

Management strategies for the zebra mussel invasion in the Ebro River basin

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Abstract

Dreissena polymorpha has invaded seven rivers in the Ebro River basin (Spain) in the period 2001-2009. In this paper, we present a compilation of management strategies directed by the Ebro Hydrographic Confederation (CHE) with the aim of showing the different steps taken by this organisation to prevent the spread of this invader. These practical procedures are accompanied by diverse educational materials to inform and make people aware of the importance of this problem. The experience gained during these years has allowed the establishment of both an adequate sampling schedule and a sampling method for larval monitoring with the purpose of detection of zebra mussel populations. On the other hand, we have observed that mechanical cleaning and drying techniques are the physical control methods most frequently used by affected users in the Ebro basin, with use of chlorine as chemical method.

Key words: *Dreissena polymorpha*, Ebro River, impact, management, navigation

Introduction

Dreissena polymorpha (Pallas, 1771) is a freshwater bivalve mollusc, established in the Ebro River basin for nearly a decade. From the earliest records registered in the lower Ebro River, with densities of 500 ind.m⁻² (Altaba et al. 2001), the expansion of *D. polymorpha* has colonized the entire axis of the Ebro River, reaching the Sobrón reservoir (Burgos) and colonizing the adjacent basins of Guadalupe, Jalón and Gállego Rivers (Aragón), Segre and Noguera Pallaresa Rivers (Catalonia) and Zadorra River (Basque Country), with densities of up to 65,000 ind.m⁻² (CHE 2004a). As a result, the zebra mussel is now present in 7 rivers in the Ebro River basin (Figure 1).

The balance of the invasion of *D. polymorpha* is rather disappointing so far, because important ecological and economic impacts continue to take place. From an ecological standpoint, when a non-native species is established in an area other than its native habitat, it causes an ecological imbalance that alters ecosystem

equilibrium. In waters from which it originates, *D. polymorpha* is not usually the dominant species. However, it becomes invasive in freshwater communities where it is established (Vorobiev 1949).

The ecological impacts result mainly from the high filtering capacity of these molluscs. Strayer (2010) reviewed and updated the ecological effects of *D. polymorpha*, emphasizing the effects on the composition and abundance of biota, in addition to increased water clarity. As a result, other biotic stressors are favoured in their development, causing significant collateral effects. An example of this problem was observed in the Lower Ebro, near the village of Ascó (Tarragona, Spain), where macrophytes cover a large part of the river. This increased abundance of macrophytes, due to increased water transparency, can act as a barrier, absorbing the flow of nutrients used by phytoplankton. Increased concentrations of soluble phosphorus and inorganic nitrogen, from dead macrophytes, is evident in the quality of the water invaded (Artigas et al. 2008).

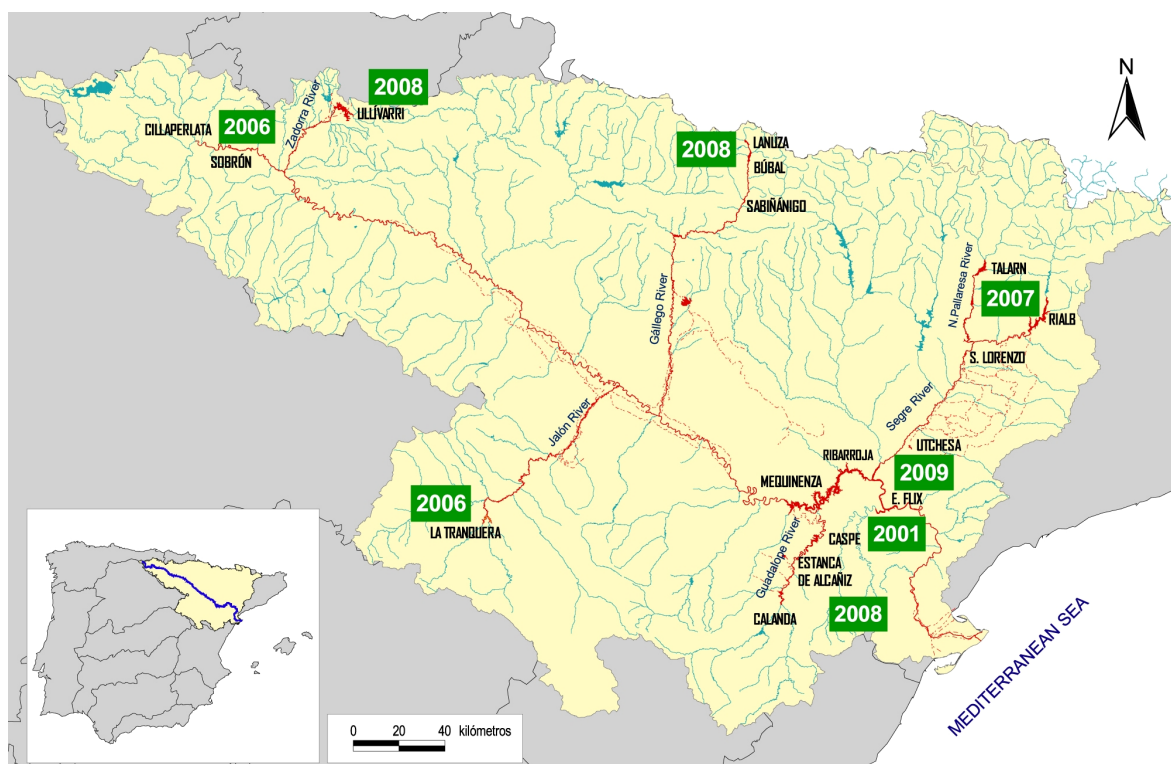


Figure 2. Known distribution of *Dreissena polymorpha* in Spain to date (April, 2010).

Native unionid mussel populations have declined due to the invasion of *D. polymorpha* in North America. Over 60 endemic mussels in the Mississippi River basin have been threatened with global extinction by the combined impacts of the *D. polymorpha* invasion and by environmental degradation (Ricciardi et al. 1998). In the Ebro River, the arrival of the zebra mussel threatens the survival of the world's largest population of the native bivalve *Margaritifera auricularia* (Spengler, 1793), located in the Ebro basin, which is now an imperiled species (Araujo 2008).

The economic impacts associated with the invasion of the zebra mussel arise from both problems in the operation of facilities affected (pipeline obstructions, interruptions) and additional costs of maintenance (cleaning activities, treatments), affecting any water user. *D. polymorpha* produces such a dense encrustations that affected facilities have to stop production to be mechanically cleaned, with attendant economic losses (Connelly et al. 2007). A single individual settles on a hard surface, and

then others colonise the space around it and even grow onto each other. The proliferation increases and can completely block pipes, vents and any holes or openings where water flows. Even if pipes and tubing are not completely blocked, they lose capacity due to section loss and increased roughness/friction (Nalepa and Schlosser 1993). In addition, there is evidence that the filaments of the byssus of zebra mussel may accelerate corrosion of joints in metal structures. Bacteria that appear between the substrate and the filaments produce an acid component through aerobic respiration which increases the corrosion of iron and steel surfaces (CHE 2007c). As a consequence, large investments from water-users are necessary. Irrigation systems, drinking water supplies, hydroelectric and nuclear plants, recreational areas and private properties are especially affected.

These costs must be added to the costs assumed by the public administrations in campaigns directed at preventing the spread of the pest and for providing information on how to act when the pest has colonised a facility.

In terms of impact on recreational use, many structures are vulnerable to zebra mussel colonisation, resulting in maintenance costs for recreational boats, jetties, floats, fishing nets, etc, and in general, any equipment in contact with the water. Also, mussels can destroy the engines of recreational craft by entering their cooling circuits.

An economical study has estimated the cost of this pest around millions of dollars since its arrival to the American continent in the 1980s (Connelly et al. 2007). The CHE conducted an economic study in 2005 which predicted a cost of around 40 millions of Euros over 20 years (CHE 2005a). This estimate is now outdated, so a new study to assess the amount spent until now is being carried out.

Reviewing the vectors of entrance of the pest in other countries, it is believed that in Spain the introduction was not due to ballast waters like in the U.S. (Johnson and Carlton 1996), or to the existence of navigation channels like in most of mainland Europe (Rajagopal et al. 2009). The precise mechanism of introduction of zebra mussel in the Ebro basin is not well-known, but the work of Rajagopal (2009) based on phylogenetic analyses between populations from different countries determined from PCR revealed that the Spanish invasion of zebra mussels was most likely from France. This introduction has been attributed possibly to the transport of recreational boats with mussel-fouled hulls or by fish transport across the Pyrenees. Another possibility could be the introduction of zebra mussel larvae with angler's food bait for catfish, a species with an important economic interest in the area Mequinenza-Ribarroja, where it was recorded for the first time. Many existing mechanisms for introduction of invasive species, both intentional and accidental, have been collected by Mills et al. (1993).

Once introduced in Spain, it is thought that most of the spread was due to the dispersal of larval stages by navigation. The first positive results of larval presence are always detected in navigable dams, not in rivers (CHE 2008). The detection of larvae in rivers, downstream of these dams, is always later. In the Ebro basin, recreational activity is plentiful in summer, so it was usual that boats navigated in several dams the same day, increasing the risk of larval zebra mussel transfer from affected bodies of water to non-affected bodies. The danger did not consist in adults attached to the boat hulls, easily visible,

but in larvae-containing water carried in the boat. As a consequence, boats must be cleaned and disinfected after sailing into affected bodies of water (Durán and Anadón 2008).

In this paper, a compilation of management works directed by the CHE is made with the aim of describing the management responses developed by this organisation to reduce the spread of this invader and its impact.

Focus of management strategy

The work carried out by the CHE has focused on three key areas: prevention, control and eradication. These actions listed on the Emergency Plan (CHE 2007a) are aimed at systematically and robustly preventing the colonisation of areas not affected to date, and reducing the effects caused by the presence of the zebra mussel in different areas affected throughout the Ebro basin.

At national level, the Ministry of the Environment of Spain prepared a National Strategy to combat the invasion (Environment Ministry of Spain 2007). The main objectives of the government are to prevent the spread of zebra mussels into uncontaminated bodies of water, as well as finding out more about the biology and behaviour of the species in order to design control measures as effective as possible.

After the detection of this invader in the basin, several studies were conducted to increase knowledge of the species and to establish appropriate management measures to curb its spread. The first steps were directed to investigate its impact in countries where the pest had already settled and to assess the pattern of physiological behaviour at our latitudes. From these physiological data, we studied the feasibility of conducting manual or mechanical extraction in affected reservoirs. Experimental work was carried out in the Ribarroja reservoir with the conclusion that these measures for large dams were too slow and labour-intensive, and therefore too expensive and impractical. The total cleaning of 1m² surface under experimental conditions took an average of about 15 minutes (CHE 2004a).

To evaluate alternative water management methods, the survival of the zebra mussel during a season of drought was studied (CHE 2005b). This study evaluated factors such as the angle of exposure to solar radiation, number of hours exposed to the sun and the relative humidity. Results showed that a higher cumulative

temperature with an increased number of hours of exposure to the sun, with adequate orientation, is crucial in obtaining high mortality rates. Another possible strategy for zebra mussel control consists of producing a drawdown in reservoirs, measure experienced in Minnesota and Pennsylvania with positive results (Grazio and Montz 2002). This measure is also supported by Navarro et al. (2006) after his research in Mequinenza reservoir. Navarro established that the distribution patterns of the zebra mussel are dominated by the hydrodynamic and thermal stratification of water layers. Therefore, any important fluctuation in the reservoir water level will cause a high death rate on the existing population.

In 2007, a study was conducted to evaluate the vulnerability of 89 reservoirs in the Ebro basin to colonization. For this purpose, an indicator called the Vulnerability Index to the Zebra Mussel (IVMC) was used. The IVMC measured the susceptibility of a body of water to being colonized by zebra mussels plus the direct risk due to its location and connection in relation to water bodies with confirmed presence of zebra mussels. This index ranges from 0 (no vulnerability) and 100 (highest vulnerability) and allows one to recognize which part of the resulting vulnerability is due to the intrinsic characteristics of the water body and which part is due to environmental and strategic importance. In applying this index, the importance of navigation as a vector for propagation of the species was considered (CHE 2007b).

The last study conducted by the CHE was based on the application of acoustic technologies in the detection and evaluation of *D. polymorpha* in Mequinenza reservoir. The hydroacoustic surveys provided high-resolution monograms with total coverage of 700 ha. This technique, joined videography, allowed detection of the location preferences of *D. polymorpha* populations in the reservoir, emphasizing a greater coverage of mussels in the first few meters depth with a maximum level to a depth of 6 m (50% coverage). At 17 m there was another sub-maximal density with 10% coverage. Between 6 and 17m, a distribution gradient of zebra mussel was observed. On a microhabitat scale, in terms of potential substrate (rocks, stones and all type of hard objects, including adult specimens of zebra mussel), there was no homogeneous colonization. There was a tendency of the mussels to colonize the lateral and inferior sides of these substrates (CHE 2009).

At present, other transmission vectors of the species are being evaluated, such as fishing equipment and swimming equipment. Identifying the likely vectors is essential in preventing the spread of this pest.

Population monitoring

Since 2004, annual campaigns of larval monitoring have been held for the early detection of the zebra mussel populations. In these campaigns, samples from different water bodies in the basin are taken using a specific protocol and then, analysed under a polarized light microscope to detect the presence or absence of zebra mussel larvae. Sampling is carried out from May to September to detect the main peak reproduction in May and June and to detect the second recruitment produced by the adults that settle in summer and grow quickly by August and September (Claudi and Mackie 1993; CHE 2006).

Results from the different annual campaigns allowed for the establishment of guidelines for the planning of sampling for the following season (A. Anadón pers. comm.). In 2009, 65 water bodies were sampled (mostly coinciding with the navigable reservoirs). Altogether, 182 sampling points were established and 1,096 larval samples were analysed. From these samples, 68 showed signs of presence of larvae but only 17 samples were confirmed, located in 5 water bodies: Sobrón, Mequinenza Flix, Azud in Fuenmayor and Utchesa (this latter was not listed as affected until the 2009 campaign).

The experience acquired year to year has allowed us to establish some guidelines to determine a sampling plan. The sampling season in which more positive results were detected and which coincides most with the peaks seen in the reproductive cycle of the species in our basin, corresponding to the months of May, June and July. However, it is more effective to extend the sampling season from June to September, coinciding with the period of greatest nautical activity in the basin. During this period, fortnightly sampling ensures an effective early detection for reservoirs since the results obtained in rivers in previous years were not decisive. To establish an adequate sampling method is equally important. The combination of a superficial sampling (sample taken from 20 cm depth) and another at depth (sample taken from the thermocline) has proved to be the most appropriate sampling. Selecting locations to

detect adult populations is also crucial, in terms of selecting sites according to the morphological, geological and dynamical characteristics of the reservoir.

Larval sampling conducted to date in the Ebro basin is based on microscopic observation, but it is considered essential to improve techniques. Techniques for detection of genetic material are being assessed as a complementary method to the traditional technique.

Regarding the monitoring of adult populations, this is performed by other local administrations with territory in the basin. Those waters with no evidence of the presence of *Dreissena* are inspected and affected water bodies are inspected to evaluate the density and evolution of populations. These tasks are performed by monthly inspection of adult specimens in the reservoirs. In the last campaign carried out in 2009, 42 were checked; the only ones colonized were in Flix, Sobrón and Calanda reservoirs.

Disinfection stations and closing of access points to affected reservoirs

After analysing the dispersal vectors in 2002, navigation rules for Ribarroja, Flix and Mequinenza were published. Rules required the disinfection of craft, entering and leaving affected water body (BOE 12.11.2002). The disinfection process involves the application of water under high pressure (160 bars) and temperature (60°C) (CHE 2002). To implement this protocol, the affected water bodies are equipped with the necessary facilities for cleaning and disinfection.

Initially, disinfection stations were built for reservoirs affected in the Lower Ebro (Ribarroja) and mobile cleaning services were promoted. At present, disinfection stations have been built or private cleaning services have been authorised in new bodies of water affected. When a new reservoir appears affected, navigation is not allowed until a new disinfection station to give service to it is built. Nowadays, there are 17 disinfection stations working. This measure is complemented with the closure of unguarded accesses to reservoirs, so that all boats go through the only point with cleaning and disinfection facilities. A total of 57 reservoirs were studied with 710 access points located (CHE 2004b).

Control and restrictions on navigation in rivers and reservoirs

The application of certain rules has been one of the main management tools used in preventing the spread of the pest in this basin. This line of work is widely developed in previous work (Durán and Anadón 2008).

Information and awareness campaigns

Much material has been published internationally about zebra mussels addressed to the general public, members of affected facilities and the educational community. In particular, two manuals presenting an overview of the zebra mussel control and eradication methods in affected facilities are recommended (CHE 2007c).

Since 2008, in response to the large number of questions about methods of control for affected facilities, the CHE decided to set up an advisory service for affected users. The aim of this service is to share the experience the Confederation has acquired since it first faced the problem in 2001.

The continued progress of the zebra mussel invasion coupled with its serious social and environmental consequences make it advisable to maintain and increase efforts at prevention, information and awareness to prevent its spreading due to inappropriate human activities, as much as possible. Educational material is available to primary and secondary schools. Since its first edition, this material has been sent to 135 primary schools (9.267 copies) and 24 high schools (1.861 copies).

On the other hand, conferences are organized, focused for different groups as centres for teachers and educational resources, fishing and sailing associations, environmental education groups and municipalities affected by the pest. These training sessions are accompanied by a travelling exhibition that moves throughout the basin.

Control methods

Many treatments exist to control zebra mussels in indoors facilities (Nalepa and Schloesser 1993; Claudi and Mackie 1994). However, a control method for open systems such as reservoirs, lakes, rivers, has not yet been found. Although eliminating the problem in facilities is successful, the colonized rivers are still active and continued foci of the species. Therefore,

research is gradually turning towards treating the original problem in the natural aquatic environment. Two research groups work in this area using the species filtration mechanism for ingestion of a control product. Aldridge with co-authors (2006) suggested a chemical control method called Biobullets. This method involves the encapsulation of active ingredients such as potassium chloride or quaternary amine compounds, coated with food material. Because of the coating, the mussel does not detect microencapsulates as a harmful substance and it does not therefore close its shell. This fact allows reducing the amount of poison used. Along these lines, Molloy (2002) was working with bacteria as potential biological control agents. *Pseudomonas fluorescens* CL145A has been selected to be highly lethal for zebra mussels. When a zebra mussel ingests high densities of this strain (live or dead), a toxin in these bacterial cells destroys the mussel's digestive system. In short, these new lines of research should be directed towards producing less harmful products for the environment.

In practice, in order to select a suitable control method for affected facilities, various issues must be taken into account, among them being the degree of control realistically achievable, the type and characteristics of the installation, the physical-chemical characteristics of the water, the larval density and the economic cost are significant.

In many European countries and in North America, where the presence of the zebra mussel has been present for many decades, governments and affected users have learned to deal with this problem counting the costs of treatment as an ongoing cost to be included in the production process. In fact, the control methods used in these countries may be considered Best Available Techniques. In Spain, the mentality is still directed towards eradicating the problem. However, gradually, the problem of zebra mussel will be recognised and managed proactively when new facilities in colonized areas are build.

In the Ebro basin, the most utilised method adopted by affected users in the Ebro basin is mechanical cleaning combined with drying techniques. Wire brushes, scrapers or other similar physical means are used. This method is widely used by the agricultural sector during the winter months, when irrigation systems are inactive because it is feasible to lower the water level in ponds or to completely empty the system. After this period, the dead mussels are

removed with brushes. The arrangement of the individual mussels must be considered, as individual mussels are more sensitive to air exposure than clumps of them. It is necessary to completely remove the remaining byssus in contact with the pumps and walls because these threads are suitable for new individuals to adhere again. This method is a short-term solution and is highly labour-intensive, but is relatively cheap and can be performed by the maintenance personnel. Other users complement these scraping techniques by using antifouling paints on pumps or turbines with good results.

To prevent the spread of the pest from the Ebro to the Cantabrian basin, a filtration system has been installed between the two basins. This method has also been selected for several areas dependant on irrigation for wine production, because strict legislation exists around this type of products. These filters constitute a physical barrier to the passage of the larvae, achieving retention of larvae of nearly 100%, without generating hazardous byproducts. This system has several stages of filter sizes of decreasing pore diameter, as in the first place, it is necessary to remove suspended solids in the raw water. The pore size filters exclusively dedicated to this purpose range from 25-50 μm .

Chemical control methods have more problems associated with them due to the byproducts generated. They must be sensitive to the aquatic ecosystem, be compatible with downstream uses of water and cannot be applied in open systems such as lakes and rivers. To reduce their emission in discharges, one measure under consideration is the establishment of the Best Available Techniques in chemical control methods, as well as defining the limits of these emissions in the aquatic environment. In any case, the most appropriate strategy to optimise treatments and minimise environmental and economic impacts is to identify periods of increased susceptibility of individuals to chemicals in each specific situation. As a consequence, with the same dose, more powerful results can be obtained.

Chlorine, as sodium hypochlorite, is the most widely used agent because of its low cost and easy availability. Because of its frequent use as primary and secondary disinfectant in the lines of drinking water treatments, chlorine is a readily available product commercially. Much literature about the dosage of chlorine to optimize effectiveness has been published (Rajagopal et al. 2003; Jenner et al. 2004). The

dosage is also different for shock treatment or in the case of prevention. The main point in common with different designs of treatment is to guarantee a minimum concentration of free residual chlorine throughout the system of distribution of 0.5 mg Cl L⁻¹ to assure the mortality of larvae. It should also be noted that chlorine also combines with organic matter, reducing the level of chlorine really effective on the larvae, so the free residual chlorine must be checked along the line of distribution. The removal of adult individuals is often done in the context of a shock treatment, where the initial concentrations of chlorine will be substantially higher than in preventive treatments.

Ozone is another chemical agent used. In fact, ozone is a well-known bactericidal agent for drinking water treatment. It has a double oxidative capacity with regard to chlorine and requires less contact time. In addition, it has no residual effect on the discharge as it quickly dissipates in the water. This fast decay, on the other hand, means a problem because to maintain the required residual oxidant in an extensive piping system, a great deal of product would be necessary (Claudi and Mackie 1994). Lewis with co-authors (1993) determined a residual dose of ozone of 0.5 mg L⁻¹ for a minimum of 5 hours to kill a 100% of larvae. In any case, ozonation equipment on site is required for this treatment.

As well as these chemicals products, some users affected in the Ebro basin utilise hydrogen peroxide in the fight against the zebra mussel. Doses of peroxide between 0.5 and 2.5 mg L⁻¹ are recommended for the inactivation of *D. polymorpha* larvae. The use of coadjuvants, such as peroxyacetic acid, favours the action of peroxide in the treatment.

Conclusions

The aim of this paper has been to group the key steps that a public organisation has developed to reduce the spread of the zebra mussel. It is important to take into account that control measures for invasive species are mostly not quantifiable in the short term, so the conclusions obtained for each strategy do not refer to measurable numbers, but to experience acquired, which marks the development of successive steps.

As a result of actions carried out since the beginning of the pest (period 2001-2009) in the Ebro basin, we can conclude that although the

strategies of the construction of disinfection stations and the closing of uncontrolled accesses are important prevention measures, in addition to research on the species and its interaction with the environment, that in fact a greater emphasis has been placed on education and outreach, both in affected areas and in areas without the presence of the pest. This is essential for the explanation of this problem and to sensitize the public to enlist their cooperation.

At the start of this or another invasion, all parties involved in management must be coordinated and work in the same direction, to avoid duplicating results and effort. Furthermore, it is of vital importance to collaborate hand-in-hand with the scientific community since the development of new technologies can be the key for a possible control and eradication of the zebra mussel.

In conclusion, not only should important actions be carried out in the affected basins, but it is essential to work with other vulnerable watersheds so that the process of invasion is delayed for as long as possible.

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