Australian Seafood Industries Pacific Oyster Mortality Syndrome (POMS) investigation into the 2016 disease outbreak in Tasmania - ASI emergency response

Matthew Cunningham

16/3/2017

FRDC Project No **2015/232**

© 2017 Fisheries Research and Development Corporation.
All rights reserved.

Australian Seafood Industries Pacific Oyster Mortality Syndrome (POMS) investigation into the 2016 disease outbreak in Tasmania - ASI emergency response

2015/232

2017

Ownership of Intellectual property rights
Unless otherwise noted, copyright (and any other intellectual property rights, if any) in this publication is owned by the Fisheries Research and Development Corporation and Australian Seafood Industries Pty Ltd.

Creative Commons licence
All material in this publication is licensed under a Creative Commons Attribution 3.0 Australia Licence, save for content supplied by third parties, logos and the Commonwealth Coat of Arms.

Creative Commons Attribution 3.0 Australia Licence is a standard form licence agreement that allows you to copy, distribute, transmit and adapt this publication provided you attribute the work. A summary of the licence terms is available from creativecommons.org/licenses/by/3.0/au/deed.en. The full licence terms are available from creativecommons.org/licenses/by/3.0/au/legalcode.

Inquiries regarding the licence and any use of this document should be sent to: frdc@frdc.com.au

Disclaimer
The authors do not warrant that the information in this document is free from errors or omissions. The authors do not accept any form of liability, be it contractual, tortious, or otherwise, for the contents of this document or for any consequences arising from its use or any reliance placed upon it. The information, opinions and advice contained in this document may not relate, or be relevant, to a readers particular circumstances. Opinions expressed by the authors are the individual opinions expressed by those persons and are not necessarily those of the publisher, research provider or the FRDC.

The Fisheries Research and Development Corporation plans, invests in and manages fisheries research and development throughout Australia. It is a statutory authority within the portfolio of the federal Minister for Agriculture, Fisheries and Forestry, jointly funded by the Australian Government and the fishing industry.

Researcher Contact Details	**FRDC contact details**
Name: Matthew Cunningham	Address: 25 Geils Court, Deakin ACT 2600
Address: Tech 3 Tasmanian Technopark, Dowsings Point, Glenorchy, 7010	Phone: 026850400
Phone: 0362723397	Fax: 020628500499
Fax:	Email: frdc@frdc.com.au
Email: matt@asioysters.com.au	Web: www.frdc.com.au

In submitting this report, the researcher has agreed to FRDC publishing this material in its edited form.

Contents

Acknowledgments

ASI would like to acknowledge FRDC for funding this and previous research projects. We would also like to acknowledge the Seafood CRC which also funded previous research into selective breeding of Pacific Oysters. We also wish to thank Peter Kube from CSIRO who provides quantitative genetics services to the ASI breeding program. Finally, we would like to acknowledge the contribution by commercial hatcheries and Oyster growers who have supported this and other projects. Shellfish Culture Ltd who has provided important operational support for ASI during this project. Josh Poke, Justin Goc and Hayden Dyke provided direct support for this project. ASI has also enjoyed support over many years from growers who are too numerous to mention.

Abbreviations

ASI: Australian seafood Industries Ltd

POMS: Pacific Oyster Mortality Syndrome

EBV: Estimated Breeding Value

CVO: Chief Veterinary Officer

Executive Summary

This report details the emergency response undertaken by Australian Seafood Industries in response to the POMS outbreak in Tasmania in January 2016. The project activities occurred from February to June 2016 in locations affected by POMS. The project had three main objectives. The first was to rescue the latest generation of ASI Pacific Oyster families located at Shellfish Culture's Pipeclay Lagoon land based facility. The second was to gather POMS mortality data from ASI family lines located in affected areas. The third was to be able to undertake activities which lead to a fast tracking of family line commercial multiplication to allow industry to have access to the best commercial families at the earliest possible time. To allow this survival data was collected, the results were analysed and compared to previously collected data in NSW and a broodstock inventory developed. This allowed ASI to swiftly provide the most resistant family lines to commercial hatchery producers to facilitate industry restocking. The outcome has been that growers will be restocking their farms with stock with useful levels of POMS resistance in the coming season. There is a clear requirement for ongoing work to both continue and accelerate the genetic gains in POMS resistance. Areas for further research and development are ensuring the gains made in POMS resistance are available nationally, developing the capability to breed from animals that have survived POMS and to develop better estimations of survival at different sizes and life stages.

Keywords

Pacific Oyster, Pacific oyster Mortality Syndrome, Selective Breeding

Introduction

The viral disease known as Pacific Oyster Mortality Syndrome (POMS) was diagnosed for the first time in Tasmania in February 2016. Based on the high mortality and rate of spread seen in previous outbreaks of this disease in NSW, it is expected that most Tasmanian oyster production will cease over the coming year in infected areas. Since the two hatcheries that produce most of the oyster spat for Australia are based in Tasmania, production in NSW and SA will also be significantly reduced. This represents a potential loss of livelihood for 400 family farms with an annual GVP of $75million.

ASI Pty Ltd is an industry-owned company with the sole responsibility of breeding POMS resistant Pacific Oysters by 2018. The company's only source of income is a voluntary levy on oyster spat sales which will be significantly impacted by the POMS outbreak in Tasmania. Oysters that are partially resistant to POMS have been produced and these oysters offer the only solution to the industry for restocking following the outbreak of POMS. Due to disruptions in the marketplace as a result of the POMS outbreak ASI income will be severely impacted which would preclude the restocking.

Objectives

1. Rescue of the latest generation of all ASI family lines by transferring selectively bred spat produced in December / January from Shellfish Culture Ltd to a quarantine facility at IMAS.

2. Assessment of the survival rate of brood stock of all ASI oyster family lines located in areas exposed to the POMS virus.

3. Multiplication of the best family lines with the highest survival rate as soon as possible to enable hatcheries to provide spat for a Progressive Industry Recovery Program.

Method

Rescue of the latest generation of all ASI family lines

On the 1st February 2016 POMS was confirmed as the causative agent for massive mortalities in cultured Pacific Oysters Lower Pittwater in Tasmania. Stock transfers had taken place to other Oyster growing regions within Tasmania including Pipeclay Lagoon. The most recent generation of ASI family lines were located at the land based nursery facilities of Shellfish Culture undergoing grow out prior to transfer to sea based culture systems. This stock was deemed to be at high risk of being exposed to POMS and as a result steps were taken to protect them. There was also a risk that Shellfish Culture may need to disinfect that site which would involve cutting off the water supply. Given that a complete stock standstill had been invoked by the Tasmanian CVO, a 3 stage methodology was developed. The first was to undertake testing to establish if the spat had been exposed to virus. The second was to put in place a water treatment system and quarantine measures to ensure that stock was protected from being infected by new virus. The third was to establish a quarantine area at IMAS at Taroona to guard against the possibility of facility shutdown at Shellfish Culture.

The spat were tested immediately for presence of POMS by PCR. The tests were conducted at the Animal Health Laboratory, Tasmanian Department of Primary Industries, Parks, Water and Environment. The tests were negative for the POMS virus. A water filtration system was then developed for the spat and commissioned on the 3rd February which was 2 days after the confirmation of POMS. A UV filtration unit with 1um prefiltration was used and the nursery system was converted to a partial recirculation system to minimise water usage and allow for maximum exposure time in the UV unit. Mortalities were observed in Pacific Oysters on the 3rd of February in the Pipeclay growing region which developed into large scale mortalities over the ensuing weeks. There continued to be no observed mortalities in the ASI family lines. A quarantine system was also established at IMAS during this time and following further retesting for POMS, which again returned negative results, the spat were transferred under permit on the 2nd March 2016.

Assessment of the survival rate of brood stock of all ASI oyster family lines located in areas exposed to the POMS virus.

Mortality assessments were conducted in Pipeclay Lagoon, Pittwater and Little Swanport which were the three POMS affected growing areas in which ASI had its family lines present. The assessments were conducted from 17th February and concluded on the 9th March. Data was collected on all year classes of ASI family lines. The data was used to generate estimated breeding values for the trait of POMS resistance for all ASI family lines. A broodstock inventory was developed for key commercial candidate families.

Multiplication of the best family lines with the highest survival rate

The generation of EBV's allowed identification of the most resistant ASI family lines and this was used as the basis for allocation of this broodstock to commercial hatcheries to allow partially resistant lines to be produced and distributed to industry. A breeding calculator tool was developed allowing commercial hatcheries to predict survival and inbreeding for potential commercial crosses of the most resistant families. Broodstock allocation began on the 11th April 2016 and is ongoing.

Results, Discussion, Conclusion

Rescue of the latest generation of all ASI family lines

The family lines remained negative to POMS until they were returned to Pipeclay lagoon in May 2016 once the POMS season was concluded. These families will be challenged against the POMS virus in the upcoming POMS season when they are approximately 12 months old. Survival rates that will allow sufficient survivors for commercial production are predicted at this age. In addition to this spat was also retained at IMAS and later sent to Georges River in NSW to also be challenged by the POMS virus. A Georges River challenge and the resultant data is very important to the program as we have 4 years of data from this site which allows estimations of cumulative genetic gains.

Assessment of the survival rate of brood stock of all ASI oyster family lines located in areas exposed to the POMS virus.

The results of the mortality showed large variation to POMS resistance. EBV's from the YC14 year class ranged from 3.6% to 85.4%. In addition to this it was also possible to calculate the increased rate of genetic gains made possible from breeding from surviving animals. Most importantly the results correlated very well to what we had seen in NSW meaning that the selection we had done on this trait previously would be directly applicable to Tasmania. See appendix 1.

Multiplication of the best family lines with the highest survival rate

Supply of these lines has now resulted in commercial production from ASI families with the highest levels of POMS resistance. Since the outbreak of POMS all hatchery production of spat has been derived from ASI broodstock. Furthermore, the ability to achieve objective 1 ensures there will be an ongoing supply of commercial and breeding program broodtock with increased levels of POMS resistance. The outcomes of this and earlier research has placed the Australian Pacific Oyster industry in a position of relative strength compared with other international industries which were unable to make genetic advances in POMS resistance prior to the arrival of the disease. See appendix 2 for Broodstock policy.

Implications, Recommendations and Further development

Specifically, the implications of this project were that crucial Research and development activities, which built on a large body of previous work, were allowed to be undertaken in a timely fashion. This allowed the outcomes that were achieved to be rolled out to industry with the urgency that the situation required.

There is a clear requirement for ongoing work to both continue and accelerate the genetic gains in POMS resistance. Areas for further research and development are ensuring the gains made in POMS resistance are available nationally, developing the capability to breed from animals that have survived POMS and to develop better estimations of survival at different sizes and life stages.

These areas of further development that have been identified formed the basis for a successful application for funding through the Future Oysters Cooperative Research Centre Project commencing in 2016

Extension and Adoption

Extension of research outcomes occurred through regular industry updates in the form of industry newsletters, ASI newsletters and attendance at industry seminars. Outcomes were also disseminated to commercial hatcheries in the form of provision of broodstock with known POMS resistance. This resulted in almost immediate adoption of the outcomes of this research. The outcomes of this and previous research have allowed Pacific Oyster growers to start the process of restocking their operations with the best available spat in terms of POMS resistance.

Project materials developed

Project materials developed as part of this project are limited to the Estimated Breeding values for POMS resistance of ASI family lines.

Appendix 1

FAM_ID	POMS_FAM_EBV_%
2014075	85.4%
2014054	65.4%
2014050	60.2%
2014072	58.0%
2013031	57.3%
2013060	55.6%
2014055	52.4%
2013085	52.0%
2013001	51.2%
2013005	49.1%
2013007	46.0%
2014002	44.9%
2014010	44.4%
2014013	43.8%
2014070	43.2%
2014046	42.7%
2014064	42.2%
2014053	41.8%
2014033	41.4%
2014071	41.0%
2013019	40.0%
2014047	39.1%
2013002	39.0%
2014085	38.9%
2014017	38.5%
2013003	37.1%
2013077	36.7%
2013021	36.6%
2013053	36.2%
2013084	35.8%
2013009	35.0%
2013052	34.9%
2013063	34.8%
2013045	34.5%
2013013	33.0%
2013032	32.8%
2014083	32.6%
2013066	32.6%
2013024	32.5%
2013027	32.5%
2013033	32.5%
2013044	32.1%
2014018	32.1%
2014057	32.1%
2014040	31.7%

2014023	30.8%
2013048	30.2%
2013017	30.2%
2014082	29.6%
2013004	29.5%
2014059	29.3%
2013006	29.0%
2013016	28.4%
2014014	28.4%
2014012	27.9%
2014052	27.9%
2014007	27.9%
2014016	27.4%
2013058	27.3%
2013022	27.0%
2014008	26.9%
2013038	26.7%
2014060	26.6%
2013078	26.6%
2014039	26.4%
2013087	26.0%
2013062	25.8%
2014009	25.5%
2013064	25.5%
2014048	24.8%
2014063	24.8%
2014084	24.6%
2013061	24.5%
2014005	23.9%
2013040	23.6%
2013014	23.6%
2014066	23.1%
2014024	23.0%
2013047	22.9%
2014021	22.3%
2014015	22.3%
2014003	22.3%
2013025	22.0%
2014004	21.9%
2013020	21.7%
2014006	21.1%
2013046	21.0%
2013015	20.8%
2014001	20.6%
2014037	20.5%
2014011	20.3%
2013065	20.2%
2014031	20.2%

2014049	20.1%
2014067	19.9%
2014058	19.3%
2013073	19.3%
2014028	18.8%
2013054	18.1%
2013059	17.7%
2014068	17.7%
2014077	17.6%
2014076	17.5%
2014029	17.0%
2014035	16.5%
2014026	16.5%
2013026	16.0%
2014062	15.9%
2014034	15.7%
2013042	15.7%
2014065	15.7%
2014022	15.6%
2013074	15.4%
2013028	15.2%
2013055	15.0%
2014038	14.6%
2013071	14.6%
2014056	14.1%
2014036	14.0%
2014030	13.8%
2014032	13.8%
2013008	13.0%
2014027	12.4%
2014069	12.1%
2013079	12.0%
2014051	11.9%
2014081	11.8%
2014061	11.5%
2013068	10.9%
2013083	10.7%
2014074	10.5%
2014019	10.1%
2013072	9.9%
2013030	9.9%
2013057	9.8%
2014045	9.8%
2013067	9.6%
2013011	9.4%
2014079	9.0%
2013018	8.9%
2013056	8.3%

2014025	8.3%
2013069	7.6%
2013076	7.3%
2014080	7.0%
2013023	6.7%
2013080	6.5%
2013081	6.1%
2013034	6.1%
2013039	5.9%
2013070	5.3%
2013036	5.2%
2013043	4.6%
2013029	4.1%
2013050	4.0%
2014078	3.6%
2013075	2.9%
2013035	2.8%
2013041	2.6%
2013082	2.2%
2013012	1.5%
2013037	1.2%
2013051	1.1%
2013049	-1.7%
2013086	-4.0%
2013010	-5.8%

Appendix 2

ASI Thoroughbred Oysters	**Broodstock Procedure** **Distribution of Broodstock to Oyster Hatcheries**

Disclaimer on "POMS Resistant" Oysters

ASI is an industry owned company that distributes selectively bred oyster broodstock to hatcheries free of charge. ASI is selecting successive generations of oysters for resistance to POMS. In conjunction with industry the target for ***POMS Resistance*** *was set at an average of 70% survival of 1 year old oysters in a family when challenged with the POMS virus. This target will not be met until 2018. However, in view of the outbreak of POMS in Tasmania. ASI is considering releasing broodstock that is* ***Partially Resistant*** *(less than 70% survival) to POMS for use by hatcheries in their breeding. Accordingly, ASI cannot and does not guarantee the survival of these oysters as their performance under commercial conditions cannot be assured.*

Background

As a result of the POMS outbreak in Tasmania, hatcheries have requested ASI to supply broodstock with the highest Estimated Breeding Values for POMS resistance (EBV-P), so that breeding and supply of partially resistant oysters can begin as soon as possible.

Many ASI broodstock and young oysters from ASI families have now been exposed to POMS in Tasmania and new information about level of resistance is available for each family. As a result, the EBV-Ps have been recalculated by CSIRO.

In the past ASI had adequate supplies of broodstock to meet demand and these were provided to hatcheries on request. However, in the current circumstances, with a significant increase in demand for ASI oysters, it may not be possible to supply all hatcheries with all the high EBV-P broodstock they request. This is because:

- Only small numbers of broodstock used in previous generations are retained by ASI
- ASI must retain some broodstock for future use in the national breeding program
- In some families, the number of these broodstock is extremely limited, due to losses incurred during normal operations.
- ASI keeps its broodstock on oyster farms in Tasmania and SA, and all families are not represented at each site.
- Only one site in Tasmania (St Helens) is POMS free. Distribution of oysters between farms is governed by movement controls implemented by the Chief Veterinary Officer, which means that broodstock held in infected areas can only be distributed to other infected areas.

Consequently ASI has developed this procedure to guide the equitable distribution of broodstock to hatcheries.

Oversight of this procedure will be by a subcommittee composed of the three independent directors of the ASI Board. The procedure will be revised as needed.

Available Broodstock
Broodstock are classified by their year class; the year in which they were produced. Hence YC13 broodstock were produced in 2013 and used for breeding from 24 months of age.

The details of ASI broodstock available are shown in Table 1 and 2, *attached*.

Supply in Tasmania
YC14
The latest available generation of ASI oysters is YC14. These oysters are immature and should be ready for use as broodstock in September 2016 or earlier depending on their growth rate. There are 14 families of YC14 with high EBV-Ps. Three of these are not available in sufficient numbers for distribution. Supplies of the remaining families are sufficient to meet the current needs of all hatcheries. A second round of distribution will occur later in 2016.

YC13
The previous generation of ASI oysters, YC13, has been used extensively in the national breeding program and therefore ASI will only have moderate requirement for YC13 broodstock and thus these mature oysters are available for distribution to hatcheries for immediate use. There are nine families of YC13 with high EBV-Ps. Three of these families are not available in sufficient numbers for distribution. Of the remaining six families, there is only one where insufficient oysters exist to distribute to all hatcheries. Consequently, the allocation process described below will be applied.

YC12
There are two families of YC12 with high EBV-Ps. The allocation process will also be applied to these two families.

POMS free zones
At this point in time only limited stock is available in POMS free zones which is St Helens. The same rules apply to other ASI broodstock. For those hatcheries with a demonstrated requirement for these oysters the broodstock will be allocated as per the described allocation process. Given the limited numbers interested parties may be required to negotiate allocation. If a negotiated position can't be achieved ASI will make the final decision. Hatcheries in infected areas must be able to prove that biosecurity requirements have been met so that broodstock supplied can be commercialised.

Supply in South Australia
YC14
The latest available generation of ASI oysters is YC14. These oysters are immature although increased growth rates in SA means they may be ready for use as broodstock prior to September 2016. There are 14 families of YC14 with high EBV-Ps. Three of these families are not available in sufficient numbers for distribution. Supplies of the remaining families are sufficient to meet the needs of all hatcheries. A second round of distribution will occur later in 2016.

YC13
The previous generation of ASI oysters, YC13, has been used extensively in the national breeding program. ASI has completed breeding with YC13 broodstock and these mature oysters are thus available for distribution to hatcheries for immediate use. There are nine families of YC13 with high EBV-Ps. Three of these are not available in sufficient numbers for distribution. Of the remaining six families, there is only one where insufficient oysters exist to distribute to all hatcheries. Consequently, the allocation process described below will be applied.

YC12
These are not available due to low numbers.

The Allocation Process
ASI's ability to provide sufficient oysters to all hatcheries depends in part on the minimum number of oysters required by hatcheries to conduct a commercial breeding run. Hatcheries have different requirements, depending on the breeding techniques used, and their volume of expected sales.

After discussions with hatcheries, is has been determined that 20 oysters is the minimum number of oysters per family required to ensure a satisfactory breeding run. Larger hatcheries will need to undertake multiple breeding runs and will therefore need more broodstock in multiples of 20.

Key Guiding Principle

After considering a range of options, the ASI Board decided that the fairest way to allocate broodstock that is in limited supply shall be in proportion to each hatchery's market share. Since the actual level of hatchery sales is unknown, an estimate will be used. Given that new hatcheries do not have past market share it was further determined that a small proportion of most broodstock families will be made available for new hatcheries. ASI reserves the right to make an assessment of supply vs likely output.

Each hatchery will be provided with a list of all available broodstock and their EBVs for all traits, as well as a *breeding predictor spreadsheet tool* (attached). Using this information hatcheries can then apply for broodstock they require using the attached form. Maximum order levels are indicated on the form.

Data on the actual supply to each hatchery will be kept confidential by ASI.

Levy Arrangements
Please note that in submitting their order, existing hatcheries will be requested to sign a statement by which they agree to continue to abide by the previously signed Deed of Undertaking.

New hatcheries will need to sign the Deed before they can be supplied.

Table 1. ASI Tas Broodstock with high EBV-Ps
(Numbers shown are individual oysters)

Year Class and Family Line	EBV-P	Location				Total	Retained only by ASI	Available to Hatcheries
		Pipeclay	Little Swanport	Pittwater	St Helens (POMS free)			
YC12.16	50%	91				91		√
YC12.44	33%	187				187		√
								√
YC13.31	57%	393				393		√
YC13.60	56%	215				215		√
YC13.85	52%	257				257		√
YC13.01	51%	128				128		√
YC13.05	49%	15				15	√	X
YC13.07	46%	196				196		√
YC13.19	40%	44				44	√	X
YC13.02	39%	53				53	√	X
YC13.03	37%	286				286		√
2014075	85%	404	204	200	0	808		√
2014054	65%	144	352	200	0	696		√
2014050	60%	0	444	200	60	704		√
2014072	58%	290	540	200	60	1090		√
2014055	52%	96	0	200	0	296	√	X
2014002	45%	155	468	200	60	883		√
2014010	44%	277	439	200	0	916		√
2014013	44%	145	437	200	60	842		√
2014070	43%	173	0	200	60	433	√	X
2014046	43%	175	0	200	0	375	√	X
2014064	42%	24	0	200	0	224	√	X
2014053	42%	0	810	200	60	1070		√
2014033	41%	144	530	200	60	934		√
2014071	41%	117	206	200	60	583		√

Table 2. ASI SA Broodstock with high EBV-Ps
(Numbers shown are individual oysters)

Year Class and Family Line	EBV-P	Location				Total	Retained only by ASI	Available to Hatcheries
		Smoky	Coffin	Cowell	Denial			
YC12.16	50%	25				25	√	X
YC12.44	33%	25				25	√	X
								√
YC13.31	57%	90				90		√
YC13.60	56%	60				60		√
YC13.85	52%	60				60		√
YC13.01	51%	0				0		X
YC13.05	49%	0				0		X
YC13.07	46%	90				90		√
YC13.19	40%	0				0		X
YC13.02	39%	90				90		√
YC13.03	37%	90				90		√
2014075	85%	212	151	200	111	674		√
2014054	65%	193	0	180	180	553		√
2014050	60%	247	144	221	0	612		√
2014072	58%	160	157	126	0	443		√
2014055	52%	232	0	0	0	232		√
2014002	45%	187	0	0	0	187		√
2014010	44%	209	130	128	0	467		√
2014013	44%	226	44	146	60	476		√
2014070	43%	193	60	216	0	469		√
2014046	43%	220	0	222	60	502		√
2014064	42%	0	0	0	0	0		X
2014053	42%	231	0	211	60	502		√
2014033	41%	169	0	197	30	396		√
2014071	41%	233	88	68	60	449		√

Broodstock Order Form – Tasmania

Year Class and Family Line	EBV-P	Total Oysters	Maximum Supply Numbers (if applicable)			Number Requested
			Existing Hatcheries	New Hatcheries	POMS free Zone	
YC12.16	50%	91	30	0	0	
YC12.44	33%	187	40	20	0	
					0	
YC13.31	57%	393	60	60	0	
YC13.60	56%	215	60	20	0	
YC13.85	52%	257	60	20	0	
YC13.01	51%	128	40	0	0	
YC13.07	46%	196	60	20	0	
YC13.03	37%	286	60	40	0	
2014075	85%	808	60	60	0	
2014054	65%	696	60	60	0	
2014050	60%	704	60	60	60	
2014072	58%	1090	60	60	60	
2014002	45%	883	60	60	60	
2014010	44%	916	60	60	0	
2014013	44%	842	60	60	60	
2014070	43%	433	60	60	60	
2014053	42%	1070	60	60	60	
2014033	41%	934	60	60	60	
2014071	41%	583	60	60	60	

Please only request broodstock numbers sufficient to meet your realistic production targets. ASI expects its broodstock to be fully utilised to the benefit of the growers and reserves the right to make an assessment of supply vs likely output.

Broodstock Order Form – South Australia

Year Class and Family Line	EBV-P	Total Oysters	Maximum Supply Numbers (if applicable)		
			Existing Hatcheries	New Hatcheries	Number Requested
YC12.16	50%	25	0	0	
YC12.44	33%	25	0	0	
YC13.31	57%	90	20	20	
YC13.60	56%	60	20	0	
YC13.85	52%	60	20	0	
YC13.01	51%	90	20	20	
YC13.07	46%	90	20	20	
YC13.03	37%	90	20	20	
2014075	85%	674	60	60	
2014054	65%	553	60	60	
2014050	60%	612	60	60	
2014072	58%	443	60	60	
2014002	45%	187	60	60	
2014010	44%	467	60	60	
2014013	44%	476	60	60	
2014070	43%	469	60	60	
2014053	42%	502	60	60	
2014033	41%	396	60	60	
2014071	41%	449	60	60	

Please only request broodstock numbers sufficient to meet your realistic production targets. ASI expects its broodstock to be fully utilised to the benefit of the growers and reserves the right to make an assessment of supply vs likely output.

Declaration:

By requesting supply of ASI broodstock I agree to continue to abide by the previously signed Deed of Undertaking.

Hatchery Business:

Signed by:

Signature:

Date: