New Approaches to Effective Mollusc Health Management

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ABSTRACT

Despite past and current efforts to prevent the spread of infectious diseases in molluscs, new outbreaks continue to be recorded; and, in endemic zones, diseases of molluscs continue to be a major constraint to the industry. Traditionally, efforts have focused on movements of live molluscs as a main underlying cause for transfers of diseases. The central paradigm of mollusc health management, from this point of view, has certainly been built on *Bonamia ostreae* and the history of its introduction into Europe from California. However, further consideration for other major diseases in other mollusc species shows how poorly this paradigm applies. In addition, new routes of disease transfer are becoming more and more of a concern, such as ballast waters and hull attachment. Furthermore, climate change is increasingly recognised as a driving force in pattern changes of the distribution of diseases. All of this is happening in the context of an increasingly active global mollusc aquaculture. It is therefore important to develop new approaches to better address mollusc health management issues. The main shift in paradigm is probably to consider not only exotic diseases but new diseases, defined as emerging, re-emerging and exotic. This paper reviews the lessons to be learnt from the past and illustrates some avenues for improvement of mollusc health management through early warning systems, diagnosis in multiplexed assays, and multi-layered information system at the ecosystem level.

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INTRODUCTION

Global aquaculture production has considerably increased over the past few decades, with mollusc aquaculture alone having doubled its production over the same period (FAO, 2006). This sector represents about a quarter of the global aquaculture production in volume. In some parts of the world, the annual growth rates of the industry may record highs of 30% or more, for species such as abalone or pearl oysters, for example. The global mollusc production is based on more than 42 different species. These species provide a source of income or a substantial contribution to livelihood in many coastal communities from developing as well as developed countries.

The increase of mollusc aquaculture production is underlined by improved hatching capacity and farming technology, increased transfers and introductions of live molluscs, species diversification and farming intensification, as well as international market development. Concomitant to this, risks of spreading pathogens and diseases around the world have considerably increased. Despite past and current efforts to prevent the spread of infectious diseases of molluscs, new outbreaks continue to be recorded; and, in endemic zones, diseases of molluscs continue to be a major constraint to the industry.

For example, losses due to *Marteilia refringens* and *Bonamia ostreae* in the European flat oyster, *Ostrea edulis*, were estimated at US$31 million in France between 1980 and 1983; the diseases led to a 90% reduction of the production and a direct loss of 20% of direct employments in this sector of industry (Meuriot and Grizel, 1984; Grizel and Héral, 1991). Both parasites continue nowadays to impede the flat oyster industry in Europe. Similarly, *Haplosporidium nelsoni* and *Perkinsus marinus* have severely impacted the American oyster, *Crassostrea virginica*, on the East Coast of the USA. *Haplosporidium nelsoni* (MSX) is believed to be responsible for 95% mortality of the native oysters in Delaware Bay between 1957 and 1959 (Ford and Haskin, 1987; Burreson and Ford, 2004). Difficulty in managing the diseases caused by the two parasites in Chesapeake Bay has led to the consideration of introducing of a non-native species, the Suminoe oyster, *Crassostrea ariakensis*, in replacement of the native one (Calvo *et al*., 2000). In Europe, finding a resistant non-native flat oyster species was less successful; and trials to acclimatise exotic species of flat oysters have shown their susceptibility to *B. ostreae* (Grizel *et al*., 1983; Le Borgne and Le Pennec, 1983; Bougrier *et al*., 1986; Bucke and Hepper, 1987; Pascual *et al*., 1991).

Traditionally, our efforts have focused on movements of live molluscs as a main underlying cause for transfers of diseases. *Perkinsus marinus*, *Marteilia refringens*, *Bonamia ostreae* and the diseases they cause are among those listed by the World Organisation for Animal Health, the OIE (OIE, 2006). Indeed, *Haplosporidium nelsoni* has also been listed by the OIE and, only very recently, was removed from the list; mainly based on consideration for restricted geographical distribution of the susceptible species. The OIE aquatic standards aim at reducing the risks arising from the introduction, establishment or spread of significant pathogens to protect aquatic animals, and aquatic resources, while supporting the international movements and trade of live aquatic animals and products.

However, new disease incursions continue to be recorded.

To illustrate this, *Bonamia ostreae* was first detected in British Colombia, Canada, in 2004, from farmed European flat oysters, *Ostrea edulis* (Marty *et al*., 2006). This haplosporidian parasite is known to occur on both coasts of the United States and it causes significant mortality in Europe. Archived samples of oysters obtained from the index farm between 1999 and 2004 were used to track *B. ostreae* back to 2003. All of the 3 infected farms had been stocked with *O. edulis* spat from the State of Washington, USA, where *B. ostreae* is endemic. On the East coast of North America, *Haplosporidium nelsoni* has been detected in Nova Scotia, Canada, in 2001, thus broadening its geographical distribution (Stephenson *et al*., 2003). It is not clearly understood whether the disease has long been present but undetected (because of non conducive environmental conditions), or it has only recently been introduced. Following detection of MSX, plasmodia of *Haplosporidium costale* (SSO), but no spores, were also detected in low prevalence and intensity in oysters from several locations in the Southern Gulf of St. Lawrence.

An old military adage has it that no plan survives contact with the enemy. The modern discipline studying mollusc diseases is still young and dates back to the mid-50s (Sparks, 2005). Its development was driven by most of the diseases listed above both in North America and Europe; Dermo disease in the early days followed by other major epidemics, MSX disease, marteiliosis, and bonamiosis. Here, I review the lessons to be learnt from the past and propose some avenues to be explored for improved mollusc health management.

CHALLENGING THE PARADIGM OF MOLLUSC HEALTH MANAGEMENT

The central paradigm of mollusc health management has certainly been built on *Bonamia ostreae* and the history of its introduction into Europe. *Bonamia ostreae* was initially described in 1979 after catastrophic mortality outbreaks in European flat oysters, *Ostrea edulis*, cultured in France (Comps *et al*., 1980; Pichot *et al*., 1980). Although French investigators coined *B. ostreae*, the parasite had apparently been previously observed in California (Katkansky *et al*., 1969; Elston *et al*., 1986). The case is very well documented that demonstrates introduction of the parasite into Europe, France and Spain, with infected oyster spat originating from California (Grizel, 1997; Cigarria and Elston, 1997). The disease has then rapidly spread through Europe along the Atlantic coast, from Spain to Great Britain and Denmark, and to the Mediterranean waters. The evidence is that translocations of infected stocks have been the main cause of transfer of bonamiosis.

How does this paradigm apply to other diseases?

In the case of *Haplosporidium nelsoni*, the scenario is slightly different. I already referred to the impact of this parasite on the American oyster. The parasite also occurs in the Pacific oyster, *Crassostrea gigas*, in California, Oregon and Washington State, on the west coast of the USA, as well as in France, Japan and Korea (Friedman *et al*., 1991; Friedman 1996; Renault *et al*., 2000; Kamaishi and Yoshinaga, 2002). In fact, it is believed that the parasite has long occurred in Asia (Kern, 1976; Burreson *et al*., 2000). It is now recognised that contact of infected *Crassostrea gigas* with the native *C. virginica* caused the spread of this parasite to what proved to be a highly susceptible host (Burreson *et al*., 2000). Although *C. gigas* is certainly the most traded species, internationally, *C. virginica* remains the only known species susceptible to *H. nelsoni*. Bearing this in mind, one could say the *Bonamia* paradigm poorly applies in this instance.

Indeed, when looking more closely to diseases emergence, those scenarios used are quite varied. The Portuguese oyster, *Crassostrea angulata*, has nearly been extinguished after the outbreak of gill disease caused by an iridovirus. The disease is believed to have been introduced into Europe with first stocks of the sister species *C. gigas* in the late 60s. The two species are now believed to be con-specific with the Japanese oysters being resistant to the virus (Grizel and Héral, 1991; Renault and Novoas, 2004). In another type of scenario, after the introduction of the Japanese carpet clam, *Ruditapes philippinarum*, in Europe, this species has demonstrated a higher susceptibility to infection with *Vibrio tapetis* (Brown Ring Disease) compared to the native carpet clam species, *R. decussatus* (Paillard *et al*., 2004). In turn, *R. decussatus* appears more susceptible to *Perkinsus olseni*.

In those cases, problems have arisen from unexpected responses of an introduced species regarding endemic pathogens or the high susceptibility of a native species challenged by healthy carriers. These few examples show that we have made a simple/single storyboard out of a range of scenarios for emergence. Furthermore, our capacity to predict any of those scenarios is extremely limited because of the limited knowledge we have and the immense number of species we deal with.

In addition, new scenarios become more and more obviously significant. Ballast water and hull transfer should be cited here. Although one must admit that there is a real lack of scientific demonstration, examples are given as for *Haplosporidium nelsoni* in Nova Scotia, *Bonamia* sp. in North Carolina, *Marteilia* sp. in Florida, *Marteilioides chungmuensis* in Darwin harbour (Stephenson *et al*., 2003; Burreson *et al*., 2004; Hine, 1996). This increasing concern is also emphasised by the number of reports of alien/invasive species in aquatic systems (Carver *et al*., 2003; Bourque *et al*., 2006). Climate change is also recognised as an important factor of the change in distribution of pathogens and diseases. Change in environmental conditions is believed to be responsible for the northward expansion of *Perkinsus marinus*. Also, sea level rise could drastically alter the coastal environment, such as salinity regime in places like Chesapeake Bay, and this, as a result, may favour local diseases or emergence of new pathogens.

These considerations show that the routes of disease spreading are far more numerous that initially expected in the context of the *Bonamia* paradigm. This, in a certain extent, may explain that despite our efforts on international trade and exports certification, diseases apparently keep spreading. Obviously, by no means should we reduce our efforts towards safe trade. However, there is a need to shift paradigm to better address the issue of mollusc health management.

In order to make that shift, I propose here to consider not only exotic diseases, as we have done so far, but new diseases. New diseases are emerging, re-emerging and exotic diseases (Bower and McGladdery, 2003). Considering this is probably not as benign as it may seem in a first attempt. Indeed, by bringing the concept new diseases under the scope of mollusc health management, we probably set the scene for new approaches.

Bearing this in mind, I foresee avenues for health management improvement by use of sentinels and development of early warning systems in surveillance, diagnosis of pathogen signatures along with host health indicators in multiplexed assays, and multi-layered information system for real-time health management at the ecosystem level.

WHAT NEW STRATEGIES?

A strategy combines specific objectives and related methodological approaches. In the previous section of this paper, I have described how there is a need to re-visit our objectives in terms of health management, and consider the new diseases as the core contention. For this to be turned into facts, it becomes obvious that emphasis is now put on surveillance of target populations. The key point, in that context, will be to enable detection of any significant change in the population health.

In the specific case of molluscs, it seems like we have very few, if any, early warning systems. During a survey performed on various mollusc species in the south Pacific, certain groups clearly appeared as very susceptible to infection with *Perkinsus* sp. (Hine, 1996). For example, members of the Arcidae (*Arca*, *Barbatia*), Malleidae (*Malleus*), Isognomonidae (*Isognomon*), Chamidae (*Chama*) and Tridacnidae (*Tridacna*) apparently tolerate relatively high prevalence and intensity rates of *Perkinsus* sp. and are likely to provide good indicators of *Perkinsus* presence in the environment. This could be used in a sentinel system to monitor the parasite burden in a specific ecosystem and prevent its impact on economically significant species. Namely, the high value the pearl oyster industry would benefit from fast tracking *Perkinsus* occurrence in a management plan.

Following the same stream of thoughts, surveillance in harbours that are involved in international (long distance) traffic or have strong connectivity with populations at risk would make a solid basis of early warning systems. With this approach, rather than trying to check some potential sources of pathogens, efforts are directed to the most probable points of entry. By doing this, index cases would most likely be very close to the source itself. This also means possible sets of actions in an attempt to mitigate potential impact or restore ecosystem integrity, when and where applicable.

The use of sentinels, or canaries, could demand to go as far as developing highly susceptible lines of animals. This may appear as a real paradox for geneticists when considering efforts to improve disease resistance is the usual mantra (Gosling, 2003). Molluscs usually show high genetic variation which is regarded as good for enhancement of desirable traits by selective breeding. While this approach has been applied towards ramping up disease resistance, so far (Ford and Haskin, 1987; Naciri-Graven *et al.*, 1998; Culloty *et al.*, 2001; Bezemer *et al.*, 2006), one could imagine such programs to provide highly susceptible lines to be deployed as sentinels. Use of caged sterile triploids for example would appropriately minimise other related risks.

Before reaching such levels of sophistication, I should stress here that basic programs for passive surveillance involving field stakeholders would greatly improve our capacity in preparedness and emergency response.

A second fold of innovative approach comes with our capacity in diagnosis. Diseases and health cannot be reduced to the simple question of presence/absence of pathogens. Although one would easily agree with this assertion, one would also admit that it does not make the reality of our diagnostic activity. In most of the cases, diagnosis is based on the presence of pathogens; this is probably the most critical drawback imposed by molecular techniques when poorly understood and mis-used.

In an era of "–omics", genomics and proteomics and sister disciplines, we could expect to overcome bug hunting and compensate the nearly absence of clinical manifestations and symptoms, by identification of key molecules (RNAs or proteins) expressing disease condition. Those markers would serve as sign-posts of host as well as pathogen physiology and state of activation. While it may be difficult to develop this for the about 65 mollusc species or so currently used in aquaculture, we may think of clusters of key molecules for the four main groups of species (oysters, clams, pectens and mussels) to be developed. Such molecules would signal disorder in the absence of specific pathogen, or in presence of usually benign infection. Such signals would assist decision making, even in the absence of specific knowledge (which is a frequent situation as illustrated by examples above).

New technologies currently enable detection of pathogens and monitoring host responses in a single assay. The muliplex fluid array system or micro-array technologies are among the most recent innovations (Adams and Thompson, 2006). The Luminex System theoretically offers simultaneous quantitative analysis of up to 100 different bio-molecules from a single drop of sample in an integrated, 96-well formatted, system. These methods are in their early stage of development but the increasing number of published articles seems to show a real trend in different fields of application (Giavedoni, 2005; Diaz and Fell, 2005).

The third component of the triad for new approaches is the use of information systems at ecosystem level as a way to predict ecosystem response to changes. Such a response may be disease emergence or re-emergence; pathogen introduction may be the change itself.

As it appears, mathematical models fail to predict host population response or provide predictions of limited accuracy. Because emergence and re-emergence are originating in local and endemic situation changes, it is important to be able to see those changes and be able to anticipate on their potential consequences. It may be interesting to think about development of a multi-layered information system providing the tools for such changes to be monitored and recorded. Roughly, layers would consist in information on environmental conditions, host populations, health status, pathogen populations, etc. Although generic systems may be developed, their use in local situation on temporal series is essential to ensure the efficiency of the systems. Such multi-layered information system would provide trends sensors that could reveal themselves as useful in now-casting, rather than fore-casting, and therefore valuable assistants in decision making process.

CONCLUSION

In conclusion, the “fortress attitude” has shown to be limited in its success to prevent spread of infectious diseases that affect molluscs. In addition to responsible movements, it sounds realistically promising to explore new approaches and put efforts to the development of efficient early warning systems, accurate health indicators and ecosystem trend sensors. Furthermore, while traditional regulation frameworks are seen by the industry as a strong impediment to its operations, the proposed approaches are likely to find a positive echo in the private sector, thus enabling real partnership towards application of shared responsibilities in mollusc health management.

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