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Editors

Rohana P. Subasinghe

J. Richard Arthur

Devin M. Bartley

Sena S. De Silva

Matthias Halwart

Nathanael Hishamunda

C. V. Mohan

Patrick Sorgeloos



Improving biosecurity: a necessity for aquaculture sustainability

Expert Panel Review 3.3

**M. Hine^{1(*)}, S. Adams², J.R. Arthur³, D. Bartley⁴, M.G. Bondad-Reantaso⁵,
C. Chávez⁶, J.H. Clausen⁷, A. Dalsgaard⁸, T. Flegel⁹, R. Gudding¹⁰,
E. Hallerman¹¹, C. Hewitt¹², I. Karunasagar¹³, H. Madsen¹⁴, C.V. Mohan¹⁵,
D. Murrell¹⁶, R. Perera¹⁷, P. Smith¹⁸, R. Subasinghe¹⁹, P.T. Phan²⁰ and R. Wardle²¹**

¹ 73, rue de la Fée au Bois, 17450, Fouras, Charente Maritime, France. E-mail: vinet.hine@orange.fr

² Institute of Aquaculture, University of Stirling, Stirling FK9 4LA, Scotland, UK.
E-mail: alexandra.adams@stir.ac.uk

³ Box 1216, Barriere, B.C., Canada V0E 1E0. E-mail: jraconsulting@xplornet.ca

^{4,5,13,19} Department of Fisheries and Aquaculture, Food and Agriculture Organization of the United Nations (FAO), Viale delle Terme di Caracalla, Rome, Italy. Devin.Bartley@fao.org, Melba.Reantaso@fao.org, Iddya.Karunasagar@fao.org, Rohana.Subasinghe@fao.org

⁶ Unidad Mazatlán en Acuicultura y Manejo Ambiental del CIAD, A.C. Av. Sábalo Cerritos s/n, Mazatlán, Sinaloa, México. E-mail: marcris@ciad.mx

^{7, 8, 14, 16} Department of Veterinary Pathobiology, Royal Veterinary and Agricultural University, Groennegaardsvej 15, 1870 Frederiksberg, Denmark. E-mail: jehc@life.ku.dk ; ad@life.ku.dk ; hm@life.ku.dk ; hm@life.ku.dk

⁹ Centex Shrimp and Dept. Biotechnology, Faculty of Science, Mahidol University, Rama VI Rd., Bangkok 10400, Thailand. E-mail: sctwf@mahidol.ac.th

¹⁰ National Veterinary Institute, POBox 750, 0106 Oslo, Norway. E-mail: roar.gudding@vetinst.no

¹¹ Department of Fisheries and Wildlife Sciences, Virginia Polytechnic Institute and State University, Blacksburg, VA 24061-0321, United States. E-mail: ehallerm@vt.edu

¹² National Centre for Marine and Coastal Conservation, Australian Maritime College, University of Tasmania, Australia. E-mail: chewitt@amc.edu.au

¹⁵ Network of Aquaculture Centres in Asia and the Pacific, Bangkok, Thailand. E-mail: mohan@enaca.org

¹⁷ Biosecurity Australia, Australian Government Department of Agriculture Fisheries and Forestry, GPO Box 858, Canberra ACT 2601, Australia. E-mail: Ramesh.Perera@daff.gov.au

¹⁸ Department of Microbiology, National University of Ireland, Galway, Ireland.
E-mail: peter.smith@nuigalway.ie

²⁰ Centre for Environment Disease Monitoring in Aquaculture-Research Institute for Aquaculture, No. 1. Bac Ninh, Vietnam. E-mail: phanvan@ria1.org

²¹ Intervet Schering-Plough Animal Health, 24-26 Gold Street, Saffron Walden, Essex CB10 1EJ. E-mail: Robin.wardle@sp.intervet.com

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* Corresponding author: vinet.hine@orange.fr

Abstract

The implementation of biosecurity measures is vital to the future development of aquaculture, if the culture of aquatic species is to make it possible to feed the global human population by 2030. Biosecurity includes control of the spread of aquatic plant and animal diseases and invasive pests, and the production of products that are safe to eat. For controls on diseases and invasive pests, it is necessary to implement programmes that involve all regional countries. Lessons from measures implemented in Asia need to be expanded/upscaled in Latin America, Africa and other emerging aquaculture regions. Such development will make countries more self sufficient and will feed local populations.

Globally, there is good evidence that aquatic animal diseases and invasive animal and plant pests are being spread by hull fouling and ballast water in shipping, and serious aquatic animal diseases by the international trade in ornamental fish. While there has been a growing awareness of the danger of ballast water transfer, hull fouling remains a serious problem. It is widely recognized that ornamental fish present a disease risk, but individual countries have tried to address this alone, and there has not been an international effort to control the trade.

Developments in genetics and molecular biology hold great potential for disease control, either by breeding for disease resistance, or by the use of rapid, specific, culture site testing. Currently, there is no evidence that the use of antibiotics in aquaculture poses a threat to human health or that antibiotic-resistant strains have developed; however, the future use of genetically modified aquatic organisms (GMOs) may negate the need for chemotherapy. Cultured aquatic organisms, selected for disease resistance or rapid growth, are likely to become more acceptable, and probably necessary, to feed the rapidly growing global population.

Most global aquaculture occurs in developing Asian countries, in which aquaculture products can harbor zoonotic parasites, and there is a need to treat such products to negate the threat of parasitic zoonoses and permit international export. Climate change is likely to be a major influence on aquaculture in the future, with impacts on coastal aquaculture through increased sea levels affecting coastlines, and acidification. To feed the growing global population, it will be necessary to culture new species, for which research on diseases and invasiveness will be necessary to acquire the information necessary to implement biosecurity measures.

KEY WORDS: *Aquaculture, Biological invasions, Biosecurity, Genetically modified organisms, Transboundary aquatic animal diseases.*

Introduction

More than 200 species are produced in aquaculture worldwide; some 25 of these are of high value and traded globally. A successful harvest can be very profitable, and this has spurred the expansion of aquaculture production in both area and geographical range. As aquaculture becomes more intensive, new diseases and other problems are likely to emerge, and old diseases will appear in new locations.

Subasinghe, Bondad-Reantaso and McGladdery (2001) in a review paper entitled “Aquaculture development, health and wealth” as part of the *Technical Proceedings of the Conference on Aquaculture in the Third Millennium* (FAO/NACA, 2001), described how disease has become a primary constraint to sustainable aquaculture production and product trade, provided some examples of the socio-economic impacts of transboundary aquatic animal diseases (TAADs) as well as measures to deal with aquatic diseases, and evaluated the effectiveness of health management programmes and what can be done to improve health management and reduce disease risks. The current review takes a broad approach to as many aspects and issues of biosecurity as possible and the role of effective biosecurity in the sustainable increase in aquaculture production.

The Food and Agriculture Organization of the United Nations (FAO) defines biosecurity as a strategic and integrated approach that encompasses both policy and regulatory frameworks aimed at analyzing and managing risks relevant to human, animal and plant life and health, including associated environmental risks (FAO, 2007a). It covers food safety, zoonoses, introduction of animal and plant diseases and pests, introduction and release of living modified organisms (LMOs) and their products (e.g. genetically modified organisms or GMOs), and the introduction of invasive alien species. It is a holistic concept of direct relevance to the sustainability of agriculture, public health and protection of the environment, including biological diversity. An essential element of sustainable agricultural development and food production, the overarching goal of biosecurity is to prevent, control and/or manage risks to life and health appropriate to the particular biosecurity sector.

Many factors are driving the current interest in biosecurity. Globalization (increase in volume and diversity) of trade in food, plant and animal products; changing food production practices and climate with new technologies; heightened awareness of biological diversity; greater demand for public health and environmental protection and other emerging issues such as rising food prices, climate change and animal welfare, are some of these. The benefits of improving biosecurity through safeguarding plant and animal life and health, enhancing food safety, promoting environmental sustainability, protecting biodiversity and a long-term strategic response to rising food prices are also recognized (Bondad-Reantaso, Lem and Subasinghe, 2009).

In aquaculture, biosecurity refers to the application of appropriate measures (e.g. proactive risk analysis) to reduce the probability of an organism spreading to individuals, populations or ecosystems, and to mitigate the adverse impacts that may result from such (Subasinghe and Bondad-Reantaso, 2006). It is concerned with management of aquatic animal health, conserving aquatic biodiversity and reducing public health risks associated with production and consumption of aquaculture products. This analysis incorporates the best information available on aspects of husbandry, epidemiology and good science.

Sections 3.11 (managing aquatic animal health), 3.13 (applying genetics to aquaculture), 3.14 (applying biotechnology) and 3.15 (improving food quality and safety) of the *Bangkok Declaration and Strategy for Aquaculture Development Beyond 2000* (Subasinghe et al., 2001) are all relevant to biosecurity. Traditionally, such concerns have been addressed using the sectoral approach to biosecurity, and what is lacking is a holistic systems approach to aquatic animal health management and biosecurity. Since the 2000 Aquaculture Millennium Conference, introduction of TAADs through global trading, and food safety and public health issues continue to challenge the aquaculture sector, and new issues have emerged. These include TAADs associated with the global trade in ornamental aquatic animals; a spread of invasive animals and plants, viruses, microbes and toxic algae by vectors; and climate change scenarios affecting biosecurity.

Implementing effective biosecurity is vital to the future development of aquaculture, if the culture of aquatic species is to make it possible to feed the global human population by 2030. Biosecurity concerns including food safety, public health risks on the use of veterinary medicines, bioinvasions and the use of aquatic GMOs are discussed in this review. Major issues and trends during the last decade are presented, followed by an elaboration of what has been achieved by different stakeholders. The outcomes of the expert panel presentation during the Global Conference on Aquaculture 2010, held in Phuket, Thailand in October 2010 are also presented. The paper concludes with a number of recommendations and the way forward.

Major biosecurity issues and trends during the last decade

Transboundary aquatic animal diseases

The health of aquatic animals is not always readily visible, as feed consumption and mortalities are hidden under water. Thus, attention is required to monitor their health. Because of the great diversity of the aquaculture sector in terms of species cultured, the range of culture environments, the nature of containment, the intensity of farming practices and the variety of culture and management systems, the task of managing aquatic animal health and biosecurity governance is particularly challenging. Once a pathogen has been introduced and becomes established in the natural aquatic environment, there is very little or no possibility for either treatment or eradication; therefore, prevention is the best strategy.

Transboundary aquatic animal diseases (TAADs) are aquatic animal diseases that are highly infectious, have the potential for very rapid spread irrespective of national borders and can cause serious socio-economic consequences. Domestic and international trade are important pathways for the introduction of TAADs. Increase in trade will also increase the risk of new mechanisms by which pathogens may be introduced and spread to new areas together with host movement. In aquaculture, many examples exist of TAADs that created serious negative impacts, including: direct production losses, direct and indirect impacts on income and livelihoods/employment, increased operating costs, restrictions on trade, impacts on biodiversity, loss of market share or investment, loss of consumer confidence, and in some cases, collapse of the sector (Subasinghe, Bondad-Reantaso and McGladdery, 2001; Bondad-Reantaso *et al.* (2005); Bondad-Reantaso, Sunarto and Subasinghe, 2007). Available estimates on losses due to TAADs, reviewed by Bondad-Reantaso *et al.* (2005), range from as low as USD17.5 million (white spot disease (WSD) of shrimp in India in 1994) to as high as USD650 million (for yellowhead virus and WSD in Thailand in 1994) to a global estimate of USD3.019 billion in losses due to shrimp diseases. In a review of disease issues in the shrimp aquaculture industry up to 2005 (Flegel *et al.*, 2008), it was estimated that production losses due to disease over the preceding 15 years amounted to approximately USD15 billion. According to a survey conducted by the Global Aquaculture Alliance, approximately 60 percent of disease losses in shrimp aquaculture could be attributed to viral diseases and approximately 20 percent to bacterial diseases (Flegel, 2006b), indicating that 80 percent of the disease losses were attributed to only two pathogen groups, with viruses having approximately four times more negative impact on production than bacteria. Movement of live aquatic animals has been recognized as a major pathway for the introduction and spread of major TAADs. Fish are the most globally traded commodity, with a world value of USD93 billion for 2007 (Bondad-Reantaso, Lem and Subasinghe, 2009).

The current period of rapid change in the international trading environment has changed the disease situation in aquaculture rapidly and in an unpredictable way. Factors contributing to the current disease situation in aquaculture include: increased globalization of trade and markets: intensification of fish-farming practices through the movement of broodstock, postlarvae, fry and fingerlings; introduction of new species for aquaculture development; expansion of the ornamental fish trade; enhancement of marine and coastal areas through the stocking of aquatic animals raised in hatcheries; the unanticipated interactions between cultured and wild populations of aquatic animals; poor or lack of effective biosecurity measures; slow detection of emerging diseases; the misunderstanding and misuse of specific pathogen free (SPF) stocks; climate change and human-mediated movements of aquaculture commodities. Indiscriminate and unregulated global movement of aquatic animals has extended the geographical range of important TAADs and has caused serious disease outbreaks (Bondad-Reantaso *et al.*, 2005).

TAADs include: (1) epizootic ulcerative syndrome (EUS), whose original distribution was in Asia and the United States of America, which has recently expanded its geographic range to Africa (in 2006 and is now present in at least four countries in the African region) affecting mainly wild and some cultured populations; (2) koi herpesvirus (KHV), which has spread infecting the important food fish the common carp (*Cyprinus carpio*), the high-value ornamental koi carp and wild carp populations; and (3) infectious salmon anemia (ISA) and sea lice that have cost the salmon-producing countries millions of dollars in losses annually. Major European oyster-producing countries have experienced severe mortality events, including losses caused by the protozoan parasite *Bonamia ostreae*, which was transported from North America, and oyster herpesvirus (OsHV-1), which has spread with culture of Pacific cupped oysters (*Crassostea gigas*). White spot disease (or white spot syndrome virus, WSSV), considered as the most serious global pathogen of cultivated shrimp, has spread to more than 20 shrimp-producing countries. Viral nervous necrosis (VNN) is an important disease of cultured and wild marine fish, affecting almost 30 species.

TAADs, risk analysis and the ornamental fish trade

The *Aquatic Animal Health Code and Manual of Diagnostic Tests for Aquatic Animals* (OIE, 2011a,b) of the World Organisation for Animal Health (OIE) both recognized the international spread of disease via trade in ornamental aquatic animals. Recent changes to the global aquatic animal disease situation, and the importance of pathogens that infect ornamental fish (primarily cyprinids) are increasingly reflected in the OIE list of diseases, which now includes KHV and EUS, as well as spring viraemia of carp (SVC) and bacterial kidney disease (BKD). The inclusion of KHV and EUS, allows competent authorities to require international health certificates indicating freedom from these diseases, thus avoiding the need for import risk analyses (IRAs).

It was generally assumed that the risk of disease introduction in importing countries by the ornamental fish trade was theoretical, and that the likelihood of negative impacts resulting from the trade was very low. This was due to an absence of hard evidence linking ornamentals to serious disease outbreaks in native populations, belief that escapes or releases of aquarium-held ornamentals into natural waters were rare, and when they did occur, the chances of ornamental fishes surviving in temperate aquatic systems was unlikely (Davenport, 2001). The pathogens of ornamental fish and invertebrates and their host specificities are very poorly known, making assessment of the risk of establishment in new aquatic environments and hosts, and their environmental impacts, difficult to assess. Governments have had difficulty in effectively regulating the highly complex ornamental trade, due to its huge volume (>1 billion ornamental fish moved annually), the large number of species involved (>4 000 freshwater and 1 400 marine species), and the large number of exporting and importing countries (>100) (Whittington and Chong, 2007). In addition, the high frequency of transshipment and relabeling obscures both the source (e.g. from wild-caught

or cultured stocks) and the country of origin (Davenport, 2001; Latiff, 2004; Arthur *et al.*, 2008). The world's largest producer, Malaysia, for example, with a 2007 production of ~558 million ornamental fish and plants, exports much of its production via Singapore (Ng, 2009). Further difficulties arise because the industry has been resistant to regulation and because many countries accept "health certificates" based on the absence of gross signs of disease, without knowledge of the health status of the production facility, the origin of stock, surveillance, or the fish being shipped having been screened for parasites and diseases.

The international trade in ornamental aquatic animals has been shown, both theoretically (through IRAs) and actually (Lumanlan *et al.*, 1992; Hedrick and McDowell, 1995; Sano *et al.*, 2004; Iida *et al.*, 2005; Sunarto and Cameron, 2005; Bondad-Reantaso *et al.*, 2005; Whittington and Chong, 2007) to pose serious risks of introducing TAADs to new areas through the movement and escape or release of infected animals. National governments, particularly of countries in semitropical and tropical latitudes, have become increasingly aware of the potential environmental and pathogen risks posed by the ornamental trade and the difficulties of accurately assessing and managing these risks. They will thus be increasingly inclined to adopt a more precautionary approach to the movements of ornamental species.

The European Union (EU) has introduced regulation of the ornamental fish trade, adopting a risk-based approach to disease control. Regulations introduced in 2008 and 2009 include conditions for marketing, certification requirements, possible vector species, a model health certificate, a list of permitted third countries, ornamental fish susceptible to listed diseases, and the suspension of imports from Malaysia of some ornamental cyprinid fishes.

Risk management for aquatic animal pathogens outside those in the OIE Code must be justified by IRA. During the past decade, several IRAs have been conducted for ornamental aquatic animals (Table 1). With the exception of the recent IRA for gourami iridovirus by Biosecurity Australia (2009), such IRAs have considered many hosts and pathogens, and have many weaknesses. Ornamental fish are a special case in live animal trade where the OIE guidelines for IRAs may need to be revised, or where countries such as Australia with very high appropriate level of protection will have to greatly reduce the number of species traded and the number of sources permitted for hazard identification and risk assessment (Whittington and Chong, 2007).

An example of a more "specific" IRA for ornamental aquatic animals is that for gourami iridovirus and related iridoviruses conducted by Biosecurity Australia (2009). The study concluded that gouramis, cichlids and poeciliids pose an unacceptably high level of risk and recommended that in addition to existing import conditions, fish in these families should either be batch tested post-

TABLE 1
Summary of risk analyses completed on ornamental aquatic animals

Risk Assessment	Commodity	Importing Country/ Exporting Country	No. Hosts Considered	No. Potential Hazards in Preliminary List	No. Hazards Fully Assessed	Hazard: Host Ratio	Hazards Fully Assessed as % of Preliminary Hazards
Khan <i>et al.</i> (1999)	Live ornamental finfish	Australia/ Global	605 genera	104	44	0.17:1	42.7%
Hine and Diggles (2005)	Ornamental fish & marine invertebrates	New Zealand/ Global	394 genera and species	>500	35	2.4:1	7.9%
Biosecurity NZ (2009) *			+158 genera	+42	+8		
			Total of approx. 1300 species	>542	43		
Biosecurity Australia (2009) **	Ornamental finfishes	Australia/ Global	All allowable taxa	29	29	–	100%

* This study was a supplement to the earlier IRA by Hine and Diggles (2005).

** IRA was restricted to consideration of gourami iridovirus and related viruses (total of 29 strains/isolates). The study considered all freshwater and marine ornamental fishes allowed for importation (currently some 284 listings; see www.environment.gov.au/biodiversity/trade-use/lists/import/pubs/live-import-list.pdf); as these include listings at the family, genus and species level, no exact number can be calculated; however, the number of potential species must be in the thousands.

arrival in Australia to show freedom from iridoviruses of quarantine concern or that importations should be approved only if they are from countries, zones or compartments known to be free of iridoviruses of quarantine concern (based on active surveillance).

TAADs in shrimp culture and other technological developments

Transboundary movements of viral pathogens is a particular problem in shrimp aquaculture. Crustaceans may carry low levels of one or more non-host specific viral pathogens, even lethal ones, as persistent infections for long periods without gross signs of disease. These active viruses can be transmitted to naïve shrimp or other crustaceans, causing lethal infections, and can also be transmitted from broodstock to apparently normal larvae and postlarvae, with subsequent disease in rearing ponds stocked with the infected postlarvae. These hidden viral infections pose a great risk when living crustaceans destined for aquaculture are moved transboundary outside their enzootic range (Flegel, 2006c). This has resulted in several major shrimp viral epizootics, most notably for *Penaeus stylirostris* densovirus (PstDNV) in *Litopenaeus stylirostris* and *L. vannamei* in the Americas (Lightner, 1996), WSSV in all cultivated shrimp in Asia and the Americas (Flegel, 2006b), Taura syndrome virus (TSV) in *L. vannamei* in Asia (Nielsen *et al.*, 2005) and more recently infectious myonecrosis virus (IMNV) in *L. vannamei* cultivated in Indonesia (Senapin *et al.*, 2007). Polyculture carries risks, such as the risk of transfer of endemic PstDNV from *P. monodon* to *L. vannamei* at the larval stage when rearing of captured *P. monodon* and exotic specific pathogen free (SPF) *L. vannamei* in Asian shrimp hatcheries. Also, *Macrobrachium rosenbergii* nodavirus (MrNV) can infect larvae of *P. monodon*

and *Fenneropenaeus indicus* causing high mortality (Ravi *et al.*, 2009), despite not causing mortality in challenged juvenile shrimp of the same two species (Sudhakaran *et al.*, 2006).

About 20 shrimp viruses have been described, some with subtypes differing in virulence, but only a few pose serious threats, and serious pathogens differ according to shrimp species. WSSV causes the greatest production losses, and it is lethal to all cultured species (Flegel, 2006a). Yellow head virus (YHV) causes serious mortalities in *P. monodon* (Boonyaratpalin *et al.*, 1993) and *L. vannamei* (Senapin *et al.*, 2010), but there are five or six subtypes and the most virulent type (YHV-1) only causes serious disease in Thailand (Wijegoonawardane *et al.*, 2008). PstDNV causes high mortality in *L. stylirostris* and stunted growth in *L. vannamei*, but has little effect on *P. monodon* (Withayachumnankul *et al.*, 2006). Most commercial stocks of *L. vannamei* are now tolerant to TSV, and PstDNV does not usually affect PL in rearing ponds. The serious viral pathogens for *L. vannamei* are WSSV, YHV Type-1 and IMNV, and for *P. monodon*, WSSV, YHV Type-1 and Laem-Singh virus (LSNV).

All these viruses exist in their shrimp and other crustacean hosts in active states, in company with other viruses, with or without visible signs of disease. A non-disease state can be converted to a disease state by various stress triggers. The first consequence arising from these facts is the possibility of transferring known (or unknown) exotic viruses to new locations together with exotic shrimp. The second is that known (or unknown) viruses may jump into the exotic imported shrimp from local crustaceans. Precautions must be taken to avoid these possibilities.

If a secure supply of uninfected postlarvae can be obtained for stocking shrimp ponds, the next biggest problem for farmers is to maintain strict biosecurity to prevent viral transmission from natural carriers to shrimp in rearing ponds, mostly by exclusion of potential shrimp and other crustacean carriers during pond preparation before stocking and during rearing after stocking. This can be accomplished simply by filtration and storage of water before it is used in rearing ponds. However, some farmers elect to use short-lived insecticides or disinfectants to treat water before it is used. Physical barriers (e.g. low fences) are often used to limit crab entry over land. Recent unpublished work in Thailand indicates that insects may sometimes be shrimp virus carriers, suggesting that ponds should be completely covered, when possible, with fine netting (i.e. equivalent to mosquito netting) to exclude insects. This has the added advantage of also excluding moribund shrimp dropped by birds from nearby outbreak ponds.

By comparison to viral pathogens, work on control of bacterial pathogens of shrimp has been less intensive and has focused mainly on farm management practices related to control of the environment in hatchery tanks and

rearing ponds. Much of this has been focused on the use of probiotics and immunostimulants. As predicted (Flegel *et al.*, 2008), development of rapid and specific diagnostic methods for major shrimp pathogens has improved steadily in the past decade. Since the reviews up to 2005 (Flegel, 2006a, 2008), more pond-side immunodiagnostic strips have been developed (Sithigorngul *et al.*, 2007) for pathogen confirmation at the prepatent or outbreak level of infection. For carrier states, more isothermal nucleic acid amplification methods have been developed for use with electrophoresis (Mekata *et al.*, 2006) or with lateral flow diagnostic strips (Jaroenram, Kiatpathomchai and Flegel, 2009). Offering test specificity and sensitivity equivalent to nested polymerase chain reaction (PCR) methods but lacking of the requirement for an expensive PCR machine, these isothermal methods provide the opportunity for more widespread application. Despite these new opportunities, more training and extension work is required to bring them to the farm level. A good model of how to achieve this can be seen in the Australian Center for International Agricultural Research (ACIAR) project (FIS/2002/075) on application of PCR for improved shrimp health management in the Asian region (Walker and Subasinghe, 2005).

In the wider application and improvement in shrimp biosecurity, much has been achieved by the implementation of good aquaculture practices (GAP), particularly via government extension workers and shrimp farmer associations, but there is still a need for more training and extension work as exemplified by the ACIAR project mentioned above. For transboundary movement of living crustaceans for aquaculture, the major problem is not with regulations but with aquaculture practitioners who ignore the regulations. A very recent example is the case of IMNV outbreaks in Indonesia (described above) initiated by illegal shrimp imports from Brazil. Clearly, laws are not enough, and there has been insufficient education to achieve a situation where everyone in the shrimp aquaculture industry believes that such activities are socially, morally and economically unacceptable.

Turning to the application of new technologies such as probiotics, immunostimulants and vaccines, there has been little change in the situation since 2005 (Flegel *et al.*, 2008). Despite the widespread use of probiotics and to a lesser extent immunostimulants in shrimp farming, there have been no published results from large-scale field trials to prove by statistical analysis that they are really effective. Field trials and more research are also needed on quorum sensing control of bacterial pathogens (Van Cam *et al.*, 2009). For so-called shrimp “vaccines” based on heterologously produced viral coat proteins, inactivated viral preparations, shrimp viral binding proteins (Ongvarrasopone *et al.*, 2008) and DNA “vaccines” (Ning *et al.*, 2009), the mechanism of protection is still unknown. Based on what is known of shrimp immunity (Flegel and Sritunyalucksana, 2010), the mechanisms are unlikely to be the same as those associated with vaccines used in fish and other vertebrates. Other recent discoveries include the efficacy of using double-stranded RNA (see Robalino *et al.* 2007 for a review) and egg

yolk antibodies (passive immunity) (Lu *et al.*, 2008) to protect shrimp from viral infections. So far, reports of all these new technologies have been based on laboratory trials, and further tests are needed to determine whether they will be efficacious in large-scale commercial applications. For more details on these technologies, readers may consult a number of recent reviews (e.g. Robalino *et al.*, 2007; Flegel and Sritunyalucksana, 2010). Very recently, it has been proposed that viral inserts in the shrimp genome may be the basis of a new type of heritable immunity (Flegel, 2009). If this proves correct, it will fundamentally change the process for selection of viral-resistant shrimp stocks.

Finally, work on shrimp molecular epidemiology has been focused largely on comparison of geographic isolates of infectious hypodermal and hematopoietic necrosis virus (IHHNV) (Tang and Lightner, 2006), TSV (Tang and Lightner, 2005), WSSV (Pradeep *et al.* 2008) and YHV (Wijegoonawardane *et al.*, 2008) and less on the more practical aspects of dynamics and risks of spread in farming systems. Work on molecular ecology (i.e. metagenomics) and biochemical engineering to control the microbial dynamics in shrimp ponds and hatchery tanks has been relatively neglected.

Disease diagnostic methods: developments, gaps in knowledge and needs

Rapid disease diagnosis is crucial to the sustainability of aquaculture, and rapid progress in biotechnology over the last decade has enabled the development and improvement of a wide range of immunodiagnostic and molecular techniques (Cunningham, 2004; Adams and Thompson, 2006, 2008), and reagents and kits have become more widely available. In recent years, methods developed for clinical and veterinary medicine have been adapted and optimized for use in aquaculture. Despite this, identification of certain pathogens is difficult to achieve, and some of the methods developed are too complicated to implement and interpret. Traditional methods of pathogen isolation and characterization tend to be costly, labour intensive, slow and may not give a definitive diagnosis. For many rapid methods, live and dead pathogens cannot be distinguished; therefore, enrichment methods and the use of live/dead kits are useful supplementary methods (Vatsos, Thompson and Adams, 2002). Interpretation of results using rapid methods should be considered with other clinical evidence. The OIE *Aquatic Animal Health Manual* (OIE, 2911b) includes standardized methods for the identification of notifiable pathogens, but for those diseases that are not included, there are no set standards. Commercial reagents and kits (Adams and Thompson, 2008) provide specific and sensitive standardized methods, but a full range of reagents or kits is not available for use in aquaculture. The cost, speed, specificity and sensitivity of assays are all extremely important to end-users. Many of the new technologies require specialized equipment and highly skilled staff, and few of the existing methodologies are suited to field testing or use in rudimentary laboratories.

Immunodiagnostic methods currently used, such as immunohistochemistry (IHC), the fluorescence antibody test (FAT) and indirect fluorescence antibody test (IFAT) enable rapid specific detection of pathogens in tissue samples without the need to first isolate the pathogen. IHC is an extension of histology, while FAT/IFAT is a more rapid, sensitive procedure. Other antibody-based methods, such as the enzyme linked immunosorbent assay (ELISA), have also been developed for use in aquaculture (Adams and Thompson, 1990). ELISA allows high throughput, and automated equipment is available, but is less sensitive than IHC and IFAT. ELISA can also be used for serology, although it has not yet been validated for any bacterial diseases in fish. Serology, however, effectively detects fish viruses, such as KHV (Adams and Thompson, 2008). Recently, lateral flow technology is widely used in clinical and veterinary medicine (Bai, *et al.*, 2006) and has been developed for use in aquaculture (Adams and Thompson, 2008). It is very rapid and sensitive, and can be used as a pond-side test. Commercially available kits for infectious salmon anemia virus (ISAV) were recently independently evaluated (Carauel *et al.*, 2010) against other methodologies (culture, IFAT, reverse transcriptase-PCR (RT-PCR) and quantitative RT-PCR (qRT-PCR)) and were found to have the highest operational specificity. This technology is simple to use, rapid (with results in less than 10 min), cheap to perform and does not require skilled operators or expensive equipment.

Molecular technologies for the detection of fish pathogens (Cunningham, 2004; Adams and Thompson 2006, 2008) generally have the highest sensitivity in detecting low numbers of micro-organisms and those that are difficult to culture. They can identify species (Pourahmed, 2008) and individual strains, and differentiate closely related strains (Cowley *et al.*, 1999). There are many variations of PCR, including nested PCR, random amplification of polymorphic DNA (RAPD), RT-PCR, reverse cross blot PCR (rcb-PCR) and RT-PCR enzyme hybridisation assay (Cunningham, 2004). Colony hybridization rapidly identifies *Vibrio anguillarum* in fish, and detects both pathogenic and environmental strains (Powell and Loutit, 2004). Real-time quantitative PCR (qRT-PCR) offers quantification and high sample throughput. Real-time PCR methods have recently been developed for a variety of significant fish bacterial pathogens (Bacázar *et al.*, 2007), and many viral pathogens (Hick and Whittington, 2010). Polygenic sequencing of specific genes following PCR identifies some pathogens where differentiation of closely related species is difficult, such as the three different genes necessary to classify some fish mycobacteria (Pourahmed, 2008). Multiplex PCR permits the simultaneous detection of *Aeromonas hydrophila*, *A. salmonicida* subsp. *salmonicida*, *Flavobacterium columnare*, *Renibacterium salmoninarum* and *Yersinia ruckeri* (Altinok, Kapkin and Kayis, 2008), and pathogens in yellowtail (*Seriola lalandi*) and sea bass (*Dicentrarchus labrax*) (Amagliani *et al.*, 2009). Loop-mediated isothermal amplification (LAMP) is faster and simpler and can detect bacterial, parasitic and viral fish pathogens. It is faster and more sensitive than conventional PCR (Notomi *et al.*, 2000) and can be performed in 90 minutes, without the use of a thermocycler, making it

suitable as a field test (Soliman and El-Matbouli, 2005). LAMP uses autocycling strand displacement DNA synthesis, using *Bst* DNA polymerase and at least four specially designed primers (two inner and two outer) to recognize six distinct sequences on the template DNA (Notomi *et al.*, 2000). The reaction time can be reduced using two further primers. Products of LAMP amplification can be visualized by eye with the addition of SYBR Green I to the mixture, or can be detected by photometry due to magnesium pyrophosphate turbidity. Some commercial LAMP kits use an enzyme substrate system to visualize the reaction on a membrane.

Fifteen fish pathogens have been discriminated using microarray technology, and several groups are working on assay development. The method involves hybridizing samples of DNA fragments (amplicons), amplified by PCR, on to specific DNA detector fragments spotted onto a solid support. A large number of DNA spots from different pathogens can be included on a single slide, allowing multiplexing for different pathogens. The method is highly sensitive, specific, has high throughput capacity, reduces costs and increases the speed of diagnosis, but is in its infancy in aquaculture (Kostić *et al.*, 2008).

Prudent and responsible use veterinary medicines

Antimicrobials

As in other animal production sectors, veterinary medicines (particularly antimicrobial agents) are used in aquaculture during both production and processing, mainly to prevent and treat bacterial diseases. Antimicrobial agents are biologically active at very low concentrations, demanding their prudent use. Of their possible adverse effects, the most important is clinically significant resistance in target bacteria, and therefore their treatment can have no beneficial effect and is imprudent. Similarly, their routine prophylactic use, particularly in hatcheries and when the cause of disease is not bacterial, is uneconomic and unjustifiable.

The enormous gains in aquaculture production capacity that have been achieved globally during the past 30 years would not have been possible without the use of veterinary medicines. All antimicrobial agents in use in aquaculture are also used in human or veterinary medicine. There are no antimicrobial agents that have been specifically developed for aquaculture use, and simple economic considerations suggest that this will always be the case (FAO, 2012b).

The *Aquatic Animal Health Code* (OIE, 2011a) recognizes that antimicrobial agents are essential for treating, controlling and preventing infectious diseases in aquatic animals. While continued access to antimicrobials is a priority, direct and indirect adverse effects must be considered.

Direct adverse effects result from the agent being in the environment of the production facility or in the marketed product. Environmental direct effects

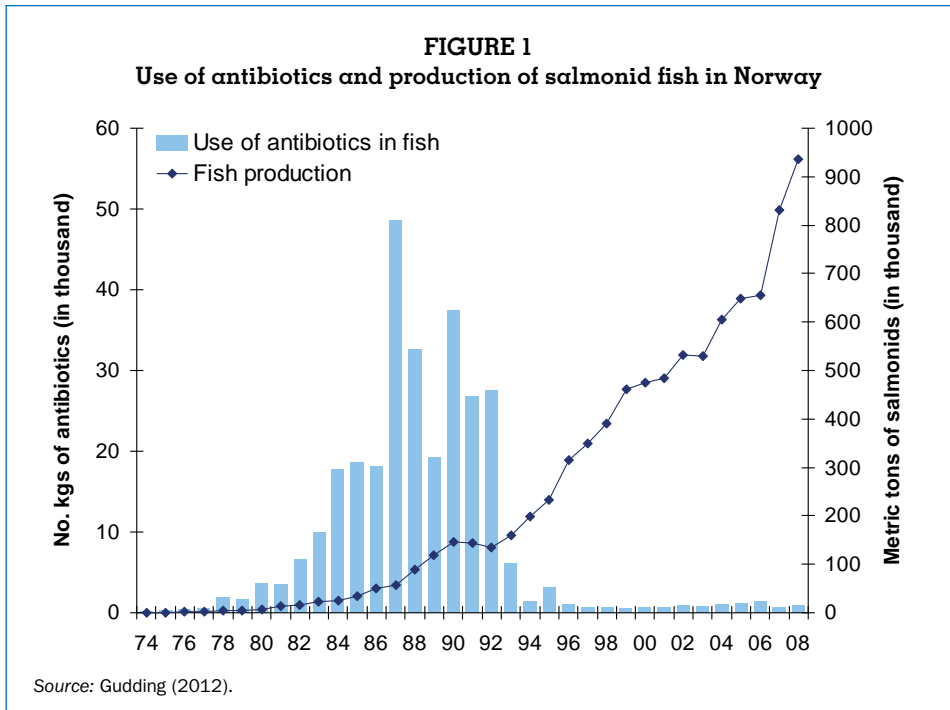
are probably small scale, local and short term. Despite a lack of reports on adverse effects on human health from agents in aquacultural products, their presence has a major influence on market acceptability and on the economics of aquaculture. In the last decade, there have been major improvements in control of residues by regulatory agencies, but a major problem relating to residues is the lack of agents with marketing authorizations (MA) for use in aquaculture. For example, there are no agents with MA for application to shrimp culture. Also, many producer countries regulate agent use by banning unacceptable agents rather than by authorizing usable agents. The setting of maximum residue levels (MRL) and recommended withdrawal times (WT) has been strongly linked to the granting of MA. A major consequence of the lack of MA is the lack of specific evidence-based regulatory MRL and WT values. MRL values can be set by processes that do not require the simultaneous granting of an MA. For example the Codex Alimentarius has set an MRL for oxytetracycline in shrimp. Knowledge of WT is necessary for the prudent use of these agents in aquaculture, and serious consideration should be given to the setting of generic WT. Although these would be conservative, they would provide some much needed guidance.

Indirect adverse effects result from the potential of antimicrobials to selectively enrich resistant variants, which must be considered in two contexts: aquatic animal therapy and human therapy. In aquatic animal health, the main problem is resistance in the bacterial target of therapeutic administration, and ample data show that the agents used in aquaculture have caused significant resistance in target bacteria. Attempts to treat an infection by a resistant bacterium are bound to fail. In human health, although resistance in agents in aquaculture may transfer to human pathogenic bacteria, there is no evidence of this. The frequency of transferable gene-encoded resistance in human pathogens may be highly complex, and limit the applicability or value of formal risk analysis. Three factors must be recognized: (i) resistant bacteria in aquaculture may derive from contamination of the water supply by land-derived resistant strains; (ii) resistant bacteria may occur in aquaculture products from postharvesting contamination; and (iii) for many of the diseases of humans associated with the consumption of fish, antimicrobial therapy is not recommended and, therefore, the occurrence of resistant variants has no relevance.

In most cases, there are no validated test protocols to determine the clinical resistance or sensitivity of target bacteria. Three largely unresolved problems include: (i) harmonization of the test protocols, (ii) setting of interpretive criteria and (iii) development of the laboratory infrastructure to perform the tests.

Vaccines

The use of antimicrobials may be significantly reduced by the use of vaccines, when possible (see Figure 1) (Gudding, 2012). Vaccination has been successful in prevention of bacterial diseases such as vibriosis, furunculosis, yersiniosis, edwardsiellosis, pasteurellosis and other Gram-negative bacterial infections.



Streptococcosis and lactococcosis, caused by Gram-positive bacteria, are preventable by vaccination, but vaccination against intracellular bacteria like *Piscirickettsia* has not been achieved. Prevention of viral diseases has been less successful, with vaccines against infectious pancreatic necrosis virus (IPNV), infectious salmon anaemia virus (ISAV) and other viruses giving some, but not acceptable protection. Vaccines have been developed for diseases of several fish species (i.e. *Salmo salar*, *Oncorhynchus mykiss*, *Dicentrarchus labrax*, *Sparus aurata*, *Ictalurus punctatus*). They are administered by injection, with or without adjuvants, and by immersion. Adjuvants are added when a strong immune response is required, as with furunculosis and most viral diseases. Oral administration of vaccines is also possible, but gives inferior results. Most vaccines are inactivated products. Live vaccines have been developed against diseases which cannot be treated by bacterins, such as a vaccine against *Edwardsiella ictaluri*. Molecular vaccines are available, and a DNA-vaccine has been licenced for use against infectious hematopoietic necrosis (IHN) in salmonids.

Immunoprophylaxis contributes to sustainability of aquaculture by reducing disease prevalence, use of antibiotics, prevalence of antibiotic-resistant bacteria, and prevalence of residues in aquacultural products. The main side effects are lesions using adjuvanted vaccines, which may be a welfare problem and may cause melanosis at the lesion site, reducing marketability. The only effective method of vaccinating small fish is by immersion or oral administration, and inactivated vaccines may be non-protective because of low antibodies

and insufficient cellular immunity. Consequently, live vaccines or recombinant vaccines for immersion or oral administration might be the only type of vaccine giving acceptable protection.

Live vaccines can be developed by attenuation of pathogenic bacteria by passages through media or tissue culture. Addition of rifampicin to the medium has been successful for attenuation of Gram-negative bacteria. Use of low-pathogenic micro-organisms as live vaccine gives protection against bacterial kidney disease (BKD) (*Renibacterium salmoninarum*). Genetic modification has been used for inactivated vaccines by insertion of genes into vectors for large production of virulence factors. Development of live vaccines can be achieved by deletion of virulence factors, making mutants which are safe to use. As vaccines for aquatic animals are released into the environment, live vaccines may pose risks. Vaccines may be developed against fungal diseases and parasites, such as epizootic ulcerative syndrome (EUS) and salmon lice (*Lepeophtheirus salmonis*), but not in the near future. Development of such vaccines will allow antibiotics and chemotherapeutants to be reserved for emergencies.

Health management tools: the manufacturer's point of view

Several types of veterinary medicines exist and are registered for aquatic species (Wardle and Boetner, 2012). These include the following:

- Vaccines – These are products that are directly or indirectly produced from the pathogen and administered to the animal to elicit a specific (lasting) immune response for the prevention of a range of mainly bacterial and viral diseases. Vaccines are widely used in intensive farming conditions world-wide. They are supplied as immersion, oral or injection preparations. Vaccines provide pathogen-specific disease prevention.
- Antibiotics – For treatment and cure of bacterial infections in fish.
- Antiparasitic products in feed or bath – For the treatment of external parasites (e.g. sea lice, *Benedenia*).
- Antifungal disinfectants – For eggs and infected fish.
- Immunostimulants designed to enhance the natural non-specific immune parameters of fish and shrimp to defend against mild infections and environmental stress that might trigger outbreaks.

The manufacture and production of medicines and health products for aquatic animals follows a tedious process that requires full engagement with producers, veterinarians and aquatic animal health professionals, feed companies, and regulatory bodies. The work transcends quality assurance programmes, best practices schemes to ensure that products are both efficacious, as well as safe for consumers, the fish farmers, the fish and the environment. The cycle for developing and managing a veterinary medicine for aquaculture follows a lengthy process starting from the identification of a disease and its underlying cause. The next steps involve finding a cure. The discovery of a compound that is effective against a pathogen leads to the product development phase. This

requires a high level of investment and expertise, and a great deal of work is undertaken with the active compound or the vaccine antigen to document its quality, safety and efficacy, addressing the regulatory requirements and above all, to ensure that control systems are in place to guarantee the same product standards throughout. The cost and complexity of the work means that for pharmaceutical products destined for use in aquaculture, the active ingredients will usually be registered for other animal species or other larger markets than aquaculture as well. Vaccines, however, are specifically developed and registered for aquaculture. The registration package covers all aspects of the product, and most of the data generated must come from the final product formulation that will be, or is intended to be placed on the market. The data cannot be extrapolated from other similar formulations or manufacturers.

Development documentation is generated covering the manufacturing processes and procedures, quality control checks and validated pass criteria for each stage of the manufacturing process. Compliance with the process and procedures is key to ensuring the consistency and reliability of the medicine being produced. This is critical for the on-farm performance, but even more importantly, to ensuring that the fish is safe and wholesome for human consumption.

Before an active ingredient can be developed into a medicine, a number of issues need to be evaluated and fully understood. These include: pharmacological properties of the active ingredient, toxicity issues, mutagenicity, carcinogenicity studies, immunotoxicity, microbial properties of residues, target animal safety and environmental issues.

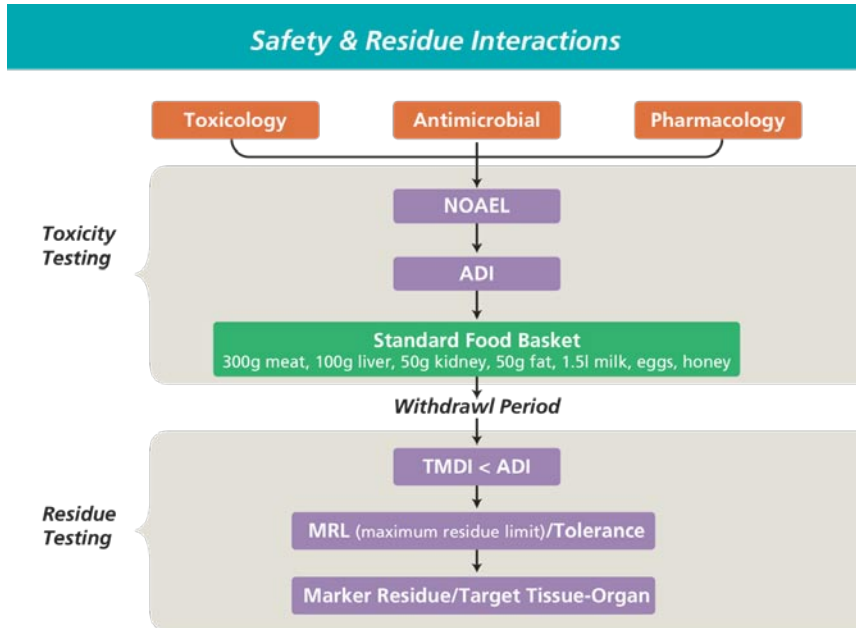
Figure 2 shows that the toxicological/safety development work allows an acceptable no observed adverse effect level (NOAEL) to be established. The acceptable daily intake (ADI) is then calculated from this level. This establishes how much of the active ingredient or its metabolites can be consumed without posing a risk to the consumer. The ADI is then compartmentalized between the components of the “standard food basket”, with fish being included in the daily meat ration (300 g). This is used to establish the maximum residue limit (MRL) that can be accepted in fish. This is measured in the edible tissues, which are considered to be the fillet, i.e. muscle with normal proportion of skin attached.

Once an MRL is established, the manufacturing company must demonstrate that the formulated product used under the recommended conditions will deplete to ensure that the active compound and or its metabolites will be at levels lower than the MRL after the defined withdrawal period has elapsed.

The implementation of the human food safety procedures is important both in the country where the fish are produced as well as in the country of destination for exported products. International (i.e. Codex Alimentarius) and national requirements have to be strictly followed to ensure that safety requirements

FIGURE 2

Diagram describing the steps and procedures required to establish an acceptable withdrawal time for a pharmaceutical medicine. (NOAEL – no observed adverse affect level; ADI – acceptable daily intake; TMDI – total maximum daily intake; MRL – maximum residue limit)



Source: Wardle and Boetner (2012).

of the importing countries are fully met. These are usually enforced by port of entry inspections. When a farm uses a registered medicine in the correct way and follows the guidelines for withdrawal, they can be confident that the use of the product does not result in a product that contains a harmful residue or causes any disruptions in the trade of foods. This approval process ensures that the medication used is safe for the consumer, the environment, the user and of course, for the fish, that it is efficacious and is produced to an approved quality standard.

Once the medicine has been approved, the manufacturing company continues to bear the responsibility for the marketing and technical support for the product. The pharmaceutical company has to follow specific pharmacovigilance responsibilities to monitor any unexpected problems (adverse reactions) which may arise with the use of the medicine in the field. In addition to the above responsibilities, the manufacturer plays an important role in supporting veterinarians and aquatic animal health professionals and farmers in achieving the best performance from the medicines that they use and rely on to achieve their production goals.

Food-borne human infections from aquatic products

Food safety also includes the elimination of food-borne human infection from aquatic products. While enterobacterial agents such as *Salmonella* do occur in fishery products, such contamination is uncommon. Non-typhoidal salmonellae cause an estimated 1.4 million illnesses in the United States of America each year, but only about 5 percent of *Salmonella* infections in the United States of America are due to seafood. Analysis of 11 312 imported and 768 domestic seafood in the United States of America during 1990–1998 revealed that 10 percent of imported and 2.8 percent of domestic raw seafood was positive for *Salmonella* and the overall incidence was 7.2 percent for imported and 1.3 percent for domestic seafood. *Salmonella* has been isolated from freshwater catfish ponds (5 percent prevalence) in the United States of America and from eel culture ponds in Japan (21 percent prevalence), and it has been found in 16 percent in shrimp and 22.1 percent in mud/water in Southeast Asia, and in 30 percent of cultured United States channel catfish and 50 percent of Vietnamese catfish.

Fishborne zoonotic trematodes (FZTs) are an emerging food safety issue in many Asian countries (Tran *et al.* 2009, Phan *et al.* 2010), particularly those with large aquaculture sectors, and are also receiving increased attention by countries outside Asia (e.g. the United States of America and Europe). The WHO and the FAO have estimated that FZTs infect more than 18 million people, with the global number of people at risk estimated to be greater than 500 million, mainly in Asian countries. Depending on the trematode species, the adult parasites infect the liver or intestine of the final host, which include humans, cats, dogs, pigs and other mammals. The adult fluke produces eggs which are excreted by the host and may contaminate the aquatic environment, where they infect snail species in which further development and multiplication occur (Skov *et al.* 2009). Free-swimming cercarial parasites are released from the snail and penetrate into the fish. The final host is then infected by eating raw or prepared fish containing infective metacercarial parasites.

Common in Viet Nam, FZTs are a significant risk to public health and safety of fish products. There has been a 9.3 fold increase in freshwater fish production in Southeast Asia, including Viet Nam, in the last few decades, with increased concern about the role of aquaculture in transmission of FZTs and a need to prevent or control the transmission of the parasites. The project Fishborne Zoonotic Parasites in Viet Nam (FIBOZOPA; <http://fibozopa2.ria1.org>) addresses this important public health and food safety problem in aquaculture. It works with research institutions, universities and government institutions within human and animal health, aquaculture and natural science to prevent FZTs in Vietnamese aquaculture. There is great variability in the prevalence and intensity of FZT metacercariae starting in fish nurseries, depending on the type of aquaculture and its location. In high-intensity culture (e.g. pangasiid catfish in southern Viet Nam), FZT metacercarial prevalence is generally less than 5 percent, whereas in

more extensive ponds (e.g. household-based carp ponds in northern Vietnam) infection rates are less than 90 percent. The parasites are mainly intestinal flukes, in particular *Haplorchis* spp. In rural Viet Nam, food fish are often taken directly from ponds, rivers and lakes, so it is important to prevent FZT infection at the preharvest level. For exported fish species, e.g. pangasiid catfish, FZT prevalence must be low enough to meet the food safety standards of importing countries. As prolonged freezing at -20 °C kills all parasites in fish products, exported frozen fish products are safe for human consumption.

Less attention has been given to animals as reservoir hosts in the epidemiology of FZTs than to humans. A FIBOZOPA study of an aquaculture community found farmers had only 0.6 percent prevalence of FZTs, but fish from aquaculture ponds had very high prevalences. Cats, dogs and pigs had FZT infections of 48.6 percent, 35.0 percent and 14.4 percent, respectively, with seven species of adult zoonotic flukes. Domestic animals are therefore reservoir hosts for FZTs (Nguyen *et al.* 2009), and drug treatment of the humans alone will not prevent transmission of FZTs to cultured fish.

Snails are critical in control and prevention of metacercariae in fish, but extensive surveys of intermediate host snails in fish ponds and other habitats have not revealed snails infected with *Clonorchis sinensis*, while several species (*Melanooides tuberculata*, *Sermyla riquetii*, *Thiara scabra*) were infected with different species of intestinal trematodes.

The potential risks for parasite transmission have been assessed in epidemiological studies in nurseries and grow-out ponds. Hazards identified include poor water quality, presence of snails, faecal contamination from infected animal and human reservoir hosts, and the use of untreated animal manure as pond fertilizer. To address these risks, an intervention study at pond level has been introduced in Viet Nam. The interventions are low cost and can be easily implemented and managed by farmers, building on their existing skills with only limited training. The programme can be integrated into general programmes on biosecurity and best management practices (BMPs) related to aquatic animal health management and to overall good farm management. As a large amount of the fish that are eaten in rural areas do not pass through a processing plant, the pond-level food safety interventions are important for the public health in the rural areas.

Use of specific pathogen free (SPF) stocks

Since the publication of the *Bangkok Declaration and Strategy for Aquaculture Development Beyond 2000*, a major revolution in shrimp cultivation has occurred, with the widespread adoption of domesticated and genetically improved whiteleg shrimp (*Litopenaeus vannamei*) as the cultivated species of choice. This has fulfilled one of the recommended interventions of the Bangkok Declaration (i.e. “developing and utilising improved domestication and broodstock management

practices and efficient breeding plans to improve production in aquatic animals”). The resulting change in shrimp aquaculture production output from approximately 1 million tonnes in 2004 to 3.2 million tonnes (more than triple) in 2007 (*FishStat plus*, FAO, 2010a) is a testament to how effective such interventions can be. On the other hand, it should not be assumed that this increase in production was due solely to introduction of the new stocks, since it was accompanied by a suite of other advances, particularly regarding biosecurity and disease control.

Use of SPF shrimp in biosecure hatcheries (i.e. hatcheries that exclude free viruses and their carriers) can virtually eliminate viral transmission risk via postlarvae used to stock rearing ponds. Biosecurity includes the need to cover outdoor nursery tanks to exclude potential insect carriers. Use of locally captured wild shrimp as broodstock for postlarval production to stock rearing ponds is always accompanied by a high risk that they will carry one or more known or unknown viruses without showing signs of infection, and that they will transmit these viruses to their offspring in shrimp hatcheries. Using captured broodstock tested for known viruses and spawned individually for individual larval rearing in biosecure facilities can reduce this viral transmission risk, but never to zero. That is the reason for mandatory development of domesticated SPF stocks for any shrimp species targeted for sustainable industrial production. Another risk for hatcheries is the continued use of live feeds. A long-term target should be to remove all live feeds from broodstock and larval diets and to substitute them with defined, dried feeds that are free of shrimp pathogens. Targets for replacement include such things as live algal feeds, *Artemia*, polychaetes and squid meat.

The paramount need for SPF domesticated shrimp stocks in sustainable shrimp aquaculture is based on a prime biosecurity issue for shrimp and other crustaceans that differs markedly from vertebrate species. The latter are often capable of clearing viral pathogens from their systems during suitable periods of quarantine. By contrast, crustaceans often carry (and share among species) one or more viral pathogens (even lethal ones) as persistent infections for long periods (up to a lifetime) without showing any gross signs of disease. Although these viruses are often present at low levels, they are active and can be passed on to other naïve shrimp or other crustaceans that may suffer lethal infections. They can also be passed from the broodstock to their grossly normal larvae and postlarvae, either naturally or in a hatchery, and this may lead to subsequent disease outbreaks in rearing ponds stocked with the infected postlarvae. This propensity of grossly normal crustaceans to carry known and unknown viral pathogens means that special precautions are needed whenever living crustaceans destined for aquaculture are translocated over large geographical distances, and especially to areas outside their natural range (Flegel, 2006c). Unfortunately, disregard for this propensity has resulted in several major shrimp virus epidemics (epizootics), most notably for *Penaeus stylirostris* densovirus

(PstDNV) (formerly called infectious hypodermal and hematopoietic necrosis virus or IHNV) in the blue shrimp (*Litopenaeus stylirostris*) and the whiteleg shrimp (*L. vannamei*) in the Americas, WSSV in all cultivated shrimp in Asia and the Americas (Flegel, 2006b), Taura syndrome virus (TSV) in *L. vannamei* cultivated in Asia (Nielsen *et al.*, 2005) and most recently, infectious myonecrosis virus (IMNV) in *L. vannamei* cultivated in Indonesia (Senapin *et al.*, 2007).

Every country should be wary of importing exotic crustaceans of any kind for aquaculture without going through the recommended risk analysis and quarantine procedures, combined with tests for unknown viruses that might be a danger to local species (Flegel, 2006c). Risk analysis is necessary to assess emerging threats from new or exotic species (Arthur *et al.*, 2009). These biosecurity measures should be applied even to exotic domesticated stocks that are SPF for a list of known pathogens. To reduce risks to the minimum, any country that imports exotic stocks for aquaculture should invest in establishment of local breeding centers comprised of properly vetted stocks that could be used for ongoing supply of broodstock and postlarvae to stock cultivation ponds. This would avoid the continual risk of importing unknown pathogens that might be associated with continuous importation and direct use of exotic stocks, even from a foreign breeding center that produces SPF stocks.

An allied issue concerns the co-cultivation of one shrimp species with one or more other shrimp species or with other crustacean species. For example, rearing of captured *Penaeus monodon* and exotic SPF *L. vannamei* in an Asian shrimp hatchery would be a good way to transfer endemic PstDNV from *P. monodon* to *L. vannamei* at the larval stage. In another example, it has recently been shown that *Macrobrachium rosenbergii* nodavirus (MrNV) (the cause of white muscle disease in *M. rosenbergii*) can infect larvae of *P. monodon* and *Fenneropenaeus indicus* and result in high mortality from white muscle disease (Ravi *et al.*, 2009), even though it does not cause mortality in challenged juvenile shrimp of the same two species (Sudhakaran *et al.*, 2006). In summary, there are good reasons to avoid mixed cultures of shrimp or other crustaceans unless one is very, very certain that negative viral interchanges are not possible.

Living modified organisms/genetically modified organisms

The rise of molecular genetics and the development of biotechnology are hallmark scientific achievements of the past three decades. Advances in biotechnology offer the potential for significant improvements in human well-being, so long as adequate measures are taken to safeguard human health and the environment. These concerns were recognized by those who negotiated the Convention on Biological Diversity (CBD), signed by most countries of the world in 1992. In Article 19.3 of the CBD, the Contracting Parties agreed to consider the need for developing appropriate procedures to address the safe transfer, handling and use of any living modified organism (LMO) resulting from application of biotechnology that may have an adverse effect on the conservation and sustainable use of

biodiversity. The Cartagena Protocol on Biosafety, a supplementary agreement to the CBD adopted in 2003, governs the movements of LMOs from one country to another. A living modified organism (LMO) is defined in the Cartagena Protocol as any living organism that possesses a novel combination of genetic material obtained through the use of modern biotechnology (UNEP, 2009). LMOs are generally considered to be the same as genetically modified organisms (GMOs). While different classes of organisms have been included in the term GMO – including organisms modified by gene transfer, chromosome set manipulation, and interspecific hybridization – discussion has focused upon transgenic organisms; hence, this contribution focuses upon transgenic aquatic organisms. A transgenic fish or shellfish bears within its chromosomal DNA a gene construct – i.e. a transgene, a gene whose expression is under novel regulation – that was introduced by human intervention. The benefits, risks, and management of risks posed by aquatic GMOs are described below.

Benefits posed by aquatic GMOs

A number of different traits have been targeted for genetic improvement via gene transfer, including growth rate, freeze resistance, disease resistance, phytate utilization, reproductive confinement and completion of biosynthetic pathways (Table 2). Most transgenic lines have not been subject to the generations of

TABLE 2
Examples of gene transfers in fish targeting aquaculture production traits

Targeted trait	Species	Transgene	Reference
Rapid growth	Atlantic salmon (<i>Salmo salar</i>)	Growth hormone	Du <i>et al.</i> , 1992
	Coho salmon (<i>Oncorhynchus kisutch</i>)	Growth hormone	Devlin <i>et al.</i> , 1994
	Common carp (<i>Cyprinus carpio</i>)	Growth hormone	Hinitz and Moav, 1999
	Mrigal carp (<i>Cirrhinus cirrhosus</i>)	Growth hormone	Venugopal <i>et al.</i> , 2004
	Mud loach (<i>Misgurnis myzolepis</i>)	Growth hormone	Nam <i>et al.</i> , 2001
	Nile tilapia (<i>Oreochromis niloticus</i>)	Growth hormone	Rahman <i>et al.</i> , 2001
	Disease resistance	Channel catfish (<i>Ictalurus punctatus</i>)	Cecropin
Grass carp (<i>Ctenopharyngodon idella</i>)		Lactoferrin	Mao <i>et al.</i> , 2004
Freeze resistance	Atlantic salmon	Antifreeze polypeptide	Hew <i>et al.</i> , 1999
	Goldfish (<i>Carassius auratus</i>)	Antifreeze polypeptide	Wang <i>et al.</i> , 1995
Phytate utilization	Nile tilapia	Phytase	Kemeh, 2004
Reproductive sterility	Rainbow trout (<i>O. mykiss</i>)	Gonadotropin releasing hormone anti-sense mRNA	Uzbekova <i>et al.</i> , 2000
		<i>zBMP2</i> , a dorsoventral developmental patterning gene	Thresher <i>et al.</i> , 2009
Vitamin C synthesis	Rainbow trout	L-gulonono- γ -lactone oxidase	Krasnov, Pikanen and Molsa, 1999

breeding needed to develop a homozygous line stably expressing the transgene. However, development of some growth hormone (GH)-transgenic lines is well advanced, and efforts to commercialize them are ongoing, including Atlantic salmon (*Salmo salar*) in the United States of America (Fletcher *et al.*, 2004), tilapia in Cuba, and common carp in China (Wu, Sun and Zhu, 2003). With the prospect of improved production efficiency, it is not surprising that some aquaculturists want to produce GH-transgenic fish commercially.

Risks posed by aquatic GMOs

Commercial aquaculture operations have a routine, often significant escape of fish through equipment failures, handling or transport operations, predator intrusion, storm damage or other mechanisms. Although farm operators attempt to prevent escapes by upgrading confinement systems, installing predator deterrent devices, and other actions, it still must be assumed that escapes will occur. Escape of cultured fish into the accessible ecosystem and ecological or genetic interactions with local intraspecific and interspecific populations pose environmental concerns (McGinnity *et al.*, 2003). Ecological concerns focus upon competition for space and food resources and direct predation (Gross, 1998). Genetic concerns include the potential breakdown of locally adapted traits through interbreeding and introgression, and range up to replacement of native stocks by cultured stocks (Saegrov *et al.*, 1997). Such concerns are posed by the prospect of producing transgenic fish in aquaculture, with additional unknowns posed by possible effects of the transgene.

Ecological risk assessment for transgenic fish is based upon case-by-case assessment of the host species, transgene, site of genomic integration, and receiving ecosystem (Kapuscinski and Hallerman, 1990). Potential hazards at issue are illustrated by empirical studies with GH-transgenic fishes. To support their rapid growth, GH transgenics require more energy, and hence will feed more actively than non-transgenic fish; for example, increased feeding rate, feeding competition and willingness to feed in the presence of a predator have been observed in Atlantic salmon (Abrahams and Sutterlin, 1999), coho salmon (Devlin *et al.*, 2004) and common carp (Duan *et al.*, 2009). The effects of introgression of a transgene into a receiving population will vary among receiving populations (Devlin *et al.*, 2001) and environmental conditions, including food availability (Devlin *et al.*, 2004), and may result in decreased demographic viability of the resulting population. Models have been developed to predict the genetic and demographic effects of interbreeding of transgenic and non-transgenic fish (Muir and Howard, 1999) but have yet to be empirically validated. General frameworks for quantifying ecological (Devlin *et al.*, 2007) and genetic (Kapuscinski *et al.*, 2007) risks have been developed. Ecological and genetic risks have not been well investigated for transgenes other than growth hormone. Further, because exact probabilities of risk are difficult or impossible to determine for all types of possible harm, it may be necessary – based on current knowledge of population genetics, population dynamics, receiving ecological communities and experience

with cultured stocks – to classify levels of concern regarding likely genetic impacts posed by cultured stocks into qualitative categories ranging from low to high.

Risk management

Under at least some circumstances, escaped transgenic fish could negatively impact accessible ecosystems and populations. The best approach for minimizing the likelihood of harm becoming realized is to minimize exposure to the hazard, in this case, escaped transgenic fish. Differences in species, production traits, receiving ecosystems and culture systems will affect the case-by-case determination of appropriate risk management measures for experimental and commercial (Mair, Nam and Solar, 2007) production systems. Risk might be managed by producing transgenic fish only under conditions of confinement; in high-risk contexts, production of transgenic fish might go forward only under conditions of strict confinement aimed at ensuring no escape of transgenic fish into the accessible ecosystem. Three non-mutually exclusive approaches to achieving confinement of aquatic GMOs include: (i) physical confinement, (ii) reproductive confinement and (iii) operations management. Achieving effective physical confinement of cultured aquatic organisms will require a combination of careful selection of production site, production system, barriers to escape of cultured organisms, and barriers to animal or human intrusion onto the site (ABRAC, 1995; Mair, Nam and Solar, 2007). Lack of reproduction would prevent loss of difficult-to-confine early life stages from the culture facility or establishment of a population of escaped transgenic fish in the accessible ecosystem. Reproductive confinement might be approached by production of monosex or triploid stocks (Mair *et al.*, 1997; NRC, 2004), although neither approach is likely to prove 100 percent effective. Transgenic approaches to reproductive confinement are under development, although progress is slow. Operations management measures are needed to: (i) ensure that normal activities of workers at the aquaculture operation are consistent with the goal of effective confinement, (ii) prevent unauthorized human access to the site and (iii) ensure regular inspection and maintenance of physical confinement systems. Combinations of risk management measures are advisable so that failure of any one measure will not lead to escape of confined stocks.

Over the past ten years, the following trends in technical advancements and development of national capacity for technology oversight have been observed. While most early gene transfer experiments targeted growth rate by introduction of growth hormone transgenes, recent work has targeted a greater range of traits, often utilizing structural genes not found in the host genome. Of relevance here, interest in promoting bioconfinement of cultured stocks led to gene transfers aimed at inducing reversible sterility (Wong and Van Eenannaam, 2008). The past ten years have seen elaboration of empirical data on risk assessment, mostly on salmonids, and to a lesser degree with model species such as medaka (Japanese ricefish, *Oryzias latipes*) and other aquaculture species such

as tilapias and carps. The range of issues posed by a proposed utilization of transgenic fish in aquaculture led to elaboration of a protocol for oversight of aquatic GMOs within a three-stage, interactive framework (Hayes *et al.*, 2007). Because all potential harms and associated pathways cannot be known and precisely predicted *a priori*, it will be necessary to update the risk analysis as knowledge accumulates using an adaptive management approach (Kapuscinski, Nega and Hallerman, 1999). The decision of whether and under what conditions production of transgenic fish would go forward will be made at the national level. Under Article 21 of the CBD and the Cartagena Protocol, signatories commit to developing and implementing policies for oversight of biotechnology. Consequently, countries including Cuba, Thailand, China, Chile and Peru are developing and implementing policy and staffing government offices that would consider applications for production of transgenic fish.

Biological invasions

Biological invasion is one area that was not addressed in the 2000 Bangkok Declaration and Strategy. The human-mediated introduction of marine species is increasingly recognized as a threat to sustainable management of marine ecosystems and the maritime economies of coastal nations (Molnar *et al.*, 2008), yet in most regions of the world, the scale and scope of marine introductions are poorly known (Carlton, 1996; Hewitt, 2002; Hewitt and Campbell, 2008). Unlike the long history of recognition of freshwater introductions, marine introductions have only been investigated over the last 40 years, during which marine and estuarine introductions have been detected worldwide (Ruiz *et al.*, 2000; Hewitt, 2003; Molnar *et al.*, 2008; Hewitt and Campbell, 2008) by literature evaluation (Carlton, 1996; Ruiz *et al.*, 2000; Rilov and Crooks, 2009) and general biodiversity surveys or targeted surveys (Coles *et al.*, 1999; Hewitt, 2002). In a recent comprehensive evaluation of global marine and estuarine invasions (Hewitt and Campbell, 2008) based on over 700 data sources, 1 781 invasive species were identified representing 27 phyla; over 55 percent of the species were arthropods, molluscs and chordates (fishes and ascidians). Using life histories and literature-based evidence, over 98 percent of the 1 781 species were linked to possible transport vectors. Where species-level information was not readily available, genus-level characteristics were used to classify morphological characteristics and habitat associations. Most species had life histories allowing transport by vessels (biofouling ~55.5 percent, ballast water ~30.8 percent, historic dry-ballast ~2.3 percent). Intentional movements (e.g. for fisheries stocking, aquaculture development, biocontrol efforts, aquarium trade, live seafood trade, scientific research) involved less than 15 percent of translocated species.

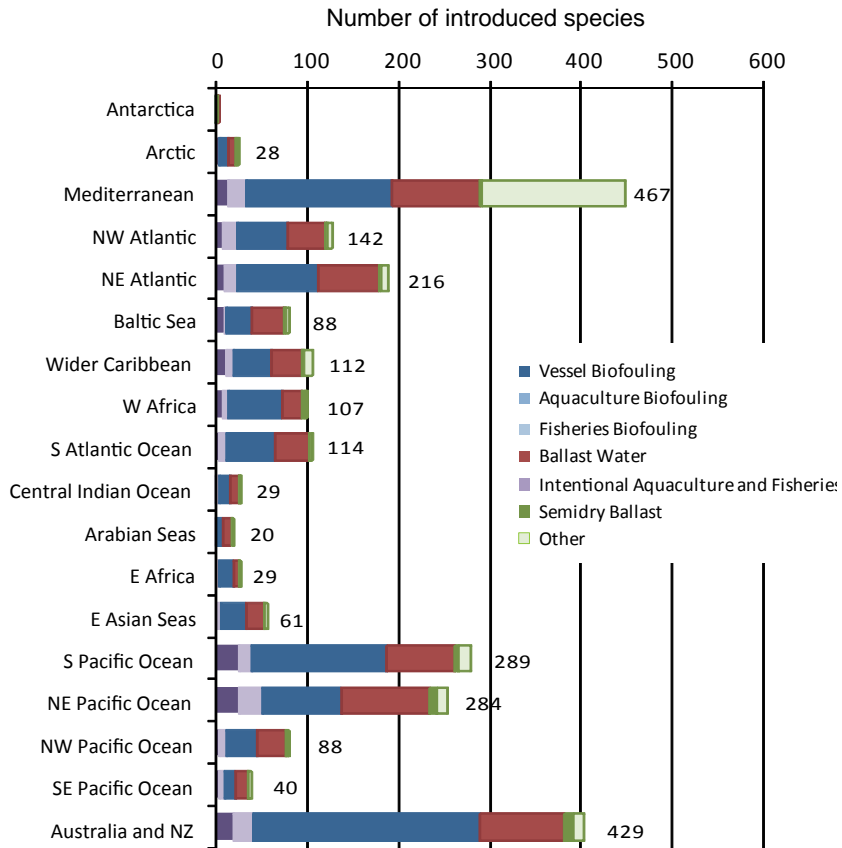
Not all bioregions of the world have experienced the same numbers or rates of biological introductions (Figure 3). An apparent acceleration of introductions, attributed to increased awareness and increasing vessel movements, has been reported in San Francisco Bay (Cohen and Carlton, 1998) and Pearl Harbor (Coles

et al., 1999), United States of America, and in Port Phillip Bay, Australia (Hewitt *et al.*, 2004) and other regions (Hewitt, 2003). Global organizations identify the need for prevention and management of transboundary marine invasions (CBD, 1992; FAO, 1995). Intentional introductions, through, for example, trade, aquaculture and live seafood, are being better controlled, and the attention is now on unintentional introductions.

The International Maritime Organization's Marine Environmental Protection Committee (IMO MEPC), adopted the International Convention on the Control and Management of Ships' Ballast Water and Sediments on 13 February 2004 (BWM, 2005). This convention aims to "prevent, minimise and ultimately eliminate the risks to the environment, human health, property and resources arising from the transfer of harmful aquatic organisms and pathogens through the control and management of ships' ballast water and sediments" through enforcement of guidelines and encouraging development of new ballast water treatment technologies (Gollasch *et al.*, 2007). Such current technologies include elimination through filtration and hydrostatic pressure, temperature, ozonation, ultra-violet (UV) light exposure and the use of chemicals. The majority of global invaders are transported as biofouling (Hewitt and Campbell 2008) comprising the living organisms associated with the external surfaces of a vessel, including protected areas (e.g. sea-chests, internal piping, anchor lockers, ballast tanks), which is highly diverse (Coutts *et al.*, 2010). Despite being one of the highest biosecurity threats to marine and estuarine environments, biofouling is not addressed internationally, although a recent IMO MEPC workplan includes guidelines for biofouling management. Management strategies rely on development of new techniques.

Qualitative risk analysis can be used when significant knowledge gaps exist (Hayes and Hewitt, 2000; Arthur *et al.*, 2009). It has been applied to marine biosecurity, including the identification of undesirable species, the evaluation of proposed intentional introductions, for import health standards (Campbell, 2008), identification of high-risk entry points (Gollasch and Leppakoski, 1999), monitoring and compliance control for transport vectors (Hayes and Hewitt, 2000) and identification of vectors (Hewitt and Campbell, 2008) (Figure 4). Risk analysis can be used for prevention, border protection and port-border response, but the quality of the analysis relies on the information available to the assessor (Carlton, 1996; Williamson, 1996; Hewitt *et al.*, 2004). Significant knowledge gaps include: (1) the absence of good baseline information in coastal zones (specifically ports and marinas); (2) knowledge of current and future trading patterns associated with transport vectors, due to new free trade associations; and (3) knowledge of the physical, ecological, environmental, economic and social (including human health) impacts. Until these gaps are filled, marine biosecurity will continue to focus on reactive, stop-gap measures, rather than the international, consistent framework established in the terrestrial environment.

FIGURE 3
Number of marine introductions (introduced and cryptogenic species) in the 18 large-scale International Union for Conservation of Nature (IUCN) marine bioregions according to contribution of specified transport mechanisms: biofouling (vessels, aquaculture species and gear, fisheries gear), ballast water, intentional introductions through aquaculture and fisheries and other. Numbers at the end of bars represent total number of introduced and cryptogenic species identified from the region

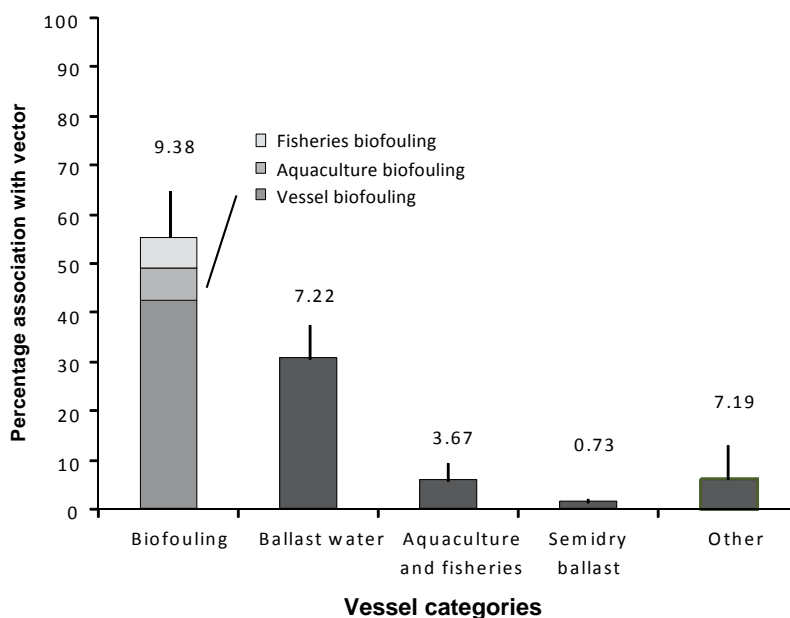


Source: Hewitt and Campbell (2008).

Climate change

Climate change is another area which was not addressed by the 2000 Bangkok Declaration and Strategy. Climate change can be the result of both natural and anthropogenic causes. Aquatic animals are very vulnerable because water is their life-support medium and their ecosystems are fragile. For example, in the case of epizootic ulcerative syndrome (EUS), temperature and rainfall are critical ecological factors for the disease. *Perkinsus olseni*, a major pathogen of molluscs, affects more than 100 host species and is temperature dependant.

FIGURE 4
Average percentage of species in each of the 18 International Union for Conservation of Nature (IUCN) bioregions with potential to be transported by major vector categories. Standard deviations of the mean for each vector are presented by error bars and numbers above the line



Source: Hewitt and Campbell (2008).

Many susceptible hosts are major food commodities. Red tides (harmful algal blooms) are influenced by climate change and spread into new locations through ballast water from ships. Climate change scenarios (e.g. sea level rise, increased incidence of storm surges and land-based run-offs, extreme weather events) that may affect biosecurity (e.g. by increasing range of pests and pathogens, intensities of their occurrence and vulnerabilities of farmed animals to diseases) will also be significant and will need to be addressed (Bondad-Reantaso *et al.*, 2005; Bondad-Reantaso and Subasinghe, 2008; Arthur *et al.*, 2009). Climate change impacts may include change in pathogen virulence and transmission, local extirpations and introductions. There is also the risk of escapes from storm-damaged facilities. The effects on parasites of climate change impacts such as alterations in host distribution, water levels, eutrophication, stratification, ice cover, acidification, oceanic currents, UV-light penetration, weather extremes and human interference also need to be understood. Climate-mediated physiological stresses such as coral bleaching and El-Niño high temperature rise may compromise host resistance and increase the occurrence of opportunistic diseases.

Expectations and commitments expressed in the Bangkok Declaration and strategy

The 2000 Bangkok Declaration and Strategy (Subasinghe *et al.*, 2001) listed the following action plans that will support the sustainable development of aquaculture:

Section 3.11 of the action plans, “Managing aquatic animal health”, includes the following:

- developing, harmonising and enforcing appropriate and effective national, regional and inter-regional policies and regulatory frameworks on introduction and movement of live aquatic animals and products to reduce the risks of introduction, establishment and spread of aquatic animal pathogens and resulting impacts on aquatic biodiversity;
- capacity building at both institutional and farmer levels through education and extension;
- developing and implementing effective national disease reporting systems, databases, and other mechanisms for collecting and analysing aquatic animal disease information;
- improving technology through research to develop, standardise and validate accurate and sensitive diagnostic methods, safe therapeutants, and effective disease control methodologies, and through studies on emerging diseases and pathogens;
- promoting a holistic systems approach to aquatic animal health management, emphasizing preventative measures and maintaining a healthy culture environment; and
- developing alternate health management strategies such as the use of disease resistant, domesticated strains of aquatic animals to reduce the impact of diseases.

Section 3.13 of the action plans, “Applying genetics to aquaculture”, includes:

- developing and utilising improved domestication and broodstock management practices and efficient breeding plans to improve production in aquatic animals.

Section 3.14 of the action plans, “Applying biotechnology”, includes:

- developing and applying biotechnological innovations for advances in nutrition, genetics, health and environmental management; and
- addressing the potential implications for aquaculture of biotechnology, including GMOs and other products, in a precautionary, safe and practical way.

Section 3.15 of the action plans, “Improving food quality and safety”, includes:

- promoting the application and adoption of international food safety standards, protocols and quality systems in line with international requirements such as the Codex Alimentarius; and

- adopting international protocols for residue monitoring in aquaculture and fishery products.

Implementation

During the last decade, aquatic animal health management and biosecurity governance has taken different forms at various levels, involving a wide range of stakeholders. This section takes a close look at examples of what has been achieved, in terms of policy and regulatory frameworks, particularly on introduction and movements of live aquatic animals, capacity building, aquatic animal health information, farm-level biosecurity and better management practices (BMPs). Examples of progress at the global, regional and national levels are presented.

Policy and regulatory frameworks

At the global level, FAO delivers aquatic animal health services under normative and field programmes working with Members, development partners, regional and international organizations, the private sector and the fish farming communities in addressing aquatic animal health biosecurity issues in aquaculture, working on the principle that prevention is better than cure and through targeted capacity building to prevent pathogen introductions. The range of work includes promoting responsible movement of aquatic animals through effective national strategies, national policies and regulatory frameworks and technical guidelines, within the framework of the *FAO Code of Conduct for Responsible Fisheries* (FAO, 1995), as a basis for enhancing compliance with regional and international treaties and instruments (FAO, 2007b); understanding and applying risk analysis to aquaculture that supports timely assessment of threats from new or expanding species (Bondad-Reantaso, Arthur and Subasinghe, 2008; Arthur, Bondad-Reantaso and Subasinghe, 2008; Arthur *et al.*, 2009); detection and identification of the emergence and spread of diseases through surveillance programmes and diagnostic services; emergency preparedness through rapid and timely response (Subasinghe, McGladdery and Hill, 2004; Arthur *et al.*, 2005); empowering and educating farmers with information and tools such as BMPs, simple and practical biosecurity measures at the farm level, as well as organization of farmers into clusters and enhancing outreach programmes to primary producers; and promoting prudent and responsible use of veterinary medicines and vaccines as a preventative strategy (FAO, 2012b). Two of FAO's statutory bodies, i.e., the Committee on Fisheries (COFI) and the Sub-Committee on Aquaculture (SCA), provide a neutral forum for discussions on global concerns affecting aquaculture development. Past sessions of COFI (COFI 28) and SCA (COFI/SCA IV and V) have highlighted the importance of aquatic biosecurity as an essential element for sustainable aquaculture development and the need to support FAO Members to improve their capacity for "preventative actions" as well as "early action capacities" when dealing with biosecurity issues and emergencies.

Between 1999 to 2002, the FAO TCP/RAS 6714(A) and 9065(A) *Assistance for the Responsible Movement of Live Aquatic Animals* – designed to address issues concerning transboundary pathogen transfer, with a view to building capacity in the Asia region for the responsible movement of live aquatic animals – was implemented by the Network of Aquaculture Centres in Asia-Pacific (NACA) with the participation of 21 countries and territories¹. During the implementation period, 12 national, 4 regional and 4 international events (training courses, workshops and consultations) were held. Important lessons from this project include the following:

- An FAO Technical Cooperation Programme paved the way for the development of an *Asia Regional Technical Guidelines on Health Management for the Responsible Movement of Live Aquatic Animals* (FAO/NACA, 2001a,b), establishment of a regional surveillance and reporting system and an aquatic animal health information system.
- Technical support services and expert consultations helped provide a solid understanding of the general principles and the essential elements contained in the technical guidelines.
- Cooperation from member governments who participated through nominated national coordinators for aquatic animal health served as the vital link on the development of national strategies and initiation or implementation of the various provisions of the guidelines.
- Various national projects and/or donor-sponsored activities assisted, to a greater or lesser extent, in monitoring the implementation aspects of the guidelines. Such activities provided information and further guidance on which elements worked well at the ground level (and those that did not) and highlighted the gaps.
- Strong collaboration with partner organizations with similar interests helped in various ways to increase understanding and also to implement the guidelines.
- A supporting implementation strategy using the concept of “phased implementation based on national needs and priorities” provided the impetus for many years of continuous and progressive work on various aspects of aquatic animal health management.
- There was strong recognition that aquaculture development needs to focus on prevention, responsible and better health management practices and maintaining healthy aquatic production (Bondad-Reantaso, 2002; Subasinghe and Bondad-Reantaso, 2008).

FAO provided emergency technical assistance on KHV to Indonesia in 2003 and on EUS in Botswana in 2007. Both activities lead to the development of national (Indonesia) and regional (seven countries bordering the Chobe-Zambezi River) technical cooperation programmes (TCPs) to assist affected countries in

¹ Australia, Bangladesh, Cambodia, China, Hong Kong China, India, Indonesia, Iran, Japan, Korea (D.R.R.), Korea (R.O.), Lao (P.D.R.), Pakistan, the Philippines, Singapore, Sri Lanka, Thailand and Viet Nam.

understanding the disease epidemiology, establishing active surveillance and reducing the risk of further spread (Bondad-Reantaso, Sunarto and Subasinghe, 2007; FAO, 2009b).

One of FAO's core mandates is to provide technical assistance towards building capacities of member governments. Through such mechanisms as TCPs, TCP facilities, programmes funded by extra-budgetary sources, unilateral trust funds and other bilateral arrangements, human and institutional capacity development have been provided both at the national and regional levels. In the Western Balkan region (Bosnia and Herzegovina, Croatia, Macedonia, Monte Negro, Serbia), a regional aquatic animal health capacity and performance survey was conducted by FAO in 2009 (Arthur *et al.*, 2011) which became the basis for developing a regional TCP programme on improving compliance with international standards on aquatic animal health (FAO, 2011). Priority areas identified include the following: building capacity in specific areas (e.g. legislation, risk analysis, surveillance (aquatic epidemiology), diagnostics, emergency preparedness/contingency planning, aquaculture development and promotion); review of national legislation to harmonize with respect to compliance with international standards of aquatic animal health; design of a regional disease surveillance programme for aquatic animal diseases; and promoting communication mechanisms and networking systems for aquaculture development. A similar exercise was done for members of the Regional Commission for Fisheries (RECOFI) (i.e. Bahrain, Iran, Iraq, Kuwait, Oman, Qatar, Saudi Arabia and United Arab Emirates) (Arthur, Reantaso and Lovatelli, 2009) which lead to the development of a regional programme for improving aquatic animal health in RECOFI member countries (FAO, 2009). The priority areas under this programme are: governance (national policy and planning, legislation and regulation), disease diagnostics (national and regional diagnostic laboratories), aquatic biosecurity (guidelines/procedures for new aquaculture species, pathogen risk analysis, disease surveillance, regional emergency response, national and regional pathogen lists, health certification, border inspection and quarantine, disease zoning); access to information (pathogen database, aquatic animal import/export database, legislation database, expert database); and regional cooperation and networking (regional Website and regional meetings). In southern Africa, FAO's work included development of an aquatic biosecurity framework for the region (FAO, 2009a) following the incursion of an exotic fish disease, EUS, in 2006 (FAO, 2009b). The process involved several regional workshops, including a high-level scoping meeting which brought together regional fisheries and veterinary authorities. Through TCPs, FAO also provided assistance to some countries (e.g. Belize, Bosnia and Herzegovina, Latvia) in developing national strategies or policy frameworks on aquatic health or assisting in revising regulations on animal health to include aquatics. The work of FAO in the Pacific region includes promotion of responsible aquaculture development and building capacity for the application of risk analysis in aquaculture implemented through several TCPs and TCP facilities.

The World Organisation for Animal Health (OIE) promotes animal health and public health, especially in the area of international trade of animals and animal products by issuing harmonized sanitary standards for international trade and disease control, by working to improve the resources and legal framework of veterinary services and aquatic animal health services and by helping OIE Members comply with OIE standards, guidelines and recommendations consistent with the World Trade Organization's (WTO) Sanitary and Phytosanitary Agreement (SPS Agreement) (Bastiansen and Mylrea, 2010). The OIE *Aquatic Animal Health Code* and *Manual of Diagnostic Tests for Aquatic Animal Diseases* (OIE, 2011a,b) continues to be updated on a regular basis, with OIE working with OIE aquatic animal disease experts and OIE Reference Laboratories. The OIE Aquatic Animal Health Standards Commission proposes appropriate methods for surveillance, diagnosis and disease prevention and control for safe trade and international movement of aquatic animals and their products with reference to the diseases listed in the OIE aquatic code. The Commission oversees the production of the code and the manual and promotes its distribution and use by veterinary and other competent authorities (Enriquez, 2010). The World Animal Health Information System (WAHIS) was set up by OIE to fulfill one of OIE's missions to ensure the transparency of the world animal health situation. There have already been agreements signed between OIE and, for example, the Organismo Internacional Regional de Sanidad Agropecuaria (OIRSA) and NACA as "Regional Cores" for WAHIS (Jebara, 2010). Recently OIE Delegates have been requested to designate focal points in several fields, including aquatic animal diseases. A network of focal points on aquatic animal diseases has been formed, with OIE providing the necessary learning and training opportunities in the role in the standard-setting process (Petrini, 2010). Another initiative is the performance of veterinary service (PVS). The OIE PVS Tool is designed to assist veterinary services to establish their current level of performance, to identify gaps and weaknesses in their ability to comply with OIE standards, to form a shared vision with stakeholders and to establish priorities and carry out strategic objectives.

At the regional level, in Asia, the Network of Aquaculture Centres in Asia-Pacific (NACA), an intergovernmental organization of 18 governments, works on the principles of cooperation and sharing regional resources among stakeholders (governments, institutions, individuals) and assists member governments to "*reduce the risks of aquatic animal diseases impacting the livelihoods of aquaculture farmers, national economies, trade, environment, and human health*". Table 3 shows the status of implementation of the *Asia Regional Technical Guidelines on Health Management for the Responsible Movement of Live Aquatic Animals* (FAO/NACA, 2001a,b) by the 21 participating Asia-Pacific governments over the last ten years.

Good progress has been made in disease diagnosis, aquatic animal health certification and quarantine, disease surveillance and reporting and farm-level

TABLE 3
Implementation of elements of the Asia Regional Technical Guidelines on Health Management for Responsible Movement of Live Aquatic Animals (FAO/NACA, 2001a,b) by Asian countries by 2008

Elements of the technical Guidelines	Progress made (Number of countries)		
	Good	Moderate	Low
Disease diagnosis	10	6	5
Health certification and quarantine measures	10	5	6
Disease zoning	3	3	15
Disease surveillance and reporting	8	8	5
Contingency planning	3	7	11
Import risk analysis (IRA)	4	4	13
National strategies and policy frameworks	11	4	6

health management, but progress in contingency planning, zoning and import risk analysis (IRA) has been rather limited. Three FAO/NACA regional workshops were held on the diagnosis of molluscan diseases. IRA was taught at an APEC Fisheries Working Group-funded project, “Capacity and Awareness Building on IRA for Aquatic Animals,” implemented by NACA during 2002–2004. IRA is being increasingly used by regional countries to make decisions on intentional introductions of live aquatic animals. AusAid has supported two aquatic animal health projects – (1) “Strengthening Aquatic Animal Health Capacity and Biosecurity in ASEAN” and (2) “Guidelines on Responsible Movement of Live Food Finfish in ASEAN”. These projects, implemented between 2006 and 2008, directly supported capacity building, harmonization and trade facilitation within the Association of Southeast Asian Nations (ASEAN). One of the most important achievements in the region was the formation of NACA’s Regional Advisory Group (AG) on Aquatic Animal Health, a select group of senior aquatic animal health specialists from the region tasked to provide high-level technical advice to NACA member governments. The AG meets annually to discuss important and emerging aquatic animal health issues affecting the Asia-Pacific region, as well as contributing vital disease information to relevant organizations such as the OIE and FAO. NACA has been contributing to the strengthening of regional health management and biosecurity through (i) capacity building (diagnostics, epidemiology, sampling, surveillance, risk analysis, contingency planning); (ii) development of resource material (technical guidelines, manuals, diagnostic guides, field identification guides, disease cards, extension brochures); and (iii) provision of technical assistance at the farm/local/national/regional levels. New issues such as food safety, emerging diseases and continued introductions of exotics to the region are being given special attention. NACA has embarked on a new regional initiative – identifying and establishing a three-tier regional resource base – to utilize the regional technical resources available to member countries. This includes, Regional Resource Experts, Regional Resource Centres and Regional Reference Laboratories for diseases not listed by the OIE. The capacity for disease diagnosis and that of the regional disease laboratories

has greatly increased in the last decade. There are now regional OIE reference laboratories for EUS, white tail disease in *Macrobrachium rosenbergii* and white spot disease in penaeid shrimp.

In the countries of Latin America and the Caribbean (LAC), unified development of regional aquatic animal health strategies is more recent than in Asia. Most LAC countries have general laws on sustainable fisheries and aquaculture containing articles related to aquatic biosecurity (e.g. programmes for aquatic animal health, aquatic food safety, reduction of environmental impacts), which may be supported by by-laws, technical norms and regulations. However, laws are often not applied because of lack of financial resources and weak decision-making, particularly in poorer countries. While legal frameworks and institutional arrangements permit exportation and importation of aquatic products, there is an urgent need for capacity building on risk analysis. In 2004, an Inter-American Committee of Aquatic Animal Health was created to fulfill the OIE international standards for aquatic animal health. Membership includes representatives from the private and public sectors (Martínez *et al.*, 2008). The objectives of the committee are to:

- establish direct contact with experts;
- develop strategies to fulfill OIE norms and guidelines and promote their application;
- improve harmonization of scientific and veterinary services;
- promote modifications to the OIE standards;
- improve diagnostic capacity;
- promote better surveillance systems;
- identify needs and promote capacity;
- strengthen structures and legal frameworks;
- make OIE notification procedures transparent in the region;
- harmonize technical methodologies;
- propose meetings on the objectives of the committee;
- identify experts and reference laboratories;
- facilitate bilateral adoption sanitary measures in relation to the OIE *Aquatic Animal Health Code*; and
- encourage the control of biological residues and veterinary drugs.

In 2008, the recommendations of the committee were to:

- define animal welfare for aquatic animals;
- identify an overseer of agreements, technical groups, and ensure regional capacity building;
- promote capacity building and training in aquatic animal health to professionals;
- promote aquatic animal health in veterinary schools; and
- in the next meeting, change the codes relating to crustaceans, molluscs, amphibians and ornamental fish.

In 2005, during an FAO/WHO Regional Conference on Food Safety for the Americas, 20 countries of LAC reported on their national food safety systems,

and eight recommendations included regional networks and harmonization with international regulations. Some countries based on *Codex Alimentarius* have codes of practice (COPs) and good management practices (GMPs) for food safety of aquaculture and fisheries products. They include measures to reduce risks of contamination with chemicals such as antibiotics, hormones, colorants, pesticides, heavy metals and additives, and to reduce the risk of contamination with pathogens of high risk to consumers. Chile, Colombia, Brazil, Mexico, Honduras and Cuba have also developed food safety training programmes.

About 70 percent of global biodiversity occurs in 12 countries, six of them being within the LAC (i.e. Brazil, Colombia, Costa Rica, Ecuador, Mexico and Peru). However, numerous aquatic organisms have been intentionally introduced into the LAC region for aquaculture and the ornamental fish trade. Around 30 invasive exotic species have been identified (Schüttler and Karez, 2008). Salmonids in Chile have had a negative impact, and have recently reached Patagonia, Argentina, and ornamentals in several countries have eliminated native species. The LAC countries need to identify native species for aquaculture, rather than importing exotic species.

Biotechnologies being used in the region include genetic improvement and control of reproduction, development of monosex populations, pathogen screening and disease diagnosis, vaccines, bioremediation, genetic selection to improve growth rate, and the use of probiotics, but adoption of new technologies is hampered by cost. Most countries have adapted regulations in agriculture and forestry to control the use of GMOs and LMOs in aquaculture, but application of these technologies is also expensive.

The Animal Health Strategy of the European Union (EU) for 2007–2013 is prevention is better than cure. The strategy involves prioritization of EU intervention (e.g. precautionary principle); modern animal health frameworks (e.g. OIE, *Codex Alimentarius*); animal-related threat prevention, surveillance and crisis preparedness; science, innovation and research (e.g. community and national reference laboratories).

The Secretariat of the Pacific Community (SPC) has given high priority to biosecurity issues. In 2007, the SPC organized a “Regional Workshop on Implementing the Ecosystem Approach to Coastal Fisheries and Aquaculture and Aquatic Biosecurity”. Two regional workshops on disease reporting (terrestrial and aquatic animals) were conducted in 2009 and 2010. These workshops have been supported and held in cooperation with regional and international partners such as FAO, EU, the Global Environment Fund (GEF), NACA, OIE and other regional partners.

Examples of actions at the national level include that of several countries in Latin America. Chile has active surveillance and contingency plans for high-risk

diseases of fish; Mexico has surveillance for shrimp, tilapia, trout, carp, catfish and molluscan diseases in collaboration with stakeholders; and Nicaragua has surveillance for shrimp diseases. Colombia uses IRAs to protect animal and plant health, quarantine implementation, aquatic health certification for imported live animals, and active surveillance for WSSV and food safety in shrimp culture. Ecuador had a system to detect WSSV in 2000 and 2001, and Peru had a surveillance programme for WSSV from 2001 to 2006. Chile, Mexico, Brazil, Ecuador, Colombia, Peru, Honduras, Nicaragua and others have level III diagnostic capacity for salmonids, shrimp and tilapia, sometimes cooperating with universities, research institutions and private companies. To harmonize methodologies with the OIE diagnostic manual, FAO, OIE and other organizations have initiated a regional project in the Americas to create a network of diagnostic laboratories maximizing national and regional resources. At the first meeting of the National Laboratories of the Veterinarian Services in the Americas in 2008, 15 conclusions and recommendations were made regarding the setting up of networks, evaluation of laboratories to meet OIE requirements, and the recognition of regional expertise. Molluscan diseases are not well known, and so an OIE Inter-American Technical Group on Molluscs comprised of seven experts from the Americas was formed to consider management of molluscan diseases (Cáceres-Martínez and Vázquez-Yeomans, 2008). An Inter-American Technical Group on Crustaceans and an Inter-American Technical Group on Fish were also formed but have yet to be activated.

Australia has a longer history of biosecurity than the LAC or other Asian countries. Australian Government frameworks aim to manage the risks of entry, establishment and spread of unwanted aquatic pests and diseases. The federal government controls the national borders to prevent the entry of pests and diseases, while the states/territories control postborder pest and disease risks. Coordination and integration of federal and state/territory government action is through two councils comprising federal and state/territory government ministers, and the New Zealand Government. The Australian Government established a taskforce comprising federal and state/territory and government agencies, stakeholders, research and environmental groups which recommended IRAs on live and dead aquatic animal commodities, to prevent introduction of exotic diseases, and the establishment of national emergency response plans to deal with exotic disease incursions. Consequently, the Australian Government established a joint government-industry Fish Health Management Committee charged with development of AQUAPLAN, a five-year (1998–2003) national strategic aquatic animal health management plan. AQUAPLAN 2005–2010 aimed to build on the 1998–2003 plan and focuses on specific issues to further improve Australia's aquatic animal health management. Federal aquatic disease risk management is primarily the role of the Australian Government Department of Agriculture, Fisheries and Forestry (DAFF), which includes the Australian Quarantine and Inspection Service (AQIS). Federal and state/territory governments and other stakeholders are implementing Australia's National

System for the Prevention and Management of Marine Pest Incursions.² It has four components: (i) a national monitoring programme for early detection of new pests, (ii) building industry and community awareness and education, (iii) research and development for development of policy and management, and (iv) evaluation and review of the effectiveness of the system. Mandatory ballast water management for international shipping, introduced in 2001, accords with International Maritime Organisation (IMO) guidelines, allowing discharge of ballast in Australian waters that has been exchanged at sea by an approved method. Vessels' records of ballast exchange are audited by AQIS. In May 2005, Australia signed, subject to ratification, the International Convention on the Control and Management of Ships' Ballast Water and Sediments. There are voluntary national biofouling guidelines, developed with marine industry stakeholders for non-trading, commercial, recreational and commercial fishing vessels and the petroleum industry (Commonwealth of Australia, 2009). Importation of live aquatic species is controlled by the Environment Protection and Biodiversity Conservation Act 1999 through a List of Species Permitted Live Import. The act is administered by the federal government and border controls (AQIS). There are several hundred species of ornamental freshwater and marine fish that can be imported. Additions to the list require stakeholder consultation on IRAs, including the likelihood of establishment of self-maintaining populations and environmental impact. In 2003, Australia's fisheries managers and stakeholders initiated A Strategic Approach to the Management of Ornamental Fish in Australia. The key recommendations include a national noxious fish species list, new management frameworks for ornamentals, better communication with stakeholders and a public awareness campaign on biosecurity risks. The strengths of Australia's current biosecurity systems and the planned improvements are expected to better position Australia to meet these challenges.

Aquatic animal health networks and information

Networking on aquatic animal health through professional societies and other relevant bodies continues to be strong, a clear demonstration of the relevance of the subject and the benefits that members receive from such networks or societies. Examples of very successful and long-standing professional societies include:

- the Japanese Society for Fish Pathology (JSFP);
- the Fish Health Section of the American Fisheries Society (FHS/AFS, 40 years);
- the Fish Health Section of the Asian Fisheries Society (FHS/AFS, 24 years);
- and
- the European Association of Fish Pathology (EAFP, at least 20 years).

Aside from the OIE Aquatic Animal Health Standards Commission (OIE AAHSC), which recently celebrated its golden anniversary in 2010, there are also newly

² www.marinepests.gov.au/national_system

emerging groups, e.g. the NACA Regional Advisory Group on Aquatic Animal Health (AG, nine years) and the International Society for Aquatic Animal Epidemiology (ISAAE, at least five years).

Major veterinary conferences include aquatic animal health as one of the keynote topics, as well as changes in veterinary curricula making aquatic animal health more explicit in educational programmes.

In terms of aquatic animal health information, the sector is continuously serviced by regional and international refereed journals such as *Diseases of Aquatic Organisms*, *Journal of Aquatic Animal Health*, *Journal of Fish Diseases*, *Fish Pathology* (Japan), *EAFP Bulletin*, and the *Diseases in Asian Aquaculture* (DAA) series, as well as disease articles in other general aquaculture publications and other subject-specific journals. There are also aquatic animal health Internet-based information systems where important disease information and databases can be accessed.

OIE provides official reports of occurrence of OIE-listed diseases based on country notifications. In the Asia-Pacific, a Quarterly Aquatic Animal Disease (QAAD) reporting system, a joint FAO/NACA/OIE-Tokyo activity, has had 21 participating regional countries since 1998. The QAAD list is revised annually by the NACA AG in cooperation with OIE and FAO. The regional QAAD lists serious emerging diseases in the region (e.g. KHV, abalone viral ganglioneuritis, WTD, IMNV), some of which are OIE-listed. Information generated from these reporting systems provides an early warning of emerging diseases and information to support IRAs and manage transboundary pathogens.

Farm-level biosecurity, better management practices and good aquaculture practices

In shrimp health, with respect to wider application and improvement in biosecurity, much has been achieved by efforts that have expanded the adoption of good aquaculture practices (GAP), particularly via government extension workers and shrimp farmer associations, but there is still a need for more training and extension work.

In LAC, farm-level biosecurity strategies include codes of practice (COPs), better management practices (BMPs), technical guidelines, standards and protocols designed to promote sustainable aquaculture. These documents contain practical strategies for site selection, water quality and source of broodstock, seed, larvae and juveniles, and include food safety, quality of animal feeds, antibiotics and chemical risks during growth and harvest, as well as good husbandry practices for fish, crustaceans and molluscs. However, there are regional disparities in the implementation of COPs and BMPs. Chile has some 20 documents on GMPs; Mexico has 19 covering aquatic health, food safety, environmental protection, cleaning and disinfection; Costa Rica has seven

documents; Colombia, Brazil and Honduras have at least three documents; Uruguay has two; Peru has one manual and five bulletins related to biosecurity. Countries with fledgling aquaculture, such as many Caribbean countries, lack biosecurity guidelines or manuals. COPs and BMPs have been initiated by national or local governments, industry groups and academic institutions. COPs, GMPs and training have been implemented by salmon farmers in Chile and by shrimp farmers in Mexico, Ecuador, Colombia, Peru, Honduras and Nicaragua, but inconsistencies in application by all farmers reduces the effectiveness of these measures.

Conclusions and recommendations arising from the expert panel presentations during the Global Conference on Aquaculture 2010

Expert Panel III.3 – Improving biosecurity: a necessity for aquaculture sustainability was one of three expert themes under *Thematic Session III on Aquaculture and Environment – Maintaining environmental integrity through responsible aquaculture*. The two others were: *Promoting responsible use and conservation of aquatic biodiversity for sustainable aquaculture development* and *Addressing aquaculture-fisheries interactions through the implementation of the ecosystem approach to aquaculture*.

The expert panel presentation made the following conclusions:

- Aquaculture development (intensification, diversification and trade) brings new challenges to sustainable development of the sector; biosecurity issues are major concerns.
- Disease intelligence, research, technologies and information have greatly improved; however, there is a need to involve especially farmers/producers into the equation for effective implementation.
- There is a need to keep pace with the aquaculture landscape in terms of species, systems, technologies and environments in order to determine appropriate biosecurity measures that can be put in place at every step of the culture cycle/value chain at all levels. It must be recognized that application of biosecurity to novel species requires considerable lead-in time for information gathering (e.g. research on diseases and potential environmental impact). Biosecurity cannot be implemented in an information vacuum.
- Efforts should be focused on prevention and maintaining healthy and safe aquatic production.
- Risk analysis is an important decision-making tool and this should be supported with infrastructure, human capacity and information.

The way forward includes the following:

- National frameworks are needed to regulate, manage and control biosecurity.

- Surveillance programmes and diagnostic services are required to detect and identify the arrival and spread of pests and diseases.
- Timely assessment of the threats from new or expanding species is essential.
- Rapid response to eradicate new pests and diseases is needed before they establish and spread.
- Standardization of science-based identification of all risk pathways and high-risk organisms, and implementation of preborder, border, and postborder measures to prevent pests and diseases from entering the country are required.
- Infrastructure, human capacity, research and information to implement the above must be improved.
- Capacity building is needed at all levels.
- Regional cooperation should be enhanced to permit disease control, based on regional as well as global disease information.
- Initiatives should be undertaken to establish new aquaculture operations, such as underwater aquaculture systems to maximize utilization of the water column and seabed, or the use of the bases of marine wind turbines to anchor sea farms.

The following were also presented as the message that will be relayed to the Fifth Session of the FAO Committee on Fisheries Sub-Committee on Aquaculture:

- International and national efforts to promote biosecurity need to better reach the grassroots levels of the industry and the community stakeholders (e.g. farmers, extension services, importers, processors, boat owners, fishermen, etc.).
- Biosecurity frameworks need to keep pace with the unprecedented level of aquaculture development in terms of species, systems and technology.
- Standards on aquatic animal health for known pathogens, aquatic pests and food safety are already available, but greater commitment by governments is needed to implement these standards.
- International standards need to be developed to address the high incidence of emerging diseases of aquatic animals and aquatic pests compared to the terrestrial scenario – there is a need to complement the pathogen/pest specific approach to biosecurity with standards that deter high-risk practices.

The way forward

Biosecurity is being challenged, and will be more challenged in the foreseeable future. The growth of the world human population and the increase in human travel, along with international trade in animal and plant products will require increased vigilance at borders to stop the spread of unwanted organisms, whether as pests causing environmental damage or as agents of epizootic disease. There is a need for border agencies to recognize that potential aquatic

pathogens and pests are more likely to be introduced through ports and the ornamental fish trade than by the traditional terrestrial routes.

The review of Subasinghe, Bondad-Reantaso and McGladdery (2001) contains many elements that are still relevant. The current review provides additional insights as to how biosecurity may be addressed in a cross-sectoral and multidisciplinary manner. Effective, coordinated and proactive biosecurity systems are the product of science-based knowledge and practices used within effective regulatory frameworks backed by sufficient resources for enforcement (FAO, 2010b). As aquaculture becomes more intensive, new diseases and other problems are likely to emerge. Aquaculture biosecurity will continue to operate at three levels; a) internationally, as recognized in the Bangkok Declaration; b) regionally, as seen through various regional activities; and c) on a small scale where variables (e.g. environment, species cultured, funding, training, economics) differ within countries in a region. A crucial consideration is how to deal with “unknowns”. There is a need to forge an effective regional and international cooperation to pool resources, share expertise and information. At the global, regional or national levels, the institution mandated to ensure biosecurity would be served well by putting emergency preparedness with advanced financial planning as their core function.

Taura syndrome virus (TSV) and infectious myonecrosis virus (IMNV) are only two examples of exotic diseases that have been introduced to the Asian region through the importation of SPF *Litopenaeus stylirostris* and *L. vannamei*, respectively. Biosecurity is an important issue in the use of SPF stocks which needs to be clearly understood by importers and farmers. Once a broodstock or postlarvae produced by an SPF facility leave that facility, they are no longer considered to have SPF status for the specific pathogens indicated, since the level of biosecurity under which they are being maintained has now decreased. Because their health status is now less certain, a new historical record for that facility must be established to support any claims of health status. Every country should be wary of importing exotic crustaceans of any kind for aquaculture without going through the recommended quarantine procedures, combined with tests for unknown viruses that might be a danger to local species (Flegel, 2006c). Risk analysis is necessary to assess emerging threats from new or exotic species. These biosecurity measures should be applied even to exotic domesticated stocks that are SPF for a list of known pathogens. To reduce risks to the minimum, any country that imports exotic stocks for aquaculture should invest in establishment of local breeding centers comprised of properly vetted stocks that could be used for ongoing supply of broodstock and of postlarvae to stock cultivation ponds. This would avoid the continual risk of importing unknown pathogens that might be associated with continuous importation and direct use of exotic stocks, even from a foreign breeding center that produces SPF stocks. In shrimp health management, which are also equally important to any other aquatic animal production

system, there is still need for improvement in many areas, including the need for:

- development of domesticated and genetically improved SPF stocks for all cultivated species;
- more widespread use and standardization of diagnostic tests;
- wider application and improvement in biosecurity;
- better control over transboundary movement of living crustaceans for culture;
- investigation of the efficacy of probiotics, immunostimulants and so-called “vaccines” in full-scale field trials;
- full understanding of the host-pathogen interaction in shrimp;
- more work on shrimp epidemiology;
- more studies on molecular ecology (i.e., metagenomics) and biochemical engineering to control the microbial dynamics in shrimp ponds and hatchery tanks.

In the Latin America and Caribbean region (LAC), no national aquatic health programme to protect aquatic organisms from disease has been developed in one document. There is a need to: (a) list the pathogens present; (b) identify OIE-listed pathogens likely to be in the region; and (c) implement disease diagnosis, health certification and quarantine, disease zoning, disease surveillance and reporting, contingency plans, IRA, capacity building, national strategies and policy frameworks, education and training, and enhancement of aquatic animal emergency disease preparedness and response (FAO/NACA, 2001a,b; Commonwealth of Australia, 2005).

On disease diagnostics, validation of new diagnostic methods is essential. Nanotechnology, currently being explored for detection of food pathogens and in clinical and veterinary diagnostics, is an area which may also have useful application in aquatic animal disease diagnosis. Gene sequencing and development of pathogen microarrays and other novel methods for use in pathogen detection in aquaculture should be continuously pursued with the objectives of improving the accuracy, sensitivity, specificity and speed of tests, and their applicability for diagnosis, screening and monitoring of health status of aquatic animals in the field.

In an ideal world, farmers would have a full “tool kit” of medicines and diagnostic services to monitor, control and prevent the diseases that threaten their stock. The tool kit would comprise of vaccines for preventing the major endemic diseases, immunostimulants and other feed additives to enhance the performance of the aquatic animals under farming conditions, and a range of treatment products to cure any new or sporadic future infections. All of these products would be fully approved, documenting their quality, efficacy and safety. The farms and industry would have the support of accurate diagnostic services and the support from veterinarians or aquatic animal health professionals – allowing them to develop and implement effective veterinary health plans and

utilize the medicines in compliance with good treatment practices and industry COPs. This is already possible in some parts of the world, and the impact has resulted in great improvements in sustainability and increased productivity, as well as improved farming efficiency. However, there are still challenges in achieving this in Asia, where there are many fish and shrimp species cultured, many diverse pathogens, a diverse environment and variable access to knowledge and information. From a manufacturer's point of view, solutions to the challenges for the sustainable use of medicines in aquaculture could include international harmonization of regulatory data requirements for approving products. Some of the particular challenges relate to the claims needed to support the use of the products in the variety of species being farmed. The provision of these practical solutions needs to be backed up with effective certification and enforcement of the regulations. In conclusion, there is an opportunity to ensure the responsible and sustainable use of medicines in aquaculture world-wide. The knowledge is available and the required products are available or can be developed. With a clear harmonized regulatory environment which will ensure globally accepted standards, the needs and expectations of the producers and the consumers for safe, efficacious medicines can be met and sustainable aquaculture can be achieved. This could include:

- the idea of crop grouping, i.e. use of representative species (e.g. Atlantic salmon) of a similar group or production environment to allow use of a medication in the whole group (e.g. salmonid fish);
- extrapolation of maximum residue levels (MRLs) from major species to minor species;
- the development of a network of facilities and experts able to disseminate and validate information to support health management in a region; and
- the development and implementation of veterinary health plans so that farmers can treat and sell their produce with confidence.

Applications of transgenic fishes, the science of risk assessment, the practice of risk management, and public policies for oversight of biotechnology are all in development. Future developments will include broader appreciation within both the aquaculture and regulatory communities of both the benefits and the risks posed by production of aquatic GMOs. Recognizing that all hazards cannot be predicted nor associated risks reliably and cost-effectively quantified, there will be a broader appreciation that biosecurity is the key issue for realizing benefits while managing risks posed by production of aquatic GMOs. Hence, granting of permits for production of aquatic GMOs will be conditional upon reaching agreement on how to manage risk by means of implementing effective confinement. The granting of the first such permits is yet before us, and will be a landmark event, especially as regards the technical conditions under which production of the stock in question is permitted to go forward. The degree to which production of transgenic fish ultimately will prove sustainable will depend upon many societal decisions as to whether, and under what conditions, to utilize transgenic technology in aquaculture.

On marine invasives, filling these knowledge gaps will allow proactive marine biosecurity measures that will be consistent with the international framework established in the terrestrial environment:

- good baseline information in coastal zones (specifically ports and marinas);
- knowledge of current and future trading patterns associated with transport vectors, due to new free trade associations; and,
- knowledge of the physical, ecological, environmental, economic and social (including human health) impacts.

The application of risk analysis is at the heart of the modern approaches to biosecurity. It offers an effective management tool where by pragmatic decisions can be made that provide a balance between competing environmental and socio-economic interests, despite limited information. This tool, however, needs research, databases and other vital sources of information and knowledge so that it can effectively support biosecurity assessments, surveillance, diagnostics, early warning, and contingency planning (Arthur *et al.*, 2009).

The efforts of FAO, OIE, WHO, the EU and regional partners such as NACA (in Asia), OIRSA (in Latin America) and SPC (in the Pacific), as well as governments' individual efforts in bringing together relevant competent authorities on biosecurity governance should be continued. Effective national biosecurity governance, regional and global partnerships and champions are needed so that the risks posed by transboundary diseases of aquatic animals and other biosecurity threats can be minimized and associated losses and other negative impacts reduced. The recommendations generated from the review and the discussions and conclusions of the Global Aquaculture Conference 2010 are not directed to one single institution or stakeholder. Addressing biosecurity which transcends national boundaries should be a shared responsibility.

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