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AN EVALUATION OF THE EFFECTIVENESS OF NANOSILVER IN SWIMMING POOL WATER TREATMENT - WATER QUALITY AND TOXICITY OF THE PRODUCT

Abstract: The possibility of applying a colloidal solution of nanosilver in the closed circuit of pool water treatment as a complementary disinfectant with chlorine compounds was presented. The applied nanosilver solution is characterized, by hygienic certificate, as having a very high biocidal effect. Samples of pool water for the control were taken from 5 points of a pool circuit. The safety of the water was appraised by comparing the bacteriological and physicochemical test results with the admissible values specified by hygienic requirements. The results show that nanosilver solution can be successfully applied for pre-coating the filter bed and supporting the disinfection system. Special attention was paid to the bacteriological purity and stability of the disinfectant concentration. The influence of concentration of colloidal nanosilver (0-25 mg/dm³) on bacterial bioluminescence, crustacean mortality and macroscopic effect of root growth and seed germination of selected plants was analysed. The results obtained were related to the current knowledge on the impact of nanoparticles on indicator organisms. It was found that due to many still unknown mechanisms of interaction and transformation of nanoparticles in living organisms, further study of this issue is necessary.

Keywords: nanosilver, the quality of pool water, pool water treatment technology, toxicological assessment

Introduction

In recent years, there has been a rapid increase in the number of public swimming pools. Due to the high demand for water recreation activities and rehabilitation in water, swimming pool facilities must function correctly, and the basis for proper operation are safe and reliable swimming pool installations.

Strict requirements regarding swimming pool water [1-5] have rendered traditional and one-stage filtration systems insufficient [6-9]. Because of this, some existing facilities are being modernized and fitted with additional devices and processes to boost the efficiency of water treatment [10, 11].

Swimming pool water treatment systems that guarantee healthy water include prefiltration, surface coagulation run in various filter beds, ultrafiltration, disinfection with sodium hypochlorite or calcium hypochlorite aided by ozonation or UV irradiation, and pH correction [6-10].

Despite the use of proven pool water purification technologies, the disinfection process is constantly being improved. Traditionally, chlorine-based products are used to disinfect the pool water, but they are the cause of harmful DBP (disinfection by-products). For this

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reason, there is a need for alternative disinfectants or disinfection processes to reduce the dosage of chlorine (as a final disinfectant) to pool water [12-14].

Pool water treatment processes may also incorporate nanosilver products. Nanosilver is increasingly being used in consumer products from washing machines and refrigerators to other devices. Silver nanoparticles are known to be excellent antimicrobial agents, and therefore they can be used as alternative disinfectant agents for the disinfection of drinking water or recreational water [15-19].

Although silver (Ag_2SO_4) in a low concentration (0.01 mg/dm^3) kills more than 99.9 % of heterotrophic bacteria in pools, the time needed to achieve this effect (about 30 minutes) does not allow its use as the main disinfectant agent [16].

This paper presents research on the use of a colloidal solution of nanosilver in the technology of swimming pool water. The main objective of the research was the analysis of the quality of swimming pool water treated with a system that incorporates: colloidal solution of nanosilver aiding the final disinfection of water with sodium hypochlorite, a vacuum filter with multilayered bed, and a low-pressure UV lamp.

Due to the increasing use of silver nanoparticles (AgNP), their emission to the natural environment may also increase. There are many publications on the harmfulness of silver, in particular for organisms living in the aquatic environment [20-23]. Depending on the environmental conditions in which nanoparticles will be found and mechanisms of oxidation and destabilization of their surface-coating agents, they can survive the process of wastewater treatment and still show their activity in the environment. It has been shown that silver nanoparticles with sizes close to 50 nm can be present in the aquatic environment, even for many months [23]. As the mechanism of toxicity of nanoparticles is still not fully understood, it is necessary to monitor the impact of this type of technology on the living organisms [24].

Therefore, the authors of the work additionally presented an ecotoxicological assessment based on the reactions of selected indicator organisms in relation to the classification of toxicity [25, 26]. The influence of colloidal nanosilver (concentration ranging from 0 to 25 mg/dm^3) on bacterial bioluminescence *Aliivibrio fischeri*, crustacean mortality *Artemia salina* and macroscopic effect of root growth and seed germination of *Lemna minor* and *Sinapis alba* were analysed. The results obtained were referred to the current knowledge on the impact of nanoparticles on indicator organisms.

Materials and methods

In the tested swimming pool, there were physiotherapy sessions for patients suffering from various injuries during the morning hours. In the afternoon, there were swimming lessons for infants and their parents. The swimming pool is equipped with a vertical water flow system with a capacity of $30 \text{ m}^3/\text{h}$ and 4 stations with massage jets. The swimming pool dimensions ($3.2 \text{ m} \times 5.4 \text{ m} \times 1.2 \text{ m}$) allow for the comfortable rehabilitation of 6 to 8 persons. The swimming pool draws water from the municipal water supply system. Water deficits resulting from evaporation, splashing and the need to wash filter beds are replenished by drawing water into the retention tank. The basic requirement for the correct circulation of swimming pool water is a closed circulation system with an active overflow. The treated water is introduced to the pool through 14 jets located at the bottom of the pool basin. The water is drained through the top overflow troughs to the retention tank. Then, the water is sucked by the circulation pump. Before the pump, there is a mesh filter, whose aim

is to capture large solid contaminants. The pump moves the water to the filter where, after the application of a disinfectant and a pH correction solution, it is directed to the swimming pool through heat exchangers. Before the filters, a coagulant solution is applied (2.5-10 % solution of aluminum hydroxide chloride). To impregnate the filter, it was precoated with 11 dm³ colloidal solution of nanosilver (hygienic certificate) on the first day of research (Fig. 1a, b). The swimming pool water treatment system uses a multilayered vacuum filter with sand and hydroanthracite bed with an area of 1 m² and height of 1.2 m. The filtrate pipe was supplemented with a low-pressure UV lamp. The treatment system is an automatic system, controlled by an analyzer that monitors the quality indicator values of the water that is drained from the swimming pool basin.

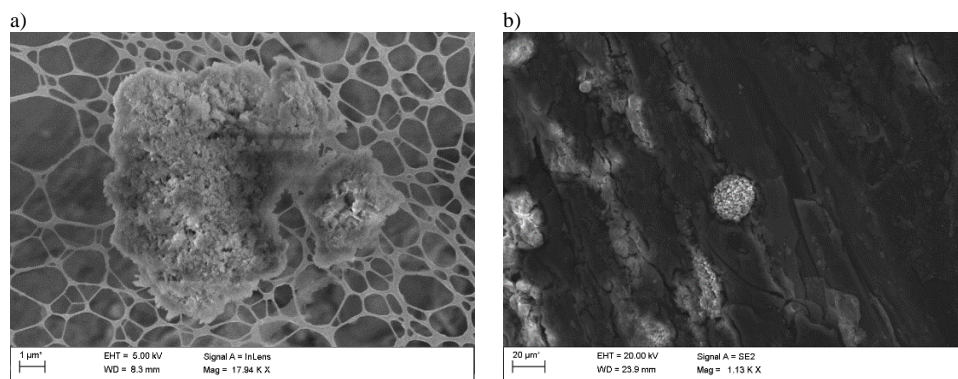


Fig. 1. Colloidal solution of nanosilver: a) as a stock solution; b) on a filter bed (Photo taken with the high-scanning electron microscope ZEISS SUPRA 35; EHT - electron high tension, WD - working distance, Signal A - signal selection register, Mag - magnification)

The samples for physicochemical and bacteriological tests were taken from five points in the pool water circuit: swimming pool basin (SP), retention tank (RT), before filter (BF), after filter and before UV lamp (AF) and water supply (WS).

The test results, collected over a 2-month period, were analysed. The water samples were subjected to bacteriological (once a week) and physicochemical (twice a week) analyses that measured basic control parameters of pool water quality [1-5].

The samples were collected and marked in accordance with applicable standards and methods [27, 28]. The physicochemical parameters that best described the water quality in the tested pool were: pH (colorimetric method-photometer DSC 400, Dinotec and potentiometric method-HQ11D Digital pH meter kit, Hach, Loveland, CO, USA), redox (potentiometric method-ORP electrode; HQ11D Digital, Hach), free and combined chlorine (DPD colorimetric method, photometer DSC 400, Dinotec and POCKET Colorimeter II, Hach), silver ion, oxidizability (*COD*), chlorides and nitrates (spectrophotometer DR5000 UV/VIS, Hach and Spectroquant[®] Pharo 300, Merck). Total organic carbon (*TOC*) and total dissolved organic carbon (*DOC*) (after membrane filter PVDF 0.45 µm) was determined using a *TOC* analyzer (TOC-LCSH/CPH, Shimadzu).

The bacteriological parameters that best described the water quality in the tested pool were: *Escherichia coli* (method compliant with: PN-EN ISO 9308-1:2004 [29]), total plate count in 36 °C after 48 hours (method compliant with: PN-EN ISO 622:2004

[30]), *Legionella sp.* (method compliant with: PN-ISO 11731-2:2008 [31]) and coagulase positive staphylococci (method compliant with: PN-EN ISO 6888-1:2001/A1:2004 [32]).

During the tests, the values for water pH, redox potential and the concentration of free chlorine and combined chlorine were read every day, directly from the screen of control and measurement device SCL DINOTEC. The obtained tests results were compared against the recommendations of DIN 19643 [1, 3], WHO [2], ZHK NIZP-PZH [4] and the permissible values specified in the decree [5] (Table 1).

Table 1

Recommendations and permissible values of swimming pool water quality

Parameter	WHO [2]	DIN 19643 [1, 3]	ZHK NIZP-PZH [4]	Dz.U. of 2015, item 2016 [5]
Total plate count in 36 °C after 48 h [CFU ¹ /1 cm ³]	<200	100	100	100
Coliform bacteria of the fecal type [CFU/100 cm ³]	<1	-	0	-
<i>Escherichia coli</i> [CFU/100 cm ³]	1	0	0	0
<i>Legionella sp.</i> [CFU/100 cm ³]	<1	0	0	0
<i>Pseudomonas aeruginosa</i> [CFU/100 cm ³]	<1	0	0	0
Coagulase-positive staphylococci [CFU/100 cm ³]	-	-	2	-
pH [-]	7.2-7.8	6.5-7.6	6.5-7.6	6.5-7.6
Redox [mV]	720	750	750	750
Free chlorine [mg Cl ₂ /dm ³]	0.5-1.2 ²	0.3-0.6	0.1-0.3	0.3-0.6
Combined chlorine [mg Cl ₂ /dm ³]	0.2	0.2	0.2	0.3
Silver [mg Ag/dm ³]	0.01 ³	-	-	-
COD [mg O ₂ /dm ³]	-	4.0 ⁴	5.0 ⁴	4.0 ³
Chlorides [mg Cl ⁻ /dm ³]	-	-	250	-
Nitrates [mg NO ₃ ⁻ /dm ³]	-	20	30	20

¹ CFU (colony forming unit); ² Depending on the method of water disinfection; ³ WHO [33]; ⁴ Difference in COD values in swimming pool water and COD in water supplementing circulation system

In extended studies, including ecotoxicological analysis, pool water samples were treated as a matrix for nanosilver solutions with selected concentrations. Before performing the bioassays, the basic physicochemical parameters of the water collected were examined (Table 2). Then, naturally (free evaporation during 120 hours) the concentration of free chlorine in water was reduced to 0.1 mg Cl₂/dm³. The presence of free chlorine may have a profound influence on the increased level of sample toxicity resulting from the destructive action of chlorine upon indicator organisms. Subsequently, colloidal nanosilver solutions with concentrations of 0.5; 5.0; 10.0; 15.0; 25.0 mg Ag/dm³ were made in pool water and a sample of raw pool water (0 mg Ag/dm³) was prepared in parallel. Ecotoxicological analyses of the prepared solutions were carried out 24 hours after their preparation.

Table 2

The values of physicochemical parameters of pool water taken for toxicological analysis

Parameter	Value
pH [-]	6.65
Redox [mV]	778
Free chlorine [mg Cl ₂ /dm ³]	0.84
Combined chlorine [mg Cl ₂ /dm ³]	0.54
TOC [mg C/dm ³]	3.411
DOC [mg C/dm ³]	3.051
COD [mg O ₂ /dm ³]	< 15
Chlorides [mg Cl ⁻ /dm ³]	282
Nitrates [mg NO ₃ ⁻ /dm ³]	2.80

The first toxicity test was performed following the *Screening Test* procedure of the MicrotoxOmni system in a Microtox analyzer, Model 500, Tigret [34], performing the function of both an incubator and a photometer. The percentage of bioluminescence inhibition relative to the control sample (bacteria not subjected to a potential toxicant) was measured after the exposure times of 5 and 15 minutes. Results after 5 and 15 min are reported (*E* - Effect [%]). The obtained results were assessed based on the toxicity classification system shown in Table 3 [25, 26]. The standard deviation of the Microtox[®] test ranges from 3.92-13.94 %.

Assessment of toxicity of pool water samples was also performed based on the mortality test (or lethality) of *Artemia salina* nauplii. The organisms were obtained from a test breed run according to own methodology and experiments presented in the literature [35, 36]. The samples of test solutions (2 cm³ each) were introduced into the test wells, followed by 10 *Artemia salina* nauplii. The number of dead or immobilized organisms was determined after 24 and 48 h of the test duration in microscopic observations. For each of the samples and an additional control sample of brine solution for nauplii cultivation, the toxicity effect *E* was calculated. The standard deviation of this mortality test was 0-4.51 %. The obtained results were compared with the results obtained in the Microtox[®] test and assigned to the appropriate toxicity class.

Table 3

Toxicity classification system

<i>E</i> [%]	Toxicity class
< 25	non-toxic
25-50	low toxicity
50.1-75	toxic
75.1-100	high toxicity

The evaluation of the macroscopic effect of nanoparticle solutions on root growth and germination of plants was based on the recommendations of the US EPA [37] for *Lemna minor* and according to the methodology of the Phytotoxkit[®] test [38] for *Sinapis alba*. Additional control samples were prepared for the test series: for *Lemna minor*, a solution in which the plants were grown and deionized water for *Sinapis alba*. Phytotoxicity assessment of nanosilver solutions with respect to *Lemna m.* was made on the basis of observations of stimulation or inhibition of the increase in the number of fronds by a 7-day test at 23 °C, with a 25 W lighting (224 lm). Relative growth rate *RGR* [1/d] was calculated as:

$$RGR = (\ln FN_t - \ln 9)/t \quad (1)$$

where FN_t - the frond number of *L. minor* on day t (7 days).

Then the E [%] was determined based on the value of the root inhibition index, IR_f :

$$IR_f = ((R_{fc} - R_{ft})/R_{fc}) \cdot 100 \quad (2)$$

where R_{fc} and R_{ft} - the rates of inhibition of the growth of fronds in the control sample and for nanosilver solutions samples, respectively.

The standard deviation of the *Lemna minor* growth inhibition test was 0-37.12 %. In *Sinapis alba* test, 4 cm³ of the samples were used. The acute toxicity phytotest took 3 days and was conducted in a laboratory incubator at a temperature of 25 °C. The macroscopic effect for *Sinapis alba* (E [%]) was evaluated based on I_{RG} root growth inhibition and I_{GS} seed germination inhibition. Calculated on the basis of I_{RG} .

$$I_{RG} = (K_c - K_s)/K_c \cdot 100 \quad (3)$$

where K_c and K_s - root lengths in control samples and solutions.

In contrast, inhibition of I_{GS} germination was calculated as:

$$I_{GS} = ((L_c - L_s)/L_c) \cdot 100 \quad (4)$$

where L_c and L_s - the number of seeds sown on the first and last days of the test.

The standard deviation of the *Sinapis alba* seed germination test was 1.36-14.56 %. The values of growth inhibition coefficients with positive values > 0 % were assumed as the signal of inhibition of plant growth, while the growth stimulation was indicated by negative values < 0 %. All biotests were performed three times, showing the value of the arithmetic mean and the standard deviation.

Results and discussion

The physicochemical and bacteriological tests of the samples of swimming pool water taken from 5 points in the pool water circuit allowed the determination that the quality of water met the requirements in this regard.

In all samples subjected to microbiological analysis, number of CFU (colony forming unit) of *Pseudomonas aeruginosa* and coagulase-positive staphylococci was found. CFU of *Escherichia coli* and *fecal coliforms* were found in water samples collected during the first day of the research, < 5 CFU of *E. coli* and < 5 CFU of *fecal coliforms*. CFU of *Legionella sp.* was detected three times in water supplementing the circuit (WS). Despite the presence of *Legionella sp.* in the water supplementing the circuit, the remaining samples - SP, RT, BF and AF - did not contain CFU of *Legionella sp.*, proving the high efficiency of the treatment and disinfection system. In all water samples after the filter and before the UV lamp (AF), the total plate count was also high, and significantly exceeded the recommended values for pool basin water, i.e., 20 CFU/1 cm³. Such a high number of CFU of bacteria in the filtrate proved that they were being washed out from the bed. Despite the use of nanosilver to precoat the filter bed, the conditions in the filter were favorable for bacterial growth. Nevertheless, CFU of bacteria at other points demonstrated the effectiveness of the disinfection system.

Figure 2 shows the average values of physicochemical parameters that supplement the bacteriological ones and co-determine if the pool is fit for use. Water pH at every collection point was within the required range, i.e., 6.5-7.6 (Fig. 2a).

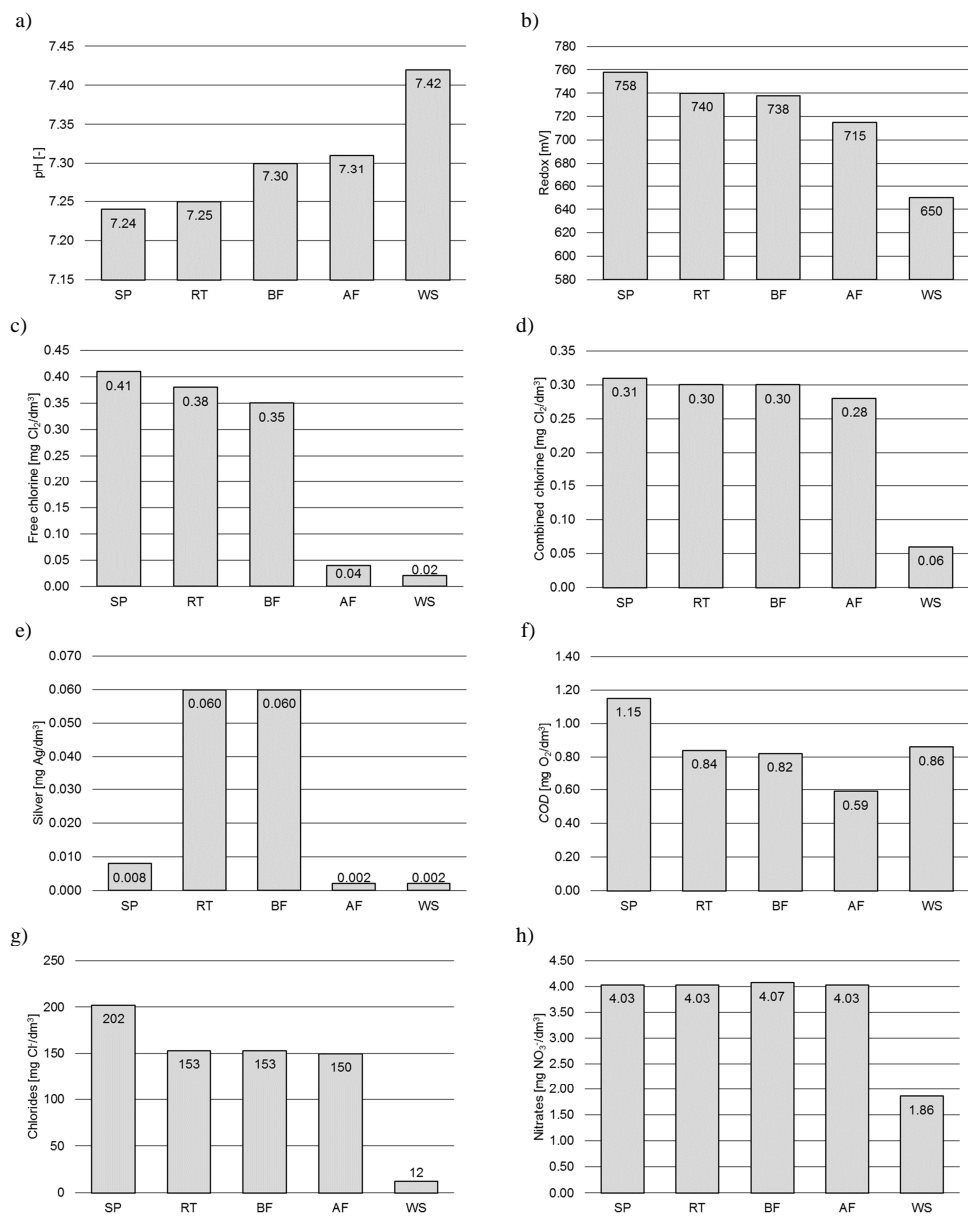


Fig. 2. Physical and chemical parameters of water quality in the tested pool circuit: a) pH, b) redox, c) free chlorine, d) combined chlorine, e) silver, f) *COD*, g) chlorides, h) nitrates in the swimming pool (SP), retention tank (RT), before filter (BF), after filter (AF) and in water from the supply system (WS)

Values of redox potential are especially important for pool basin water. The values obtained during *in situ* tests were within the range of 725-765 mV (on average: 758 mV), which proved that the bathers were sufficiently protected against contamination (Fig. 2b).

The concentrations of free chlorine in the pool water (Fig. 2c), due to the automatic dosage of NaOCl solution, were stable and within the range of 0.38-0.43 mg Cl₂/dm³ (on average: 0.41 mg Cl₂/dm³). A systematic decrease of free chlorine was observed in the subsequent parts of the pool circuit. Filtering the water through a filtration bed with an anthracite layer resulted in a decrease in the concentration of free chlorine, on average, by 88.68 %.

Due to the adverse effects on the bathers [39-42], the permissible content of combined chlorine in pool water according to [1-4] is 0.2 mg Cl₂/dm³, and according to [5] is 0.3 mg Cl₂/dm³. There were no complaints from the bathers when the concentrations of bounded chlorine in water samples taken from the pool basin exceeded the concentrations stipulated in DIN19643, on average, by 0.11 mg Cl₂/dm³ (Fig. 2d).

Because the circuit water was supplemented with 11 dm³ of colloidal solution of nanosilver, the presence of silver was determined as an additional parameter. It should maintain microbiological stability in the pool basin, and be neutral to the bathers. According to the recommendations of the WHO [33], the content of silver in drinking water should not exceed 0.01 mg/dm³. The concentrations of silver in the pool circuit water were, on average: 0.002-0.008 mg/dm³ (Fig. 2e).

During the tests, in the majority of samples, oxidizability index (*COD*) was below 1.0 mg O₂/dm³ (Fig. 2f) and the permissible value is 5 mg O₂/dm³. Oxidizability above the permissible value (9.46 mg O₂/dm³) was detected in only one water sample from SP. In the same water sample, the value of total organic carbon (*TOC*) also exceeded the permissible value, and amounted to 14.5 mg C/dm³. The high contamination level of the water in the analysed sample was also confirmed by the high concentration of chlorides, 282 mg Cl⁻/dm³.

The content of chlorides and nitrates in the tested water was satisfactory (Fig. 2g, h). However, during the filtration cycle, they were concentrated in the pool basin. As the water supply was low in concentrations of chlorides (12 mg Cl⁻/dm³), it was determined that the intake of "fresh" water for the pool circuit was not sufficient. This fact was also confirmed by the concentration of nitrates, which increased with the filtration time.

Silver nanoparticles, AgNP, are particularly easy to interact with bacterial membranes due to their small size [43]. AgNP toxicity is associated with the release of both Ag⁺ ions and particles, resulting in particle-related toxicity and/or „the Trojan horse effect” [23, 44]. However, it is difficult to determine what form of toxicity predominates in this effect. It is strongly dependent on the form of nanoparticles, their size, charge, structure, type of organism with which they interact, and such environmental parameters as pH, ionic strength or NaCl concentration [22, 45]. For example, toxicity of nanoparticles in relation to *Escherichia coli* is associated in the first phase of contact with molecules (first 6 h of exposure), then within 48 h Ag⁺ ions are released, which are also responsible for the destruction of these bacteria [23].

Nanosilver penetrates into the cell. AgNP as the source of Ag⁺ ions is responsible for the oxidative stress associated with the generation of reactive oxygen species, causing damage to cellular components, including DNA damage, activation of antioxidant enzymes and damage to the cell membrane [23, 24, 44].

The Microtox[®] test showed a significant decrease in bacterial bioluminescence under contact with nanosilver solutions. The raw pool water, without the addition of nanoparticles, already showed a toxic effect at 68 % after 5 minutes of exposure and over 73 % after 15 minutes (Fig. 3a). However, the toxic effect of nanosilver solutions ranged from 63 to 90 % after 5 minutes of exposure and from 77 to 96 % after 15 minutes. It should be emphasized that both the increasing concentration of nanosilver and the prolongation of the contact time had a significant impact on the increase of toxicity of the samples relative to the bacteria *Aliivibrio fischeri*. The obtained results confirm the tendency presented by Echavarrri-Bravo et al. who observed the effect of silver nanoparticles on the population of marine bacteria. The increase in AgNP contact time with microorganisms caused a decrease in their population. The tested product inhibited bacterial growth after exceeding the concentration of 0.072 mg/dm³ [46].

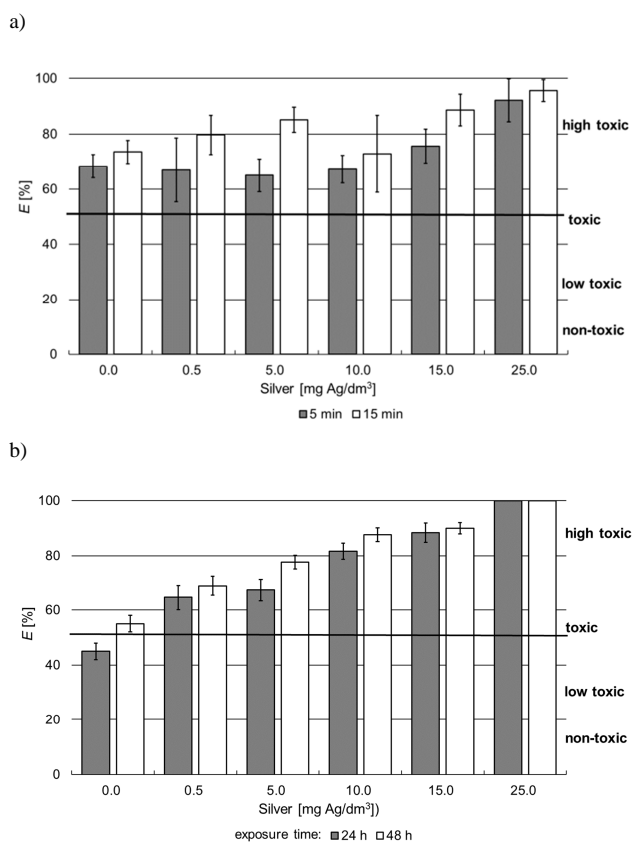


Fig. 3. Classification of toxicity of pool water samples tested with silver nanoparticles in tests: a) Microtox[®], b) mortality of *Artemia salina*

As regards the subliminal microorganisms, it should be borne in mind that silver nanoparticles, which will be transported to sea waters, have less stability and may have different effects [44]. One of the phenomena is related to the presence of Cl⁻ ions. In the

chlorination process the solubility of biologically available silver forms is significantly reduced. The result is the formation of AgCl, which in turn reduces the release of Ag⁺, but leaves such a product to be transported in the environment for a long time [23]. An important factor activating the toxicity of nanoparticles, which may be important in photometric measurements is the phenomenon of light induction and transformation of nanoparticles. This speeds up the oxidation of their surface and the release of Ag⁺ [23, 24].

There was also a clear relationship between the concentration of nanoparticles and the increase in mortality of *Artemia salina* nauplii and the prolonged contact time (Fig. 3b). Initially, the toxicity was low - 44 % with an exposure time of 24 hours. However, with an increase in observation time to 48 h, the mortality of individuals increased to 55 %. The addition of nanoparticles at a concentration of 0.5 mg Ag/dm³ caused an increase in mortality of test organisms to over 60 %. *Artemia salina* nauplii turned out to be highly sensitive to high concentrations of nanosilver in the tested samples (one hundred percent mortality of individuals at 25.0 mg Ag/dm³) This organism has a higher sensitivity to the presence of silver nanoparticles than *A. fischeri* bacteria, but one should have in mind that the contact time with the toxicant was longer.

Because *Artemia salina* belongs to crustaceans, its sensitivity can be compared with another popular indicator organism in this group - *Daphnia magna*. This species also shows high sensitivity to elevated concentrations of silver nanoparticles. Approximately 60 % mortality of individuals was observed, at a concentration of 1.6 µg Ag/dm³ [20]. Jemec et al. [47] also found that the concentration range from 0.001 to 0.1 mg Ag/dm³ has a highly toxic effect on a wide group of indicator organisms, at different levels of the organization - algae, crustaceans, fish.

The effect of silver nanoparticles on plants can be both negative and positive. Unlike bacteria and animal organisms, the inhibition of plant growth is more affected by particles with diameters above 20 nm. This is due to the fact that smaller AgNPs stop mainly in the root phase, while larger ones migrate deeply through plant tissues [48]. Rhizofiltration is the first place of interaction between the toxic substance and the cell walls of the plant, there are adsorption and precipitation processes [49]. The main factor inhibiting growth here is the overproduction of reactive oxygen species, which often leads to a reduction in the amount of chloroplasts, but it does not contribute to a clear reduction in the number of photosynthetic pigments [21].

The research carried out with the use of *Lemna minor* allowed us to observe the strong destructive impact of nanoparticles on the plant's fronds. In a sample of pool water without the addition of nanosilver, the stimulation of the growth of fronds was observed at -15 % (Fig. 4a). With the increase in the dose of nanoparticles in the solution, the growth of fronds was inhibited (*E* in the field 90-100 %), at doses of 15 and 25 mg Ag/dm³. Their discoloration and disintegration of plant tissue were also observed. In the case of low-rooted aquatic plants, it is not possible to partially stop the toxicant on the roots. In addition, *Lemna minor* samples were irradiated during the entire duration of the test, which may have contributed to the induction and growth of AgNP toxicity [24].

A slight effect of the concentration of nanoparticles was observed on *Sinapis alba* germination as at a concentration of 15 mg Ag/dm³, the average value was 6.7 %, and at 25 mg Ag/dm³ only 3.3 %. Because the relationship between a toxicant's negative impact and plant sprouting may not be a good indicator of inhibition, root growth should be analysed to describe the inhibitory properties of nanoparticles [48]. The inhibition of root growth was observed in full range during *Sinapis alba* test (Fig. 4b). In test tubes that did

not contain nanoparticles, the average toxic effect was 50 %. The root inhibition value was gradually increased and at a concentration of 25 mg Ag/dm³ nanoparticles it was over 89 %. This indicates that nanoparticles are only one of the factors that inhibit root growth of *Sinapis alba* in pool water samples.

In the case of testing the properties of silver nanoparticles on plant development, the observed effect depends largely on the species of the plant we are exploring, its size and the environment in which it is located (water or soil). Genetic differences of indicator organisms may contribute to obtaining contrasting test results.

The form of nanoparticles and the type of surface-coating agents, with stability and ability to react under the influence of environmental factors, can have a significant influence on the size of the toxic effect [43, 44, 50].

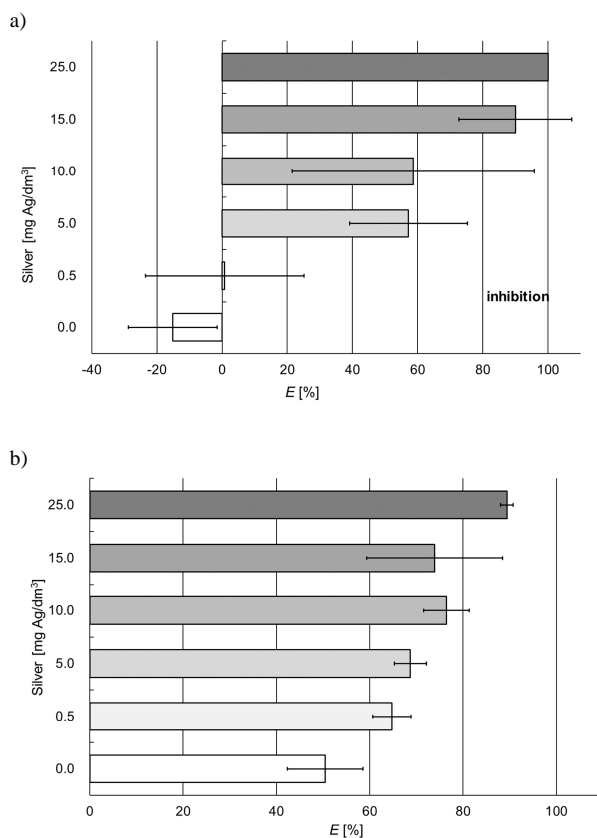


Fig. 4. Macroscopic effect of silver nanoparticles on selected indicator plants: a) *Lemna minor*, b) *Sinapis alba*

Conclusion

The tests performed indicate the effectiveness of the proposed pool water treatment system, incorporating the dosing of nanosilver colloidal solution, a vacuum filter with

multilayered bed and the irradiation of the circuit water with UV light. The parameters of water quality in the pool basin were compliant with requirements in this regard.

- The low levels of silver in pool water samples - about 0.008 mg/dm^3 - did not constitute a risk to the health of bathers. Silver concentrations of up to 0.1 mg/dm^3 can be tolerated in the case of silver salts to maintain the bacteriological quality of drinking water [51].
- Despite the use of the colloidal solution of nanosilver as a bacteriostatic product, it was found that favorable conditions for the development of bacterial colonies were present in the bed. The anthracite and sand filtration bed facilitated the growth of bacteria, which then were washed out to the filtrate.
- Although CFU of bacteria in the filtrate samples was high, water from the pool basin contained only 1 CFU/1 cm^3 (permissible number: 100 CFU/1 cm^3). Thus, the two-step disinfection (UV+NaOCl) was sufficient to ensure safe bath.
- The redox values further confirmed the effectiveness of protecting the pool water against bacteriological contamination.
- Additionally, a systematic decrease in free chlorine concentration was observed in water samples taken from the subsequent parts of the pool circuit (filtering the water through a filtration bed with an anthracite layer decreased it by 88.6 %) and the systematic increase of chlorides and nitrates during the filtration cycle indicated that an insufficient amount of water was taken into the pool circuit.
- Toxicity of nanoparticles depends to a large extent on their concentration, form of occurrence and environmental factors in which they will be found. The ecotoxicological effect that has been reported in the presented work is related, among others, to the used concentrations, which significantly exceeded those that were found in the real samples. Due to many still unknown mechanisms of interaction and transformation of nanoparticles in living organisms, further study of this issue is necessary. Because toxicity can be generated on many levels, it is necessary to assess: oxidative stress, cytotoxicity, genotoxicity, growth and development of plant shoots.

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