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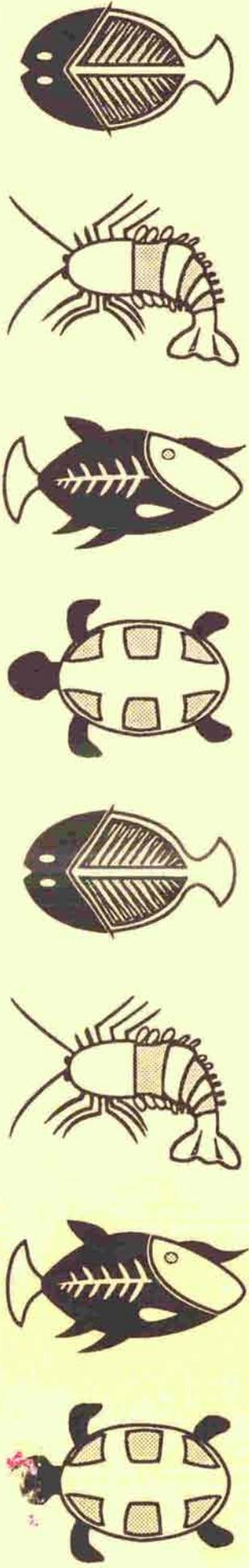
# PERSPECTIVES IN AQUATIC EXOTIC SPECIES MANAGEMENT IN THE PACIFIC ISLANDS

VOLUME II

INTRODUCTIONS OF AQUATIC ANIMALS TO THE PACIFIC ISLANDS:  
DISEASE THREATS AND GUIDELINES FOR QUARANTINE

by

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Australia



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South Pacific Commission  
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## PREFACE

The quarantine of fish and marine invertebrates, or rather the general lack of it, has been a nagging worry to fisheries administrators in the Pacific Islands for some time. In contrast to the terrestrial field, there have been no 'horror stories' about introducing devastating diseases or parasites to the aquatic environment of the developing Pacific Islands, but this has always been balanced by the fear that we spend so little time studying the ocean that we would not even notice if something were wrong. This report, by Dr John Humphries, was commissioned by the South Pacific Commission Resource Assessment Section with funding from the Australian Centre for International Agricultural Research. It puts the situation into perspective and provides the basic principles and guidelines for aquatic animal quarantine services in the Pacific Islands.

This work is part of the fulfilment of a request from the island countries of the region, at the South Pacific Commission's 23rd Regional Technical Meeting on Fisheries in 1991, to provide information that could be used by Pacific Island governments in the setting-up of aquatic quarantine measures. The draft of this document was discussed at the SPC's 25th Regional Technical Meeting on Fisheries, in March 1994, and the meeting made the following recommendation to the SPC Committee of Representatives of Governments and Administrations:

*Noting the disease risk posed by increasing numbers of transfers of exotic species into the Pacific Islands, the meeting endorsed the interim Guidelines and Principles for Regional Aquatic Animal Quarantine detailed in Annex 2 of Working Paper 11 towards the development and harmonisation of aquatic animal quarantine mechanisms in the region, and recommended that the Secretariat monitor progress in the development of Pacific Island quarantine mechanisms to enable the Interim Guidelines to be reviewed at the next Regional Technical Meeting on Fisheries.*

The Committee of Representatives of Governments and Administrations supported this recommendation to the 34th South Pacific Conference.

This report is the second volume in a planned three-part series on various aspects of introductions of aquatic organisms to the Pacific Islands, coordinated jointly by the South Pacific Regional Environmental Programme and the South Pacific Commission. The first volume, by Dr Lucius G. Eldredge, presented a history of transfers of aquatic organisms in the Pacific Islands region, while Volume III will deal with the assessment of ecological risks associated with exotic aquatic species introduction.

Julian Dashwood  
Manager, Fisheries Programme  
South Pacific Commission

## SUMMARY

Diseases of social, economic and ecological importance affecting fish, crustaceans and molluscs have spread widely as a result of failures to impose appropriate quarantine and health certification procedures. Within the Pacific Islands region, the need for quarantine of plants and domestic animals introduced into the region is well recognised but until recently, little emphasis was placed on quarantine of aquatic animals. Many aquatic animals have been introduced or transferred within the region and on occasions, diseases and pests have accompanied such movements. A spectrum of diseases and parasites has been described in the region. Many aquatic species present are likely hosts to serious diseases which occur elsewhere. Freedom from disease confers major social, economic and ecological advantages and a responsibility exists to protect the fisheries resources of the region from spread or introduction of disease. The implementation of a practical system of quarantine offers an opportunity to protect fisheries from disease within the South Pacific region.

This study is presented as a basis for the development of a regional strategy for the quarantine of introduced aquatic animals. The basis of the strategy is the evaluation of candidate species on a case-by-case basis, with implementation of quarantine commensurate with the relative risk of disease introduction for that species.

Quarantine is necessary for all transferred species. As different species present different risks with regard to introduction of exotic diseases, increased levels of quarantine and disease testing may be necessary with high-risk species. Minimum requirements for the quarantine of aquatic animals are described. Factors determining the relative risk of disease posed by different species entry are discussed and additional measures which may be taken to protect against higher risk species are presented.

Minimum quarantine requirements described include:

- Evaluation of each transfer on a case-by-case basis,
- Risk categorisation of introduced species,
- Pre- and post-transfer quarantine and disease examination,
- Health certification by a competent authority,
- A minimum total time in quarantine for lower-risk species,
- A correspondingly longer time in quarantine with more stringent disease testing for higher-risk species,
- Secure quarantine facilities for holding aquatic animals during transfer,
- Development of legislation on which to base quarantine,
- Compilation and maintenance of a regional disease database,
- Training in diagnostic procedures for recognition of diseases in aquatic animals.

# RÉSUMÉ

Des maladies des poissons, crustacés et mollusques qui ont des répercussions sur les plans social, économique et écologique, se sont propagées un peu partout, faute de mesures de contrôle et de certification zoosanitaires suffisantes. En Océanie, la nécessité du contrôle phytosanitaire et zoosanitaire d'espèces introduites dans la région est bien admise, mais jusqu'à récemment, le contrôle zoosanitaire d'espèces aquatiques n'a guère retenu l'attention. De nombreux animaux aquatiques ont été introduits dans la région ou transférés d'un endroit à l'autre de celle-ci, et à quelques occasions, ces déplacements ont entraîné avec eux des maladies et des parasites dont l'étude dresse un inventaire pour la région. De nombreuses espèces aquatiques qu'on y trouve sont vraisemblablement porteuses de maladies graves qui ont été apportées d'ailleurs. L'absence de maladie comporte d'importants avantages sur les plans social, économique et écologique; la protection des ressources halieutiques de la région contre la propagation ou l'introduction de maladies est donc une grande responsabilité. A l'occasion de la mise en place d'un système de contrôle sanitaire effectif, il convient de sauvegarder les ressources halieutiques du Pacifique Sud de la maladie.

L'objet de la présente étude est de servir de point de départ à l'élaboration d'une stratégie régionale en matière de contrôle zoosanitaire d'espèces aquatiques introduites. Cette stratégie se fonde sur l'évaluation, au cas par cas, des espèces dont l'introduction est souhaitée, assortie de mesures de contrôle zoosanitaire dont la sévérité est proportionnelle aux risques d'introduction de maladies que présente l'espèce.

Le contrôle phytosanitaire s'impose pour toutes les espèces transférées, car les risques d'introduction de maladies exotiques sont différents d'une espèce à l'autre, et une augmentation de la durée de l'isolement et une intensification du dépistage des maladies s'imposent donc pour les espèces présentant des risques élevés. En tout état de cause, le contrôle zoosanitaire des espèces aquatiques doit répondre à des exigences minimales. L'étude examine les facteurs permettant d'établir le risque relatif que présentent les diverses espèces pour ce qui est de l'introduction de maladies et décrit des mesures supplémentaires qui peuvent être prises pour se protéger contre les espèces présentant les risques les plus élevés.

Les exigences minimales en matière de contrôle zoosanitaire sont les suivantes :

- évaluation individuelle de chaque transfert,
- catégorisation des risques que présentent les espèces introduites,
- isolement avant et après le transfert et dépistage de maladies,
- certification zoosanitaire par un organisme compétent,
- durée minimum de l'isolement total pour les espèces présentant des risques moins élevés,
- isolement plus long, proportionnel au risque, et dépistage plus rigoureux pour les espèces présentant des risques élevés,
- installation pour l'isolement des animaux aquatiques en cours de transfert,
- élaboration d'une législation sur laquelle fonder le contrôle zoosanitaire,
- compilation et tenue d'une base de données régionale sur les maladies,
- formation aux méthodes diagnostiques afin de reconnaître les maladies des animaux aquatiques.

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# INTRODUCTION

On many occasions over the past century, spread of serious disease has occurred as a direct result of transferring live aquatic animals to new locations without imposing basic quarantine precautions. Often, the introduced diseases have had catastrophic effects on aquaculture, commercial fisheries and the aquatic ecosystem. The international spread of diseases and pests of aquatic animals is extensively documented and is the subject of long-standing concern (Albaladejo and Arthur 1989, Andrews 1980, Bauer 1991, Farley 1988, Hoffman 1970, Humphrey 1988, Langdon 1990, Lightner *et al* 1989a, Lumanlan *et al* 1992, Rosenthal 1980, Reichenbach-Klinke 1984, Stewart 1991, Turner 1988). Numerous recommendations and guidelines for the implementation of quarantine of aquatic animals have been made and precedents established (Arthur 1987, Davy and Chouinard 1983, Davy and Graham 1979, DeKinkelin and Hedrick 1991, Grizel 1989, Langdon 1990, Roberts 1981, Rohovec 1983, Turner 1988). Many countries, however, continue with limited, ineffectual or no quarantine when introducing aquatic animals.

Within the Pacific Islands region, including Hawaii, more than 140 species of fish and aquatic invertebrates have been intentionally introduced or transferred over the past 200 years. These movements have been for commercial, social or ecological purposes (Eldredge 1992,1994) and, with few recent exceptions, have occurred with little regard to the introduction or spread of diseases or pests. For countries in which fisheries resources and industries are of social and economic importance, protection from diseases and pests should be a high priority of government. The importance of quarantine in preventing the spread of diseases and pests of animal and plant species has been recognised in the Pacific Islands region for many years (Anon 1951, 1977, 1980, 1982, 1984a, 1987, Morschel 1988a,1988b). Quarantine for introduced fish and aquatic invertebrates has, however, been neglected, despite the social, economic and ecological importance of these species and despite their frequent introduction from other regions.

This study is presented as a basis for the development of a regional strategy for quarantine of aquatic animal species. The basis of the strategy is the evaluation of candidate species on a case-by-case basis, with implementation of quarantine commensurate with the relative risk of disease introduction for that species. The study suggests minimum requirements for the quarantine of aquatic animals within the Pacific Islands region, examines risks of introducing disease and describes more stringent quarantine measures which may be imposed as necessary. The serious consequences of importing aquatic animals without quarantine are reviewed. The disease status of fish and aquatic invertebrates in the Pacific Islands region, including Hawaii, is summarised, providing background information for the implementation of quarantine for transfers within the region, as well as a basis for the definition of exotic diseases.

## DISEASE THREATS POSED BY INTRODUCED AQUATIC ANIMALS

A vast number of serious diseases and pests of fish and aquatic invertebrates is known. The majority of these are not described in the Pacific Islands region and are thus exotic. In addition, many genera are present which are hosts to a wide range of exotic diseases. The greatest risk of introducing exotic disease is posed by the movement of live aquatic animals (Langdon 1990). Thus, introductions of aquatic animals to the region are always accompanied by the risk of the introduction and establishment of serious exotic diseases.

A far greater risk of disease introduction exists when animals are taken from the wild, where their health status is poorly defined and they will likely be carrying diseases, parasites or pests. Animals from hatcheries or farms, where the disease status is generally known, represent less risk, especially where quarantine and health certification procedures are implemented, although serious diseases may still be present. Whenever possible, it is desirable to introduce animals from farmed or hatchery sources which are subject to quarantine and health certification.

Numerous examples exist whereby diseases and pests have been introduced and established as a result of importing or transferring live fish, fertile fish eggs, live aquatic crustaceans and live aquatic molluscs between countries and within regions. Many diseases affect marine and estuarine species. Diseases may also affect both freshwater and marine species. Thus importation of freshwater species may introduce diseases of marine species, and *vice versa*. Risks of introducing diseases apply equally to freshwater and marine species.

Many marine and freshwater fish and aquatic invertebrate species have been introduced into the Pacific Islands region and Hawaii for stock enhancement programs, food purposes, aquaculture, biological control and as ornamental species (Eldredge 1993, Gerberich and Laird 1968). Live bait fish are widely used (Smith 1974), presenting a special case with regard to potential introductions of disease. As early as the 1770s, catfish were introduced to Guam from the Philippines, and transfers of fish, crustaceans and molluscs have continued to the present (Eldredge 1993). A partial listing of these species is given in Appendix I. The introduced species are often of commercial significance and have originated from regions known to have major diseases. Many of the introduced species are known hosts of serious diseases exotic to the region.

# INTERNATIONAL SPREAD OF IMPORTANT DISEASES

Numerous important diseases of marine and estuarine aquatic animals have spread as a result of the international movements of fish, crustaceans and molluscs. A partial list of these diseases is shown in Table 1. The importance and means of introduction of these diseases are briefly described as examples of the serious consequences which may occur from failure to impose effective quarantine.

## Diseases caused by viruses

**Viral haemorrhagic septicaemia (VHS)**, is a viral disease of rainbow trout *Oncorhynchus mykiss* and other species which causes severe mortalities and high economic losses in marine and freshwater fish (Wolf 1988). VHS was introduced into trout farms in Czechoslovakia and Bulgaria with imported fertile eggs and was common in live trout imported into Switzerland (Bauer 1991, Meier 1984). **Lymphocystis**, an infection of marine and freshwater fish which may be fatal in young stock (Wolf 1988), is common in imported ornamental fish (Ashburner 1975, Boustead 1982, Durham and Anderson 1981, Lawler and Ogle 1977). **Infectious pancreatic necrosis (IPN)** is a highly infectious, often fatal disease of farmed and wild freshwater and marine fish. The virus also occurs in marine molluscs and crustaceans. IPN has spread through imports of infected fish or fish eggs to many countries including Britain, Denmark, France, Germany, Spain, Italy, Japan, Switzerland, China, South Africa and Chile (Bauer 1991, Bragg and Combrink 1987, Ledo *et al* 1992, McAllister and Reyes 1984, Meier 1984, Tong and Hedrick 1990, Wolf 1988, Yulin and Zhengqui 1987).

Introduced live penaeid prawns are well known for spreading viral diseases into many countries, including the Pacific Islands and Hawaii (See below). These diseases primarily affect juvenile farmed prawns, but are carried by wild, adult prawns. **Monodon baculovirus (MBV)**, a disease causing high mortalities (Lightner *et al* 1983, Sparks 1985) was introduced into Israel, USA, Brazil, Ecuador and the Pacific as a result of importing live prawns (Colorni *et al* 1987, Lightner *et al* 1989a,b). **Infectious hypodermal and haematopoietic necrosis virus (IHHNV)** causes deaths in sub-adult prawns (Lightner *et al* 1985, Sparks 1985) and was similarly introduced into Israel, USA, Philippines, Venezuela, Ecuador, Peru, Mexico and the Pacific (Colorni *et al* 1987, Lightner *et al* 1989b). **Hepatopancreatic parvo-like virus (HPV)**, a virus associated with mortalities in cultured prawns (Lightner *et al* 1985), was detected in quarantine in prawns imported into Israel (Colorni *et al* 1987) and has been introduced into Brazil, Ecuador and the Pacific (Lightner *et al* 1989b).

A severe viral disease of molluscs, **Portuguese oyster iridovirus-like disease**, virtually eliminated the Portuguese oyster from the French coast. The disease occurred in Britain in 1969 in oysters imported from Portugal (Alderman and Gras 1969, Hill 1984, Sparks 1985).

## Diseases caused by bacteria

**Furunculosis** caused by *Aeromonas salmonicida* is characterised by skin ulceration and generalised infections of many marine and freshwater fish. It is a major problem in aquaculture (Austin and Austin 1987) and may result in loss of export markets. The agent likely entered Europe in imported salmonid fish (Stewart 1991). It recently spread to Norway in imported Atlantic salmon (Stewart 1991). It entered Australia in imported goldfish *Carassius auratus* and has since spread widely (Humphrey and Ashburner 1994). **Epitheliocystis** is a common infection of freshwater and marine fish which may cause deaths, especially in young fish (Wolf 1988). Epitheliocystis has spread in imported tropical marine ornamental species and in red sea bream *Pagrus major* imported into Japan (Miyazaki *et al* 1986, Wolf 1988). **Fish tuberculosis or mycobacteriosis** caused by *Mycobacterium* sp. causes nodular lesions, weight loss and death and is recorded in more than 150 marine and freshwater fish species; some *Mycobacteria* of fish cause disease in humans (Austin and Austin 1987). The agent was introduced to a major fish hatchery in Australia with imported chinook salmon *Oncorhynchus tshawytscha* eggs (Ashburner 1976) and is common in ornamental fish. Bacterial kid-

ney disease (BKD) caused by *Renibacterium salmoninarum* is a serious disease of farmed and wild marine and freshwater salmonid fish, which may result in high losses and loss of markets. Spread is primarily due to movements of infected fish (Austin and Austin 1987), but salmonid eggs imported into Chile were considered the source in that country (Sanders and Barros 1986). Vibriosis caused by *Vibrio* sp. may cause heavy losses, especially of hatchery-reared, larval and juvenile marine fish, crustaceans and molluscs (Austin and Austin 1987). *Vibrio* sp. were imported into Malaysia in sea bass (Wong and Leong 1987). *Vibrio cholera* (non-01) was associated with high mortalities in goldfish *Carassius auratus* imported into Australia from Singapore (Reddacliff et al 1993), and *Vibrio anguillarum*, a serious pathogen of marine fish species, may have been introduced into Japan with eels imported from France in 1975 (Austin and Austin 1987).

**Diseases caused by fungi**

**Epizootic ulcerative syndrome** (EHS) is an invasive, ulcerating fungal disease of the skin and muscle of a range of freshwater and estuarine fish species which has caused high losses and extremely adverse social and economic impacts throughout much of South East Asia. A similar condition occurs in Papua New Guinea (Roberts et al 1986, Uwate 1983). EUS is thought to have spread in imported live food fish, ornamental fish or prawns (Frerichs 1988, Lumanlan et al 1992, Shariff and Subasinghe 1992).

**Diseases caused by protozoa**

**Brooklynelliasis** is a disease caused by *Brooklynella hostilis*, a gill parasite of marine fish characterised by deaths, especially in farmed or aquarium fish (Wootton 1989, Lom and Dykova 1992). A *Brooklynella* sp. was imported into Singapore on red sea bream (Anon 1987b). **Oodiniasis** is an infection with *Oodinium* sp. which are common, serious parasites of the skin and gills of marine fish, especially in aquaria and in aquaculture (Wootton 1989, Hoffman 1970).

**Table 1: A partial listing of important diseases of marine and estuarine aquatic animals spread by the international movement of fish, crustaceans and molluscs**

|  |  |
|--|--|
| <p><b>Viral Diseases of Fish</b></p> <p>Viral haemorrhagic septicaemia<br/>Lymphocystis<br/>Infectious pancreatic necrosis</p> <p><b>Viral Diseases of Crustaceans</b></p> <p>Monodon baculovirus<br/>Infectious hypodermal and haematopoietic necrosis virus<br/>Hepatopancreatic parvo-like virus</p> <p><b>Viral Diseases of Molluscs</b></p> <p>Portuguese oyster iridovirus-like disease</p> <p><b>Bacterial Diseases of Fish</b></p> <p><i>Aeromonas salomocida</i><br/>Epitheliocystis<br/><i>Myobacterium</i> sp.<br/><i>Renibacterium salmoninarum</i><br/><i>Vibrio</i> sp.</p> <p><b>Fungal Diseases of Fish</b></p> <p>Epizootic ulcerative syndrome</p> | <p><b>Protozoal Diseases of Fish</b></p> <p><i>Brooklynella hostilis</i><br/><i>Oodinium</i> sp.<br/><i>Trichodina</i> sp.</p> <p><b>Protozoan Diseases of Molluscs</b></p> <p><i>Minchinia armoriana</i><br/><i>Haplosporidium nelsoni</i> (<i>Minchinia nelsoni</i>)<br/><i>Marteilia refringens</i><br/><i>Bonamia ostreae</i></p> <p><b>Crustacean Diseases of Molluscs</b></p> <p><i>Mytilicola intestinalis</i><br/><i>Mytilicola orientalis</i></p> <p><b>Pests of Marine Molluscs</b></p> <p><b>Oyster drills</b><br/><i>Urosalpinx cinerea</i><br/><i>Ocenebra erinacea</i><br/><i>Purpura cloviger</i></p> <p><b>Worms</b><br/><i>Pseudostylochus ostreaophagus</i><br/><i>Polydora websteri</i></p> |
|--|--|

*Oodinium* sp. have spread widely on many fish species, including ornamental species (Boustead 1982, Gratzek *et al* 1976, Hoffman 1970). **Trichodiniasis** is a common, serious disease of freshwater and marine fish caused by *Trichodina* sp. which are common serious skin and gill parasites (Wootton 1987, Lom and Dykova 1992). *Trichodina* sp. have spread frequently in ornamental and other live fish and have often caused serious disease in their new location (Alimon *et al* 1993, Ashburner 1976, Boustead 1982, Djajadiredja *et al* 1983, Hoffman 1970, Langdon 1990, Lumanlan *et al* 1992, Wong and Leong 1987).

*Minchinia armoricana* causes a serious disease of European oysters *Ostrea edulis* characterised by progressive wasting (Sparks 1985). This parasite was introduced from European oysters imported to the Netherlands from France (Van Banning 1977, 1979). *Haplosporidium nelsoni* (*Minchinia nelsoni*) caused catastrophic disease outbreaks in American oysters *Crassostrea virginica* in the USA (Sparks 1985). The agent was thought to have been introduced to the USA in oysters introduced from Asia. (Andrews 1980). *Marteilia refringens* is the cause of severe disease outbreaks and deaths of European oysters and also may infect other molluscs (Sparks 1985). *M. refringens* was transferred from France to Spain and Holland in imported oysters *Ostrea edulis* (Sparks 1985, Van Banning 1979). *Bonamia ostreae*, the cause of heavy mortalities in European oyster growing areas, has spread through imports of oyster seed stock to the Netherlands and Spain. Its occurrence in Ireland may be due to illegal importation of oysters from France and in England is likely due to introductions from continental Europe (Sparks 1985, Stewart 1991).

#### Diseases caused by crustaceans

*Mytilicola* spp. are copepod parasites of the intestinal tract of marine molluscs and have caused catastrophic mortalities in infected hosts (Sparks 1985, Bauer 1991). *Mytilicola intestinalis* has spread from the Mediterranean to Northern Europe and caused massive mortalities in mussels. It has also been introduced into Scotland (Sparks 1985). *Mytilicola orientalis* causes loss of condition and high mortalities among a number of marine mollusc species and has caused devastating losses in the oyster industry. *Mytilicola orientalis* was introduced in seed oysters *Crassostrea gigas* transplanted from Japan to the USA and France (Andrews 1980, Bauer 1991, Rosenthal 1980, Sparks 1985).

#### Pests of marine molluscs

The gastropod predatory oyster drill *Urosalpinx cinerea* was imported to the United Kingdom with *Crassostrea virginia* (Andrews 1980). The Japanese oyster drill, *Ocenebra japonica*, is an accidental introduction accompanying imports of oysters from Japan and is well-established in North American oyster beds (Andrews 1980, Rosenthal 1980). *Ocenebra erinacea*, another gastropod pest, was introduced into South Africa with *Ostrea edulis* imported from Europe (Rosenthal 1980). *Purpura cloviger*, an oyster drill, invaded the west coast of North America with imports of Japanese oysters (Rosenthal 1980).

*Pseudostylochus ostreaophagus* is a serious parasite of oysters and mussels, and was introduced to the West Coast of North America with imports of shellfish from Japan (Andrews 1980). *Polydora websteri*, a mudworm of oysters, may have been introduced into Australian waters in the 1890's with imports of oysters from New Zealand (Nell and Smith 1988) and has also been introduced into the Pacific.

# REGIONAL SPREAD OF DISEASES

## Spread of diseases

Braley (1991) reported that diseases of pearl oysters have been transferred between atolls in the Tuamotu group, and that these diseases had affected other bivalve molluscs. Further, Eldredge (1994) noted that the disease had been spread by transfers of shells as well as animals.

Viral diseases of penaeid prawns have been introduced to the Pacific Islands region and Hawaii with imported stock. *Monodon baculovirus* was introduced into French Polynesia and subsequently imported into Hawaii with post-larval *Penaeus monodon* from several countries (Lightner *et al* 1983, 1989b). Similarly, infectious hypodermal and haematopoietic necrosis virus, a cause of serious disease of Penaeid prawns, especially *Penaeus stylirostris*, was introduced to Tahiti, Guam and Hawaii (Lightner *et al* 1989b). Hepatopancreatic parvo-like virus and a reo-like virus have also been introduced into Hawaii (Lightner *et al* 1989b).

## Spread of pests and predators

The introduction of aquatic animals themselves has had adverse effects on existing animal and plant communities. The freshwater snails *Pomacea canaliculata*, *Pomacea bridgesi* and *Pila conica* were intentionally introduced to Hawaii for aquaculture, with subsequent complaints of damage to taro crops. *Pomacea* spp. introduced elsewhere for aquaculture have become serious pests of rice and other crops (Cowie 1992). A freshwater crayfish *Procambarus clarkii*, introduced into Hawaii, has also become a threat to the taro (*Colocasia esculenta*) crop. *Polydora nuchalis*, a polychaete worm pest which causes accumulations of tube masses and sediment in penaeid shrimp and oyster ponds, was likely introduced into Hawaii with shrimp imported from Mexico. Similarly, *Polydora websteri*, the cause of 'mud blisters', was likely introduced into an intensive oyster facility in Hawaii with the introduction of *Crassostrea virginica* from the USA, as well as in oysters introduced from elsewhere on the islands (Bailey-Brock 1990).

Pyramidellid gastropod parasites, a major problem in tridacnid clam culture, were described in batches of tridacnid clams imported into Guam, Fiji and Hawaii by Govan (1992a,b). This author correctly noted that in some cases it was difficult to tell whether the predators found in ocean-nurseries were introduced or were part of the local population.

## Regional spread of diseases

Reports of disease incursions into the Pacific Islands and Hawaii are infrequent, despite the many introductions of aquatic animals. This absence of disease may possibly reflect inadequate surveillance rather than the true status. Examples are known, however, even where quarantine regulations were present (Bailey-Brock 1990). Further, unexplained mortalities of imported species are also reported (Eldredge 1994), suggesting that imports have not been free from disease, or imports have succumbed to diseases or parasites already in the region.

# INDIGENOUS DISEASE INVENTORY

Protection of national and regional fisheries from disease is the primary objective of quarantine and requires a detailed knowledge of the diseases and pests which affect aquatic animal species within the region for two reasons:

1. To prevent spread of existing diseases between islands within the region.
2. To define diseases exotic to the region and implement appropriate quarantine practices to prevent their introduction.

Diseases have been reported in the region which are of particular significance and have caused serious losses. Many other agents have been identified but have not yet been associated with disease.

A description of diseases which have occurred within the Pacific Islands region, including Hawaii, and a partial listing of pathogens, parasites, diseases and pests which have been recorded are given in Appendix II.

The compilation and maintenance of a database of this nature, listing diseases, species affected and geographical location is essential for disease control purposes and is strongly recommended. A regional authority such as the South Pacific Commission with its library and data information resources might compile such a database for the region.

# INTERNATIONAL QUARANTINE PRACTICES AND PROCEDURES

There are no common or uniform international requirements for imports or exports of aquatic animals, with the exception of the European Union (EU) countries which are currently harmonising import requirements. Quarantine and health certification requirements for imported live aquatic animals are the responsibility of importing countries. Increasing attention is, however, being directed at establishing a level of standardisation and uniformity in approaches to quarantine and disease control through organisations including the International Office of Epizootics (OIE), the International Council for Exploration of the Sea (ICES) and the European Inland Fisheries Advisory Council (EIFAC).

Recently, the international importance of the prevention of spread of diseases of aquatic animals has been stressed, exemplified by recommendations arising from the 17th Conference of the OIE Regional Commission for Asia, the Far East and Oceania, namely:

1. That importing countries should seek protection from aquatic animal diseases by requesting veterinary and phytosanitary certificates for aquatic animals and products.
2. That countries in the region strengthen their competence in the diagnosis, control, prevention and eradication of economic and zoonotic diseases of aquatic animals.

Further, several authorities including Arthur (1987), DeKinkelin (1992), DeKinkelin and Hedrick (1991) and Turner (1988) have discussed guidelines on which international quarantine practices may be based. Thus, much recent attention is focussed on the prevention of the spread of aquatic animal diseases by the implementation of effective quarantine measures at the individual country level.

Two international codes of practice exist on which national policies for aquatic animal quarantine may be based: the International Animal Health Code of the OIE and the Codes of Practice of, and Manual of Procedures for Consideration of Introductions and Transfer of Marine and Freshwater Organisms of ICES/EIFAC (DeKinkelin and Hedrick 1991, Turner 1988). These codes are recommended as a basis for health certification and disease control for aquatic animals, but they are not binding. Individual countries are free to accept, modify or reject any or all parts of the codes.

**Table 2: Diseases of fish, crustaceans and molluscs listed by OIE (June 1993)**

| Fish Diseases                                     | Molluscan Diseases                     |
|---|--|
| Viral haemorrhagic septicaemia                    | Bonamiasis                             |
| Spring viraemia of carp                           | Haplosporidiosis                       |
| Infectious haematopoietic necrosis                | Perkinsosis                            |
| Salmonid herpes virus (Type 2)                    | Iridoviruses                           |
| Renibacteriosis <i>Renibacterium salmoninarum</i> | Mikrocytosis <i>Mikrocytos mackini</i> |
| Ictalurid herpes virus (Type 1)                   |  |
| Epizootic haematopoietic necrosis virus           |  |
| Edwardsiellosis <i>Edwardsiella ictaluri</i>      |  |

OIE is an international animal health organisation comprising 110 member countries worldwide. It promotes awareness on the occurrence, progress and control of serious animal disease problems, and publishes an Animal Health Code which recommends procedures for disease surveillance for domestic and international trade. In addition, OIE produces a manual of recommended diagnostic techniques with standardised methods for the implementation of health surveillance schemes.

The OIE, through its Fish Diseases Commission, has included pathogens of fish, crustaceans and molluscs in the Animal Health Code (Table 2) and is currently producing recommended diagnostic techniques for fish and aquatic invertebrates. These documents may be used as models on which standard health certification systems for international trade in live aquatic animals might be based (DeKinkelin 1992).

The ICES / EIFAC Code includes inspection and certification, quarantine and pathology, and environmental impact considerations. Only are mentioned codes detail practical requirements for quarantine and health certification for movements of aquatic animals (Turner 1988, DeKinkelin and Hedrick 1991). The ICES/EIFAC Code defines a different set of diseases of importance, exemplifying difficulties in attaining international agreement on quarantine practices and procedures.



# NATIONAL QUARANTINE PRACTICES AND PROCEDURES

There are as many differing sets of quarantine practices and procedures as there are individual countries. Policies are frequently under revision, considerable variations occur between countries and little uniformity is evident. Some countries have highly restrictive practices designed to ensure importation of aquatic animals free from specific pathogens; others have quarantine legislation which is inadequately enforced; others still have no restrictions or quarantine requirements. More rigorous regimes of quarantine and health certification, exemplified by the Canadian Fish Health Protection Manual of Compliance (Anon. 1984), require two years quarantine of breeding fish with biannual laboratory testing for viral, bacterial and other diseases in each age class prior to export. Export of disinfected ova and certification by a signatory accredited by the Government of Canada are also required.

The onus is thus on the importing country to establish its own requirements for aquatic animal quarantine, taking into account the endemic disease status, the need to protect fisheries resources from exotic disease incursions and the need to maintain export markets based on healthy stocks of fish or aquatic invertebrates.

# REGIONAL QUARANTINE PRACTICES AND PROCEDURES: PACIFIC ISLANDS AND HAWAII

## Precedents

The importance of protection of plant and domestic animal species from exotic diseases and pests through an effective system of quarantine has long been recognised and recommended for the Pacific Islands region. Requirements for provision of designated ports of entry, approved quarantine premises, declarations and inspections of goods and manifests, issuance of quarantine permits, holding, treatment and release from quarantine, seizure and destruction, specific exclusions from quarantine, pre-import sanitary certification, prohibited species, and provisions for penalties have been generally accepted (Morschel 1988a, 1988b, Anon. 1977, 1980, 1984a). Implementation of regional quarantine has been further assisted by the 'Quarantine Advisory Leaflet' and 'Ag Alert' series published by the South Pacific Commission, publications containing information on new agricultural pest and disease outbreaks and topical information on animal health and plant protection matters.

In recent years, the need for quarantine of aquatic animals entering the Pacific Islands region has been recognised. In 1985, concerns at the Regional Technical Meeting on Fisheries of the South Pacific Commission regarding potential diseases associated with introductions and transfers of marine species, especially tridacnid clams, resulted in recommendations for the adoption of interim quarantine guidelines. These recommendations included introductions from within the natural distribution range of the species, the use of quarantine with filtered, sterilised water preceding transfer, transfer of young stages, water filtration for the quarantine facility and disposal of water after transfer, destruction of stock with diseases or predators, sterilisation of all equipment, and appropriate certification of compliance by a competent exporting authority (Adams 1993).

The need for extreme caution in considering introductions was emphasised by Anon. (1988), at which time pre- and post-transfer quarantine and assurance of freedom from diseases, parasites and associated species was suggested. Morschel (1988b) in revising quarantine regulations for Micronesia proposed that fish and lower animals be subject to quarantine. Identification of potential infectious agents, quarantine precautions to prevent spread of such agents, and the establishment of a regional centre for diagnosis and dissemination of information on diseases and quarantine procedures were recommended in 1988 by Copland and Lucas (1988). Govan (1992a) stressed the importance of adopting South Pacific Commission guidelines for preventing transfers of pests when moving Tridacnid clams. Eldredge (1994) stressed the potentially destructive effects of disease introductions. Braley (1991) in addressing a specific disease problem, recommended that pearlshell spat for Tokelau be obtained from regions other than Okinawa as there was less chance of introducing new parasites or viruses.

## Quarantine legislation: Hawaii

Quarantine for imports of aquatic animals is dependent on the species to be imported, life stage, current knowledge of pests, predators and pathogens, and disease history. The introduction into Hawaii of all live animal and plant species is under the jurisdiction of the Department of Agriculture, State of Hawaii and the relevant agencies of the United States Government. Requests for aquatic animal introductions are treated in a similar manner as requests for other types of animals, with consideration of import requests by specialist subcommittees. Unless exempted, most aquatic animals imported for aquaculture or fisheries resource development are held in isolation or closed systems until inspected and found free of exotic pests, predators or pathogens. Such imports are then released from the isolation area, but are required to be kept in captivity, with subsequent periodic examinations. Effluent from the holding areas must be disinfected or placed into a well system (Brock 1986).

### **Quarantine practices: Pacific Island countries**

The first recorded regional example of quarantine restrictions on aquatic animals occurred when a highly fatal, apparently infectious disease of pearl oysters resulted in restrictions on the transfer of infected oysters from atolls in the Gambier group and Tuamotu group (Anon. 1985). Eldredge (1994) further described the attempt to control the spread of the disease by the French Polynesian Ministry of the Sea.

The most noteworthy example of planned introductions with due regard for quarantine and the prevention of disease incursions was the program used for international transfers of giant clams. Guidelines for the international transfer of giant clams and other molluscs were adopted under Recommendation #7 at the 17th South Pacific Commission Regional Technical Meeting on Fisheries. The program, comprising pre- and post-transfer quarantine and health certification, has been used successfully to minimise the introduction of pathogens and parasites whilst facilitating the export of larval and juvenile *Tridacna* sp. and *Hippopus* sp. (Braley 1992, Govan 1992b, Humphrey 1988, Ledua and Adams 1988, Norton *et al* 1993a).

Ledua and Adams (1988) described the implementation of these guidelines for post-import quarantine of *Tridacna gigas* reintroduced to Fiji, and stressed the feasibility and desirability of using small-scale quarantine facilities in Pacific islands for use with proposed introductions.

### **Human health considerations: zoonotic diseases**

Many diseases and parasites of fish and aquatic invertebrates are capable of infecting humans, with serious and/or fatal consequences (Deardorff and Overstreet 1991). Such infections are usually acquired by eating raw or partially cooked animals. Examples of such zoonotic infections exist in the region (Laird 1961). Protection of humans from disease, especially where fish and shellfish are important components of diets, should be a major consideration of regional and national quarantine.

# GENERAL DISCUSSION

## **Intent of quarantine**

An effective and responsible system of animal quarantine should facilitate trade and transfers of animals and their products whilst minimising entry of diseases, pathogens or pests exotic to the importing country. Quarantine programmes must therefore establish whether candidate species for transfer are free of prescribed or other pathogens or pests, and expedite their introduction and release with a reasonable assurance of freedom from disease.

Quarantine programmes should not be unnecessarily rigorous or expensive so as to preclude imports or encourage smuggling or illegal importations; neither should they be so lax as to be ineffective in detecting and preventing entry of diseases. Such programmes should be readily implemented and be cost-effective, commensurate with the degree of difficulty in determining freedom from disease.

## **Benefits of introduced species**

Many social and economic benefits have accrued through the importation of aquatic animal species. Governments and quarantine authorities have a responsibility to consider proposals to introduce aquatic animals and to support such activities where benefits are likely.

## **Benefits of disease freedom**

South Pacific countries are fortunate in that many of the serious diseases of aquatic animals elsewhere are not recorded; the regional absence of such agents is an asset worth protecting. Specific benefits of maintaining relative freedom from disease include:

- Increased productivity,
- Decreased costs,
- Market accessibility,
- Pathogen-free broodstock.

Benefits of relative freedom from disease, combined with the regional fisheries and aquaculture resources, place the Pacific Islands in a commanding situation with regard to aquaculture and future breeding and export of disease-free aquatic animals.

## **The case for aquatic animal quarantine**

Within the region numerous species of aquatic animals are present, both introduced, or resident, which are susceptible to many exotic diseases. Also, diseases occur within the region which should be restricted from spreading further. Thus, a compelling case exists for the formulation and implementation of quarantine practices and procedures to protect regional fisheries by preventing incursions of exotic disease in introduced or transferred aquatic animals into or within the region,

## **Variable factors and circumstances**

Protocols for quarantine and health certification of aquatic animals are difficult to formulate due to variable factors which influence the risk of introducing disease. These variable factors include age, species, source, history, known disease status, reliability of health certification, the known diseases of the candidate species, the status of exporting country or region, and facilities and capabilities of importing and exporting authorities.

### Quarantine location

As multiple sets of circumstances arise in relation to quarantine options for transfers of aquatic animals, one of three sets of circumstances will generally apply to any such transfer.

1. **Quarantine primarily in country of origin.** Where the exporting country and the exporting farm can demonstrate strict quarantine, a high level of assurance with regard to freedom from disease and full and authoritative compliance with import requirements. This option is preferable, as most testing can be conducted prior to transfer, with minimum risk of introducing disease to the importing country.
2. **Quarantine in countries of origin and entry.** Where lesser levels of assurance of freedom from disease in the country of origin apply, where the spectrum of pathogens and parasites is poorly defined for the candidate species, or where latent infections are likely. Quarantine in the importing country is an added precaution against the entry of asymptomatic infections of low prevalence which may become manifest at a later date.
3. **Quarantine primarily in country of entry.** Where an exporting country cannot demonstrate the required capability for quarantine and testing of stocks, this should be carried out in the importing country. A higher risk of escape of disease accompanies this option.

### Quarantine on a case-by-case basis

All proposals for introductions of aquatic animals should be considered, and appropriate quarantine implemented on a case-by-case basis. Such an approach resolves unnecessarily stringent or excessive quarantine, and also allows the implementation of more stringent measures in the case of high-risk introductions.

A single quarantine protocol which would address all variable circumstances for every proposed transfer of aquatic animals is thus not possible to formulate. For example, an oyster species transferred as spat reared in a quarantine facility, and subject to detailed testing for disease from a region free of diseases of concern may be introduced with a minimum of concern. Conversely, mature oysters of the same species, obtained from the wild, from a region known to be infected with serious diseases, with no quarantine or disease testing, would pose a major risk and necessitate stringent quarantine measures.

### Disease threats: ranking of risk

The disease threat posed by imported aquatic animals will vary considerably, depending on the variable circumstances noted. As such, differing levels of quarantine stringency will apply in order to minimise introduction of diseases and pests.

It is possible to semi-quantify the relative risk involved under different sets of circumstances in order to evaluate the level of quarantine stringency which should be applied to a particular case. A score system which may be used for this purpose is shown in Table 2, page 13. On this basis, aquatic species proposed for transfer may be classified into lower or higher risk categories.

### Stringency of quarantine

Options and measures available to ensure the introduction of disease free aquatic animals are unrestricted and may be imposed on potential imports in multiples of ever-increasing levels of isolation, segregation, sampling and health-testing programmes and procedures. Often, the cost or complexity of such procedures effectively precludes the importation of a species. Thus, the level of stringency applied to a proposed transfer should be commensurate with the relative risk of entry of disease.

### **Minimum quarantine practices**

The majority of disease agents spread by international transfers of aquatic animals have been external protozoan or metazoan parasites. Many are readily observable and may be treated. In the majority of these cases, the application of inexpensive, fundamental quarantine and treatment may well have prevented the incursions of these agents.

A minimum level of quarantine and health certification is therefore essential in preventing spread of diseases. Such procedures can be basic in nature and are not necessarily technically or economically demanding, but can be effective in preventing unwanted spread of diseases.

Minimum requirements are discussed below, and additional measures which may be taken with high-risk introductions are further described in Appendix III.

### **Compliance with international guidelines**

Quarantine practices, where possible, should comply with recognised practices and guidelines, especially in regard to sampling and testing.

### **International obligations and courtesies**

Importing nations should advise neighbouring countries of the intention to import at an early stage, to avoid potential conflicts with regards to species and disease introductions, especially where introduction into one country might adversely affect another. A regional authority such as the South Pacific Commission might play a role in this area.

### **Designation of importing authority**

A government or semi-government authority should be designated and a mechanism defined to assess proposals to transfer, evaluate likely outcomes of transfers, the suitability of proposed procedures, the role of parasites and diseases of the candidate species and to recommend to government appropriate actions. The authority should be a forum for lodgement of objections to the proposed movement. The same authority should specify quarantine conditions and requirements. This authority may also consider ecological implications of proposed transfers, or may seek this information elsewhere.

The importing authority must be represented by individuals with expertise in the fisheries and veterinary sciences and quarantine, as transfers involve detailed knowledge from these fields (See Interdisciplinary Collaboration). In many countries, a national specialist committee or working group is appointed for this purpose. Often, one government department responsible for animal quarantine or fisheries retains final authority guided by recommendations of the committee. Examples include the Fisheries Research Directorate, Department of Fisheries and Oceans, Canada (Anon 1984b), the State of Hawaii Department of Agriculture (Brock 1986), and the Australian Quarantine and Inspection Service.

Authoritative international bodies such as the International Council for Exploration of the Sea (ICES) may also assume the role of an importing authority, providing advice to governments on introductions and transfers (Turner 1988). The SPC is well placed to provide similar advice to the Pacific Islands region.

### **Regional legislation and uniform practices**

Legislation is essential to support quarantine for introductions or transfers of aquatic organisms. Legislation should empower quarantine authorities in importing countries to monitor, confiscate, destroy, enter facilities and regulate all aspects of the transfer and introduction process.

Quarantine practices that are common across a geographic region, such as the South Pacific, confer considerable benefits with regard to ease of movement within, and protection from diseases from outside, the region.

### **Problems of disease diagnosis and diagnostic requirements**

Some level of diagnostic capability and expertise is essential to satisfactorily certify that candidate species are free from diseases and pests. In many cases it is possible to make preliminary examinations and assessments using basic equipment. Subsequent collaboration with more specialised laboratories can usually be arranged, if required for the diagnosis of viral and bacterial diseases and to undertake histopathological examinations. Therefore, in line with the provision of basic quarantine facilities, basic laboratory resources and skills can provide vital first-line protection against introductions of pests and diseases, with additional resources being utilised as necessary.

Aquatic animals often carry infectious micro-organisms in the absence of disease and satisfactory diagnostic procedures may not be available. The detection of latent infections may be most difficult, and may require specialised laboratory resources and trained personnel. Many diseases affecting fish are poorly characterised, and new diseases are certain to emerge, providing additional complexities to disease diagnosis. Thus, importation of fish and aquatic invertebrates presents major challenges for quarantine, fisheries and veterinary authorities.

### **Interdisciplinary collaboration**

The transfer of fish into new locations impinges on many specialist scientific disciplines. As such, authoritative comment and involvement is essential from fisheries and veterinary scientists, as well as quarantine personnel, in developing and implementing aquatic animal quarantine and health-testing programmes. Further, this collaboration should extend to scientific and quarantine staff in countries of export, and may involve the international scientific community where disease or ecological matters arise.

### **Ecological considerations**

Consideration of aquatic animal introductions or transfers raises issues relating to ecological impacts. Quarantine should address prevention of disease or pests only in the transferred species; Ecological impacts of proposed introductions should be the responsibility of personnel qualified to make such assessments. A strong case exists for an expert committee which can address both issues.

### **Destruction of animals**

Animals in quarantine incurring diseases of quarantine importance, unknown diseases or diseases refractory to treatment should be destroyed, and all associated materials sterilised or disinfected. A decision to destroy should be based strictly on disease concerns, should be made by the importing government after due consideration by the importing authority, and should be supported by appropriate legislation.

### **Dynamic nature of aquatic animal quarantine**

Knowledge relating to diseases of aquatic animals is constantly changing. Therefore, any system of quarantine must be capable of adapting to new or changing circumstances.

# GUIDELINES AND PRINCIPLES FOR REGIONAL AQUATIC ANIMAL QUARANTINE

## General concepts

1. A compelling case exists for quarantine of aquatic animals in view of the regional freedom from many serious diseases, the history of serious disease introductions elsewhere and the importance of fisheries resources to the Pacific Islands.
2. Quarantine should facilitate the transfer of aquatic species, be practical and readily implemented, should utilise readily available facilities, and be cost efficient. It should not be excessively restrictive to trade or impose unnecessary requirements.
3. The stringency of quarantine should be consistent with the relative risk of introducing disease with the candidate species. A standard of quarantine should be applied to all introductions or transfers, with increased stringency as the risks of introducing diseases in the candidate species increase. Classification into lower-risk and higher-risk categories is essential.
4. Quarantine should be implemented on a case-by-case basis which evaluates all circumstances and variable factors relating to the proposed introduction or transfer. A full disease history of the candidate species, including a detailed review of specific pathogens, parasites, diseases and pests, and the status of these agents in the source country or region of origin should be compiled.
5. A protocol should be then developed which includes the formulation or application of specific tests or diagnostic procedures which are considered necessary to detect specified disease agents, numbers and times of sampling, methods of sampling and other specific instructions to be followed in relation to the candidate species. Compliance with this protocol should form the basis of the Certificate of Health issued by the testing and certifying authority.
6. Quarantine and health certification procedures should be conducted pre-transfer and post-transfer. Where possible, the major component of quarantine and health examination should be conducted pre-transfer.
7. Where resources or information are insufficient to reasonably permit the safe introduction of a species with minimum quarantine, especially those species known to harbour major pathogens, then increased stringency of quarantine and health certification should be applied or the transfer stopped. A full and reasonable explanation for such actions should be given to the proponents of the transfer.
8. Conversely, where a case for transfer with reasonable assurances of freedom from disease or adverse ecological impact exists, importing and exporting authorities should support, encourage and expedite such transfers, imposing minimum quarantine requirements commensurate with relative risk.
9. Quarantine protocols should generally comply with international standards of sampling and testing. They should, however, satisfy the requirements of the importing country or region in the first instance.
10. Quarantine and health certification protocols should be developed in collaboration between fisheries scientists, veterinary scientists and quarantine authorities.

11. An importing authority with an advisory role to governments on quarantine issues should be formed. This body should act as the forum for all issues relating to imports of aquatic animals and should consist of authorities in fisheries and veterinary science, quarantine and aquatic ecology.
12. Legislation should be present on which national quarantine can be implemented or enforced, including the destruction of animals in quarantine.
13. Development of quarantine protocols requires a detailed knowledge of the disease status of aquatic animals within the region, and the nature and range of specific exotic diseases which may affect or be carried by the candidate species. In this regard a national or regional data-base should be developed which may be continuously updated as new information becomes available.
14. In the first instance, considerable levels of assurance for freedom from disease are achievable by holding and observing animals in quarantine, testing for parasites and treating if necessary. Access to more specialised laboratories and resources may be necessary to diagnose certain diseases.
15. Sampling for diseases should be done on a statistical basis on at least two occasions during the quarantine period, with testing immediately prior to transfer.
16. Testing for disease of concern should employ the most sensitive available methods.
17. All animals which appear unhealthy or are diseased during the transfer period in quarantine should be examined by a competent authority and the cause of the disease determined.
18. Training in disease recognition, basic diagnostic techniques and the identification of protozoan and metazoan parasites is highly desirable for quarantine staff as an aid to implementation of effective quarantine.
19. Quarantine practices and procedures must be capable of changing with new circumstances.
20. Quarantine and freedom from disease in introduced species should be treated as a separate issue from the ecological or environmental impacts of such movements.

Many guidelines and recommendations are available on which quarantine policies and practices may be based (Anon. 1984b, Braley 1992, DeKinkelin 1992, DeKinkelin and Hedrick 1991, Govan 1992b, Humphrey 1988, Langdon 1990, Ledua and Adams 1988, Norton *et al* 1993a, Rohovec 1983, Turner 1988).

#### **Minimal quarantine requirements**

The following are minimum quarantine requirements which should apply to all transfers or introductions, depending on the risk of entry of disease. Additional measures which should be undertaken, depending on an evaluation of the risk of disease entry (Table 3), are discussed further in Appendix III.

#### **Transfer of embryonic or juvenile animals**

The range of diseases that may be carried generally increases with age. Eggs, embryonic or juvenile life stages should therefore be transferred. Further, it is generally easier to maintain young animals in captivity under quarantine conditions. Animals should be of the same age and be derived from the same source or population.

### Transfers on a batch by batch basis

Candidate species should be transferred on a batch-by-batch basis: a batch is a group of animals of the same age derived from the same population and maintained as an individual group. Mixing of animals, water or equipment between batches should be avoided.

### Total time in quarantine

Time spent in quarantine should be determined by the relative risk of the transferred species introducing exotic disease (Table 3).

It is not possible or desirable to stipulate exact times during which animals should be held in quarantine as the total time required pre- and post-transfer will depend on a detailed evaluation of all variable factors relating to the candidate species. For **lower-risk species**, a minimum total time in the order of several weeks to several months is likely, with prolonged, even lifetime quarantine for certain **higher-risk species**.

### Pre-transfer quarantine

Animals for transfer should be collected and placed in a quarantine facility for health examination and disease testing. Treatment should be used as necessary prior to transfer (see treatments and medications).

Animals must be critically examined for evidence of disease. Subsamples of the population must be taken for examination for parasites or other pathogenic agents on at least one occasion prior to transfer. Any animal which shows signs of disease should be examined and the cause of the disease determined or the transfer should be aborted.

**Table 3: Semi-quantitative scoring system for assessment of quarantine stringency for imports of aquatic animals**

|                                    |                                   | Risk Category  | (Score) |
|------------------------------------|-----------------------------------|--|---------|
|                                    |                                   | Lower  | Higher  |
| Age at transfer                    | Egg                               | + (1)  |         |
|                                    | Larvae or juveniles               | + (1)  |         |
|                                    | Adult                             |  | +(100)  |
| Source                             | Farm or hatchery                  | + (1)  |         |
|                                    | Wild-caught                       |  | +(100)  |
| Geographic origin                  | Within natural range              | + (1)  |         |
|                                    | Outside natural range             |  | +(100)  |
| Country or regional disease status | Free of specified diseases        | + (1)  |         |
|                                    | Status uncertain                  |  | +(100)  |
|                                    | Specified diseases present        |  | +(100)  |
| Diseases in candidate species      | Major diseases not described      | + (1)  |         |
|                                    | Recognised host to major diseases |  | +(100)  |
| Interpretation                     | <i>Score</i>                      | <i>Quarantine strategy</i>   |         |
|                                    | < 105                             | Minimum quarantine   |         |
|                                    | 200-400                           | Higher stringency (e.g. giant clam protocol)                               |         |
|                                    | > 400                             | Prolonged quarantine and testing of parent stock with transfer of progeny. |         |

### **Post-transfer quarantine**

Animals should enter quarantine in the importing country for health examination and disease testing.

Animals must be critically examined for evidence of disease. Subsamples of the population must be taken for examination for parasites or other agents. Any animal which shows signs of disease should be examined and the cause of the disease determined. If the cause cannot be determined, or if pathogens or parasites of importance are found, the transfer should be aborted.

### **Approvals and permits**

In order to prevent the introduction of disease or undesirable species, a system of permits and approvals is necessary between responsible authorities in importing and exporting countries. A system of approvals and permits which might be used is given in Appendix III.

### **Release from quarantine**

Animals must not be released from quarantine in the importing country until approval is given by the certifying authority. Only animals which are healthy and do not pose a disease risk should be released from quarantine.

### **Quarantine inspection procedures**

Each consignment of transferred animals should be inspected by an official appointed by the importing authority upon entry to ensure that imported species comply with any specified conditions and that no other extraneous animals or plants are present. This person should ensure all requirements for quarantine are met.

### **Quarantine facilities and management**

A physically-secure, isolated quarantine facility is required pre-transfer and post-transfer to hold candidate species. Pre-transfer, the facility should provide for the health examination of the candidate species with minimal risk of infection by further diseases. Post-transfer, the facility should provide for health examination and disease testing while minimising escape of any diseases which may have inadvertently been imported with the transferred species.

***Physical security.*** Physical security of the quarantine facility should be ensured to prevent against inadvertent entry by unauthorised persons and possible loss or release of quarantined animals. The facility should be located within or adjacent to an existing fisheries or animal health station, preferably with 24 hour-supervision. The facility should be isolated by a fence or other structure; it should be lockable, with access only to designated personnel.

***Location.*** Tanks, ponds, pools or other containers of an appropriate size and volume for the introduced candidate species should be constructed in a location physically isolated from other aquaculture facilities, waterways, ponds or dams. Construction and siting should be such that, in the event of an accidental spill or discharge, no water or animals therein will gain access to natural waterways. The siting of the facility on sand, with construction of surrounding raised barriers of sand or soil is a satisfactory safeguard.

***Intake of water.*** Water should be derived from clean unpolluted sources. Incoming water should be filtered during pre-transfer quarantine to prevent introductions of diseases and parasites during the quarantine period.

***Discharge of water.*** All water leaving the facility should be regarded as potentially infected. As such, provision for containment in a sump or pond, with subsequent chemical disinfection or discharge into a land-based pit or pond is necessary. Effluent must not be discharged directly into natural waterways.



**Water flow and plumbing.** The post-transfer facility should be constructed such that an air lock is present on the inflow in order that accidental discharge via the inlet piping is impossible.

**Water management.** Wherever possible, recirculating systems should be installed, with replenishment of water on a regular basis as required.

Continuous flow systems in the post-transfer facility should be avoided due to problems of disinfection or sterilisation of large volumes of effluent water. If such systems are used, strategies described in Appendix III should be adopted, including filtration, chemical disinfection or irradiation.

**Equipment.** All equipment used, for example nets, containers, pipes, hoses, pumps, should remain within the facility and not be used for any other purpose unless disinfected or sterilised.

**Laboratory area.** A covered, enclosed area which can be used as a laboratory is necessary to prepare samples and undertake microscopic examinations. Containers and reagents should be available to collect samples for dispatch to other laboratories for further examination if necessary.

### **Disease diagnosis and health examinations**

The microscopic examination of wet preparations of specimens taken from candidate species to diagnose or exclude the presence of external protozoan and metazoan parasites is a minimum requirement.

Microscopic examination for such agents is readily undertaken by individuals with a basic knowledge of biology and access to dissecting and compound microscopes. Training in the recognition of protozoan and metazoan parasites of fish and aquatic invertebrates, at least in broad taxonomic groups, as a basis for treatment, would be an advantage.

All animals which die or are unhealthy should be examined. Access to more specialised laboratory facilities and personnel with experience in fish and shellfish diseases is desirable to assist if disease problems are not resolvable with the facilities available. A number of such laboratories are available where detailed, microbiological and pathological examinations may be undertaken.

A number of illustrated text books on the subject are available and should be referred to.

Health examinations are necessary to detect sub-clinical or inapparent infections in the population. At least one such examination should be conducted pre-transfer and at least one such examination conducted post-transfer. The number of animals sampled should be in accordance with standard sampling procedures, and the number taken commensurate with the known or suspected disease prevalence of specific diseases.

### **Freedom from specific diseases**

A checklist of diseases or parasites known to affect the candidate species should be compiled and used as a basis for health certification for freedom from such diseases. The use of specific diagnostic procedures and tests may be necessary, and circumstances may require more stringent quarantine restrictions than the minimum specified.

### **Treatment and medication**

Many diseases, especially the common diseases caused by external protozoans and metazoan agents, can be treated using simple, readily-available chemicals.

Commonly-used techniques for treatment include baths of salt or formalin, decreased salinity or short-term freshwater baths for marine species. Other reagents may be used and may be acquired specifically for the project.

Note, however, that many parasites, especially the internal parasites, cannot readily be treated.

Holding of stock from the wild often results in severe outbreaks of external protozoal or metazoan parasites: This should be prevented by an initial treatment of animals on entering the quarantine facility.

#### **Health certification**

A permit or certificate from a responsible authority in the country of origin verifying the origin of the stock, the stage to be exported, the quarantine and disease history, and assurance that all specified tests, procedures and sampling have been conducted in accordance with the requirements of the importing country should be forwarded to the importing authority prior to transfer of the animals.

#### **Release from quarantine**

Only animals which are healthy should be released from quarantine.

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# APPENDIX I

A partial listing of intentional and unintentional introductions of fish and aquatic invertebrates to islands of the South Pacific region and Hawaii (from Eldredge 1994).

(m = marine species; f = freshwater species)

## Fish

- Albula vulpes* bonefish (m)  
*Anabas testudineus* climbing perch (f)  
*Anchoa compressa* (m)  
*Ancistrus* sp. bristle-nosed catfish (f)  
*Aristichthys nobilis* bighead carp (f)  
*Arius* sp. (f)  
*Astronotus ocellatus* cichlid (f)  
*Barbus semifasciatus* Chinese barb (f)  
*Betta brederi* fighting fish (f)  
*Bidyanus bidyanus* silver perch (f)  
*Carassius auratus* goldfish (f)  
*Caranx* sp. jacks (m)  
*Centropyge flavissimus* lemonpeel angelfish (m)  
*Cephalopholus guttatus* (m)  
*Cephalopholus urodeta* (m)  
*Channa (Ophiocephalus) striata* striped snakehead (f)  
*Cichla ocellaris* cichlid (f)  
*Cichlasoma nigrofasciatum* convict cichlid (f)  
*Cichlasoma* sp. cichlid (f)  
*Cichlasoma spilurum* Cutter's cichlid (f)  
*Clarias batrachus* walking catfish (f)  
*Clarias macrocephalus* catfish (f)  
*Colossoma macroporum* pacu (f)  
*Corydoras aeneus* bronze corydora (f)  
*Cromileptes altivelis* (m)  
*Ctenopharyngodon idella* grass carp (f)  
*Cyprinus carpio* common carp (f)  
*Dorosoma petenense* (m)  
*Epinephelus fasciatus* (m)  
*Epinephelus hexagonatus* (m)  
*Epinephelus merra* (m)  
*Epinephelus spiniger* (m)  
*Fundulus grandis* (m)  
*Gambusia affinis* mosquitofish (f)  
*Herklotsichthys quadrimaculatus* (m)  
*Hypophthalmichthys molitrix* silver carp (f)  
*Hypostomus* sp. suckermouth or armoured catfish (f)  
*Hypseleotris klunzingeri* western carp gudgeon (f)  
*Ictalurus punctatus* catfish (f)  
*Kuhlia rupestris* (m)  
*Lates calcarifer* barramundi (f)  
*Leiopotherapon unicolor* spangled perch (f)  
*Leoporinus fasciatus* blackbanded leporinus (f)  
*Lethrinus* sp. (m)  
*Lutjanus fulvus* toau (m)  
*Lutjanus gibbus* (m)  
*Lutjanus guttatus* snapper (m)  
*Lutjanus kasmira* taape (m)  
*Macquaria (Plectroplites) ambigua* golden perch (f)  
*Macquaria colorum*  
*Macquaria novemaculata* Australian bass (f)  
*Mesopristes argenteus* silver grunter (f)  
*Micropterus dolomieu* bass (f)  
*Micropterus salmoides* bass (f)  
*Misgurnus anguillicaudatus* Oriental weatherfish (f)  
*Morone saxatilis* striped bass (m)  
*Mugil cephalus* mullet (m)  
*Mugiligobius parvus* (m)  
*Neopomacentrus violascens* (m)  
*Omobranchus elongatus* (m)  
*Oncorhynchus mykiss* rainbow trout (f)  
*Oncorhynchus tshawytscha* Chinook salmon (m)  
*Oreochromis aureus* blue tilapia (f)  
*Oreochromis hornorum* (f)  
*Oreochromis mossambicus* tilapia (f,m)  
*Oreochromis niloticus* Nile tilapia (f)  
*Osphronemus gourami* giant gourami (f)  
*Osteoglossum* sp. arowana (f)  
*Parablennius thysanius* (m)  
*Parioglossus* sp. (m)  
*Peckoltia* sp. armoured catfish (f)  
*Pelvicachromis pulcher* (f)  
*Percalates colonorum* estuary perch (f)  
*Petroscirtes breviceps* blenny (m)

*Plecoglossus altivelis* (m)  
*Poecilia latipinna* (f,m)  
*Poecilia mexicana* (f,m)  
*Poecilia reticulata* guppy (f)  
*Poecilia* sp. topminnow (f)  
*Poecilia vittata* (f,m)  
*Potamalosa richmondia* freshwater herring (f)  
*Pteroplichthys multiradiatus* armoured catfish (f)  
*Pterophyllum* sp. angelfish (f)  
*Puntius filamentosus* blackspot barb (f)  
*Puntius gonionotus* Javanese carp (f)  
*Puntius semifasciolatus* (f)  
*Puntius seali* barb (f)  
*Retropinna semoni* Australian smelt (f)  
*Salmo trutta* brown trout (f)  
*Salvelinus fontinalis* brook trout (f)  
*Sardinella marquesensis* Marquesan sardine (m)  
*Sarotherodon melanotheron* (m)  
*Serrasalmus* sp. Pirhana (f)  
*Synodontis* sp. African catfish (f)  
*Tandanus tandanus* freshwater catfish (f)  
*Tilapia rendalli*  
*Tilapia* sp. tilapia (f)  
*Tilapia zillii* redbelly tilapia (f)  
*Trachystoma petardi* freshwater mullet (f)  
*Trichogaster pectoralis* snakeskin gourami (f)  
*Trichogaster trichopterus* three-spot gourami (f)  
*Upeneus vittatus* goatfish (m)  
*Valamugil engeli* mullet (m)  
*Xenentodon cancila* Asian needlefish (f)  
*Xiphiphorus maculatus* southern platyfish (f)

### Crustaceans

*Atergadopsis immigrans* crab (m)  
*Callinectes sapidus* blue crab (m)  
*Glabropilumnus seminudus* crab (m)  
*Macrobrachium lar* freshwater prawn (f)  
*Macrobrachium rosenbergii* freshwater prawn (f)  
*Metapenaeus ensis* greasyback shrimp (m)  
*Panopeus herbsti* crab (m)  
*Panopeus pacificus* crab (m)  
*Penaeus japonicus* Kuruma prawn (m)  
*Penaeus indicus* Indian white prawn (m)  
*Penaeus aztecus* northern brown shrimp (m)

*Penaeus merguensis* banana prawn (m)  
*Penaeus monodon* giant tiger prawn (m)  
*Penaeus stylirostris* blues shrimp (m)  
*Penaeus semisulcatus* green tiger prawn (m)  
*Penaeus vannamei* whiteleg shrimp (m)  
*Procambrius clarkii* freshwater crayfish (f)  
*Schizophrys aspera* crab (m)  
*Scylla serrata* mangrove or red crab (m)

### Bivalve Molluscs

*Cellana mazatlanica* limpet (m)  
*Corbicula fluminea* Asiatic freshwater clam (f)  
*Crassostrea belcheri* oyster (m)  
*Crassostrea echinata* Australian oyster (m)  
*Crassostrea gigas* Pacific or Japanese oyster (m)  
*Crassostrea iredalei* Philippine oyster (m)  
*Crassostrea virginica* American oyster (m)  
*Cytheria meretrix* Japanese clam (m)  
*Hippopus hippopus* horse's hoof clam (m)  
*Ostrea edulis* European or flat oyster (m)  
*Perna viridis* green mussel (m)  
*Pinctada fucata martensi* Japanese pearl oyster (m)  
*Pinctada maxima* gold-lip shell pearl oyster (m)  
*Pinctada margaritifera* black-lip pearl oyster (m)  
*Pteria penguin* winged pearl shell (m)  
*Saccostrea cucullata tuberculata* Mangrove oyster (m)  
*Saccostrea commercialis* Sydney rock oyster (m)  
*Tapes japonica* Japanese or Manila clam (m)  
*Tridacna derasa* smooth giant clam (m)  
*Tridacna gigas* giant clam (m)  
*Tridacna squamosa* fluted clam (m)

### Gastropod Molluscs

*Cipangopaludina chinensis* Asian vivipariid snail (f)  
*Haliotis cracherodii* black abalone (m)  
*Haliotis rufescens* red abalone (m)  
*Lymnaea viridis* freshwater snail (f)  
*Pila conica* Freshwater ampullariid snail (f)  
*Pomacea bridgesi* apple or golden snail (f)  
*Pomacea canaliculata* apple or golden snail (f)  
*Pomacea paludosa* apple or golden snail (f)  
*Trochus niloticus* large trochus (m)  
*Turbo marmoratus* green snail (m)

## APPENDIX II

### Diseases of aquatic animals in the South Pacific and Hawaiian Islands: A summary and checklist

While records of pathogens, parasites and pests affecting fish and aquatic invertebrates within the Pacific region are scarce, a small but growing body of data exists, demonstrating that, as elsewhere, disease is a major limiting factor in production of these species. Many agents have been described unassociated with disease, but should be regarded as potential pathogens under different circumstances, especially aquaculture (Table I).

#### Viruses and Viral Diseases

A picorna-like virus is the cause of mortalities in larval barramundi *Lates calcarifer* in Tahiti (Renault et al 1991). The penaeid shrimp viruses, infectious hypodermal and haematopoietic necrosis virus and monodon baculoviruses, are recorded (Lightner 1988, Lightner et al. 1983), but their regional significance is uncertain, although they are associated with high mortalities elsewhere.

#### Bacteria and Bacterial Diseases

Bacterial necrosis and heavy mortalities of cultured larval *Penaeus* sp. and *Macrobrachium* sp. was described in Tahiti, with *Vibrio alginolyticus* causing 'white pleura disease' in older animals (Aquacop 1977). Bacterial necrosis of cultured larval giant clams *Tridacna* sp. associated with *Vibrio* sp., *Aeromonas* sp. and *Plesiomonas* sp. may be a significant problem (Braley 1992).

Rickettsial infections appear common in giant clams *Tridacna* sp. and *Hippopus hippopus*. Mortalities in adult broodstock associated with rickettsial infections from Micronesia (Norton et al. 1993) are recorded, but asymptomatic infections appear common, rarely invoking a granulocytic inflammatory response in juvenile animals (Braley 1992).

Winter mortality, a condition of young juvenile *Tridacna gigas* and *Tridacna maxima*, occurs when clams are exposed to temperatures less than 20°C, and is associated with an intracellular bacterium of unknown identity in surviving animals (Braley 1992, Norton et al. 1993). The condition has resulted in heavy mortalities

#### Fungi and Fungal Diseases

Fungal invasion of juvenile *Penaeus* sp. and associated mortalities were reported by Aquacop (1977) to be due to *Lagenidium* sp. and *Sirolopidium* sp., with gill invasion by *Fusarium* sp. in *Penaeus japonicus* broodstock, resulting in chronic mortalities.

Uncharacterised epiphytic fungi were associated with shell damage in cultured giant clams (Anon. 1991).

#### Protozoa and Protozoal Diseases

Aquacop (1977) reported low grade mortalities and gill necrosis in *Penaeus aztecus* and *Metapenaeus ensis* associated with an uncharacterised amoeboid organism. A *Marteilia* or *Paramarteilia* - like sp. is the cause of nodular lesions in *Tridacna maxima* in Fiji (Braley 1992).

*Perkinsus* sp. appear common in *Tridacna* sp., and *Hippopus hippopus*, but are unassociated with disease (Braley 1992, Norton et al 1993b).

### Algae and Algal Diseases

Uncharacterised epiphytic algae were associated with shell damage in cultured giant clams (Anon. 1991).

### Platyhelminths and Platyhelminth Diseases

**Turbellaria.** A turbellarian parasite *Pseudostylochus* sp. was associated with high mortalities in the oyster *Crassostrea gigas* imported from Japan into Vanuatu for experimental aquaculture (Hallier 1977). *Stylochus* sp. have resulted in high mortalities through predation in oysters *Crassostrea gigas* and *Crassostrea virginica* (Eldredge 1993). *Stylochus* sp. (*S. matatasi*) is a predator of giant clams in the Solomon Islands (Anon 1991).

**Polychaetes.** The mudworm *Polydora* sp. caused severe shell damage in imported oysters *Crassostrea gigas* in Vanuatu and French Polynesia (Eldredge 1993, Hallier 1977). *Polydora websteri* contributed to the collapse of aquaculture of the oyster *Crassostrea virginica* in Hawaii (Bailey-Brock 1990). Polychaete worms were also associated with deaths and shell deformities in cultured giant clams (Anon. 1991).

### Porifera (Sponges) and Poriferan Diseases

The sponge *Cliona* sp. resulted in severe shell damage to imported Japanese oysters *Crassostrea gigas* in Vanuatu (Hallier 1977). Shell damage associated with uncharacterised epiphytic sponges is also reported in cultured giant clams (Anon 1991).

### Crustacea and Crustacean Diseases

Predation of cultured tridacnid clams by crabs, probably xanthid crabs, is reported in the Solomon Islands (Govan 1992a) and xanthid, portunid and diogenid crabs are noted by Govan et al. (1993) as serious predators of tridacnid clams in the region.

### Molluscs and Molluscan Diseases

Predation of cultured giant clams by ranellid gastropods of the genus *Cymatidium*, is a major problem throughout the region (Anon 1987, Eldredge 1993, Govan et al 1993). The oyster drill *Cymatium* sp. killed stocks of oyster *Crassostrea gigas* in Tonga. Infection and predation with pyramidellid gastropod parasites including *Tathrella* sp. and *Turbonilla* sp. is a serious problem of cultured giant clams in Palau, Papua New Guinea, Solomon Islands and Guam (Cumming 1988, Braley 1992, Govan 1992b). At least 20 gastropod predators of giant clams have been identified, which feed on tissues and present major problems for cultured giant clams. These include the genera *Cantharus*, *Vexillum*, *Cymatium*, *Pleuropoca*, *Chicoreus*, *Cronius*, *Morula*, *Thais*, *Tathrella*, *Turbamilla* and *Melo* (Anon. 1991).

### Diseases of Uncertain Aetiology

A 'virus' disease, initially reported in the Gambier group, was reported to be responsible for mass mortalities in pearl oysters *Pinctada margaritifera* on Takapoto atoll, Tuamotu Group, French Polynesia, with severe socio-economic losses. It is uncertain on what basis the 'virus' was diagnosed. The disease spread in cultured black lip pearl oysters in the Tuamotus following transfer of stocks among atolls (Anon. 1985, 1988, Braley 1991, Cabral 1989a, 1989b, Eldredge 1994). Braley (1991) reported the disease in pearl oysters and other bivalve molluscs in the Tuamotu group which had been transferred between atolls. More recently Cabral (1989b) considered the aetiology to be a complex of uncharacterised factors.

Mass mortalities characterised by mantle retraction and death in wild and cultured giant clams *Tridacna gigas* and *Hippopus hippopus* occurred during 1992 in the Eastern Solomon Islands (Gervis 1992).

A bacterial-like agent identified in juvenile *Tridacna gigas* has been associated with deaths after periods of cold weather (Braley 1992).

Epidemics of a disease in several fish species characterised by ulcerating skin lesions, and resembling epizootic ulcerative disease, was reported by Coates (1984) in the Sepik Region of Papua New Guinea. The disease was reported to cause hardship to people higher in the river systems due to depletion of stocks.

**Table 4. A partial checklist and bibliography of pathogens, parasites, pests and diseases recorded in fish and aquatic invertebrates in the South Pacific and Hawaiian regions (Key to countries: AS = American Samoa; CI = Cook Islands; F = Fiji; FP = French Polynesia; FSM = Federated States of Micronesia; G = Guam; H = Hawaii; MI = Marshall Islands; NI = Norfolk Island; PNG = Papua New Guinea; SI = Solomon Islands; T = Tonga; TV = Tuvalu; U = Ubiquitous; V = Vanuatu; Y = Yap)**

#### Viruses and Viral Diseases

| Aetiological Agent            | Disease        | Host Species                       | Distribution | Reference/s   |
|-------------------------------|----------------|------------------------------------|--------------|---|
| <i>Baculovirus penaei</i>     | Mortalities    | 'wild penaeid shrimp'              | H            | Lightner et al 1989                                     |
| Hepatopancreatic parvovirus   |                | Penaeid shrimp                     | H            | Lightner et al 1989                                     |
| IHHNV*                        |                | <i>Penaeus</i> sp.                 | FP, F, H     | Eldredge 1994,<br>Lightner 1988,<br>Lightner et al 1989 |
| Lymphocystis                  |                | <i>Sargocentron punctatissimum</i> | H            | Anderson et al 1988                                     |
| <i>Monodon baculovirus</i>    |                | <i>Penaeus monodon</i>             | FP, H        | Lightner et al 1983, 1989                               |
| Picornavirus                  | Encephalopathy | <i>Lates calcarifer</i>            | FP           | Renault et al 1991                                      |
| Rhabdovirus (uncharacterised) |                | <i>Penaeus</i> sp.                 | H            | Lu et al 1991   |
| Reo-like virus                |                | <i>Penaeus japonicus</i>           | H            | Lightner et al 1989                                     |

\* Infectious hypodermal and haematopoietic necrosis virus

#### Bacteria and Bacterial Diseases

| Aetiological Agent          | Disease            | Host Species                     | Distribution | Reference/s                       |
|-----------------------------|--------------------|----------------------------------|--------------|-----------------------------------|
| <i>Aeromonas</i> sp.        | Bacterial necrosis | <i>Tridacna</i> sp.              | Ubiquitous   | Braley 1992                       |
| <i>Plesiomonas</i> sp.      | Bacterial necrosis | <i>Tridacna</i> sp.              | Ubiquitous   | Braley 1992                       |
| <i>Rickettsia</i> sp.       | Rickettsiosis      | <i>Tridacna</i> sp.              | F, T, FSM    | Braley 1992,<br>Norton et al 1993 |
| <i>Rickettsia</i> sp.       | Rickettsiosis      | <i>Hippopus hippopus</i>         | F, T, FSM    | Braley 1992,<br>Norton et al 1993 |
| <i>Vibrio alginolyticus</i> | White pleura       | <i>Penaeus</i> sp.               | FP           | Aquacop 1977                      |
| <i>Vibrio</i> sp.           | Bacterial necrosis | <i>Tridacna</i> sp.              | Ubiquitous   | Braley 1992                       |
| Uncharacterised             | Bacterial necrosis | <i>Macrobrachium rosenbergii</i> | FP           | Aquacop 1977                      |
| Uncharacterised             | Bacterial necrosis | <i>Penaeus</i> sp.               | FP           | Aquacop 1977                      |
| Uncharacterised             | Winter mortality   | <i>Tridacna gigas</i>            | Ubiquitous   | Braley 1992,<br>Norton et al 1993 |
| Uncharacterised             | Winter mortality   | <i>Tridacna maxima</i>           | Ubiquitous   | Braley 1992,<br>Norton et al 1993 |

**Table 4 (cont'd). A partial checklist and bibliography of pathogens, parasites, pests and diseases recorded in fish and aquatic invertebrates in the South Pacific and Hawaiian regions**

**Fungi and Fungal Diseases**

| Aetiological Agent      | Disease          | Host Species             | Distribution | Reference/s   |
|-------------------------|------------------|--------------------------|--------------|---------------|
| <i>Fusarium</i> sp.     | Gill mycosis     | <i>Penaeus japonicus</i> | FP           | Aquacop 1977  |
| <i>Lagenidium</i> sp.   | Systemic mycosis | <i>Penaeus</i> sp.       | FP           | Aquacop 1977  |
| <i>Sirolopidium</i> sp. | Systemic mycosis | <i>Penaeus</i> sp.       | FP           | Aquacop 1977  |
| Uncharacterised         | 99% mortality    | 'Oysters'                | H            | Eldredge 1994 |
| Uncharacterised         | Shell damage     | 'Giant clams'            | SI           | Anon 1991     |
| Uncharacterised         | Shell damage     | <i>Trochus niloticus</i> | Ubiquitous   | Eldredge 1994 |

**Protozoa and Protozoal Diseases**

| Aetiological Agent                 | Disease             | Host Species                    | Distribution | Reference/s                       |
|------------------------------------|---------------------|---------------------------------|--------------|-----------------------------------|
| <i>Cryptocaryon irritans</i>       | External parasitism | <i>Lates calcarifer</i>         | FP           | Aquacop et al 1989                |
| <i>Cryptocaryon irritans</i>       | External parasitism | <i>Caranx ignobilis</i>         | FP           | Aquacop et al 1989                |
| <i>Cryptocaryon irritans</i>       | External parasitism | <i>Siganus argenteus</i>        | FP           | Aquacop et al 1989                |
| <i>Cryptocaryon irritans</i>       | External parasitism | <i>Epinephelus microdon</i>     | FP           | Aquacop et al 1989                |
| <i>Cryptocaryon irritans</i>       | External parasitism | <i>Coryphaena</i> sp.           | FP           | Aquacop et al 1989                |
| <i>Haemogregarina bigemina</i>     |                     | <i>Clinus perspicillatus</i>    | NI           | Laird 1958a                       |
| <i>Haemogregarina bigemina</i>     |                     | <i>Mugil</i> sp.                | SI           | Laird 1958a                       |
| <i>Haemogregarina bigemina</i>     |                     | <i>Trypterygion rufopileum</i>  | NI           | Laird 1958a                       |
| <i>Haemogregarina mugili</i>       |                     | <i>Awaous ocellaris</i>         | SI           | Laird 1958a                       |
| <i>Haemogregarina mugili</i>       |                     | <i>Stenogobius genivittatus</i> | SI           | Laird 1958a                       |
| <i>Haemogregarina salaritasi</i>   |                     | <i>Salaria periphthalmus</i>    | F            | Laird 1958a                       |
| <i>Hartmanella tahitiensis</i>     |                     | <i>Saccostrea commercialis</i>  | FP           | Angell 1986,<br>Eldredge 1994     |
| <i>Henneguya vitiensis</i>         |                     | <i>Leiognathus fasciatus</i>    | F            | Laird 1950                        |
| <i>Marteilia/Paramarteilia</i> sp. | Focal lesions       | <i>Tridacna maxima</i>          | F            | Braley 1992,<br>Norton et al 1993 |
| <i>Perkinsus</i> sp.               |                     | <i>Hippopus hippopus</i>        | F            | Braley 1992                       |
| <i>Perkinsus</i> sp.               |                     | <i>Tridacna</i> sp.             | F            | Braley 1992,<br>Norton et al 1993 |
| <i>Trichodina</i> sp.              | Gut infection       | <i>Acanthurus xanthopterus</i>  | PNG, H       | Basson et al 1990                 |
| Uncharacterised amoeba             | Gill invasion       | <i>Metapenaeus ensis</i>        | FP           | Aquacop 1977                      |
| Uncharacterised amoeba             | Gill invasion       | <i>Penaeus aztecus</i>          | FP           | Aquacop 1977                      |

**Monogenea and Monogenean Diseases**

| Aetiological Agent  | Disease | Host Species                   | Distribution | Reference/s       |
|---|---------|--------------------------------|--------------|-------------------|
| Phylum PLATHYHELMINTHES<br>Class TREMATODA<br>Order MONOGENEA |         |                                |              |                   |
| <i>Alloposeudaxine</i> sp.                                    |         | <i>Katsuwonus pelamis</i>      | NC           | Rohde et al 1980  |
| <i>Alloposeudaxinoides vagans</i>                             |         | <i>Katsuwonus pelamis</i>      | NC           | Rohde et al 1980  |
| <i>Daitreosoma parva</i>                                      |         | <i>Ambassis miops</i>          | SI           | Laird 1958c       |
| <i>Diplectanum melanesiensis</i>                              |         | <i>Epinephelus merra</i>       | F, V         | Laird 1958b       |
| <i>Microcotyle</i> sp.  |         | <i>Mugil oligolepis</i>        | V            | Laird 1958c       |
| <i>Neobenedenia melleni</i>                                   |         | <i>Oreochromis mossambicus</i> | H            | Kaneko et al 1988 |
| <i>Neothoracocotyle acanthocybii</i>                          |         | <i>Acanthocybium solandri</i>  | NC           | Rohde et al 1980  |

**Digenea and Digenean Diseases**

| Aetiological Agent  | Disease | Host Species                  | Distribution | Reference/s      |
|---|---------|-------------------------------|--------------|------------------|
| Phylum PLATHYHELMINTHES<br>Class TREMATODA<br>Order DIGENEA |         |                               |              |                  |
| <i>Clinostromum</i> sp. (metacercaria)                      |         | <i>Oxyeleotris herwerdeni</i> | PNG          | Coates 1984      |
| <i>Plagioporus</i> sp. (metacercaria)                       |         | <i>Porites compressa</i>      | H            | Aeby 1991        |
| <i>Syncoelium filiferum</i>                                 |         | <i>Katsuwonus pelamis</i>     | NC           | Rohde et al 1980 |
| <i>Stephanostomum</i> sp. (metacercaria)                    |         | <i>Ambassis miops</i>         | SI           | Laird 1958       |

**Table 4 (cont'd). A partial checklist and bibliography of pathogens, parasites, pests and diseases recorded in fish and aquatic invertebrates in the South Pacific and Hawaiian regions**

**Nematodes and Nematode Diseases**

| Aetiological Agent                | Disease    | Host Species                  | Distribution | Reference/s        |
|-----------------------------------|------------|-------------------------------|--------------|--------------------|
| <i>Echinocephalus overstreeti</i> | Parasitism | <i>Taeniura melanospilos</i>  | FP           | Beveridge 1991     |
| <i>Echinocephalus overstreeti</i> | Parasitism | <i>Urogymnus asperrimus</i>   | MI           | Beveridge 1991     |
| <i>Eustrongylides</i> sp.         |            | <i>Oxyeleotris herwerdeni</i> | PNG          | Coates 1984        |
| <i>Eustrongylides</i> sp.         |            | <i>Ophieleotris aporos</i>    | PNG          | Coates 1984        |
| Uncharacterised                   | Parasitism | <i>Epinephelus microdon</i>   | FP           | Aquacop et al 1989 |

**Cestodes and Cestode Diseases**

| Aetiological Agent            | Disease          | Host Species                     | Distribution | Reference/s            |
|-------------------------------|------------------|----------------------------------|--------------|------------------------|
| <i>Dasyrhynchus talismani</i> | Aortic infection | <i>Thunnus albacares</i>         | H            | Brill et al 1987       |
| <i>Floriceps minacanthus</i>  | Parasitism       | <i>Carcharhinus melanopterus</i> | K            | Richmond & Cairra 1991 |
| Uncharacterised               | Parasitism       | <i>Epinephelus microdon</i>      | FP           | Aquacop et al 1989     |

**Turbellaria and Turbellarian Diseases**

| Aetiological Agent  | Disease             | Host Species                 | Distribution | Reference/s                    |
|---|---------------------|------------------------------|--------------|--------------------------------|
| Phylum <b>PLATYHELMINTHES</b><br>Class <b>TURBELLARIA</b> |                     |                              |              |                                |
| <i>Pseudostylochus</i> sp.                                | Predation/mortality | <i>Crassostrea gigas</i>     | V            | Eldredge 1994,<br>Hallier 1977 |
| <i>Stylochus matatasi</i>                                 |                     | 'Giant clams'                | SI           | Anon 1991                      |
| <i>Stylochus</i> sp.                                      | Predation           | <i>Crassostrea gigas</i>     | H            | Eldredge 1994                  |
| <i>Stylochus</i> sp.                                      | Predation/mortality | <i>Crassostrea virginica</i> | H            | Eldredge 1994                  |

**Polychaetes, Polychaetal Diseases and Pests**

| Aetiological Agent                                | Disease        | Host Species             | Distribution | Reference/s                    |
|---|----------------|--------------------------|--------------|--------------------------------|
| Phylum <b>ANNELIDA</b><br>Class <b>POLYCHAETA</b> |                |                          |              |                                |
| <i>Polydora nuchalis</i>                          |                | 'Oysters'                | H            | Bailey-Brock 1990              |
| <i>Polydora websteri</i>                          | Shell damage   | 'Oysters'                | H            | Bailey-Brock 1990              |
| <i>Polydora</i> sp.                               | Shell damage   | <i>Crassostrea gigas</i> | V, FP        | Eldredge 1994,<br>Hallier 1977 |
| <i>Polydora pacifica</i>                          | Shell damage   | 'Pearl oysters'          | P            | Eldredge 1994                  |
| Uncharacterised                                   |                | 'Giant clams'            | SI           | Anon 1991                      |
| Uncharacterised                                   | High mortality | <i>Pinctada maxima</i>   | PNG          | Eldredge 1994                  |

**Crustacea and Crustacean Diseases**

| Aetiological Agent                               | Disease             | Host Species                     | Distribution | Reference/s              |
|--|---------------------|----------------------------------|--------------|--------------------------|
| Phylum <b>CRUSTACEA</b><br>Class <b>COPEPODA</b> |                     |                                  |              |                          |
| <i>Argulus</i> sp.                               |                     | <i>Oxyeleotris herwerdeni</i>    | PNG          | Coates 1984              |
| <i>Argulus papuensis</i>                         |                     | <i>Bunaka herwerdeni</i>         | PNG          | Rushton-Mellor 1991      |
| <i>Branchiella thynni</i>                        |                     | <i>Acanthocybium solandri</i>    | NC           | Rohde et al 1980         |
| <i>Caligus bonito</i>                            |                     | <i>Katsuwonus pelamis</i>        | NC           | Rohde et al 1980         |
| <i>Caligus productus</i>                         |                     | <i>Acanthocybium solandri</i>    | NC           | Rohde et al 1980         |
| <i>Caligus productus</i>                         |                     | <i>Katsuwonus pelamis</i>        | NC           | Rohde et al 1980         |
| <i>Jusheyhoea moseri</i>                         | Parasitism          | Macrourid fishes                 | H            | Kabata 1991              |
| <i>Renocila loriae</i>                           | External parasitism | <i>Apogon (Pristiapogon)</i> sp. | PNG          | Williams & Williams 1992 |

**Table 4 (cont'd). A partial checklist and bibliography of pathogens, parasites, pests and diseases recorded in fish and aquatic invertebrates in the South Pacific and Hawaiian regions**

**Molluscs, Molluscan Diseases and Pests**

| Aetiological Agent          | Disease   | Host Species             | Distribution         | Reference/s                                    |
|-----------------------------|-----------|--------------------------|----------------------|--|
| <b>Phylum MOLLUSCA</b>      |           |                          |                      |  |
| <i>Burtsa granularis</i>    | Predation | Tridacnid clams          | U                    | Govan et al 1993                               |
| Pyramidellid parasites      | Predation | 'Giant clams'            |                      | Govan 1992b                                    |
| <i>Cymatium aquatile</i>    | Predation | Tridacnid clams          | TV, U                | Govan et al 1993                               |
| <i>Cymatium muricinum</i>   | Predation | Tridacnid clams          | AS, CI, FSM          | Anon 1987<br>Eldredge 1994                     |
| <i>Cymatium nicobaricum</i> | Predation | Tridacnid clams          | U                    | Govan et al 1993                               |
| <i>Cymatium pileare</i>     | Predation | Tridacnid clams          | U                    | Govan et al 1993                               |
| <i>Cymatium</i> sp.         | Predation | 'Giant clams'            |                      | Anon 1991                                      |
| <i>Cymatium</i> sp.         | 'Drill'   | <i>Crassostrea gigas</i> | T                    | Eldredge 1994                                  |
| <i>Chicoreus palmarosae</i> | Predation | Tridacnid clams          | U                    | Govan et al 1993                               |
| <i>Chicoreus ramosus</i>    | Predation | Tridacnid clams          | U, Y                 | Eldredge 1994                                  |
| <i>Tathrella iredalei</i>   | Predation | <i>Tridacna</i> sp.      | SI, G, PNG<br>FSM, P | Cumming 1988,<br>Braley 1992,<br>Eldredge 1994 |
| <i>Sabia conica</i>         | Scaring   | <i>Trochus niloticus</i> | U                    | Eldredge 1994                                  |

**Porifera and Poriferan Diseases**

| Aetiological Agent | Disease      | Host Species             | Distribution | Reference/s  |
|--------------------|--------------|--------------------------|--------------|--------------|
| <i>Cliona</i> sp.  | Shell damage | <i>Crassostrea gigas</i> | V            | Hallier 1977 |

**Non-infectious Diseases**

| Aetiological Agent          | Disease             | Host Species                | Distribution | Reference/s |
|-----------------------------|---------------------|-----------------------------|--------------|-------------|
| Decreased irradiation (?)   | Bleaching           | <i>Tridacna</i> sp.         | Ubiquitous   | Braley 1992 |
| Decreased irradiation (?)   | Bleaching           | <i>Hippopus hippopus</i>    | Ubiquitous   | Braley 1992 |
| Decreased temperature <20°C | Chilling            | <i>Tridacna</i> sp.         | Ubiquitous   | Braley 1992 |
| Air supersaturation         | Gas bubble disease  | <i>Tridacna</i> sp.         | Ubiquitous   | Braley 1992 |
| Increased temperature >35°C | Heat stress         | <i>Tridacna</i> sp.         | Ubiquitous   | Braley 1992 |
| Nitrogen deficiency         | Nitrogen deficiency | <i>Tridacna</i> sp.         | Ubiquitous   | Braley 1992 |
| Decreased salinity          | Death, gill lesions | <i>Hippopus porcellanus</i> | Ubiquitous   | Braley 1992 |
| Increased salinity >45ppt   | Shell closure       | <i>Tridacna</i> sp.         | Ubiquitous   | Braley 1992 |

Table 4 (cont'd). A partial checklist and bibliography of pathogens, parasites, pests and diseases recorded in fish and aquatic invertebrates in the South Pacific and Hawaiian regions

## Diseases of Unknown Aetiology

| Aetiological Agent | Disease            | Host Species                        | Distribution | Reference/s  |
|--------------------|--------------------|-------------------------------------|--------------|--|
| Unknown            | Muscle necrosis    | <i>Macrobrachium rosenbergii</i>    | H            | Akiyama et al 1982   |
| Unknown            | Mass mortalities   | <i>Tridacna gigas</i>               | SI           | Gervis 1992  |
| Unknown            | Mass mortalities   | <i>Hippopus hippopus</i>            | SI           | Gervis 1992  |
| Unknown            | 22% mortalities    | <i>Crassostrea iredalei</i>         | F            | Eldredge 1994  |
|                    | Deformed nauplii   | <i>Penaeus</i> sp.                  | FP           | Aquacop 1977   |
| Bacteria (?)       | 'Whirling disease' | <i>Penaeus</i> sp.                  | FP           | Aquacop 1977   |
| Unknown            | High mortalities   | <i>Ostrea edulis</i>                | T            | Eldredge 1994  |
|                    | Ulceration         | <i>Toxotes</i> sp.                  | PNG          | Coates 1984  |
|                    | Ulceration         | <i>Scatophagus</i> sp.              | PNG          | Coates 1984  |
|                    | Ulceration         | <i>Anguilla bicolor pacifica</i>    | PNG          | Coates 1984  |
|                    | Ulceration         | <i>Ophieleotris aporos</i>          | PNG          | Coates 1984  |
|                    | Ulceration         | <i>Oxyeleotris herwerdeni</i>       | PNG          | Coates 1984  |
|                    | Ulceration         | <i>Zenarchopterus kampeni</i>       | PNG          | Coates 1984  |
|                    | Ulceration         | 'Mullet'                            | PNG          | Coates 1984  |
|                    | Ulceration         | 'Arid catfish'                      | PNG          | Coates 1984  |
|                    | Ulceration         | 'Gudgeons'                          | PNG          | Coates 1984  |
|                    | Ulceration         | <i>Acanthopagrus berda</i>          | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Ambassis nalua</i>               | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Bunaka</i> sp.                   | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Cinetodus froggati</i>           | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Datnioides quadrifasciatus</i>   | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Eleotheronema tatradaactylum</i> | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Glossogobius giurus</i>          | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Hexanematichthys lentaspis</i>   | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Hexanematichthys</i> sp.         | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Hexanematichthys danielsi</i>    | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Hexanematichthys latirostris</i> | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Johnius belengeri</i>            | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Kurtus gulliveri</i>             | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Lates calcarifer</i>             | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Liza dussumieri</i>              | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Lobotes surinamensis</i>         | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Lutjanus</i> sp.                 | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Oxyeleotris</i> sp.              | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Parambassis gulliveri</i>        | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Polydactylus sheridani</i>       | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Priopidichthys gymnocephalus</i> | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Scatophagus argus</i>            | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Scutengraulis seratchleyi</i>    | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Selentoca multifasciata</i>      | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Tilapia mossambica</i>           | PNG          | Uwate 1983   |
|                    | Ulceration         | <i>Toxotes chartareus</i>           | PNG          | Uwate 1983   |
| Uncharacterised    | Mass mortalities   | <i>Pinctada margaritifera</i>       | FP           | Anon 1985, Braley 1991, Cabral 1989a, 1989b, Eldredge 1994 |
| Uncharacterised    | Mass mortalities   | 'Molluscs'                          | FP           | Anon. 1985, Braley 1991                                    |
| Uncharacterised    | Lymphoma           | <i>Penaeus vannamei</i>             | H            | Lightner & Brock 1987                                      |

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## APPENDIX III

### General principles and guidelines for aquatic animal quarantine: Minimal requirements and conditions for increased stringency

#### Animals for Transfer

The aim of quarantine should be the transfer of specific disease-free stock, with quarantine and health testing in the country of entry an additional precaution against inadvertent entry of disease. In general, the range of diseases and parasites that may be carried is greater with age. Many such agents are carried in the absence of disease and may be particularly difficult to detect. Thus, the intent should be to transfer embryonic or juvenile life stages in all cases.

The best method to ensure disease free stock is to breed the juvenile life stages from broodstock held in quarantine.

- i. Where possible, the candidate species should be transferred as fertilised ova, or as larval or juvenile stages.
- ii. Stocks of candidate species may be established in a quarantine facility, tested for pathogens, parasites and pests, and subsequently transferred for further evaluation in quarantine in the country of entry.
- iii. Stocks of candidate species for breeding purposes should be established in an approved quarantine facility. The progeny of this stock should be considered for transfer, subject to quarantine and examination for pathogens, parasites and pests as specified.
- iv. Candidate species are collected and transferred directly to quarantine facilities in the importing country. This option necessitates thorough evaluation of disease freedom in quarantine. Animals may be imported with or without health certification and placed in quarantine for further evaluation. In the case of mature animals imported with minimal health certification, such animals should not be released from quarantine; rather, they should be used for breeding purposes, with progeny subject to intensive disease-testing procedures.
- v. Where continued imports are likely, broodstock should be established from imported stocks in an approved quarantine facility in the importing country to obviate the need for continued imports from elsewhere.

#### Approvals and Permits

In order to prevent the introduction of disease, extraneous species, illegal substances or ecologically undesirable species, a formal system of permits and approvals is necessary between responsible authorities in importing and exporting countries.

- i. An *Import Request* should be forwarded by the proponent seeking the transfer to the importing authority in the importing country, providing full details of the proposed transfer and seeking permission to proceed with the transfer.
- ii. An *Approval to Proceed* should be issued by the importing authority in the importing country if, after due consideration of biological, ecological and health risks posed by the proposed transferor, valid reasons as to why the import should not proceed cannot be found. Reasons for rejection of the request should be clearly explained to the proponent. The approval to proceed should clearly and in detail specify the nature of the quarantine, disease testing and

health certification which will be necessary to import the candidate species. These criteria should be developed in consultation with authorities on fish disease and quarantine matters.

- iii. A *Certificate of Health*, duly signed by qualified and designated Government officer/s in the country of export verifying that all requirements for health testing and quarantine have been met should be obtained by the importing authority directly from the exporting authority.
- iv. A *Permit to Import* should then be issued by the importing authority upon satisfactory compliance with specified quarantine, disease testing, health certification and receipt of the *Certificate of Health*. A permit to import should accompany each batch of animals which are transferred.
- v. The importing authority should be advised immediately of any occurrences or diseases, breaches of quarantine or other circumstances which may impact on the security of the candidate species for transfer. Such occurrences may mitigate against the transfer.

### Quarantine Procedures

- i. Broodstock for breeding purposes should be selected from animals with no known history or evidence of disease. Preferably, they will have been maintained under isolated conditions and have been subject to regular examination for pathogens and parasites.
- ii. Any animal selected for broodstock which appears unhealthy should be removed and a detailed pathological examination conducted.
- iii. In the case of fish or crustaceans, prophylactic treatment for external protozoa and metazoa should be conducted. In the case of molluscs, the external shell of each animal should be scrubbed and cleaned to remove all epiphytic animals and plants. Other prophylactic treatments should be applied at the discretion of the importing or exporting authority, provided that pathogens and parasites of quarantine concern are not masked by such treatments.
- iv. Tanks, containers or other holding or culture facilities should be thoroughly disinfected prior to the introduction of the broodstock or stocks for transfer, after removal of stocks and after any outbreaks of disease resulting in losses or destruction of stocks.
- v. Handling and environmental stresses should be minimised at all times.
- vi. In the case of animals spawning in quarantine with the F1 generation proposed for export, ova and sperm should be collected or removed from the tanks immediately after spawning, and fertilization and subsequent incubation conducted in clean facilities supplied by an independent water supply.
- vii. Feeding of broodstock should preferably be done using sterile foodstuffs or, where live foodstuffs are required, the feed should be reared in the laboratory and be free from potential pathogens.
- viii. Animals bred in quarantine for export should best be maintained and monitored for a minimum period of 6 months and preferably 12 months. During this time, periodic sampling of animals for laboratory examination should be conducted and other tests or examinations conducted to determine the presence of disease (see Disease Testing).
- ix. The occurrence of pathogens refractory to treatment, unexplained mortalities or lesions consistent with infectious microorganisms in any batch should be considered grounds for destruction or non-export of that batch.
- x. The occurrence of disease in animals within the facility should result in immediate notification of the importing authority if any animals have been exported.

### Quarantine Facilities and Management

A physically secure quarantine facility should be mandatory for pre-transfer holding and testing of candidate species. Such a facility is necessary to avoid introduction of pathogens and parasites to the stocks in quarantine from the surrounding environment.

The quarantine facilities should best be located in areas remote from aquaculture establishments, natural waterways and other potential sources of contamination.

- i. Incoming water should be sterilised or disinfected to minimise entry of pathogens. This may be achieved in several ways:
  - a. Use of spring, ground, artesian or well waters which contain no flora and fauna prior to intake,
  - b. Filtration to 1 micron, and/or
  - c. Ultraviolet sterilisation, and/or
  - d. Ozone sterilisation.
- ii. A facility based on recirculation of water and biological filtration should be used.
- iii. All candidates for transfer should be maintained in batches, with groups or age classes isolated with respect to water supply, equipment and handling.
- iv. All other animals and birds should be excluded from the quarantine facility.
- v. All ponds, tanks and other containers should be individually identified.
- vi. Daily records of all operating procedures and records for each batch of animals must be accurately maintained and made available for inspection.
- vii. Access to the facility should be restricted to trained, authorised staff who should be under the supervision of appropriately qualified individual staff to ensure compliance with operational requirements.
- viii. Entry and exit for personnel operating the facility should be through an appropriate disinfection station, with footbaths, showers and changes of clothing as a minimum requirement.
- ix. Strict quarantine and disinfection should be maintained by personnel when moving between batches of groups of animals within the facility, including disinfection of hands, arms, legs and feet or boots.

### Disease Testing and Health Examinations

Some measure of laboratory examination is required to detect pathogens and parasites. It is not possible to certify freedom from parasitic protozoa or metazoa without microscopic examination, and examinations for freedom from viral, bacterial and fungal pathogens may require access to specialised laboratories.

- i. Wherever possible, the candidate species should be obtained from a production facility or area that has a documented history of testing and freedom from prescribed diseases, for a period of at least 1–2 years.
- ii. Sampling for pathogens, parasites and pests should be conducted whilst the candidate species is in quarantine.
- iii. Sampling should be carried out under the supervision of a fish health officer or other competent person appointed or approved by the importing authority.



- iv. Sample size should be based on achieving a 95% probability of detecting an agent in a particular batch or lot, with an assumed incidence of the carrier state.
- v. Additional testing should be undertaken using known susceptible test species cohabited with the introduced candidate species, or other native species as appropriate.
- vi. Testing should be undertaken in an appropriately equipped laboratory, and should be supervised by a competent person appointed or approved by the importing authority, who is familiar with the nature of pathogens and parasites being sought.
- vii. Testing for disease should consist of examining sub-samples of the population for specific pathogens or parasites as determined for the candidate genus or species. In addition to specifying freedom from known pathogens of quarantine concern, criteria should be established which will render the candidate species unsuitable for export or release from quarantine, even in the absence of definitive identification of an infectious agent: Such criteria should include
  - a. The occurrence of gross, histological or ultrastructural tissue changes of a proliferative, necrotic, inflammatory or degenerative nature associated with, typical of, or consistent with viral, bacterial, fungal, protozoan or metazoan agents, including the occurrence of viral-induced inclusion bodies, and including agents which may not yet be known or described.
  - b. The occurrence of viral, bacterial, fungal, protozoan or metazoan agents known to be pathogenic for other classes of marine aquatic animals.
  - c. The occurrence of unexplained lesions, tissue changes or mortalities.

The presence of commensal or symbiotic organisms including bacteria and protozoa should not be considered a primary reason for preventing transfer of the candidate species (Braley 1992).

- viii. Disease testing may be done by laboratories remote from the quarantine facility provided logistics for dispatch of samples are addressed satisfactorily, ensuring delays are not incurred in receipt of samples which may render examinations futile.
- ix. Any and all individuals which display signs of disease or are otherwise abnormal or die should be subject to a clinical and/or laboratory examination to determine the cause of disease.
- x. A clear understanding of the exact numbers of samples required, their nature, i.e. fresh, on ice, fixed, and the times of collection, and the collecting and examining personnel.
- iv. Close collaboration between the collecting staff and the examining staff is necessary.
- v. Considerations as for pre-transfer testing apply.

### **Treatments and Medications**

The collection and holding of stock from non-captive sources will often result in fulminant outbreaks of external protozoan or metazoan parasites. This should be prevented by an initial prophylactic treatment of broodstock on entering the quarantine facility.

- i. The subsequent occurrence of disease or pests in the candidate species while in quarantine should not necessarily terminate the programme. If the causative agent is diagnosed, and is amenable to treatment, such treatment should proceed, and if successful, and if acceptable to the importing authority, transfer procedures should continue.

- ii. If the outbreaks of disease cannot be controlled, or are due to specific pathogens of quarantine concern, the diseased stocks should be destroyed and disposed of in an approved manner.
- iii. If specified pathogens of concern are identified during quarantine, which are not amenable to treatment, stocks should be destroyed and disposed of in an approved manner. In this case, full and thorough disinfection and sterilisation procedures should be initiated to remove all infectious material from the facility.
- iv. Prophylactic treatment of broodstock or stocks introduced into the quarantine facility should be undertaken to eliminate external protozoa and metazoa.
- v. The imported species, where feasible, should be treated on arrival in a manner to disinfect and remove potentially infectious surface or systemic micro-organisms.

#### **Health Certification**

- i. A permit or certificate from a responsible authority in the country of origin verifying the origin of the stock, the stage to be exported, the quarantine and disease history, and assurance that all specified tests, procedures and sampling have been conducted in accordance with the requirements of the importing country should ultimately be forwarded to the importing authority prior to issuance of a Permit to import.
- ii. Certification that inspections, testing procedures and other requirements must be carried out by qualified personnel duly authorised by the exporting and importing countries. For this purpose, individual, with appropriate training and experience should be nominated as accredited signatories for the quarantine programme.

#### **General principles: Post-Transfer Considerations**

##### *Quarantine Procedures*

Inspection and authorisation of each consignment on arrival at point of entry by a Quarantine Officer:

- i. Point-of-entry inspection should be conducted to ensure compliance with specified conditions, numbers, species and presence of extraneous materials or substances.
- ii. The inspecting authority at point of entry should have fully completed permit-to-import forms, together with appropriate documentation from exporting country.
- iii. Following arrival, depending on the specific requirements for the species and the conditions stipulated for entry, release under the direction of the importing authority of imported animals should be to:
  - a. A quarantine facility for further inspections and disease testing
  - b. Directly for culture or release to the environment according to the importing conditions.
- iv. Close collaboration and coordination between quarantine/importing authorities and the proponent/s of the transfer is essential to ensure the efficient quarantine clearance and entry into the holding facility with minimal delays. Individuals from the importing authority and the proponents' organisations should be nominated for this purpose.
- v. Release from the quarantine facility should be done following thorough certification of freedom from disease and authorisation by the importing authority.

### *Quarantine Facilities and Management*

Post-transfer quarantine is necessary as a safeguard against inadvertent transfers of pathogens and parasites in candidate species subject to intensive quarantine and health certification procedures pre-transfer; is essential for candidate species not subject to intensive quarantine and disease testing.

Quarantine facilities must provide for the microbiologically secure housing of aquatic animals, with appropriate means of sterilisation and disinfection of all wastes and effluent to prevent escape of potential pathogens into the environment of the importing country.

- i. All effluent from quarantine establishments receiving and holding transferred species must be sterilised or otherwise treated and disposed of in an approved manner which will ensure the inactivation of all infectious micro-organisms present. This may be by chemical disinfection, discharge into a sump or desiccation in porous ground remote from the aquatic environment.
  - iii. Following arrival and point-of-entry quarantine inspection, transport water, packing materials, containers and associated shipping items must be sterilised and disposed of in an approved manner which will ensure inactivation of any infectious agents present.
  - iv. Acclimation of transferred eggs, larvae, adults to environmental conditions following arrival should be done in a manner which minimise mixing of transport water and final holding waters.
  - v. Incoming water should be sterilised or disinfected. This may be achieved in several ways:
    - a. Use of spring, ground, artesian or well waters which contain no flora or fauna prior to intake,
    - b. Filtration to 1 micron, and/or
    - c. Ultraviolet sterilisation, and/or
    - d. Ozone sterilisation.
- or
- vi. Water used shall be recirculated, filtered biologically and physically and UV sterilised, with water for exchanges treated in an appropriate manner on discharge.
  - vii. Facilities must be approved and regularly inspected by a competent government authority to ensure effectiveness and compliance with specified requirements.
  - viii. Water quality in the facility should be monitored at all times, or at regular intervals, to ensure that mortalities in quarantine are not due to adverse environmental conditions, but rather to disease agents. Records should include temperature, ph, nitrite, ammonia, nitrate, oxygen at a minimum. Such monitoring should apply to recirculated water, as well as water taken from the external environment.
  - ix. All mortalities and cases of clinical disease should be carefully recorded and investigated. All such cases should be reported immediately to the regulatory authority, and if appropriate, be referred for detailed laboratory examination. The cause of all mortalities should be ascertained and reported in writing.
  - x. Full accountability of all animals must be maintained, with written records or losses, means of disposal and reasons for losses.
  - xi. Daily records of all operating procedures and records for each batch of animals must be accurately maintained and made available for inspection.

- xii. Transferred species within the facility must be maintained on a batch-by-batch basis, with housing in individual tanks or compartments, and precautions taken to exclude transfer of water, stock or pathogens between batches. For this purpose, separate equipment must be available for each batch or group; it should not be transferred between groups.
- xii. No equipment should enter or leave the facility without sterilisation or disinfection.
- xiii. To ensure compliance with operational requirements, access to the facility should be restricted to trained, authorised staff who should be under the supervision of appropriately qualified individuals.
- xiv. Entry and exit for personnel operating the facility should be through an appropriate disinfection station, with footbaths, showers and changes of clothing as a minimum requirement.
- xv. Personnel operating the quarantine facility should not visit other aquaculture establishments within three days of being on the facility.
- xvi. Disposal of all solid wastes must be done in approved manner so as to ensure that infectious agents do not escape from the facility.
- xvii. Adjacent, physically-isolated laboratory facilities should be present for inspection and sample preparation. The facility should be readily disinfected and all wastes, effluent and packaging processed as above.
- xviii. Unauthorised persons should not be permitted on the facility at any time.
- xix. All other animals should be excluded from the quarantine facility.



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