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Evaluation of candidate biocides to control the biofouling Asian clam in the drinking water treatment industry: An environmentally friendly approach

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ABSTRACT

The biofouling Asian clam *Corbicula fluminea* greatly affects freshwater-dependent facilities in Europe and North America, including in the Great Lakes region. As chlorination has become increasingly restricted, finding alternative control agents is a priority. Due to the species' epifaunal nature, the body of knowledge on *Dreissena polymorpha* is larger than that on the Asian clam, and there is a tendency to assume that mitigation methods should work similarly for both species. However, this generalisation is inaccurate, and the optimisation of Asian clam control relies on species-specific toxicological data. This paper reports information on the potential of three candidate biocides for *C. fluminea* control: (i) the cationic polydiallyldimethylammonium chloride (polyDADMAC), (ii) potassium chloride and (iii) aluminum sulphate. While these chemicals may be employed in a range of contexts, they are particularly suitable for the highly regulated drinking water industry. LT_{50} values ranging from 284.3 h, for polyDADMAC applied at 10 mg/L, to 855.1 h, for an aluminum sulphate concentration of 11 mg/L, were obtained. Ecotoxicological standard tests with *Pseudokirchneriella subcapitata* and *Daphnia magna* suggested that, amongst the three biocides, potassium chloride is the one representing lower potential environmental hazard (with, for example, 48 h-EC_{50} higher than 1 g/L for *Daphnia* immobilisation, which compares to values of 20.2 and 112.5 mg/L for polyDADMAC and aluminum sulphate, respectively). The three chemicals are promising control agents at dosages compatible with waterworks' operational requirements with polyDADMAC having the highest biocidal activity, but also posing more environmental risks.

Introduction

The freshwater Asian clam (*Corbicula fluminea*) is a damaging invasive species and a biofouling organism of great concern (DAISIE, 2008a). Native to Southern/Eastern Asian and African aquatic systems, the species began to disperse across North America in the first half of the 20th century and reached Europe in the early 1980s (DAISIE, 2008a; Mackie and Claudi, 2010). It is well established in both continents, including in the Great Lakes region, but it continues to spread.

In addition to the negative impacts on the infested ecosystems (Sousa et al., in press), the Asian clam greatly affects freshwater-dependent industries (Mackie and Claudi, 2010; Pimentel et al., 2005). Drinking water treatment plants, power stations and irrigation systems are especially vulnerable to the species' biofouling activity (Isom, 1986; Rosa et al., 2011). For instance, these small clams, generally of no more than 3.5 cm in the adult stage, have been reported to be responsible for shutting down large power plants in the USA (Isom, 1986). Some of the problems experienced by industrial facilities as a result of *C. fluminea* infestations include pipe and equipment blockage, reduced efficiency of water cooling systems, increased corrosion, safety hazards when fire protection units are affected, and plant operation disruption associated with the need for biofouling removal (Mackie and Claudi, 2010; Rosa et al., 2011). While the overall economic impacts of invasive bivalves are difficult to quantify, Pimentel et al. (2005) predicted that, in the USA, the annual losses for the Asian clam are in the billion dollar range. In Europe, significant economic losses have also been experienced, but the documentation of the problem is scarcer compared to North America (Jenner et al., 1998; Rosa et al., 2011).

Several control methods, with varying degree of effectiveness, may be applied to manage *C. fluminea* in the industrial environment (Mackie and Claudi, 2010). Chemical treatment, involving the use of biocides, is the central to most control programmes. It tends to be more economical, efficient and versatile than alternative methods (e.g. the use of filters, physical cleansing) as it can be easily applied in existing facilities and allows the protection of the entire system against a range of biofouling organisms.

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Until not long ago, chlorine was the control agent traditionally selected, but it has been losing favour with regulators due to the generation of carcinogenic and ecotoxic trihalomethanes and the impacts on non-target biota (AISE, 2009; Emmanuel et al., 2004; Fisher et al., 1999; Mackie and Claudi, 2010; Maugh, 1981). As a result, the search for alternative biocides for Asian clam control has been pursued and is of major interest (Belanger et al., 1991; Bidwell et al., 1995; Mackie and Claudi, 2010; McMahon and Lutey, 1988; McMahon et al., 1993).

Very often *C. fluminea* coexists with the zebra mussel *Dreissena polymorpha*, another damaging invasive freshwater bivalve with similar ecological and economic impacts (DAISIE, 2008b; Mackie and Claudi, 2010). Furthermore, zebra mussel's epifaunal nature exacerbates its activity as a biofouler, turning it into the primary focus of many research initiatives, and thus the data and literature concerning this species are considerably more vast. For these reasons, when looking for alternatives to chlorine-based control, there is a general trend to assume that management practices and mitigation methods that work for the zebra mussel should perform similarly for the Asian clam. However, the two bivalves differ in key features of their biology (Karatayev et al., 2005), and hence such a generalisation can be inaccurate. For example, *C. fluminea* has been reported to be more tolerant than *D. polymorpha* to the nonoxidising surfactant dodecylguanidine hydrochloride/n-alkyl dimethylbenzyl ammonium chloride (DGH/QUAT), cadmium, zinc and potassium dihydrogen phosphate (Bidwell et al., 1995; Fisher et al., 1991; Marie et al., 2006). The opposite order of sensitivities was found for exposures to n-alkyl-dimethyl benzyl ammonium chloride (polyquat) and 2-(thiocyanomethylthio) benzothiazole (TCMTB) (McMahon and Chase, 1997; McMahon et al., 1993). The different sensitivities of the two bivalves should thus be a key premise for the development of chemical treatments. Only by building on species-specific toxicity data it is possible to optimise control with cost-effective dosing and minimal environmental impacts.

The aim of the present study is to provide information on the potential of a series of candidate biocides for *C. fluminea* control, and thus contribute to the broadening of the toolbox available for pest managers to specifically deal with the species. While the biocides assessed here may be useful in various industrial scenarios, special attention was given to the highly regulated drinking water treatment facilities. In these facilities, pest control is particularly challenging due to the strict limitations on chemical dosing to ensure the quality and safety of potable water. In this context, polydiallyldimethylammonium chloride (polyDADMAC), aluminum sulphate and potassium chloride were selected for evaluation. These chemicals have already proven to be effective against the zebra mussel (Costa et al., 2011; Mackie and Claudi, 2010; Mackie and Kilgour, 1995; Waller et al., 1993; Wildridge et al., 1998), and they are promising for use in waterworks because their presence in potable water has been generally regulated (CSN EN, 2004, 2008; EC, 1998; WHO, 2011). Moreover, polyDADMAC and aluminum sulphate are attractive control agent candidates for the drinking water treatment industry because they can be employed for treatment purposes (coagulation/flocculation) other than pest mitigation, with clear advantages coming from the use in the system of multiple function chemicals.

An integrated approach to the assessment of alternative biocides benefits from the early consideration of the chemicals' effects on non-target biota, with environmental safety being a key requirement for registration and licensing (EC, 2006). From this perspective, the candidate control agents were evaluated not only in terms of their performance against the Asian clam, but also regarding their environmental selectivity. Short-term exposures of the green microalga *Pseudokirchneriella subcapitata* and the cladoceran zooplankter *Daphnia magna* were carried out, focusing on growth inhibition (OECD, 2011) and immobilisation (OECD, 2004), respectively. These species are recommended as part of the test battery generally adopted in early stages of xenobiotic risk assessment (EC, 2002). Besides being recognised as potentially sensitive non-target species, microalgae and daphnids are key representative species regarding trophic regulation within freshwater food webs which supports their use as indicators to address the likelihood of environmental impairment.

The toxicological studies presented in this paper have direct practical relevance for the optimisation of Asian clam control programmes. The data provided will be particularly useful for the drinking water treatment facilities operating in the North American Great Lakes region, threatened by the pest (OISAP, 2013; Waterworks, New York State Federation of Lake Associations, Inc., 2011).

Material and methods

Chemicals

PolyDADMAC, a high charge density cationic homopolymer of diallyldimethylammonium chloride, was dosed as Floquat FL 4440 (SNF AMBIENTAGUA, Santo Tirso, Portugal), a product containing 40% (w/w) of the polymer. Potassium chloride was supplied by Merck (Darmstadt, Germany) as laboratory-grade reagent 99% (w/w) purity. Aluminum sulphate was dosed as a commercial solution 8.2% (w/w) alumina (QuimiTécnica, Estarreja, Portugal).

Test organisms

During the winter, adult *C. fluminea* individuals were collected from a canal in Mira, Portugal (N40°25′06.90″/W8°44′13.18″), where a well-established population (density above 2000 clams/m^2) is available. Clams were collected by sieving sediment into a 1-mm mesh bag. Individuals with shell length in the range 20–30 mm were selected and immediately transported in field water to the laboratory, where they were gradually acclimated to and kept until use in dechlorinated municipal water, at 20 ± 2 °C, under a 16:8 h light dark photoperiod cycle and continuous aeration. The water was fully renewed once a week, and the clams were fed ad-hoc with *P. subcapitata* suspensions.

P. subcapitata was cyclically cultured in non-axenic batch culture in synthetic MBL-Woods Hole medium, at 20 ± 2 °C, under permanent illumination (Stein, 1973). Monoclonal cultures of *D. magna* (clone A, sensu Baird et al., 1989a) were reared in synthetic ASTM hard water medium (ASTM, 1980), supplemented with a standard organic additive (Baird et al., 1989b) and vitamins (Elendt and Bias, 1990). Temperature and photoperiod were kept constant at 20 ± 2 °C and exposed to 16:8 h light dark photoperiod. *Daphnia* cultures were renewed three times per week and fed with *P. subcapitata* suspensions (3×10^5 cells/mL).

Molluscicidal activity against *C. fluminea*

C. fluminea was treated with polyDADMAC, potassium chloride and aluminum sulphate to determine the mortality profiles resulting from exposure to reference chemical concentrations. Treatments lasted until 100% mortality or 2 months in the cases when full mortality was not achieved. A replicated semi-static design was employed with a blank control plus two concentrations per chemical.

As both polyDADMAC and aluminum sulphate are licensed for potable water treatment, the respective test concentrations were set to comply with normal waterworks operation. In the case of polyDADMAC, the maximum allowed treatment concentration (10 mg/L as generally established by CSN EN (2008) and set by DWI (2012) for the United Kingdom in particular) and the concentration typically employed (2 mg/L; CSN EN, 2008) were tested. Regarding aluminum sulphate, the treatment concentration generally depends on raw water quality, with no legal maximum being imposed (CSN EN, 2004; DWI, 2012). According to WHO (2011), a health-based limit of 0.9 mg/L could be derived for residual aluminum in drinking water, but this value largely exceeds those achieved under normal optimised operating conditions. In this study, aluminum sulphate concentrations of 11 and 18 mg/L were tested. These fall into the common concentration range in waterworks — generally not less than 1 mg/L (CSN EN, 2004) and more often 2 to 5 mg/L (WHO, 2011) as aluminum. Potassium chloride is not part of typical waterworks operation, and hence the respective