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Inactivation of microalgae in ballast water with pulse intense light treatment

Daolun Feng *, Jidong Shi, Dan Sun

College of Ocean Science and Engineering, Shanghai Maritime University, Shanghai 201306, PR China

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ABSTRACT

The exotic emission of ballast water has threatened the coastal ecological environment and people's health in many countries. This paper firstly introduces pulse intense light to treat ballast water. 99.9 ± 0.09% inactivation of *Heterosigma akashiwo* and 99.9 ± 0.16% inactivation of *Pyramimonas* sp. are observed under treatment conditions of 350 V pulse peak voltage, 15 Hz pulse frequency, 5 ms pulse width and 1.78 L/min flow rate. The energy consumption of the self-designed pulse intense light treatment system is about 2.90–5.14 times higher than that of the typical commercial UV ballast water treatment system. The results indicate that pulse intense light is an effective technique for ballast water treatment, while it is only a competitive one when drastic decreasing in energy consumption is accomplished.

1. Introduction

Ballast water is used to maintain the stability, trim and structural integrity of unladen ships (Hua and Liu, 2007). During voyage, ballast water is frequently loaded into a ship in one port, and discharged in another exotic port, which poses substantial risk of introducing nonindigenous species (Drake et al., 2007). The introduced nonindigenous species may adversely affect ecological system, cause economical losses and threat people's health (Tsolaki and Diamadopoulos, 2010).

Therefore, in order to eliminate those negative impacts, the International Maritime Organization formulates "The International Convention for the Control and Management of Ship's Ballast Water and Sediments" (IMO, 2004). Subsequently, different treatment methods were proposed to inactivate viable microorganisms in ballast water (Tsolaki and Diamadopoulos, 2010). Normally, the treatment process consists of two stage treatment: firstly, physical solid–liquid separation techniques, mainly hydrocyclone and filtration that may be enhanced by chemical coagulation, are introduced to remove sands and viable microorganisms with large size. Subsequently, physical or chemical treatment methods are incorporated to inactivate the residual viable microorganisms in ballast water (Lloyd's Register, 2010). However, none of them can meet the needs of safety, environmental friendliness, easy maintenance, low cost and effectiveness simultaneously and strictly (Gregg et al., 2009; Nengye and Frank, 2012), and more concerns should be focused on the environmental soundness of the treatment methods (Bowmer and Linders, 2010).

Pulse intense light (PIL) is considered to be one of the most promising non-thermal sterilization techniques (Gómez-López et al., 2007; Oms-Oliu et al., 2010) in food industry. PIL technology use short time pulses (100–400 μs) with an intense broad spectrum between 100 and 1100 nm (Ferrario et al., 2013) to inactivate microorganisms. Even though the peak power of each pulse is high, the total pulse energy is relatively low because of its short duration (Barbosa-Canovas et al., 2011). In comparison with the traditional UV inactivation, PIL technique shows several extra advantages: firstly, no toxic substances in xenon lamp in contrast to that of mercury in standard UV lamps (Schaefer et al., 2007); secondly, the UV irradiance of xenon lamp is about three or four orders of magnitude higher than that of UV lamp (Schaefer et al., 2007), which means faster and stronger inactivation ability; thirdly, microorganisms that expose to pulse intense light exhibit no tailing to their survival curves (Dunn et al., 1997; Martínez et al., 2013).

The vast research on PIL technology is focused on the inactivation of bacteria, yeast, fungi, viruses in food, food container, food package, etc. (Gómez-López et al., 2007; Elmnasser et al., 2007; Oms-Oliu et al., 2010). Recently, degradation of organic compounds in wastewater (Baranda et al., 2012; Moreau et al., 2013)

* Corresponding author at: Shanghai Maritime University, 1550 Haigang Avenue, Pudong New District, Shanghai 201306, PR China. Tel.: +86 (021) 38284332; fax: +86 (021) 38284342.
E-mail address: fengdaolun@aliyun.com (D. Feng).

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and sterilization for final cleaning in hospital (Levin et al., 2013) are also researched.

While, to the best of our knowledge, there is no research on applying PIL technique to treat ballast water. For ballast water that undergoes a typical pre-filtering treatment is colorless and transparent, which is an ideal liquid for PIL inactivation (Barbosa-Canovas et al., 2011; Gómez-López et al., 2007; Krishnamurthy et al., 2007). Therefore, PIL technique shows the potential to replace the widely used continuous UV treatment system to inactivate viable microorganisms in ballast water.

The work here is to introduce PIL technique into ballast water treatment. In addition, since there is no research on the inactivation of microalgae that is naturally different from previous investigated microorganisms (Gómez-López et al., 2007; Elmnasser et al., 2007; Oms-Oliu et al., 2010) and is the predominant species in ballast water (Steichen et al., 2012), two typical red tide microalgae: *Heterosigma akashiwo* and *Pyramimonas* sp. are chosen to test the treatment efficacy.

2. Materials and methods

2.1. Microalgae and culture medium

H. akashiwo and *Pyramimonas* sp. are purchased from Ocean University of China. *f*/2 medium is applied to culture both microalgae (Guillard and Ryther, 1962), while there is no $Na_2SiO_3{\cdot}9H_2O$ for *Pyramimonas* sp.

2.2. Pulse intense light inactivation experiments

Inactivation of microalgae is performed in a self-designed PIL inactivation set-up (Fig. 1). The experimental process is the following: Firstly, culture medium with *H. akashiwo* and *Pyramimonas* sp. at their logarithmic growth stage is diluted with artificial sea water, and stored in raw water tank as untreated ballast water. Then, dosing pump (GM0120PQ1MNN, Milton Roy Industrial (Shanghai) Co., LTD, China) is started to drive the untreated ballast water into a cylindrical treatment chamber with inner diameter of 4.15 cm and length of 20.80 cm, where the viable microalgae is inactivated by PIL generated from a xenon lamp (outer diameter 0.8 cm and lighting length 12.0 cm). The hydraulic retention time of the ballast water in treatment chamber is about 0.53 s for a flow rate of 1.74 L/min. Theoretically, turbulent flow is predominant within the treatment chamber. The xenon lamp is triggered by an ordered pulse power source. The treated water is finally collected by the treated water tank. Samples are taken from two points to test the viable microalgae concentration before and after inactivation. Also the treated water temperature is recorded by an OMEGA® thermometer (HH500RA, Omegadyne, Sunbury Ohio).

2.3. Determination of viable microalgae concentration

Flow Cytometry (Cyflow Cube 6, Partec GmbH, Münster, Germany) is used to determine the low viable microalgae concentration in ballast water accurately. A Guava® ViaCount® reagent (Guava Technologies Inc., Millipore, USA) is used to distinguish the viable and non-viable cells, nucleus-containing debris, and other impurities based on the differential permeability of DNA-binding dyes in the ViaCount® reagent. 10 mL water sample is added into 40 mL centrifugal tube and is well agitated to keep it homogeneous. Subsequently, 400 μL homogeneous sample and 400 μL Viacount® reagent are added into a sample tube, and is well agitated. After 30 min staining in the dark and under temperature below 10 °C, the stained sample is diluted by 200 μL 0.2 μm-filtered sea water, and then numerated by flow cytometry.

2.4. Calculation of inactivation percentage

The inactivation percentage is calculated by the following equation:

$$\text{Inactivation percentage}\ (\%) = (I - D)/I \times 100\%$$

where I is the concentration of viable microalgae before inactivation (cell/mL) and D is the concentration of viable microalgae after inactivation (cell/mL).

2.5. Calculation of energy consumption

The energy consumption (W_s, J/m^3) for PIL inactivation is calculated by the following equation:

$$W_s = (F \times f)/Q \times 60 \times 1000$$

where F is the input single-pulse power (J/pulse), f is the pulse frequency (Hz) and Q is the volumetric flow rate (L/min).

2.6. Statistical analysis

All experiments are performed in triplicate and data are expressed as mean ± standard deviation (SD). Analysis of SD is performed using OriginPro 7.5 software.

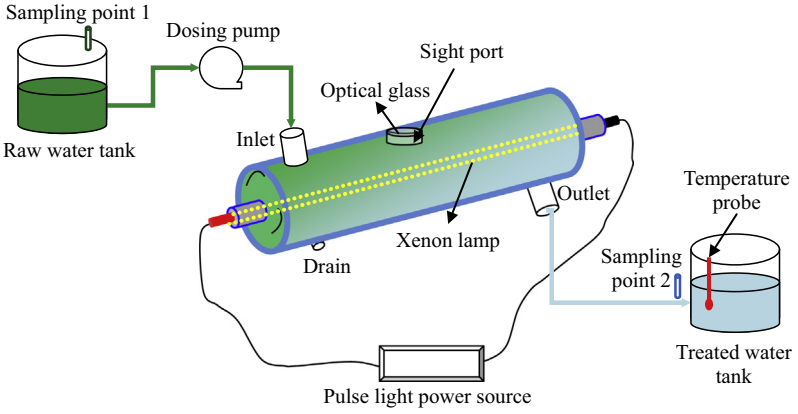

Fig. 1. Schematic of pulse intense light inactivation set-up.

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