water and Ecological Problems of the Russian Academy of Sciences, 65, Kim-Yu-Chen St., Khabarovsk, 680000, Russia

[c] Nature Committee of State Department, 14, Kirov Ave, Tomsk, 634050, Russia

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with 50 mL of the solution of 2,4-D and 50 mL of wastewater sludge, at 24–26°C under stationary conditions. For absorbance spectra recording, the particles of sludge were separated from the culture liquid by filtration through membrane filters (0.2 µm pore size; Vladipor, Russia). For non-irradiation or irradiation batch experiments, 2,4-D solution was added to mineral salt medium of the following composition (grams per liter of distilled water): KNO_3, 2; $MgCO_4$, 0.4; NaCl, 2; K_2HPO_4, 2 (in the ratio 1:1). The salt medium and distilled water for all microbiological experiments was autoclaved at 121°C. 2,4-D solution with activated sludge was incubated at 22 ± 2°C for 20 h to 28 days.

HA fractions were obtained from Fluka Chemical Co. The solutions of HAs were prepared in according to the literature procedure (7). The desired amount of HAs was dissolved in 0.1 M NaOH aqueous solution. The solution was sonicated for 15 min at 40°C in an ultrasonic bath. This procedure was repeated after 24 h. The effective concentration of the dissolved HAs in 0.001 M NaOH aqueous solution remained below 10^{-3} g l^{-1}.

The UV-radiation sources used for photochemical investigations were: (1) a DRT-240 high-pressure mercury lamp (Hg) and three barrier discharge excilamps [purchased from the Institute of High Current Electronics of the Siberian Branch, Russian Academy of Sciences (9)]. These were KrCl, XeBr and XeCl excilamps emitting maximum UV-radiation at 222, 283 and 308 nm, respectively. The parameters and choice of the lamps are discussed elsewhere (9). The exposure time was varied from 1 to 60 min at room temperature (23–25°C) under static conditions. Pre- and post-irradiation electronic absorption and fluorescence spectra were recorded by a conventional procedure using UV–Vis Unicam spectrometry and a Cary Eclipse spectrofluorimetry, at 25 ± 1°C in air equilibrated solutions. Fluorescence excitation wavelengths were 330 and 360 nm; 330 nm is the fluorescence excitation wavelength of emission of the assumed photolysis product of 2,4-D transformation and 360 nm is the fluorescence excitation wavelength of emission of HAs (10).

To determine the concentration of 2,4-D after UV-irradiation, the samples were acidified with HCl to pH = 1 and extracted by diethyl ether. The extracts were evaporated in the air flow to a volume of 0.5 ml. The chromato-mass-spectrometric analysis of the samples was performed on a Finnigan Model Trace DSQ facility (Thermo Electron Chromatography and Mass Spectrometry Division, USA). Determination conditions were as follows: column, Trace TR-5MS; temperature, 100°C (5 min); heating rate 10°C min^{-1} to 180°C (5 min), then heating rate 100°C/min to 300°C (1 min); carrier gas, helium.

The concentration of carbon dioxide was determined on a Chromatron GCHF 18.3 chromatograph with a thermal conductivity detector. Determination conditions were as follows: column, 9 mm in length and 3 mm in diameter packed with Spherochrom impregnated with a dibutyrate triethylene glycol stationary phase; carrier gas, helium (3 dm^3/h); column temperature, 35°C; detector temperature, 50°C; evaporator temperature, 50°C; and sample volume, 1 mL.

The 2,4-D phototransformation rate in water and in the presence of HAs was determined by evolution of the initial bands of 2,4-D absorbance at 230, 256 and 285 nm vs irradiation time. Bioluminescence measurements of toxicity of non-irradiated 2,4-D solution were performed with an Angstrem chemiluminometer (design office Real, Novosibirsk, Russia) using the bioluminescence assay, which is based on lyophilized luminous bacteria *Photobacreium phosphoreum* and produced at the Institute of Biophysics (Krasnoyarsk, Russia) (11). The bioluminescence intensity in the control (I_0) was compared with the bioluminescence intensity recorded in the presence of 2,4-D (I). The toxicity of 2,4-D solutions was expressed as I/I_0. It is generally assumed that $I/I_0 \geq 1$ is not toxicity, $I/I_0 > 0.7$ is weak toxicity, $I/I_0 = 0.5$ is average toxicity, and $I/I_0 < 0.3$ is acute toxicity. The degree of detoxification of 2,4-D solutions with activated sludge treatment was characterized using the detoxification coefficient $K = I_{max}/I$, where I_{max} is the maximal bioluminescence intensity recorded in the presence of 2,4-D after UV-irradiation. All results of the bioluminescence test were corrected for the 'optic-filter effect'.

Results and discussion

Photodegradation of 2,4-D in pure water

The maximum of absorption of molecular 2,4-D is located at 283 nm (see Fig. 1). The photodegradation of 2,4-D in oxygen-saturated aqueous solution in the absence of HAs was carried out by UV-irradiation from different sources. There were changes in the shape of the spectrum with increasing UV-irradiation time. The experimental details are presented in Table 1. It was observed that irradiation resulted in a decrease in absorbance peaks of 2,4-D at 256 and 283 nm (see Table 1 for details, nos. 9–16). As can be seen, the lowest photoreaction rate appeared with the use of the XeCl excilamp (λ_{rad} = 308 nm). Long irradiation times were necessary to observe a transformation because 2,4-D weakly absorbs in this wavelength. The highest 2,4-D conversions were achieved using the KrCl excilamp. When low initial concentration ($C = 1 \times 10^{-4}$ M) of 2,4-D was used, the results obtained with both the XeBr and KrCl excilamps were similar. When high initial concentration ($C = 1 \times 10^{-3}$ M) of 2,4-D was used, the results obtained with both the Hg lamp and KrCl excilamp were similar. According to chromatography data, after irradiation of the XeBr and KrCl excilamps, the 2,4-D conversion (99%) was achieved. Based on the chromato-mass spectra data, we concluded that 2,4-dichlorophenol was the main photoproduct of 2,4-D (see Fig. 2). It is in good agreement with reported data (1). The total quinones concentration was also obtained by basic photometric analytic procedure using benzosulfonic acid standard (see Table 2). However, the structures of these products

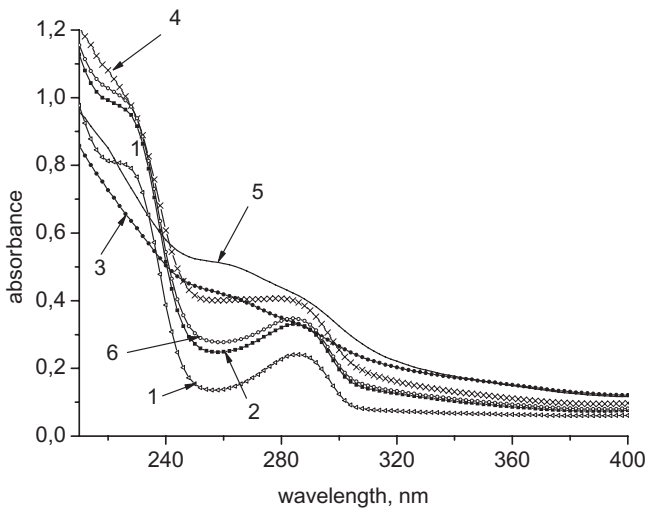

Figure 1. Absorbance spectra of 2,4-D solution in the absence (1) and in the presence of HAs (2) after UV-irradiation treatment for 32 min: 3 Œ by the Hg lamp; 4 Œ by the XeBr excilamp; 5 Œ by the KrCl excilamp; 6 Œ by the XeCl excilamp.

Table 1. Spectral-fluorescent characteristics of 2,4-D solution (10^{-4} M) and HAs after UV irradiation

No.	Solution	The action of UV irradiation		Absorbance, D/D_0			Fluorescence	
		Lamp, λ_{rad}	Time, min	At 230 nm	At 256 nm	At 285 nm	λ_{max} (nm)	I
0	HAs in 0.001 M NaOH	—	—	1	1	1	520	1.5
1	aqueous solution	Hg	15	1	1	1	480	1.4
2			30	0.9	0.9	1	480	1.1
3		KrCl	15	1	1	1.1	500	1.4
4			30	0.9	1	1	500	1.4
5		XeCl	15	1	1.1	1.2	520	1.4
6			30	1	1.1	1.1	520	1.2
7		XeBr	15	0.8	1	1.1	500	1.2
8			30	0.8	1	1.1	500	1.1
9	2,4-D in water	—	—	1	1	1	—	—
10		Hg	15	0.5	11	1	425	0.5
11			30	0.7	11.2	2	—	—
12		KrCl	15	0.5	15	2	450	0.8
13			30	0.8	2	1	—	—
14		XeCl	15	0.8	2.5	1	—	—
15			30	1	6	1.1	—	—
16		XeBr	15	1	10	1.2	450	0.5
17			30	0.5	11	1	425	0.5
18	2,4-D in water + HAs	—	—	1	1	1	510	1.8
19		Hg	15	0.7	2.2	1.1	510	1.6
							470	1.7
							425	0.6
20			30	0.6	1.8	0.9	470	1.81
							440	2
21		KrCl	15	0.8	2.3	1.3	425	0.6
22			30	0.6	2.3	1.2	450	0.9
23		XeCl	15	1	1.6	1	410	0.3
24			30	1	1.8	1	410	0.3
25		XeBr	15	1	1.2	1.1	425	0.8
26			30	0.9	1.4	1.2	425	1.4

The fluorescence excitation wavelength was 365 nm. D_0 is the intensity of absorbance of unirradiated solution, D is the intensity of absorbance irradiated solution and I is the intensity of fluorescent band (rel. un.).

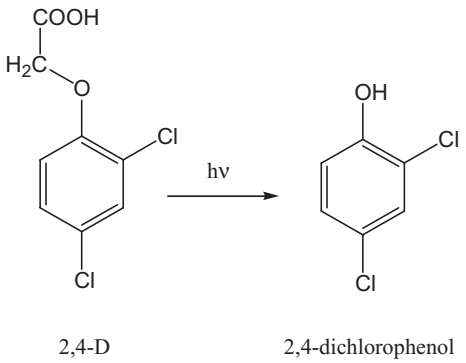

Figure 2. Scheme of phototransformation of 2,4-D in water.

were not identified. In order to estimate the efficiency of the physical treatment for the removal of 2,4-D, the fluorescence data after exposure to UV-irradiation were compared. According to fluorescence spectra, after UV-irradiation of 2,4-D solution the fluorescent photoproducts were formed (see Figs 3 and 4). 2,4-D in water did not have fluorescence. This may be because secondary photoproducts from the photolysis of 2,4-D were formed. Our results indicate (see Figs 3 and 4), that photoproducts did not differ in the pathway for photodegradation of 2,4-D upon irradiation by the KrCl and XeBr excilamps.

The experimental values of I/I_0 are collected in Table 3. The 2,4-D present in the solution reduced the intensity of bioluminescence of the test bacteria $I/I_0 = 0.66$ (see Table 3, no. 1); that is, the solution was toxic. The bioluminescent analysis of the toxicity of solutions containing 2,4-D showed that their toxicity was decreased during UV-irradiation by both the Hg lamp and XeCl excilamp ($I/I_0 \approx 1$), detoxification coefficient $K > 1$ (see Table 3, nos 4 and 11); that is, photodetoxication took place. The effect of detoxication was maximum upon irradiation with the XeCl excilamp for 15 min. The solutions of 2,4-D exposed to the KrCl excilamp radiation were possessed the greatest toxicity (see Table 3, no. 20).

Irradiation of 2,4-D in the presence of humic acids

In this chapter we also describe the dependence of the fluorescence intensity of HAs upon 2,4-D quencher concentration. The

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Stern–Volmer plot (12) is linear, which indicated that only one type of quenching occurs. From the slope of the Stern–Volmer plot, one can calculate that $K = 70$ м$^{-1}$. This is the value expected for the diffusion-controlled bimolecular rate constant between 2,4-D and HAs, which indicated efficient quenching by 2,4-D. Also this is the value expected for the diffusion-controlled bimolecular rate constant between macromolecule and quencher (12).

Table 2. The total quinones concentration in 2,4-D solutions exposed to UV irradiation from exciplex lamps

No.	t_{exc} (min)	UV irradiation	
		KrCl excilamp	XeBr excilamp
1	15	6.2×10^{-6} м	6×10^{-6} м
2	30	2×10^{-5} м	7.5×10^{-6} м
3	60	5×10^{-5} м	2.5×10^{-5} м

We observed weak changes in the HAs absorption spectra under UV-irradiation from mercury and exciplex lamps around 256 nm, while the absorption intensity around 285 nm was affected by the XeCl excilamp irradiation (see Table 1, nos 0–8). On exposure to UV-radiation the fluorescence bands were observed to appear one after another around 520 → 500 → 480 nm. The fluorescence intensity decreased in the fundamental band around 520 nm, as the exposure time was increased. It appears that some of the HA fragments were phototransformed too.

The wavelength at 256 nm is the absorbance region of assumptive phototransformation products of 2,4-D (see Fig. 1). According to the absorbance data (see Fig. 5), in the presence of HAs the phototransformation of 2,4-D was not effective in contrast to irradiative solution in the absence of HAs. When 2,4-D was irradiated with the Hg lamp in the presence of HAs, both direct photolysis and photoinduced reactions could occur and the photoinduced transformation was the main route. That is why

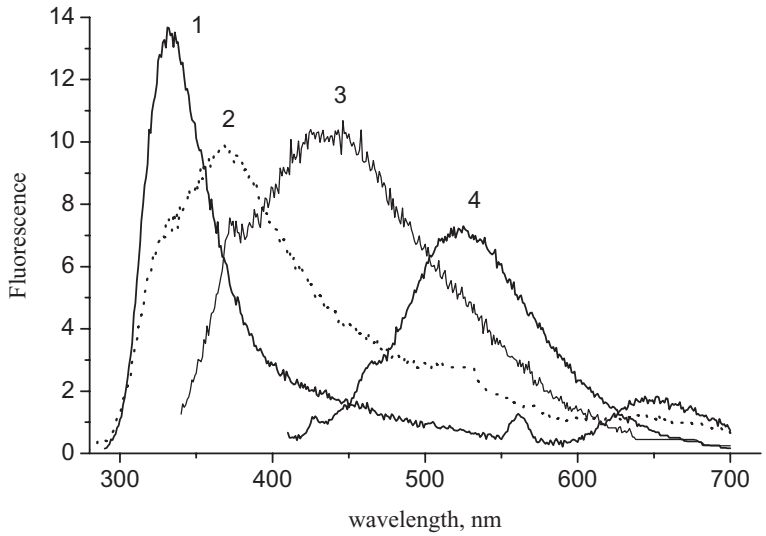

Figure 3. Fluorescence spectra of 2,4-D in water at a concentration 10^{-3} м after exposure to the XeBr excilamp radiation ($E = 1.7$ J cm^{-3}) for 60 min. The fluorescence excitation wavelengths are 280 nm (1), 260 nm (2), 330 nm (3) and 400 nm (4).

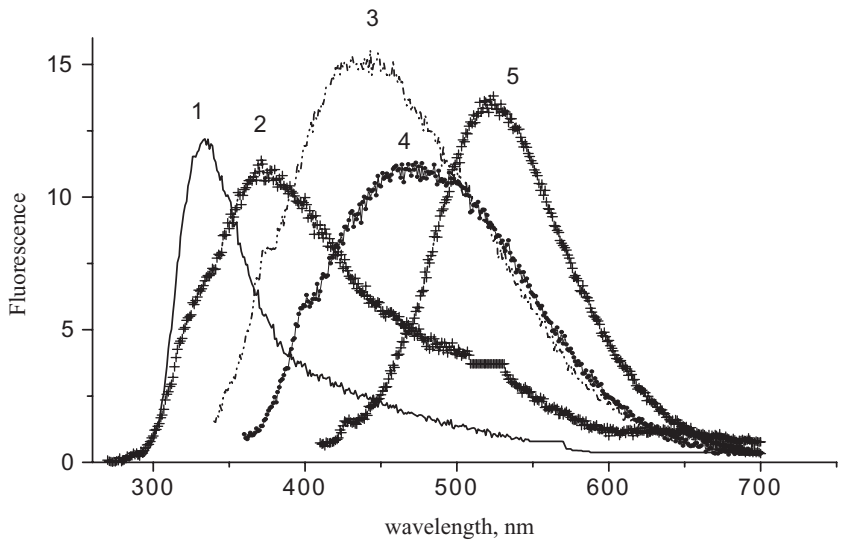

Figure 4. Fluorescence spectra of 2,4-D in water at a concentration of 10^{-3} м after exposure to the KrCl excilamp radiation ($E = 1.9$ J cm^{-3}) for 60 min. The fluorescence excitation wavelengths are 280 nm (1), 260 nm (2), 330 nm (3), 350 nm (4) and 400 nm (5).

Table 3. Bioluminescence analysis of the toxicity of 2,4-D solutions at a concentration of 10^{-4} M after UV irradiation and activated sludge treatment (AST)

No.	Sample	I/I_0
0	Control	1
1	in water	0.66
2	+ 15 min irradiation with the Hg lamp	0.81
3	+ 30 min irradiation with the Hg lamp	1.18
4	+ 60 min irradiation with the Hg lamp	0.95
5	+ 60 min irradiation with the Hg lamp + 68 h AST	1.22
6	+ 30 min irradiation with the Hg lamp + 20 h AST	1.25
7	+ 30 min irradiation with the Hg lamp + 116 h AST	1.05
8	+ 60 min irradiation with the Hg lamp + 116 h AST	1.08
9	+ 60 min irradiation with the Hg lamp + 20 h AST	1.23
10	+ 15 min irradiation with the Hg lamp + 14 days AST	1.07
11	+ 15 min irradiation with the XeCl excilamp	1. 03
12	+ 30 min irradiation with the XeCl excilamp	0.71
13	+ 60 min irradiation with the XeCl excilamp	0.79
14	+ 15 min irradiation with the XeCl excilamp + 20 h AST	1.25
15	+ 30 min irradiation with the XeCl excilamp + 20 h AST	1.25
16	+ 30 min irradiation with the XeCl excilamp + 116 h AST	1.29
17	+ 60 min irradiation with the XeCl excilamp + 116 h AST	1.18
18	+ 15 min irradiation with the KrCl excilamp	0.79
19	+ 30 min irradiation with the KrCl excilamp	0.62
20	+ 60 min irradiation with the KrCl excilamp	0.42
21	+ 30 min irradiation with the KrCl excilamp + 20 h AST	1.36
22	+ 30 min irradiation with the KrCl excilamp + 116 h AST	1.06
23	+ 60 min irradiation with the KrCl excilamp + 68 h AST	1.23
24	+ 60 min irradiation with the KrCl excilamp + 116 h AST	1.06

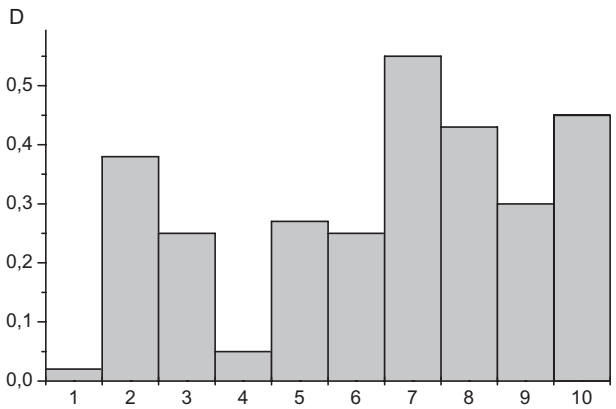

Figure 5. The comparison of the absorbance intensity at 256 nm of 2,4-D in water (1) without (1–5) and in the presence (6–10) of HAs with no preliminary UV-irradiation (1, 6) and pre-irradiation for 30 min: 2, 7 – by the KrCl lamp; 3, 8 – by the XeBr lamp; 4, 9 – by the XeCl lamp; 5, 10 – by the Hg lamp.

the degradation of 2,4-D solution within HAs after action UV-irradiation with the Hg lamp was accompanied by the formation of the new product fluorescing at 440 nm (see Table 1, nos 10 and 20).

When 2,4-D was irradiated in the presence of HAs the sets of fluorescent photoproducts were different (for example, see Table 1, nos 10 and 19) than in distilled water. In the presence of HAs the fluorescent products of 2,4-D at 410 and 440 nm were defined. HAs apparently catalyzed the formation some different fluorescent photoproducts of 2,4-D after UV-irradiation treatment by excilamps.

Synergetic degradation of 2,4-D by integrated photo- and activated sludge treatment

Biodegradation of 2,4-D was estimated according the respiration activity of microbial complex of activated sludge by CO_2 chromatograph analysis. In the presence of non-irradiated and pre-irradiated solutions of 2,4-D ($C = 10^{-4}$ M) respiration activity of sludge increased in comparison to control (see Fig. 6). This confirms that products of 2,4-D photolysis were not toxic for microorganisms, i.e. destructors. From using two resources of UV-radiation the respiration activity was higher with the sequential photo-biodegradation of 2,4-D using the XeBr excilamp.

Sequential photo-biodegradation allows reduction of the toxicity of pre-irradiated 2,4-D solutions, $I/I_0 > 1$, detoxification coefficient $K = 1.6–3.4$ (see Table 3, nos 6, 14 and 22). The mechanism of detoxification with activated sludge is very complex. First of all the biological treatment based on biochemical and partly on biophysical processes occurs (13). Then the complexity of HAs leads to a variety of physical–chemical behaviors and chemical interactions (14). In addition, upon exposuree to light HAs can promote pollutant degradation via the generation of many chemical transients (15). The presence of HAs in 2,4-D solution was not inhibited the respiration activity of microbial complexes of activated sludge.

According to chromatography data, the sequential action of UV-irradiation with the XeBr excilamp for 1 h and after 7 days of

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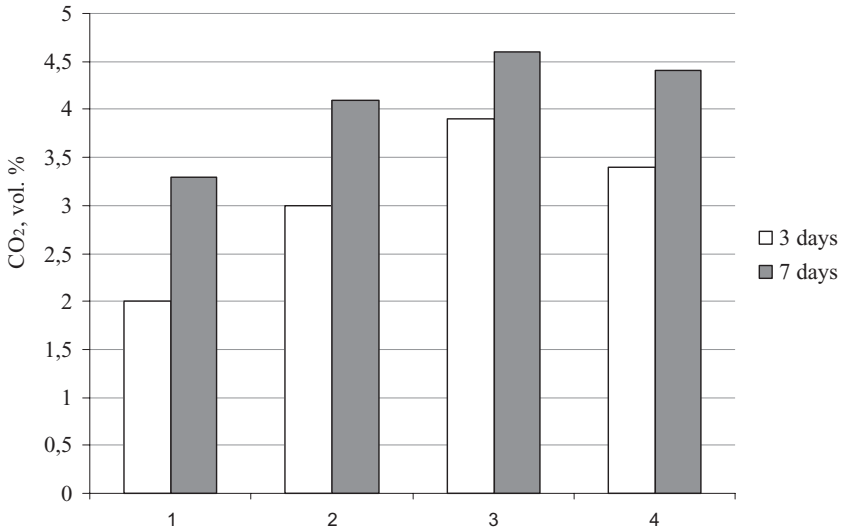

Figure 6. The comparison of the respiration activity of activated sludge microorganisms without (1) and in the presence of 2,4-D (2–4): 1, control; 2, 2,4-D; 3, 2,4-D irradiated with XeBr excilamp for 30 min; 4, 2,4-D irradiated with KrCl excilamp for 30 min.

activated sludge treatment is highly recommended for 2,4-D removal from water.

Conclusion

The results of our study imply that it is possible to optimize the configuration of biological wastewater treatment plants to improve the removal of some herbicides in wastewater before they are discharged into the environment. The wavelength of UV-irradiation influences the toxicity of 2,4-D solutions.

Acknowledgments

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