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Research Article

Zebra mussel control using Zequanox® in an Irish waterway

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Abstract

Due to the invasion of zebra and quagga mussels in European and North American waters, there is a need for an environmentally benign mussel control method to replace chlorine and other currently used control products. Zequanox® is a natural product comprised of *Pseudomonas fluorescens* strain CL145A, which effectively controls zebra and quagga mussels. The objective of this study was to demonstrate an effective method of zebra mussel control in inland waterways using Zequanox. Water quality was monitored to determine any negative impacts and to observe product dispersion. A curtain made of an impermeable material was placed in the Grand Canal at Tullamore Harbour sealing off two 8 × 0.5 m sections of canal wall under the bridge, and a control site was chosen further down the docking area. Both sections were treated with Zequanox at a concentration of 150 mg active substance/L for an 8 hour treatment period. Water quality was monitored in the treatment area and in the selected control area before, during, and after treatment. Naturally settled and seeded adult zebra mussels were observed for mortality in the treatment and control areas and juveniles were monitored for survival in both the treatment and control areas. Naturally settled adult mussel numbers were reduced by approximately 46% in treatment side 1, and 65% in treatment side 2, seeded adult mussel mortality reached 75% in treatment side 1 and 56% in treatment side 2. These results demonstrate that under the optimum conditions Zequanox effectively controls zebra mussels in open water.

Key words: Grand canal, invasive mussel control, water quality

Introduction

The zebra mussel, *Dreissena polymorpha* (Pallas, 1771), is an invasive, aquatic bivalve shellfish, which has impacted freshwater ecosystems and water abstraction in all invaded countries including Ireland (Minchin et al. 2002; Lucy 2010; Lucy et al. 2013). The zebra mussel arrived in Ireland in the early 1990's (Minchin and Moriarty 1998) in the lower River Shannon on the hulls of boats, most likely attached to used leisure crafts from Britain (Pollux et al. 2003). Inland waterway systems (canals) in Ireland have allowed for movement of the zebra mussel both of its own accord and by accidental movement, largely attributed to boaters and recreational anglers (Minchin et al. 2005). Not only is the zebra mussel causing problems for Ireland's rivers and lakes through their role as ecosystem engineers (Karatayev et al. 2002), but industries are also suffering from the high costs of controlling these mussels (Aldridge et al. 2004). Currently chlorine is the most commonly used control method (Mackie and Claudi 2010); however, its use is limited and is only suitable in enclosed systems (intake pipes) as it is a non selective general biocide and is lethal to all living organisms. Presently the only control method for zebra mussels in inland waterways is physical removal, and therefore, there is a need for a more efficient management option.

Marrone Bio Innovations (MBI), a company specialising in the development and commercialisation of natural biocides in Davis, CA, USA, is the commercial license holder for the invasive zebra and quagga mussel (dreissenid) control product Zequanox®. The active ingredient in Zequanox is killed *Pseudomonas fluorescens* strain CL145A cells, which is lethal to dreissenid

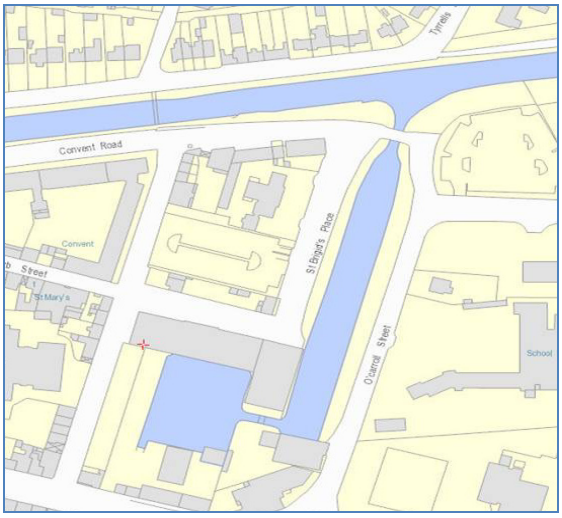

Figure 1. Tullamore Harbour, Co. Offaly, Ireland.

Mussels, but studies show it has minimal to no impact on other aquatic organisms (Molloy et al. 2013a). *Pseudomonas fluorescens* is present world-wide and commonly found in food. In nature, it is a harmless bacterial species that is known to protect the roots of plants from disease (Marrone Bio Innovations 2012). Ecotoxicology studies were carried out in the Institute of Technology Sligo and in the USA, where Zequanox was tested on a number of aquatic species. No negative effects were observed at concentrations required to sufficiently control zebra mussels (150 mg active ingredient/L) (Marrone Bio Innovations Ecotoxicology Studies 2012). Additionally, Molloy et al. (2013b) carried out a number of non target trials using the active ingredient in Zequanox (*Pseudomonas fluorescens* CL145A) and again found no negative impacts to the organisms tested at concentrations required to control zebra mussels.

In March, 2012 the United States Environmental Protection Agency registered Zequanox for use in the USA in enclosed or semi-enclosed systems. In 2011, successful Zequanox trials were conducted within the cooling water system of Davis Dam in Bullhead City, Arizona in the USA, and in 2012 within the cooling water system of DeCew II Generating Station of Ontario Power Generation in St. Catharine's, Ontario, Canada. MBI also conducted a successful open water trial in Deep Quarry in DuPage County, Illinois, USA in 2012; this open water trial was similar to the canal trial described in this report.

Figure 2. Impermeable curtains used to hold treated water within treatment area along canal walls. Photograph by Sara Meehan.

Tullamore Harbour is part of the Grand Canal, connecting the east of Ireland to the Shannon River navigation in central Ireland. It was traditionally used for transporting goods via barge boats, and now is solely used for leisure purposes (Byrne 2007). The Grand Canal at Tullamore Harbour has a zebra mussel infestation spanning from under the bridge, along the harbour branch of the canal, and into a harbour and dock area (Figure 1).

A pilot demonstration trial using Zequanox was conducted under the bridge in the Grand Canal at Tullamore Harbour. The objectives of this trial were to firstly demonstrate an effective method of zebra mussel control in inland waterways and secondly trial a method which could be used for zebra mussel fouled jetties, pontoons and navigational structures.

Materials and methods

Experimental set-up

This trial was conducted under the bridge at Tullamore Harbour (53°27'82"N, -7°48'86"W) where dreissenid infested canal walls on both banks were treated with Zequanox to test its effect on settled juveniles, seeded adult mussels and naturally settled adult mussels. The areas of the canal were labeled treatment side 1, treatment side 2 (treated areas under the bridge) and control. Two impermeable curtains were set up to enclose the treatment area (canal wall). These curtains were comprised of an impermeable material (scaffband), which was weighted down with stainless steel chains at the bottom and attached to aluminum at the sides, with foam used to seal in the containment area (Figure 2).

Figure 3. PVC plates used to monitor juvenile survival. Photograph by Sara Meehan.

The curtains were on average 7.70 m in length, 0.45 m in width and 1.31 m in depth, so that approximately 4.5 m^3 (4500 L) of water was enclosed along each concrete wall. The curtains were set up one day in advance of treatment to allow the mussels to acclimatise and resume normal feeding behavior prior to treatment.

The infested canal walls under the bridge at Tullamore Harbour were treated with Zequanox at a target concentration of 150 mg active substance (a.s.)/L (active substance is synonymous with active ingredient). The target concentration was maintained for 8 hours. This treatment concentration and duration was based on the results of trials carried out in North America and in Ireland (Meehan et al. 2013).

Juvenile mussel collection

PVC plates were deployed in Lough Key (53°593'04"N, 08°16'46"W) on July 23rd, 2012 to gather juvenile zebra mussel settlement, as this lake is known for high settlement (Lucy 2005). These plates were removed from Lough Key on September 2nd, 2012 and an initial baseline count was made. These plates were then transported to the Grand Canal at Tullamore Harbour and placed in the two treatment areas and the control area on weighted rope (Figure 3). Juvenile plates were counted 24 hours after treatment then daily followed by weekly until juvenile settlement reached zero.

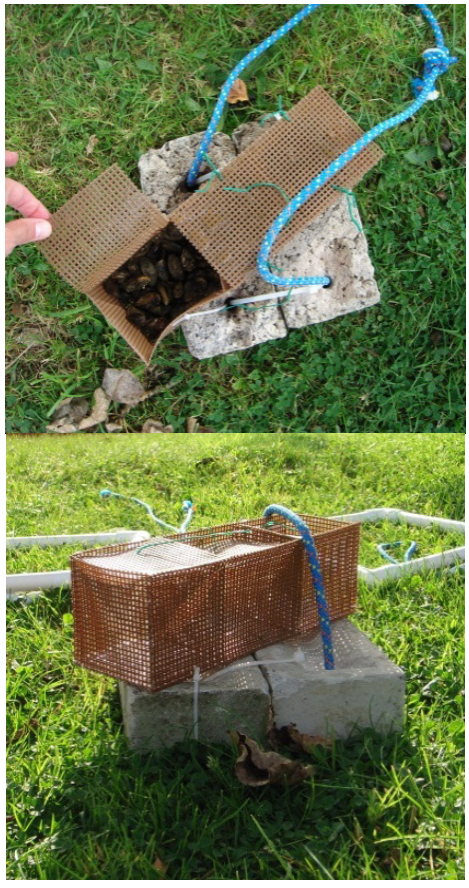

Figure 4. Mesh cages to hold seeded adult mussels (26 cm in length). Photograph by Bridget Gruber.

Adult mussel collection

Adult zebra mussels were collected from the Grand Canal at Tullamore via a long-handled scraper (Minchin 2007; Minchin et al. 2002) and by hand removal from the wall while wading. Healthy mussels were then seeded into three mesh cages (mesh size 3mm), each containing three compartments housing 50 mussels each (Figure 4). These mesh cages were attached to bricks via cable ties. Floating rope was then tied to the bricks so the cages could be easily removed from the canal using a boat hook; this method was developed so the cages would not be visible to the public as they were to remain in the

canal for an extended period of time. Once the mesh cages were ready, they were left to acclimatise overnight in the canal. One cage was placed in the control area, and one in each treatment area. Mussels were checked for mortality before treatment and any dead ones were removed and replaced with live healthy ones. Mussels were presumed dead if shells were open and did not close after being gently prodded. After treatment seeded adult mussels were counted first daily then weekly for seven weeks.

Naturally settled adult mussels

The number of naturally settled adult mussels in the two treated areas and the control area was estimated prior to treatment using 25 cm × 25 cm quadrats. Three quadrats per defined area were used to estimate mussel settlement/m^2. Quadrats were placed at random and at different depths by divers. Divers counted the number of live mussels within each quadrat. A record of the exact spot the quadrats were placed was kept by measuring its distance from a pre-determined point along the bank and the depth at which the quadrat was placed. Photographs were also taken so that the same quadrats could be counted again after treatment. Quadrats were re-counted seven weeks after treatment.

Zequanox application

The curtains were placed in the canal 24 hours prior to treatment to allow the naturally settled mussels to resume normal behavior after the disturbance of the curtain placement. Twenty four hours after the curtains were placed in the canal (before treatment), dissolved oxygen (DO) inside the curtained areas had significantly reduced and was approximately 3 mg/L lower than the DO outside of the curtains. This was likely due to the natural diurnal cycle and flow restriction. Therefore, treatment side 1 was aerated with bubblers until the curtains were removed to ensure DO stayed at background levels, whilst on treatment side 2, DO was not controlled and no aeration occurred. This experimental design allowed us to quantitatively determine if observed mortality could be attributed to Zequanox, or whether the observed mortality could be attributed to low DO levels. It also allowed us to infer if water quality conditions impacted zebra mussel ingestion of Zequanox.

Zequanox, a dry powder formulation (as registered in the US), was used to treat the canal walls. The powder was mixed on-site with canal water to create the following stock solution concentration:

$C_1V_1 = C_2V_2$ where
C_1 = target treatment concentration (mg a.s./L)
V_1 = volume of treatment area (4500 L)
C_2 = stock concentration (100 g a.s./L)
V_2 = volume of stock concentration to be applied (L)

For each curtained off area a total of 675 g a.s. of Zequanox was mixed with 6.75 L of canal water using a small hand blender to achieve a concentrated product solution of 100 g a.s./L. This solution was slowly poured into the curtained off area so as to evenly distribute the product. Once all of the product was in the water, a wooden paddle was used to gently mix the treated water to achieve an even distribution of product within the treated area. As turbidity and treatment concentration have a linear relationship (Meehan et al. 2013), turbidity inside the curtains was monitored throughout the application process using a Hach 2100Q portable turbidimeter to ensure the target concentration was reached and maintained.

As flow in the canal increased, nominal leakage of product from within the curtain occurred and concentrations within the treatment area decreased. This leakage likely occurred due to an increase in wind speed or the passing of a barge along the canal. In order to maintain a target concentration of 150 mg a.s./L, additional product was mixed in two stages and added.

After the 8 hour treatment period in which Zequanox concentrations were maintained at 150 mg a.s./L, the curtains were then held in place for a further 16 hours (but no additional product was added) making the hold time 24 hours in total. This additional hold time allowed for natural degradation of the product. Studies indicate that, once Zequanox is wetted, it biodegrades rapidly and the efficacy significantly decreases after 8 hours in water, and after 24 hours in water it is no longer efficacious. After the 24 hour hold time, the curtains were removed and, based on water quality measurements, the product dispersed to non-detectable levels within the canal system.

Water quality measurements

Turbidity inside the treatment area was monitored throughout the application and post-treatment period with a Hach 2100Q portable turbidimeter;

as turbidity and concentration are correlated this ensures that the target concentration was reached and maintained throughout the application period, and that Zequanox had dispersed to non-detectable levels after the curtains were removed.

Additional water quality measurements were taken before treatment, during treatment (at 4 and 8 hours), 24 hours after treatment before the curtain was removed, and 24 hours after the curtain was removed. These water quality measurements included: temperature, dissolved oxygen (DO), pH, turbidity, biological oxygen demand (BOD), and total organic carbon (TOC).

Dissolved oxygen, pH and temperature were measured with an Orion 5 star meter. The analysis of BOD and TOC was subcontracted out to Alcontrol Laboratories. Method 5210B, AWWA/APHA, 20th Ed., 1999; SCA Blue Book 130 was used to determine BOD. US EPA Method 415.1 and 9060 was used to determine TOC.

Results

Juvenile mussels

Figure 5 and Table 1 show the mean juvenile counts for the treatment and the control areas. Juvenile numbers were high (over 8,000/m^2) 48 hours in advance of the trial. Between 48 hours and the first count carried out after treatment, survival dropped considerably for both the treated juveniles and the control juveniles. After this initial drop, juvenile survival in the treated areas continued to decrease, while juvenile survival in the control area stayed approximately the same between 05/09/12 and 07/09/12.

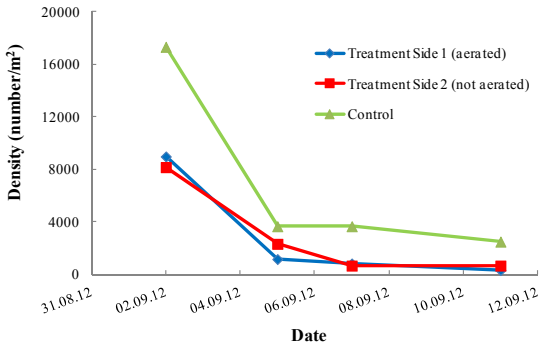

Figure 5. Mean density of juveniles before and after Zequanox treatment.

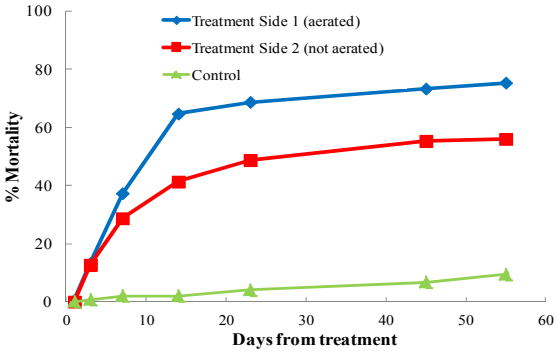

Figure 6. Seeded adult mussel mortality after treatment with Zequanox.

Adult mussels

Seeded adult mussels

After 55 days, treatment side 1 had 75% seeded adult mussel mortality and treatment side 2 had 56% mortality. The mortality in the control was 9% (Figure 6).

Naturally settled adult mussels

Table 2 shows the mean number of naturally settled mussels before and after treatment within the treatment areas and the control. The mean number of live adult mussels decreased by approximately 46% in treatment side 1, and by approximately 65% in treatment side 2. The mean number of live mussels decreased by 15% in the control area (there was one less mussel observed in the control area after treatment).

Water quality

In treatment areas 1 and 2 the temperature ranged from 17.8 to 18.6°C; in the control area the temperature ranged from 17.1 to 19.6°C (Table 3). In treated areas, pH varied between 7.58 and 8.03, similar to the range seen in the control area (7.76–7.88). Dissolved oxygen in treatment side 1 (aerated side) ranged from 5.6 to 7.68 mg/L. Dissolved oxygen levels in treatment side 2 (not aerated) ranged from 2.38 to 7.58 mg/L. In treatment side 2, 24 hours after treatment, DO dropped to 2.38 mg/L. Once the curtain was removed DO levels increased to 7.58 mg/L (background levels). Biological oxygen demand in the treated areas ranged between < 2 and 103 mg/L. Total organic carbon ranged from 20.7 to 49.5 mg/L. The turbidity in the treated areas before treatment was < 3 NTUs. During treatment, the turbidity in

Table 1. Mean density of juveniles before and after Zequanox treatment with standard deviation (juveniles/m^2).

Date	Treatment Side 1 (aerated)	SD	Treatment Side 2 (aerated)	SD	Control	SD
02/09/2012	8983	4820	8167	2593	17333	3300
05/09/2012	1167	236	2333	943	3666	2828
07/09/2012	833	236	667	0	3667	0
11/09/2012	333	471	667	471	2500	1179
% Survival	4		8		14	

Table 2. Mean density of naturally settled adult mussels (live adult mussels/m^2) before and after Zequanox treatment with standard deviation (SD).

Date	Treatment Side 1 (aerated)	SD	Treatment Side 2 (aerated)	SD	Control	SD
03/09/2012	1000	662	272	136	69	37
22/10/2012	539	272	96	34	59	24
% Mortality	46		65		15	

Table 3. Water quality measurements before, during (4 and 8 hours) and after treatment (before and after curtain removal).

Sample Date & Time	Location	Turbidity (NTU)	Temp (°C)	BOD (mg/l)	TOC (mg/l)	pH	DO (mg/l)
Before treatment							
4-Sep, 09:30	Control	4.97	17.1	<2	22.8	7.76	7.22
04-Sep, 09:30	Treated 1	2.72	17.8	<2	21.7	8.03	5.6
04-Sep, 09:30	Treated 2	2.97	17.9	<2	20.7	7.82	3.84
4 hrs into treatment							
04-Sep, 14:00	Control	4.26	19.3	<2	21.6	7.85	8.2
04-Sep, 15:00	Treated 1	109	18.4	91.5	49.5	7.59	7.15
04-Sep, 14:10	Treated 2	125	18.2	71	43.3	7.68	4.29
8 hrs into treatment							
04-Sep, 18:09	Control	3.74	19.6	<3	21.1	7.83	8.44
04-Sep, 18:56	Treated 1	127	18.5	103	31.9	7.93	7.63
04-Sep, 18:20	Treated 2	59.9	18.6	28.6	23.1	7.85	5.08
24 hrs after treatment; before curtain removal							
05-Sep, 07:45	Control	8.78	17.9	<2	20.8	7.87	6.85
05-Sep, 08:00	Treated 1	26.5	18.3	13.1	22.3	7.84	7.68
05-Sep, 07:55	Treated 2	32.3	18.5	17.1	25.3	7.58	2.38
24 hrs after curtain removal							
06-Sep, 12:00	Control	5.12	17.9	3.2	21.6	7.88	7.18
06-Sep, 12:00	Treated 1	9.31	18	3.65	22.7	7.63	7.42
06-Sep, 12:00	Treated 2	8.19	18.1	<2	22.3	7.85	7.58

the treated areas increased and ranged between 59.9 and 127 NTUs. Approximately 24 hours after treatment, prior to curtain removal, turbidity decreased to 26.5 and 32.3 in the treated areas. Once the curtains were removed, within 24 hours, turbidity decreased to 9.31 and 8.19 NTUs. The turbidity of the control throughout the 48 hour monitoring period ranged from 3.74 to 8.78 NTUs.

Discussion

Juvenile mussel survival

Juvenile survival on the treated plates and the control plates initially declined after treatment. After this decline, control survival leveled out and survival on the treated plates continued to drop. There is no way to determine if any of the mortality during the initial decline in survival is due to Zequanox treatment therefore it must be assumed that it is due to outside influences namely the transportation of the plates to the treatment site. However the continued decline of settlement on the treated plates was due to Zequanox as the control survival was maintained. These results parallel studies conducted by MBI at Davis Dam (Arizona, US) where a decline in juvenile survival on settlement plates treated with Zequanox was observed, and a study carried out in Sligo, Ireland (a demonstration trial for a

water treatment plant) where juvenile survival after treatment with Zequanox decreased (Meehan et al. 2013). It is also important to note that seasonal plates are known to underestimate total natural settlement but are considered a good proxy (Lucy et al. 2005). The initial high mortality in both the treated and control plates is not representative of what would happen in a real time application as there would be no movement of settlement plates from one site to the other. Therefore further research is necessary to examine the effects of Zequanox on settled juveniles in situ.

Adult mussel mortality

Seeded mussel mortality was observed in treatment side 1 (aerated) and 2 (not aerated); however, mortality was greater on treatment side 1 (75%) than side 2 (56%). Several factors may have contributed to this difference. The lower DO levels on treatment side 2 may have disturbed the mussel's feeding, by causing them to shut their valves as a response to unfavourable conditions, as is the case with intermittent chlorination (Rajagopal et al. 2003). Zequanox must be ingested by the mussels to have an effect. Mixing and aeration may also have contributed to the difference in mortality, making Zequanox more bioavailable throughout the treatment area. On treatment side 2 only hand mixing aided in the distribution of the product whereas aeration on treatment side 1 may have helped to more evenly distribute Zequanox.

A decrease in naturally settled mussels after treatment with Zequanox was observed; however, in contrast to the seeded mussel mortality, more mortality occurred in treatment side 2 (65%) than in treatment side 1 (46%). This may have been due to the aeration bubblers and air tubing on treatment side 1 being located close to the wall thus disturbing the mussel's causing them to shut their valves and cease feeding. The seeded adult mussels on aerated side 1 were located at the bottom of the canal away from the direct interference of the aeration system and this could account for the difference in mortality between the seeded and naturally settled mussels.

Water quality

No negative impacts from Zequanox treatment to temperature or pH was observed. The temperature range seen in the treated and control areas was consistent with the natural diurnal and seasonal cycles in Ireland. The slightly higher temperatures in the control area was likely due to that area being in direct sunlight while the treated areas were under the bridge and therefore had less sun exposure. The difference in sunlight had no apparent impact on pH levels. The zebra mussels in this study (seeded and naturally settled) at all sites were present at depths of between 1.0–1.5 m and due to low water transparency were at naturally low light levels. In fact the divers required torch light to take samples on both sampling dates. Therefore sunlight is not considered a varying environmental factor in this study.

During treatment, the turbidity in the treated areas increased (since Zequanox is made up of organic material, turbidity was expected to increase significantly) and ranged between 59.9 and 127 NTUs. After treatment was terminated, but prior to curtain removal, turbidity, as expected, began to decrease due to natural degradation of the product. Once the curtains were removed, within 24 hours, turbidity dropped to control levels.

Aeration sufficiently controlled DO levels in treatment side 1. In treatment side 2, 24 hours after treatment, DO dropped to 2.38 mg/L. This was expected as Zequanox is comprised of dead bacterial cells that degrade in the natural environment causing a decrease in DO, particularly in low flow environments. However, once the curtain was removed and flow was restored, DO increased to background levels.

TOC increased in treated areas four hours into the treatment; however, by eight hours TOC levels were decreasing to background levels. This increase again was expected because Zequanox is primarily made up of particulate organic matter. TOC levels decreased as degradation of the product took place. Since Zequanox is organic in nature, biochemical oxygen demand also followed a similar pattern, increasing at 4 hours into treatment and then decreasing as time passed and Zequanox degraded.

Environmental monitoring before, during, and after treatment indicated there was minimal impact to water quality in the canal. Though TOC, BOD, and turbidity temporarily increased during treatment in the enclosed treatment areas, by 8 hours, measurements were decreasing and returned to background levels 24 hours after treatment once Zequanox had naturally biodegraded.

Conclusion

Presently the only zebra mussel control option for canals in Ireland is mechanical removal. This study shows that Zequanox effectively controlled up to 75% of zebra mussels in an Irish canal. Though Zequanox is not yet registered in the EU, it has potential as an alternative control option for Irish waterways; the results of the study show that when Zequanox is applied under the correct conditions (sufficient DO levels and minimal disturbance to the mussels) it can be an effective zebra mussel control method for inland waterways and structures.

Future recommendations for a similar trial would include aeration in all enclosures ensuring that the aeration occurs a sufficient distance from settled mussels so as to cause minimal disturbance. Also, settlement plates should be removed less frequently and allowed more time to acclimatise after plate transportation so as to avoid high levels of control mortality. This trial was the first canal treatment with Zequanox and the methods used here support further development of similar application techniques for static, contained, and open water treatments.

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References

Aldridge DC, Elliott P, Moggridge DG (2004) The recent and rapid spread of the zebra mussel (*Dreissena polymorpha*). *Biological Conservation* 119: 253–261, http://dx.doi.org/10.1016/j. biocon.2003.11.008

Byrne M (2007) The grand Canal and Tullamore. Offaly Historical and Archaeological society. http://www.offaly-history.com/articles/78/1/The-Grand-Canal-amp-Tullamore/Page1.html (Accessed 27 August 2013)

Karatayev AY, Burlakova LB, Padilla DK (2002) The impact of zebra mussels on aquatic communities and their role as ecosystem engineers. In: Leppäkoski E, Olenin S, Gollasch S (eds), Invasive aquatic species of Europe: Distribution, Impacts and Spread. Kluwer Scientific Publishers, Dordrecht, pp 433–446, http://dx.doi.org/10.1007/978-94-015-9956-6_43

Lucy FE, Burlakova LE, Karatayev AY, Mastitsky SE, Zanatta DT (2013) Zebra mussel impacts on unionids: A Synthesis of Trends in North America and Europe. In: Nalepa T, Schloesser D (eds), Quagga and Zebra Mussels. Taylor and Francis, pp 623–646

Lucy FE (2010) Zebra Mussels: Review of ecology and impacts since invasion in Ireland. In: Mackie G, Claudi R (eds), Monitoring and control of macrofouling molluscs in freshwater systems 2nd Edition. CRC Press, Boca Raton, FL., pp 386–389

Lucy F, Sullivan M, Minchin D (2005) Nutrient levels and the zebra mussel population in Lough Key. ERTDI Report Series No. 34. EPA, Wexford. Available at: http://www.epa.ie/downloads/pubs/research/water/epa_zebra_mussels_ertdi34_final.pdf (Accessed 11 December 2012)

Mackie GL, Claudi R (2010) Monitoring and control of macrofouling mollusks in freshwater systems, 2nd Edition. CRC Press, Boca Raton, FL, 508 pp

Marrone Bio Innovations (2012) http://www.marronebioinnovations.com/products/brand/zequanox/ (Accessed 16 Sept 2013)

Marrone Bio Innovations Ecotoxicology Studies (2012) http://www.marronebioinnovations.com/lightray/site/wp-content/uploads/2012/09/ZQ_EcotoxStudies_0413_web.pdf (Accessed 16 Sept 2013)

Minchin D (2007) Rapid coastal survey for targeted alien species associated with floating pontoons in Ireland. *Aquatic Invasions* 2: 63–70, http://dx.doi.org/10.3391/ai.2007.2.1.8

Minchin D, Lucy F, Sullivan M (2005) Ireland a new frontier for the zebra mussel *Dreissena polymorpha* (Pallas). *Oceanological and Hydrobiological Studies* 34 (Suppl. 1): 19–30

Minchin D, Lucy F, Sullivan M (2002) Zebra mussel: Impacts and Spread. In: Leppakoski E, Gollasch S, Olenin S (eds), Invasive Aquatic Species of Europe: Distribution, Impacts and Spread. Kluwer Scientific Publishers, Dordrecht, pp 135–146, http://dx.doi.org/10.1007/978-94-015-9956-6_15

Minchin D, Moriarty C (1998) Zebra mussels in Ireland. Fisheries Leaflet 177, Marine Institute, Dublin, 11 pp

Meehan S, Lucy FE, Gruber B, Rackl S (2013) Comparing Zequanox® and chlorine as zebra mussel control strategies in an Irish drinking water treatment plant. *Management of Biological Invasions* 4: 113–122, http://dx.doi.org/10.3391/mbi.2013.4.2.03

Molloy DP, Mayer DA, Giamberini L, Gaylo MJ (2013a) Mode of action of *Pseudomonas fluorescens* strain CL145A, a lethal control agent of dreissenid mussels (Bivalvia: Dreissenidae). *Journal of Invertebrate Pathology* 113(1): 113–115

Molloy DP, Mayer DA, Gaylo MJ, Burlakova1 LE, Karatayev AY, Presti1 KT, Sawyko PM, Morse JT, Paul EA (2013b) Non-target trials with *Pseudomonas fluorescens* strain CL145A, a lethal control agent of dreissenid mussels (Bivalvia: Dreissenidae) *Management of Biological Invasions* 4: 71–79, http://dx.doi.org/10.3391/mbi.2013.4.1.09

Pollux B, Minchin C, Van Der Velde G, Van Alen T, Van Der Staay SYM, Hackstein J (2003) Zebra mussels (*Dreissena polymorpha*) in Ireland, AFLP-fingerprinting and boat traffic both indicate an origin from Britain. *Freshwater Biology* 48: 1127–1139, http://dx.doi.org/10.1046/j.1365-2427.2003.01063.x

Rajagopal S, Van der Velde G, Van der Gaag M, Jenner HA (2003) How effective is intermittent chlorination to control adult mussel fouling in cooling water systems? *Water Research* 37: 329–338, http://dx.doi.org/10.1016/S0043-1354(02)00270-1