EPIZOOTIC INVESTIGATION INTO THE DETECTION OF BONAMIOSIS IN NATIVE OYSTERS (*OSTREA EDULIS*) IN A NEW AREA IN IRELAND DURING AUTUMN 2006

Cheslett, D.L.,[1] McKiernan, F.,[1] Hickey, C.,[1] Collins, E.,[1] Geoghegan, F.[1]

1. Marine Institute, Rinville, Oranmore, Co. Galway, Ireland

Contents

SUMMARY

The Republic of Ireland obtained Approved Zone status for the List II (91/67/EEC) shellfish diseases bonamiosis (*Bonamia ostreae*) and marteiliosis (*Marteilia refringens*) under Commission Decision 2002/300/EC in 2002. At that time approval was given to the entire coastline for marteiliosis, and for the entire coastline except Clew Bay, Galway Bay, Cork Harbour and Ballinakill Harbour for bonamiosis. Between 1993 and 2002 bonamiosis had remained confined to these areas. However in 2002 bonamiosis was detected in a new area (Achill Sound). Shortly after, in 2003, the disease was discovered in the nearby oyster beds of Blacksod Bay. The ensuing epizootic investigation found the most likely source of infection to have been either the movement of boats between Achill, Blacksod Bay and Clew Bay or movements authorised before it was known the disease was present in other parts of the country (22).

In the spring of 2005 *Bonamia ostreae* was again detected in a new area, this time in Lough Foyle. Lough Foyle is situated in a cross border catchment area, with part of the lough falling in Donegal in the Republic of Ireland and the remainder in County Derry in Northern Ireland. A joint epizootic investigation involving representatives from both sides of the border was undertaken. The most likely source of infection was identified as an unauthorised movement of mussels from Clew Bay to Lough Foyle. It was believed that a number of oyster spat may have been attached to the mussels (25).

The nearest native oyster fishery to Lough Foyle is in Lough Swilly in County Donegal. The two loughs are in close proximity, with less than eight miles separating them at their closest point. Movements of shellfish had had taken place between the two sites prior to the identification of bonamiosis in Lough Foyle, following which a program of increased surveillance was put in place in Lough Swilly.

In both the spring and autumn of 2006, samples of 150 oysters were taken from Lough Swilly and submitted to the Marine Institute for screening under Directive 91/67/EEC. The sample submitted in the autumn originated from two oyster beds and was split into

two samples of 75. One of these samples tested positive for *Bonamia ostreae* at a prevalence of 17.3%.

As a result of the positive finding, all movements of shellfish out of the Lough were immediately suspended by the Irish authorities, and an epidemiological investigation was initiated under Article 5 of Council Directive 95/70/EC. The EU Commission and the OIE were informed, and Lough Swilly lost its approved zone status for bonamiosis under Commission Decision 2007/104/EC.

The details and findings of the epidemiological investigation are summarised in this report. Further monitoring of the development and geographical spread of *Bonamia ostreae* within Lough Swilly is ongoing.

1. INTRODUCTION

1.1 Bonamiosis

Bonamiosis is a disease caused by a number of haplosporidea including *Bonamia ostreae, B. exitiosa* and *B. roughleyi* (Formerly *Mikrocsytos roughleyi)* (1-7). *Bonamia ostreae* is found in the Northern Hemisphere whilst *Bonamia exitiosa* and *Bonamia roughleyi* are found in the Southern Hemisphere. The disease is also known as micro cell disease, haemocyte disease of flat oysters (*B. ostreae*), haemocyte disease of dredge oysters (*Bonamia exitiosa*) or winter mortality (*B. roughleyi*).

Bonamia ostreae naturally occurs in *Ostrea edulis* and *O. conchaphila* (= *O. lurida*), and has been found in *O. puelchana, O. angasi, Tiostrea chilensis* (= *Tiostrea lutaria*), and *Crassostrea rivularis* (18) when they are moved into *Bonamia ostreae* endemic zones. This protistan parasite infects the haemocytes of the oyster, where it can quickly become systemic, with overwhelming numbers of parasites coinciding with the death of the oysters.

Bonamiosis first appeared in Europe in France in 1979 (7, 11) and has since spread to most *Ostreae edulis* cultivating areas in Europe including the Netherlands in 1980 (12), the United Kingdom in 1982 (13), Spain in 1984 (14), and Ireland in 1987 (15). It has been responsible for considerable losses amongst populations of native, flat oysters, and has, in conjunction with the parasite *Marteilia refringens,* been in part responsible for the decline of the industry in a number of countries.

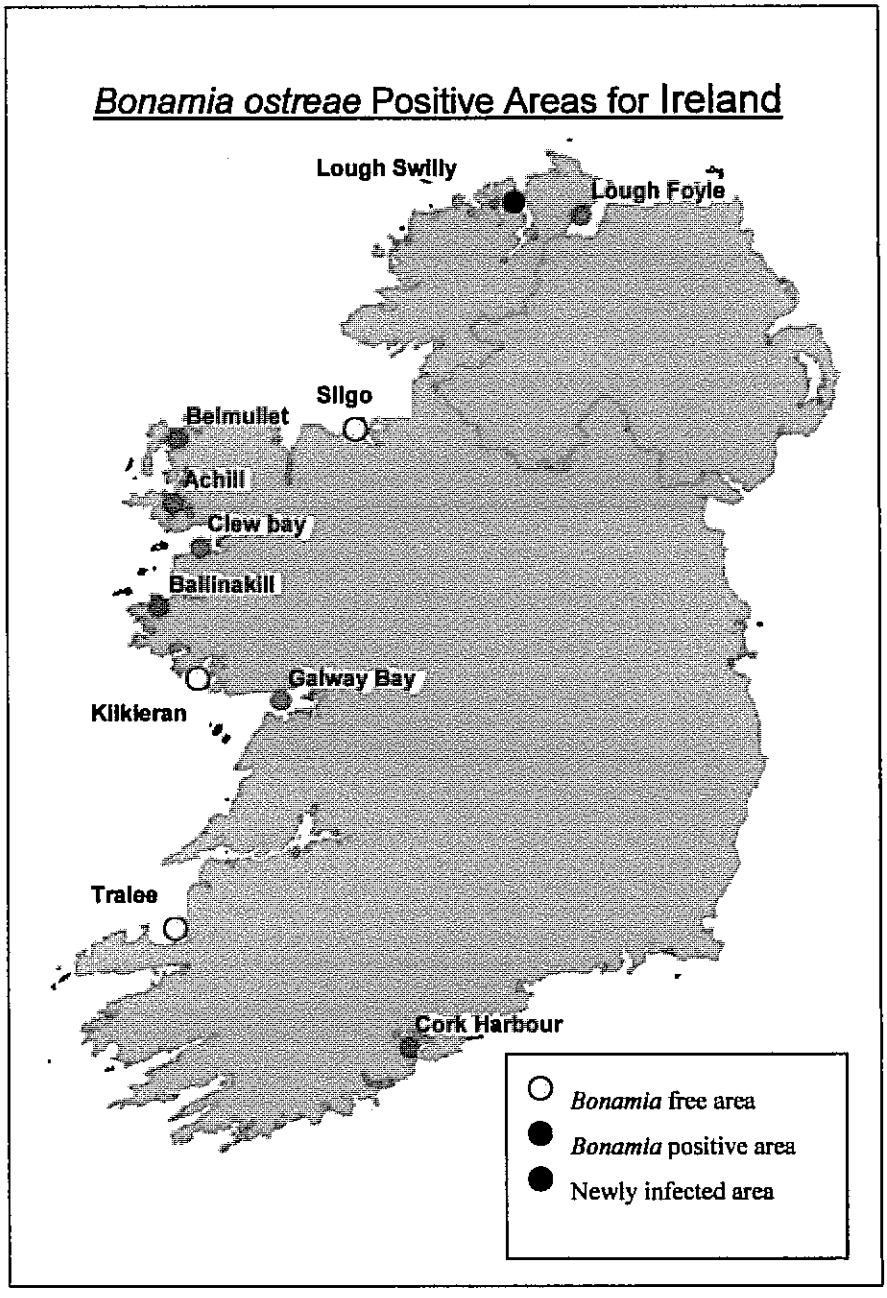
The spread of the disease has been attributed to movements of native oysters. Once it has been introduced it has generally proved impossible to eradicate. Even in areas left fallow for years the disease reappeared when oysters were reintroduced (16, 17). An exception to this was observed in Limfjorden in Denmark where the parasite was detected in a small population of *O. edulis*. It was believed the oysters had been recently introduced, and so they were immediately removed from the water. The parasite has not been detected there since. Similarly the Scottish authorities are currently attempting to

eradicate *Bonamia ostreae* from Loch Sunart, following its detection there last year in a small population of oysters which had been introduced into the loch. As there are no wild populations of native, flat oysters in the Loch, it is hoped that, by removing all the oysters that were introduced, it may be possible to eradicate the disease..

Methods for detection of the parasite are described in the Office Internationale des Epizooties (OIE) manual for diagnostic tests. These include heart smears, histology, PCR and *in situ* hybridisation (ISH). (33)

1.2 Bonamiosis in the island of Ireland

Figure 1: Location of the *Bonamia ostreae* positive areas in Ireland



Bonamia ostreae was first detected in Ireland in Cork Harbour in 1987, in a site where oysters were being intensively cultured. In the last 20 years the parasite has gradually spread to the majority of wild, native oyster fisheries within the country, a total of eight bays are now positive for the parasite. Figure 1, shows the bays where there are wild and cultured native, flat oyster fisheries and their status with regard to bonamiosis.

The first area affected by bonamiosis was Cork Harbour in 1987. There were high mortalities on site, particularly in the older age groups. In the 4 year old oysters, for instance, mortalities reached levels of around 90% (19). It is believed that the parasite had been introduced earlier than this and could have been contributing to mortalities that had begun at the site a number of years before (20). The source of the infection proved impossible to determine.

In 1988, bonamiosis was detected in a wild oyster fishery in Clew Bay. The parasite was identified in a consignment of oysters exported to France. No mortalities had been observed in Clew Bay, and it was a number of years before the infection was confirmed at a low prevalence. In Clew Bay there are a number of beds with low stocking densities, and up to present times no major mortalities have been reported.

In 1989, *B. ostreae* was detected in Galway Bay in association with high mortalities (70-80% in one bed)(21).

In 1993, *B. ostreae* was detected in two out of four samples collected from Ballinakill Harbour. The samples had been taken, as part of the testing regime to obtain Approved Zone status and it is not known when, or how, the oysters became infected.

The next new occurrence of the disease was in 2002, when it was identified during routine testing in Achill Sound. Following the identification of the parasite in Achill Sound, surveillance of stocks in the nearby beds of Blacksod Bay was increased, and in the spring of 2003, *B. ostreae* was also detected in one of these beds in Loughmore, Blacksod Bay (22).

In the spring of 2005, *B. ostreae* was detected in Lough Foyle. This resulted in increased surveillance of oysters in the nearby Lough Swilly where the parasite was identified in Autumn 2006.

1.3 Statutory Testing & Movements

1.3.1 EU Legislation

Statutory testing is carried out under Directive 91/67/EEC, which governs the placing on the market of live aquaculture animals. bonamiosis (*Bonamia ostreae*) and marteiliosis (*Marteilia refringens*) are listed in Annex I (List II) of the Directive, and it is on the basis of freedom from these diseases, that movements are controlled within the Community. Movements of susceptible species of molluscs are permitted only between areas of equal health status, or from areas of higher health status to areas of lower health status.

Under Article 14 of Directive 91/67/EEC all aquaculture mollusc species were considered as possible vectors of bonamiosis or marteiliosis until otherwise demonstrated. However, this had a huge potential impact on trade and resulted in the publication of Commission Decision 93/54/EEC. This amended Article 14 to allow movements of mollusc species from non-approved to approved zones:

"where practical experience and/or scientific evidence has shown that there is no passive transmission of the disease when aquaculture animals, their eggs and gametes, not belonging to the susceptible species referred to in Annex A, column 2, list II, are moved from a non-approved zone to an approved zone."

Decision 93/169/EC lists *Crassostrea gigas* as not susceptible to either *B. ostreae* or *M. refringens.* In 2003, Commission Decision 2003/390/EC, establishing special conditions for placing on the market of aquaculture animal species extended the list of species not considered susceptible to *B. ostreae* to include *Crassostrea gigas*, *Mytilus edulis* and

Mytilus galloprovinciallis, *Ruditapes decussatus* and *Ruditapes philippinarum*. This Decision takes into account scientific evidence (29, 30) and recommendations from the OIE. The possible vector status of other aquaculture species (*P. maximus*, *Haliotis sp.*) is not known and they are therefore still considered to be susceptible species.

1.3.2 Legislation and Testing in Ireland

The legal framework in Ireland is provided through a number of Statutory Instruments (S.I.s).

Council Directive 91/67/EC is transposed in Irish law by the European Communities (Aquaculture Animals and Fish) (Placing on the Market and Control of Certain Diseases) Regulations 1996, S.I. No. 253 of 1996. This S.I. provides the mechanism for restricting movements, from non-approved zones into approved zones or those undergoing testing programmes. It was amended by S.I. No 614 of 2006, the European Communities (Aquaculture Animals and Fish) (Placing on the Market and Control of Certain Diseases) (Amendment) Regulations of 2006, which added Lough Swilly to the list of zones to which additional movement restrictions apply.

Council Directive 95/70/EC is transposed into Irish law by the European Communities (Minimum Measures for the Control of Certain Diseases affecting Bivalve Molluscs) Regulations 1999, S.I. No. 26 of 1999.

Under the Molluscan Shellfish (Conservation of Stocks) Regulations 2006, S.I. No. 345 of 2006, the dredging, fishing for, or relaying of shellfish may only take place under licence, issued by the Minister. (It may be noted that this S.I. has replaced the previous instrument, the Molluscan Shellfish (Conservation of Stocks) Order 1987, S.I. No. 118 of 1987, following the passing of the new Sea-Fisheries & Maritime Jurisdiction Act in 2006).

The Department of Communications, Marine & Natural Resources (DCMNR) is the Competent Authority in Ireland with responsibility for controlling movements of shellfish within, into and out of the country.

Movements of shellfish for the purposes of on growing, must be authorised by the DCMNR, to ensure compliance with fish health legislation, and where applicable, for the management and conservation of wild stocks.

The Marine Institute is the designated National Reference Laboratory for Molluscan Diseases. Testing is carried out for bonamiosis and marteiliosis in line with Commission Decision 2002/878/EC. Cytological examination of heart imprints is the primary method used for the detection of *B. ostreae*, and the scoring system used to assess the intensity of infection, is based on that described by Bachere *et al.*, 1982 (23). Histological examination, PCR and ISH (24) are used, as required, to aid in confirmation of the presence of the disease.

1.3.3 Approved Zone Status for Bonamiosis

The Irish programme, to obtain Approved Zone status, was submitted to the EU Commission in 1993, and was approved by Commission Decision 93/56/EEC. The application requesting approved zone status, detailing the testing carried out, was submitted to the Commission in 2001 and approval was granted under Commission Decision 2002/300/EC. Under this Decision, approval was given for bonamiosis for the entire coastline except Cork Harbour, Galway Bay, Clew Bay and Ballinakill Harbour. Approved status was subsequently lost for Achill Sound in 2002 (Commission Decision 2003/378/EC) and for Loughmore, Blacksod in 2003 (Commission Decision 2003/729/EC) and again for Lough Foyle in Spring 2005 (Commission Decision 2005/748/EC). Following the detection of bonamiosis in Lough Swilly in Autumn 2006 the EU Commission were notified, and approved zone status was lost under Commission Decision 2007/104/EC.

2. Detection of *Bonamia ostreae* in Lough Swilly

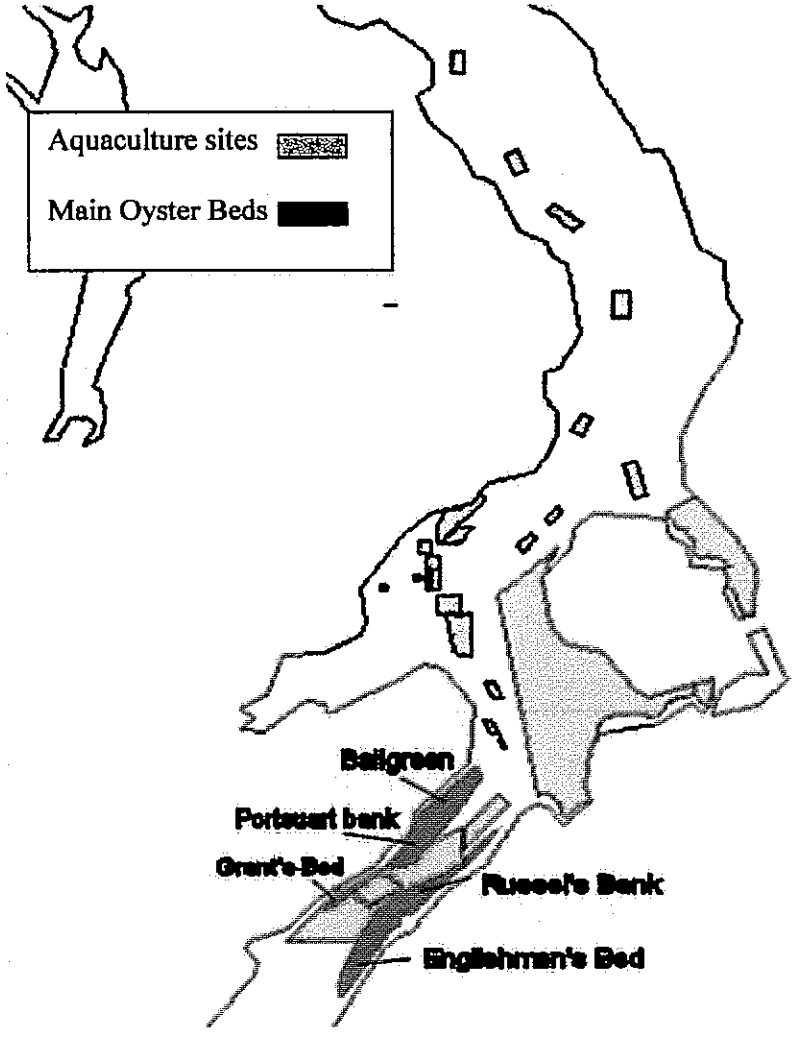
2.1 Lough Swilly

Lough Swilly originated as a flooded river valley and stretches 40 kilometers into County Donegal from the Atlantic Ocean. It is surrounded on the east by the Inishowen Peninsula and on the west by the Fanad Peninsula. It is accessible by boat traffic at all stages of the tide, as far upstream as Letterkenny, with depths ranging from 15 to 50m at the deepest points, and tidal changes between 1.4 and 3.7m.

The main wild mollusc fisheries within the Lough are blue mussels (*Mytilus edulis*) and native oysters (*Ostrea edulis*). However the majority of the mussels grown in Lough Swilly nowadays are cultivated in licensed plots. A small quantity of *C. gigas* is also cultivated.

The existence of an oyster fishery in the lough has been recorded for centuries. In a report into incidents of food poisoning in shellfish in 1904 Went mentioned the existence of the fishery at the early part of the 17th century (26). More recently a survey carried out by Bord Iascaigh Mhara (BIM) in 2001 attempted to define the discrete areas that comprise the flat oyster fishery within the lough. Similarly surveys carried out by BIM have attempted to define the mussel resource areas. Figure 2 shows the principal oyster beds fished and the aquaculture sites in Lough Swilly. The aquaculture sites are predominantly mussel sites.

Figure 2. Map of aquaculture sites / oyster beds



2.2. Testing for *B. ostreae* in Lough Swilly

Until 2001 the Marine Institute was unable to obtain samples of *O. edulis* from Lough Swilly, as the fishery was not extensively commercially exploited. In 2001, the Marine Institute (MI) commenced testing 150 oysters twice a year to ensure that Lough Swilly was free of the disease. The results of this testing and all subsequent testing up until the detection of Bonamia are summarised in table 1.

Table 1: Results of testing, for *B. ostreae,* carried out by the MI in Lough Swilly since 2001

YEAR	SEASON	NO OF OYSTERS	*BONAMIA* RESULT	YEAR	SEASON	NO OF OYSTERS	*BONAMIA* RESULT
2001	Spring	Not available		2004	Spring	30, 30	Negative
	Autumn	150	Negative		Autumn	60	Negative
2002	Spring	150	Negative	2005	Spring	30, 30	Negative
	Autumn	150	Negative		Autumn	73, 78	Negative
2003	Spring	150	Negative	2006	Spring	75, 75	Negative
	Autumn	30, 30	Negative		Autumn	75*, 75**	* 13/75 positive ** Negative

Following the detection of *B. ostreae* in Lough Foyle, the risk of neighbouring Lough Swilly becoming infected was deemed to have increased. As a result, increased sampling, at a level of 150 oysters twice annually, was undertaken, from the autumn of 2005 onwards. This initial sample and the sample taken the following spring both tested negative, but the sample taken in the autumn of 2006 tested positive. In each case the sample of 150 was made up of two samples of 75 from two different areas within Lough Swilly.

The principal oyster beds fished in Lough Swilly over the years, were Ballgreen and Russels Bank, and hence testing has focused on these two beds. A sample of 75 oysters was submitted from each of these beds in the autumn of 2006. One sample of 75 was found to be positive for *B. ostreae*. In follow up to the detection of *B. ostreae* in the sample from Ballgreen additional samples were collected from these two beds, as well, as from a third bed, Portstuart Bank. Further samples were also collected in the spring of 2007. All samples were screened using cellular imprints and the results are summarised below in table 2.

Table 2: *B. ostreae* in Lough Swilly: autumn 2006 – spring 2007 tests showing levels and prevalence of infection

Date Received	Bed Sampled	No. of Oysters	No. Positive by imprint	Prevalence of infection	Level of infection**
5/10/06	Ballgreen / Russel's Bank	75	13	17.3%	Level 1 – 30.8% Level 2 – 53.8% Level 3 – 7.7% Level 4 - 7.7%
5/10/06	Ballgreen / Russel's Bank	75	0	0%	0%
17/11/06	Russel's Bank	64	0	0%	0%
17/11/06	Portstuart Bank	63	6	9.1%	Level 1 – 83.3% Level 4 - 16.7%
30/11/06	Ballgreen	75	10	13.3%	Level 1 – 80% Level 2 – 20%
13/4/07	Russel's Bank	50	0	0%	0%
13/4/07	Ballgreen	50	9	18%	Level 1 – 44.4% Level 2 – 44.4% Level 4 – 11.1%
13/4/07	Englishman's Bed	50	0	0/%	0%

Two oyster beds have thus far been identified as positive. Subsequent testing will aim to investigate other beds within the Lough, with the objective of building a comprehensive picture of the distribution of the disease within the Lough, as well as monitoring its spread over time.

2.3. Measures taken by the Irish Authorities

2.3.1 Notification & Dissemination of Information

The Aquaculture Policy Section of the Irish Department of Communications, Marine and Natural Resources (DCMNR) informed the European Commission of the suspicion of bonamiosis in Lough Swilly on the 9th November 2006. Following confirmation of the

presence of *Bonamia ostreae* by the CRL the EU Commission were notified by the DCMNR of these definitive findings on the 27/11/06. The OIE were notified of the finding on the 29/11/06.

2.3.2 Movement restrictions

Shellfish movement restrictions were put in place in Lough Swilly following suspicion of the presence of bonamiosis. These restrictions have been maintained following confirmation of the presence of the parasite, and legislation has been introduced by the DCMNR to implement the provisions of Commission Decision 2007/104/EC (See 1.3.2 above)

3. EPIZOOTIC INVESTIGATION

Under Article 5 of Directive 95/70/EC, if one of the pathogens specified in Annex A, list II of 91/67/EC is confirmed in an approved area then an epizootic investigation must be carried out. The purpose of this investigation is:

- to determine the possible means of contamination of the new area
- to determine whether molluscs have left the area for reimmersion elsewhere in the period preceding detection of the pathogen.

Following confirmation of the presence of *Bonamia ostreae* in Lough Swilly an epizootic investigation was therefore carried out.

3.1 Investigation into the source of infection in Lough Swilly

The Investigation was composed of two parts. The first part focused on the examination of testing and movement records, held by the DCMNR and Marine Institute, in relation to Lough Swilly. The second part of the investigation involved a series of meetings and

interviews, both in person and by phone, with a representative section of the shellfish industry based in Lough Swilly. The interviews were based on a questionnaire, that was put together, to investigate the possible sources of infection for Lough Swilly. In addition some of these questionnaires were also completed and returned to the Marine Institute anonymously.

3.1.1. Testing for *B. ostreae* in Lough Swilly

Table 1 on page 12 summarises the testing carried out in Lough Swilly from 2001-2006. Testing had focused on the Ballgreen and Russel's Bank beds. These results indicate that there was no evidence of the parasite within the lough until the end of 2006.

3.1.2. Authorised Shellfish Movements into Lough Swilly

Movement documents were examined for a ten-year period from 1996 to 2006 preceding the detection of bonamiosis in Lough Swilly. This information is summarised in Tables 3 and 4. The aim was to investigate the likelihood that an authorised movement could have been responsible for the introduction of *B. ostreae* into the Lough.

Table 3: Authorised shellfish movements into Lough Swilly from other sites within Ireland, 1996 - 2006

Year	Source	Species	Quantity
1996	Irish Sea	*M. edulis*	2000T
1997	Irish Sea	*M. edulis*	3000T
1998	Irish Sea	*M. edulis*	Up to 1000T
2000	Irish Sea	*M. edulis*	Up to 1800T
2000	Irish Sea	*M. edulis*	Up to 2000T
2000	Irish Sea	*M. edulis*	Up to 750T
2001	Irish Sea	*M. edulis*	500T
2001	Irish Sea	*M. edulis*	500T
2002	Woodstown Strand, Co Waterford	*C. gigas*	150,000 oysters
2003	Woodstown Strand, Co Waterford	*C. gigas*	2T
2003	Lough Foyle, Co Donegal	*M. edulis*	1928T
2003	Woodstown Strand, Co Waterford	*C. gigas*	3.5T
2004	Lough Foyle, Co Donegal	*M. edulis*	60T

2005	Bantry Bay, Co Cork	*M. edulis*	100T
2005	Bantry Bay, Co Cork	*M. edulis*	200T
2005	Bantry Bay, Co Cork	*M. edulis*	200T
2006	Bantry Bay, Co Cork	*M. edulis*	200T

Table 4: Authorised shellfish movements into Lough Swilly from other EU countries, 1996-2006

Year	Source	No of consignments	Species	Quantity (Individuals)
1996	Guernsey	6	*C. gigas*	954,000
1996	Cumbria, England	1	*C. gigas*	50,000
1997	Guernsey	2	*C. gigas*	500,000
1998	Guernsey	1	*C. gigas*	500,000
1999	Guernsey	2	*C. gigas*	600,000
2000	Guernsey	3	*C. gigas*	1,110,000
2000	Cumbria, England	1	*C. gigas*	300,000
2001	Guernsey	2	*C. gigas*	2,250,000
2002	Guernsey	1	*C. gigas*	500,000
2002	Cumbria, England	4	*C. gigas*	3,000,000
2003	Cumbria, England	7	*C. gigas*	4,000,000
2003	Guernsey	3	*C. gigas*	565,000
2004	Cumbria, England	3	*C. gigas*	1,600,000
2005	Cumbria, England	8	*C. gigas*	3,081,000
2005	Zone VII / VIII, France	3	*C. gigas*	3,500,000
2005	Guernsey	1	*C. gigas*	20,000
2005	Zone VIII, France	1	*C. gigas*	150,000

Over the last 10 years there have been no authorised movements of *O. edulis* into Lough Swilly. Movements have taken place, to supply the aquaculture sites within the lough with *M. edulis* and *C. gigas.* Under current legislation neither of these species is considered to be susceptible to, or responsible for, the transmission of bonamiosis, and hence movements of either species are permitted from non-approved to approved zones.

Crassostreae gigas

All except four consignments of *C. gigas* originated from approved zones, and hence the risk that *B. ostreae* was introduced with these transfers, is negligible. Four consignments

originated from areas in France, where *B. ostreae* is known to occur. Commission Decision 93/169/EC allows for the movement of *Crassostrea gigas* from non-approved to approved zones for *Bonamia ostreae* on the basis that there was scientific and practical evidence that they could not passively transmit the disease. Studies carried out subsequently (29, 30) using histology and cellular imprints to detect the parasite, also aimed to demonstrate whether *C. gigas* could act as a carrier of the disease. Both studies concluded that *C. gigas* could neither develop the disease nor transmit the parasite to naïve oysters, after exposure of the *C. gigas* to *B. ostreae*. However, as these conclusions were based on methods that allowed for detection of a conventional form of the parasite, the authors cautioned that the presence of an unrecognised form of the parasite could not be ruled out. Molecular based methodologies are now available for the detection of *B. ostreae* that appear to be more sensitive than traditional methods, as well as allowing for the detection of forms of the parasite which may differ from that observed in *Ostreae edulis*. A number of studies are now underway to address this issue such as Marine Institute funded Project ST/05/25. This project, currently underway in UCC, aims investigate the ability of a number of commercial species, including *C. gigas* to act as carriers of the disease, using molecular methodologies in conjunction with the traditional histology and cytopathology methods to detect the parasite. Results presented at the 13th International conference of the EAFP by both the UCC research group and by a research group from the Centro de Investigacions Marinas in Spain have demonstrated the presence of *B. ostreae* DNA in tissue samples taken from *C. gigas* that were exposed to *O. edulis* infected with *B. ostreae*. The UCC trial also aims to investigate whether these exposed *C. gigas* can transfer the parasite back to *O. edulis*. If *C. gigas* are able to harbour the parasite and transfer it back to *O. edulis* then the movement of *C. gigas* from non-approved zones may pose a risk for the transmission of *B. ostreae*.

Mytilus edulis

The latent period, between introduction of bonamiosis into an area and detection of the parasite in oysters, is thought to be at least 3 months (28, 34-37). A number of consignments of mussels have been moved into Lough Swilly from Lough Foyle. Although Lough Foyle was approved for bonamiosis at the time that these movements,

took place, it is possible that Lough Foyle had already become infected before some of these movements occurred. Mussels have been shown in previous studies not to be susceptible to or responsible for the transmission of the disease. These studies, as with those for *C. gigas*, relied on histology and cellular imprints so with the advent of new, more sensitive molecular methodologies, subsequent studies may find that this is not the case.

Hitchhiker species

Movements of shellfish may also present a risk for disease transfer through the presence of other hitchhiker species. Bivalve species have been found attached to other shellfish such as *O. edulis*. In the recent investigation into the outbreak of bonamiosis in Lough Foyle, it was concluded that the most likely source of infection had been the unauthorised movement of a consignment of *M. edulis* from a *B. ostreae* positive area to Lough Foyle with *O. edulis* spat attached (25). In Lough Foyle, the oyster and mussel beds are found in close proximity, and in some areas the beds are overlapping. Oyster spat will readily settle on and attach to the shells of other species and so it is not surprising that in a recent survey of the lough carried out by CEFAS oyster spat was noted on the shells of mussels. If these mussels are dredged and relayed the oyster spat will be moved along with the mussels. The potential for introduction of bonamiosis through this route cannot be ignored. Even rigorous pre-movement inspections cannot be guaranteed to detect all "hitchhiker" species in a consignment.

Recent studies carried out at UCC have indicated that there may be a role for benthic macro invertebrates and zooplankton in the life cycle of the parasite (31). Potentially these species could also be passively transferred with consignments of shellfish.

3.2. Questionnaire to Industry

The main shellfish industries within Lough Swilly include culture of *M. edulis* and *C. gigas*, as well as exploitation of the wild, native oyster fishery by fishermen. A

questionnaire was designed which aimed to target those involved in these industries, in an attempt to determine how the lough became infected. The questionnaire was administered to aquaculture operators and fishermen through a series of formal and informal meetings, telephone interviews, and by post, in the early months of 2007. Six mussel growers, one *C. gigas* grower and seventeen native oyster fishermen were consulted in this survey. This was deemed to be a representative cross section of those involved in the shellfish industry as it covered the majority of mussel growers, the only *C. gigas* grower active at the time and the majority of oyster fishermen active at the time the survey was administered. Questions focused on the following areas:

- Unauthorised movements of shellfish into the Lough for on-growing
- Movements of shellfish prior to sale for human consumption
- Movements of boats / equipment / cultch

A number of those involved in sales and distribution of oysters were also questioned in follow up to the survey of shellfish producers and fishermen.

A copy of the questionnaire is included in Appendix A.

3.2.1 Unauthorised movements of shellfish for on-growing

Movements of shellfish into and out of Lough Swilly are subject to authorisation by the DCMNR. However, the possibility that unauthorised movements had taken place could not be ruled out. The questionnaire included questions, aimed at identifying whether additional movements of shellfish had taken place, which did not appear on the official records. In some instances, operators declared movements, which they had carried out themselves, which did not appear on the official statistics. In other instances movements that had been carried out by other companies, were cited. If a company was named as having moved stock without a permit, confirmation was sought from that operator

3.2.1.1 *Ostrea edulis*

The fishery in Lough Swilly is essentially a wild fishery, exploited by a fishing Co-operative, as well as by a number of handpickers who remove oysters from the shore. Three movements, which were not authorised, and so do not appear on the official statistics, were identified. The Oyster Co-op, in conjunction with a mussel operator brought *O. edulis* seed into Lough Swilly in 2001. This consignment originated from an approved zone, in County Sligo, that continues to remain free of the disease. This consignment is therefore not likely to have harboured the parasite. Additionally, native, flat oysters were brought in to an aquaculture site for on growing in the 90s. They originated from the Limfjorden in Denmark as well as from Tralee Bay in the south west of Ireland. Both these sites are approved and again these consignments are not likely to have been responsible for the introduction of the disease to Lough Swilly.

3.2.1.2. *Mytilus edulis*

Traditionally, mussel seed has been sourced from areas of natural spat settlement within Lough Swilly. In recent years, however, as mussel aquaculture increased within the Lough additional seed has also been sought from the Irish Sea. This is an approved zone for bonamiosis and hence the risk from these movements is negligible, even with the potential for hitchhiker species.

One mussel operator admitted to moving stocks of mussels between bays, including Lough Swilly, Belfast Lough and Lough Foyle. Some of the movements appear on the official records (See section 2.5.2 above), but from the interviews, held with both mussel and oyster fishermen, it seems likely that there have been more movements than appear on the records. Movements from Lough Foyle, which lost its approved zone status in 2005, could have posed a threat of introduction of the disease.

3.2.2 Movements of shellfish prior to sale for human consumption

Once fished oysters are often held in bags on the shore until the fishermen have sufficient to sell. The majority of fishermen fishing for oysters in Lough Swilly, fish lough Swilly only, therefore all of the oysters that they hold originate in the Swilly. There have been

three oyster fishermen who have fished in Lough Swilly as well as else where, but they did not hold oysters from other areas in Lough Swilly.

One incidence of a movement of winkles into Lough Swilly was cited about 3 yrs ago though who brought these in or where they originated from could not be established. One of the oystermen surveyed stated that a consignment of winkles had been brought in to Lough Swilly and reimmersed about three years previously. He was not, however, able to give any further details on where these had originated from or who had brought them in, it was therefore not possible to establish the significance of this movement. No data has been published with regard to the ability of winkles to transmit bonamiosis, and without any further information it is not possible to conclude what if any risk was posed by this movement.

Through the administration of the questionnaires two practices came to light that could have put Lough Swilly at an increased risk in relation to the potential introduction of disease. The first involves the return of undersize oysters, and the second relates to the collection of oysters for sale.

1. A number of fishermen sell oysters to a buyer located on the shores of Lough Foyle. Oysters from Lough Foyle are also sold to this same buyer. Oysters are graded on site, and prior to the detection of bonamiosis in Lough Foyle, undersize oysters had, on occasion, been returned to Lough Swilly after grading. Good management practices were in place on site to ensure that Lough Swilly oysters only were returned. These included colour coding of the bags holding oysters, as well as physical separation of storage areas and confining grading of oysters from each site to different days. A number of years ago undersize oysters would have been reimmersed just outside the premises in Lough Foyle prior to being sent back to Lough Swilly. The buyer in question stated that this practice stopped about five years prior to the detection of bonamiosis in Lough Foyle in 2005. However, Undersize oysters, continued to be returned to Lough Swilly up until the detection of *B. ostreae* in Lough Foyle, but these animals were returned the same day or the following day, and according to those interviewed, were not submerged in the water.

The equipment used for the grading was the same for both sets of oysters. Oysters from the two areas have been graded on different days for a number of years and the equipment was rinsed down at the end of each day. It is therefore unlikely that the disease could have been transmitted through the use of common equipment in the grading process and the subsequent return of undersize oysters to Lough Swilly. Following the detection of *Bonamia ostreae* in Lough Foyle, procedures were introduced to prevent the return of undersize oysters to Lough Swilly such as the pre-grading of oysters before they were sent to the buyer in Lough Foyle.

2. The second issue, which emerged in relation to sales of shellfish, was the collection of the shellfish by buyers. The patterns of sales and collection of oysters for sale were studied. Three respondents indicated that buyers collected from multiple locations, and these could be mixed loads of *M. edulis*, *O. edulis* and other species. This was substantiated by some of the buyers. Oysters or mussels are sometimes collected from the piers at Rathmelton, Buncrana, Inch and Rathmullen. On occasion, trucks wait for some time at the pier, during which time water could be discharged straight into Lough Swilly. These same trucks often collect *O. edulis* from Lough Foyle because due to its close proximity it makes logistical sense to collect from both loughs in one trip. In this way water harbouring the parasite from Lough Foyle could have found its way into Lough Swilly. In a recent study of the effects of temperature and salinity on the survival of the parasite outside the host Arzul et al (2007) demonstrated a 58% survival rate after a week for the parasite in seawater under experimental conditions (32). These trucks collect molluscs for consumption within the same day from both loughs, and hence it is quite likely that this water could support the survival of the parasite quite readily for the time it would take for the water to make its way into Lough Swilly from nearby Lough Foyle.

3.2.3 Movements of boats, equipment and cultch

Movements of shellfish dredgers between sites were considered to have been the cause of the spread of bonamiosis from the Grevelingen to the Oosterschelde in the Netherlands (Van Banning, 1991). Following the introduction of the *B. ostreae* to the Grevelingen in

1980, movements of boats and equipment came under strict control, and yet in 1988 the disease was detected in the Oosterschelde. Whether it was *B. ostreae* attached to gear, boats, or in mud or the introduction of infected oysters that finally led to the outbreak in the Grevelingen, is not known. Similarly it is believed that movements of boats and equipment from a *B. ostreae* positive area may have caused the introduction of bonamiosis to Achill sound and its subsequent spread to Blacksod Bay.

The aim of the interviews held with the fishermen and aquaculture operators, and through the completion of questionnaires the objective was to determine the extent of movements between approved and non-approved zones. Seventeen respondents said they were aware of other commercial boats from outside Lough Swilly using the lough. Of these, fourteen cited mussel dredgers, which moved back and forth between Lough Swilly and Lough Foyle. One mussel operator said that he moves stock from Lough Foyle to Lough Swilly. Only one movement from Lough Swilly to Lough Foyle appears on the official records (See Table 5). Again it would appear that there are more movements than appear on the official records.

Table 5: Authorised Shellfish Movements out of Lough Swilly from 1996 to 2006

Date	Destination	Species	Quantity
19/09/2001	Inverin	*O. edulis*	30T
01/10/2003	Inverin	*O. edulis*	30T
10/10/2002	Inverin	*O. edulis*	20T
31/05/2005	Galway Bay	*M. edulis*	1T
27/02/2006	Lough Foyle	*M. edulis*	850T

Another respondent advised that there are potentially a number of mussel operators from outside Lough Swilly who fish for mussel seed in Lough Swilly in areas of natural spat settlement, and would relay elsewhere. It seems likely from indications given in the interviews, that some of this seed is being relayed to Lough Foyle and hence that there are boats moving between these two areas. Eight respondents indicated that they had observed either mussel dredgers, which had originated in the Foyle discharging ballast

water into the Swilly or sailboats and other pleasure craft, which had originated in other areas discharging bilge water into the Swilly. These claims could not be substantiated.

Ballast water has often been implicated in the spread of disease agents. For example, the bacterium *Vibrio cholerae* is believed to have been transported from Asia to Latin American coastal waters, through discharges of ballast water. The spread of other organisms, such as the Southeast Asian dinoflagellates of the genera *Gymnodinium* and *Alexandriu*m, has also been attributed to discharges of ballast. Although there is no published information on the potential for the spread of *B. ostreae* by ballast water it would seem logical to assume that provided the parasite can survive outside of its host, that it could be transferred in ballast water. As noted above, survival of the parasite under experimental conditions has been shown for up to a week outside the host (32). Hence the potential for transfer of *B. ostreae* via ballast water seems possible. If as suggested such practices are occurring in Lough Swilly then the introduction of *B. ostreae* to Lough Swilly via discharges of ballast water or bilge water, cannot be ruled out.

Other commercial fishing boats fishing for salmon, crab and other shellfish may also move in and out of the Swilly, but it was not clear from the responses given what other locations these boats may fish.

Fourteen respondents indicated that pleasure craft movements into and out Lough Swilly took place, and one respondent gave additional detail suggesting that these craft may moor in Lough Foyle for some time, before moving and that there existed therefore the potential for spat settlement on the hulls, which could be transferred to Lough Swilly.

Introduction of cultch to Lough Swilly was also investigated. Three respondents stated that they were aware that cultch had been relayed around 2001-2002 at Portstuart Bank but that this had originated in Lough Swilly. No other cultch has been introduced into the Lough as far as either fishermen or those working for government agencies in the area were aware. This mode of transmission was therefore ruled out.

3.3 Potential spread of bonamiosis from Lough Swilly

Table 5 above shows that very few authorised movements of shellfish, for relaying, have taken place from Lough Swilly. Those of concern are movements to a dispatcher in Rossaveal, which is close to Kilkieran Bay (one of the few remaining approved zones in the country). The last movement applied for was in 2003, well before the date of detection of *B. ostreae* in Lough Swilly. Testing in Kilkieran has taken place twice annually throughout this period and so far no *B. ostreae* has been detected. The area will continue to be monitored for the parasite.

4. CONCLUSIONS

4.1 Conclusions: Possible origin of bonamiosis in Lough Swilly

A number of practices were identified which could have posed a risk of introducing bonamiosis into Lough Swilly, though it is not possible to conclude which, if any, was responsible for the introduction of the parasite.

These included:

- **Authorised and unauthorised movements of shellfish considered not responsible for the transmission of bonamiosis from areas where *Bonamia ostreae* is known to occur.**

  Imports of *C. gigas* from France may pose a risk of carrying the parasite directly into Lough Swilly. Preliminary results of two studies, presented at 13th EAFP meeting in September 2007, have raised the possibility that *C. gigas* could act as a carrier of the disease. Further work will be required to show this conclusively.

  M. edulis were moved from Lough Foyle to Lough Swilly before bonamiosis was identified in Lough Foyle. It is possible that the disease was present for some time before it was detected. The risk identified was from potential spat

attachment, and from the close proximity of mussel and oyster beds which could result in the transfer of *O. edulis* along with the *M. edulis*.

- **Return of undersize oysters to Lough Swilly from Lough Foyle prior to the identification of the disease in Lough Foyle.**

  A small risk was identified from the return of undersize oysters, which had been graded on the same site, and using the same equipment as oysters from Lough Foyle. In addition, although the practice of reimmersing oysters was believed to have stopped a number of years before *B. ostreae* was identified in Lough Foyle it is not known how long bonamiosis may have been present in Lough Foyle before its detection. Claims were made that the practice of reimmersing the oysters had continued up to more recent times but it was not possible to substantiate this claim.

- **Discharge of Ballast**

  A number of respondents claimed that Ballast water taken up in Lough Foyle had been discharged into Lough Swilly by mussel dredgers operating in both areas. This claim could not be substantiated. There are no published data on the potential for transfer of *B. ostreae* via ballast, but *B. ostreae* has been shown to survive experimentally in seawater for up to a week. Thus it would be safe to assume that, if this practice had occurred, then it would have posed a risk for the introduction of the disease.

- **Movements of boats between *Bonamia ostreae* positive areas and Lough Swilly.**

  This practice has been identified on previous occasions in both the Netherlands and Ireland as having the potential to spread bonamiosis. Both pleasure craft and commercial boats are operating in both areas. It was noted in one interview that boats may be moored in Lough Foyle for prolonged periods before entering Lough Swilly and that there exists the very real potential for spat settlement on the hulls of such boats, making this a very real possibility for the transfer of the disease between the two loughs.

- **Discharge of water from trucks collecting shellfish for sale.**

It was noted, in some interviews by respondents, that trucks may sometimes wait on the piers to load shellfish, and that these trucks may contain species including *O. edulis* collected in other areas. One of these areas could be the positive area of Lough Foyle. Given that the parasite may exist in seawater for a week this does pose a risk for the spread of the disease.

It is clear that all of these practices pose a risk. The close proximity of Lough Swilly to Lough Foyle and the close links of various sectors of the shellfish industry to both loughs increased the risk to Lough Swilly. Attempts were made by some members of the industry to mitigate against these risks, but the information collected during the course of this investigation suggests that this may not have been sufficient. However it is entirely possible that by the time that *B. ostreae* was detected in Lough Foyle in 2005, the disease had already been transferred to Lough Swilly. Research is still ongoing to improve the sensitivity of the methods available for the detection of the parasite and the circumstances surrounding the spread of bonamiosis to Lough Swilly highlight the need for such improvements in methodology.

4.2 Conclusions: Possible Spread of Bonamiosis from Lough Swilly to other areas

As can be seen from Table 5 above there have been very few authorised movements of shellfish out of Lough Swilly. Nothing further emerged from the questionnaires that gave cause for concern in this area. The main concern, therefore, are the movements of *O. edulis* out of Lough Swilly destined for Rossaveal near Kilkieran Bay. The company in question exports *O. edulis* for human consumption to Holland, France and Spain, but had in the past applied annually to move oysters in case any needed to be reimmersed temporarily before export. When asked whether oysters had been taken in from Swilly and reimmersed, they stated that it was very unlikely. Generally oysters taken from Lough Swilly are exported directly following collection from Donegal. However as they have no records to indicate what shellfish had been temporarily reimmersed they could not say categorically that this had not taken place. The oyster beds at Kilkieran are

approximately 30-35 miles from their site in Rossaveal so the risk posed by this site to these beds is minimal. The Marine Institute will however continue to monitor closely the beds at Kilkieran to ensure that they remain free of the disease and no further consignments of oysters from Lough Swilly will be permitted to be reimmersed at Rossaveal.

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6. **Appendix A: Lough Swilly**

QUESTIONNAIRE

February 2006

What species of shellfish do you farm / fish?

Native Oysters	☐	Section 1A-1D & Section 4
Mussels	☐	Section 2A-2C & Section 4
Other Shellfish	☐	Section 3A-3C & Section 4

Please complete the relevant sections.

Section 1: Native Oysters

1A. Exploitation of Wild Beds

Native Oysters

1a. Do you fish for native oysters?

Yes ☐ No ☐

1b. If so where do you fish?

Lough Swilly Yes ☐ No ☐

Elsewhere Yes ☐ No ☐

If elsewhere please specify_____

2. What time of year do you fish for Native Oysters?

In Lough Swilly_____

Elsewhere (Please specify location and dates)_____

_____

_____

_____

B. Nature of Wild Oyster Beds

Densities

3. {a} What is the width of your dredge?_____

_____

_____

{b} How long do you dredge for at any given time?

_____

_____

{c} How many oysters would you get in a normal tow from the areas listed below?

Russels Bank	☐	Channel Bed	☐	Burt Bed	☐
Ballgreen	☐	Ray Bridge Bed	☐		
Portstuart Bank	☐	Inch Bed	☐		

1C. Movements of Oysters and Equipment

4. Have you or your company ever brought native oysters into Lough Swilly for reseeding of the beds? Please give details of dates, age of stock (if known), quantities, source of stock and where in Lough Swilly it was relayed.

Date	Quantity	Age	Source	Destination

5. Are you aware of anyone else moving stocks of native oysters into Lough Swilly? Please give any details you have in terms of dates quantities, source and destination within L. Swilly.

_____

_____

_____

_____

_____

6. Do you currently use your boat / dredger for fishing purely in Lough Swilly?

Yes ☐ No ☐

If no, where else do you fish besides Lough Swilly?

_____

_____

_____

7. Would you fish in Lough Swilly and elsewhere at the same time of year?

Yes ☐ No ☐

8. How often, by what means and where would you clean your decks / dredgers / other equipment?

_____

_____

_____

9. Was your boat / Dredger ever used previously for fishing other locations and if so when and where was it used?

_____

_____

_____

_____

10. Do you use your boat / dredger for fishing any other species of shellfish?

Yes ☐ No ☐

If yes please specify_____

11. Are you aware of other boats fishing for native oysters in Lough Swilly and other areas? Is so please give as much details as you can in terms of dates and locations

_____

_____

_____

1D. Sales and Distribution

12. Do you sell any native oysters for on growing in Ireland or elsewhere? (Please give details of dates, quantities, source and destination)

Date	Quantity	Age	Source	Destination

13. Over the last 5 years where have you fished and where were the harvested oysters sold to?

In Lough Swilly:

Time of year	Location	Sold to

Elsewhere

Time of year	Location	Sold to

14. Where do you land the stock?

_____

_____

15. How is the stock transported to the final destination (for each destination given above)?

_____

16. Is this stock reimmersed at the final destination or en route that you are aware of?

17. Do you use any other companies to sell stock for you?

Yes ☐ No ☐

18. If so would they collect stock from multiple locations?

Yes ☐ No ☐

19 Do these companies collect other species in addition to native oysters in the same load?

Yes ☐ No ☐

Please give details

20. In the process of transporting this stock is any water likely to be discharged into other water bodies including Lough Swilly. Please give details of other locations where the transport trucks may stop

21. How, where and for how long do you hold your stock prior to sale for human consumption?

22. Have you moved *Ostrea edulis* for human consumption, which were held in tanks close to other water bodies, either within or outside of Lough Swilly prior to marketing?

Yes ☐ No ☐

If yes, please specify when they were moved, where purification took place, approximate tonnage, and what part of the Lough the oysters were from.

Section 2: Mussels

2A. Exploitation of Mussel Beds

Mussels

1a. Do you fish for fish / farm mussels?

Yes ☐ No ☐

1b. If so where do you fish?

Lough Swilly Yes ☐ No ☐

Elsewhere Yes ☐ No ☐

If elsewhere please specify_____

2. Do you fish wild mussels beds or licensed sites for market size mussels?

Wild ☐ Licensed sites ☐

3. Do you fish for any other Shellfish or have you done so in the last 5 years?

Yes ☐ No ☐

Please specify what species, where, when and how

_____

_____

4. Where do you get your mussels seed from?

Lough Swilly ☐ Elsewhere ☐

Please specify_____

2B. Movements of Shellfish and Equipment

7. Do you own your own boat / dredger

Yes ☐ No ☐

If yes, do you use the same boat/equipment for fishing Lough Swilly and elsewhere?

Yes ☐ No ☐

8. Would you fish in Lough Swilly and elsewhere at the same time of year? (Please give details)

_____
_____
_____

9. If you do not own your own boat, how do you fish for mussels?

_____
_____

10. How often, by what means and where would you clean your decks / dredgers / other equipment?

_____
_____
_____
_____

11. Was your boat / Dredger ever used previously for fishing other locations and if so when and where was it used?

_____
_____
_____
_____
_____

12. Do you use your boat / dredger for fishing any other species of shellfish?

Yes ☐ No ☐

If yes please specify_____

13. Have you or your company ever brought mussels into Lough Swilly for on growing? Please give details of dates, age of stock (if known), quantities, source of stock and where in Lough Swilly it was relayed.

Date	Quantity	Age	Source	Destination

14. Have you or your company ever brought other shellfish into Lough Swilly for on growing. Please give any details you have in terms of dates quantities, source and destination within L. Swilly.

_____
_____
_____
_____

2C. Sales and Distribution

15. Over the last 5 years where have you fished and where were the harvested mussels sold to?

In Lough Swilly:

Time of year	Location	Sold to

Elsewhere

Time of year	Location	Sold to

16. How is the stock transported to the final destination (for each destination given above)?

_____
_____
_____
_____
_____

17. Where do you land the stock?

_____
_____
_____

18. Is this stock reimmersed at the final destination or en route that you are aware of?

_____
_____

19. Do you use any other companies to sell stock for you?

Yes ☐ No ☐

Please specify_____

20. If so would they collect stock from multiple locations?

Yes ☐ No ☐

21. Do these companies collect other species in addition to native oysters in the same load?

Yes ☐ No ☐

Please give details

_____

_____

_____

22. In the process of transporting this stock is any water likely to be discharged into other water bodies including Lough Swilly. Please give details of other locations where the transport trucks may stop

_____

_____

_____

_____

23. How, where and for how long do you hold your stock prior to sale for human consumption?

_____

_____

_____

24. Do you sell any stock for on growing in Ireland or elsewhere? (Please give details of species, dates, quantities, source and destination)

Yes ☐ No ☐

Species	Date	Quantity	Age	Source	Destination

Section 3: Other Shellfish

3A. Exploitation of Wild Beds / Shellfish Aquaculture

1a. What species do you farm / fish / gather?

Please give details_____

_____

_____

1b. Is this confined to Lough Swilly

Lough Swilly Yes ☐ No ☐

Elsewhere Yes ☐ No ☐

If elsewhere please specify_____

2. What time of year do you fish?

In Lough Swilly_____

Elsewhere (Please specify location and dates)_____

_____

_____

_____

Do you use the same boat/equipment for fishing Lough Swilly and elsewhere (Please give details)_____

_____

_____

3B. Movements of Shellfish and Equipment

5. Do you own your own boat / dredger

Yes ☐ No ☐

6. If yes, do you use the same boat/equipment for fishing Lough Swilly and elsewhere?

Yes ☐ No ☐

7. Would you fish in Lough Swilly and elsewhere at the same time of year?

Yes ☐ No ☐

Please give details

_____

_____

_____

8. If you do not own your own boat, how do you fish /harvest?

_____

_____

9. How often, by what means and where would you clean your decks / dredgers / other equipment?

_____

_____

_____

10. Was your boat / Dredger ever used previously for fishing other locations and if so when and where was it used?

_____

_____

_____

_____

11. Have you or your company ever brought shellfish into Lough Swilly for on growing? Please give details of dates, age of stock (if known), quantities, source of stock and where in Lough Swilly it was relayed.

Date	Quantity	Age	Source	Destination

3C. Sales and Distribution

12. Do you sell any stock for on growing in Ireland or elsewhere? (Please give details of species, dates, quantities, source and destination)

Species	Date	Quantity	Age	Source	Destination

13. Where are your main markets for sales for human consumption?

14. How is the stock transported for sale and is it relayed at it's final destination or elsewhere after transportation?

15. How, where and for how long to you hold your stock prior to sale for human consumption?

16. Have you moved stock for human consumption, which were held in tanks close to other water bodies, either within or outside of Lough Swilly prior to marketing?

Yes ☐ No ☐

If yes, please specify when they were moved, where purification took place, approximate tonnage, and what part of the Lough the stock was from.

17. Have you ever held stock in storage tanks or in bags on the shore in the vicinity of Lough Swilly, which have originated from other areas?

Please specify when, where tanks / bags were located, origin of stock

18. Do you use any other companies to sell stock for you?

Yes ☐ No ☐

19. If so would they collect stock from multiple locations?

Yes ☐ No ☐

20. Do these companies collect other species in addition to your stock in the same load?

Yes ☐ No ☐

Please give details

21. In the process of transporting this stock is any water likely to be discharged into other water bodies including Lough Swilly. Please give details of other locations where the transport trucks may stop

Section 4: General

1. Are you aware of other commercial fishing boats using Lough Swilly for harvesting of mussels, oysters or other shellfish?

Yes ☐ No ☐

Please give details including any other sites they may fish

_____
_____
_____
_____
_____
_____
_

2. Do other pleasure craft use Lough Swilly and would they spend prolonged periods in other areas prior to Lough Swilly?

_____
_____
_____
_____

3. Are you aware of ballast water being discharged into Lough Swilly and do you know where these boats may have originated?

_____
_____
_____

4. Has cultch been introduced into Lough Swilly?

If yes, please specify when was it introduced, it's origin and where in the Lough it has been used.

_____
_____
_____
_____

5. Are you aware of *Ostrea edulis* being held in storage tanks or in bags on the shore in the vicinity of Lough Swilly, which have originated from other areas?

Please specify when, where tanks/bags were located, origin of oysters.

6. Have you or are you aware of other shellfish being reimmersed temporarily in Lough Swilly prior to sale for human consumption which originated in either L. Swilly or elsewhere. Please give details of species, approximate quantities dates and where stock originated from?

7. Are you aware of any other shellfish being held in storage tanks in the vicinity of Lough Swilly? If so please give as much detail as you have in relation to dates, origin and species

8. Are you aware of anyone moving stocks of shellfish into Lough Swilly for on growing? Please give any details you have in terms of dates quantities, source and destination within L. Swilly.

9. Do you have any information about densities of oysters on the beds either gathered personally or from surveys etc.? Please give details including sources of information

10. Have you ever observed any abnormal or unexplained mortality in any of the oyster beds

Yes ☐ No ☐

If yes please give details:

Date	Bed	Percentage mortality	Other information

11. Additional Information. Please give any other information that you feel may be relevant to this investigation.