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REPORT OF THE

WORKING GROUP ON INTRODUCTIONS AND TRANSFERS OF MARINE ORGANISMS

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1 OPENING OF THE MEETING AND INTRODUCTION

The 1997 meeting of the ICES Working Group on Introductions and Transfers of Marine Organisms (WGITMO) was held at IFREMER-URAPC Aquaculture Research Laboratory, La Tremblade, France, from April 22 to April 25, 1997. The objectives of the 1997 meeting were reviewed; the agenda for the meeting was considered and approved (Annex 1). At this meeting, there were 13 participants representing 8 Member Countries. In addition 17 invited guests were present (Annex 2).

Many members of WGITMO had attended the previous day's separate meeting of the joint ICES/IOC/IMO Study Group on Ballast Water and Sediments (SGBWS), and brief summary and review remarks were made. In particular, it was noted that WGITMO supports the recommendation of SGBWS that SGBWS convene for a second year to discuss research and management programs on ballast water and sediments and other ship-mediated vectors with a view toward increased international cooperation and coordination.

2 TERMS OF REFERENCE

The terms of reference for the 1997 meeting of the WGITMO (ICES C.Res. 1996/2:15:7) were to:

1) continue the assessment of potential marine biocontrol activities and the risks thereof, through the invitation to biocontrol researchers to the 1997 WGITMO meeting, e.g., researchers working on the control of the invasive seaweed *Caulerpa taxifolia* or the invasive comb jellyfish *Mnemiopsis leidyi,* with a goal of formulating a brief review of the efficacy of various other measures (mechanical, chemical, or otherwise) that have been employed to control exotic species invasions;

2) continue to assess the disease and ecological implications arising from the introduction and transfer of aquarium (ornamental) marine and estuarine species into ICES Member Countries;

3) prepare and undertake risk assessment retrospectives for selected case histories of introduced marine plants, invertebrates, and fish in ICES Member Countries, in order to provide a stronger basis for future considerations of newly proposed introductions and transfers;

4) assemble materials for a Directory of Vectors Involved in the Introduction and Transfer of Marine and Estuarine Organisms, to be published in the ICES Cooperative Research Report series;

5) report on the current status of fish, shellfish, algal, and other introductions in and between ICES Member Countries, through submission of annual National Reports, and in particular through continued overviews of:

6) the status of the culture of the marine seaweeds *Porphyra yezoensis* on the Atlantic coast of North America and of *Undaria pinnatifida* on the Atlantic coast of Europe, and its subsequent dispersal and establishment in other ICES Member Countries,

7) the potential and risk of dispersal of the introduced Mediterranean alga *Caulerpa taxifolia* into Atlantic waters,

8) the increasing expansion and dominance of the American marine worm *Marenzelleria viridis*,

9) the increasing activities with GMOs in ICES Member Countries, and

10) the continued development and coordination of cooperative databases on introductions and transfers of marine and brackish water organisms.

11) assess the risk of disease transfer via movements of stocks which are tolerant to the agents responsible for significant diseases.

The Working Group will report to ACME before its June 1997 meeting.

Representatives of IMO, IOC, ICS, OIE, and EIFAC will be invited, at their own expense, to attend specific sessions pertinent to their interests.

3 UPDATE ON NEMO ACTIVITIES

The work of the Baltic Marine Biologists (BMB) Working Group: Nonindigenous Marine and Estuarine Organisms (NEMO) was considered.

Dr Sergej Olenin (Centre for System Analysis, Klaipeda University, Klaipeda, Lithuania) reports as follows:

Through the Nordic Council of Ministers' Office in Vilnius (and with support from Dr E. Leppäkoski of Finland) a small amount of funds was received for preparatory work to organise the next BMB WG NEMO meeting. It is planned for Klaipeda in the beginning of 1998. Prior to that an Internet home page on the Baltic NEMOs will be created, and as many entries collected as possible. A network of Baltic scientists working on NEMO is organised and functioning (although only in a limited fashion, as to date there is essentially no support for this activity). Nevertheless, interest in the problem is growing. In the near future, for instance, the Lithuanian Parliament will discuss a "Marine Protection Law" wherein ballast water issues are included.

Recently S. Olenin was in Kaliningrad at the Fishery Research Institute and at the local Technical University for Fishery and Maritime Industry. He met with specialists willing to cooperate in research studies. New contacts also appeared (or old ones have been renewed) in St. Petersburg, Moscow, and at some research institutes and universities in the River Volga region (such as Yaroslavl and Kazan). These activities are being undertaken because it is clear that Eastern Europe is a very interesting and a very important area from an invasive ecology point of view for the following reasons (and all of which suggest that the need for such a research network is of great importance):

1) it is an area of origin of many alien species which have invaded the New World and other distant regions;
2) it is a transitional area from Ponto-Caspian and Ural/Sabiria/Baikal regions to the North-Western Europe and further, overseas;
3) it is an area where many "experiments with nature" have been performed in the last several decades. Until recently, voluminous data on these introductions have been stored and are unpublished or, being printed in local languages, are unreachable (unreadable) for the greater international scientific community and environmental managers.

4 STATUS OF RECOMMENDATIONS FROM 1996 MEETING

The ICES Advisory Committee on the Marine Environment (ACME) deliberations on the WGITMO 1996 Gdynia Report were read and noted.

In addition to C.Res. 1996/2:15:7 on the Terms of Reference for the 1997 meeting in La Tremblade (see Section 2, above) the following C.Res. 1996/2:15:9 was also made:

That: A Study Group on Marine Biocontrol of Invasive species (SGMBIS) will be established under the chairmanship of Dr J. Carlton (USA) and will work by correspondence to review information relevant to the potential biological control of marine invasive species, taking into account recent interest in this issue in the Black Sea, France, Australia, and elsewhere. Emphasis should be placed initially upon the potential control of the invasive seaweed *Caulerpa taxifolia* in the Mediterranean Sea by the proposed introduction of exotic herbivorous seaslugs, because of the potential for this alga (and its herbivores) to spread into ICES regions. The Study Group should include the following members: Dr M.I. Campbell (Canada), Dr H. Grizel (France), Dr M. Héral (France), Prof. E. Leppäkoski (Finland), Dr D. Minchin (Ireland), Dr S.D. Utting (UK), Prof. I. Wallentinus (Sweden), Dr P. Wells (Canada), Dr R. Thresher (Australia), and Dr C. Maillard (France) (Dr Maillard will provide oceanographic information). The Study Group will report to the meeting of WGITMO in April 1997.

5 ICES CODE OF PRACTICE, 1994

In addition to the English/French version of the ICES Code of Practice on the "Introductions and Transfers of Marine Organisms 1994", the Code has been translated into German, Swedish and Finnish. The WGITMO decided to explore the possibility of translating the Code into four other languages, namely Norwegian, Spanish, Portuguese and Welsh.

It was further noted that it might be possible to put the "Code of Practice" on the ICES web site, or at least a reference to it with information on how to get a copy of the Code.

The Code had also been included in other publications:

Minchin, D. 1996. Management of the introduction and transfer of marine molluscs. aquatic conservation. Marine and Freshwater Ecosystems, 6: 1–16.

Minchin, D. 1996. The impact of aquatic introductions on native populations, pp. 20–30. *In* Council of Europe 'Convention on the Conservation of European Wildlife and Natural Habitats' Colloquium on Conservation, Management and Restoration of Habitats for Invertebrates: Enhancing Biological Diversity, Killarney, Ireland 26–29 May 1996.

WGITMO noted that the 1994 version of the Code of Practice was no longer the ICES/EIFAC Code (the most recent version of the EIFAC Code was published in 1989) and that ICES WGITMO should aim once again to harmonise an ICES/EIFAC version of the Code.

6 STATUS OF NEW COOPERATIVE RESEARCH REPORTS

The manuscripts for three Cooperative Research Reports, namely, 'Summary of Introductions in ICES Member Countries as of 1990', 'Guidebook for Code of Practice' and 'Aalborg 1995 Ballast Water Symposium', have been or are about to be submitted to ICES for publication.

7 OVERVIEW OF NEW EUROPEAN INITIATIVES

7.1 Intermediate North Sea Ministerial Meeting on the Integration of Fisheries and Environmental Issues

The Assessment report prepared for the North Sea Intermediate Ministerial meeting on the integration of fisheries and the environment contained details of the impact from introductions of non-indigenous species. It considered them in relation to different vectors of introduction, including ballast water, aquaculture and stock enhancement (leading to the possibility of genetic degradation). The work of ICES, EIFAC and IMO was recognised. The Basis report contained details of introductions, threats to the environment, measures taken and planned by each of the North Sea States. The Statement of Conclusions, as agreed by Ministers, agreed that strategies should be adopted to apply a precautionary approach to all human activities that involve non-indigenous species and GMOs.

7.2 EC Regulation No. 338/97

The new EC Regulation on the protection of species and wild fauna and flora by regulating trade therein—CITES (The Convention on Trade in Endangered Species)—338/97, contains specific reference to the control of alien species. Article 3(2)(d) states that Annex B shall contain 'species in relation to which it has been established that the introduction of live specimens into the natural habitat of the Community would constitute an ecological threat to wild species of fauna and flora indigenous to the Community'. Such species will therefore require import and export permits (Article 4 (2)(a) and impose restrictions on countries of origin (Article 4(6)(d). To date the only species added to Annex B specifically for this purpose are the American bullfrog and the red-eared terrapin, both traded by the pet shop trade with invasive capacities (seen in the UK).

Independently, IUCN has prepared a position statement on Trade in Alien Species for presentation to the CITES Conference of the Parties in June 1997, supporting a proposal by the USA and New Zealand encouraging CITES to: 'Recognise that non-indigenous species can pose significant threats to biodiversity...; recognise that CITES may play a significant role in this issue; and other recommendations .... including implementation of IUCN's Invasive Species Specialist Group guidelines for the prevention of biodiversity loss due to biological invasion.

7.3 OSPAR Convention

The following report was prepared by K. Jansson (Sweden):

Within the OSPAR Convention (Oslo and Paris Conventions for the Prevention of Marine Pollution) alien species are on the agenda of the Working Group on Impacts on the Marine Environment (IMPACT). At IMPACT 1996, Sweden presented an overview of national activities concerning alien species in the Convention area (i.e., the Northeast Atlantic). The report was based on a questionnaire to Contracting Parties seeking national information on alien species,

including, inter alia, information on relevant research activities, strategies for the development of monitoring programmes and sampling/measurement techniques. Taking into account that many Contracting Parties agreed on the importance of continuing work on this topic, IMPACT 1996 made a number of proposals concerning alien species to its parent committee ASMO (Environmental Assessment and Monitoring Committee). At its meeting in April 1997, ASMO, i.a., decided that:

* the report on alien species should be updated and used as input to the QSR 2000 (Quality Status Report of the Convention Area in the year 2000),

* monitoring of alien species will be included in the Commission's Joint Assessment and Monitoring Programme (JAMP), and that,

* ICES will be asked to consider a reporting format for alien species. As a first step, identification of the status of species (as, e.g., native, alien or cryptogenic; incidental or established) could be included as a parameter in existing programmes (such as monitoring of phytoplankton and benthos) and taxonomic lists.

ASMO also noted that, in relation to measures concerning intraregional dispersal of alien species via shipping, the ICES observer at IMPACT had suggested that this issue could be taken into consideration by the Joint ICES/IOC/IMO Study Group on Ballast Water and Sediments and/or other appropriate bodies. The suggestion by IMPACT that the IMO Guidelines be adopted as an OSPAR Recommendation was however not deemed appropriate by ASMO.

8 NATIONAL REPORTS

National reports were received from Canada, Finland (by mail), France, Germany, Ireland, Norway, Poland (by mail), Sweden, UK (England and Wales) and USA. National Reports (Annex 3) contain details of new laws and regulations, deliberate releases, accidental introductions and transfers, live imports, live exports, planned introductions, and meetings. References cited in the National Reports, and elsewhere in the report, are in the Bibliography (Annex 6).

8.1 CANADA

* In 1996 the Federal Department of Fisheries and Oceans authorised the importation of a maximum of 100 *Porphyra yezoensis* from Maine to a private aquaculture site in Grand Manan, New Brunswick.

* Two major escapes of triploid rainbow trout (4,500 and 20,000 fish) occurred from Bay D'Espoir, Newfoundland. In the same area, escapes of rainbow trout and Atlantic salmon from cages may have occurred associated with 'super-chilling' in cages.

* There was continued spread of the zebra mussel (*Dreissena polymorpha*), the oriental mystery snail (*Cipangopaludina chinensis malleata*) and the spiny water flea (*Bythotrephes cederstroemi*) and the amphipod *Echinogammarus ischnus* in inland lakes.

* Eight vials of transgenic Atlantic salmon milt were transferred from New Brunswick to a transgenic research facility on Prince Edward Island.

* Permission was given to the New Brunswick aquaculture industry to import eggs of a European strain (Mowi strain, landcatch strain) of Atlantic salmon from Maine, USA. It is the intention that fish will be held in land-based facilities to reduce the risk of escape.

8.2 FINLAND

* 100,000 elvers were imported from England via Swedish quarantine facilities and released into inland waters.

* Salmonid eggs and juveniles and newly-hatched and 1-2 year old whitefish (*Coregonus lavaretus*) were released into the Baltic for fisheries management projects.

8.3 FRANCE

* A presentation on the 'Introduction of the Asian shore crab *Hemigrapsus penicillatus*' was given by Dr Pierre Noel, as follows:

A crab previously unknown in the European fauna was discovered in La Rochelle (France) in 1994. This mud crab was identified as *Hemigrapsus penicillatus*, a Northwest Pacific Ocean species. It appears to be spreading rapidly. It has now (early 1997) been detected from Laredo, Spain (43°25'N, 03°20'W) to Fromentine, France (46°53'N, 02°09'W) in sheltered areas, under stones in the mid-littoral zone. It is locally abundant, with a density of up to 10 to 20 individuals per square metre, particularly in estuaries and ports where it tolerates slimy -- muddy zones. In its native area its distribution involves cold (Northern Japan, Korea) to tropical (Taiwan, Hong Kong) habitats. Being tolerant to both high and low temperatures (even at the freezing point), it is likely that it could colonise most of the European and North African coasts within a short period of time.

* In August 1996, IFREMER, La Tremblade was named as the European Reference Laboratory for molluscan bivalve diseases (under EU Directive 95/70/CE).

* Natural populations of *Undaria pinnatifida* have decreased, especially in northern Brittany. Limited cultivation of this alga occurs in southwest France.

8.4 GERMANY

* No new non-indigenous species were recorded in marine or brackish waters.

* The importation of juvenile sturgeon from Russia for commercial purposes was continued.

8.5 IRELAND

* The exotic copepod *Mytilicola orientalis* has become established in Dungarven Bay and continues to be introduced in importations of oyster spat.

8.6 NORWAY

* The abundance of the Red King crab (*Paralithodes camtschatika*) in the Barents Sea, and the northern Norwegian/Russian coastal areas appears to have doubled and the size of mature crabs is larger than in their native habitat.

* In Russia there is interest in introducing the blue crab (*Paralithodes platypus*) to the Barents Sea.

* Dense patches of the brown seaweed *Sargassum muticum* have been found in shallow bays in Rogaland, western Norway indicating an extension of this alga northward.

8.7 POLAND

* The round goby (*Neogobius melanostomus*) was not reported from any new localities but surveys in 1996 were not extensive.

8.8 SWEDEN

* Further spread of the polychaete worm *Marenzelleria viridis* has occurred and it has been found at depths of 60 m in the Baltic Sea.

* Since 1993, the brown alga *Sargassum muticum* has extended its range 100 km southwards on the Swedish west coast.

* In 1996, the dinoflagellate *Alexandrium minutum* was recorded for the first time in the eastern Skagerrak. It was abundant at the end of June. Cysts of the dinoflagellate *Gymnodinium* cf. *G. catenatum* were abundant in sediment samples taken along the coast of Bohuslän.

8.9 UNITED KINGDOM: ENGLAND AND WALES

* Beds of naturally settled Manila clams *Venerupis philippinarum* were found in Poole Harbour on the south coast of England. The clams were outside the designated areas for aquaculture. The shell length indicated that clams were from a successful spawning in 1994.

* An attempt was made to physically remove attached plants of the brown alga *Sargassum muticum* from Strangford Lough in the summer 1996, but it is still too early to determine how successful this was.

* The brown kelp seaweed *Undaria pinnatifida* has been found in two new sites on the south coast of England.

8.1.10 UNITED STATES OF AMERICA

* The National Invasive Species Act of 1996 (NISA 1996), which updates the 1990 Nonindigenous Aquatic Nuisance Prevention and Control Act and upgrades the level of ballast water management, was passed by the US Congress in October 1996.

* The European oyster *Ostrea edulis* continues to spread its range on the Atlantic coast.

* Established and reproducing populations of the red alga *Grateloupia doryphora* (native to the Pacific) were found for the first time in Narragansett Bay, Rhode Island.

9 STATUS OF *PORPHYRA YEZOENSIS* IN THE GULF OF MAINE (USA)

Dr Ike Levine reviewed the progress of the regular monitoring programme for natural recruitment of the Japanese red seaweed *P. yezoensis* in the vicinity of the culture areas and the future development of the culture programme (Annex 4). Electrophoretic techniques have been used to distinguish between the introduced *P. yezoensis* and the native *Porphyra* species. From 18 plants that were collected from outside the culture area, only 3 were *P. yezoensis* (see Figure 4 of the report, attached as Annex 4). Although monospores can recruit, *P. yezoensis* does not outcompete the native species and monospores do not survive the winter. Therefore, the risk of natural spread of this alga from the designated culture sites would appear to be low.

Future developments include genetic modification of native species of *Porphyra* although the WGITMO was assured that all culture of any genetically modified strains would be in contained, land-based culture facilities.

10 AUSTRALIA'S NATIONAL CENTRE FOR RESEARCH ON INTRODUCED MARINE PESTS (CRIMP)

Dr Ron Thresher described the reasons for the inception of CRIMP, its objectives, organisation, areas of interest and responsibilities. Since it was set up three years ago, CRIMP has expanded rapidly (see Annex 5).

11 MARINE BIOCONTROL SESSION

This session included a number of presentations on introduced species that have reached the status of being pests as well as much discussion on methods that have been proposed for biocontrol of such pest species and the associated risks entailed. Experience from the Australian research programmes on marine biocontrol, probably the most advanced research programme in the world, was particularly relevant during this session.

11.1 REVIEW AND DISCUSSION OF REPORT OF THE "STUDY GROUP ON MARINE BIOCONTROL AND INVASIVE SPECIES"

The WGITMO reviewed the report of the 'Study Group on Marine Biocontrol and Invasive Species' and after discussion a number of amendments and modifications were suggested which will be considered by the Study Group for inclusion in the final report. (Note: The full deliberations of the Study Group and its report will be presented as a separate item and are not included in this WGITMO report.)

The WGITMO supported the recommendation of the ICES Study Group on Marine Biocontrol of Invasive Species (SGMBIS) that a permanent international "Working Group on The Control of Marine Pests" be formed, as initially proposed by GESAMP, which would advise and recommend strategies on the prevention and post-introduction research and control of introduced species. This Group could be formed by a possible consortium of GESAMP, ICES, and other international agencies.

11.2 MARINE BIOCONTROL IN AUSTRALIA

Dr Louise Goggin gave the following presentation to the WGITMO on the status of several marine introductions to Australia that are now considered as pest species and for which marine biocontrol research programmes have been or are likely to be proposed:

Asian Brown Kelp *Undaria pinnatifida*

A desktop study indicated that there are few natural enemies of this Asian seaweed. Physical removal of *U. pinnatifida* from a marine reserve area was undertaken. The success of this attempt will be determined in the next two months. A preliminary trial with 9 herbicides showed limited success against the sporophyte. A second more extensive trial has been commissioned to test the effect of other herbicides against the gametophyte.

Mediterranean Polychaete Worm *Sabella spallanzani*

Die-backs of *S. spallanzani* occurred in Port Philip Bay, Victoria and Western Australia in late 1996. No causative agent was found. A desktop study found few natural enemies which are likely to be useful for biological control.

North Pacific Seastar *Asterias amurensis*

A protozoan, the scuticociliate *Orchitophrya* sp., is being investigated as a biocontrol agent. The species identity of the isolate from *Asterias amurensis* from Japan has not been determined but morphology indicates that it differs from isolates from Belgium from other hosts. Molecular data link the ciliate from Japan to that from the seastar *Pisaster ochraceus* from Vancouver. Further investigation of the host specificity and life cycle of the ciliate is ongoing. Few other natural enemies have been identified. Physical removal may be necessary to restrict the seastar to the Derwent estuary.

European Shore Crab *Carcinus maenas*

The parasitic barnacle *Sacculina carcini* is under investigation as a possible control agent for *C. maenas* in Australia. A desktop study indicated that other natural enemies such as ciliates, parasitic dinoflagellates (*Hematodinium* spp.) and viruses may be worthwhile investigating. Physical removal will be attempted at Falmouth in Tasmania.

11.3 THE INFLUENCE OF THE PARASITIC CIRRIPEDE *SACCULINA CARCINI* ON ITS BRACHYURAN HOST *CARCINUS MAENAS* WITHIN ITS HOME RANGE

Dr Dan Minchin gave a summary of his studies on the morphological externae of *Sacculina carcini* in Irish waters based on observations over a twenty-year period. The main influence would appear to be on the crab's predation capabilities. Reductions in the mean crab size arising from infection together with reduced chelae power indicate an approximate reduction in the predation of on-bottom culture of oysters, mussels and scallops of 5-25%. Infested crabs behave differently to normal crabs, proportionally more were captured in traps at night, at greater depths and more were captured by dredge than by trap. Parasite prevalence appeared to be unrelated to numbers captured in traps and ranged in different populations from 0.3% to 64.4%. Within one sea inlet the prevalence on infested crabs over <10 km ranged from 0% to 21%, with the lowest levels in shallow brackish estuarine areas and the highest in the bay centre in deeper water. It is likely that hydrographic features are important for levels of infestation and may explain why in some inlets levels are consistently low. *Sacculina* sp. are associated with *Cancer, Atelycyclus* and other portunid crabs in Irish waters.

In Irish waters *S. carcini* does not infest the majority of the *C. maenas* in a population and so crab populations remain viable, although there is some reduction in larval output because infested females do not produce eggs. The taxonomy of *Sacculina* is not well understood because of poor morphological features, principally of the externa which could be modified by crab abdomen shape and size. In the absence of genetic studies it is unclear whether *S. carcini* has a broad host range.

The introduction of *S. carcini* to Australian waters is unlikely to contain range expansions of *Carcinus maenas* or significantly control their numbers, and it could perhaps be accommodated in some Australian portunid species. However, infection impairs *C. maenas* feeding capabilities and is likely to reduce the reproductive output to different levels in each crab population affected. Overall biological control using *S. carcini* still has potential risks and few benefits.

11.4 MARINE BIOCONTROL IN THE BLACK SEA

Controlling the comb jellyfish (ctenophore) *Mnemiopsis leidyi* in the Black Sea

It was noted that the Group of Experts on the Scientific Aspects of Marine Environmental Protection (GESAMP) of the UN, UNEP, FAO, UNESCO, WHO, WMO, IMO and IAEA had produced in January 1997 its report on the "Opportunistic settlers and the problem of the ctenophore *Mnemiopsis leidyi* invasion in the Black Sea" (Reports and Studies no. 58).

A summary of this work is as follows:

The carnivorous comb jellyfish *Mnemiopsis leidyi* was introduced from North America to the Black Sea in the ballast water of ocean-going ships. The first specimens were found in 1982, and it was found again in 1986 and continuously thereafter. The invasion of *Mnemiopsis* is considered to be a major element in the profound decline—starting in 1989—of the endemic anchovy and other fisheries in the Black and Azov Seas. As a result, it was concluded that "A reduction in *Mnemiopsis leidyi* population levels is essential ... [and that there is a] .... need for immediate action..." (GESAMP 1997, p. 46). GESAMP held two meetings in 1994 and 1995 at UNEP in Geneva, Switzerland, to consider the economic and ecological impact of the invasion and control strategies.

The analysis of biocontrol options at these meetings yielded the following results:

(1) There are no known parasites or diseases of *Mnemiopsis* that appear to reduce its population sizes. One abundant parasite, the larval stage of a sea anemone, apparently causes little damage to the combjelly, while in turn these sea anemone larvae occur in such blooms as to create "intense reactions in swimmers." This parasitic sea anemone was not introduced with *Mnemiopsis* into the Black Sea.

(2) There are no known species-specific predators on *Mnemiopsis*. However, the large jellyfish *Chrysaora quinquecirrha* feeds voraciously on *Mnemiopsis*. But it also feeds on a wide variety of zooplankton (including larval fishes), and is itself a frequent serious menace on swimming beaches when occurring in large numbers. *Chrysaora* does not occur in the Black Sea.

(3) The comb jelly *Beroe* eats gelatinous zooplankton, including salps (pelagic tunicates [urochordates]) and ctenophores. Native *Beroe* do not occur in the Black Sea. *Beroe* do occur in the Mediterranean Sea, to where *Mnemiopsis* has already spread.

(4) Many North American fish feed on ctenophores (which do not have stinging cells, unlike true jellyfish). The butterfish *Peprilus* feeds extensively on gelatinous zooplankton. One species, *Peprilus triacanthus*, has been suggested as the major controller of *Mnemiopsis* populations in Narragansett Bay, Rhode Island, USA (on the south side of Cape Cod). GESAMP (1997) noted that,

"For any introduction of fish predators, the following prerequisites are generally agreed on:

i) the fish must be commercially exploitable, with both domestic and export potential,

ii) the fish must be disease free and quarantined,

iii) it must feed heavily on *Mnemiopsis leidyi*,

iv) existing facilities and skills for its culture must be available in the Black Sea region, and

v) before any release is undertaken, there must be basic knowledge on its feeding specificity, environmental tolerance, ecological impact and suitability for maintenance, rearing and transport in the Black Sea region."

GESAMP (1997) proposed the following strategies:

1) Native fish, such as the horse mackerel *Trachurus mediterraneus*, should be managed so as to enhance their populations. Enhancement of these stocks would "most likely" have an impact on *Mnemiopsis* , since the mackerel feed on ctenophores. Enhancement could be achieved by a decrease in fishing effort and a reduction in pollution. GESAMP (1997) concluded that "It is strongly recommended that this course of action be studied".

2) Non-native fish, such as the cod *Gadus*, the butterfish *Peprilus*, and the salmon *Oncorhynchus*, could be introduced (under a plan that includes the above-named prerequisites). GESAMP (1997) concluded, "The Group does not regard these as necessarily the best candidates, and they are used only as examples." A considerable suite of complexities and challenges would attend the introduction of any of these three fish examples. For example,

   a) Cod and salmon are omnivorous, with the latter eating "small pelagic fish" such as anchovy; the report added, that, however, "[the salmon is] more valuable than anchovy".

   b) Cod cannot live in temperatures above 14 °C, which means that it would be unable to feed on the bulk of the *Mnemiopsis* population residing in summer in the warmer surface layers.

   c) Butterfish reproduction and biology are poorly known, its eggs and larvae may be eaten by *Mnemiopsis*, and the butterfish may enter the Mediterranean.

3) A specific comb jelly predator, the American comb jellyfish *Beroe* sp., could be introduced. However, *Beroe* would probably also eat the native Black Sea comb jellyfish *Pleurobrachia* and other gelatinous zooplankton. *Beroe* does not enter salinities as low as *Mnemiopsis* (and therefore it is not likely to control *Mnemiopsis* in regions of lower salinity).

In conclusion, GESAMP (1997) noted that "The Group is well aware of the potential problems that may be created by the introduction of exotic predators on *Mnemiopsis leidyi*."

Finally, GESAMP also stated that the formation of a "permanent Working Group on the Control of Marine Pest Invasions is recommended through a global mechanism such as GESAMP. This group would advise and recommend strategies on the prevention and post-introduction research and control of introduced species, and would communicate with other international agencies."

11.5 MARINE BIOCONTROL IN FRANCE

Controlling the green algae *Caulerpa taxifolia* in the Mediterranean Sea

A presentation on the potential biocontrol of the invasive green seaweed *Caulerpa taxifolia* in the northwest Mediterranean Sea was made by Dr Alexandre Meinesz and his student Thierry Thibaut. A summary is as follows:

The giant coenocytic chlorophyte (green alga) *Caulerpa taxifolia* (Vahl) C. Agardh is a tropical alga that was introduced into the Mediterranean Sea in 1984. Since then, it has spread vegetatively and by the end of 1996 it had affected 3000 hectares. The introduced strain is very ubiquitous and perennial and out-competes most Mediterranean algae. It also invades the seagrass beds and eliminates a large number of invertebrates. The alga contains toxins (caulerpenyne) and as a result it is not grazed by herbivores.

Since the affected area is so large, it does not allow the use of physical or chemical controls to eradicate the *Caulerpa*. Thus, biological control was considered. Potential control organisms are the specific predators of *Caulerpa*. These are seaslug molluscs (sacoglossans). In the Mediterranean Sea, two indigenous species feed on *C. taxifolia*; these are *Oxynoe olivacea* (Rafinesque, 1814) and *Lobiger serradifalci* (Calcara, 1840). However, there is the drawback that these two species have pelagic larvae in their life cycle, with the result that larvae would be distributed by currents and dense populations of seaslugs would not develop on the target areas, that is, the *Caulerpa* meadows. Intensive juvenile culture is possible, but 500 to 1000 seaslugs per square meter are required to obtain an efficient feeding pressure to control *C. taxifolia*. However, such cultures have a high cost and the release of millions of seaslugs per hectare with a homogeneous distribution is technically very difficult.

For this reason, sacoglossans with a benthic development have been studied for three years. The biology of the Western Atlantic Ocean tropical species *Elysia subornata* (Verrill, 1901) and *Oxynoe azuropunctata* (Jensen, 1980) was studied. These species are specific to the genus *Caulerpa* with which it has a strong coevolution. They do not bite the algae but make a hole in the cell wall and suck out the cytoplasm. They accumulate caulerpenyne toxin in order to defend

themselves against predators. They store chloroplasts in their tissues where the chloroplasts can stay functional for more than a month. Biological characteristics (longevity, reproductive rates, amount of *C. taxifolia* consumed) favour the selection of *E. subornata* as a potential biocontrol agent. A sterile culture of egg-masses from two successive generations allowed Professor Meinesz's laboratory to obtain a culture following the recommendations in the ICES Code of Practice. The laboratory is now waiting for agreement from the French Government for a preliminary experiment in the open sea.

After this presentation, the WGITMO discussed the potential for *Caulerpa taxifolia* to spread (either naturally or by the human-mediated transport mechanisms, e.g. on boats, the aquarium trade) to other ICES Member Countries, in particular to the Mediterranean coast of Spain in the immediate future. Since there had been no National Reports or official national representative from Spain attending the WGITMO meeting since the meeting in Lisbon in 1992 it was decided that the Chairman should write to the Spanish delegate to request information on the current status of *C. taxifolia* in Spanish waters.

It would appear that the strain of *C. taxifolia* that is in the Mediterranean can survive in conditions that it cannot tolerate in its native habitat. It has been found in areas of the Mediterranean where the sea water temperature drops to 10 °C in the winter (the stolon remains alive even at temperatures less than 10 °C) although growth only occurs at 15 °C.

11.6 RISK ASSESSMENT

11.6.1 RISK ASSESSMENT RESEARCH AT CRIMP (AUSTRALIA)

Dr Ron Thresher of the Centre for Research on Introduced Marine Pests (CRIMP) in Hobart, Tasmania, Australia, described the steps that needed to be followed before any proposed biocontrol programme could be implemented in Australia. The following are important:

- safety is paramount: the biocontrol agent should be host-specific,
- consultation with, and approval from community, governmental and environmental agencies is essential at all stages,
- cost-benefit analysis should be carried out, and,
- formal risk assessment protocols should be established.

In Australia, any proposed introduction for biocontrol purposes has to go through a two stage process:

Stage 1—approval to import for testing which takes 3-5 years,

Stage 2—approval to release which takes an additional 3-5 years.

Even before Stage 1 is reached it has to be demonstrated that all other methods of control (physical removal, chemical control, use of native biocontrol species, transgenic control) have been considered. Impact studies must also demonstrate that there will be no negative impacts from the proposed introduction of a marine biocontrol agent.

11.6.2 PREPARATION OF HISTORICAL RISK ASSESSMENT STUDIES

Owing to insufficient time, this item in the Agenda was postponed and has been included in the proposed agenda for the 1998 meeting.

12 RISK OF DISEASE TRANSFER VIA MOVEMENTS OF STOCKS WHICH ARE TOLERANT TO THE AGENTS RESPONSIBLE FOR SIGNIFICANT DISEASES

A request was received from WGPDMO to assess the risk of disease transfer via movements of stocks which are tolerant to the agents responsible for significant diseases (see Section 2, above). The relevant background to this request is as follows, from Section 9 of the 1997 Working Group on Pathology and Diseases of Marine Organisms report:

Progress On Development Of Resistance In Oysters To Commercially Significant Diseases

WGPDMO discussed progress on the development of resistance to commercially significant diseases in American oysters (*Crassostrea virginica*), European oysters (*Ostrea edulis*) and Pacific oysters (*Crassostrea gigas*). It was

concluded that resistance, in the classic sense (see definitions in 1997 WGPDMO Report, Annex 9a), has not been demonstrated for any oyster disease to date, however, there are clear cases of the development of tolerance to significant infectious agents, e.g., *Haplosporidium nelsoni* and *Bonamia ostreae*. Only one case has demonstrated complete disappearance of clinical infections, namely, Malpeque disease of *C. virginica*. Information on oyster tolerance of commercially significant disease agents was investigated to determine which factors may be inhibiting or enhancing the development of tolerance to these agents (1997 WGPDMO Report, Annex 9a).

Conclusions

Naturally developed tolerance has been demonstrated by *C. virginica* to the causative agent of Malpeque disease (unknown etiology), *H. nelsoni* (cause of MSX disease) and, possibly, *Perkinsus marinus* (causative agent of Dermo disease). There is also evidence of natural development of tolerance by *O. edulis* to *B. ostreae*. In the latter case, stocks from the Northeast Pacific showed 26% mortality compared with 99% in native populations. In The Netherlands, outbreaks of bonamiasis in 1988 decimated over 90% of oysters in Lake Grevelingen. Surveys of surviving oysters demonstrated a parallel decline in prevalence of *B. ostreae* infection (WGPDMO Annex 9b). Since oyster populations are beginning to build up again, it is important to determine if the decline in *B. ostreae* infection is due to acquired tolerance by the oyster, or inhibition of parasite proliferation by low host densities.

Selective breeding is commonly used to accelerate production of oysters tolerant to specific infectious pathogens. This method of selection and/or over selection (see definition in WGPDMO Annex 9a) has led to inbreeding in certain stocks. This was particularly notable in *C. virginica* stock selected for tolerance to *H. nelsoni* infection, which showed enhanced susceptibility to *P. marinus*. The pathogen-specific tolerance trait may have been induced by inbreeding or by selection of a defence mechanism unsuited for control of a different pathogenic agent. Regular outbreeding with broodstock that have developed a natural tolerance to one or more infectious agents is, therefore, advisable. Areas within epizootic zones, which are closed to exploitation, allow natural tolerance to build up and can be used for outbreeding, as well as production of seedstock for local use.

Tolerant oysters frequently show no evidence of infection. Thus, they have the potential to be healthy carriers of infectious agents. Subclinical infections have been linked to the spread of *B. ostreae* throughout both coasts of the USA, as well as to the spread of MSX from Chesapeake Bay to Delaware and Maine on the east coast of the USA. Thus, selectively-bred stocks should be restricted to enzootic areas and not transferred to non-endemic areas.

WGPDMO recommends:

a) that ICES Member Countries are encouraged to undertake studies to investigate persistence of *B. ostreae* in low densities of European oysters and refer their results to the WGPDMO for evaluation;

b) that assessment of the risk of disease transfer via movements of stocks which are tolerant to the agents responsible for significant diseases be reviewed by the WGITMO.

Dr Henri Grizel reviewed the issue and made a presentation to WGITMO. WGITMO reached the following conclusions:

1) Tolerance or resistance in oysters can be developed naturally but under specific conditions (e.g., modifications of the environmental conditions, or of the pathogenicity of the pathogen, or of the physiology and/or immunology system of the host that enables it to build up a natural immunity). Before it can be claimed that tolerance or resistance is permanent and transmissible (i.e., from F1 to F2 to F3, etc.) it would be necessary to check this through challenge trials using experimental infections of the pathogen (different origin of host and pathogen must be included). If the tolerance is confirmed, then the second step should be to confirm this new character of tolerance and then check if this character can be transmitted to the progeny. The third step is to verify that the tolerance or the resistance is real (e.g., as a result of strong immunity barriers) and not due to the absence of sensitivity to the pathogen (i.e., the oyster is a vector of the pathogen without any disease symptoms being evident). In the latter case, the pathogen can be carried alive in tolerant individuals and if these individuals are moved to a pathogen-free area, then the pathogen can be spread to this area.

2) Tolerance or resistance can also be acquired through a genetic selection programme. The problem is quite similar to that described above. The main difference is the possibility of characterising in the first instance the genetic profile of the broodstock. If an effective resistant strain (non-receptive and non-sensitive) or only a tolerant strain is obtained, then either should be managed in a similar way to the naturally tolerant individuals (see paragraph 1 above).

WGITMO recognises that tolerant individuals can still be vectors of the pathogens. Therefore, only eyed larvae or the juvenile stages produced in a hatchery from broodstock with a demonstrated tolerance to pathogens (demonstrated in challenge trials) and reared under quarantine conditions should be transferred to pathogen-free areas.

In general, WGITMO continues to note its concern relative to the transfer of fish and shellfish stocks within the EC and from other countries, relative to the potential for the accidental introduction of exotic species, particularly those that would lead to disease and pest problems.

13 DISEASE AND ECOLOGICAL IMPLICATIONS ARISING FROM THE INTRODUCTION AND TRANSFER OF AQUARIUM (ORNAMENTAL) MARINE AND ESTUARINE SPECIES

Information on the status of the movements of ornamental marine and estuarine species for in the aquarium industry trade is included in some of the National Reports (Germany, Sweden and UK (England and Wales); see Annex 3). Further discussion included the following:

As was noted in the 1996 WGITMO report, the WGITMO recognises that trade in ornamental (aquarium) marine and estuarine species is largely unregulated, and there is little information available on the species involved and to where they become distributed. Limited information on national legislation/guidelines/codes of practice on ornamentals was included in the 1996 report.

Dr Clare Eno informed the WGITMO that in the UK the trade association, the Ornamental Fish Industry (OFI), is keen to promote good practice, which it achieves through posters, reports and codes of practice. The OFI keeps its members well informed on legislative developments, both nationally and internationally. Their Chief Executive has recently published a paper on trade in ornamental fish (Davenport, 1996). One of its members, the Tropical Marine Centre, based in Chorleywood, near Heathrow, is the largest importer of marine species in Europe and acts as a distribution centre for much of Europe (see National Report for England and Wales, Annex 3). They maintain very high standards and are very amenable to discussion and would consider very seriously any approach by ICES.

In view of the potential disease and ecological risks involved through the accidental or intentional release of ornamental species from either public or private aquaria (particularly the spread of the serious disease epizootic ulcerative syndrome (EUS) from the Indo-Pacific region), the WGITMO concluded that it would be of value to continue to assess the scope of the problem, how the risks may be reduced, where the likely entry points are, and the possibilities for controlling/preventing such introductions. It was stressed that, in addition to professional dealers, large informal sectors exchange species using aids such as the World Wide Web, catalogues and trade and hobby magazines.

The WGITMO is particularly concerned about the potential introduction of the green alga *C. taxifolia* to other ICES Member Countries. The continuing expanding range of *C. taxifolia* in the Mediterranean Sea and the cold tolerance of this population may enable it to colonise other areas in the North Atlantic Ocean. Establishment on the Atlantic coasts of Europe may be possible by range expansion through the Straits of Gibraltar (such as being transported by shipping activities) or by means of specimens discharged into the sea from aquarium facilities (see Section 11.4, above).

In conclusion, the WGITMO considered the potential for the spread of *C. taxifolia* to other ICES Member Countries to be extremely high, particularly as it is a species that is widely available through the aquarium trade. Information in relation to its potential introduction to other Member Countries should be obtained. It was thus considered that WGITMO should conduct a general survey to determine the extent to which the green alga *Caulerpa taxifolia* is being held in public and research aquaria in ICES Member Countries, under what temperature conditions, and the nature of effluent treatment, and also the extent to which this alga is available for purchase within the private aquarium hobby within ICES Member Countries.

More broadly, it was felt to be important that WGITMO develop a questionnaire and expanded terms of reference for a more general review of the non-native temperate marine and brackish-water organisms being held in public and research aquaria in ICES Member Countries, under what temperature conditions, and the nature of effluent treatment, and also the extent to which non-native temperate animals and plants are available for purchase within the private aquarium hobby within ICES Member Countries.

On a related matter, S. Gollasch (Germany) noted that it might be of value to include, as a new section in future National Reports, information on the importation of non-indigenous species in public aquaria. Typically, of course, such species are imported without plans to reach open waters and have therefore not been included previously in National Reports. However, it is clear from a number of cases that such species can accidentally be released into the wild and cause

negative effects to native species and the environment, as the example of the subtropical seaweed *Caulerpa taxifolia* in the Mediterranean Sea shows. Thus it could be helpful to determine the potential for an accidental release of an exotic species by public aquaria if the diversity of exotic species being held by such institutions were to be known. Thus, a section entitled "Activities in the Aquarium Trade" could be added to the reports. In the long run, it would be helpful to know if a newly discovered exotic species is or was kept in a regional or local public aquarium, in order to determine whether incomplete protection systems exist.

WGITMO noted Dr Gollasch's suggestion, and felt that it would be useful to add such as section to the National Reports.

14 COOPERATIVE INTERNATIONAL DATABASES

In the 1996 WGITMO report of the Gdynia meeting, a number of international databases were listed. The advantages and need for international databases (for the standardisation of methods for inventories and research studies, as an information source for new researchers, and as a link between researchers worldwide) were discussed further at the present meeting. Dr K. Jansson (Sweden) kindly shared with WGITMO her continued compilation of international data sources on biodiversity with a particular focus on invasive species.

The proposal for an 'International Network for Marine Invasion Research' (INFORMIR) was brought to the attention of WGITMO and is outlined below:

14.1 International Network For Marine Invasion Research (INFORMIR)

The following proposal from the USA (submitted by Dr Gregory Ruiz (Smithsonian Environmental Research Center) and Dr James T. Carlton (Williams College -- Mystic Seaport, Connecticut) relative to an "International Network for Marine Invasion Research" was presented by J.T. Carlton and received with interest. The statement is quoted below:

"Rationale for INFORMIR:

"Over the past decade, the extent and impact of non-indigenous species (NIS) invasions have received increasing attention throughout the world. For marine (including estuarine) habitats, this has resulted in a series of parallel studies in many different countries that examine patterns, mechanisms, and consequences of NIS invasions. This convergence in research programs and priorities is most evident in the area of ballast water ecology and management, but it is also occurring in many other areas (e.g., NIS inventories, impact studies, and so forth).

"The information now emerging from these parallel research programs begins to provide exciting comparison across a "global network" of sites. Importantly, it is only through such a comparative approach that we can derive generalities from individual studies, measuring significant patterns of variation among regions and testing predictions about invasion processes on various spatial scales. Development of effective management strategies to reduce the risks and impacts of NIS depends upon understanding both these generalities and sources of variation (that is, those factors that influence the success and impact of invasions).

"The organisation of a coordinated effort is well underway for ballast water research, facilitated strongly by international groups such as ICES and IMO. As a result of frequent meetings about ballast water issues within and among countries, research is increasingly being developed and compared with multiple countries around the world. Although there is still much that could be done to enhance comparative ballast water research through a global network, efforts are already underway to describe and develop standard approaches that promote such comparisons.

"In contrast, coordinated research on patterns and impacts of NIS in marine and estuarine habitats lags far behind. There are few data available to compare directly the abundance, distribution, or ecological effects of NIS among communities within a country or among countries. This results both from the relatively few studies (until recently) in this area and from the lack of standardisation. With the apparent increase of research programs to develop NIS inventories and measure various attributes of invasions around the world, we believe there is a strong benefit to establish an "International Network for Marine Invasion Research" (INFORMIR) that can be used to coordinate comparative and collaborative invasion research in this area especially.

"The proposal is thus to establish the "International Network for Marine Invasion Research" (INFORMIR) to help coordinate and develop standard approaches to measure patterns and impacts of NIS invasions in marine and estuarine habitats. The goal of the network is not to constrain or limit ways in which people study invasion process, but instead to

help promote opportunities for comparative and collaborative research among groups who wish to pursue broad scale comparisons on invasions throughout the world.

"Although its exact nature and function are still evolving, primary "Network" goals include (but are not limited to):

"* Development of standard measures of NIS invasion patterns and impacts that can be included in research programs to allow direct comparisons among studies around the world;

"* Creation of a forum for regular interaction to discuss results and ideas, foster collaboration, as well as develop novel coordinated approaches to extend the scope and interpretation of invasion research;

"* Development of funding strategies to implement coordinated international measures of invasion processes across a global network.

"At this time, we wish to gauge the level of interest among our colleagues to participate in this 'International Network'. We have received strong interest from invasion research programs in Australia, New Zealand, and the Baltic states and will soon attempt to implement some aspects of this program. Any research groups interested in participating and having comments on this proposed Network are invited to contact Dr Ruiz or Dr Carlton."

WGITMO briefly discussed the above proposal and it was felt that there was interest among individual researchers in contributing to this concept. No Working Group-level action was, however, taken at this time.

15 DIRECTORY OF TRANSPORT VECTORS

15.1 Overview of transport vectors

The WGITMO discussed an initial outline submitted by Dr J.T. Carlton of subject areas to be included in the proposed "ICES Handbook of Human-mediated Dispersal Vectors". Once completed, the handbook would be a comprehensive review of transport vectors, covering such vectors as ships, aquaculture and the aquarium industries, scientific research, and recreation. The handbook will be of value both to researchers and to managers and policy makers. The handbook and outline were discussed at some length, and it was concluded that between this and the next WG meeting members would begin to compile detailed information relative to the vector categories in the proposed outline. It was further noted that input and cooperation from interested colleagues in EIFAC could prove must useful, and Dr D. Minchin volunteered to carry this proposal to the appropriate parties at EIFAC.

15.2 Introductions by Ships' Fouling

Dr Dan Minchin (Ireland) presented some specific remarks on the current status of introductions via the vector of fouling assemblages on ships' hulls. Since the general usage of "antifouling paints" (organotin-based antifouling coatings) the biomass of fouling organisms on ships has been significantly reduced. Nevertheless, significant populations of some species can remain and survive on paint coatings that have been exposed for over a year. Alternative non-toxic applications have been and are being tested and it is likely that these will lead to a new generation of coatings for use on some vessel designs.

While much recent attention has been given to ballast water, the presence of fouling organisms and their ecological consequences have been given less modern attention. Ships' fouling may still lead to the introduction of non-native species. Further and remote expansions of species from newly established populations may readily gain access to new localities with the regular, fast and often expanding shipping activities. Attention should be given to fouling organisms and their potential for distribution and expansion, because once ballast water can be adequately treated, ships' fouling may again become the single most important means of species' introduction.

WGITMO discussed Dr Minchin's paper and concluded that ship fouling should be given detailed attention in the proposed "ICES Handbook of Human-Mediated Dispersal Vectors".

16 POSSIBLE THEME SESSION FOR THE YEAR 1999 OR 2000 ICES ANNUAL SCIENCE CONFERENCE

The concept of a possible Theme Session on 'Marine Biological Invasions: Retrospectives for the 20th Century, Prospectives for the 21st Century' to be convened for the 1999 (Stockholm) or 2000 (Belgium) ICES Annual Science Conference was strongly supported by the WGITMO. The purpose of this Theme Session would be to gain an overview of the issues that have engaged ICES Member Countries on the introductions and transfers of marine organisms for the past 20 years—since the first reconvened meeting of the ICES Working Group on Introductions and Transfers of Marine Organisms (1979)—and to gain insights into the challenges that face ICES Member Countries in the opening decades of the 21st century. In addition, speakers should be invited who would address issues concerning the precautionary approach, risk assessment, and the modelling of invasion processes.

17 STATUS OF ACTIVITY WITH GENETICALLY MODIFIED ORGANISMS (GMOS) IN ICES MEMBER COUNTRIES

WGITMO delegates were reminded that National Reports should include current status of any work on GMOs in ICES Member Countries and that GMOs are now specifically covered in the 1994 Code of Practice. An example would be studies on salmon in Canada and the UK (Scotland) that have enhanced rates of growth. Such genetically modified salmon may soon be available for commercial cultivation in Canada (for example, see Entis, 1997), as discussed below.

Dr M. Campbell (Canada) noted that the Canadian draft transgenics/GMO policy mentioned in the 1996 WGITMO report remains in a draft format pending further minor revisions. Canada is using the transgenic research facility in Prince Edward Island as a test case for the policy, that is, how the containment requirements and conditions can be applied and how practical and economical they are in actual use. Relative to the actual facility (Aqua Bounty Farms, a division of A. F. Protein; see Annex 6), they are proceeding with a programme aimed at developing broodstocks of Atlantic salmon, rainbow trout and, if successful, Arctic charr, that will breed "true" with the gene construct being reliably passed to F1s (or further on) which will be available to the aquaculture industry as sterile fish. To date, the only method for sterility discussed is triploidy. The company envisions a further 3-5 years before they will have a marketable transgenic fish (smolts most likely) for the industry.

Dr Inger Wallentinus (Sweden) brought to the attention of the WGITMO research into genetically modified microalgae (at least three of which are marine species) for the biotechnology industry, intended for a range of commercial applications including bioremediation, aquaculture, and the production of fuels and specialty chemicals. Dr Ike Levine had also outlined his work on *Porphyra* species to enhance production traits (Annex 4).

Dr Maurice Héral (France) reported that, under DG VI legislation in the EU, there is a proposal under discussion to prevent research on and the use of transgenic organisms that involve growth-promoting hormones. This action will be relevant to EU countries and is in response to public pressure against GMOs.

18 ACTION LIST

WGITMO identified the following as its current list of Action Points. These are also presented in Annex 7 and are in addition to those elements proposed for action in the Recommendations of the Working Group for its 1998 meeting. WGITMO members and the WGITMO Chairman shall:

* Contact the Russian delegate over the proposed introduction of blue crabs (*Paralithodes platypus*) from the Kamtschatka regions to the Barents Sea, relative to potential ecological impacts and relative to standard ICES procedures for the evaluation of proposed new introductions.

* Contact the Spanish delegate for information on the current status of *Caulerpa taxifolia* in Spanish waters, particularly its spread in the Mediterranean.

* Determine the possibility of translating the "ICES Code of Practice on the Introduction and Transfer of Marine Organisms 1994" into Norwegian, Spanish, Portuguese and Welsh.

* Determine the possibility of placing the "ICES Code of Practice", or a reference to it with information on how to obtain the Code, on the ICES web page.

* Gather detailed information and data for the Handbook of Dispersal Vectors, and seek the input and cooperation of any interested EIFAC colleagues in doing so.

* Conduct a general survey to determine the extent to which the green alga *Caulerpa taxifolia* is being held in public and research aquaria in ICES Member Countries, under what temperature conditions, and the nature of effluent treatment, and also the extent to which this alga is available for purchase within the private aquarium hobby within ICES Member Countries.

* Continue to prepare historical risk assessment studies for selected case histories of introduced marine plants, invertebrates, and fish in ICES Member Countries.

19 RECOMMENDATIONS TO ICES COUNCIL

The recommendations appear in Annex 8 of this report.

20 CLOSING OF THE MEETING

A final review of the 1997 terms of reference was made and the proposed agenda and action points for 1998 were considered. The offer from Dr Bas de Groot to hold the meeting in The Hague in The Netherlands was accepted. Final draft recommendations were discussed, revised and approved by the majority of WGITMO participants. (Owing to an air and rail strike in France on 25 April some participants had to leave before the end of the meeting to ensure their homeward journey; as a result, all WGITMO members were then contacted the next week to review and comment on all the above recommendations again.)

The Chairman thanked all of the WGITMO members and guests for their dedicated work and thanked Dr Philippe Goulletquer and his colleagues for hosting the 1997 meeting in La Tremblade. All participants agreed that the meeting had been extremely well organised. The Chairman adjourned the meeting at 12.30 hrs, Friday, 25 April.

ANNEX 1

AGENDA

22 April 1997 Tuesday

9:00	**Opening Session** Welcoming and Introductory Comments by Chair Welcome by IFREMER Officials Appointment of Rapporteur Introduction of All Participants and Guests Logistical Announcements (meals, telephone, FAX, photocopying,transportation,etc.) (P. Goulletquer) Review of Agenda and changes, corrections, additions *1997 Terms of* ***Reference***
9:30	**Previous Work and the Code of Practice** Review of 1996 Gdynia Report and Addenda/Errata (if any) Update on NEMO Activities (see handout) Status of Recommendations from 1996 Gdynia meeting (see handout) The ICES Code of Practice: Status of Translations and other Matters Status of new Cooperative Research Reports: ¤ Summary of Introductions in ICES Member Countries as of 1990 ¤ Guidebook for Code of Practice ¤ Aalborg 1995 Ballast Water Symposium
9:50	**Overview of new European initiatives (C. Eno, K. Jansson, and others)** North Sea Intermediate Ministerial Meeting Statement of Conclusions EC Directive on Environmental Impact Assessment EU regulation to implement CITES IUCN and CITES COP OSPAR Questionnaire (Alien marine species)
10:00	**National Reports and Literature Dissemination** Canada (M. Campbell, R. Randall) Finland (submitted by mail) France (P. Goulletquer, H. Grizel, M. Heral, et al.) including a presentation on the invasion of the Asian shore crab *Hemigrapsus penicillatus* (P. Noel)
10:30	**Coffee Break (20 minutes)**
10:50	**National Reports (continued)** Germany (S. Gollasch) Ireland (D. Minchin) Norway (A. Jelmert, K. Jorstad) Poland (submitted by mail) Spain (M. Ribera) Sweden (B. Holmberg, K. Jansson, I. Wallentinus)
11:55	**Group Photo**
12:00	**Lunch**
1:15	(sharp) Reconvene for the Afternoon Session **National Reports (continued)** UK England and Wales (S. Utting, C. Eno) USA (J. Carlton)
3:30	**Coffee break**
4:00	**Brief Status Reports, as Available:** Status of the Atlantic worm *Marenzelleria* introductions in Europe Status of the Asian kelp *Undaria pinnatifida* in Europe
4:15	**Australia's National Centre for Research on Introduced Marine Pests** (CRIMP) (R. Thresher)
5:00	**Status of *Porphyra yezoensis* in the Gulf of Maine** (USA) (I. Levine)
5:45	**Adjourn**
6:15	**Reception at City Hall, Hosted by Mayor of La Tremblade**

23 April 1997 Wednesday

9:00	Review of Previous Day, and Today's Agenda Announcement about Afternoon Field Trip (P. Goulletquer)
9:10	Marine Biocontrol Session: ¤ Review and Discussion of the Report of the "Study Group on Marine Biocontrol of Invasive Species"
10:05	Marine Biocontrol Research in Australia (L. Goggin)
10:30	Coffee Break (20 minutes)
10:55	Marine Biocontrol Research in France and Status of *Caulerpa taxifolia* Invasion in the Mediterranean (A. Meinesz) (Marine Biocontrol Session will be continued tomorrow morning)
11:30	End of morning session, return to hotel
12:00	At Hotel: change for field trip, pick up box lunches provided WGITMO FIELD TRIP: Oyster mariculture with the introduced Pacific oyster (*Crassostrea gigas*)
12:30	Board boat for field trip to the oyster farms of Marennes-Oléron Bay
2:30	Return to La Tremblade: Visit oyster packing houses
5:00	Adjourn

24 April 1997 Thursday

8:45	Announcements and Agenda Review
8:55	Marine Biocontrol (continued) Marine Biocontrol in the Black Sea: The GESAMP Report on the Comb Jellyfish (Ctenophore) *Mnemiopsis leidyi* (J. Carlton) Questions and Discussion
9:15	Marine Biocontrol: The Influence of the Parasitic Cirripede *Sacculina carcini* on its Brachyuran Host *Carcinus maenas* within its Home Range (D. Minchin) Questions and Discussion
9:45	Completion of Review of the Study Group Report on Marine Biocontrol, including Review, Discussion,and Further Questions on the Presentations from Australia, France, and Ireland, and the GESAMP Black Sea Report
10:30	Coffee Break
11:00	Complete Review of the Study Group Report Conclusions and Final Comments from Working Group
11:30	Risk Assessment Research at CRIMP (Australia) (R. Thresher) Questions and Discussion TOR: Risk Assessments: Postpone until 1998 (see Friday morning)
12:00	Lunch
1:33	(sharp) Reconvene for the Afternoon Session TOR Item f): Risk of Disease Transfer via Movements of Stocks which are Tolerant to the Agents Responsible for Significant Diseases (see handout)
2:00	Disease and Ecological Implications Arising from the Introduction and Transfer of Aquarium (Ornamental) Marine and Estuarine Species
2:30	Cooperative International Data Bases Reference items from Study Group on Ballast Water and Sediments: # Inventory of databases relevant to ballast water issues # "International Network for Marine Invasion Research" (INFORMIR)
3:00	Discussion and Planning: "Directory of Transport Vectors" Overview of Transport Vectors (J. Carlton) Ship Fouling (D. Minchin)
3:30	Coffee Break
4:00	Writing Session (all participants)
5:30	Adjourn

25 April 1997 Friday

9:00	Announcements and Today's Agenda Discussion: Possible Theme Session for 1999/2000 ICES MEETINGS: "Marine Biological Invasions: Retrospectives for the 20th Century, Prospectives for the 21st Century"
9:20	TOR: Additional Material for Completion and Discussion: * Status of Activity with GMOs in ICES Member Countries
9:40	*Presentation, Discussion, and Editing of Recommendations* Assignments for Risk Assessment Project Assignments for Directory of Transport Vectors Project Additional Assignments
10:00	Principal Agenda Items for 1998 WG Meeting Place and Time for 1998 Meeting (Invitation from The Netherlands) Discussion of 1999 and 2000 Meetings
10:55	Concluding remarks by Chair
11:00	Adjournment of Meeting

ANNEX 2

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ANNEX 3

NATIONAL REPORTS

NATIONAL REPORT FOR CANADA

1. LAWS AND REGULATIONS

The Department of Fisheries and Oceans continues to work on amendments to the Fish Health Protection Regulations (FHPR). The 'interim amendment', referred to in the Canadian Report for 1995, has been published in the Canada Gazette, which allows the public a 30 day period for review before the amendment goes forward for final approval. The amendment should be implemented in the spring of 1997. Once approved this amendment to the existing regulations will embody a 'like-to-like' philosophy whereby facilities with FHPR pathogens may be permitted to transfer fish into the country or to another province providing that the recipient facility already has those specific agents.

Canada's specific 'Shellfish Health Protection Regulations' are at a stage where a draft is ready to be distributed for technical review.

The Province of Manitoba has drafted a new 'Fishing Licensing Regulation' which has sections which will prohibit the sale of live native species in the aquarium trade. It is hoped that this regulation can be finalised for implementation in the spring of 1997.

2. DELIBERATE RELEASES

2.1. FINFISH

Significant numbers of eggs and fish (rainbow trout, *Oncorhynchus mykiss*; chinook salmon, *O. tshawytscha*; sockeye salmon, *O. nerka*; coho salmon, *O. kisutch*; Atlantic salmon, *Salmo salar*; brown trout, *S. trutta*; cutthroat trout, *S. clarki*; Arctic charr, *Salvelinus alpinus*; brook trout, *S. fontinalis*; lake trout, *S. namaycush*; splake, *S. fontinalis* X *S. namaycush*; northern pike, *Esox lucius*; walleye, *Stizostedion vitreum*; smallmouth bass, *Micropterus dolomieui*; striped bass, *Morone saxatilis*; brown bullhead, *Ictalurus nebulosus*; yellow perch, *Perca flavescens*) continue to be transferred into the country, between provinces and intra-provincially in support of aquaculture (not intended to be released into the natural environment) and enhancement programmes (released into the natural environment). In general these movements receive rigorous scrutiny by regional/provincial introductions and transfers committees which consider disease, genetic and ecological risks. In addition, all transboundary movements of salmonids must also satisfy the Canadian FHPR. Some points worth mention:

The Saskatchewan Department of Environment and Resource Management imported 25,000 smallmouth bass (*Micropterus dolomieui*) fingerlings from North Dakota for release into three isolated water bodies. An additional 200 adult smallmouth bass were imported from Manitoba and released into one water body.

The Manitoba Department of Natural Resources deliberately releases to waters where they do not already occur: brook trout (*Salvelinus fontinalus*), brown trout (*Salmo trutta*), rainbow trout (*Oncorhynchus mykiss*), Arctic charr (*Salvelinus alpinus*), Arctic charr X brook trout hybrid (*S. alpinus* X *S. fontinalus*), and splake (*S. fontinalis* X *S. namaycush*). None of these species is capable of reproduction in the waters where they are stocked. Also, walleye (*Stizostedion vitreum*) and northern pike (*Esox lucius*) are stocked in Manitoba to create new self sustaining populations or to replace populations that have been destroyed by winter-kill.

A small number (<10) juvenile shovel-nosed sturgeon (*Scaphirhynchus platorynchus*) and a small number (2 or 3) juveniles of an unidentified species of sturgeon from Russia were found at a culture facility in northern Ontario. Neither sturgeon is on the list of species acceptable for culture in Ontario. These exotic fish have been secured at the culture facility and a future course of action is now being considered.

2.2. INVERTEBRATES

Movements of invertebrates also receive rigorous scrutiny by regional/provincial introductions and transfers committees which consider disease, genetic and ecological risks before permission is given. Of note:

In Atlantic Canada, American oysters (*Crassostrea virginica*) European oysters (*Ostrea edulis*), blue mussels (*Mytilus edulis*), soft-shell (*Mya arenaria*), hard-shell (*Mercenaria mercenaria*) and bar clams (*Spisula solidissima*) and giant sea scallops (*Placopecten magellanicus*) were transferred as seedstock throughout the region in 1996. As in previous years, all official movements of shellfish destined for hatchery-use or for relay in open water are screened for parasites, pests and diseases, prior to transfer.

In British Columbia Manila clam (*Tapes philippinarum*) and Pacific oyster (*Crassostrea gigas)* seed continue to be imported for beach-seeding purposes, from certified sources in the Pacific northwest USA.

2.3. ALGAE AND HIGHER PLANTS

Canada presently has no federal regulations (Fisheries or Agriculture) which can prohibit or control the movements of marine plants. Efforts are underway to have marine plants added to the 'Fishery (General) Regulations' made under the Fisheries Act. It is hoped that by 1998 DFO will have a legislative authority to manage introductions and transfers of marine algae and higher plants.

In 1996 the Federal Department of Fisheries and Oceans authorised the importation of a maximum of 100 nori (*Porphyra yezoensis*) nets from Coastal Plantations International, Inc., Maine to a private aquaculture site in Grand Harbour, Grand Manan, New Brunswick.

A commercial company, Ocean Produce International (OPI) has been growing SEA PARSLEY™, a natural variation of dulse (*Palmaria palmata*), since April 1995. OPI has an exclusive agreement with the National Research Council (NRC) of Canada to commercially market the variant that NRC first discovered in 1978 on Grand Manan Island, New Brunswick. NRC has maintained the seaweed in culture since 1978 and has confirmed that it is a male plant natural variation of *P.palmata*. In 1996 there was a request to set up a commercial SEA PARSLEY™ operation in New Brunswick using OPI technology (salt water tanks in land-based greenhouses) and seaweed stock. The Federal Department of Fisheries and Oceans has indicated that DFO permission is not required for this type of transfer since ".... 'Sea Parsley' is a naturally occurring mutation of dulse....".

3. ACCIDENTAL INTRODUCTIONS AND TRANSFERS

An ongoing research project at the Department of Fisheries and Oceans' Maurice Lamontagne Institute aims to assess the risks for the introduction of non-indigenous species through ballast water discharges in the St. Lawrence River estuary and the Gulf of St. Lawrence. The evaluation of these risks is important in light of current Canadian regulations and guidelines which allow ballast water exchanges by foreign ships in the Gulf of St. Lawrence in situations where it is not feasible to exchange ballast water in the Atlantic Ocean. (Note: Research summary presented at the April 21, 1997 meeting of the ICES/IOC/IMO Study Group on Ballast Water and Sediments.)

A Transport Canada/DFO representative is a member of the Steering Committee of the US funded Great Lakes Ballast Technology Demonstration Project. A filtration unit has been installed on a Canadian ship (the *Algo North*) and will be evaluated during the 1997 shipping season. Transport Canada has conducted a ballast water sampling study in the Welland Canal. DFO Science has initiated a study to examine the possibility of using organic acids to treat residual ballast.

3.1. FINFISH

Surveillance for ruffe (*Gymnocephalus cernuus*) in 1996 detected no change in expansion, but ruffe were captured again at the three sites at the extreme edge of its range: Ontonagon River, Michigan and Thunder Bay, Ontario in Lake Superior, and the port of Alpena, Michigan in Lake Huron. A total of 30 ruffe were captured at the Alpena site in 1996, up from the 3 ruffe captured in 1995 when ruffe were first detected in Lake Huron. The goal of the US Ruffe Control Program now emphasises preventing the spread of ruffe into watersheds other than the Great Lakes.

A special Symposium on the Biology and Management of Ruffe was held in Ann Arbor, Michigan during March 1997. Experts from several countries were invited to participate in the Symposium. The Symposium was designed to 'enhance the current understanding of the ruffe infestation and its implications to North America', and will hopefully 'result in cost effective management decisions'. Included in the discussions was an overview of ruffe biology; a synthesis of Eurasian ruffe research; ecology, distribution and impacts of ruffe in the Great Lakes; and a consideration of control, management and policy.

The round goby (*Neogobius melanostomus*) has not expanded its range across the north side of Lake Erie, as it has on the south shore (U.S. side) probably due to the lack of commercial ports on the north side. There was a confirmed report of gobies in the Welland Canal joining Lake Erie and Lake Ontario. In 1996, the tubenosed goby (*Proterorhinus marmoratus*) has become very rare on the south side of Lake St. Clair, being replaced by the round goby.

A European flounder (*Platichthys flesus*) was caught by a commercial fisherman in Thunder Bay, Lake Superior in February 1997. This represents the fourth sighting of this species in Lake Superior in the last 15 years (it has also been collected in Lake Erie and Lake Huron). Believed to be another ballast water invader, this fish is a marine species that regularly penetrates fresh water in Europe. It is believed that each sighting represents an individual ballast water introduction.

Two major escapements of rainbow trout (4,500 and 20,000 fish) occurred from 'holed' sea cages in the Bay D'Espoir aquaculture area of Newfoundland in the spring of 1996. All fish were believed to be triploids.

The Newfoundland Department of Fisheries, Food, and Agriculture reported that in January 1996, 470,000 rainbow trout and 130,000 Saint John River strain Atlantic salmon perished due to 'super-chilling' in cages in Bay D'Espoir. Aquaculture operators in the area feel that many of these fish did ultimately escape.

3.2. INVERTEBRATES

The situation where hard-shell clams (*Mercenaria mercenaria notata*) were 'unofficially' introduced directly into Atlantic Canadian waters from a shellfish hatchery in Maine in the early 1990's was mentioned in last year's report. Despite scientific advice to the contrary, approval was given for a portion of the clams to be transferred to a New Brunswick Department of Fisheries and Aquaculture facility in northern New Brunswick for use as broodstock. A histopathological examination of these broodstock, done after they had spawned, found the labyrinthulid-thraustochytrid, preliminary assigned the name "QPX" (quahog parasite X), which has also been observed in local hard-shell clams (*M. mercenaria*). Larvae from the broodstock were allowed limited distribution to New Brunswick aquaculturists willing to try out the notata clams in preliminary grow-out trials. In 1996 Prince Edward Island declined to participate until the outstanding, 'unscreened', notata which were still in a Nova Scotia hatchery (20 -25 animals from the original unofficial import) had been dealt with (destroyed or examined for disease). Although the Nova Scotia clams have spawned, there is a current moratorium on movement of notata from Nova Scotia to elsewhere in Atlantic Canada until the situation is better resolved.

The zebra mussel (*Dreissena polymorpha*) was found in eight new inland lakes in Ontario in 1996. Four of these lakes are isolated from previously known to be infested waterways indicating that overland transport has occurred.

The oriental mystery snail (*Cipangopaludina chinensis malleata*) was discovered in Stoney Lake, Ontario in 1996. This snail has now been found in several of Ontario's inland lakes. It is believed to have been introduced to the Great Lakes in the 1930's.

The spiny water flea (*Bythotrephes cederstroemi)* was found in Pigeon Lake in the Kawarthas Lakes region of Ontario in 1996. It is now established in 17 inland Ontario lakes as well as in all of the Great Lakes.

The amphipod *Echinogammarus ischnus*, native to the Ponto-Caspian region was found in the Detroit River during September, 1995 by John Witt of the University of Guelph, Guelph, Ontario, Canada. A manuscript on its occurrence has been accepted for publication in the Canadian Journal of Fisheries and Aquatic Sciences. By September 1996, Echinogammarus was found to be the dominant amphipod on rocky substrate in the Detroit and St. Clair Rivers, and along the north shore of Lake Erie to the Welland Canal and Niagara River mouth on Lake Ontario. The new amphipod has rapidly replaced *Gammarus fasciatus* as the dominant amphipod in the Welland Canal. In Europe, this exotic species has recently migrated to the Rhine and is often found to be closely associated with Dreissena clumps. The presence of an exotic amphipod among the *Gammarus* in the Detroit River and Lake Erie has been suspected by several benthic researchers since 1993. The probable mode of introduction is ballast water discharge, possibly at the same time as the introduction of the goby and *Dreissena* species into Lake St. Clair area.

The New Zealand Mud Snail (*Potamopyrgus antipodarum)* remains common in the south west end of Lake Ontario. The distribution of this hydrobiid snail in Lake Ontario is being published by Zaranko et al. in the Canadian Journal of Fisheries and Aquatic Sciences. The occurrence of *Potamopyrgus* in the Great Lakes was reported in the 1996 ICES report.

3.3. ALGAE AND HIGHER PLANTS

At the 1996 meeting we reported that a seaweed identified as *Codium fragile tomentosoides* had only recently been detected in Atlantic Canadian waters and the belief was that it was introduced with shellfish from the USA. At that time available evidence suggested that it appeared to only be abundant in Mahone Bay, Nova Scotia (south of Halifax on the Atlantic coast) and, despite evidence of some minor dispersal to adjacent areas, it did not appear to have spread much. In 1996 there were numerous reports of *Codium* being found in Gulf of St. Lawrence waters with a confirmation of its existence during the fall when samples were collected, firmly attached to American oysters (*Crassostrea virginica*), from a shellfish growing site in the Lennox Island Channel in western Prince Edward Island.

In 1997 federal and provincial biologists are planning to conduct surveys for *Codium*. As well, the Prince Edward Island Department of Fisheries and Environment is intending to distribute information on the seaweed, along with a photograph, to fishermen and aquaculturists with a request that anyone suspecting that they have found the seaweed contact them. It is also hoped that a graduate student with Dr Tony Chapman at Dalhousie University in Halifax will be conducting survey and impact research on this seaweed.

European frog-bit (*Hydrocharis morsus-ranae*), which was first imported into Canada from Switzerland by Agriculture Canada in 1932, continues to spread in southern Ontario. It was discovered in Balsam Lake in the Kawarthas lakes region in 1996.

4. LIVE IMPORTS NOT LIKELY TO BE RELEASED OR USED IN AQUACULTURE

The following organisms were imported into Canada for research, display or for human consumption. The organisms are generally held in quarantine or other containment facilities, and are either destroyed after research is completed or used as food. Importation of these organisms under the above conditions is not believed to pose a significant risk to Canadian fisheries resources. The list of organisms imported for food is not complete. Recording food organisms is low priority because of the low risk of impact on wild resources.

British Columbia

Species imported for bioassays, live table market or display purposes included: red abalone, spiny lobster, spotted prawn, white sturgeon, and American eels (these eels were brought for immediate transshipping to Asia only). Note: transfers of strictly freshwater species are not included and as the Transplant Committee has less control over imports for food and table market sales, lists for these purposes may not be complete.

Saskatchewan

Imports include live crabs and lobsters for human consumption as well as many aquarium species. The Province does not regulate these importations.

Manitoba

Inconnu (*Stenodus leucichthys*), were imported for research along with numerous unmonitored imports of species for the aquarium trade (aquarium dealers are advised of the list of prohibited species under the Manitoba Fisheries Regulations of 1985). As well, 1500 lbs. of live tilapia were imported for the food market.

Ontario

The University of Guelph imported Arctic charr (*Salvelinus alpinus*) fry from the Yukon for research purposes; these fish are being maintained in an enclosed isolated facility.

Live species imported and held in various types of holding units for human consumption are the same as reported in last year's report and include: lobster, tilapia, crab (unspecified), oysters (unspecified), striped bass, rock lobster, freshwater bass, clams (unspecified), bighead carp, scallops (unspecified), conch, cod, blue crab, blackfish, bluefish, butterfish, flounder, squid, eels, herring, mullet, catfish and miscellaneous 'fresh fish'.

Quebec

Rainbow trout (*Oncorhynchus mykiss*) and brown trout (*Salmo trutta*) were imported, from unspecified sources, for use as bioassay fish in the pulp and paper industry.

Nova Scotia

White sea urchins (*Lytechinus pictus*) from California; amphipods (*Eohaustorius estuarius*) from Oregon; cuttlefish (*Sepia officinalis*) from Woods Hole, Massachusetts; pacu (*Piaractus mesopotamicus*) from Brazil and tambaqui (*Colossoma macropomum*) from Brazil were imported for research purposes at the Bedford Institute of Oceanography and Dalhousie University.

New Brunswick

Atlantic tomcod (*Microgadus tomcod*) gametes (eggs and milt) were imported from New York for research at the DFO lab in Moncton.

Prince Edward Island

Atlantic salmon (*Salmo salar*) from New Brunswick; coho salmon (*Oncorhynchus kisutch)* from British Columbia; striped bass (*Morone saxatilis*) from Massachusetts and carp (*Cyprinus carpio*) from New Hampshire; were imported to a vaccine research facility.

Eight vials of milt from transgenic (chinook salmon growth hormone with eel pout antifreeze promoter) Atlantic salmon males were transferred from Newfoundland (4 males) and New Brunswick (4 males) along with 'normal' milt (2 males) and eggs (2 females) from New Brunswick to a transgenic research facility on PEI.

Newfoundland

Several importations of sea urchins (unspecified species) from California and British Columbia, rotifers (Brachionus plicatilis) from Israel and 14 importations of rainbow trout (*O. mykiss*). All of these animals were imported for research purposes and were destroyed either during or upon completion of the experiments for which they were imported.

5. LIVE EXPORTS TO ICES MEMBER COUNTRIES

Canadian aquaculturists continue to ship Atlantic salmon (*Salmo salar*), Arctic charr (*Salvelinus alpinus*), brook trout (*Salvelinus fontinalis*) and rainbow trout (*Oncorhynchus mykiss*) eggs and fish to the USA subject to US Title 50 fish health conditions. In 1996 an aquaculturist in Quebec also sent shipments of Arctic charr and brook trout eggs to France and Arctic charr eggs to Germany (brook trout eggs also went to Italy). A company in the Yukon also sent Arctic charr eggs to Germany, Ireland and the USA. Manitoba exported approximately 50 gallons of dace (*Phoxinus* sp.) to North Dakota, USA for use as live bait fish. Of interest also is the Saskatchewan government's shipment of 4 million walleye (*Stizostedion vitreum*) eyed eggs to the People's Republic of China. Saskatchewan is planning to ship more walleye and cisco (*Coregonus artedii*) eggs to the People's Republic of China in 1997.

6. PLANNED INTRODUCTIONS

6.1. FINFISH

Continued importations and transfers of salmonids for aquaculture, enhancement and research purposes from other provinces in Canada and from sources in the USA are likely. Of note:

As reported last year, proponents in the Province of Alberta continue to investigate the potential use of triploid grass carp (*Ctenopharyngodon idella*) to control aquatic vegetation in irrigation canals. Grass carp have been imported from the United States for research purposes. The Regional Non-Native Species Introductions Committee of the Federal/Provincial/Territorial Freshwater Fisheries Liaison Committee (members from Manitoba, Saskatchewan, Alberta and the Federal Department of Fisheries and Oceans), met several times during 1996 to discuss this issue. A risk assessment protocol was developed (based on and modified from a protocol prepared by the US Aquatic Nuisance Species Task Force), and risks associated with the use of grass carp in Alberta are being considered. This activity is ongoing at present.

Manitoba plans to import 250,000 muskellunge (*Esox masquinongy*) eggs from Minnesota. If the import goes ahead, it is planned that the fish will be stocked in western and southeastern Manitoba. All previous stockings of muskellunge, and there have been many, have failed to produce self-sustaining populations.

The Ontario Ministry of Natural Resources is planning to import wild coho salmon eggs (200,000 eggs) from New York waters of the Lake Ontario drainage in 1997.

The New Brunswick Department of Natural Resources & Energy is planning to use a Maine strain of landlocked salmon (*Salmo salar)* in selected lakes currently being stocked with landlocked salmon. This use of Maine salmon has been reviewed and approved by the Federal Department of Fisheries and Oceans.

The Federal Department of Fisheries and Oceans has given the New Brunswick aquaculture industry permission, to import a European strain (Mowi strain, landcatch strain) of Atlantic salmon from sources in Maine, USA into New Brunswick. A total of 300,000 eyed eggs have been imported through disease quarantine facilities to three aquaculture sites. The fish will be restricted to land-based facilities pending the successful development of 'containment procedures' which will reduce escapement risks to an acceptable level.

6.2. INVERTEBRATES

Continued importations and transfers of invertebrates for aquaculture, enhancement and research purposes from other provinces in Canada and from sources in the USA are likely. Of note:

Juvenile bay scallops (*Argopecten irradians*) will be transferred from a hatchery in New Brunswick to an open water site, also in New Brunswick, for grow-out trials.

Hard-shell clam (*Mercenaria mercenaria notata*) juveniles will be transferred from previously approved experimental grow-out sites in New Brunswick to other aquaculturists' sites in New Brunswick, Nova Scotia and Prince Edward Island. All of these animals originated from the broodstock which had been moved to the provincial facility in northern New Brunswick and which had received a post-spawning histopathological examination (see 3.2, above).

6.3. ALGAE AND HIGHER PLANTS

There is interest in expanding nori (*Porphyra yezoensis*) culture on Grand Manan Island, New Brunswick. The Regional Introductions & Transfers Committee has recommended approval with the same conditions as for last year, i.e. that nets only come from the approved Coastal Plantations International, Inc. site in Maine. As well, interest has been expressed by aquaculturists in Prince Edward Island and Newfoundland in the possibility of nori culture in those provinces; no decisions have been made.

Ocean Produce International (OPI) continues to aggressively market 'franchises' for the culture of the seaweed SEA PARSLEY™, a natural variation of dulse (*Palmaria palmata*), through trade shows and industry publications (e.g. Atlantic Fish Farming) throughout the Maritimes.

7. MEETINGS, CONFERENCES, SYMPOSIA OR WORKSHOPS

Under an environmental cooperation agreement between the Province of British Columbia and the State of Washington, parallel working groups have been developing a collaborative approach to increasing protection against negative impacts due to the introduction of non-indigenous species for their shared waters in Puget Sound and the Strait of Georgia. Consultation for this has included federal, provincial and state governments, aquaculture industries, port authorities, academic specialists, and other interest groups. The strategy under development addresses both intentional and unintentional introductions. It endeavors to safeguard the resources of the shared waters, without closing the door to new species being introduced for culture. It also recognizes the regulatory burden faced by the aquaculture and shipping industries, and seeks to establish collaborative, non-regulatory approaches where appropriate, based on a commitment from industry and government to meet the intent of the strategy. A workshop was held in spring 1996 to review theoretical aspects of species introductions and to bring together interested parties to discuss potential mechanisms for regulation of species introduction.

Great Lakes Panel on Aquatic Nuisance Species, Ann Arbor, MI, USA, April 1996, December 1996.
Planning Workshop on North American Non-Indigenous Freshwater Fishes, Gainesville, FL, USA, April 1996
National Workshop on Exotic Species, Hull, PQ, Can., March 1996

Ruffe Control Committee Meeting, Alpena, MI, USA, July 1996
IUCN, World Conservation Union, Alien Species Workshop, Montreal, PQ, Can., October 1996
Seventh International Zebra Mussel and Aquatic Nuisance Species Conference held in New Orleans, January 1997.
International Symposium on Biology and Management of Ruffe, Ann Arbor, MI, USA, March 1997.

The DFO Great Lakes Laboratory for Fisheries & Aquatic Sciences (GLLFAS) in Burlington, Ontario established an 'Exotic Species Committee' in 1996.

The 'Regional Non-Native Species Introductions Committee' of the Federal/Provincial/Territorial Freshwater Fisheries Liaison Committee (members from Manitoba, Saskatchewan, Alberta and the Federal Department of Fisheries and Oceans) met on several dates to develop a 'Risk Assessment Protocol for the Introduction of Non-Native Species of Fish'. The protocol is being applied to the use of grass carp in Alberta (see section 6.). The group was also asked to consider the risks associated with the live food fish industry.

Reported Prepared and Collated by

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NATIONAL REPORT FOR FINLAND

1.0 LAWS AND REGULATIONS

Regulations concerning import of live aquaculture and fishery animals and products from EU countries and from non-EU countries have been modified according to EU directives.

In accordance with the EU's position, all export and import of live farmed fish and gametes are prohibited between the EU and Finland. Imports from countries outside the EU are also prohibited from 1995-1997.

2.0 DELIBERATE RELEASES

Deliberate releases into the Baltic for fisheries management purposes were as follows in 1996:

3.7 million salmon (*Salmo salar*) juveniles

0.7 million salmon eggs

1.7 million sea trout (*Salmo trutta m. trutta*) juveniles

1.4 million sea trout eggs

5.3 million 1-2-summer-old whitefish (*Coregonus lavaretus*)

31 million newly-hatched whitefish

As in previous years, veterinary authorities allowed the import of elvers from England via Swedish quarantine. 110 000 elvers were released into inland waters in southern Finland in 1996.

3.0 ACCIDENTAL INTRODUCTIONS AND TRANSFERS

3.2 Invertebrates

The Ponto-Caspian cladoceran *Cercopagis pengoi*, recorded since 1995, was commonly reported in the open Gulf of Finland. It has been found to contribute significantly in the diet of the Baltic herring.

Since 1995 the bivalve ("zebra mussel") *Dreissena polymorpha* has been recorded in the eastern parts of the Gulf of Finland. The abundance of the species seems to be very low.

4.0 LIVE IMPORTS

4.1 Fish

See 2.0.

4.2 Invertebrates

As in previous years, aquarium shops and some restaurants and stores may import live marine animals such as oysters, lobsters and crabs for sale or consumption without the authorization of the Veterinary Department because it is obvious that they cannot survive in natural Finnish waters.

4.3 Algae or Higher Plants

None apart from aquarium plants.

5.0 LIVE EXPORTS to ICES Member Countries

5.1 Fish

Veterinary certificates for export purposes of live fish and eggs have been written as follows:

- Fertilized eggs of Atlantic salmon to hatcheries in Estonia and Denmark
- Fertilized eggs and fingerlings of rainbow trout in Russia (mostly Karelia)
- Fertilized eggs of rainbow trout in Chile

6.0 PLANNED INTRODUCTIONS

6.1 Fish

All export is prohibited from 1995-1997, when the IHN and VHS examinations will be carried out. However, elvers are planned to be imported annually from Swedish quarantine.

6.2 Invertebrates

None

6.3 Algae and Higher Plants

None

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NATIONAL REPORT FOR FRANCE

1.0 Laws and regulations.

The EU Directive 95/70/CE (December 1995) was implemented in France by the State decree from the Ministry of Agriculture, Fishery and Food (N°2742) in August 1996. The Directive specified the minimal controls over molluscan bivalve diseases. The list of national reference laboratories was included and the European Reference Laboratory specified, namely IFREMER La Tremblade. The latter has scheduled for next September a European working group with the National Reference laboratories to implement and coordinate this directive.

2.0 Deliberate Releases.

2.2 Invertebrates

The Manila Clam*Tapes philippinarum*

Following its introduction for aquaculture purposes during the 1980s, this species has spread naturally to the extent that a public fishery is now operating on a yearly basis. Landings reached 4 000 to 5 000 metric tons in 1996. A fishery management plan is currently under process. The Manila Clam *T. philippinarum* is now widely distributed. Both the Manila and the native European species *T. decussatus* occur concomitantly on the same beds, with an overlap between the populations. However no hybrids have been yet observed.

2.3.Algae.

Undaria pinnatifida

Natural subtidal *Undaria* populations have decreased significantly, especially in the Northern part of Brittany. A new ecosystem equilibrium has been reached following predation by a gastropod *Gibbula* sp.

Limited *Undaria* cultures (southwest of France) using longline techniques still exist without significant *Undaria* sightings in the nearby vicinity. Although small algae were found on the nearby mussel longlines, no overwintering occurred.

Accidental introductions and transfers.

3.1.Crustacean

Introduction of the Asian Shore Crab *Hemigrapsus penicillatus*

Please see the special presentation by Dr P. Noel in the main WGITMO Report, section 8.1.3.

4. Live imports.

4.1. Invertebrates

Based on the farming industry, Pacific cupped oysters *C. gigas* were imported from Ireland, Portugal and the U.K.

Flat oysters *O. edulis* were also imported from the former Yugoslavia, Croatia and the Netherlands.

5. Live exports.

5.1. Finfish

A significant quantity of finfish species were exported at the juvenile stage for aquaculture purposes: the flatfish turbot *Scophthalmus* to Spain and Italy and *Dicentrarchus labrax* to Greece and Italy.

A large increase in the elver public fishery was observed during the 1996-1997 winter season. All the elvers were exported for aquaculture purposes to Japan, South Korea and the People's Republic of China.

5.2. Invertebrates.

Pacific *C. gigas* cupped oysters were exported to Ireland and Portugal for farming purposes.

Report prepared by:

P. Goulletquer

NATIONAL REPORT FOR GERMANY

3.0 Accidental Introductions and Transfers

In 1996 no new non-indigenous species are reported from German marine or brackish waters. The following paragraphs review important aspects of previously found exotic species.

3.2 Invertebrates

Teredo navalis

The further spread of the shipworm *Teredo navalis*, occurring in the North Sea since 1700 and in the Baltic since 1875, was observed in the Baltic. Its distribution extends eastwards to the Island of Hiddensee and Rügen. This phenomenon combined with an exceptionally high population density in some areas (but no mass occurrence) raised the interest of the public after damaging different wooden harbour installations along the German part of the Baltic coast. Several newspaper articles, TV and radio interviews were published. The extended distribution of *T. navalis* in the Baltic Sea endangers several sunken sailing ships, as do zebra mussels (*Dreissena polymorpha*) in the Great Lakes of North America. Specialists try to salvage these several centuries old ships as fast as possible. The presence of these old ships in this area and the absence of *T. navalis* in their wooden construction is an evidence for the spread of the further limited distribution of the shipworm in the Baltic.

Marenzelleria cf. viridis and *M. wireni*

The introduced polychaete originally identified as *M. viridis* is now believed to belong to the different species *M. cf. viridis* and *M. cf. wireni*. *M. cf. viridis* is found in the Baltic since the early 1980s; the exact first record of *M. cf. wireni* in the North Sea is not known. It is believed that this species occurred earlier than *M. cf. viridis*. The taxonomical expertise is difficult due to the lost of the original holotypes.

The population density of *M. cf. viridis* and *M. cf. wireni* in German waters seems to be balanced. No further spread or mass reproduction nor any kind of negative effects on native species or the environment is documented. Both species were introduced unintentionally.

Anguillicola sp.

In some lakes of northern Germany the reported incidence of the swimbladder nematode infestations of eels introduced by live eel imports is slightly reduced. This nematode is spreading eastwards, being recognized by scientists from Hungary (Lake Ballaton).

Dreissena polymorpha

As in many other countries the Zebra Mussel (the translated German name is "Hiker Mussel") is spreading further. Increased population densities are mentioned in some areas of southern Germany.

4 Live Imports

4.1 Fish (Sturgeon)

Importation of juvenile sturgeon from Russia has continued for commercial purposes. A few small scale companies are still raising sturgeons. A few records of wild caught exotic sturgeon species (mostly *Acipenser baeri*) continued to be reported to the authorities of northern Germany (Schleswig-Holstein). In an attempt to save the common sturgeon *A. sturio* , a few specimens have been transferred from France to Berlin, where they are kept in a quarantine station in late 1996.

- Live Imports for Public Aquaria -

The opening of two oceanaria / public aquaria along the German part of the Baltic coast (Timmendorfer Strand in 1996 and Fehmarn 1997) has lead to the transfer and introduction of numerous species. The aquarium systems operate with

pre-treated brackish water, taken from the Baltic. A study on accompanying fauna (including parasites and disease agents) will be implemented shortly by the Institut für Meereskunde, Kiel.

4.2 Invertebrates

The oyster aquafarm located on the most northern German island (Sylt) of the Wadden Sea imported several 10,000 juvenile specimens in 1996. The exact number of the imported *Crassostrea gigas* is not known. The policy of this oyster farm is to sell the oysters as German Wadden Sea product, but by another source we know that they do import oysters from Ireland.

7 Meetings, Conferences, Symposia or Workshops on Introductions and Transfers

- Symposium of the "Society for the Protection of the German North Sea Coast" (Schutzgemeinschaft Deutsche Nordseeküste e.V.)

May 6. Agenda on commercial practice of native bivalves in northern Germany (Schleswig-Holstein). Relations between ecology and economy. One side issue probably will be the interest of cultivating non-indigenous species.

- Meeting of the Retired Seamen Club, Hamburg

May 22. The international shipping as vector for further species introductions

- Colloquium of the Institut für Hydrobiologie und Fischereiwissenschaften & Biologische Anstalt Helgoland

June 9 . Faunal distortion by international shipping

8 Bibliography

The first Ph. D thesis in the field of non-indigenous species introduced by ships was completed in 1996 by S. Gollasch at the Zoologisches Institut und Zoologisches Museum der Universität Hamburg. It contains the zoological results of the 4 year German shipping study. The first description of a new species (Turbellaria) found in a hull sample of the above mentioned shipping study was published in 1996.

Prof. Rosenthal's bibliography is finished now and includes 11.000 entries. Present problems of publishing on CD-ROM will delay the progress further. It is intended to provide the working group chairman of the WGITMO and the ICES headquarter as well as the EIFAC working party with an unpublished paper copy for internal use. This will be tabled during the ICES Annual Science Meeting 1997.

Reported prepared by

Dr S. Gollasch and Prof. Dr H. Rosenthal

NATIONAL REPORT FOR IRELAND

2.0 DELIBERATE RELEASES

2.2 Invertebrates

The abalones*Haliotis discus hannai* and *H. tuberculata* continue to be cultivated on west, south-west and south Irish coasts.

The clam*Venerupis philippinarum*, cultivated from hatchery seed on all Irish coasts.

The oyster*Crassostrea gigas*, all cultivated from hatchery seed imported from Normandy (to Counties: Clare, Kerry, Louth and Waterford) Arcachon (to County Waterford) France and Guernsey. All of the initial French half grown oysters introduced in 1993 have been marketed.

3.0 ACCIDENTAL INTRODUCTIONS AND TRANSFERS

3.2 Invertebrates:

Mytilicola orientalis

The copepod *Mytilicola orientalis* has become established in one bay, Dungarvan Bay, on the southern Irish coast, where in 1993 half grown Pacific oysters had been transferred from France. This copepod continues to be imported with oyster spat.

American oysters

An unapproved introduction of American oysters to Ireland from Long Island Sound was intercepted in air freight. The sample of 10kg contained the commensal pea crab*Pinnotheres ostreum* (5% prevalence), the slipper snails*Crepidula plana* and *Crepidula fornicata* were common (despite being brushed and cleaned) and egg cases of the predatory oyster drill*Urosalpinx cinerea* were found. The intended recipient was anxious to develop a trade in Europe for American oysters. The risk is that American oysters although intended for human consumption would become re-laid somewhere in Europe and act as a vector for these pests and diseases MSX and *Dermoscystidium.*

Italian mussels

A consignment of 11 tonnes of rope-grown market sized mussels from the Venice Lagoon (Chioggio) were transported to Ireland for processing but were refused because these had spawned. The haulier disposed of the mussels on the shore within 0.5km of an oyster farm. The sample obtained from the shore a week later was made up mainly of dead mussels, some live individuals have been subjected to histology. The shells of these mussels were clean.

American lobsters (*Homarus americanus*)

Live lobsters from North America although not presently entering Ireland are allowed apparent unhindered access to other Member Countries. Lobster imports to Europe may increase as part of a new trade agreement with North American states. There are concerns that imported lobsters will become placed in impoundments in the sea, even if commercial operators are advised not to do so, and thereby introduce American lobster diseases. Ireland, supported by the EU, has a significant investment in the ranching of European lobsters to help develop opportunities for local communities.

4.0 LIVE IMPORTS

4.1 Fish

Atlantic salmon eggs were imported from Scotland (16 million) and Tasmania (0.5 million). Rainbow trout eggs were introduced from Denmark (4.7 million). Arctic char eggs were imported from Canada (0.3 million).

4.2 Invertebrates

Pacific oyster (*Crassostrea gigas*) spat were imported in 67 consignments from England (49.7 million), 32 consignments from Guernsey (10.2 million) and 14 consignments from France (19.6 million). Five consignments of clam spat Venerupis philippinarum (6.0 million), and one consignment of 30,000 native oysters were introduced from England. A single consignment of 0.25 million scallops *Pecten maximus* were introduced from the west coast of Scotland.

5.0 LIVE EXPORTS TO ICES MEMBER COUNTRIES AND OTHER AREAS.

5.1 Fish

No significant changes recorded since previous year.

5.2 Invertebrates

No significant changes recorded since previous year.

7.0 MEETINGS

Bern Convention, Killarney, 26-29 May 1996.

ICES Statutory Meeting, Reykjavik, Iceland. October 1996.

Reported prepared by:

D. Minchin

NATIONAL REPORT FOR NORWAY

Status of releases and introductions

1.0 Laws and regulations

A new law for regulations of marine stock enhancement/sea ranching is now under evaluation, and decision about the presentation of the revised proposal in the Parliament will be taken in 1997.

2.0 Deliberate releases

2.1 Fish

Two fish species (*Salmo salar; Gadus morhua*) are now studied through the Norwegian Sea Ranching Program (PUSH). The main interest in the different projects is to obtain reliable estimates for recapture rates as base line for an overall evaluation of the program, focusing on the potential for commercial activities.

2.2 Invertebrates

As part of the Norwegian Sea Ranching Program a total of 130 000 microtagged lobster (*Homarus gammarus*) juveniles were released around the Kvitsoy islands from 1990 to 1994. All catches in the lobster fishery in the area have been monitored, and the hatchery derived lobsters have been identified. In the 1996 catches, the fraction of cultured lobster above legal size was 37%. The lobsters below legal size was dominated by cultured lobsters (69%).

Investigations on the introduced Red King crab (*Paralithodes camtschatika*) in the Barents Sea and the north Norwegian and Russian coastal area, are now carried out jointly between PINRO (Murmansk), Institute of marine Research, IMR (Bergen) and Fiskeriforsk (Tromso). Based on a trial fishery and several research vessel surveys in 1996, the information available (M. Hufthamar, IMR, pers. comm.) suggest a doubling of abundance estimates in the distribution area which is also indicated from the catch rates from the commercial trial fishery. The mature crab population now includes a large proportion of very big crabs, also compared with native populations in the north Pacific. Egg-carrying females are caught from the Russian border to Tanafjord, and tagging/recapture experiments indicate a westward migration. The rapid expansion in distribution area are possibly enhanced by big specimens including egg-carrying females.

In Russia there are also interests to introduce Blue crab (*Paralithodes platypus*) from the Kamtschatka regions to the Barents Sea. According to Yu. Orlov (VNIIRKH, Moscow) this species was evaluated for introduction to the Atlantic in the 1930s, and was considered to have high potential for adaptation success compared to the Red King crab. Such new introduction plans should, however, be evaluated by international scientific organisations including ICES.

In 1986-1987 there was some interest in Norway for cultivation of the Manila clam (*Ruditapes philippinarum*) and two commercial hatcheries started production. Juveniles were mainly produced for export but also released into the wild environment at six different localities along the Norwegian coast. In recent investigations at the release areas (Mortensen and Strand, in prep.) large live specimens of Manila clam were found at the two most southern localities. These individuals were all mature, but no successful reproduction in terms of juveniles was detected.

3.0 Accidental introductions and transfers

3.2 Invertebrates

The slipper limpet (*Crepidula fornicata*) (unintentionally introduced from North America through oyster imports) was first detected in the wild environment in Denmark and Germany in 1872. The first specimen was found in 1962 in Norwegian waters at the Skagerak coast. Since then, several observations of this species have been reported, mainly in the Skagerak region. In summer, 1996, a new live individual was found at Kvitsoy, in the western part of Norway (K. Sjotun, IMR, pers. comm.). This indicates an increase in the distribution area, in a western and northern direction.

During 1996 a Russian research vessel found the first specimens of snow crab (*Chionoecetes opilio*) in bottom trawl catches at Goose Bank in the southeastern part of the Barents Sea. According to Dr S. Kuzmin at PINRO, Murmansk, the first individual was a female, and later in the same year, four additional specimens (all males) have been caught

mainly in the same region. Two of these are available in the Museum of Moscow University. This species is widely distributed in the northwest Atlantic and it has been speculated that this species may have been introduced into the northeast Atlantic by ballast water recently.

3.3 Algae and higher plants

Sargassum muticum is now well established in the southern part of the Norwegian coast (Skagerak) and has also been observed occasionally on the western coast as far north as Hordaland. In summer 1996, dense aggregations of this alga was found in a number of shallow bays in the coastal area of Rogaland in western Norway. This indicates a northern movement of the established populations of this species.

7.0 Meetings

The Norway/UN Conference on Alien Species, hosted by the Royal Norwegian Ministry of the Environment, was held in Trondheim, 1-5 July 1996.

In connection with the overall reporting from the Norwegian Sea Ranching Program (PUSH), the first international symposium on 'Stock Enhancement and Sea Ranching' will be arranged in Bergen, September 8-11, 1997. A web-page for the meeting is available at http://158.37.91.10/2N/sear/hav97.html

Report prepared by:

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NATIONAL REPORT FOR POLAND

Distribution of the Round Goby *Neogobius melanostomus*

We spent last year mainly working up the findings from previous investigations. The distribution of the round goby is probably similar to that shown in the report from last year. We do not have any information about it from new localities, but investigations during this past year were not very extensive. In this year we will start tagging experiments. We hope that these experiments will give us new and valuable information

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NATIONAL REPORT FOR SWEDEN

1.0 LAWS AND REGULATIONS

No changes.

The Swedish Environmental Protection Agency is developing a general policy concerning introduced species and genetically modified organisms (to be completed during 1997).

The translation of the ICES Code of Practice into Swedish has resulted in a publication available from the National Board of Fisheries (Anon. 1996).

2.0 DELIBERATE RELEASES: Species deliberately released or held in open waters

2.1 Fish

Salmon, sea trout; elvers imported from England (Severn)

3.0 ACCIDENTAL INTRODUCTIONS AND TRANSFERS

3.2 Invertebrates

Marenzelleria viridis

The polychaete *Marenzelleria viridis* has been present in the Baltic Sea since the early 1980s. The first observation in Swedish waters was made in the northeastern part of the Hanö Bight, province of Blekinge, in 1990. Observations of its expansion along the Swedish coast of the Baltic Sea have since been made in national, regional and local monitoring programmes.

The northward expansion has taken *Marenzelleria* up through the Kalmar Sound. In 1995 it was observed in the Västervik area, but in 1996 it had retreated back to its 1994 limit at the mouth of the Emån River (Smith 1996). To the south, in the Hanö Bight, its numbers are still relatively low.

The first observations of *M. viridis* along the Swedish coast of the Bothnian Sea (at Norrbyn, south of Umeå) were made in 1995. The size of the individuals (4-5 cm) indicated that they had been present in the area for some years. In 1996 an increase in abundance was noted, and *Marenzelleria* is believed now to be well established in the Norrbyn area. Some stray observations have also been made further south, along the Ångermanland coast. Due to the prevailing direction of the currents (counter-clockwise), it is likely that *Marenzelleria* will arrive to the northernmost part of the Gulf of Bothnia by way of Finland. (K. Leonardsson, Umeå University; pers. comm.).

In 1994 the first observations of *Marenzelleria* in the inner archipelago of Stockholm were made, with a grand total of three specimens from three different sampling stations. By 1996 it was present in seven out of eight possible locations, with maximum abundances of 95 individuals/m^2, and at depths ranging from 5 to 40 metres. It is now also present at some stations in the "middle" archipelago (down to 60 metres depth), but it has not (yet?) been reported from open coastal waters. (A. Stehn, Stockholm Water Company; pers. comm.)

The opossum shrimp *Hemimysis anomala*, reported from Finnish waters in 1992, was observed in great numbers outside the Askö Marine Research Station (south of Stockholm) in 1995.

3.3 Algae and Higher Plants

Sargassum muticum

During the summer of 1996 a survey of the distribution of the Japanese brown algae *Sargassum muticum* was performed along the Swedish west coast and the results were compared with areas visited in 1993 (Karlsson 1997). The distribution of attached plants had expanded about 100 km south during that three year period. The species was found growing in large amounts in the cooling water discharges from the nuclear power plant at Ringhals, northern province of Halland, the northern Kattegat. It was estimated to have been there for at least some years (first observations by fishermen in

1992-1993) and were more coarsely built than elsewhere and often epiphytized by bryozoans and filamentous red algae. Intensive fishing activities were hypothesized to have contributed to the dispersal in the southern part of the west coast where the currents mainly are north going.

The southernmost locality for attached plants was found to be in the middle part of the province of Halland (Träslövsläge) south of the city of Varberg (the E Kattegat). The plants in these more brackish areas were fertile and similar in morphology to those along the northern parts of the Swedish west coast. The colonization has also proceeded to the inner parts of the archipelagoes in the province of Bohuslän, where it was first found attached in 1987. By forming large and dense populations the plants also act as a barrier for water movements, leading to water stagnation and enhancements of growth of ephemeral algae.

Phytoplankton

In 1996 the potential PST-producing dinoflagellate *Alexandrium minutum* was observed for the first time at the Swedish west coast (the E Skagerrak), being abundant during the end of June (Lindahl and Edler 1997). The species is previously not known as phytoplankton from the North Sea/Skagerrak area but has been recorded from the Atlantic coast from France (Brittany), Spain and Portugal as well as in the Mediterranean, E USA, Japan, Australia and New Zealand. However, live cysts of this species occurred in nine out of ten sediment samples collected in the autumn of 1995 along the coast of Bohuslän (Persson and Godhe 1997). Also the potentially toxic dinoflagellate *Gymnodinium cf. catenatum* occurred abundantly in the sediment samples (live cysts at seven and as dead from all ten stations). Still no motile cells of *Gymnodinium cf. catenatum* have been found in the area and the origin of the cysts is not known (cf. earlier reports).

4.0 LIVE IMPORTS

4.1 Fish

Elvers from England (Severn). Quarantine regulations followed.

4.2 Invertebrates

Lobsters from U.S.A and Canada. Oysters from France.

For scientific purpose crabs were imported from Australia (CRIMP) to test for the infection by the native parasite *Sacculina carcini* at the Kristineberg Marine Research Station on the Swedish west coast. The crabs were hold at specific facilities, all animals to be destroyed after the experiment and all shipping material destroyed upon arrival and seawater in the shipping container disposed of on land. Only males were imported (most of them juvenile) constituting 20 specimens each of *Carcinus maenas* (Australian population originally introduced there, Portunidae), *Ovalipes australiensis* (Portunidae), *Paragrapsus laevis* (Grapsidae); 12 specimens of *Portunus pelagicus* (Portunidae); 3 specimens of *Charybdis* sp. (Portunidae) and 1 specimen of *Nectocarcinus* sp.

5.0 LIVE EXPORTS to ICES Member Countries

5.1 Fish

ICES countries		Outside ICES	
Eggs to:			
Belgium	Salmon	Chile	Rainbow trout
France	Grayling	Taiwan	Salmon, Trout
Germany	Arctic charr		
Scotland	Rainbow trout		

Live fish to:

Norway Salmon, Trout

Live eels for consumption to:

Denmark Israel

Germany Italy

The Netherlands, Poland, Spain

5.2 Invertebrates

Blue mussels to Denmark and The Netherlands

7.0 MEETINGS, Conferences, Symposia or Workshops on Introductions and Transfers

International Symposium Paris, France, March 13-15, 1997 "Dynamics of invasive marine species: application to the expansion of *Caulerpa taxifolia* in the Mediterranean"

Seminar at the Swedish Environmental Protection Agency, Stockholm, May 6, 1997 "Polyps, Ports and Pilotage - Presentation of Queensland's 'EcoPorts'-concept and ballast water risk assessment in Australia.

PhD Course August 5-14, 1997 on "Ecology of marine invasions and introductions" arranged by NorFA and Åbo Academy, Finland.

Report prepared by

Bo Holmberg, Kristina Jansson and Inger Wallentinus

NATIONAL REPORT FOR U.K., ENGLAND AND WALES

2.0 DELIBERATE RELEASES

2.2 Invertebrates

Pacific oyster and Manila clam seed were produced in commercial hatcheries in England and Guernsey. Seed oysters were planted out at many sites around the coasts of England and Wales; Manila clam seed were planted out at one commercial site in Poole Harbour. Approximately 500 tonnes of Pacific oysters and 20 tonnes of Manila clams were harvested.

3.0 ACCIDENTAL INTRODUCTIONS AND TRANSFERS

3.2 Invertebrates

Natural settlement of Manila clams has occurred in Poole Harbour. It was reported that 3-4 year classes may be present and a preliminary survey was carried out by the Centre of Environment Fisheries and Aquaculture Science (CEFAS) in April 1997 to assess the extent and age structure of clams that have settled in the intertidal area. From the mean shell length of clams (39-44 mm) it would appear that clams are from one age group from a 1994 spawning. No small clams were found at the 7 sites surveyed in the Harbour. Some of the patches of wild clams have been fished out by commercial fishermen. (Unconfirmed information gave an estimate of 20 tonnes since autumn 1996.) Manila clam seed have been planted out in Poole Harbour in increasing numbers since cultivation of this species began in the late-80s (1989-1993, 1 to 2 million seed; 1994-1995, 6 and 8 million seed).

The discovery of 2 new introduced species in Cardiff docks in the winter of 1996.

The Zuiderzee or dwarf crab, *Rhithropanopeus harrisii,* was first found in Roath Docks, Cardiff, South Wales, in 1996. Searches have revealed that it is present throughout the dock system in salinities of 12–15. It is not known where the introduction came from, although this species is native to the east coast of America. It probably arrived in association with shipping, possibly in ballast water or clinging to hulls. Ships arrive in Cardiff Docks from all over the world, including coal barges from the U.S.A. It is found throughout much of continental Europe and was first observed in the Zuiderzee, The Netherlands in the 1870s.

A brackish water zebra mussel (of the family Dreissenoidea) *Mytilopsis leucophaeta* has very recently (end of 1996) been identified from Cardiff docks, in South Wales (Oliver, Mettam and Holmes, in press). A full size range has been found amongst a total of 26 individuals. This species was described from Belgium (under a different name) in 1835 and is present in other European countries, but it is not clear whether it has arrived in Britain from the continent or its native America.

3.3 Algae and Higher Plants

Undaria pinnatifida (attached plants) has been found at two additional locations along the south coast of England, namely Torquay (Devon) and the Isle of Wight, as well as at the original site of introduction in the Hamble (see previous National Reports). Plants have also been found on Jersey in the Channel Isles. Owing to the distance between sites, these represent sites of new introductions rather than natural spread from the original site.

Sargassum muticum was cleared from Strangford Lough, Northern Ireland during summer 1996. To date, no attached plants have been seen but sea water temperatures have been too low for it to grow extensively.

4.0 LIVE IMPORTS

4.1 Fish:

Approximately 20 million rainbow trout eggs were imported from disease-free sources. The majority of eggs were imported from the northern hemisphere, mainly Northern Ireland, Denmark and the Isle of Man while imports from the Republic of South Africa arrived in June - August to satisfy the demand for eggs in late spring and early summer.

The UK imports 4 tonnes of wild caught marine ornamentals (equivalent to 350,000). Trade involves around 1,000 species. Most of the species are only in transit and are rarely kept for more than 2-3 weeks before they are exported to European and world-wide markets. (Information from Trade Press - Fish Farmer, April/May 1997).

4.2 Invertebrates:

Seed Pacific oysters were imported from Guernsey for on-growing.

5.0 LIVE EXPORTS to ICES Member Countries

5.1 Fish:

Approximately 200,000 turbot juveniles were reared on the Isle of Man, some for export to Southern Ireland. Export of juveniles for on-growing in Galicia, Spain has ceased now that hatcheries in Spain and France have started to produce juveniles locally.

Trade in live elvers caught in the River Severn continued.

Tropical Marine Centre in England claims to being Europe's main importer and wholesaler of a wide range of ornamental fish, soft corals, molluscs and crustacea. The customer base in the UK extends to 650 retailers and wholesalers, livestock is supplied to Europe on a daily basis and to world-wide markets every week. The Centre also cultures a few of the higher volume species (clown or anemone fish, Amphiprion sp.) intended for the home and global markets. Three quarters of the output went overseas in 1995.

5.2 Invertebrates:

A commercial hatchery in north-west England exported 77 consignments of Pacific oyster seed (totalling approximately 70 million individuals) to Southern Ireland and 6 consignments (totalling 5 million seed) to Guernsey; 1 consignment of Manila clam seed was exported from the same hatchery to Ireland.

(See 5.1 for information on ornamentals).

6.0 PLANNED INTRODUCTIONS

6.2 Invertebrates

The shellfish industry continue to express interest in introducing Pacific oysters from the Pacific coast of the USA through quarantine to boost the gene pool of broodstocks held in the UK and in introducing tetraploid oysters from the USA. No firm proposals have been made.

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Aquarium trade in UK (from Fish Farmer, March/April 1997).

UK imports 4 tonnes of wild-caught marine ornamentals (equivalent to 350,000). Trade involves around 1,000 species.

No large-scale culture units are known in Europe except the Tropical Marine Centre in England, which is situated in Chorleywood close to Heathrow Airport. It is on the site of an old trout farm. Systems run on synthetic sea water made up with freshwater from a borehole.

Less than a dozen different species are grown but these species are potentially the higher volume species intended for the home and global markets. Three quarters of the output went overseas in 1995. A preponderance of clown or anemone fish, *Amphiprion* sp. are reared. Other species held on site include sea-horses and ornamental shrimp but these are not grown in commercial quantities as yet.

In the next 5-10 years, 30-40 species may be commercially cultured which could represent 10-15 % of the total value of the market. (Current global trade is 180 tonnes wet weight, excluding water and packaging. At present, most marine ornamentals are harvested from the wild.)

Tropical Marine Centre also claims to being Europe's main importer and wholesaler of a wide range of ornamentals, including soft corals, molluscs and crustacea. The customer base in the UK extends to 650 retailers and wholesalers, livestock is supplied to Europe on a daily basis and to world-wide markets every week. Most of the species are only in transit and are rarely kept for more than 2-3 weeks.

Legislation on importation of ornamentals into the UK is through licences issued by the Ministry of Agriculture, Fisheries and Food (see 1995 ICES WGITOM report). Some species, such as hard corals come under the CITES (Convention on International Trade in Exotic Species) regulations. Trade is not banned but is monitored. Permits are issued by the Department of the Environment.

Report prepared by:

S. Utting

NATIONAL REPORT FOR UNITED STATES OF AMERICA

1.0 LAWS AND REGULATIONS

The National Invasive Species Act of 1996 (NISA 1996) was passed by the U.S. Congress in October 1996. A copy of this law was distributed. The law updates the 1990 Nonindigenous Aquatic Nuisance Prevention and Control Act, and upgrades the level of ballast water management (vide discussion under the Joint ICES-IOC-IMO Study Group on Ballast Water and Sediments, 1997 meeting).

2.0 DELIBERATE RELEASES

2.1 FISH

The State of New Hampshire reports no further captures of chinook salmon (*Oncorhynchus tshawytscha*) in 1996. Any captures in 1997 would be expected to be the final fish returning from 1992 and 1993 plantings. No further plantings are planned, and the Pacific salmon program has ceased.

2.3 ALGAE

Work continues in the State of Maine with outplantings of cultured red algae *Porphyra yezoensis*. Reference is made to a separate report filed by Coastal Plantations, Inc.

3.0 ACCIDENTAL INTRODUCTIONS AND TRANSFERS

Ballast Water Update

Please see the Joint ICES-IOC-IMO Study Group on Ballast Water and Sediments report for a summary of USA activities on ballast water research and management.

A new book, published in October 1996 by the National Academy of Sciences Press, has also now appeared on ballast water management, entitled Stemming the Tide . This work is the product of a Committee on Ballast Water appointed by the National Research Council (an information flyer was distributed).

3.1 FISH

Extensive studies continue on the invasion of the Round Goby, the Tube-Nosed Goby, and the Ruffe in the Great Lakes of the U.S. and Canada (vide list of materials distributed at the end of this report).

3.2 INVERTEBRATES

Ostrea edulis Continues to Expand Range on American Atlantic Coast

The edible European flat oyster *Ostrea edulis* continues to spread, it appears, along the northeast coast of the United States. Intentional plantings in the 1940s resulted in small permanent populations in the State of Maine. However, since the late 1980s, established populations have been reported in Rhode Island and more extensively along the Maine coast. About 1996, *Ostrea edulis* was reported from the Great Bay Estuary, New Hampshire (L. Harris, University of New Hampshire).

Japanese Crab *Hemigrapsus sanguineus*

Numerous studies are now underway on the biology and ecology of this Asian crab on the Atlantic coast of North America. Two Ph.D. students, one at the University of New Hampshire, and the other at the University of Connecticut, are both commencing doctoral dissertations on this invasion. *Hemigrapsus*, first found in New Jersey in 1988, now occurs from Massachusetts to North Carolina.

Chinese Mitten Crab *Eriocheir sinensis*

The Chinese mitten crab is now well established and expanding in San Francisco Bay, California. Cohen and Carlton (1996) report in detail upon its occurrence and probable methods of introduction.

3.3 ALGAE

The European and Pacific Ocean (native to the latter) red alga *Grateloupia doryphora* has now been found to be established and reproducing on the Atlantic coast of North America. Populations were first found in 1996 in Narragansett Bay, Rhode Island. Studies are underway by Professors M. M. Harlin (University of Rhode Island) and M. Villalard-Bohnsack (Roger Williams College).

Materials distributed with USA National Report 1997

Sea Grant. 1996. Gobies: Cyberfish of the 90s (D. J. Jude)

Sea Grant. 1995. Round Gobies Invade North America (J. E. Marsden and D. J. Jude)

Sea Grant. 1996. The Zebra Mussel: Answers to Commonly Asked Questions

(C. R. O'Neill and D. B. MacNeil)

Sea Grant. 1997. *Dreissena*! (newsletter) volume 7, no. 6

Zebra Mussel Watch cards

Ruffe Watch cards

Round Goby Watch cards

Zebra Mussel Information Clearinghouse information leaflet

Color maps showing:

Number of fish species introduced into HUC4 Drainages in the United States

(10/96) (HUC = "Hydrologic Unit Code 4", which is a fine-resolution scale)

Florida Watersheds Reported with *Hydrilla verticillata* in public waters (10/96)

Zebra mussel (*Dreissena polymorpha*) distribution in North America (1/97)

Confirmed Round Goby Sightings (*Neogobius melanostomus*) (11/96)

Confirmed Ruffe Sightings (*Gymnocephalus cernuus*) (3/97)

Nonindigenous Aquatic Species (NAS): The National Nonindigenous Aquatic Species Geographic Information System information sheet

Reprints of Carlton 1996a, b, d, e; Carlton and Mann, 1996; Johnson and Carlton, 1996; Cohen and Carlton, 1997, Pierce et al., 1997

NEWSLETTERS

Three useful newsletters continue to be produced:

"Zebra Mussel Update", from the Sea Grant Institute, University of Wisconsin-Madison, 1800 University Ave., Madison WI 53705-4094 (FAX: 614-292-4364)

"Aquatic Nuisance Species (ANS) Digest" from the Freshwater Foundation, Gray Freshwater Center, 2500 Shadywood Road, Navarre Minnesota 55331

"Dreissena!", from the New York Sea Grant Zebra Mussel Clearinghouse (vide copies distributed).

All three cover a wide variety of marine and freshwater topics, and the first two are free.

Reported prepared by

J.T. Carlton
with contributions from
NOAA/Sea Grant: New York Sea Grant Program (Brockport NY)
USGS Biological Resources Division (Gainesville FL)

ANNEX 4

REPORT ON *PORPHYRA YEZOENSIS*

April 8,1997

Dr. James T. Carlton, Chairman
ICES Working Group on Introductions & Transfers of Marine Organisms
Maritime Studies Program, Williams College - Mystic Seaport
50 Greenmanville Avenue
P.O. Box 6000
Mystic, CT 06355-0990

Dear Dr. Carlton:

As per the letter dated January 10, 1994 from Emory D. Anderson, General Secretary of the International Council for the Exploration of the Sea (ICES) to Mr. William Brennan, Commissioner of the Maine Department of Marine Resources (MDMR), the following is Coastal Plantations International's (CPI) fourth annual report to the ICES - Working Group on Introductions and Transfers of Marine Organisms (ICES-WGITMO).

Culture Sites

1992: Two culture sites were established; Johnson Cove and Mathews Island (See Figure 1, "92" delineation's). In Johnson Cove a 24 net system was assembled. The system was removed within 60 days of assemblage due to regulatory restraints. A 30 net system was established just off of Mathews Island which was maintained from July - December, 1992.

1993: Three culture sites were established. Two in waters off of Eastport, Maine USA and one site in Harbour de Lute, Campobello Island, New Brunswick Canada (See Figure 1, "93" delineations). The Eastport sites, just east and north of Goose Island, were established in June and removed in December, 1993. The Canadian effort was established in late September 1993 on the aquaculture lease site of Mr. John Mallack.

1994: The two Maine lease sites established in 1993 were utilized in 1994. The lease site just north of Goose Island has been shifted approximately 600 feet due west. The lease site east of Goose Island has been shifted approximately 300 feet to the east to establish a 1320 foot buffer zone between CPI cultivation lease sites and the seabird nesting areas on Goose and Spectacle Islands. An additional 80 acre tract, Huckins Ledge, was permitted and utilized as CPI's nursery tract. It is located 4000 yards west south west of Goose Island, just west of Seaward Neck on "Huckins Ledge" in waters off of Lubec, Maine (See Figure 1,"94" delineation)

1995: The three 1994 CPI culture lease sites were similarly utilized in 1995. Additionally, as part of a National Marine Fishery Service (NMFS) grant, CPI established a small, 15 net, pole farm during the 1995 growing season. The farm was located just north of Mathews Island and is designated by "T" in Figure 1. The experimental system was reassembled in 1996 at the company's Goose Island Aquaculture lease site.

The company's effort to train displaced fisherman, (CPI offered a Nori Farming Training Seminar and a six month nori farming training course) has resulted in the formation of the Blue Hill Nori farming Cooperative. Their efforts have consisted of deploying six nori nets with *Porphyra yezoensis* in 1994, and eight nori nets in 1995 in the Blue Hill, Maine area, see Figure 2. 1996 was to be their first attempt at a commercial size operation. Difficulties in permitting resulted in zero cultivation during the 1996 cultivation season. The MDMR's staffing difficulties have been resolved and the commissioner awarded the necessary aquaculture lease site permit for the Blue Hill effort in December 1996. The Blue Hill Nori farming cooperative is on schedule for the 1997 cultivation season.

Additional 1996 efforts included the establishment of a 20 net test polyculture system at the Connors Aquaculture Deep Cove, Eastport, Maine salmon lease site, and the licensing of a 100 net effort by a fisherman in Grand Manan, New Brunswick. The nori:finfish integrated polyculture system was installed relatively late in the 1996 growing season (October) but the nori quality, as measured by color, growth rates and nitrogen content was significantly greater than the Maine Nori Company's nori harvested from its monoculture operations. A cooperative study of nori samples from the mono and polyculture systems was initiated by researchers from the University of Connecticut (Dr. Charles Yarish) and University of New Brunswick (Dr. Thierry Chopin). CPI is still awaiting receipt of the final results of the study.

Cultivation/Reproduction:

The *Porphyra yezoensis* cultivation season is limited by the minimum growing temperatures of 6-7° C. The cultivation season in the waters of Cobscook Bay usually comprises the first week in June to the first week in December. The 1996 season commenced June 5, 1996, two weeks later than 1995 season due in part to lower temperatures experienced at our Huckins Ledge lease site. The nets deployed were seeded in September 1995 at our Eastport facility and the Huckins Ledge nursery site. The nets were then returned to shore and stored in the company's freezer awaiting the following cultivation season. Monospore production was observed from both our indoor facility in addition to the outdoor nursery site (Huckins Ledge). Evidence of monospore production was observed from June through November during the nursery phase of 1996's seeding process. Successful recruitment was evident upon the seeded nets, cultivation system support ropes and the anchor lines.

Adjacent salmon cages were examined monthly for evidence of successful monospore recruitment. Two sets of salmon cages adjacent to our nori sites consist of 1. 2500 yards south and 2. 3500 yards west, southwest of our Spectacle and Goose Island Sites. Cages 1 and 2 are 6000 yards west and 2000 yards south, respectively, of our Huckins Ledge site. No evidence of *Porphyra yezoensis* on the cage structure nor mooring lines were observed.

As stated in CPI's 1996 report to the ICES WGITMO, a study entitled "Establishment of a Monitoring Program for the Mariculture of the Non-Indigenous Seaweed *Porphyra yezoensis* in the Gulf of Maine" has been and continues to be sponsored by CPI. The study, conducted by Dr. Donald Cheney and two graduated students from Northeastern University, has completed its' first phase (Appendix I). Their conclusions reflect similar results as those previously reported by CPI. The conclusions are: 1) *P. yezoensis* plants are present but uncommon on the shoreline adjacent to the CPI farm during the farming season, 2) local *Porphyra* species out-recruit *P. yezoensis* on our netting substrates, and 3) there is no evidence to date that *P. Yezoensis* will over-winter in Cobscook Bay and replace local *Porphyra* species. CPI will continue to support this study and an research proposal which expands the original Northeastern University study entitled "Effect of Nori Aquaculture on the Marine Flora of Cobscook Bay and Selected Sites Within the Gulf of Maine" submitted to the University of New Hampshire/Maine Sea Grant College Program by multiple Principle Investigators from University of New Hampshire and Northeastern University (Appendix II)

Recruitment:

Determination of natural or anthropogenic dispersal of *Porphyra yezoensis* has been accomplished this year by the participants of the Northeastern University study. Artificial substrates were placed adjacent to CPI's Goose Island site < 100 yards from CPI nets. (Figure 1, Appendix I). From prepared substrates samples were collected in the fall of 1996 which resulted in the identification of *P. yezoensis* in 1 of 20 nori plants collected. Over-wintering potential of *P. yezoensis* was examined from the March 1997 collections analyses, which resulted in zero of twelve *Porphyra* plants being classified as *P. yezoensis.*

Future Plans

CPI is in the initial stages of planning nori cultivation efforts outside of the ICES WGITMO's mandated "waters of the Gulf of Maine". Sites off the coasts of Rhode Island, Massachusetts, Connecticut and New York are being considered. *Porphyra yezoensis* or other nonindigenous *Porphyra* is **NOT** intended for these cultivation initiatives. Results from the $ 1.2 million Sea Grant College Program research grant has been the domestication of indigenous New England *Porphyra* species. *Porphyra amplissima* collected from Cobscook Bay, Maine and domesticated at the labs of the University of Connecticut will be tested this cultivation season at

CPI's Eastport aquaculture site. CPI anticipates its next farming effort will be installed prior to the 2000 cultivation season.

Please feel free to contact my office if you, the working group or ICES have any questions concerning this matter. A letter of confirmation to the present commissioner of the Department of Marine Resources is all that the MDMR requires for CPI to proceed. We appreciate the time and effort on our behalf and welcome a site inspection by any and all of the members of the ICES working group.

Yours very truly,

Ira A. Levine, Ph.D.
President

IAL:kab
Enclosures

cc: Robin Alden, Commissioner, Maine Dept. Marine Resources
Steve Crawford, Farm Manager, CPI's Maine Nori Company

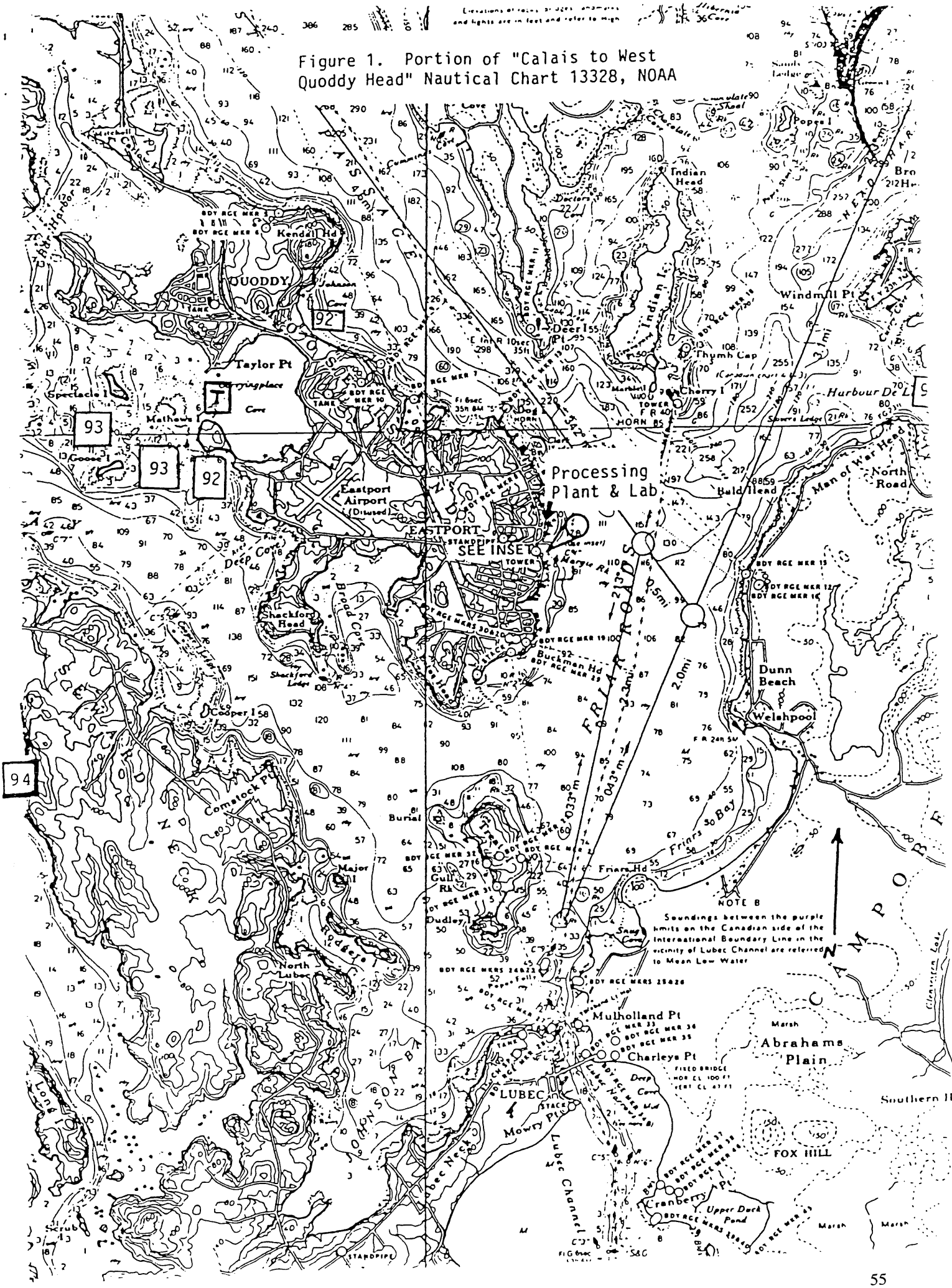

Figure 1. Portion of "Calais to West Quoddy Head" Nautical Chart 13328, NOAA

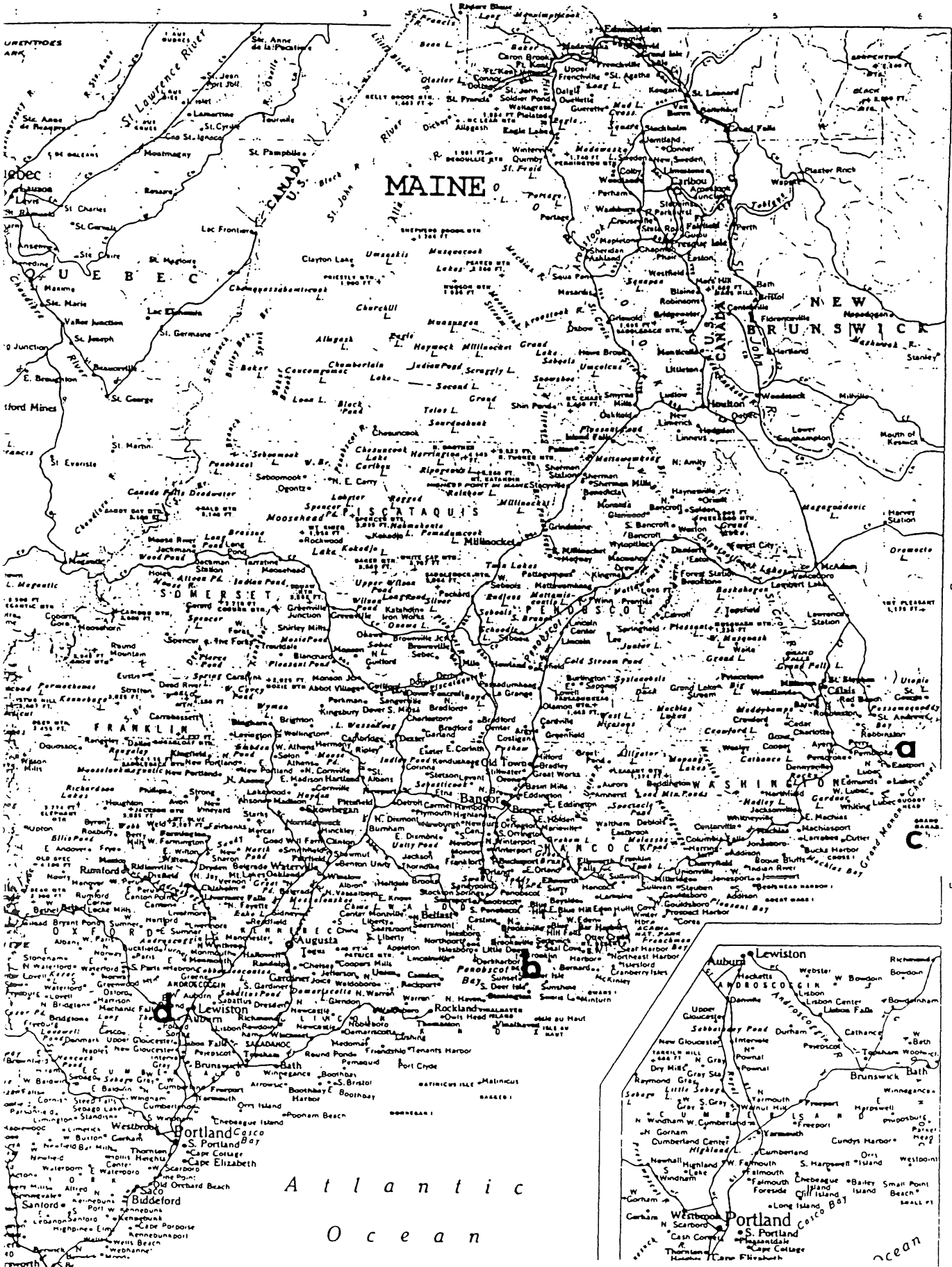

Figure 2. State of Maine Map: a. Location of CPI nori farming and processing activities; b. Experimental efforts of the Blue Hill Nori Farming Cooperative; c. Proposed site of 1996 Grand Manan, New Brunswick nori farm; d. Location of CPI corporate headquarters and laboratory.

Establishment of a Monitoring Program for the Mariculture of the Non-indigenous Seaweed *Porphyra yezoensis* in the Gulf of Maine: Preliminary Results

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and

Ira Levine
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Prepared April 2, 1997

Abstract

A monitoring program was initiated in the summer of 1996 to monitor the establishment and spread of the Japanese nori species *P. yezoensis* in the area adjacent to the site where it is being farmed by Coastal Plantations International. A two-fold approach was used which involved the sampling of both natural substrates and special netting substrates deployed to "collect" recruits from monospores. Based upon our findings to date, it appears that: 1) *P. yezoensis* plants are present but uncommon on the shoreline adjacent to the CPI farm during the farming season, 2). local *Porphyra* species out-recruit *P. yezoensis* on our netting substrates, and 3). there is no evidence to date that *P. yezoensis* will over-winter in Cobscook Bay and replace local *Porphyra* species.

Background

Although seaweed mariculture is a multibillion dollar business in Japan and throughout much of Asia, there is only a single company growing seaweeds in the United States today, Coastal Plantations International of Eastport and Poland, Maine. CPI has been growing the edible red alga *Porphyra* (or nori) on nets in Eastport, Maine, since 1991. The particular species of nori they farm, called *Porphyra yezoensis*, comes originally from Japan. Because of its non-indigenous nature, CPI had to obtain permits from a variety of international, national and state agencies to grow this seaweed in the Gulf of Maine. The permit process took a considerable amount of time, in part because of concerns that were raised over the potential impact of introducing a non-indigenous *Porphyra* species into northern New England waters. However, the permit applications were ultimately approved after CPI was able to provide data indicating that the temperature and daylength conditions in the area of the intended farm site (Cobscook Bay, Maine) would not permit conchospore release and therefore sexual reproduction.

In August of 1996, we initiated a small investigation to monitor the establishment and spread of *Porphyra yezoensis* in the vicinity of CPI's farm site. The goals of this study were simply to determine how abundant *Porphyra yezoensis* is in the vicinity of CPI's farm site after five years of farming, and to determine the extent to which *P. yezoensis* was able to recruit, over-winter and become established in northern Maine waters. We used a two-fold approach which encompassed both field sampling of *Porphyra* populations adjacent to the farm, as well as deploying artificial (netting) substrates to "collect" *Porphyra* recruits from monospores. In this report we describe our preliminary findings after the first seven months of study.

Methods

Field Collections

In August of 1996, we conducted a preliminary survey of the *Porphyra* populations found in the intertidal zone adjacent to CPI's nori farm (see Figure 1). We especially focused on a small island, called Matthews Island, that is the closest intertidal area to the nori nets. Matthews Island is exposed only at low tide, and receives current flow from the direction of the nets for a good portion of the tidal cycle. Matthew Island consists predominantly of mud and gravel with assorted boulders in the high intertidal zone. *Ascophyllum* and *Fucus* dominate the rocky surfaces, while large patches of *Ulva* and filamentous greens grow on the mud and gravel in the lower intertidal zone. *Porphyra* plants were found growing on the gravel in the lower intertidal zone where they formed large "slicks" and, to a lesser extent, growing epiphytically on *Ascophyllum* and *Fucus* in the mid to upper intertidal zone.

Because our bias was towards finding any *P. yezoensis* that might be present, we conducted non-random sampling and collected as many plants as possible that had *P. yezoensis* - like characteristics; that is, monostromatic, lanceolate morphotypes with *P. yezoensis* - like pigmentation. Samples were also collected from nets of a salmon farm approximately a mile away. Plants from both sites that appeared to have a*P. yezoensis* - like appearance were brought back to the laboratory for taxonomic determination using microscopic examination and isozyme electrophoretic analysis.

Isoenzyme Electrophoresis

Due to the morphological similarities of the monostromatic *Porphyra* species, isoenzyme electrophoretic analysis was employed to distinguish the non-indigenous *Porphyra yezoensis* from *Porphyra* species native to Cobscook Bay. Using the staining solution for the enzyme phosphoglucose isomerase (PGI), we have established a genetic "marker" for unequivically distinguishing the *P. yezoensis* strain farmed by CPI from local species (see Figure 2). On each gel we ran, we used a genetically uniform stock of conchocelis of *P. yezoensis* as a reference marker, which consistently bands as a single band (PGI-1; Figure 2) lower than those of the local species. The conchocelis stock we use belongs to the same seed stock that is used by CPI to seed their nets each year. Three other loci have been identified for the native species *P. umbilicalis* and *P. miniata*. *P. umbilicalis*, another monstromatic species, exhibits two bands (PGI-2 and PGI-3; Figure 2) which occur higher than PGI-1 and appear either singly or together (see Figure 2). *P. miniata*, a distromatic species, exhibits a single band above that of PGI-3 (not shown in Figure 2).

Use of Artificial (Netting) Substrates to Collect Recruits

The ability of *P. yezoensis* to spread to sites adjacent to the farm via the release of monospores was investigated through the deployment of special settling substrates, nori netting. Specifically, we deployed sections of the same type of Chinese netting that is used for growing plants on CPI's farm. Since such netting is known to be a preferential settling material for *P. yezoensis* spores, we presume it will act as an optimal substrate for testing the relative recruitment ability of *P. yezoensis* vs. that of local species.

Netting was affixed to four rocks on Matthews Island and one rock on an adjacent piece of the mainland nearby CPI's farm in August, 1996, which is approximately in the middle of their growing season (see Figure 1). Small pieces of netting, approximately 1 m^2 square were affixed to cleaned rock surfaces using bolts, cable ties and marine epoxy. In addition, a larger piece of netting, approximately 10 feet long was strung between two 6 foot metal "sign posts" (thus resembling a volleyball net) on Matthews Island and the mainland as well (see Figure 3). Samples of netting substrates were collected in January and March and sent to us for *Porphyra* species identification.

Results

More than 100 plants were collected in our August, 1997, field survey of Matthews Island. Upon microscopic examination, all of these plants proved to be monostromatic. They varied in shape from lanceolate forms resembling *P. yezoensis* to umbilicate forms typical of *P. umbilicalis*, the latter being far more common. Plants collected from the nets of the nearby salmon farm were distromatic and deep pink in color, which we have tentatively identified as *P. miniata*.

Isoenzyme electrophoretic analysis was run on several umbilicate-shaped plants and eighteen plants that most resembled *P. yezoensis* in their lanceolate form and pigmentation. The results are summarized in Figure 4. All of the lanceolate plants attached to gravel we tested did not exhibit the PGI-1 band characterisitc of *P. yezoensis*, which suggests that they were not *P. yezoensis*. However, 3 lanceolate plants that were attached to *Ascophyllum* did show the PGI-1 band, suggesting they did belong to *P. yezoensis*. All of the umbilicate shaped plants we tested did not show PGI-1, but rather PGI-2 and/or PGI-3, typical of *P. umbilicalis*. Thus, it appears from these preliminary findings that the majority of the *Porphyra* plants found on Matthews Island (at least during August) belong to a local species, with the possibility of a small percentage of plants belonging to *P. yezoensis*.

The netting substrates that were deployed in August on Matthews Island and the adjacent piece of the mainland held up well through January, when samples were collected and sent to us by Chris Bartlett. Fouling of the January netting samples sent us varied greatly by location, from 70% *Enteromorpha*, *Ulva* and *Chaetomorpha* coverage to no algae at all. *Porphyra* was less abundant and was found growing on five of the seven netting samples. A summary of which sites had the greatest number of *Porphyra* blades is shown in Figure 5. Altogether, a total of 42 *Porphyra* specimens were found recruited onto our January netting substrates. Twenty of these plants were of sufficient size (ie. greater than 1 cm long) to be analyzed eletrophoretically. Only one of these twenty plants demonstrated the PGI-1 band characteristic of *P. yezoensis* (Figure 6). The remainder of the plants showed banding patterns of either *P. umbilicalis*or that of another, highly seasonal, local *Porphyra* species, *P. purpurea.*

In March, we received another set of netting samples from Matthews Island and the adjacent piece of the mainland from Chris Bartlett. Fewer samples could be sent to us than in January because of damage to one of our "volleyball" nets and one of our rocks caused by urchin dragging activity in the area around Matthews Island. Nevertheless, 13 *Porphyra* specimens were found recruited onto the netting samples we received (Figure 5). Twelve of these plants were of sufficient size to be analyzed electrophoretically. None of these plants this time, however, demonstrated the banding pattern characteristic of *P. yezoensis*, but rather appeared to belong to *P. umbilicalis* or *P. purpurea* (Figure 6). Thus, although we recognize that our netting "sample size" was small, we saw no evidence for the ability of *P. yezoensis* to over-winter in the vicinity of the farm site. We hope to validate this observation further by making another field survey on Matthews Island in May, before the next CPI growing season begins.

Conclusions

Based upon our preliminary findings, it appears that: 1). *P. yezoensis* plants are present but uncommon on the shoreline adjacent to the farm during the farming season; 2). local *Porphyra* species are able to out-recruit *P. ezoensis* on our artificial (nori netting) substrates; and 3).there is no evidence to date that *P. yezoensis* will over-winter in Cobscook Bay and replace local species.

Acknowledgements

We gratefully acknowledge the support of Sea Grant #NA46RG0434 and the help of Chris Bartlett and Steve Crawford.

Figure 1

MAP OF SUBSTRATES

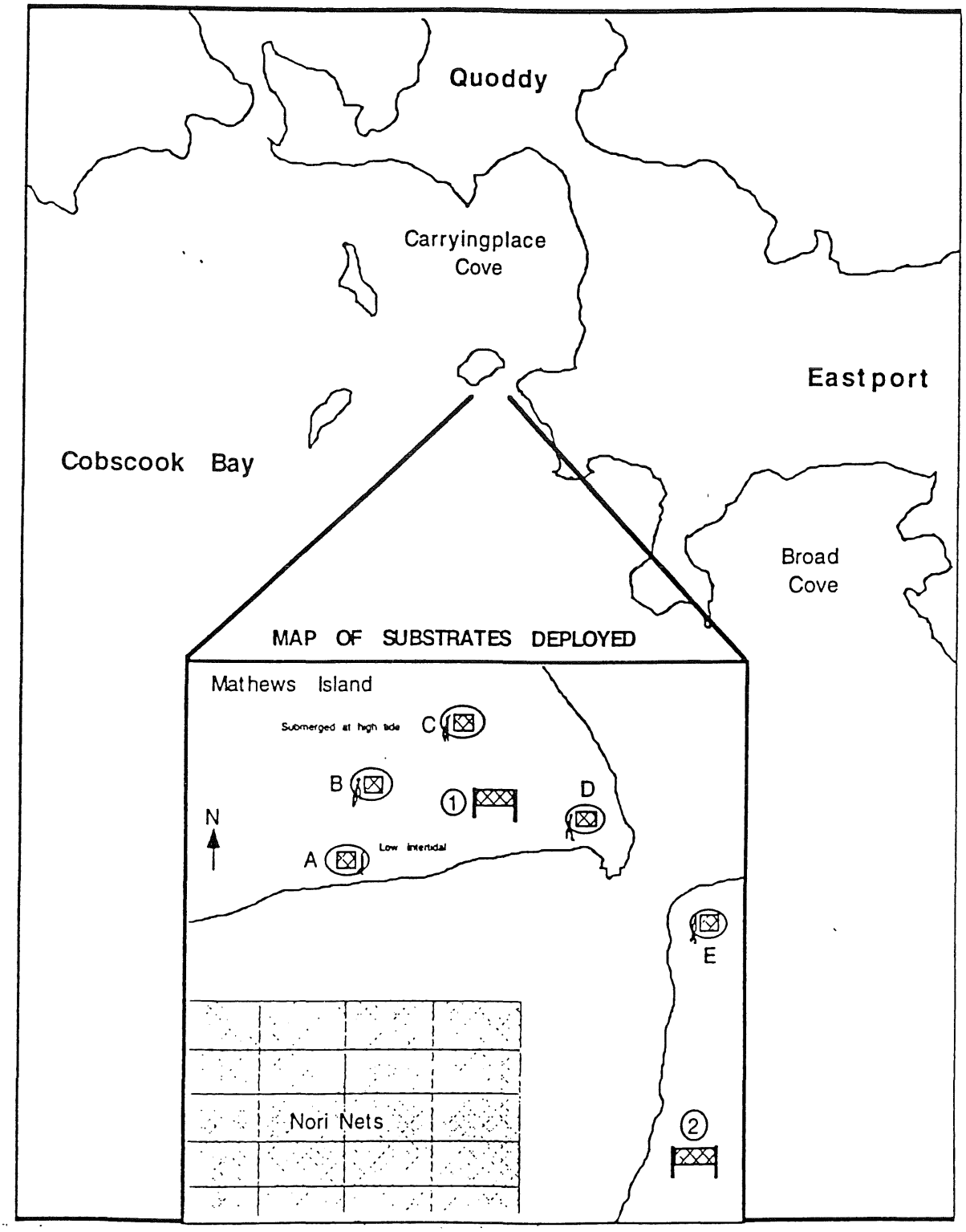
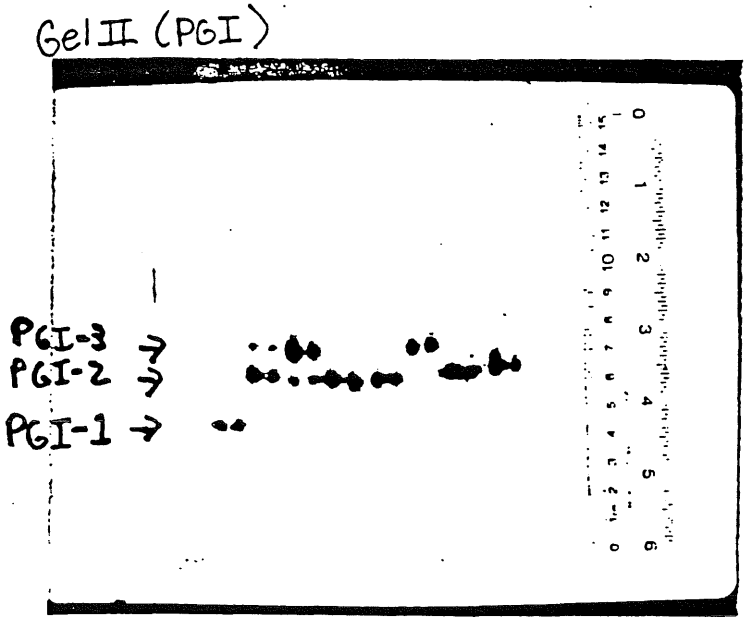


Figure 2

Sample of Isoenzyme Electrophoresis

(Phosphoglucose Isomerase Stain)



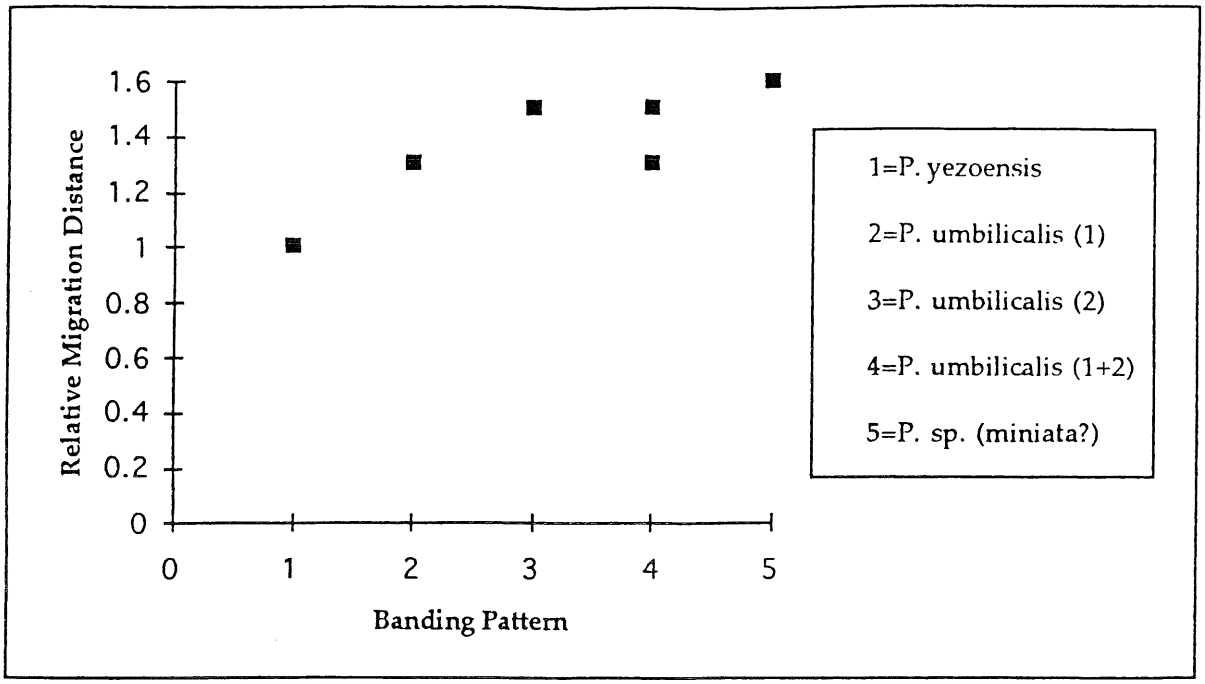
Columns 1 &2 - P. yezoensis controls; columns 3-16 - P. umbilicalis

Figure 3

"Volleyball" style nori netting set out to collect recruits

Figure 4

MAP OF ISOENZYME LOCI



ANALYSIS OF FIELD SURVEY SPECIMENS

(NOTE: Specimens analyzed do not represent a "random" sample. Of the 100+ plants collected, only those with P.yezoensis-like morphology and pigmentation were analyzed electrophoretically.)

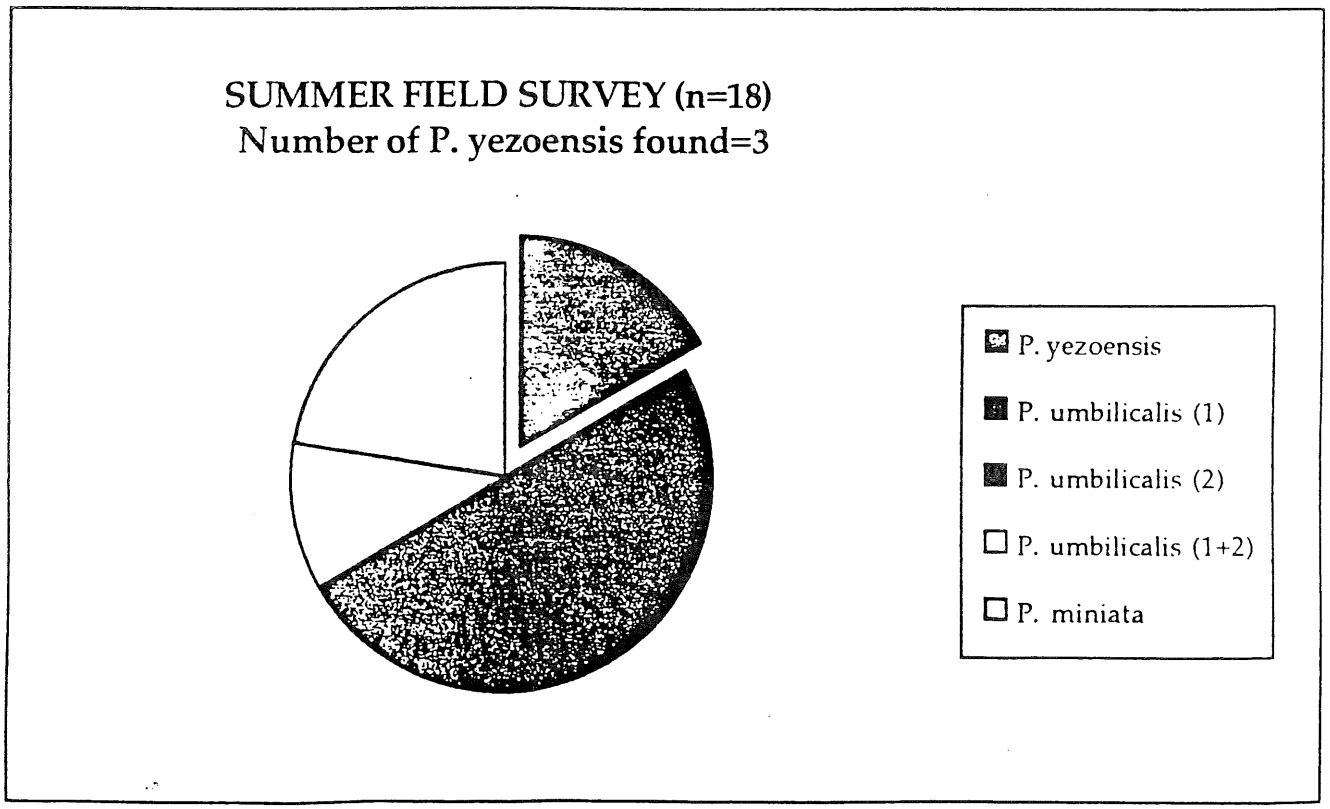


Figure 5

SUBSTRATE RECRUITMENT

Substrate (Distance from farm)	Fall Porphyra Recruitment	Spring Porphyra Recruitment
Net 1 (30 m)	14	X
Net 2 (20 m)	19	9
Rock A (20 m)	4	X
Rock B (40 m)	1	1
Rock C (60 m)	0	0
Rock D (30 m)	4	2
Rock E (30 m)	0	1

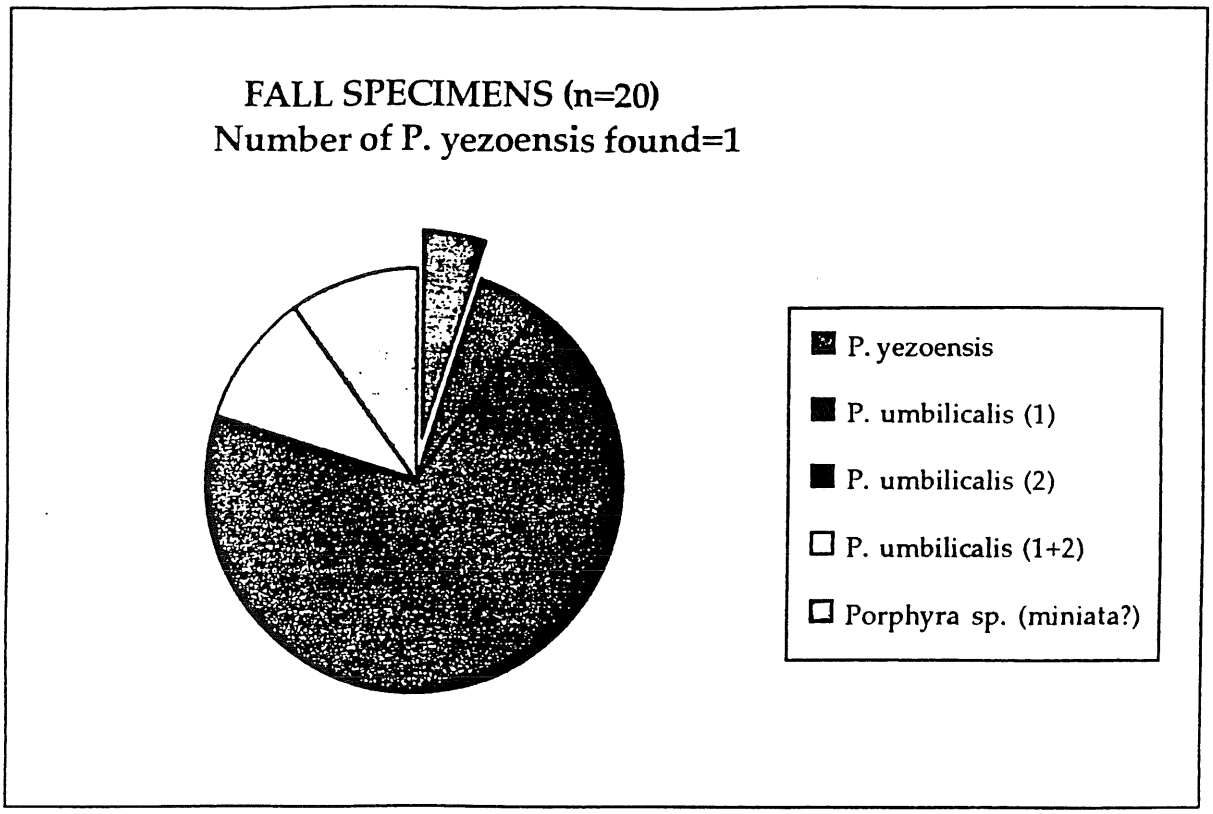
X=Destroyed by urchin draggers

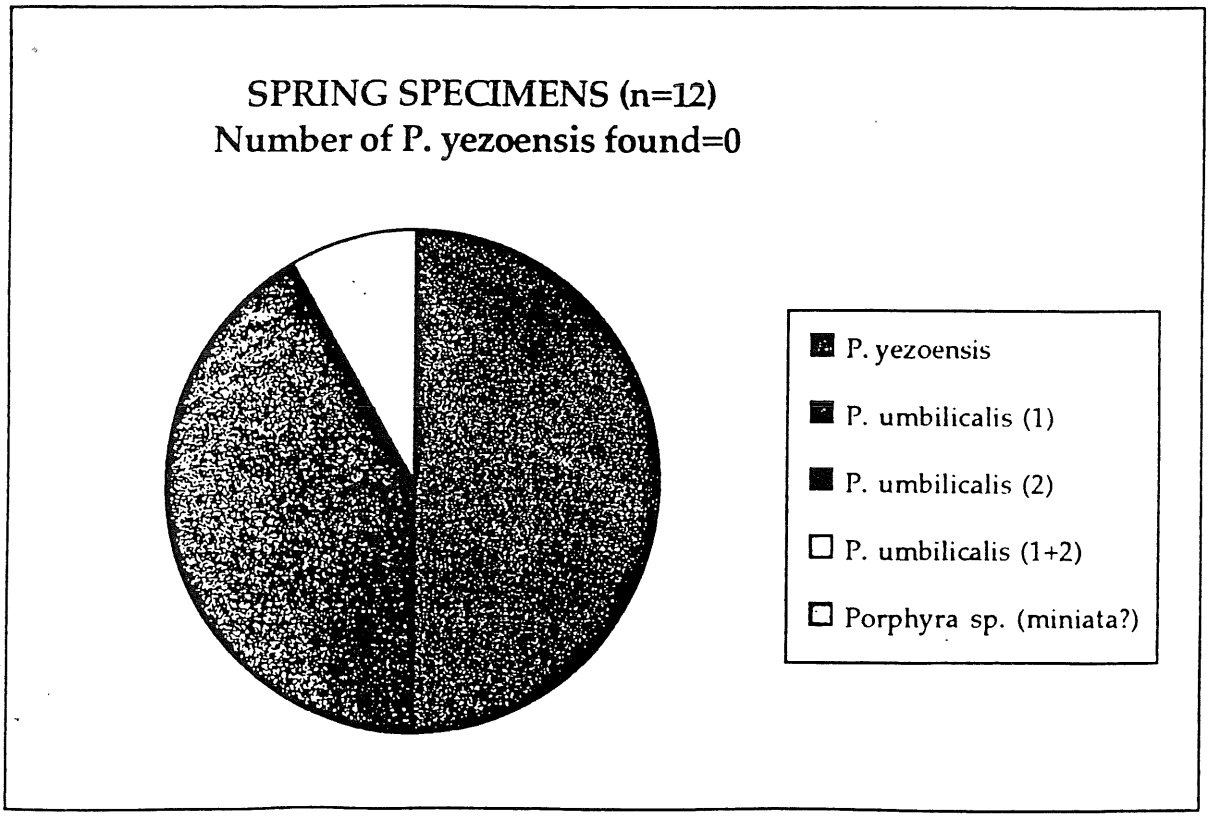
PHOTO A: (right)
Fall 1996 Recruitment Sample
from Net 1

PHOTO B: (below)
Spring 1996 Recruitment Sample
from Net 2 (in field)

Figure 6

ANALYSIS OF SUBSTRATE SPECIMEN





EFFECT OF NORI AQUACULTURE ON THE MARINE FLORA OF COBSCOOK BAY AND SELECTED SITES WITHIN THE GULF OF MAINE

Submitted to the

University of New Hampshire/University of Maine
Sea Grant College Program
For Funds available to Support Non-indigenous Species Research

by

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SEA GRANT PROJECT SUMMARY
UNH/UM SEA GRANT PROGRAM

TITLE: Effects of Nori Aquaculture on the Marine Flora of Cobscook Bay and Selected Sites Within the Gulf of Maine

PROJECT NUMBER: REVISON DATE:
GRANT NUMBER: INITIATION DATE: September 1, 1997
SUB PROGRAM: COMPLETION DATE: August 30, 1999
PRINCIPLE INVESTIGATOR(S): A. S. KLEIN (2 mo) and A. C. MATHIESON (2 mo)
AFFILIATION: DEPT. BIOCHEMISTRY & MOLECULAR BIOLOGY, UNH; JACKSON ESTUARINE LAB & DEPT PLANT BIOLOGY, UNH
ASSOCIATE INVESTIGATOR(S): C. D. NEEFUS (2 mo), D. P. Cheney (0.6 mo)
AFFILIATION: OFFICE OF BIOMETRICS, UNH; MARINE SCIENCE CENTER, NORTHEASTERNUNIVERSITY, NAHANT, MASSACHUSETTS
PROPOSED FED FUNDS: $181,201 PROPOSED MATCH FUNDS: $68,907
CURRENT FED FUNDS: CURRENT MATCH FUNDS:
TOTAL FED FUNDS TO DATE: TOTAL MATCH FUNDS TO DATE:
CURRENT PASS THRU FUNDS:
RELATED PROJECTS: PARENT PROJECTS:
SEA GRANT CLASSIFICATION:
KEYWORDS: ALGAE, AQUACULTURE, INTRODUCED, GENETICS, SEAWEEDS

OBJECTIVES: To assess the impact of the fledging nori aquaculture industry on the marine flora of Cobscook Bay and selected sites within the Gulf of Maines by: (1) examining dispersal and persistance of Porphyra yezoensis at sites adjacent to current and new cultivation sites; (2) developing genetic profiles for the U-51 strain that has been cultivated during the past seven years within Cobscook Bay, as well as additional strains of P. yeozensis proposed for use by Coastal Plantations Inc. (CPI) in their expanded aquaculture program; and (3) establishing baseline information regarding the distribution and abundance of indigenous Porphyra species within the Cobscook Bay cultivation and nursery areas, plus proposed new sites (e.g. the Bagaduce River near Castine, Maine).

METHODOLOGY: (1) Intensive monthly surveys will be made in order to assess the seasonal and spatial abundance patterns of Porphyra yezoensis and indigenous species surrounding diverse nori aquaculture sites within the Gulf of Maine; (2) Molecular methods based upon PCR-amplification of species-specific alleles will be used to positively type (identify) species; (3) Amplified Fragment Length Polymorphisms (AFLP) will be used to develop genotype profiles for the various cultivars of P. yeozensis, as well as to examine the occurrence of indigenous Porphyra species at each aquaculture site. Molecular methods are expected to be more accurate than traditional morphological/ reproductive features because of the plant's extreme phenotypic plasticity and infrequent occurrence of critical reproductive stages. They are also amenable to the high through-put required for this type of ecological survey.

RATIONALE: 1) To provide regulatory agencies and the aquaculture industry with a scientific basis for making decisions about the introduction of aquaculture strains/species in relation to the management of coastal resources; (2) To establish a scientific basis for measuring the impact of mariculture of economically important seaweeds on native species and the marine environment.

ACCOMPLISHMENTS: We have developed a variety of molecular techniques with diverse taxa of Porphyra during our recent Sea Grant Enhancement project dealing with nori cultivation. Further we have extensive field experience throughout the Gulf of Maine, including the Cobscook Bay and Bagaduce River (Castine) areas.

BENEFITS: The results of our molecular (i..e. DNA) and ecological studies should characterize the dispersal and immediate impact of Porphyra yezoensis (i.e. U-51 & other strains) on indigenous Porphyras surrounding aquaculture sites in the Gulf of Maine. They should also provide population and ecological information required for development of aquaculture regulatory policies for Porphyra.

GENERAL INTRODUCTION

A Regional Approach to the Problem of Marine Invasions: The problem of marine biological invasions by non-indigenous species is a complex one that, while national in scope, is addressed most effectively through a broad-based regional approach. The complexity of marine invasions is due to the diversity of vectors, taxonomy, life history, and habitats among the invading species, plus diverse environmental, social, and economic impacts of these invasions that can range from negligible to catastrophic. A problem of this magnitude requires a comprehensive, regional approach that will provide the following benefits: (1) an examination of invasions of multiple, non-indigenous species within a biogeographic region, from origination and establishment through their spread and subsequent impact, will allow a more comprehensive understanding of the mechanisms governing aquatic invasions; (2) integration of findings within this region will permit stronger comparisons between areas and ultimately produce a broader, more comprehensive perspective of this national dilemma; and (3) communication between researchers and "end users" in management and industry within a region will provide the basis for a more coherent regional policy for predicting, preventing, and mitigating introductions.

New England as a Premier Site to Study Marine Invasions: The coastal zone from Maine to Long Island Sound offers an unparalleled region in which to conduct experimental and observational studies to understand the dynamics of marine biological invasions. This ocean-dominated region, which is united by a common geography, hydrography, and climate, is composed of a broad range of habitats that exhibit various stages of human disturbance and biological invasion. Because New England possesses the longest history of intensive waterborne international trade and coastal development in the United States, it also has the longest documented history of marine invasions of any U. S. region (J. T. Carlton, pers. comm). This history records what may be the oldest documented marine invasion in the U.S. (the green crab Carcinus maenas, pre-1820), as well as one of the most recent (the red alga Grateloupia doryphora, 1996). Thus, the New England coastline represents a unique database to understand the tempo and mode of invasions.

Due to its Holocene glacial history, most of New England's coastline has been colonized naturally only within the past 10,000 years, causing a relatively low diversity of its biota. The relative simplicity of the system, where susceptibility to non-indigenous species has already been demonstrated, is well-suited to understand and to predict invasion impacts. Overlaying this low natural biotic diversity are the human-induced coastal alterations of the past 350 years, which have created subsystems even more depauperate in species, and thus, potentially even more susceptible to invasions. Further, coastal development pressures are intense, causing strong economic and social incentives for increasing mariculture (e.g., using non-native stocks of seaweeds and oysters) and raising concerns about the release and establishment of new exotic species. By integrating our findings within this region, we can make stronger comparisons between regions and ultimately gain a broader, more comprehensive

perspective of invasions.

A Diversity of Species, Habitats, and Approaches: A coordinated approach to the study of non-indigenous marine invaders in the northeastern region necessarily requires research involving a diversity of species, habitats, and methodologies. Invasions in this region involve organisms from a wide range of plant and animal divisions/phyla and a diversity of habitats, including intertidal salt marshes, rocky shores, offshore open ocean, and subtidal systems. These species, which range in morphology, reproductive strategy, and trophic mode, have, through a variety of mechanisms, significantly impacted native biota, fisheries' resources, and coastal recreation areas. Studies from the New England region will utilize a range of approaches to address the transport of these species and their impacts, including broad-scale biogeographic comparisons, manipulative experiments in the lab and field, and genetic analysis. This regional strategy, which explicitly incorporates a diversity of organisms, habitats, and methodologies will contribute substantially to the goal of forging a broader understanding of the threats that exotic species pose to coastal systems.

Solutions to Social and Economic Impacts: The economy of New England is critically dependent on marine resources, both directly from fisheries and aquaculture and indirectly from recreation and tourism. Introductions of non-indigenous species may jeopardize these marine resources and the economies that depend on them. For example, recently introduced species in subtidal habitats have the potential to seriously reduce landings and associated revenues of important fisheries species, including sea urchin and clam. Substantial losses of seasonal tourism revenues may result from recent introductions of seaweeds, which produce repellent beach detritus that deters beach goers. Human health may be directly impacted as in the case of shellfish contamination by toxin-producing dinoflagellates. These toxic microorganisms may cause substantial economic hardship to local communities if both wild and cultured shellfish beds are closed to harvest. Therefore, the projects addressed in the New England region concern species with a wide range of potential economic and societal impacts. The proposals will also significantly advance the development of effective prevention techniques (e.g., ballast water control), treatment and cleanup procedures, and will aid in the redesign or retrofitting of industrial equipment.

A Dialogue Between Researchers, Industry, and Management: To facilitate the regional coordination of research and education/outreach efforts related to marine biological invasions and to ensure an efficient communication network, a New England Marine Bioinvasions Regional Coordinating Committee (NEMB/RCC) will be established. Open meetings will be held and all stakeholders including representatives from the private sector, industry, government officials, and scientists will be encouraged to attend. In particular, all academic researchers on marine invasions in the New England area will be encouraged to attend these workshops, which would be held annually. Funding will be requested through individual grantee's travel budgets and through discretionary funds of individual state Sea Grant offices. The meetings will consist of updates of all research activities, news of recent or

expanding invasions, relevant studies from other regions, and educational and outreach program advances, with a particular emphasis on intra- and inter-state coordination. These meetings will also provide a critical opportunity to understand timely end-user needs and desires in order to refocus or enhance research directions.

Facilitating the Dialogue between Scientists and Stakeholders: Identifying and understanding the effects of non-indigenous species in marine ecosystems presents a special challenge to coastal resources managers who are responsible for developing policy and for stakeholders who may be affected by regulations designed to protect ecosystem integrity. Bringing together stakeholders with researchers early in the process of developing research agendas provides an opportunity for investigators to understand and integrate concerns and needs of managers and stakeholders; further it allows stakeholders to become aware of the limitations of science and the difficulties of translating results into useful information. Sea Grant Marine Advisory Programs are uniquely placed to facilitate information exchange through identification of stakeholders, coastal resource managers, and specific problems and concerns in the states. The Massachusetts Institute for Technology Sea Grant Marine Advisory Program will take the lead in developing a process for identifying how specific research data (e.g. on single species or methods of entry) can be used to develop policy. Tools that are likely to be useful to policy managers and stakeholders include developing maps of sensitive areas, developing risk analysis approaches, integrating life history data of successful invaders into potential risks, and generally providing a guide to the information through printed word and internet access.

INTRODUCTION/JUSTIFICATION FOR PORPHYRA STUDIES

The Genus Porphyra: Nori, the Japanese name for Porphyra, is a sizable genus of blade-forming red algae that grows on rocky, cold- and warm-temperate shorelines throughout the world (Brodie et al., 1996; Lüning, 1990). Most species are seasonal annuals (cf. Mathieson and Hehre, 1986), and they may live in some of the highest and driest reaches of the intertidal zone. The plant, which is also known as 'laver' (England), 'kim' (Korea), 'sluckum' (native Americans of the Pacific Northwest), and 'karengo' (New Zealand), is a major source of food for humans and the most valuable maricultured seaweed in the world. Approximately 15 billion sheets of nori were produced in 1992 (N. Takaoka, per. comm.), with an annual value of over $1.8 billion U. S. dollars (Jensen, 1993). Porphyra is primarily used as the reddish-black wrapping around 'sushi' rolls, which consist of chopped, pressed, and toasted blades, plus rice and other ingredients. Nori also serves as a preferred commercial source of the red pigment r-phycoerythrin, which is utilized as a fluorescent 'tag' for DNA and microscopic evaluations. Porphyra is currently the focus of a billion-dollar aquaculture industry in Japan, Korea, and the People's Republic of China, while a recent multi-investigator Sea Grant Enhancement project is addressing ways to help and improve current efforts to cultivate nori in the Northeastern United States (see Current and Pending Support).

Most members of the genus Porphyra have a biphasic life history that alternates

seasonally between foliose, gametophytic blades (i.e. nori) and microscopic, shell-boring sporophytic filaments, the conchocelis phase. Meiosis either occurs during the initial germination of conchospores (e.g. Porphyra purpurea) or presumably during conchospore production (cf. Mitman and van der Meer, 1994). Some Porphyras lack a conchocelis stages and exhibit 'direct' life histories; thus, blades are produced directly from germinating monospores (Guiry; 1990; Kornmann, 1961; Kornmann and Sahling, 1991). In culturing the asiatic P. yezoensis (i.e. strain U-51), the conchocelis stage is maintained under controlled conditions in the lab and its conchospores are used to "seed" the cultivation nets. The nets are then transferred to the field where the grow-out of foliose gametophytes occurs. The primary means of dispersal of Porphyra is via non-motile spores, which are either produced from its blade or conchocelis stage (e.g. carpospores, monospores, and conchospores). In most cases, the dispersal of viable red algal spores is thought to be quite limited (Guiry, 1990; Neushul, 1972; North, 1970; Norton et al., 1982; Zechman and Mathieson, 1985). Hence, the long-range dispersal of Porphyra would probably be associated with multiple transfers (generations) of spores. It should also be noted that fertile plants usually do not retain their reproductive potential upon becoming pelagic (cf. Fritsch, 1945; Lüning, 1990; Norton and Mathieson, 1983).

Porphyra Aquaculture in the Gulf of Maine: The establishment of the present nori industry in the Gulf of Maine (i.e. by Coastal Plantations Inc., Poland Spring, ME) was stalled for several years by delays in the permit processes of several agencies (i.e. ICES, state and federal) based on concerns over the potential ecological impact of introducing a non-native seaweed to New England waters. The fundamental reason for the delay was the lack of any scientific basis for predicting environmental effects. Although a number of phycologists in the Northeast were contacted, insufficient scientific information was available to predict whether wild populations of Porphyra yezoensis might become established and pose a threat to local ecosystems. Aside from the dispersal patterns outlined above, the establishment of wild populations of non-native or genetically manipulated species depends on their physiology, hydrographic conditions, intra- and inter-specific competition, disease resistance, etc. (Lüning, 1990). Ultimately, the permit applications were approved after the applicants were able to provide data indicating the combination of temperature and daylength conditions in the specified area (Cobscook Bay, Maine) would prevent the sexual reproduction of P. yezoensis.(I. Levine, pers. comm; Mumford, 1990).

For the nori industry to grow, it will be necessary to expand geographically. Thus, Coastal Plantations Inc. would like to use the Cobscook Bay area that was initially covered in its permits primarily as a nursery area to produce seeded nets for other growers in the Northeast; at this point they have apparently requested and obtained approval for an expansion of permits throughout the Gulf of Maine (I. Levine, pers. comm.). The potentials for introductions of genetically improved strains of Porphyra yezoensis may also be requested in the future (e.g. hybrids of P. yezoensis x native species with altered growth but not reproductive characteristics) as genetic manipulatioin studies are currently being conducted (Watson et al., 1996). At present, we lack crucial information for predicting key population dynamic characteristics of

any introduced Porphyra genotype into our coastal waters (e.g. their dispersal, recruitment and survivorship).

Porphyra yezoensis has now been under cultivation in Cobscook Bay, Maine for approximately seven years. The U-51 strain of Japanese P. yezoensis is currently being farmed in Maine due to a variety of growth, taste, and processing characteristics. Recently the state of Maine has agreed to expand the leasing of P. yezoensis beyond Cobscook Bay (e.g. the Bagaduce River-Castine area) as well as allow the introduction of additional cultivars of the same species. The Province of New Brunswick is also preparing to license nori aquaculture (T. Chopin, pers. comm). Therefore concerns about introduction of a non-indigenous species have been raised again in relation to expansion of the industry to sites where conditions may be favorable for sexual reproduction. The potential environmental effects of introducing reproductively viable non-native species of Porphyra remain unknown. Although we lack any scientific basis for predicting population dynamics for introduced Porphyra genotypes, there are a number of molecular marker systems that could be applied to this problem (see Molecular Methods below).

Impact of Porphyra yezoensi on Native Northwest Atlantic Porphyra's: One consideration in accessing the impact of Porphyra yezoensis mariculture on the ecosystem, is whether the non-indigenous species could displace native species of nori. Under a current Sea Grant Enhancement subproject (Neefus, Mathieson & Klein), we have begun to catalog native Porphyra species geographically and temporally throughout the Gulf of Maine and Long Island Sound. An immediate issue raised in this survey work, is the difficulty of distinguishing both native and non-indigenous species. That is, the taxonomy of Porphyra is notoriously difficult throughout the world, particularly within the North Atlantic and North Pacific (Bird and McLachlan,. 1992; Brodie et al., 1996; Lindstrom, 1993; Lindstrom and Cole, 1992a, b; Mitman, 1992; Mitman and van der Meer, 1994). In discussing species differentiation for British Porphyras, Brodie et al. (1996) emphasize that frond morphology within species is highly variable and that there are probably more species in this geography than previously recorded (cf. Parke and Dixon,1976). Bird and McLachlan (1992) record six species of Porphyra (i.e. P. amplissima, P. linearis, P. leucosticta, P. miniata, P. purpurea, and P. umbilicalis) and note that several "form-species" may exist. They speculate that there may be an underestimation of species richness for this geography. A similar situation may exist in New England and the mid-Atlantic (i.e. to Virginia), where each of these species is recorded, except for P. purpurea (cf. Hehre and Mathieson, 1970, 1993; Mathieson and Hehre, 1986; NUSCO, 1992; Schneider and Searles, 1991; Schneider et al., 1979; Taylor, 1957). In comparing the North Atlantic and North Pacific species of Porphyra, Lindstrom and Cole (1992a, 1993) emphasize that there are a number of taxonomic problems, particularly for "similar" species occurring in both geographies. At this point we know relatively little about the population genetics, taxonomy, or phylogeny of Porphyra and other Bangialean red algae, which could aid in the management and conservation of these valuable resources.

Molecular Taxonomy: Stiller and Waaland (1993) recently used PCR amplification

of the 18S genes, followed by restriction analysis, to generate riboprints and thereby examine species richness among Porphyra in the Northeastern Pacific. Bird and colleagues (1994) also used the 18S nuclear ribosomal gene to assess members of the taxonomically difficult red algal family Gracilariaceae, finding an unusually high level of intragenus sequence variation (~15%). Thus, there is as much sequence divergence for the 18S gene within the Rhodophyta as seen among the fungi or between green plants and green algae together (Ragan et al. 1994). The sequence of the 18S rDNA has been determine for more than 10 species of Porphyra (Friel et al., unpublished results; Oliveira et al. 1995; Ragan et al. 1994).

Because the species richness of Porphyra in the North Atlantic is probably underestimated, we (Klein, Neefus & Mathieson) are determining the 18S rDNA sequence for native species of Porphyra from diverse sites within the Gulf of Maine, Long Island Sound, and adjacent coastal waters of New Brunswick and Nova Scotia (see below). An essential component to this molecular identification scheme has been to examine the sequence diversity for the rDNA with each morphologically recognized species. In so doing, we found minimal rDNA sequence diversity among 10 accessions of both P. linearis and P. amplissima, while analogous comparisons of "Porphyra umbilicalis" showed two distinct groups separated by a 14% sequence divergence. Such results suggest that a second "cryptic" taxon is evident, which has formerly gone undifferentiated (Friel et al., 1996). PCR primers, specific to each taxon were designed and used to screen more than 200 "P. umbilicalis" samples (Figure 1). Each primer pair amplified rDNA from one or more Porphyras . This molecular screen also established that the cryptic "P. umbilicalis" was widely distributed along the New England coast, and occasionally is found sympatrically with the 'type'- P .umbilicalis. Detailed culture, life history, and ecological surveys are underway to better distinguish these two morphologically similar species (Sperr et al., 1996; Yarish et al., 1996, 1997 ; Yarish & Mathieson, unpublished results).

Similar surveys for rDNA sequence diversity within species are underway for native North Atlantic species, Porphyra purpurea, P. miniata and P. leucosticta, as well as the cultivated U-51 strain of P. yezoensis. At the conclusion of these surveys, we will be in a position to develop allele-specific PCR primers to selectively identify at least seven Porphyra taxa. Single gene traits would ordinarily not be sufficient for species classification. However, our sequence data have been correlated with specific life history patterns (Yarish, unpublished results), plus differences in multi-gene isozyme loci (Neefus, unpublished results).

The identification of Porphyra species by DNA-based methods has several advantages over traditional morphological classification and potentially over other biochemical means for typing species. Alpha taxonomy has proved to the single-most time consuming aspect of our current survey of Porphyra species diversity in the Gulf of Maine and Long Island Sound (Mathieson, pers. comm). Reproductive tissues, essential to accurate morphological classification, are often restricted to certain temporal and climatic windows, and are frequently unavailable for morphological assessment (Bird and McLachlan, 1976; Brodie et al., 1996). The two goals of our

proposed study are : (1) to provide an intensive survey of native taxa of Porphyras; and (2) to assess the dispersal of the non-indigenous cultivars in areas surrounding P. yeozensis lease sites. The proposed molecular techniques (see below) will allow us to positively identify hundreds of samples from each cultivation area. The high throughput and highly accurate identification of species should override costs concerns for molecular analysis. Identification by ribosomal DNA markers may be confirmed with other genetic loci- e.g. chloroplast or mitochondrial.

Genetic Profiles for Non-indigenous and Native Species of Porphyras: Hyper-variable molecular markers such as Randomly Amplified Polymorphic DNAs (i.e. RAPDs) have proven very effective for genotyping or identifying variation between different cultivars of Porphyra yezoensis (Dutcher and Kapraun, 1994; Mizukami et al., 1996; Patwary et al., 1993). Unfortunately, the error and noise levels associated with RAPDs may be problematic for localized phylogenetic studies (van Oppen et al., 1996). Microsatellite DNAs are also effective molecular markers distinguishing genetic relationships in plants, but they require cloning and sequencing, which is both time-consuming and expensive (Perkin Elmer, 1995).

The AFLP technology provides 'DNA fingerprints' that are characteristic and reproducible (Perkin Elmer, 1995). Markers are hyper-variable, because they target non-coding regions of the genome (Vos et al., 1996). AFLPs have been used for several applications, ranging from strain identification (Walsh-Weller et al., 1997) to mapping crop genomes. Genomic DNA is cleaved with both a rare and frequent cutting restriction enzyme, while double-stranded oligonucleotide adapters are ligated to the digested DNAs. The adapters allow selective amplification of genomic fragments with specialized oligonucleotide primers. There is a second round of DNA amplification, this time with fluorescent-dye-labeled primers. Fluorescent tagging allows computer monitoring of multiple AFLP profiles of up to 36 separated samples/gel, using an ABI DNA sequencing. Multiplexing by fluorescent tags expands the number of markers that may be resolved per sample in each gel run.

PROJECT OBJECTIVES AND SCIENTIFIC HYPOTHESES:

The dispersal and persistance of Porphyra yeozensis cultivars will be measured at the existing nori aquaculture site in Cobscook Bay and at any newly licensed sites (e.g. the Bagaduce River near Castine, Me). Three complementary hypotheses will be tested: (1) There is minimal dispersal and persistence of the non-indigenous P. yezoenesis; (2) There is dispersal but minimal persistence of P. yezoenesis around the aquaculture lease sites; and (3) There is dispersal and persistance of P. yezoenesis but no apparent impact of the distribution and abundance of native Porphyra species surrounding the aquaculture lease sites. By comparing survey data at a site that has experienced cultivation for seven years (Cobscook Bay) and newly leased site(s), we will obtain information that would otherwise be unobtainable- i.e. no pre-operation evaluations were made. Furthermore we will obtain baseline genotype information regarding native populations of Porphyra (see below), the U-51 strain of P. yezoensis that has been previously cultivated, plus other proposed strains (per. comm. I. Levine,

CPI). Such "genetic profiling" will provide invaluable insights into the population dynamics of both native and cultivated species.

RESEARCH PLAN:

Biological Surveys: Periodic biomass samples of Porphyra yezoensis and native Porphyra taxa will be collected near CPI's Eastport grow-out and nursery sites, plus other proposed aquaculture locations (e.g. Bagaduce River-Castine area). The samples, which will be collected consecutively during an 18 month period, will be used to quantify seasonal patterns of biomass (g dry wt/m^2) and species composition from "seeded" nets and solid substrata near the aquaculture site(s). Replicated samples of a uniform net surface area (~6-10 cm^2) will be collected. From these monthly samples ~50-75 fronds will be enumerated with respect to species composition (i.e. P. yezoensis versus native Porphyras), growth and major morphometric features (i.e. stature, coloration, sexuality, etc.). Stratified and replicated biomass samples (1/16m^2 quadrats) will be taken from a variety of rocks, pilings, etc. (cf. Gonor and Kemp, 1978; Greig-Smith, 1964; Kershaw, 1973; Lavery et al., 1991), with these quadrats being positioned upstream, downstream and within the middle of the aquaculture sites. A GPS or global positioning system would be employed to document the location of these sites. Sampling will also be done within the upper, middle, and lower shoreline in order to assess vertical, as well as temporal patterns of native and possible P. yezoensis escapes. The recruitment of new Porphyra plants on these denuded quadrats (i.e. native and P. yezoensis) will be followed throughout the project (i.e. two years), allowing an assessment of temporal dynamics of colonization, species interactions, and "mature" community structure after 23 months of regrowth (cf. Burns and Mathieson, 1972; Mathieson and Burns, 1975).

All of the biomass samples will be returned to the Jackson Estuarine Laboratory where they will be initially sorted to species, measured (stature), "sexed", and weighed (biomass/m^2). As a large number of samples will be assessed by molecular methods (>200 samples/site/month, below) only a few representative samples (10-15 plants) will be prepared as herbarium voucher specimens. Several relevant treatises will be employed to make initial taxonomic delineations (e.g. Bird and McLachlan, 1992; Brodie et al., 1996; Conway, 1964; Kapraun, 1984; Kapraun et al., 1991; Kornmann and Sahling, 1977, 1991; Krishnamurthy, 1972; Kylin, 1949; Lindstrom and Cole, 1993; Rueness, 1977; Scagel et al., 1986; Schneider and Searles, 1991; Taylor, 1957). Most of these accounts emphasize cell thickness (monostromatic versus distromatic), numbers of plastids/cell, external morphologies (lanceolate, linear, oblong, ovate, etc.), coloration, reproductive patterns (monoecious versus dioecious, presence or absence of distinct zones, etc.), ecology (epiphytic versus lithophytic, intertidal versus subtidal), and seasonal occurrence. After being initially identified, the samples will be delivered to A. Klein for DNA analyses (see below). Ultimately herbarium voucher specimens, which will be enumerated both morphologically and via DNA, will be prepared (i.e. with a proper name, site, date, collector, etc.) and deposited as permanent reference collections within the Albion R. Hodgdon Herbarium (NHA) at UNH.

The recruitment of Porphyra yezoensis and other native nori species will also be assessed by placing special netting, specifically made for Porphyra spore settlement, within the intertidal zone near CPI's farm. These studies will compliment the destructive biomass sampling described above, as well as provide an easy way to assess the relative potential recruitment of species near the farm site both seasonally and spatially. In addition, preliminary studies by D. Cheney and colleagues (August, 1996 to January, 1997) demonstrated the successful application of this procedure for "collecting" P. yezoensis recruits or "escapes," as well as the use of isozyme techniques in distinguishing this taxon from other local species (cf. Cheney, 1975; Roberts et al. 1997). Hence, we are optimistic that the molecular techniques outlined below will be successful in screening for different native species, as well as strains of P. yezoensis. It should also be noted that in their preliminary studies only one P. yezoensis frond was found amongst twenty-nine Porphyra plants that recruited onto netting near the aquaculture site .

These initial recruitment studies will be continued and supplemented as follows: (1) by establishing netting periodically to determine seasonal recruitment patterns; (2) by establishing netting at different distances from the farm site in order to assess spatial patterns; (3) by establishing netting near the nursery farm and the grow-out farm, as spores are likely to be produced by plants at both sites; (4) by removing netting during and after the farming season. All of the above materials would be returned to the Marine Science Center (Northeastern University) for initial analysis and testing. That is, the relative abundance of Porphyra yezoensis recruits versus native species would be initially determined based upon microscopic examination (see above), with the samples then being transferred to UNH for DNA analyses (see below). Based upon the destructive sampling and detailed recruitment studies, we should be able to determine if P. yezoensis recruits can survive in the field and how well they compete with local species. Hence, these studies will be helpful in determining to what degree, if any, P. yezoensis cultivation poses a threat to local Porphyra species.

Molecular Methods: Field samples of Porphyra blades are often covered with various epiphytic and epizoic organisms (e.g. diatoms, seaweeds, and invertebrates), all of which can contribute large amounts of DNA to a bulk genomic DNA isolation (C. J. Bird, pers. comm.) and go undetected in routine examination (Goff and Coleman, 1995; Klein and Smith, 1996). Mechanical and chemical methods have been used to reduce epiphyte loads on seagrasses (Zimba, 1996) and could also be used on freshly collected samples of Porphyra.

DNA-based analyses begin with the isolation of DNA. Fresh and frozen tissues are ground, and DNAs are isolated following a basic CTAB extraction procedure, supplemented with a phenol chloroform extraction (Stiller and Waaland, 1993). These DNAs represent total genomic preparations containing nuclear, chloroplast, and mitochondrial DNAs. As materials are processed in microcentrifuge tubes very small tissue samples are needed (<50 mg). Such methods permit DNA isolations from several dozen samples each day. DNAs will be distributed in a grid pattern on

microtiter plates in order to facilitate multiple PCR screens. Attention will be paid to maintain detailed records for samples and their correspondig DNAs (see above). The DNAs should be stable for several years when stored at ultralow temperatures; therefore they will represent archival collections suitable for future temporal and longitudional studies.

Allele-specific oligonucleotide primers have been designed to amplify portions of the 18S rDNA gene from two morphologically cryptic Porphyra umbilicalis taxa (Friel et al., 1996). Fragment amplification occurs only when the appropriate template is present in the DNA sample. Other primer pairs will be designed for the additional native and non-native strains of P. yezoensis. Up to 96 DNA samples may be amplified simultaneously. Amplification products will be separated on agarose gels and scored against authentic markers. It should be feasible to multiplex some PCR-primer pairs which would produce distinct size PCR fragments for different species. Several hundred samples can be screened simultaneously on three large agarose gels. Molecular identification of individual samples will be verified by partial rescreening of DNA templates. Data will be combined with the morphometric and ecological assessments (see above), allowing an enumeration of temporal variation in abundance and microsite variation for native species. In addition, they will provide a thorough assessment of the dispersal and persistence of the non-indigenous species, P. yezoensis.

Genetic Profiles: AFLP profiles will be developed primarily for DNAs isolated from haploid blade tissue. Genetic chimerism (cf. Mitman and van der Meer, 1994) and contaminating DNA templates present two potential hazards to applying standard population statistics, or population statistic methods derived for hyper-variable molecular markers, to Porphyra AFLP profiles. Another concern about AFLP's is whether the markers are "portable". That is, variable but comparable within subpopulations. In general AFLPs segregate as Mendelian traits (Perkin Elmer, 1995). Polymorphisms for AFLPs markers can be scored by similarity indices (e.g. Jaccard's Index) that may for example be able to discriminate subpopulations (Skroch et al., 1992). Markers that are consistent and polymorphic across each species will be treated as alleles.

Overall, comparison of AFLP markers will be used to examine major genetic components of native Porphyras near the aquaculture sites by: (1) estimating genetic variability and structure in populations; (2) evaluating temporal and geographical patterns of genetic variability within populations. In addition, AFLP profiles will be used to evaluate whether other strains of P. yezoensis have different patterns of dispersal and persistence than U-51, the initially licensed strain.

EDUCATIONAL IMPACT

The project will provide two years of support and training for 1.5 graduate students. It is intended that some of this work will provide the basis for their graduate theses. Coordinated field and molecular studies of natural and introduced seaweed species, represent new and important areas of research. The multi-instutional, multi-discipinary scope of the project will provide a unique

educational opportunity.

AVAILABLE RESOURCES

The proposed research will draw upon multi-institutional research facilities and individuals with diverse backgrounds dealing with the biology, biochemistry, ecology, genetics and aquaculture of seaweeds.

Personnel (also see attached BIOGRAPHICAL SKETCHES)

Anita S. Klein: She is molecular biologist in the Department of Biochemistry and Molecular Biology at UNH. Dr. Klein has applied molecular genetic techniques to population genetics of conifers and to examine, characterize and manipulate mutations in maize. She is also collaborating with a consortium of New England phycologists to measure the genetic and taxonomic diversity of Northwest Atlantic species of the red alga Porphyra, with a goal to adapt local species for successful aquaculture.

Arthur C. Mathieson: He has been involved with and published papers on a wide variety of ecological and aquaculture studies dealing with seaweeds (see attached C.V.). Dr. Mathieson also has extensive knowledge of the distribution and phenology of New England seaweeds that will be fundamental to the proposed study.

Christopher D. Neefus: He has a broad range of knowledge regarding the population genetics of seaweeds, including Gracilaria, Porphyra, and Laminaria based upon previous/current Sea Grant NSF funded projects. He is also part of UNH's Office of Biometrics and has consulted on research design and the use of appropriate statistical methods for data analysis for a wide range of biological and genetic studies.

Donald P. Cheney: He has broad range of knowledge and experience dealing with the taxonomy, ecology, and biotechnology of seaweeds. Currently he is involved with a variety of genetic manipulation studies of Porphyra and other economic seaweeds, as well as the biomonitoring of P. yezoensis.

FACILITIES, EQUIPMENT AND OTHER RESOURCES:

Hodgdon Herbarium (NHA): The Herbarium was recently moved to newly renovated space within the Biological Sciences Center (Spaulding Hall) at UNH. Long-term storage of critical materials is available, and herbarium exchanges can be made through its Director, Dr. Garrett Crow, a "higher plant" plant systematist in the Department of Plant Biology. Arthur Mathieson is the curator of the Algal Herbarium, which currently contains ~63,000 specimens. This invaluable resource, which is one of the largest in North America, has specimens from throughout the world, with particular emphasis on New

England. At this point we are cataloguing all algal specimens with a "user-friendly" database program, File-Maker. The program is supported by several computer units on campus, which means they provide both updated versions of the program, as well as periodic tutorials. Dr. Chris Neefus, an Associate Investigator on this project and part of UNH's Office of Biometrics, has played a major role in organizing and initiating this database program.

Jackson Estuarine Laboratory (JEL): The Laboratory, which is the home base for Arthur Mathieson, has equipment for microscopic, culture, and analytical investigations, including compound and dissecting scopes, growth chambers, a greenhouse facility, computers, freezers, ovens, balances, a Global Positioning System (GPS), plus other diverse field instruments. Much of the initial identification and processing of herbarium voucher specimens will be done at JEL. Voucher specimens will subsequently be transferred (deposited) to the Hodgdon Herbarium on campus.

Molecular Genetics Laboratory: Dr. Klein's molecular biology laboratory at UNH contains one programmable thermocycler equipped for microtiter PCR-Tm, multi-channel pipettors, and agarose electrophoresis apparatuses for large scale throughputs of DNAs for population studies using RAPD and RFLPs. Other equipment is available for standard DNA manipulations: incubators, ovens, a -80°C freezer, etc. Automated DNA sequencing, plus AFLP, RFLP, and DNA sequencing, are available through the UNH Sequencing Facility. Her laboratory has utilized RFLP and DNA sequencing methods for studies of plant population genetics and phylogenetics (Bobola et al., 1992a, b; Jianhua et al., 1997). She is currently engaged in molecular phylogenetic and population genetic research on diverse organisms, including North American Picea, Cupressaceae, Hamalidaceae, and Porphyra (see BIOGRAPHICAL SKETCHES). Dr. Thomas Kocher, a colleague in the Zoology Department at UNH, has successfully implemented AFLP technology and would be accessible for technical expertise.

Office of Biometrics: This Office, which is Dr. Neefus' primary home at UNH, is involved in a variety of consultation on research design, as well as the use of appropriate statistical methods for data analysis for a wide range of biological investigations (see BIOGRAPHICAL SKETCHES-CDN). The Office houses three network file servers that are the center of an extensive interdepartmental computer network serving ten departments of UNH's College of Life Science and Agriculture (COLSA). Dr Neefus is credited with the design and implementation of the network. The Office also maintains an HTML server that hosts a major portion of COLSA's Website. A portion of these facilities would be utilized for the development of the proposed project's information system. The Office and Dr. Neefus have also been responsible for the development of a wide range of network enabled computer software, including database systems, computer aided instruction applications, lab automation software, and automated species identification tools.

BUDGET JUSTIFICATION

The "lion" share of the budget is obviously committed to salaries as 1.5 students and four investigators will be involved with the project (ASK, ACM, CDN, & DC). Because of financial limitations one of the investigators (ACM) will not draw any salary, even though he will work a minimum of 1 months/year. Another individual (DPC) will receive ~0.3 month compensation/year. The cost-share from the three UNH P.I.s is significant, being ~$34,943 for the two year project. The graduate stipend will used to support aUNH student who will be primarily involved with DNA evaluations (see above). He/she will be supervised by A. S. Klein and A. C. Mathieson. An hourly support budget of $10,000/year will be used to support a half-time graduate student (i.e. from Northeastern University), plus aUNH work-study student. The former student would help with the extensive field and recruitment studies, while the latter would assist with the processing of extensive field samples and do some DNA assessments. Most of the travel funds will be needed for the acquisition of specimens, as monthly sampling will be conducted at a minimum of two field two sites (Eastport and Castine, Maine); $800 is put aside for professional travel during the second year of the project.With respect to equipment, we plan to purchase a new PCR machine during year one (~$4,500), as ASK's current unit would not survive the projected study. We will also purchase a "Newton" recording pad to aid with field documentations. The costs for the molecular analysis, including fees for the automated DNA sequencer, are obviously the most expensive supply items. Additional supply expenses will be associated with the acquisition of field, computer, and herbarium studies.

A subcontract will be established with Northeastern University, providing D. Cheney with some modest summer stipend (see above), plus partial support for a graduate student, and some field supplies. Most of this funding will be utilized for the field and recruitment studies outlined above. Similar Sea Grant subcontracts have been previously established with other New England institutions.

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ANNEX 5

1997 STATUS REPORT ON AUSTRALIA'S NATIONAL CENTRE FOR RESEARCH ON INTRODUCED MARINE PESTS (CRIMP)

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In 1994, the Australian Department of Industry, Science and Technology provided funds to the Commonwealth Scientific and Industrial Research Organisation (CSIRO), Australia's national science organisation, to establishment of a new, national Centre for Research on Introduced Marine Pests (hence, CRIMP). The principal arguments in favour of such a centre were, first, that it would identify and fund a single lead agency with primary responsibility for the problem of introduced marine pests and second, that the CSIRO had a long and successful involvement with management of introduced terrestrial pests, and hence a range of multi-disciplinary skills that could be brought to bear on the problem. Funding was about Australian $ 1.3 million/year, with an commitment explicitly of five years support and implicitly of indefinite support.

The objectives of the Centre are:

i) To develop and promote implementation of tools for earlier warning, better prediction, and more effective assessment of risks and costs of marine pest species introduced to Australia.

ii) To develop new methods or improve existing measures to control the spread and minimise the impacts of introduced marine species.

The amount of new funds provided dictated a staged approach to achieving these objectives. To determine priorities, CSIRO formally established a small advisory committee, consisting of key clients in government and industry, as well as several senior scientists from other organisations very familiar with the issues. Discussions with the committee led to a research plan that has three main thrusts: (i) an assessment of the scale of the problem in Australian waters; (ii) the development and evaluation of management options for existing pest species; and (iii) provision of advice and assistance to the Australian Ballast Water Management Advisory Committee's (ABWMAC) initiatives to minimise the risk of domestic translocation of exotic species and the rate of new introductions.

In the three years since its inception, CRIMP has expanded from two part-time researchers to twelve permanent staff (five Ph.D.s), seven term staff, nine students and, this last year, four long-term (more than five months) visiting international researchers. As well, CRIMP has or is supporting research at several Australian, American and European universities.

CRIMP is organised into three groups: Invasion Processes, Impacts and Demographics and Pest Management, as well as a small group managing the Centre (all of whom are involved in the research, as well). Current initiatives in each, and their status, are as follows:

Invasion Processes

i) Intensive survey of Port Phillip Bay (south-eastern Australia) - a detailed, multi-year, multi-investigator evaluation of introduced species in a major metropolitan embayment. In assessment and write-up stage, for delivery as a CRIMP Technical Report by the end of 1997.

ii) Targeted pest surveys of other Australian ports - a joint initiative between CRIMP, ABWMAC and the Australian port authorities, to obtain semi-quantitative information on the distribution among and within ports of a set of identified "pest" and exotic species. Planned to be national in coverage. Seven ports have been done to date, 5 more are planned for this year, and more are scheduled for following year. Coverage to date has been primarily temperate ports, but this year will move to tropical regions (Queensland) and thereafter to tropical and sub-tropical ports in Western Australia. This project is likely to lead to a national port monitoring program, currently being designed.

iii) Community-based coastal monitoring - a joint effort between CRIMP, the Australian Department of the Environment, ABWMAC and state fisheries agencies, to develop a national network of regional coordinators will link scientific and management agencies to local community groups for rapid detection of new introductions. Funding secured and position of national liaison officer being filled.

iv) Development of a general risk assessment framework for ballast water introductions - a joint CRIMP and ABWMAC initiative. We are developing a modular risk analysis framework that uses the port survey information, data on ballast tank configurations and management and biological data on pest species to calculate for each vessel and voyage the risk of introducing a new pest introductions. The risk assessment will be used to determine the level of management action required for each vessel. The basic model has been constructed and will be tested in coming year.

v) Development of robust methods for sampling ballast tanks - a joint CRIMP-ABWMAC initiative, which examines the distribution of planktonic organisms in ballast tanks, determines the accuracy and precision of different techniques for obtaining representative samples, and then tests the system against a target organism of known densities in the up-take water. On-board trials are underway. The project is scheduled for completion in late 1997.

As well, the Invasion Processes group is involved in a study (with BHP Transport) of the effectiveness of heat treatment as a means of 'sterilising' ballast water, a preliminary study of hull fouling as a transport vector across the Tasman Sea and studies on the ability of organisms to survive in ballast tanks. Major studies on the effects of port management techniques on colonisation success and the role of hull fouling (in general) as a transport vector are scheduled to start in 1997/98.

As part of this project, CRIMP has also produced a comprehensive guide (which is being up-dated annually) to the introduced marine species of Australia and hosts a marine pest list-server.

Impacts and demographics

i) Impact and population dynamics of the European crab, *Carcinus maenas* - Now in its second year, we are documenting the distribution and abundance of this nominated pest species in Australian waters, and undertaking experimental studies to determine its impacts on inshore communities. In the last year, six scientists were involved in this study on a near full-time basis. At the end of the austral summer field season, CRIMP hosted an international workshop on *Carcinus* impacts and management. Several publications on *Carcinus* impacts are in planning stages. Work on the project, which is aimed to underpin a control attempt, will continue over the next several years.

ii) Impacts and Spawning dynamics of the northern Pacific seastar, *Asterias amurensis* - Two Ph.D. students are undertaking studies on the impacts of this seastar in SE Australia, and assessing spawning behavior, fertilisation success and recruitment variability as a lead towards incorporating this information into a pest management program. Both students are now in the second year of their studies.

As well, CRIMP is supporting small scale studies on the impacts of the New Zealand screw shell, *Maoricolpus roseus* (which is the only introduced species we know of that has successfully spread to the open continental shelf) and the Japanese kelp, *Undaria pinnatifida*, and has completed impacts reviews of introduced marine pests for the ABWMAC.

Pest Management

CRIMP is still recruiting specialist staff to develop this project, for which we have just appointed an experienced project leader (Dr. Nic Bax). Current initiatives include

i) Evaluation of biocontrol options against the northern Pacific seastar, *Asterias amurensis* - Following an evaluation of natural pests on this seastar in its native range, we have commenced detailed work on the biology of the best prospect at this point - an endoparasitic ciliate, nominally in the genus *Orchitophrya*, that attacks, castrates and may kill male seastars. Two seasons of work has been undertaken on the ciliate in Japan, and collaborative studies with Canadian and European scientists are currently underway. We have also surveyed in detail the native Australian seastars and the introduced population of *A. amurensis* for parasites, as a baseline for a potential biocontrol initiative and to search for native species that might be effective against it.

ii) Development of pest management options against the European crab, *Carcinus maenas* - Preliminary studies are underway in Sweden looking at the host specificity of the parasitic castrator, *Sacculina carcini,* as a first step towards evaluating its potential as a biological control agent. Similar studies are planned for a range of potential control agents that have been identified for the crab at the CRIMP-sponsored workshop. We have also identified a range of native Australian parasites that infect the introduced crab, and are examining their potential as possible control options. A large scale attempt at physical removal, as a means of testing the effectiveness of a pest management program, is planned to commence in about two years time.

The *Carcinus* work, in general, is being done as a collaborative effort with scientists in Sweden, Denmark and the USA.

iii) Evaluation of physical removal and herbicidal treatments for control of the introduced Japanese alga, *Undaria pinnatifida* - One trial has been completed on application of herbicides to sporophytes (with limited success), a second project looking at a similar approach to gametophytes has been commissioned, and a trial local eradication using physical removal of sporophytes is underway.

As well, CRIMP has begun to examine the potential of transgenic techniques for controlling pest species (joint projects with other CSIRO Divisions). CRIMP also plans to host an international workshop on management of introduced marine pest populations, at this stage scheduled for early 1998.

ANNEX 6

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ANNEX 7

ACTION LIST

WGITMO identified the following as its current list of action items:

* Contact the Russian delegate over the proposed introduction of blue crabs (*Paralithodes platypus*) from the Kamtschatka regions to the Barents Sea, relative to potential ecological impacts and relative to standard ICES procedures for the evaluation of proposed new introductions.

* Contact the Spanish delegate for information on the current status of *Caulerpa taxifolia* in Spanish waters, particularly its spread in the Mediterranean.

* Determine the possibility of translating the "ICES Code of Practice on the Introduction and Transfer of Marine Organisms 1994" into Norwegian, Spanish, Portuguese and Welsh.

* Determine the possibility of placing the "ICES Code of Practice", or a reference to it with information on how to obtain the Code, on the ICES web page.

* Gather detailed information and data for the Handbook of Dispersal Vectors, and seek the input and cooperation of any interested EIFAC colleagues in doing so.

* Conduct a general survey to determine the extent to which the green alga *Caulerpa taxifolia* is being held in public and research aquaria in ICES Member Countries, under what temperature conditions, and the nature of effluent treatment, and also the extent to which this alga is available for purchase within the private aquarium hobby within ICES Member Countries.

* Continue to prepare historical risk assessment studies for selected case histories of introduced marine plants, invertebrates, and fish in ICES Member Countries.

ANNEX 8

RECOMMENDATIONS TO ICES COUNCIL

The following recommendations to the Advisory Committee on the Marine Environment (ACME) were formulated by the WGITMO:

1) WGITMO supports the recommendation of the ICES Study Group on Marine Biocontrol of Invasive Species (SGMBIS) that a permanent international "Working Group on The Control of Marine Pests" be formed, as initially proposed by GESAMP, which would advise and recommend strategies on the prevention and post-introduction research and control of introduced species. This Group could be formed by a possible consortium of GESAMP, ICES, and other international agencies.

2) WGITMO supports the recommendation of the joint ICES/IOC/IMO Study Group on Ballast Water and Sediments (SGBWS) that SGBWS convene for a second year to discuss research and management programmes on ballast water and sediments and other ship-mediated vectors with a view toward increased international cooperation and coordination.

3) WGITMO should conduct a general survey to determine the extent to which the green alga *Caulerpa taxifolia* is being held in public and research aquaria in ICES Member Countries, under what temperature conditions, and the nature of effluent treatment, and also the extent to which this alga is available for purchase within the private aquarium hobby within ICES Member Countries.

4) FAO/EIFAC should be contacted by ICES with the view of establishing a coordinated effort between EIFAC and ICES to harmonize the ICES 1994 Code of Practice on the Introduction and Transfer of Marine (and Freshwater) Organisms with the earlier EIFAC Code of Practice.

5) WGITMO recommends that a Theme Session, entitled "Marine Bioinvasions: Retrospectives for the 20th Century, Prospectives for the 21st Century", be convened for the 1999 or 2000 ICES Annual Science Meeting, to gain an overview of the issues that have engaged ICES Member Countries on the introductions and transfers of marine organisms for the past 20 years, since the first reconvened meeting of the ICES Working Group on Introductions and Transfers of Marine Organisms (1979), and to gain insights into the challenges that face ICES Member Countries in the opening decades of the 21st century.

6) The Working Group on Introductions and Transfers of Marine Organisms (Chair: Dr J.T. Carlton, USA) will meet in The Hague, Netherlands, from 1–3 April, 1998 to:

   a) make detailed plans for the Theme Session on "Marine Bioinvasions: Retrospectives for the 20th Century, Prospectives for the 21st Century" for the 1999 or 2000 ICES Annual Science Conference;

   b) pursue progress on the harmonization of the joint EIFAC-ICES Codes of Practice, through an invitation to the Chairman of the EIFAC Working Party on Introductions to meet with WGITMO;

   c) continue the assessment of the potential marine biocontrol activities through review of current proposed programmes in the ICES area, and through the invitation to the Marine Biocontrol Risk Assessment Specialist of the Australian CSIRO/CRIMP to meet with WGITMO,

   d) review the findings of the Caulerpa Aquarium Review conducted in 1997-1998 by WGITMO members, relative to the status of the importation, holding, and disposal of *Caulerpa* in waters of ICES Member Countries,

   e) continue to prepare historical risk assessment studies for selected case histories of introduced marine plants, invertebrates, and fish in ICES Member Countries, in order to understand the ecological and other environmental effects of commercially used exotic species introductions into ICES Member Countries, so that the types of risk can be identified retrospectively and thus be used as a basis for aiding future management decisions;

   f) continue work on a "Directory of Vectors Involved in the Introduction and Transfer of Marine and Estuarine Organisms", which work reviews the diversity, nature, and specific roles of vectors that are important in the

transportation of exotic marine and brackish water organisms, in order to provide decision-makers with ready access to data sources that may aid in risk assessments, when specific vectors are proposed or come into play that have previously not existed or have not yet been recognized in individual ICES Member Countries;

g) report on the current status of fish, shellfish, algal, and other introductions in and between ICES Member Countries, through:

   i) submission of the National Reports,

   ii) updates on the issues of transport of exotic organisms through ships' ballast water and sediments, and other ship-associated vectors, through information provided by the joint ICES/IOC/IMO Study Group on Ballast Water and Sediments,

   iii) review of the status of the current and projected distribution, ecological impacts and commercial use in ICES Member Countries of introduced marine and brackish water organisms, such as the marine seaweeds *Porphyra yezoensis*, *Undaria pinnatifida*, *Sargassum muticum*, and *Caulerpa taxifolia*, and the marine invertebrates *Marenzelleria viridis*, *Hemigrapsus penicillatus,* and *Dreissena polymorpha*,

   iv) review of the status of research on genetically modified organisms (GMOs) in ICES Member Countries, with particular attention to the current status of transgenic salmon in aquaculture,

   v) continued coordination of cooperative databases on introductions and transfers of marine and brackish water organisms,

   vi) continued communication with and cooperation with the Baltic Marine Biologists Working Group on Nonindigenous Estuarine and Marine Organisms, and

   vii) develop a questionnaire and expanded terms of reference for a more general review of the non-native temperate marine and brackish-water organisms being held in public and research aquaria in ICES Member Countries, under what temperature conditions, and the nature of effluent treatment, and also the extent to which non-native temperate animals and plants are available for purchase within the private aquarium hobby within ICES Member Countries.