

Inhibition of *Daphnia magna*'s occurrence in drinking water treatment process by controlling its phototactic behavior

Tao Lin, Yiwen Tan and Wei Chen

ABSTRACT

Cladocera zooplankton as carriers of bacteria result in biological risk due to their occurrence in drinking water treatment systems. In this paper, bench-scale experiments were performed to investigate the inhibition effect on *Daphnia magna* (*D. magna*) by controlling its phototactic behavior. The results showed that UVA had a negative effect on the phototaxis of *D. magna*, indicating an active movement away from light source, while blue light was positive in inducing phototactic behavior. The water quality could influence the phototactic behavior of *D. magna*. When the turbidity value was higher than 10 NTU or total organic carbon (TOC) concentration was beyond 4 mg/L, the phototaxis of *D. magna* to UVA (25 $\mu\text{W}/\text{cm}^2$ intensity) or blue light (1,000 Lux intensity) was significantly weakened. It was difficult for *D. magna* to offset the effect of water flow by its phototactic movement when the flow rate was higher than 10 mm/s. According to the above results, with suitable process parameters in full-scale experiments, the occurrence of *D. magna* in the effluent of sedimentation tank and activated carbon filter was obviously inhibited by the UVA irradiation and blue light induction, respectively.

Key words | blue light, *Daphnia magna*, drinking water treatment, phototaxis, UVA

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INTRODUCTION

In recent years, there have been many reports on the occurrence of cladocera zooplankton in drinking water sources (Chen *et al.* 2012; Bichai *et al.* 2014; Pascual *et al.* 2014). Cladocera exhibit great vitality and transfer potential, and can easily penetrate treatment processes to enter drinking water (Bichai *et al.* 2014). Cladocera can be seen by the naked eye, which would cause consumers to feel that the water is unsanitary. As well, zooplankton may harbor viable microorganisms which are protected from disinfection during drinking water treatment. Therefore, the occurrence of cladocera in drinking water treatment processes poses a threat to human health. Zooplankton have frequently been found in the effluent from waterworks in southern China (Tao *et al.* 2016), with *Limnoithona sinensis* (*L. sinensis*) and *Daphnia magna* (*D. magna*) being the predominant species. Complete removal of cladocera in

waterworks' effluent is vital and previous studies mainly focused on how to completely inactivate zooplankton via disinfection. However, cladocera have a strong resistance against oxidation and are not effectively inactivated by conventional disinfection methods, such as chlorination. It has also been well documented that cladocera have been shown to provide an effective bacterial refuge against external stressors, particularly UV radiation and chlorination (Bichai *et al.* 2008; 2014; Lin *et al.* 2014; Tao *et al.* 2016). Bichai *et al.* (2008) found that zooplankton clouds carry *Giardia cysts* and *Cryptosporidium oocysts* against disinfection and enter pipe networks, and then pathogenic protozoa attached to zooplankton were re-released to the water. Lin *et al.* (2014) found that as a carrier, *L. sinensis* could provide protection to attached bacteria from disinfection.

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Some effective approaches have been reported as how to eliminate zooplankton from the drinking water treatment process. Schreiber et al. (1997) found that the optimization of backwashing could reduce the zooplankton density in the effluent of sand filters. *Mesocyclops leuckarti* of zooplankton can be effectively removed in water by chlorine dioxide preoxidation combined with a conventional drinking water treatment process (Zuo et al. 2006). In China, a strengthening conventional water treatment combined with preoxidation was used to control zooplankton. However, these present methods have disadvantages, such as the instable removal effect, high operation cost, resurrection of zooplankton and disinfection by-products. Therefore, it is essential to explore a new approach to effectively control the penetration of zooplankton in the drinking water treatment process.

Biological studies have shown that zooplankton have phototaxis behavior (moving to the light source or escaping from the light source) in certain lighting conditions, which is the main reason for the vertical migration of zooplankton in natural waters (Dupont et al. 2009; Williamson et al. 2011). Cladocera have shown UV avoidance behavior, i.e., a negative phototaxis movement away from the light source (Aarseth & Schram 1999; Speckmann 2000). Smith & Macagno (1990) studied the spectral sensitivity of freshwater *D. magna*, indicating a decreasing absolute sensitivity with the wavelengths ranging from 348 nm to 608 nm. Fischer et al. (2006) found that *D. magna* has a significant positive phototaxis to visible light while it is negative to UV especially at 340 nm. Vega & Pizarro (2000) evaluated the occurrence of oxidative stress in cladoceran *D. longispina* exposed to UVA and UVB radiation. However, there is still no research on the elimination of zooplankton occurrence in drinking water treatment processes by controlling their phototactic behavior. Phototaxis behavior of zooplankton could be affected by the wavelength of light, light intensity, and illumination time, which have distinct differences in influencing different species of zooplankton. Similarly, the phototaxis of zooplankton has also been affected by the concentration of organic matter and turbidity in water, which in natural waters are different from those in the drinking water treatment process.

In this paper, the phototaxis of *D. magna* was investigated when it was exposed to UV irradiation or visible

light induction. The influences of turbidity, organic matters, and water flow rate on *D. magna*'s phototaxis were analyzed. The feasibility was discussed of effectively inhibiting the penetration of zooplankton by controlling its phototaxis in the drinking water treatment process. A full-scale experiment was then conducted to verify the effectiveness of this approach.

MATERIALS AND METHODS

D. magna sampling

Zooplankton were collected from the effluent water of a sedimentation tank in a waterworks in Wuxi, China, using a zooplankton net (pore size = 0.074 mm). The predominant species, *D. magna* (2.5 mm body length), was chosen as the experimental species. Mature *D. magna* were initially selected from a natural population under a stereoscope and then artificially cultivated in the laboratory. Adult pairs were isolated in an aerated glass aquarium with a volume of 5 L at constant temperature (24 ± 1.5 °C) and a photoperiod of 14 h light/10 h dark. The culture medium (per liter sterilized pure water) comprised 0.147 g CaCl₂, 0.840 g NaHCO₃, 0.0075 g KCl, 0.0203 g MgCl₂, 0.5 g dried yeast, and 15 mL of a log-phase algal culture (*Chlorella vulgaris* at a cell density around 10^6 – 10^7 cells mL⁻¹). Three-quarters of the water in the culture tanks was replaced every 24 h (Gong et al. 2013).

The experimental equipment

Two different experimental set-ups were designed to simulate the flow condition of horizontal sedimentation tank and vertical filters, respectively. Both experiments were carried out in two patterns, i.e., the quiescent water experiment and the flowing water test. The pump and the valves were used to adjust the water flow rate in the flowing water test. To simulate a horizontal sedimentation tank, the experimental set-up was designed according to De Meester & Cousyn's (1997) reports, as shown in Figure 1(a). It consisted of five chambers, expressed by Q, L, M₁, M₂, and R, respectively, which were made of black opaque plastic except the facet closest to the light source which was made of quartz glass

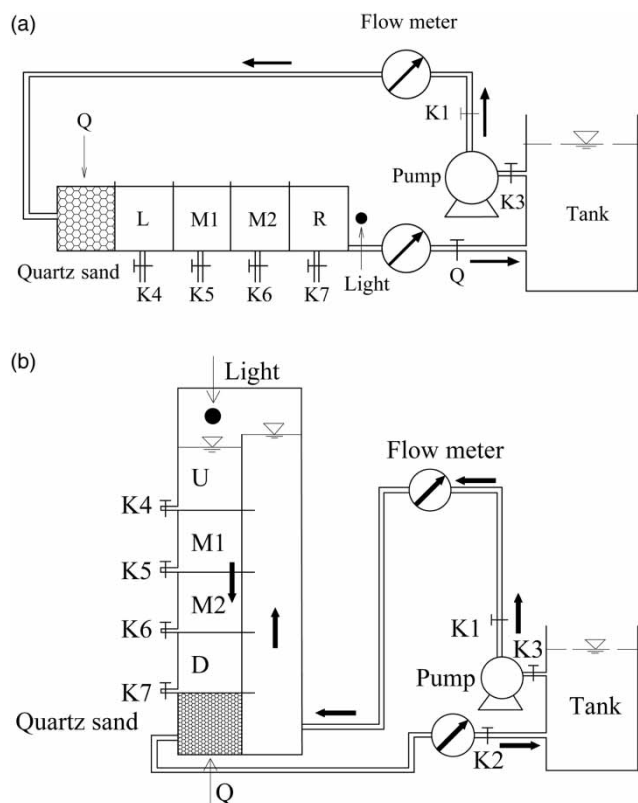


Figure 1 | (a) Horizontal experimental set-up and (b) vertical experimental set-up. K1–K7 are the water valves, in which K1–K3 were used to control the water flow rate while the rest were used to discharge water and collect *D. magna* after irradiation. L, M₁, M₂, R, U, and D are the chamber numbers.

(95% light transmittance) to ensure ultraviolet (UV-light) passed through. Chamber Q was filled with quartz sands in the flowing water test, working as a flow equalization zone. A plankton net (pore size = 0.074 mm) was set between chamber Q and chamber L, which only allows water to flow through. As well, plankton nets were set in the flow channel of chamber R to retain *D. magna* in the system. The initial light intensity could be adjusted by changing the horizontal distance of the lamp to the facet. There was a water outlet (diameter = 1.5 cm) in the bottom of every chamber. Only chamber M₂ had a detachable cover, where *D. magna* could be added into the set-up. The plastic partition could be inserted into the slot between every two adjacent chambers so as to count the number of *D. magna* in every chamber at the end of the experiment.

To simulate the vertical flow of filters, the experimental scheme is shown in Figure 1(b). The set-up consisted of five chambers (U, M₁, M₂, D, and Q, 15 cm × 15 cm × 15 cm),

which were made of black polymethyl methacrylate. Chamber Q was filled with quartz sands, working as a flow equalization zone. A plankton net (pore size = 0.074 mm) was set between chamber Q and chamber D to only allow water to pass through. The light intensity could be adjusted by changing the distance between the lamp and the water surface. *D. magna* was added into the set-up from the top chamber U with a detachable cover, i.e., a plankton net (pore size = 0.074 mm). The steel partition could be inserted into the slot between every two adjacent chambers for counting the number of *D. magna* at different chambers.

According to previous studies (Smith & Macagno 1990; Aarseth & Schram 1999; Speckmann 2000), three kinds of ultraviolet light (Philips CLEO PL-L36 W UVA, 320–400 nm, peaks at 365 nm; Philips CLEO PL-L36 W UVB, 280–315 nm, peaks at 311 nm; Philips TUV36 W, 200–280 nm, peaks at 254 nm) were used for negative phototaxis experiment and three kinds of visible light (blue light, 434 ± 5 nm; green-yellow light, 525 ± 5 nm; orange-red light, 608 ± 5 nm) for positive phototaxis experiment. An ultraviolet radiation meter and illumination-meter (both purchased from Photoelectric Instrument Factory of Beijing Normal University) were used for the measure of light intensity.

The added substances

Ultrapure water (Milli-Q system; Millipore Corp., USA) was used throughout this study. Kaolin was added to the water to attain turbidity. The turbidity was measured by a turbidity meter (HACH 2100Q; HACH Corp., USA). Dried, powdered NOM (International Humic Substances Society (IHSS), Golden-Colorado) was used as organic matters source. Total organic carbon (TOC) was measured by a TOC analyzer (HACH IL550; HACH Corp., USA). Water samples were aerated for 1 h using aquarium pumps to ensure sufficient dissolved oxygen (DO) prior to experiments.

In the quiescent horizontal experiments, 50 *D. magna* together with artificial water are introduced into the system from the water inlet of chamber M₂. Subsequently, *D. magna* were kept in the dark for 5 min to eliminate the impact of external factors on phototaxis. The light was

turned on and the light intensity adjusted to the desired level on facet A, which was the closest facet to the light. After irradiating for a certain time, the light was turned off, the partition was inserted between each two chambers and a plankton net (pore size = 0.074 mm) was fixed beneath every water outlet to collect effluent *D. magna*, which were counted using a microscope. Fifty *D. magna* in the absence of illumination were used as a blank control. In the horizontal flowing water test, all experimental conditions were similar to those of quiescent water experiments except for using a pump to adjust the water flow according to the desired flow rate.

In quiescent water vertical experiments, 50 *D. magna* are introduced into the device from chamber U, which was then covered with a plankton net (pore size = 0.074 mm). In the flowing water test, a pump was used to adjust the water flow rate. In this study, the experimental procedures were the same as in horizontal experiments.

Experimental parameters and phototaxis measurement

In this study, the experimental conditions were controlled at a pH value of 6.5 to 7.5 by NaOH and HCl solutions, and temperature around $24 \pm 1.5^\circ\text{C}$ according to the actual treatment process. Turbidity of 1, 5, 10, 15, and 20 NTU and the TOC of 2, 4, 6, and 8 mg/L were adopted to investigate the effect of water quality on phototaxis of *D. magna*.

A pre-test showed that *D. magna* could obviously perceive the stimulation of UV irradiation at an intensity of $10 \mu\text{W}/\text{cm}^2$, therefore 5, 10, 15, 20, and $25 \mu\text{W}/\text{cm}^2$ UV-light intensities were used in the phototaxis experiment, where the exposure times were 1, 2, 3, 4, and 5 min. The water flow rate was 0, 0.83, 1.67, 2.5, and 3.33 mm/s in vertical experiments and 0, 5, 10, 15, and 20 mm/s in horizontal experiments, respectively. To ensure the reliability of the results, the experiment was performed in three parallels, so three replicate measurements of each sample were taken and the average value determined ($p < 0.05$).

A pre-test showed that in the quiescent water, *D. magna* tended to be an even distribution in the horizontal direction but a centralized distribution in the bottom of the vertical experimental equipment. In the horizontal experiment, the phototaxis was measured according to the methods

described by Michels & Meester (2004). The phototactic behavior was characterized by the phototactic index as shown in Equation (1):

$$\text{PI} = \frac{(R - L)}{(R + M_1 + M_2 + L)} \quad (1)$$

where L, M_1 , M_2 , and R represent the number of *D. magna* in each chamber. The phototactic index ranges from 1 as extremely positive behavior to -1 as extremely negative phototactic behavior (Meester 1991).

In the vertical experiment, the phototaxis measurement was characterized by a phototactic index as shown in Equation (2):

$$\text{PI} = \frac{U}{(U + M_1 + M_2 + D)} \quad (2)$$

where U, M_1 , M_2 , and D represent the number of *D. magna* in each chamber. The phototactic index ranges from 1 as extremely positive behavior to 0 as extremely negative phototactic behavior.

The full-scale experiment

The inhibiting effect of *D. magna*'s penetration in the actual drinking water treatment process was examined by controlling its phototactic behavior in a full-scale experiment. In the horizontal sedimentation tank, UVA irradiation was used to inhibit the leakage of *D. magna* by controlling its negative phototaxis. The light intensity was $25 \mu\text{W}/\text{cm}^2$. The UV lamps were vertically arranged at a distance (about 20 meters) in front of the catchment area. The layout of the UVA irradiation trial in the horizontal sedimentation tank is shown in Figure 2.

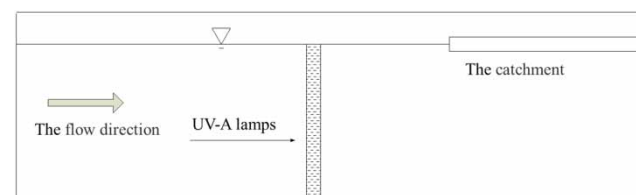


Figure 2 | Layout of UVA irradiation in the horizontal sedimentation tank.

In the activated carbon filter, blue light induction was used to inhibit the leakage of *D. magna* by controlling its positive phototaxis. The light intensity was 1,000 Lux. The lamps were evenly arranged above the water surface (about 20 cm). The layout of the blue light induction trial is shown in Figure 3.

Turbidity value, TOC concentration, and flow rate of water in the sedimentation tank or activated carbon filter were detected. The effluent of the sedimentation tank or activated carbon filter was collected at 1, 10, and 24 hours after the trial began and then the water samples were used to detect *D. magna* counts.

RESULTS AND DISCUSSION

The effect of light wavelength on phototaxis

Three kinds of ultraviolet light and three kinds of visible light were used to investigate the negative and positive phototaxis, respectively. As shown in Figure 4, there was behavior associated with negative phototaxis in *D. magna* exposed to UV irradiation. At the same irradiation intensity and exposure time, UVA obviously affected negative phototaxis, PI reaching -0.9 at $25 \mu\text{w}/\text{cm}^2$ intensity with 5 min irradiation, while UVB and UVC had less influence. The negative phototaxis effect was increased with the increasing UV irradiation intensity or irradiation time. Studies have shown that *D. magna* has a simple eye and a compound eye. The simple eye is a simple structure of photoreceptors, which can feel the intensity and the direction of light (Arendt & Wittbrodt 2001; Pichaud & Desplan 2002). *D. magna's* perception of light is the most important reason for diel vertical migration (Jensen et al. 1999;

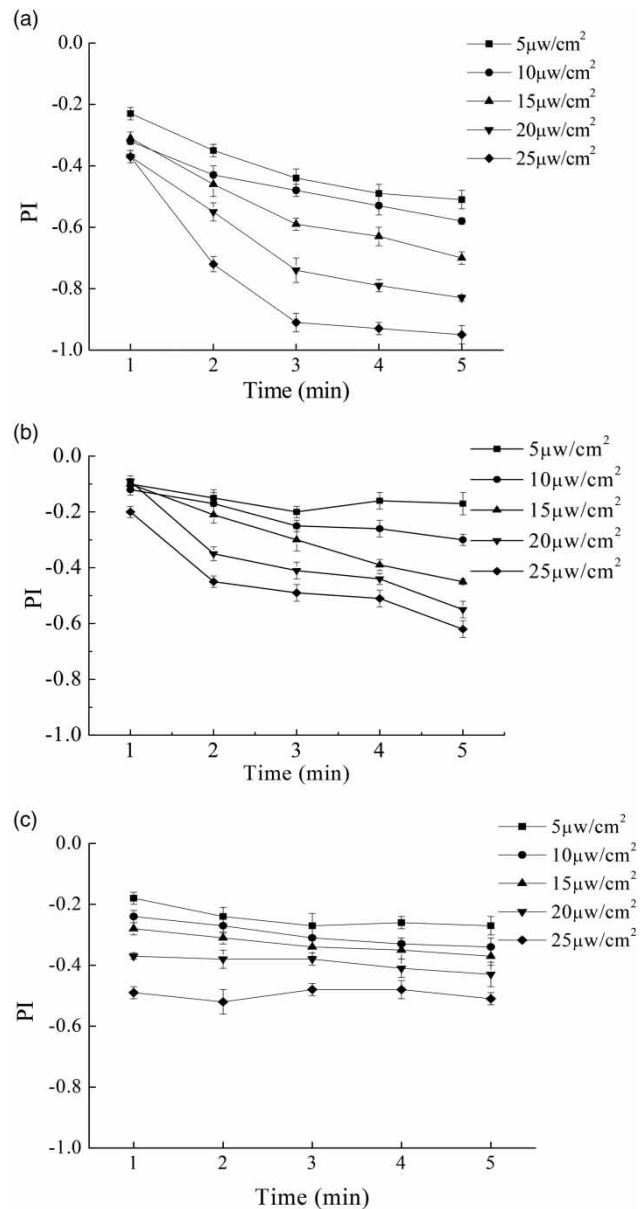


Figure 4 | The effect of UV (a) UVA, (b) UVB, and (c) UVC lights with different light intensities on negative phototaxis of *D. magna*.

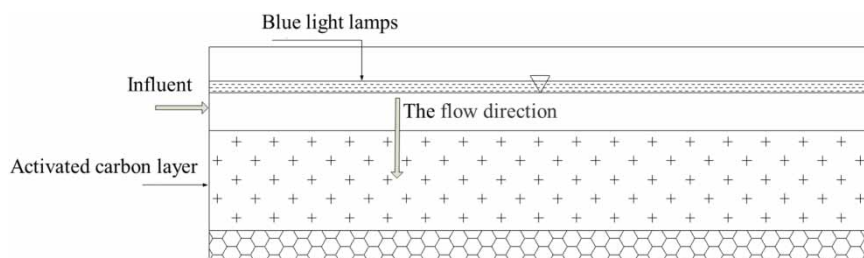


Figure 3 | Layout of blue light induction in the activated carbon filter.

Ringelberg 1999; Williamson et al. 2011). The wavelength of UVA is longer than others, which allows the light more easily to pass through water to be sensed by *D. magna*.

The results in Figure 5 show the positive phototaxis of *D. magna* exposed to visible lights. It can be seen that *D. magna* have an obvious positive phototaxis with exposure to blue light irradiation, PI reaching 1.0 at the

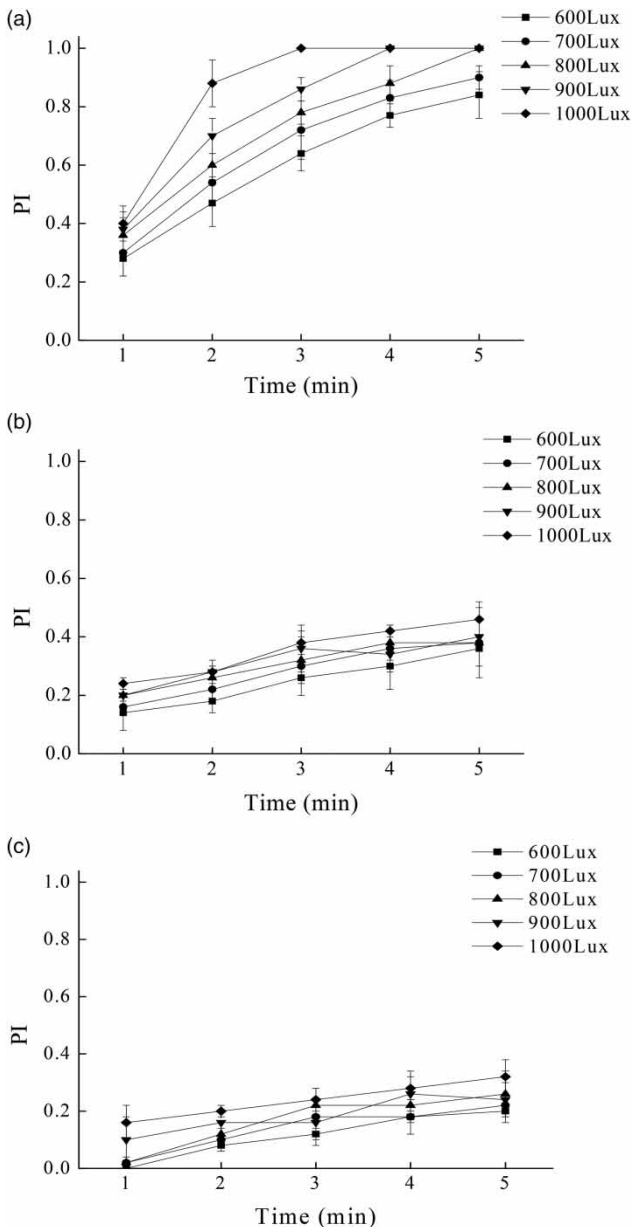


Figure 5 | The effect of visible lights with different light intensities on positive phototaxis of *D. magna*: (a) blue light (434 ± 5 nm), (b) green-yellow light (525 ± 5 nm), and (c) orange-red light (608 ± 5 nm).

intensity of 800–1,000 Lux with 3 min exposure. However, *D. magna* showed a weak positive phototaxis under the orange or red light irradiation. Natural water usually appears blue (Lythgoe 1984) and the blue photoreceptor was confirmed in *D. magna*'s simple eye (Martin & Mote 1982; Smith & Macagno 1990). Blue light has the strongest transmission capacity in the water. Therefore, it is easier to be sensed than other visible lights by *D. magna* (Mostajir et al. 1999).

According to the above experiments, the UVA at 25 $\mu\text{W}/\text{cm}^2$ intensity with 5 min irradiation and blue light at 1,000 Lux intensity with 3 min irradiation were selected for subsequent study in negative and positive phototaxis experiments, respectively.

The transmission of light in the water

The perception of *D. magna* to light is closely related to the transmission of light in water, which is mainly influenced by the water quality, such as the turbidity and TOC. Due to the low turbidity of experimental water samples, light transmission in the water is mostly in line with Lambert–Beer's law. According to Lambert–Beer's law, the light absorption in the solution is expressed by Equation (3):

$$A = \lg I_0 - \lg I = kbc \quad (3)$$

where A is absorbance, I_0 is the initial light intensity, I is the light intensity after flow through one certain thickness of liquid layer, k is proportional constant, b is the thickness of liquid layer, and c is the concentration of the solution.

A hypothesis was put forward to simulate the transmission of light in water, as shown in Equation (4):

$$I = I_0 e^{f(a,b,c)} \quad (4)$$

where I_0 is the initial light intensity, I is the light intensity after flow through a certain thickness of liquid layer, a is the turbidity, b is the thickness of liquid layer, and c is TOC concentration.

According to the results of experiments, the functional expressions were established as Equations (5) and (6) for describing the transmission of UVA and blue light in

water, respectively. The boundary conditions were given according to the experimental condition in the process of developing mathematical models. The detailed derivation process is shown in the Supplementary material (available with the online version of this paper).

$$lgI = lgI_0 + (0.0185 - 0.0018a + 10^{-5}a^2 - 0.033c)b$$

$$(1 \text{ NTU} \leq a \leq 20 \text{ NTU}, \quad 0 \text{ cm} \leq b \leq 60 \text{ cm},$$

$$2 \text{ mg/L} < c < 8 \text{ mg/L}) \quad (5)$$

$$lgI = lgI_0 + (-0.0149 - 0.0011a + 7 \times 10^{-6}a^2 - 0.006c)b$$

$$(1 \text{ NTU} \leq a \leq 20 \text{ NTU}, \quad 0 \text{ cm} \leq b \leq 60 \text{ cm},$$

$$2 \text{ mg/L} < c < 8 \text{ mg/L}) \quad (6)$$

Four random experiments were carried out to estimate the accuracy of the two models. The results are shown in Table 1. The relative errors were lower than 10%, meaning that the two models are feasible to analyze the influence of turbidity and TOC on phototaxis.

The effect of turbidity on phototaxis

Water turbidity is often used to investigate the characterization of scatter or absorption of light in water samples.

The phototaxis variation of *D. magna* was investigated under the different desired turbidity at irradiation of 25 $\mu\text{W}/\text{cm}^2$ UVA or 1,000 Lux blue light. As shown in Figure 6, when the turbidity was less than 10 NTU, *D. magna* showed great negative phototaxis to the induction of UVA irradiation. After 3 min irradiation, PI stabilized at about -0.95 . However, when the turbidity was higher than 15 NTU, the negative phototaxis of *D. magna* to UVA irradiation could be obviously weakened.

Water turbidity is mainly caused by inorganic and organic matters such as soil, microbes, and suspended particles, which can affect the transmission of light in water. The transmission of UVA is closely relative to the amount of suspended particulate matters in water (Rahman et al. 2014). The presence of suspended particles not only absorbs part of the light but also impedes or changes the transmission of light (Hongve & Åkesson 1998). When turbidity was higher than 10 NTU, the particle concentration weakened *D. magna*'s phototaxis behavior.

The results in Figure 7 show the effect of turbidity on its positive phototaxis when *D. magna* was exposed to blue light. When the turbidity was within 10 NTU, *D. magna* showed a significant positive phototaxis, PI stabilizing at around 1.0 after 5 min irradiation. When the turbidity

Table 1 | Comparison between model values and measured values of random experiments

Model	Turbidity (a, NTU)	TOC (c, mg/L)	Position (b, cm)	Model I	Measured I	Relative error (%)
Model for UVA ($\mu\text{W}/\text{cm}^2$)	5.0	0.5	0	500.0	500.0	0.0
			15	396.0	380.4	4.1
			30	313.7	325.4	-3.6
			45	248.4	240.0	3.5
			60	196.8	204.8	-3.9
			60	196.8	204.8	-3.9
	12.0	1.7	0	500.0	500.0	0.0
			15	68.0	68.8	-1.2
			30	9.3	10.0	-7.4
			45	1.3	1.3	-6.3
			60	0.2	0.2	1.7
			60	0.2	0.2	1.7
Model for blue light (Lux)	3.0	1.1	0	600.0	600.0	0.0
			15	255.3	261.6	-2.4
			30	108.7	110.5	-1.7
			45	46.2	48.2	-4.1
			60	19.7	20.2	-2.5
			60	19.7	20.2	-2.5
	13.0	2.7	0	600.0	600.0	0.0
			15	130.3	128.4	1.5
			30	28.3	29.3	-3.4
			45	6.1	6.3	-2.5
			60	1.3	1.4	-2.0
			60	1.3	1.4	-2.0

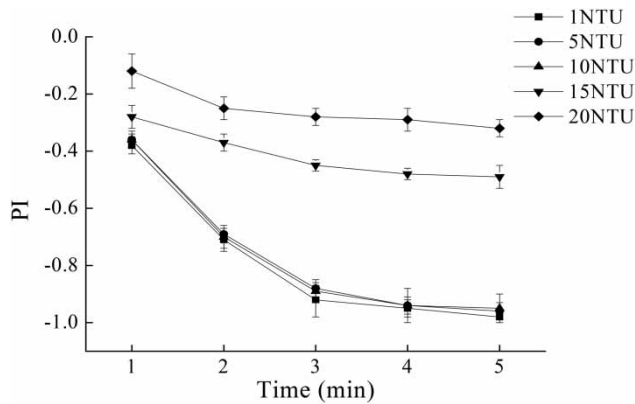


Figure 6 | The effect of turbidity on negative phototaxis of *D. magna* (I_0 of UVA = 25 $\mu\text{W}/\text{cm}^2$, TOC = 0 mg/L, quiescent water).

reached 15 NTU, the positive phototaxis of *D. magna* was weakened but still showed an observed positive effect unlike that under the irradiation of UVA. With the same turbidity, the phototactic behavior of *D. magna* was more affected by 1,000 Lux of blue light than 25 $\mu\text{W}/\text{cm}^2$ of UVA.

The effect of TOC concentration on phototaxis

As shown in Figure 8, when TOC concentration ranged from 0 mg/L to 4 mg/L, *D. magna* showed an obvious negative phototaxis to UVA irradiation. After 3 min irradiation, PI reached to around -1 , indicating that all *D. magna* were far away from the UV-light source. However, when TOC was 6 mg/L, PI dropped to -0.4 . This showed that when TOC concentration was higher than 4 mg/L, the negative

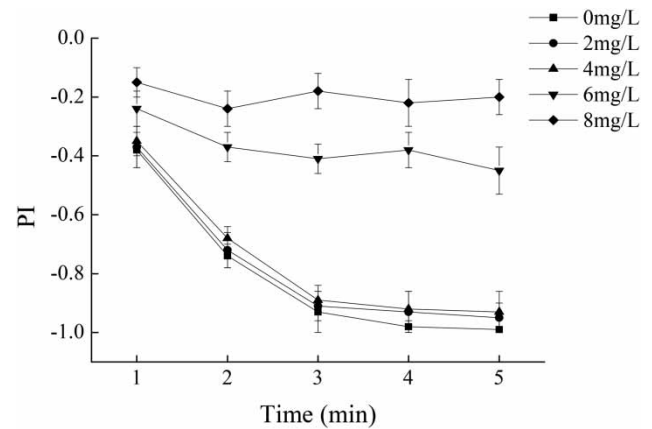


Figure 8 | The effect of TOC concentration on negative phototaxis of *D. magna* (I_0 of UVA = 25 $\mu\text{W}/\text{cm}^2$, turbidity = 0 NTU, quiescent water).

phototaxis was obviously weakened. This is because the organic matter in water may absorb a large amount of UVA. Studies have shown that organic matters have a strong ability to absorb UV-light due to their complex structures and various functional groups, which is one of the reasons that marine zooplankton can avoid UV damage by vertical migration (Lythgoe 1984; McCarty & Aieta 1984; Smith & Macagno 1990; Eaton 1995). In addition, the water may be colored by organic matter, thereby further weakening the UV-light transmission (Smith & Macagno 1990).

The results in Figure 9 show that the changes of TOC concentration also had an impact on the positive phototaxis of *D. magna*. When it ranged from 0 mg/L to 4 mg/L, PI increased significantly with the increasing exposure time.

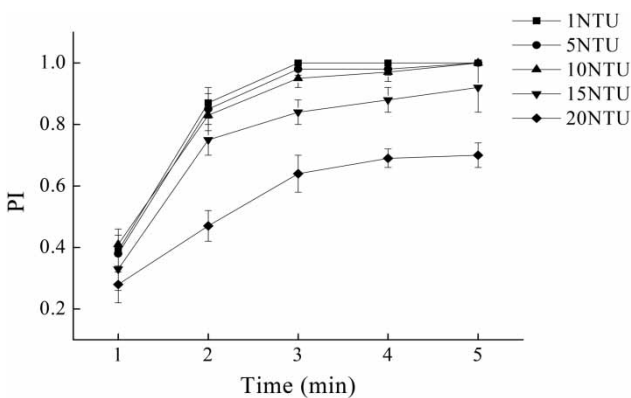


Figure 7 | The effect of turbidity on positive phototaxis of *D. magna* (I_0 of blue light = 1,000 Lux, TOC = 0 mg/L, quiescent water).

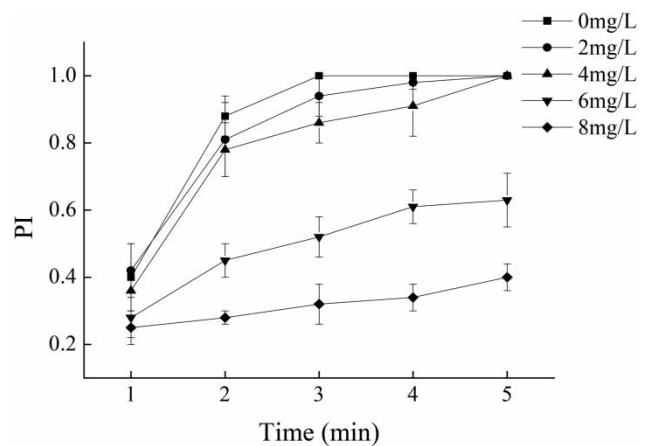


Figure 9 | The effect of TOC concentration on positive phototaxis of *D. magna* (I_0 of blue light = 1,000 Lux, turbidity = 0 NTU, quiescent water).

When TOC concentration was 6 mg/L, PI still stabilized at around 0.6. The organic matters in water can absorb part of the blue light due to their functional groups and special chemical bonds (McCarty & Aieta 1984; Eaton 1995). However, *D. magna* have special photoreceptors to sense blue light, which has a strong light transmission capability (Martin & Mote 1982; Mostajir et al. 1999). Therefore, although blue light is weakened, *D. magna* can still sense it and swim close to the light source.

The effect of water flow rate on phototaxis

Figure 10 shows the effect of the water flow rate on the negative phototaxis of *D. magna*. Turbidity of 5 NTU and TOC of 3 mg/L were set according to the water quality near the sedimentation tank's outlet. When the flow rate was lower than 5 mm/s, it had little effect on negative phototaxis and PI could reach -0.95 , indicating that *D. magna* could swim against this water flow to avoid ultraviolet radiation. However, when the flow rate was up to 15 mm/s or 20 mm/s, *D. magna* almost lost the ability of resistance to flow and were carried by the water close to the light source. During observing the swimming behavior of *D. magna*, it was found that *D. magna* did not swim in a straight line but in a helix of a millimeter-sized diameter around an axis. This was also demonstrated by Jékely et al. (2008). It is a special action of *D. magna* to feel the light intensity in different directions, which is favorable for *D. magna* against the carrying capacity of water flow. The swimming speed of

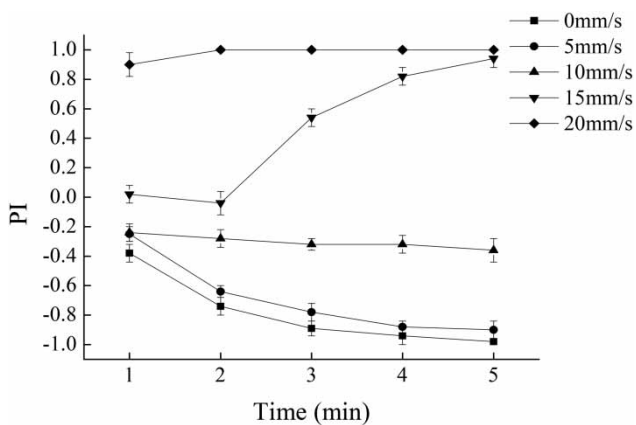


Figure 10 | The effect of water flow rate on negative phototaxis of *D. magna* (I_0 of UVA = $25 \mu\text{W}/\text{cm}^2$, turbidity = 5 NTU, TOC = 3 mg/L).

D. magna is about 10 mm/s. Actually, 15 mm/s is a designed flow of sedimentation tanks in some waterworks in China. Therefore, it is necessary to reduce the water flow rate to a desired one for preventing *D. magna* occurring in the effluent of sedimentation tanks by the negative phototactic induction of *D. magna* exposed to UVA irradiation.

As shown in Figure 11, *D. magna* showed a strong positive phototaxis to induction of blue light at all tested flow rates. Turbidity of 1 NTU and TOC of 3 mg/L were set according to the water quality in the filters. This means that when the flow rate ranged from 0 mm/s to 3.33 mm/s, *D. magna*'s phototactic behavior to blue light was hardly affected. When the flow rate was within 1.67 mm/s, PI reached 0.98 after 3 min irradiation, indicating that 98% of *D. magna* were upstream to the water surface. When the flow rate was up to 2.5 mm/s and 3.33 mm/s, PI still reached 0.9 after 4 min irradiation of blue light. In fact, the actual filtration rate of filters was lower than 2.5 mm/s in most waterworks in China. It is therefore feasible to use blue light as the induction source to draw *D. magna* to the water surface of filters so as to inhibit their occurrence in the effluent. Then, *D. magna* would flow out from filters together with backwash water.

Application to control the occurrence of *D. magna*

According to the above results, the inhibiting effect on *D. magna*'s occurrence in the effluent of the water treatment

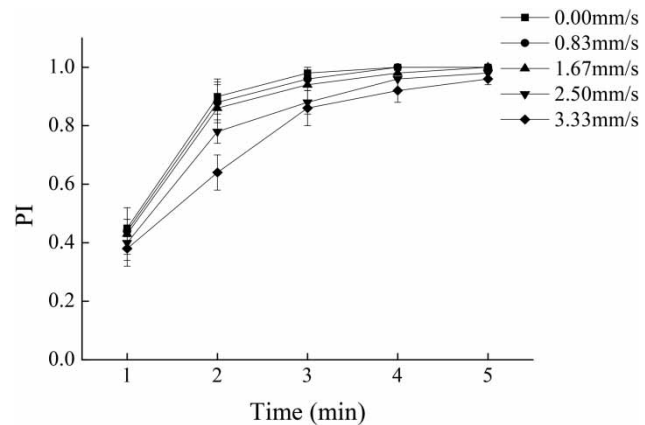


Figure 11 | The effect of water flow rate on positive phototaxis of *D. magna* (I_0 of blue light = 1,000 Lux, turbidity = 1 NTU, TOC = 3 mg/L).

process was examined by controlling its phototactic behavior in a full-scale experiment. As is shown in the above investigations, if turbidity value is higher than 10 NTU or

TOC concentration is beyond 4 mg/L, the phototaxis of *D. magna* can be weakened in waters. Therefore, the influent water qualities of the sedimentation tank and activated carbon filter were measured in an actual waterworks. As shown in Figure 12(a), in the sedimentation tank, the turbidity value was beyond 10 NTU from August to October and TOC concentration was higher than 4 mg/L from July to September. In order to improve the influent water quality, the enhanced coagulation was used to reduce the turbidity and preoxidation was conducted to remove organic matter. The dosage of coagulant (aluminum sulfate) was increased from 40 mg/L to 60 mg/L. The feed water was oxidized with ozone. The dosage of ozone was 0.8 mg/L and the contact time was 10 min. The effects of enhanced coagulation and preoxidation are shown in Figure 12(a). After the enhanced coagulation and preoxidation, the influent water quality near the UV lamp was suitable for the phototaxis control of *D. magna* in the sedimentation tank. Additionally, the flow rate in the sedimentation tank was no higher than 10 mm/s by adjusting the hydraulic load. As shown in Figure 12(b), in the activated carbon filter, the influent water quality was good enough to control *D. magna* phototactic behavior. The water flow rate in the activated carbon filter was less than 3.33 mm/s.

After installing the lamps, the effluents of the sedimentation tank and activated carbon filter were sampled at 1, 10, and 24 h performance after the trial began. The result is shown in Table 2. It was obvious that the irradiation of UVA and blue light hold back the occurrence of *D. magna* in effluent of the sedimentation tank and activated carbon filter. When the flow rate is

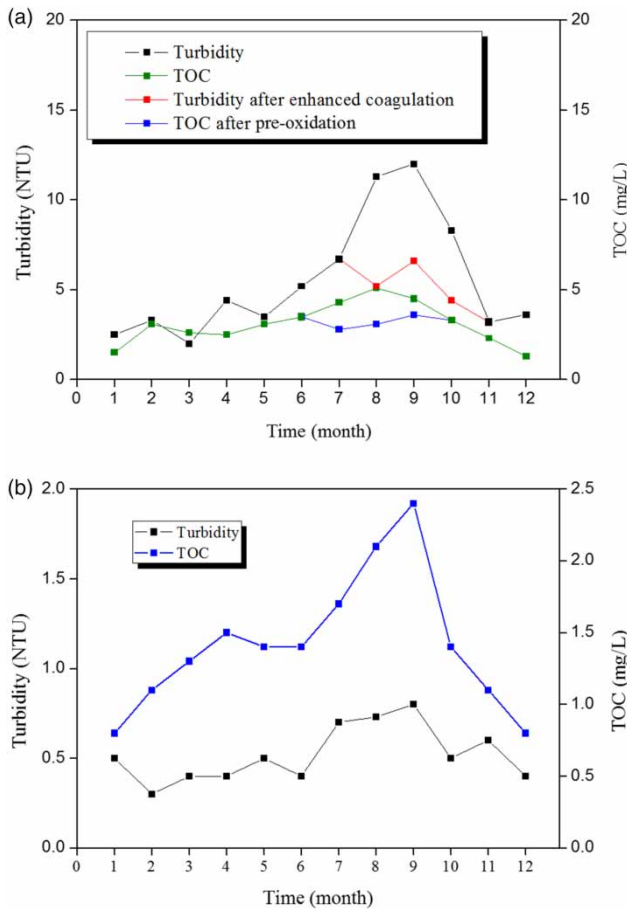


Figure 12 | The water quality in (a) the sedimentation tank and (b) activated carbon filter.

Table 2 | Application instances of UVA and blue light

Unit	Light condition	Turbidity (NTU)	TOC (mg/L)	Flow rate (mm/s)	<i>D. magna</i> in effluent (without light)			<i>D. magna</i> in effluent (with light)		
					1 h	10 h	24 h	1 h	10 h	24 h
Sedimentation tank	UVA 25 $\mu\text{w}/\text{cm}^2$	2.1	1.5	4.5	4 ^a	15	7	-	-	-
		1.8	1.4	6.6	10	2	5	1	-	-
		3.7	2.5	8.2	12	8	7	2	-	1
Activated carbon filter	Blue light 1,000 Lux	0.7	2.4	2.8	8	14	4	-	-	-
		0.5	1.0	2.5	3	6	11	1	-	-
		0.8	1.8	3.3	13	4	9	-	-	-

^aThe number of *D. magna* in every liter of water.

-, not detected.

lower than 10 mm/s, *D. magna* could resist the carrying capacity of water.

CONCLUSIONS

D. magna displayed different phototactic behaviors when they were exposed to light with different wavelengths. UVA (320–400 nm) was used for inducing a negative phototaxis of *D. magna* while blue light (434 ± 5 nm) was used for positive phototaxis. After 3 min irradiation, PI reached $-0.95/+1$ with $25 \mu\text{w}/\text{cm}^2$ UVA/1,000 Lux blue light, respectively. The phototactic behavior of *D. magna* was affected by water quality significantly. If turbidity was higher than 10 NTU or TOC was more than 4 mg/L, the phototaxis of *D. magna* was weakened due to the reduced permeability of light. The water flow rate also affected the phototaxis of *D. magna*. It was difficult for *D. magna* to resist to 10 mm/s of flow rate by its self-movement under the horizontal flow. Thus, it is necessary to reduce the water flow rate to less than 10 mm/s for preventing *D. magna* occurring in the effluent of a sedimentation tank by the negative phototactic induction of *D. magna* exposed to UVA irradiation. However, the phototaxis of *D. magna* was seldom affected by the downward water flow in the filters (flow rate ≤ 3.33 mm/s). In a full-scale experiment, the occurrence of *D. magna* in effluent of a sedimentation tank (adjusted flow rate < 10 mm/s) or activated carbon filter was obviously inhibited by a negative phototaxis induced by UVA irradiation and a positive phototaxis induced by blue light, respectively.

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