The Effects of Pulsed Streamerlike Discharge on Cyanobacteria Cells

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***Abstract*—Recently, cyanobacteria blooms (or water blooms) occurred on the surface of water bodies frequently and extensively due to eutrophia of the water. That has posed more and more serious environmental problems worldwide. In this paper, the effects of pulsed streamerlike discharge on *M. aeruginosa* cells are reported, which are one genus of cyanobacteria and ease to form water blooms. A stainless needle with a diameter of 30 μm was employed as a point discharge electrode, which is 15-cm apart from the cylinder cathode, and a 2-μs 160-kV pulse was applied. A pulsed streamerlike discharge was obtained in the water filled with cyanobacteria cells (named as sample water in this paper). From the experimental result, it can be found that the discharge collapsed the intracellular-structure gas vesicles in the *M. aeruginosa* cells, and the colonies of the cells sank to the bottom of the discharge chamber and rotten gradually.**

***Index Terms*—Cyanobacteria, *M. aeruginosa*, pulse-forming network, streamerlike discharge.**

I. Introduction

CYANOBACTERIA (often referred to as blue-green algae) are members of a group known as eubacteria or true bacteria [1]. They are a frequent component of many freshwater and marine ecosystems. Under certain conditions, especially where waters are rich in nutrients and exposed to sunlight, cyanobacteria may multiply to high densities—a condition referred to as a water bloom [8]. Large blooms of cyanobacteria can clog intake pipes and filter lines and are aesthetically unappealing. When a bloom dies in a pond or shallow lake, severe oxygen depletion can produce objectionable odors and even cause fish kills. Some cyanobacteria produce substances which are extremely toxic and are capable of causing serious illness or even death if consumed [1]. Recently, cyanobacteria outgrow rapidly and can easily form blooms in lakes and agricultural ponds because of the eutrophication. Therefore, cyanobacteria blooms have posed a serious environmental problem all over the world.

Some treatment options of cyanobacteria blooms have been researched and developed. Chemical compounds [1], ultrasonic [2], microwave [3], and mollusk were applied to eliminate cyanobacteria cells in the water. The use of streamerlike discharge in the gas to realize bacterial decontamination is a firmly established method. The application of this kind of discharge in liquids has been developed [7]. In this paper, we report the effects of streamerlike discharge in water on cyanobacteria cells.

A Blumlein-type pulse-forming network (B-PFN) was employed to provide a 2-μs 160-kV pulsed voltage to a point-to-cylinder electrode geometry and generate a tremendously high electric field and formed streamerlike discharge in a sample water, which was collected from Hikawa Dam in Kumamoto, Japan.

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Color version of Figs. 1, 2, and 4–10 are available online at http://ieeexplore.ieee.org.

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II. Experimental Apparatuses

A. Sample Water From Hikawa Dam

Fig. 1 shows the landscape [Fig. 1(a)] of the Hikawa Dam and the state of the lake surface [Fig. 1(b)] in August 2005. It can be found that the lake is green in color, and many small green particles were floating on the surface of the water. From the microscopical observation, it was confirmed that the small green particles were the colonies of cyanobacteria cells, and the major genus was *M. aeruginosa*. *M. aeruginosa*, which is 3.2–6.6 μm in diameter, is a typical genus of toxic cyanobacteria [4]. Fig. 2(a) and (b) shows the bright field photomicrographs [Fig. 2(a) with $\times$400 magnification and Fig. 2(b) with $\times$1000 magnification] of *M. aeruginosa* taken by the microscope system (BX60; Olympus, Japan) equipped with a digital camera (DXM1200; Nikon, Japan). It can be observed from Fig. 2(a) that *M. aeruginosa* in the sample water appeared black in color. Numerous *M. aeruginosa* cells gathered together and formed the colonies, and the colonies appeared to be small green particles and floated in the water. Fig. 2(b) shows a single cell of *M. aeruginosa* with 1000 times of magnification. It can be seen that the mucilaginous sheath (MS) surrounded the cells and the gas vesicles (GVs) showed blackish appearance due to the refraction of background light. GVs are the special intracellular structure of the bloom-forming cyanobacteria genus [4]. GVs are filled with gas. Therefore, cyanobacteria cells can float toward the water surface to position in optimal sunlight condition for growth [5]. The electrical conductivity of the sample water was 11.6 mS/m.

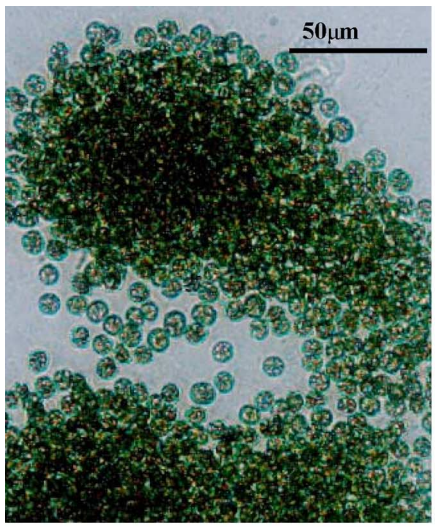
B. Pulsed Streamerlike Discharge System

Fig. 3 shows the schematic diagram of the pulsed power generator used in this paper. The generator is consist of a

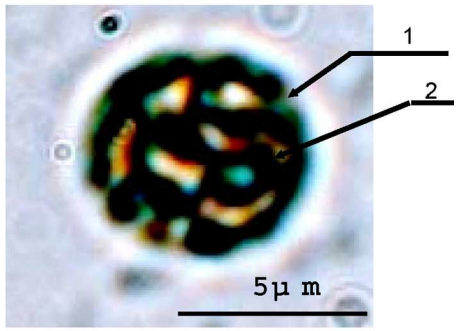
(a)

(b)

Fig. 1. (a) Landscape and (b) surface of Hikawa Dam.



(a)



(b)

Fig. 2. *M. aeruginosa* sampled from Hikawa dam. (a) Colonies of the cells. (b) Single cell, where 1: MS and 2: GVs.

positive dc source (E, HDV-50K3US; Pulse Electronic Engineering Co., Japan), the B-PFN, and a pulse transformer (PT). A triggered spark gap switch was used as a closing switch of B-PFN. B-PFN had 15 stages of LC ladder, which were composed of 20 nF of capacitor and 0.2 μH of inductor. The characteristic impedance $(2\sqrt{L/C})$ and the pulsewidth $(2N\sqrt{LC})$ of B-PFN, calculated from capacitance (C) and inductance (L) of the LC ladder, and number (N) of LC ladder stages, were approximately 6 Ω and 1.9 μs, respectively. The FINEMET (FT-3H; Hitachi Metals, Japan) was utilized as the core material of PT. PT had five of the winding ratio (windings: secondary windings $= 1:5$). Therefore, the total characteristic impedance of the generator was computed at $150\,\Omega\,(6\,\Omega \times 5^2)$.

Fig. 4 shows the configuration of the discharge chamber used in this paper. The needle-to-cylinder electrode was utilized as the discharge chamber. The needle electrode with 30 μm in tip curvature was put concentrically into the cylinder electrode with 300 mm in inner diameter and 180 mm in length. To enhance the electric field at the tip of needle electrode, the needle was covered with polyethylene insulator, except the tip part. In this paper, the discharge chamber was filled with the sample water from Hikawa Dam. The output from the pulsed power generator was connected to the needle electrode, and the cylinder electrode of the discharge chamber was grounded. The

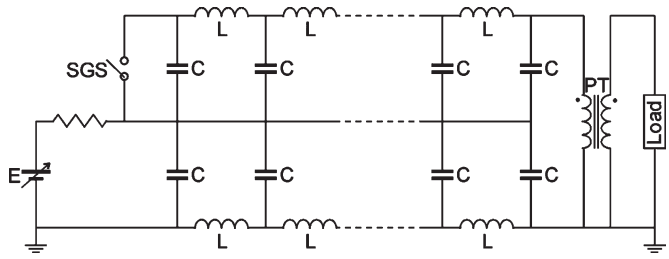


Fig. 3. Schematic diagram of the pulsed power generator.

charging voltage into B-PFN was fixed at +25 kV. In this case, the positive polarity of the pulsed voltage was applied to the needle electrode. For the utilization of the maximum flux swing of the PT core, the reset current of 2 A was flown near the PT core. The applied voltage to and the discharge current through the discharge chamber were measured using a voltage divider (EP-100 K; Pulse Electronic Engineering Co., Japan), which was connected between the needle and the cylinder electrodes, and a current monitor (Model 110 A; Pearson Electronics, USA), which can be located upon returning to the ground. The signals from the voltage divider and the current monitor were recorded in a digitizing oscilloscope (54512 B; Hewlett Packard, USA) with a 200-MHz bandwidth.

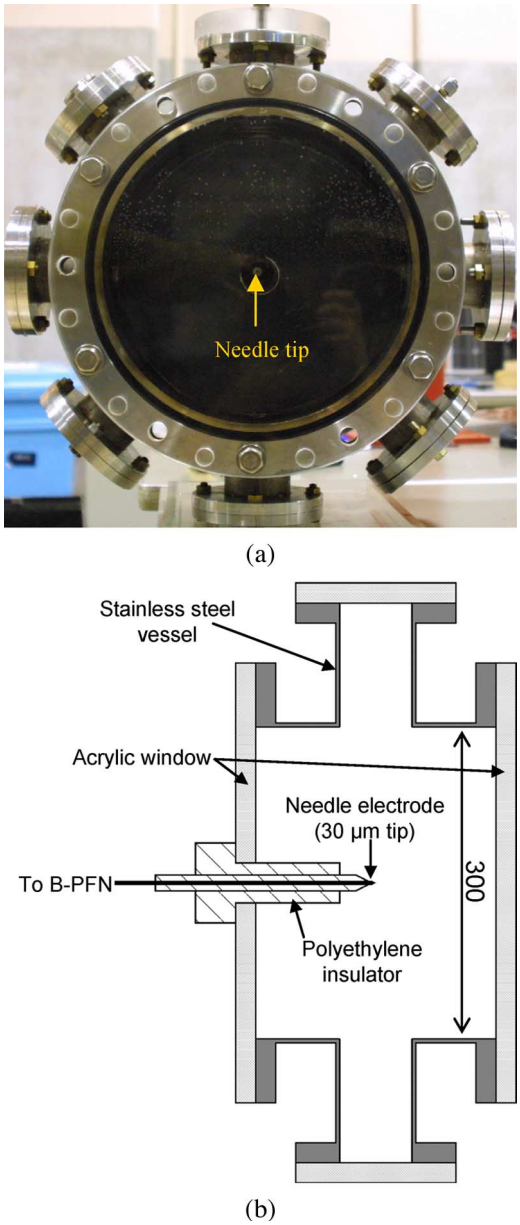


Fig. 4. Configuration of the discharge chamber, where (a) is the front view and (b) is the side view of the chamber.

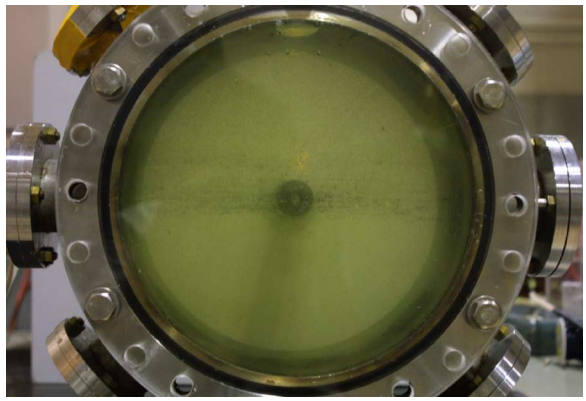


Fig. 5. Discharge chamber appearance soon after the sample water is filled in.

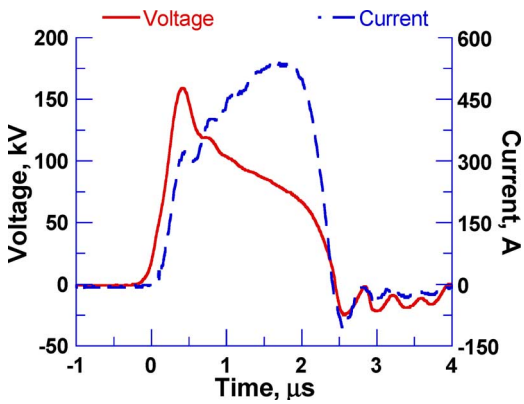


Fig. 6. Applied voltage to and discharge current through the chamber.

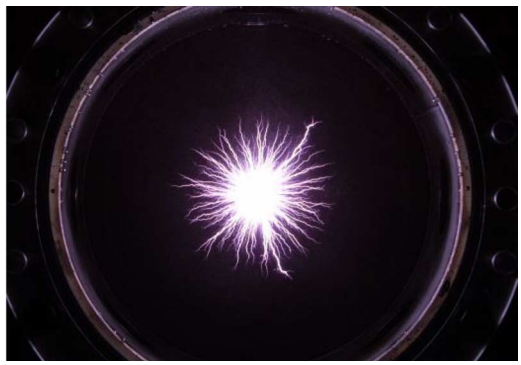
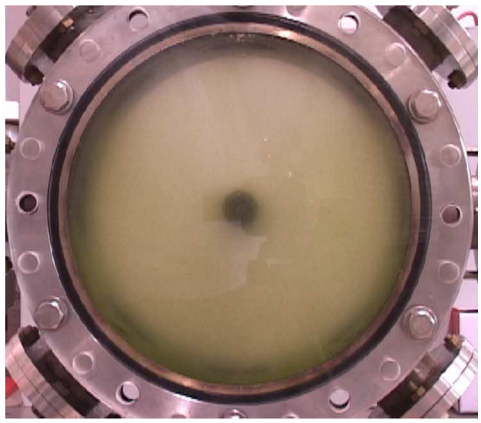


Fig. 7. Discharge appearance in the chamber.

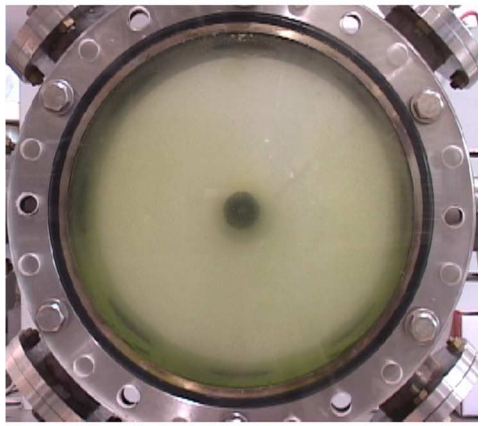
III. Experimental Results and Discussions

A. Discharge in the Sample Water

Fig. 5 shows the photograph of the discharge chamber soon after it was filled with the sample water from Hikawa dam. It can be seen that the colonies of *M. aeruginosa* were suspended in the entire discharge chamber. The colonies gradually float up to the upper side of the chamber when it is kept still for about 30 min. It should be noted that the pulsed streamerlike discharge was applied to the sample water at the very moment when it was filled in the chamber in this experiment. Fig. 6 shows the waveforms of the applied voltage to and discharge current through the discharge chamber with 160-kV and 500-A peak values, respectively. The input energy into the discharge chamber was calculated at 83 J/pulse. Fig. 7 shows the appearance of a single discharge in the tap water, with a conductivity of 11.6 mS/m. It was confirmed that the streamerlike discharges spread from the positive needle electrode toward the ground cylinder electrode and did not shift to an arc discharge. The diameter of the discharge area was about 100 mm. Fig. 8 shows the time dependence of the appearances of the sample water after a single discharge application. It can be seen that the colonies of *M. aeruginosa* cells gradually sank to the bottom of the discharge chamber. Most of the *M. aeruginosa* colonies arrived at the bottom after about 2 h. The temperature of the sample water did not change during the experiment.



(a)



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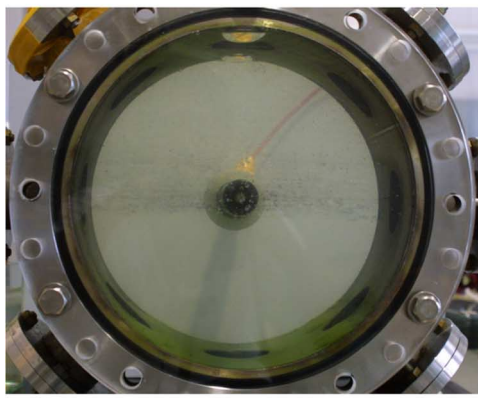

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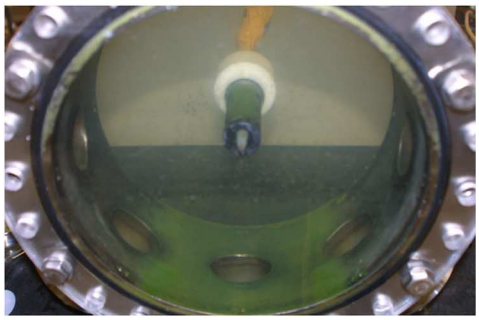
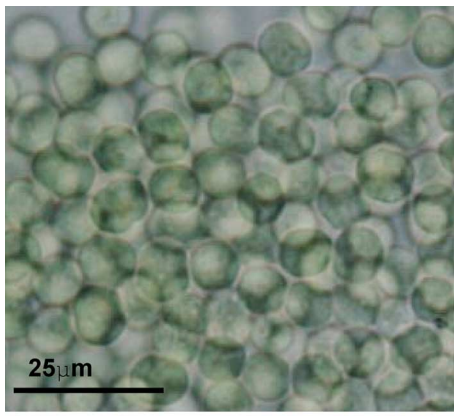
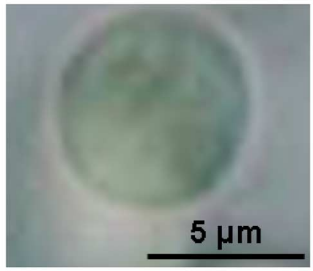


(d)

Fig. 8. Time dependence of the sample water after applying discharge. (a) 30 min. after discharge; (b) 60 min. after discharge; (c) 120 min. after discharge; (d) 120 min. after discharge.



(a)



(b)

Fig. 9. *M. aeruginosa* cells appearances after applying discharge. (a) Color appearance of the cells; (b) Single cell appearance.

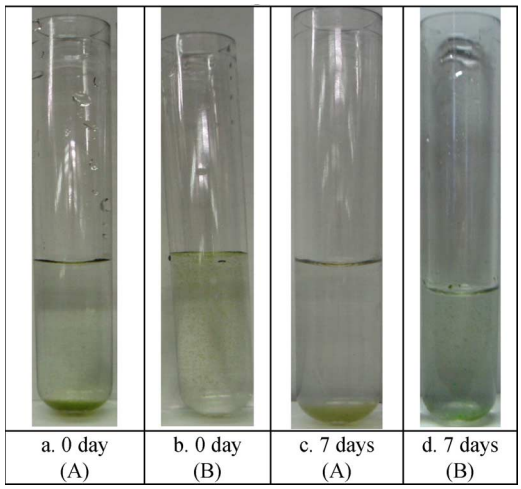


Fig. 10. Cultured *M. aeruginosa* cells with and without applying discharge. (a) and (b) Appearances of the cultured cells at the very moment when the cells were put in the medium. (c) and (d) Appearances of the cultured cells after one week, where "A" means after applying discharge and "B" means without applying discharge.

B. Diagnosis of M. aeruginosa

In order to diagnose, a part of the discharge-treated *M. aeruginosa* cells were collected from the bottom of the discharge chamber, and the microscopical observation was carried out. Fig. 9 shows the bright-vision photomicrographs [Fig. 9(a) with $\times$400 magnification and Fig. 9(b) with $\times$1000 magnification]. In the result of the comparison of Fig. 3 with Fig. 9, it can be seen that the GVs in the *M. aeruginosa* cells disappeared after applying discharge because GVs were collapsed by the pulsed

streamer discharge. At the same time, the *M. aeruginosa* cells with and without applying discharge were cultured in 24 °C BG-11 medium, with shaking and 24-h fluorescent shining, respectively. Fig. 10(a) and (b) shows the appearances of the discharge chamber at the very moment when the cells were put in the cultured medium. Fig. 10(c) and (d) shows the appearances of the cultured cells after one week, where "A" means with applying discharge and "B" means without applying discharge. It was observed that *M. aeruginosa* cells, after applying discharge, have changed color from green to yellow after one week. It can be said that the cells were killed and became rotten. In contrast, the cells, without applying discharge, were still living after one week. There are two possible reasons of the death of the *M. aeruginosa* cells after discharge. One is the streamer discharge, which led to the death of the cells. The other one is the discharge which weakened the cells, and the cells were catabolized by other bacteria or plankton in the sample water.

C. Discussion

It is well known that the pulsed streamerlike discharge in water can cause four physical phenomena such as the intense discharge current from the needle electrode to the ground, the free radical formation in the plasma, the ultraviolet radiation, and the shockwave generation [6]. The main impact factors in the *M. aeruginosa* treatment experiment should be considered. In this paper, a single streamer discharge was applied to the discharge chamber filled with sample water. It is obvious from Fig. 7 that the discharge did not spread to the entire space of the discharge chamber. The free radicals formation, such as OH^-, H^-, O^-, and HO_2^-, were considered to cause effect insignificantly. Because these radicals are formed by high-energy state electrons in the streamer head, at the same time, they can form product molecules H_2 and H_2O_2 or reform water. Therefore, these radicals cannot diffuse far away in the water column, and the UV radiation did not seem to play a role either. As a matter of fact, the method using UV radiation to eliminate bacteria from algal cultures has been reported [9]. Therefore, free radicals formation and ultraviolet radiation was not considered as the main factors in this paper. As a conclusion, the shockwave and discharge current due to the pulsed streamerlike discharge in water are the effective factors for the GVs collapsed, and the *M. aeruginosa* cells sank to the bottom of the discharge chamber.

IV. Conclusion

The effects of the pulsed streamerlike discharge in water on *M. aeruginosa* cells from the Hikawa Dam, Kumamoto, Japan were described in this paper. The following conclusions have been deduced.

1) After applying a single discharge, almost all the *M. aeruginosa* cells sank to the bottom of the discharge chamber since the GVs of the cells collapsed.
2) By comparison of the *M. aeruginosa* cells with and without applying discharge, which were cultured in BG-11 medium, respectively, the death of the cells after applying discharge was observed.
3) There exists a possibility that the pulsed streamerlike discharge will become one of effective options for cyanobacteria blooms treatment.

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References

[1] N. Scott, *Algae, Cyanobacteria and Water Quality*. Saskatchewan, Canada: Agriculture and Agri-Food Canada, 2002. Water Quality Matters.
[2] H. Hao, M. Wu, Y. Chen, J. Tang, and Q. Wu, "Cyanobacterial bloom control by ultrasonic irradiation at 20 kHz and 1.7 MHz," *J. Environ. Sci. Health, Part A, Toxic/Hazardous Substances and Environmental Eng.*, vol. 39, no. 6, pp. 1435–1446, 2004.
[3] S. Sato, *Report of "Project for Water Environment Renovation of Lake Kasumigaura*," The Science and Technology Promotion Foundation of Ibaraki. Section 2.2-(8), In Japanese. [Online]. Available: http://www.i-step.org/kasumi/project/joint_research/index.htm
[4] A. Howard, "Problem cyanobacteria blooms: Explanation and simulation modeling," *Trans. Inst. Br. Geogr.*, vol. 19, no. 2, pp. 213–224, 1994.
[5] A. Mlouka, K. Comte, A. Castets, C. Bouchier, and N. Marsac, "The gas vesicle gene cluster from microcystis aeruginosa and DNA rearrangements that lead to loss of cell buoyancy," *J. Bacteriol.*, vol. 186, no. 8, pp. 2355–2365, 2004.
[6] H. Akiyama, "Streamer discharge in liquids and their applications," *IEEE Trans. Dielectr. Electr. Insul.*, vol. 7, no. 5, pp. 646–653, Oct. 2000.
[7] A. Abou-Ghazala, S. Katsuki, K. H Schoenbach, F. C. Dobbs, and K. R. Moreira, "Bacterial decontamination of water by means of pulsed-corona discharges," *IEEE Trans. Plasma Sci.*, vol. 30, no. 4, pp. 1449–1453, Aug. 2002.
[8] I. Chorus and J. Bartram, *Toxic Cyanobacteria in Water: A Guide to Their Public Health Consequences, Monitoring and Management*. ISBN 0-419-23930-8C 1999, published on behalf of WHO by: F & FN Spon,11 New Fetter Lane, London EC4) 4EE.
[9] R. Mehta and K. Hawxby, "Use of ultraviolet radiation to achieve bacteria-free algal culture," in *Proc. Okla. Acad. Sci.,* vol. 57, pp. 54–60, 1977.

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