This document was created in response to a Freedom of Information request made to CSIRO.

FOI Number: FOI2011/69

Date: 31 July 2012

Request: Documents relating to the outbreak of a herpes-like virus in Victoria abalone, now known as Abalone Viral Ganglioneuritis ("AVG") which commenced during December 2005 (the "Victorian Outbreak")

Documents: Part 3 – Documents 51-76

For more information, please refer to CSIRO's FOI disclosure log at www.csiro.au/FOILog
DOCUMENT 51

EXEMPT IN FULL – s 47B(a)
DOCUMENT 52

EXEMPT IN FULL – s 47B(a)
Hi Nick,

We had negotiated that if we could get $30,000.00 from any source and put it through Victoria State then FRDC could match it to make a total of $60,000.00 for this financial year. The industry has not been able to raise any funds to date but I believe that it is still under discussion - but the financial year is now almost over. Certainly FRDC preproposal(s) have been prepared and whether they are processed out-of-cycle is also being considered - but nothing definite.

Mark

MARK CRANE Ph.D.
Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
Private Bag 24
Geelong Vic 3220

International Phone: +61 3 52 275118
International Fax: +61 3 52 275555
email: mark.crane@csiro.au

-----Original Message-----
From: Elliott, Nick (CMAR, Hobart)
Sent: Wednesday, 7 June 2006 09:24
To: Crane, Mark (LI, Geelong)
Subject: Abalone virus / any progress

Hi Mark

I have had no response from the FFF office on possible support. I will check with them.

Have you and others had any luck with FRDC?
Nick
Mehdi,

I have revised the preproposal as discussed. Included are all the costs for AAHL. I have included salaries for technical support (salaries and operating) for Attwood. You will need to put in values for your components which will increase the total.

The breakdown is as follows:

AAHL Salaries: $193,661
AAHL Operating: $41,339
AAHL Travel: $2,000
AAHL In-kind: $151,842

Attwood Salaries: $42,000
Attwood Operating: $10,000
Attwood Travel: $2,000
Attwood in-kind: $21,000

Total to date: $463,843

FRDC's contribution is $291,000. If we get any cash contributions then this will be reduced.

Hope this makes sense.

Cheers

Mark

MARK CRANE Ph.D.
Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
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Geelong Vic 3220

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e-mail: mark.crane@csiro.au

-----Original Message-----
From: Mehdi.Doroudi@dpi.vic.gov.au [mailto:Mehdi.Doroudi@dpi.vic.gov.au]
Sent: Thursday, 8 June 2006 14:03
To: Crane, Mark (LI, Geelong)
Agreed as discussed.

Regards
Mehdi Doroudi - DVM, PhD
Research Director
Marine & Freshwater Systems - PIRVic
Department of Primary Industries

Ph: +613 5258 0272  Fax: +613 5258 0270
Mobile: +61400 845 406

Email: mehdi.doroudi@dpi.vic.gov.au
2A Bellarine Hwy, Queenscliff VIC 3225 Australia
PO Box 114, Queenscliff VIC 3225 Australia

How about this Mehdi:

A 2-year project to achieve:

1. Development of management strategies and control options
2. Sequence genes and develop conventional PCR (1.0 FTE for 24 months at AAHL)
3. Use sequence to develop ISH (1.0 FTE for 12 months at VicDPI)

The real-time PCR would be dropped from this project to reduce the size of the project (but we may develop it in-house - anticipating future surveillance needs)

If you agree, I can revise our part of the preproposal and the research budget.

Mark

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Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
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Geelong Vic 3220

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email: mark.crane@csiro.au
Mark,

Thanks for the message.

I understand the importance of reliable diagnostic tests (specifically as confirmatory tests). However, when it comes to PCR, it is more important to have access to a rapid test (than reliable) or tests that can detect virus while abalone is not clinically affected. This will definitely help in the development of management strategies, however, control measures could still be developed in the absence of a PCR test.

To be able to go forward from here, we need to make a decision how we want to pursue this pre-proposal. The possible options are:

1) To submit two separate pre-proposals and wait for the FRABs and FRDC’s comments. This means that you will need to submit the current pre-proposal as it stands to VICFRAB by tomorrow and I will take care of the management concept.

2) To incorporate my comments into the current pre-proposal and add additional person to deliver on diagnostic work within 12 months or to extend the diagnostic side of the project for another 12 months.

Please indicate which approach would you prefer “option 1 or 2”.

Regards

Mehdi Doroudi - DVM, PhD
Research Director
Marine & Freshwater Systems - PIRVic
Department of Primary Industries

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Mobile: +61400 845 406

Email: mehdi.doroudi@dpi.vic.gov.au
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PO Box 114, Queenscliff VIC 3225 Australia

---Original Message---
From: Mehdi.Doroudi@dpi.vic.gov.au [mailto:Mehdi.Doroudi@dpi.vic.gov.au]
Sent: Thursday, 8 June 2006 13:18
To: Crane, Mark (LI, Geelong)
Cc: malcolm.lancaster@dpi.vic.gov.au; Corbeil, Serge (LI, Geelong)
Subject: RE: URGENT - Comments on the pre-proposal

Mark, 

Apologies for the lack of effective communication - I made the assumption that I was communicating with Victoria via Malcolm but in retrospect we should have cc’d you on everything.

The only problem I have with this proposal is that it will take more than 12 months to develop a rapid and reliable detection and identification procedure (PCR-based) - minimum 2 years would be required. Information we have to-date indicates that this is a new virus - the ostreid herpesvirus PCR primers do not recognise this abalone herpesvirus and therefore we would have to start from ground zero. If everything went smoothly it might be quicker (18 months) but rarely do things go smoothly in research. One option would be to put more
than one person on the project here at AAHL.

Unfortunately without the diagnostic tools everything else is more difficult. I do not know how you can put in fully effective management systems without rapid and reliable diagnostics.

Mark

MARK CRANE Ph.D.
Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
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Geelong Vic 3220

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International Fax: +61 3 52 275555
email: mark.crane@csiro.au

-----Original Message-----
From: Mehdi.Doroudi@dpi.vic.gov.au [mailto:Mehdi.Doroudi@dpi.vic.gov.au]
Sent: Thursday, 8 June 2006 10:33
To: Crane, Mark (LI, Geelong); Malcolm.Lancaster@dpi.vic.gov.au
Subject: URGENT - Comments on the pre-proposal

Mark / Malcolm,

Your preproposal is about a long-term achievement (4 years of study) and is just focused on diagnostic techniques. I understand that diagnosis is your area of expertise and you may not have the answers to other questions, however, both industry and Vic DPI are after short-term resolutions to this outbreak. Today our stakeholders are after management solutions. There would always be a chance to develop long-term programs on this virus (PhD, POSTDOC etc.) the same as QX, Perkinus, Haplosporidium etc. From my perspective, the current focus should be on what could be achieved in a short period of time (max 12 months).

I was under the impression that you guys will discuss with me any further development of FRDC proposals on this matter (I think that we agreed on that previously) but obviously it did not happen until I approached you a couple of days ago. It is important to communicate effectively as we achieve more if we share our resources and expertise.

I have asked VicFRAB to extend the deadline for this proposal until lunchtime this Friday. There may be a chance to have an early start if the final proposal is approved by FRDC. Cash contribution could be provided through both Vic government and industry. I need you to go through my comments as soon as possible and let me know if we have a consensus on what I have proposed below or that you would prefer to pursue your prepropossl as it is now.

Specific Comments on Pre-Proposal:

Title:
Developing Management Strategies for Herpes-Like Virus Infection in Abalone

Leading Agency:
Vic DPI (PIRVic)

Collaborative agencies:
AAHL, CSIRO and Industry (both aquaculture and wild sector).

Principle Investigators:
Dr Mehdi Doroudi
Dr Serge Corbeil (Mark, please advise if we could have more than one PI)

Proposed Co-Investigators:
Dr Mark Crane
Dr Malcolm Lancaster

Duration of the project:
12 months

Budget:
To be reviewed if we agree on the proposed changes.

Need:
First paragraph, last sentence could be changed to "If appropriate management strategies including rapid diagnostic techniques are not developed this emerging virus may have the potential to adversely impact Australian abalone industry".

Prior to the last paragraph add following as a new paragraph:
In order to protect Victoria's valuable abalone industry, there is a need to develop specific management strategies which incorporates disease monitoring, detection, response and control measures both in farm and wild stocks. These include the implementation of routine health management procedures and a system for the exclusion of virus. Health management strategies for both farmed and wild abalone sectors and processing plants could be developed to improve productivity of farmed sector and to protect the health status of wild population of abalone. The application of effective detection and exclusion or control methods incorporates into a workable biosecurity plan to minimise the impacts of this virus.

The last paragraph of 'Need' should be simplified.

Objectives:
To improve physical, chemical and biological measures of biosecurity within abalone farming systems to prevent the introduction and spread of virus

To develop a "rapid and reliable diagnostic and detection method"

To develop a "code of practice" for commercial divers to avoid further spread of virus in wild population

To develop a "practical biosecurity program" for abalone processing plants

Mark, to save each other's time, please let me know if you are happy with my suggestions made so far then I will have more to contribute into Industry and Management Consultation, Direct Benefits, Design and Methodology etc.

The deadline is tomorrow. Your prompt response will be appreciated.

Regards

Mehdi Doroudi - DVM, PhD
Research Director
Marine & Freshwater Systems - PIRVic
Department of Primary Industries

Ph: +613 5258 0272  Fax: +613 5258 0270
Mobile: +61400 845 406

Email: mehdi.doroudi@dpi.vic.gov.au
2A Bellarine Hwy, Queenscliff VIC 3225 Australia
PO Box 114, Queenscliff VIC 3225 Australia
AQUATIC ANIMAL HEALTH SUBPROGRAM

2007-08 Preliminary Research Proposal

Project Title
Aquatic Animal Health Subprogram: Development of management strategies for herpes-like virus infection of abalone

FRDC Strategic challenge identification
Natural Resources Sustainability

Co-Principal Investigators Contact Details
Title: Dr Mehdi Doroudi Organisation: Vic DPI (PIRVic) Mailing Address: PO Box 114, Queenscliff VIC 3225
Phone No: 03 5258 0272 Fax No: 03 5258 0270 Email: serge.corbeil@csiro.au

Title: Dr Serge Corbeil Organisation: CSIRO Mailing Address: Private Bag 24, Geelong, Vic, 3220
Phone No: 03 5227 5254 Fax No: 03 5227 5555 Email: serge.corbeil@csiro.au

Commencement and completion date
Commencement date: 01-02-2007
Completion date: 31-01-2009

Preliminary Budget

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Need
In December 2005/January 2006, a disease outbreak caused high mortality rates in abalone from two farms in Victoria. A third Victorian farm also experienced disease but to a lesser extent. The abalone species affected by the outbreak are *Haliotis laevigata*, *H. rubra*, and *H. laevigata x H. rubra* hybrid. Histopathology performed on moribund animals indicated a ganglioncritis – infiltration of haemocytes in multiple ganglia and nerves (cerebral ganglion affected, also other ganglia and nerves). Examination by electron microscopy revealed the presence of a herpes-like virus in the pleuropedal ganglion. Preliminary transmission studies, carried out within AAHL’s high biosecurity facility, indicated that this emerging virus is highly pathogenic and can be transmitted to healthy abalone through the water column. In addition, the viral suspension remains infectious after undergoing a dilution up to 1 in 100. So far, the virus has not grown in tissue culture. Attemps by the farm managers to eliminate the disease from the abalone farms have failed partly due to the lack of detection methods specific to the aetiological agent which would allow early diagnosis of infected animals. If appropriate management strategies including rapid diagnostic techniques are not developed this emerging virus may have the potential to have a significant adverse impact on the Australian abalone industry.

In order to protect Victoria's valuable abalone industry, there is a need to develop specific management strategies which incorporate disease monitoring, detection, response and control measures both in farm and wild stocks. These include the
implementation of routine health management procedures and a system for the exclusion of virus. Health management strategies for both farmed and wild abalone sectors and processing plants could be developed to improve productivity of farmed sector and to protect the health status of wild population of abalone. The application of effective detection and exclusion or control methods incorporates into a workable biosecurity plan to minimise the impacts of this virus.

Access to diagnostic tests that are rapid, reliable and sensitive is of fundamental importance for effective control/management of disease outbreaks. In addition to surveillance tools such as PCR, better procedures/reagents for disease diagnosis are required. Presence of histological lesions provides a presumptive diagnosis. The development of in situ hybridisation probe(s) for the localisation of abalone herpesvirus within histological lesions will provide a means for definitive diagnosis to be made. The development of molecular tools and reagents will allow researchers and industry to rapidly and specifically detect and locate the virus in abalone tissues therefore providing a vital means for diagnosis and facilitating a better understanding of the epidemiology of this disease, leading to more efficient management of disease outbreaks.

**Objectives**

1. To improve physical, chemical and biological measures of biosecurity within abalone farming systems to prevent the introduction and spread of virus.
2. To develop a “code of practice” for commercial divers to avoid further spread of virus in wild populations.
3. To develop a “practical biosecurity program” for abalone processing plants.
4. To identify nucleic acid sequences of the emerging abalone herpesvirus (via PCR-based gene amplification and sequencing) necessary for the development of diagnostic tools (e.g. conventional PCR, TaqMan PCR, ISH probe).
5. To develop and validate PCR assays for the detection of the abalone virus.
6. To develop an in situ hybridisation assay specific for the abalone herpesvirus.
7. To document an Australian and New Zealand Standard Diagnostic Procedure and submit for external review.

**Industry and Management consultation**

The abalone Growers Association of Victoria strongly supports the proposal.

VicFRAB
Abalone Subprogram
AAH Subprogram
NSW FRAB
QFIRAC
SAFRAB
WA FRAB
Tas FRAB

**Direct benefits and beneficiaries**

1. All sectors of the abalone wild-capture and aquaculture industries (farmers, divers, processors) will directly benefit from increased biosecurity developed through the implementation of this project.
2. The diagnostic tests will be available to detect and identify the emerging virus in abalone (*Haliotis* spp.), and other potential host mollusc species.
3. Export certification services will be available to industries that wish to develop export markets as well as translocate farmed stock between regions without transmitting disease. In addition, should pathogenic agents be detected during health surveys, industries and State officers will be able to make informed decisions with regards to brood stock translocation, stock destruction etc. Specifically, mollusc aquaculture industries and State agencies will be able to develop health surveillance programs in collaboration with AFDL. In addition, Australia will be better prepared to negotiate with international trading partners on issues concerned with the importation of disease free molluscs from Australian sources.

**Estimated Flow of Benefits**

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Project Design and Methodology
- Training workshops for government and industry representatives will be held to develop practical biosecurity measures that are agreed to by both government and industry.
- The subtractive DNA hybridisation method will be used to clone viral gene fragments from infected abalone tissues. Cloned gene fragments will be sequenced and blasted against gene data banks in order to find existing homologous viral genes.
- Primers will be synthesised to perform a walking PCR strategy in order to get appropriate gene length for the development of PCR assays.
- Herpesvirus nucleic acid sequences suitable for use as a diagnostic probe will be developed for in situ hybridisation.
- Infection trials of healthy abalone will be performed within the AAHL biosecure facility to determine which abalone tissues are the most appropriate for sampling during an active surveillance program. For example, wild broodstock.
- Procedures for the detection and identification of the abalone virus will be incorporated into an Australian and New Zealand Standard Diagnostic Procedure (ANZSDP) and submitted to SCAHLS for review and publication.

Research Capability and Experience

Mehdi Doroudi

Serge Corbeil: BSc MSc PhD. Eleven years experience in aquatic animal disease research and diagnosis (viral, protozoan and bacterial diseases). Nine years experience in molecular diagnosis (conventional PCR, real-time PCR, gene sequencing) and immunodiagnosis of aquatic animal diseases. Five years experience in vaccine R&D for mammalian and fish diseases.

Previous FRDC Projects

Relevant Publications

Please forward the Preliminary Research Proposal in a MS-Word format via e-mail to:

Ms Joanne Slater
Coordinator, Aquatic Animal Health Subprogram

E-MAIL: joanne.slater@csiro.au

No later than cob on Friday 23 June 2006.
DOCUMENT 55

EXEMPT IN FULL – s 47B(a)
Thanks Mark. We will further discuss it with the industry today. Let's hope there will be a significant level of cash contribution from both industry and DPI. I will keep you guys posted with the outcomes. Regards

Mehdi Doroudi - DVM, PhD
Research Director
Marine & Freshwater Systems - PIRVic
Department of Primary Industries

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Mobile: +61400 845 406

Email: mehdi.doroudi@dpi.vic.gov.au
2A Bellarine Hwy, Queenscliff VIC 3225 Australia
PO Box 114, Queenscliff VIC 3225 Australia

To: Mehdi.Doroudi@dpi.vic.gov.au
cc: malcolm.lancaster@dpi.vic.gov.au, Serge.Corbell@csiro.au
Subject: RE: URGENT - Comments on the pre-proposal

Mehdi,

I have accepted most of your changes - corrected some minor grammatical/typographical errors - and reduced so me blurb so that it was less than 3 pages. I have also added some extra salary for 1.0 FTE at Attwood for one year, based on Malcolm's email.

Hope this is the final version :-)

Cheers

Mark

MARK CRANE Ph.D.
Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
Private Bag 24
Geelong Vic 3220
2007-08 Preliminary Research Proposal

Project Title
Aquatic Animal Health Subprogram: Development of management strategies for herpes-like virus infection of abalone

FRDC Strategic challenge identification
Natural Resources Sustainability

Co-Principal Investigators Contact Details

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<th>Organisation</th>
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<td>Dr Mehdi Doroudi</td>
<td>Vic DPI (PIRVic)</td>
<td>PO Box 114, Queenscliff VIC 3225</td>
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<td>Dr Serge Corbeil</td>
<td>CSIRO LI</td>
<td>Private Bag 24, Geelong, Vic, 3220</td>
<td><a href="mailto:serge.corbeil@csiro.au">serge.corbeil@csiro.au</a></td>
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Commencement and completion date
Commencement date: 01-02-2007
Completion date: 31-01-2009

FRDC Budget

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Need
Recent disease outbreaks in Victorian abalone farms associated with high mortality rates have been shown to be caused by a previously unknown herpes-like virus. The abalone species affected by the outbreak are *Haliotis laevigata*, *H. rubra*, and *H. laevigata x H. rubra* hybrid. The disease has now been detected in wild abalone populations in the vicinity of one of the affected farms.

Histopathology performed on moribund animals indicated a ganglionitis – infiltration of haemocytes in multiple ganglia and nerves (cerebral ganglion affected, also other ganglia and nerves). Examination by electron microscopy revealed the presence of a herpes-like virus in the pleuropedal ganglion. Preliminary transmission studies, carried out within AAHL’s high biosecurity facility, indicated that this emerging virus is highly pathogenic and can be transmitted to healthy abalone through the water column. In addition, the viral suspension remains infectious after undergoing a dilution up to 1 in 100. So far, the virus has not grown in tissue culture.

Attempts by the farm managers to eliminate the disease from the abalone farms have failed partly due to the lack of detection methods specific to the aetiological agent which would allow early diagnosis of infected animals. If appropriate management strategies including rapid diagnostic techniques are not developed this emerging virus may have the potential...
to have a significant adverse impact on the Australian abalone industry.

In order to protect Australia’s valuable abalone industry, there is a need to develop specific management strategies which incorporate disease monitoring, detection, response and control measures both in farm and wild stocks. These include the implementation of routine health management procedures and a system for the exclusion of virus. Health management strategies for both farmed and wild abalone sectors and processing plants could be developed to improve productivity of farmed sector and to protect the health status of wild populations of abalone. The application of effective detection and exclusion or control methods incorporates into a workable biosecurity plan to minimise the impacts of this virus.

Access to diagnostic tests that are rapid, reliable and sensitive is of fundamental importance for effective control/management of disease outbreaks. In addition to surveillance tools such as PCR, better procedures/reagents for disease diagnosis are required. Presence of histological lesions provides a presumptive diagnosis. The development of in situ hybridisation probe(s) for the localisation of abalone herpesvirus within histological lesions will provide a means for definitive diagnosis to be made. The development of molecular tools and reagents will allow researchers and industry to rapidly and specifically detect and locate the virus in abalone tissues therefore providing a vital means for diagnosis and facilitating a better understanding of the epidemiology of this disease, leading to more efficient management of disease outbreaks.

**Objectives**

1. To improve physical, chemical and biological measures of biosecurity for abalone farms to prevent the introduction and spread of virus.
2. To develop a “code of practice” for commercial divers to avoid the introduction and further spread of virus in wild populations of abalone.
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Previous FRDC Projects


Relevant Publications

Ann,

As discussed by telephone, Victoria and AAHL have revised the preproposal to include development of specific management strategies which incorporate disease monitoring, detection, response and control measures in both farm and wild stocks.

In order to keep costs as low as possible we have limited the development of diagnostic tests to the most essential (AAHL may develop further tests in-house for other specific purposes). Nevertheless, the project remains relatively big with a significant budget. Industry cash contributions will be required to reduce the cost to FRDC and to ensure that this project proposal is funded. Mehdi (representing the lead agency) will be following up on these financial issues.

Serge will be logging the preproposal on FISHNET some time today and I will forward the preproposal to other relevant FRABs.

Regards

Mark

FRDC Abalone herpesvirus pre-p..

MARK CRANE Ph.D.
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Australian Animal Health Laboratory
CSIRO Livestock Industries
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Geelong Vic 3220

International Phone: +61 3 52 275118
International Fax: +61 3 52 275555
email: mark.crane@csiro.au
AQUATIC ANIMAL HEALTH SUBPROGRAM

2007-08 Preliminary Research Proposal

Project Title
Aquatic Animal Health Subprogram: Development of management strategies for herpes-like virus infection of abalone

FRDC Strategic challenge identification
Natural Resources Sustainability

Co-Principal Investigators Contact Details

Title: Dr Mehdi Doroudi Organisation: Vic DPI (PIRVic) Mailing Address: PO Box 114, Queenscliff VIC 3225
Phone No: 03 5258 0272 Fax No: 03 5258 0270 Email: mehdi.doroudi@dpi.vic.gov.au

Title: Dr Serge Corbeil Organisation: CSIRO L1 Mailing Address: Private Bag 24, Geelong, Vic, 3220
Phone No: 03 5227 5254 Fax No: 03 5227 5555 Email: serge.corbeil@csiro.au

Commencement and completion date
Commencement date: 01-02-2007
Completion date: 31-01-2009

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Need
Recent disease outbreaks in Victorian abalone farms associated with high mortality rates have been shown to be caused by a previously unknown herpes-like virus. The abalone species affected by the outbreak are Haliotis laevigata, H. rubra, and H. laevigata x H. rubra hybrid. The disease has now been detected in wild abalone populations in the vicinity of one of the affected farms.

Histopathology performed on moribund animals indicated a ganglion neuritis – infiltration of haemocytes in multiple ganglia and nerves (cerebral ganglion affected, also other ganglia and nerves). Examination by electron microscopy revealed the presence of a herpes-like virus in the pleuropedal ganglion. Preliminary transmission studies, carried out within AAHL’s high biosecurity facility, indicated that this emerging virus is highly pathogenic and can be transmitted to healthy abalone through the water column. In addition, the viral suspension remains infectious after undergoing a dilution up to 1 in 100. So far, the virus has not grown in tissue culture.

Attempts by the farm managers to eliminate the disease from the abalone farms have failed partly due to the lack of detection methods specific to the aetiological agent which would allow early diagnosis of infected animals. If appropriate management strategies including rapid diagnostic techniques are not developed this emerging virus may have the potential
to have a significant adverse impact on the Australian abalone industry.

In order to protect Australia's valuable abalone industry, there is a need to develop specific management strategies which incorporate disease monitoring, detection, response and control measures in both farm and wild stocks. These include the implementation of routine health management procedures and a system for the exclusion of virus. Health management strategies for both farmed and wild abalone sectors and processing plants could be developed to improve productivity of farmed sector and to protect the health status of wild populations of abalone. The application of effective detection and exclusion or control methods incorporates into a workable biosecurity plan to minimise the impacts of this virus.

Access to diagnostic tests that are rapid, reliable and sensitive is of fundamental importance for effective control/management of disease outbreaks. In addition to surveillance tools such as PCR, better procedures/reagents for disease diagnosis are required. Presence of histological lesions provides a presumptive diagnosis. The development of in situ hybridisation probe(s) for the localisation of abalone herpesvirus within histological lesions will provide a means for definitive diagnosis to be made. The development of molecular tools and reagents will allow researchers and industry to rapidly and specifically detect and locate the virus in abalone tissues therefore providing a vital means for diagnosis and facilitating a better understanding of the epidemiology of this disease, leading to more efficient management of disease outbreaks.

Objectives

1. To improve physical, chemical and biological measures of biosecurity for abalone farms to prevent the introduction and spread of virus.
2. To develop a "code of practice" for commercial divers to avoid the introduction and further spread of virus in wild populations of abalone.
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Previous FRDC Projects


Relevant Publications

Hi Mark

I have just returned from a meeting in France and while there I visited the IFREMER laboratories at La Tromblade to see their oyster research. During various discussions the issue of viruses came up and some discussion was held on abalone herpes-like virus. My contact there Pierre Boudry mentioned one of his colleagues (not there at the time) had worked on this issue both in oysters and in abalone in Taiwan. I cannot find my notes right know with his name (may have been Tristan Renault) but I did ask Pierre to pass on your contact details to him – so he may already have contacted you. If not, and if you are not already aware of that group I can follow things up with Pierre.

In your plans with the Vic DPI and industry, has thought been given to the development of a suitable biosecure challenge facility that could be utilised for the breeding program work?

How are the plans going for funding?

Cheers
Nick
Only research they were the creatures that we got directly from Portland to infect the naive abs with in the LAF and I thought it was worth a go as they were the freshest samples we would ever get.

-----Original Message-----
From: Crane, Mark (LI, Geelong)
Sent: Tuesday, 11 July 2006 2:42 PM
To: Williams, Nette (LI, Geelong)
Subject: RE:

Nette,

I do not have any info on 06-0442. Is this research only rather than a diagnostic sample?

Mark

MARK CRANE Ph.D.
Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
Private Bag 24
Geelong Vic 3220

International Phone: +61 3 52 275118
International Fax: +61 3 52 275555
email: mark.crane@csiro.au

-----Original Message-----
From: Williams, Nette (LI, Geelong)
Sent: Tuesday, 11 July 2006 14:35
To: Crane, Mark (LI, Geelong)
Subject:

06 0442

Abalone from "known infected abalone from Portland" that I tried to isolate virus form.

Do I need to do a report or is all this covered previously?

Nette
Hi Malcolm,

The FRDC Aquatic Animal Health Subprogram rec'd the DPI preproposal. I noted that there is no ISH probe development in it - does this mean that DPI VIC are no longer interested in a probe? As Subprogram Leader I am going to have to give advice to the preproposal PIs, following assessment of the preproposals and Serge is going to be asking for some direction.

Mark

MARK CRANE Ph.D.
Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
Private Bag 24
Geelong Vic 3220

International Phone: +61 3 52 275118
International Fax: +61 3 52 275555
e-mail: mark.crane@csiro.au
Mehdi,

We can always tailor our proposals to a specific budget. Getting a PCR is easy. Getting a PCR that does what you want and demonstrating that is the case is not so easy. I am not sure how Attwood can do that especially since they seem to be under the misapprehension that aquatic herpes-like viruses are like mammalian herpesviruses.

MARK CRANE Ph.D.
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email: mark.crane@csiro.au

-----Original Message-----
From: Mehdi.Doroudi@dpi.vic.gov.au [mailto:Mehdi.Doroudi@dpi.vic.gov.au]
Sent: Wednesday, 9 August 2006 16:46
To: Crane, Mark (LI, Geelong)
Subject: Re: Abalone Up-date

Mark,

I agree with the points that you have made in relation to the evaluation of PCR techniques etc. However, we need to remember that the main reason for industry to ask for a new proposal (from Attwood) was the costs associated with the development of the techniques.

I will discuss your email with the abalone reference group on next Monday but I am sure that they will say the same thing that AAHL proposal is too expensive.

You may want to discuss the possibility of collaborative work between AAHL and Attwood with either Malcolm or Catherine Ainsworth prior to Adelaide meeting. I will probably be in Adelaide.

Regards

Mehdi Doroudi - DVM, PhD
Research Director
Marine & Freshwater Systems - PIRVic
Department of Primary Industries

Ph: +613 5258 0272 Fax: +613 5258 0270
Mobile: +61400 845 406
Hi Mehdi,

I just thought I would up-date you on the latest concerning FRDC and abalone herpesvirus diagnosis. Because there were two potentially competing applications (Malcolm's and Serge's) the FRDC Aquatic Animal Health Subprogram committee deferred evaluation. It is my impression that this issue will be discussed at the Abalone Workshop in Adelaide and, maybe, some progress can be made there. I explained to the committee as well as I could what had been happening re: DPI VIC/AAHL etc. AAHS recommended revised application(s) to be submitted after the Adelaide Conference.

AAHL is still keen to work on the virus - evaluation of any PCR technique will require experimental infections as a source of infected material. Infection trials will need to be undertaken to titrate the virus and come up with some semi-quantitative infective dose. Without this the specificity and sensitivity of the diagnostic methods cannot be assessed.

Crispian (FRDC) recommended coordination through Peter Appleford’s group.

What is your opinion on how we can progress this issue sensibly? Malcolm has left it up to me to present the papers at the Adelaide Conference - he has a conflict in his diary and cannot make it. I am not sure how other interested States/industry groups/FRDC will view this. As you know, I am happy/keen to collaborate but there needs to be open communication up front rather than what happened previously.

Mark

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Project Leader
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International Phone: +61 3 52 275118
International Fax: +61 3 52 275555
e-mail: mark.crane@csiro.au
Hi Serge,

I have found that there were two lots of abalone in the February trial. SAN 06-00442 were the diseased specimens from Portland and 06-00441 were healthy specimens from Indented Head. They were both used in the transmission trial. As I understand it all of the specimens under SAN 06-00441 were used in the February trial. Of the specimens under SAN 06-00442 Mark has noted on the file that 10x were to be used live in the co-habitation trial, 10x were to be sacrificed and have their ganglia dissected out for use in that trial, 20x were to be frozen intact, 2x were to be sent to EM for examination, 2x were to go to Histology for examination and 6x were to be sacrificed and inoculated onto cell cultures in an attempt to grow the herpes virus.

Therefore the SAN that these samples come from depends on whether the abalone are from Indented Head (06-00441) or from Portland (06-00442). A new number would be reasonable to catalogue specimens created for each separate experiment. That is too late for the experiments already conducted, but it looks like SAN 06-00441 would cover any samples taken from the Indented Head specimens both before and after they were included in the co-habitation trial. I expect the specimens from SAN 06-00442 were just used to deliver the virus and not sampled at any stage. If so the whole experiment could easily be catalogued under SAN 06-00441.

Does that answer your question?

John

-----Original Message-----
From: Corbeli, Serge (LI, Geelong)  
Sent: Friday, 11 August 2006 12:20  
To: Young, John (LI, Geelong)  
Subject: RE: san

Hi John,

The samples were from the February cohab transmissibility exp. Therefore I will tell Diane that the san number is 06-00442, can you confirm this?

Serge

-----Original Message-----
From: Young, John (LI, Geelong) 
Sent: Friday, 11 August 2006 11:13 
To: Corbeli, Serge (LI, Geelong) 
Subject: RE: san

Hi Serge,

I sure can, but would like more information. I entered into LIMS a job 06-00442 in February. Though I have nothing recorded on what that was for I guess it was the original cohabitation transmissibility and infection work. I also entered jobs 60-1154 to 60-1156 in April to test the transmissibility of the abalone herpes virus through contaminated water. No samples have been recorded in LIMS for these experiments. If these samples are from those experiments I can add them as extra samples for analysis by the histo group. If they are new experiments we should give the whole experiment a new number then add these samples to that.

Regards,

John

-----Original Message-----
From: Corbeli, Serge (LI, Geelong)
Hi John,

I have 3 pieces of abalone, originating from the recent infection experiments carried out in the LAF, that I have fixed and brought to the histo lab for embedding and sectioning. Can you provide me with a san number for these even though they were not diagnostic samples sent to us?

Thanks,

Serge
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**PRICING DECISION STATEMENT**

**Type of Activity**
- Consulting or Technical Service
- Research - Contract
- Research - Collaborative - subject to CN
- Research - Collaborative - NOT subject to CN

| COSTS: |  |
| Direct Costs (less Costs of Sub-contracts) | 26,322 |
| Indirect Costs | 8,023 |
| Cost of Sub-Contracted Activities | - |
| Full-cost Price | 34,345 |
| Competitive Neutrality | 5,220 |
| Full-cost Price plus competitive neutrality | 39,565 |

**PRICE:**
- Cash Contribution | 15,400 |
  59% DIRECT COSTS ;  45% TOTAL COSTS
- CSIRO - Livestock Industries In-kind Contributions | 18,945 |

**Justification (if applicable) (2):**

Prepared by: ___________________________  Recommended by: ___________________________
Project Leader  Commercial Manager

Approved by: ___________________________  Date: ___________________________
Delegate

**NOTES:**

1. Other forms of Consideration include long term value of licence fees and royalties
2. Justifications for prices lower than Full-cost Price include National interest (specify it) and commercial considerations
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From: Crane, Mark (LI, Geelong)
Sent: Friday, 18 August 2006 2:46 PM
To: Elliott, Nick (CMAR, Hobart)
Cc: Walker, Peter (LI, Geelong); Corbeil, Serge (LI, Geelong); McColl, Ken (LI, Geelong)
Subject: Further developments re: Abalone herpesvirus

Follow Up Flag: Follow up
Flag Status: Completed

Nick,

Harry Peters (EO, Western Victorian Abalone Divers Association) and I managed to catch up yesterday. Apparently, with funding from the industry, the association is trying to get three mollusc disease experts (Carolyn Friedman, Mike Hine and Tristan Renault) from overseas to come to Australia for a workshop on the abalone herpesvirus. I informed Harry that we had already been in contact with Tristan and were attempting to set up an initial collaboration and I also offered AAHL to host the workshop. Harry is trying to arrange for the o/s experts to visit the Victorian farms and then follow up the farm visits with laboratory sessions (histopathology, electron microscopy). He is hoping to do this for the week of 16-23 September. It is likely that at least one day would be spent at Attwood but he understood that it would be useful to have the visitors come to AAHL.

He also stated that the industry are more confident in CSIRO delivering the PCR than Attwood (but CSIRO was too expensive) - he seemed to think that there were internal conflicts within DPI VIC - for example, he knows that an initial report on the epidemiology of the disease was prepared in January but was not released until much later - around 6 months later - and suggested that the industry would have responded differently had they known the contents of the report. I also indicated that we could submit a "PCR-only" proposal which would be cheaper than the all-encompassing project. I suggested that he needed to let FRDC know what industry wanted and which organisation should be contracted to undertake the work.

In addition, I had a phone call today from Kevin Ellard (DPIW, Tasmania) - he needed some clarification on some of our results for a report he was writing for the CVO. He said that the department had met with industry earlier this week to discuss the department's management of the situation. Interestingly, the industry asked the department who should undertake the development of a diagnostic PCR and AAHL was recommended.

Kevin and I discussed other basic questions that needed to be answered and he was in favour of a much broader project rather than a PCR-only project. Some basic questions needing to be answered are, of course, stability of the virus in the environment, chemical inactivation, comparison with the Taiwanese virus etc. He seemed to think that we could get direct funding from the industry. Clearly, this issue is, now, seen as an industry-wide problem and not just a Victorian problem - perhaps, contrary to Victoria's belief, Victoria do not need to be the lead agency. Moreover, outside of DPI Victoria, stakeholders seem to be saying that the Victorian response to the virus has, in fact, added to the problem rather than help resolve it.

Perhaps the meeting in Adelaide will be instrumental in clarifying the research needs and the potential sources of funding even more than I originally thought. Perhaps, since the wild-capture industry is much bigger than the aquaculture industry and they are deeply concerned about this virus, they will be more influential and may contribute more to the discussion as well as to the funding debate.

Will Serge receive an evaluation of the preproposal from TASFRAB, at some stage?

Cheers

Mark

MARK CRANE Ph.D.
Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
Private Bag 24
Geelong Vic 3220
Hi Mehdi,

I just thought I would update you on the latest concerning FRDC and abalone herpesvirus diagnosis. Because there were two potentially competing applications (Malcolm’s and Serge’s) the FRDC Aquatic Animal Health Subprogram committee deferred evaluation. It is my impression that this issue will be discussed at the Abalone Workshop in Adelaide and, maybe, some progress can be made there. I explained to the committee as well as I could what had been happening re: DPI VIC/AAHL etc. AAHS recommended revised application(s) to be submitted after the Adelaide Conference.

AAHL is still keen to work on the virus - evaluation of any PCR technique will require experimental infections as a source of infected material. Infectivity trials will need to be undertaken to titrate the virus and come up with some semi-quantitative infective dose. Without this the specificity and sensitivity of the diagnostic methods cannot be assessed.

Crispian (FRDC) recommended coordination through Peter Appleford’s group.

What is your opinion on how we can progress this issue sensibly? Malcolm has left it up to me to present the papers at the Adelaide Conference - he has a conflict in his diary and cannot make it. I am not sure how other interested States/industry groups/FRDC will view this. As you know, I am happy/keen to collaborate but there needs to be open communication up front rather than what happened previously.

Mark

MARK CRANE Ph.D.
Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
Private Bag 24
Geelong Vic 3220

International Phone: +61 3 52 275118
International Fax: +61 3 52 275555
email: mark.crane@csiro.au
Mark,

Please find attached a copy of Anna Mouton's Itinerary while she is in Australia. The basis of her visit to Australia is for her to provide DPI with an independent review of the epidemiology study undertaken to date.

On Wednesday, 20 September 2006 both Anna and myself intend to visit AAHL from 12:00-1:00pm. Can you please confirm this time is suitable for yourself.

Regards,

Mehdi Doroudi - DVM, PhD
Research Director
marine & Freshwater Systems - PIRVic
Department of Primary Industries

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Mobile: +61400 845 406

Email: mehdi.doroudi@dpi.vic.gov.au
2A Bellarine Hwy, Queenscliff VIC 3225 Australia
PO Box 114, Queenscliff VIC 3225 Australia

(See attached file: Itinerary - Dr Anna Mouton - Sept 2006.doc)
Dr Anna Mouton

ITINERARY

Department of Primary Industries

SATURDAY, 16 SEPTEMBER 2006

TBA

Dr Mouton flight Capetown, South Africa to Melbourne, Australia

SUNDAY, 17 SEPTEMBER 2006

TBA

Dr Mouton arrives at Tullamarine, Melbourne Airport
Mehdi Doroudi and Hugh Millar to collect Dr Mouton and drive her
accommodation.
Accommodation: Airport Motel, Attwood

MONDAY, 18 SEPTEMBER 2006

9-10:30am
Introductory meeting with Mehdi Doroudi, Peter Appleford, Hugh
Millar, (Sally Ridge, Andrew Cameron), Attwood

10:30-12:30pm
Mehdi Doroudi & Anna Mouton – Drive to Portland to visit CS Farm

5:00pm
Drive to Warrnambool. Stay in Warrnambool Monday night

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Internal DPI workshop – suggested participants, in addition to
introductory meeting, invite Malcolm Lancaster, Mike Jeffers, Dallas
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9-11:00am  Meet with Abalone Reference Group

11-5:00pm  Dr Mouton to work on consultancy report

SATURDAY, 23 SEPTEMBER 2006

TBA  Return Flight to South Africa
Mark

As outlined below Anna Mouton is going to be in Australia. We were wondering if she may be appropriate for the epidemiology for the Perkinsus project, PI Geoff Liggins.

Your thoughts?

Regards

Crispian

-----Original Message-----
From: emma.young@dpi.vic.gov.au [mailto:emma.young@dpi.vic.gov.au] On Behalf Of Mehdi.Doroudi@dpi.vic.gov.au
Sent: Friday, 25 August 2006 1:23 PM
To: Crispian Ashby
Cc: Patrick Hone; Peter.Appleford@dpi.vic.gov.au; Hugh.Millar@dpi.vic.gov.au
Subject: Visiting Abalone Disease Expert

Crispian,

As requested please find attached a copy of Anna Mouton's brief CV and her Itinerary while she is in Australia.

The basis of this invitation is for her to provide DPI with an independent review of the epidemiology study undertaken to date.

We are going to form a technical scientific forum on Thursday, 21 September in order to invite relevant abalone disease experts from other jurisdictions to share our knowledge and seek their input in the further research and development required to overcome this disease. This exercise will have significant input into the current short-term FRDC project (development of management strategies for herpes virus infection in abalone).

As you know the wild abalone sector is planning to invite overseas experts at the same time. If you need further information on the nature of this invitation and the terms of reference for them you would need to speak to Harry Peeters (ph: 0417 119 577). Harry, as I understand, has been organising this visit. I encouraged Harry Peeters to liaise with us in terms of the maximum achievement from bringing all these experts together, however I personally believe this exercise is unnecessary.

Regards,

Mehdi Doroudi - DVM, PhD
Research Director
Marine & Freshwater Systems - PIRVic
Department of Primary Industries
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2A Bellarine Hwy, Queenscliff VIC 3225 Australia PO Box 114, Queenscliff
VIC 3225 Australia

(See attached file: Anna Mouton - Resume.doc) (See attached file:
Itinerary
- Dr Anna Mouton - Sept 2006.doc)

This email has been scanned by the MessageLabs Email Security System.
For more information please visit http://www.messagelabs.com/email

The Fisheries Research and Development Corporation plans, invests in and manages fisheries research and development throughout Australia. It is a federal statutory authority jointly funded by the Australian Government and the fishing industry. If I sent this e-mail message to you in error, please accept my apologies. I would appreciate a return e-mail or telephone call (02 6285 0400) so that I can prevent the error from happening again. I ask that you do not distribute or print any part of a wrongly sent e-mail message, or take any action as a result of knowing its contents, but that you destroy all copies and any attachment(s).
Your cooperation is appreciated.
RESUME: Anna Mouton

A. Personal details

Title : Dr

Full names : Anna-Louise Mouton

Address : PO Box 967
           Stanford
           7210 South Africa

Contact number : +27 28 341 0391

Nationality : South African

B. History

I obtained a BVSc from the University of Pretoria in 1995 and a BSc from the University of South Africa in 2002. After I qualified as a veterinarian, I initially worked in private practice, both in England and South Africa. From September 1996 to the end of January 2001, I was with the Onderstepoort Veterinary Institute, primarily to do diagnostic bacteriology and was the deputy head of the reference veterinary bacteriology laboratory for the country. My involvement in aquatic diagnostics began at Onderstepoort and I initiated a health management program for the abalone industry. During this time, I spent a month training under Carolyn Friedman at the Bodega Marine Laboratory of the University of California. After Onderstepoort, I was with the Department of Agriculture in the Western Cape, based at their regional veterinary laboratory in Stellenbosch. I was responsible for all aquatic diagnostic services, including investigation of disease and mortalities in fish and shellfish, running health management programs for abalone and koi production units, consulting to the commercial aquaculture sector, and advising government bodies on aquaculture policy. During this time, I developed a vaccine against streptococcus in trout which is still successfully used by the Mpumalanga trout industry. In addition, I was responsible for diagnostic bacteriology and food hygiene services. Since October 2002, I have been directly employed by the Abalone Farmers’ Association of South Africa under an industry government partnership in conjunction with Marine and Coastal Management of the Department of Environmental Affairs and Tourism.
C. Papers presented

- **The occurrence of bacterial fish diseases in South Africa**
  Fourth Congress of the Aquaculture Association of South Africa
  Stellenbosch, South Africa, 1997

  repeated by invitation
  Regional Workshop on Aquaculture
  Lilongwe, Malawi, 1997

- **Health management and disease surveillance in abalone, *Haliotis midae*, in South Africa**
  Fourth International Abalone Symposium
  Cape Town, South Africa, 2000

- **Undesirable aliens: the importation of fish diseases**
  Fifth Congress of the Aquaculture Association of South Africa
  Pretoria, South Africa, 2000

- **The current status of abalone health management in South Africa**
  and
  **Histological changes associated with stress in intensively cultured South African abalone, *Haliotis midae***
  Fifth International Abalone Symposium
  Qingdao, China, 2003

  repeated by invitation
  Tenth Annual Abalone Aquaculture Workshop
  Port Lincoln, Australia, 2003

- I was invited as guest speaker to the Abalone Health Workshop held in conjunction with the Australasian Aquaculture Conference in 2004. I gave twelve talks on various issues related to abalone health.

- **Factors affecting the prevalence of gut associated parasites in the South African abalone, *Haliotis midae***
  Sixth International Abalone Symposium
  Puerto Varas, Chile, 2006
D. Research interests

I am currently involved in the projects listed below.

- **The epidemiology of parasites infecting South African abalone, *Haliotis midae*, in western Cape aquaculture facilities**
  - principal investigator

- **A national survey of abalone (*Haliotis midae*) health**
  - principal investigator

- **Oxygen and ammonia as limiting factors in abalone (*Haliotis midae*) aquaculture systems: husbandry, handling protocols, energetics and growth**
  - coresearcher

- **The characterization of an intracellular bacterium infecting the digestive gland of the South African abalone, *Haliotis midae***
  - cosupervisor

- **The characterisation of an intracellular protozoan parasite infecting the digestive gland of abalone, *Haliotis midae***
  - cosupervisor

E. Present responsibilities

I am currently responsible for all aspects of the abalone health management program. The program includes routine herd health, diagnostic examinations and disease surveillance. The primary aim of the program is to optimise abalone health for production purposes. Abalone movement within South Africa is not officially regulated, but the Abalone Farmers' Association of South Africa has adopted a voluntary stock movement protocol. I developed this protocol and am responsible for its implementation, including all health examinations of abalone. In addition, I act as advisor to Marine and Coastal Management, the official regulatory body, on importation of live abalone. Other responsibilities include compiling the annual industry survey, which is a survey of the technical aspects of abalone production. Results are presented on a comparative basis, with each participating unit receiving their figures relative to the industry as a whole.
Dr Anna Mouton

ITINERARY

Department of Primary Industries

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11-5:00pm  Dr Mouton to work on consultancy report

SATURDAY, 23 SEPTEMBER 2006

TBA  Return Flight to South Africa
Hi Mehdi,

Thank you for the invitation which I accept and I look forward to participating in the Scientific Forum.

Cheers

Mark

MARK CRANE Ph.D.
Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
Private Bag 24
Geelong Vic 3220

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International Fax: +61 3 52 275555
email: mark.crane@csiro.au

-----Original Message-----
Sent: Friday, 8 September 2006 13:58
Subject: Abalone Virus - Scientific Forum - 21 September 2006
Importance: High

Dear All,

The outcomes of the recent investigation into unusual mortality of abalone at four Victorian aquaculture farms and wild abalone in close proximity to one of the affected farms have confirmed the presence of a virus as the cause of mortality. The virus causes ganglioneuritis in abalone. An epidemiology study has been undertaken to provide us with a better understanding of the nature of this outbreak. The Department of Primary Industries Victoria and Western Abalone Divers Association have invited international experts including Dr Anna Mouton (from South Africa), Dr Carolyn Friedman (from USA), Dr Mike Hine (from New Zealand) and Dr Tristan Renault (from France) to provide us with an independent review of the epidemiology study undertaken to date and further advice.

To maximise the benefits and to share our current knowledge and information with each other, DPI / WADA would like to extend an invitation to you to participate in an Abalone Virus - Scientific Forum on Thursday, 21 September 2006 from 10:00 am to 3:00 pm at DPI, 1 Spring Street, Melbourne. The purpose of the forum is:

to provide you with an update on the current status of the outbreak and its management response and

to identify / discuss the current knowledge gaps and research priorities
required to address future biosecure management issues.

There are limited places available for the forum. Please nominate a representative from your organisation to attend. RSVPs are due by COB, Friday 15 September 2006. Please RSVP to Caroline McGowan either by email at caroline.mcgowan@dpi.vic.gov.au or by phone on 03 5258 0266. Please note that you are responsible for any costs incurred.

An agenda for the forum will be forwarded closer to the date.

I look forward to seeing you on the 21st September.

Regards,

Mehdi Doroudi - DVM, PhD
Research Director
Marine & Freshwater Systems - PIRVic
Department of Primary Industries

Ph: +613 5258 0272  Fax: +613 5258 0270
Mobile: +61400 845 406

Email: mehdi.doroudi@dpi.vic.gov.au
2A Bellarine Hwy, Queenscliff VIC 3225 Australia
PO Box 114, Queenscliff VIC 3225 Australia
Hi Alex,

This coming Monday 18th September AFDL is hosting a meeting to discuss the herpes-like virus outbreak in abalone. Members of the abalone diving association will be present as well as 3 international experts on mollusc diseases (Tristan Renault from Ifremer, Carolyn Friedman from Washington Uni and Mike Hine from NZ). I thought you could be interested in coming along to have a chat with these experts and show few EM photos of our herpes-virus on Power Point. The meeting will be held in the board room at around 11:30 am (exact time is not available because our guest will be driving from Port Fairie). Let me know if you are interested in attending the meeting so that can arrange lunch for the appropriate number of people.

Serge
Hi guys:

A number of international (and national) abalone experts are visiting Victoria next week, as you are no doubt aware. On Tuesday 19th September there will be a histopathology session here at Attwood, using our facilities to present microscopic images to a crowd. You are all welcome to participate, bring slides etc. The session kicks off at 10:00 am, and will not be restricted to ganglioneuritis, although that is the major issue. This international group will also be involved in assessing research requirements for abalone ganglioneuritis. After they have made their recommendations then the various Victorian players will have to decide what they want and what resources will be allocated.

Given that the FRDC deadline is not till 1st Nov, do we have to decide today what shape our proposal needs to take?

Regards,
Malcolm Lancaster
Good Afternoon

Please find attached a copy of the agenda for the Abalone Virus - Scientific Forum on Thursday, 21st September 2006.

For those of you who have not done so already, could you please advise of your attendance by COB today, 15 September?

We look forward to seeing you at the forum.

Regards

Mehdi Doroudi - DVM, PhD
Research Director
Marine & Freshwater Systems - PIRVic
Department of Primary Industries

Ph: +613 5258 0272 Fax: +613 5258 0270
Mobile: +61400 845 406

Email: mehdi.doroudi@dpi.vic.gov.au
2A Bellarine Hwy, Queenscliff VIC 3225 Australia
PO Box 114, Queenscliff VIC 3225 Australia
(See attached file: Agenda - Abalone Virus - Scientific Forum 21 September 2006.pdf)
ABALONE VIRUS - SCIENTIFIC FORUM

AGENDA

Thursday, 21 September 2006
Conference Room, Ground Level
1 Spring Street, Melbourne

09.30   Registration

10.00   Welcome, Introduction & Background
        Chair - Dr Peter Appleford, Executive Director Fisheries Victoria

10.30   DPI's Response
        Dr Hugh Millar, Chief Veterinary Officer, Biosecurity Victoria

11.00   Epidemiological aspects
        Dr Paul Hardy-Smith, Managing Director, Panaquatic Health Solutions Pty Ltd

1130   Open Forum with Panel Experts
        Dr Peter Appleford - Chair
        Dr Carolyn Friedman, United States of America
        Dr Judith Handlinger, Tasmania
        Dr Mike Hine, New Zealand
        Dr Anna Mouton, South Africa
        Dr Tristan Renault, France

12.30   Lunch

13.15   Research undertaken to date
        Dr Mark Crane, Project Leader, AAHL Fish Diseases Laboratory, CSIRO

1345   Current research proposals
        Dr Mehdi Doroudi, Research Director, Marine & Freshwater Systems, PIRVic

1415   Future directions
        Dr Peter Appleford, Executive Director Fisheries Victoria

1430   Open Forum with Panel Experts
        Dr Peter Appleford - Chair
        Dr Carolyn Friedman, United States of America
        Dr Judith Handlinger, Tasmania
        Dr Mike Hine, New Zealand
        Dr Anna Mouton, South Africa

15.00   Close
Hi Caroline,

I did get the copy of the epidemiology report - thanks.

Also - my powerpoint presentation for the scientific forum is too big to send as an attachment - would it be ok to bring it on CD or on a USB memory stick on the day?

Mark

MARK CRANE Ph.D.
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Australian Animal Health Laboratory
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email: mark.crane@csiro.au

-----Original Message-----
Sent: Tuesday, 19 September 2006 10:16
To: Crane, Mark (LI, Geelong)

Wonderful - thanks Mark. Hope you had a great time away from the office.

Did you get the copy of the epidemiology report I sent you by express post?

Kind Regards

Caroline

Mehdi Doroudi - DVM, PhD
Research Director
Marine & Freshwater Systems - PIRVic
Department of Primary Industries

Ph: +613 5258 0272 Fax: +613 5258 0270
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Email: mehdi.doroudi@dpi.vic.gov.au
2A Bellarine Hwy, Queenscliff VIC 3225 Australia
PO Box 114, Queenscliff VIC 3225 Australia
Hi Caroline/Mehdi,

As discussed with Mehdi last week - I will be participating. I have only just got back from a few days away and will get my presentation to you asap.

Cheers

Mark

MARK CRANE Ph.D.
Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
Private Bag 24
Geelong Vic 3220

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Abalone herpesvirus: Background

Histopathological examination on moribund animals indicated a ganglionitis.

Examination by electron microscopy revealed the presence of a herpes-like virus in the pleuroperitoneal ganglion.

A similar case was reported in cultured abalone, *Haliotis diversicolor supertexta*, in Taiwan (Lien 2003) (Chang et al., *J. Aquat Org. 65*: 23-27 2005).

Immediate Research Need

As discussed with DPI Victoria: Information on the virus transmissibility was required in order to assist implementation of effective management strategies to control the disease and/or eradicate the virus.

Two infection trials were planned and undertaken.

Herpes-like virus transmission trial 1

Objectives:

- Confirm that the virus isolated from sick abalone can cause disease in healthy abalone.
- Confirm Henle-Koch's postulates.
- Determine if moribund abalone (virus infected) can transmit the virus to healthy uninfected abalone through the water column.
Herpes-like virus transmission trial 1

Method:
Inocula were prepared from 6 frozen (infected) and 9 moribund abalone
Ganglia were dissected, pooled (1 frozen group and 1 fresh group), homogenised, centrifuged, supernatant filtered and kept at 4°C until inoculation into healthy abalone.

Treatment Groups (12-15 abalone/group):
1) Co-habitation with sick abalone
2) Injected with frozen virus (100uL)
3) Injected with fresh virus (100uL)
4) Injected with DMEM only (100uL)

Experiments were carried out in 100 L plastic tanks containing 80 L aerated salt water (sea). Water flow rate through the system was 3 Litres/hour.

Herpes-like virus transmission trial 1

- An extra tank contained healthy naïve abalone as negative (no treatment) control
- Health status and mortality was recorded daily
- Histological examination and electron microscope analyses were performed on some moribund animals.

Herpes-like virus transmission trial 1: Results

Graph showing cumulative mortality rates over time.
Conclusions

1. The virus is transmitted through the water column from sick abalone to the healthy abalone.

2. The virus is highly pathogenic, killing abalone within a few days of infection.

3. Injection of the virus in the abalone foot causes disease and mortality.

4. The virus remains virulent and pathogenic after being frozen at -80°C.
Crane Scientific
Forum part 2....

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Project Leader
AAHL Fish Diseases Laboratory
Australian Animal Health Laboratory
CSIRO Livestock Industries
Private Bag 24
Geelong Vic 3220

International Phone: +61 3 52 275118
International Fax: +61 3 52 275555
email: mark.crane@csiro.au
Objective:
To determine if dilution of contaminated water is a sufficient means of disease control

Method:
Sample 200L of farm water (ongoing abalone mortalities) and transport to AAHL
Expose healthy abalone to various dilution levels (100%, 10%, 1%, 0.01%, 0.001%) of farm water for a 48 hour period
Record morbidity and mortality
Perform histological examination of moribund animals

Results:
• No disease or mortality in any of the experimental groups
• Positive control group (virus-injected abalone) demonstrated typical clinical signs and mortality

Conclusions:
1) There was no (or little) virus present in the water at the time of sampling (virus titres may fluctuate in a farm setting, particularly in flow-through systems) and/or
2) The transportation of the water to AAHL affected the viability of the virus present in the water and/or
3) The virus in the water was present at a titre that was too low to cause a productive infection in the abalone
Control Experiment:
Co-habitation of healthy abalone with diseased animals (held in basket) until approx. 50% mortality
Exposure of healthy abalone to various dilution levels (100%, 10%, 1%, 0.01%, 0.001%) of simulated farm water from the co-habitation group for a 48 hour period
Record morbidity and mortality
Perform histological examination of moribund animals

Conclusions
The virus remained infectious to animals even after a 1 in 100 dilution (although only one of the duplicate tanks was affected at this dilution compared with both tanks affected at 100% and 10% levels)
Even though the dilution factor at the outlet of the farm is greater than 1 in 100 and while dilution reduces infectious dose, it remains possible that wild mollusc species could become infected by virus released into the environment

Results
Cumulative mortality in abalone exposed to virus-infected water:

- Coraline
- 100% water
- 10% water
- 1% water
- 0.1% water
- 0.01% water
- Uninoculated control

Days post exposure

What we know:
- The disease can be transmitted horizontally
- The virus is transmitted through the water column – direct contact is not required for transmission
- The virus is highly pathogenic, killing abalone within a few days of infection
- Injection of the virus in the abalone foot causes disease and mortality
- The virus remains virulent and pathogenic after being frozen at -89°C
- Cannot be grown in fish cell lines
- Oyster herpesvirus PCR negative
Abalone Herpes-like virus
Research Needs

What we need to know:
Geographic range
Is it the same as the Taiwanese virus?
Host range
Distribution and prevalence
Infectious dose
Sensitivity to physico-chemical conditions
Stability in the environment
Routes of transmission
Tissue distribution
Mechanisms of resistance, if any
Control methods
Crane Scientific
Forum part 11...

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(12-15 abalone/group)
1) Co-habitation with sick abalone
2) Injected with frozen virus (100uL)
3) Injected with fresh virus (100uL)
4) Injected with DMEM only (100uL)
Experiments were carried out in 100 L plastic tanks containing 80 L aerated salt water (sea). Water flow rate through the system was 3 Litres/hour.

Herpes-like virus transmission trial 1

- An extra tank contained healthy naïve abalone as negative (no treatment) control.
- Health status and mortality was recorded daily.
- Histological examination and electron microscope analyses were performed on some moribund animals.

Herpes-like virus transmission trial 1: Results
Conclusions

1. The virus is transmitted through the water column from sick abalone to the healthy abalone

2. The virus is highly pathogenic, killing abalone within a few days of infection

3. Injection of the virus in the abalone foot causes disease and mortality

4. The virus remains virulent and pathogenic after being frozen at -80°C
Mark

I've made a couple of format changes (bullet points etc - hope that is ok). I've saved it as a pdf version should you want to forward it electronically to anyone.

Please let me know if the attached is ok?

Yours Regards

(See attached file: Mark Crane Presentation - Scientific Forum.pdf)

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Hi Caroline,

You should have rec'd 2 emails with my presentation in 2 parts; part 1 (labelled ...part 11) is the first half and part 2 is the remainder. If you could let me know that you have rec'd these OK and you can put these together that would be great.

Cheers
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Abalone Herpesvirus – Results of current research

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Abalone herpesvirus: Background

- Histopathological examination on moribund animals indicated a ganglioneuritis.

- Examination by electron microscopy revealed the presence of a herpes-like virus in the pleuropedal ganglion.

- A similar case was reported in cultured abalone, *Haliotis diversicolor supertaxa*, in Taiwan (Jan 2003) (Chang et al., *Dis Aquat Org* 65: 23-27 2005)
Immediate Research Need

- As discussed with DPI Victoria: Information on the virus transmissibility was required in order to assist implementation of effective management strategies to control the disease and/or eradicate the virus.

- Two infection trials were planned and undertaken.
Objectives:

- Confirm that the virus isolated from sick abalone can cause disease in healthy abalone
- Confirm Henle-Koch's postulates
- Determine if moribund abalone (virus infected) can transmit the virus to healthy uninfected abalone through the water column
Herpes-like virus transmission trial 1

Method:

- Inocula were prepared from 6 frozen (infected) and 9 moribund abalone
- Ganglia were dissected, pooled (1 frozen group and 1 fresh group), homogenised, centrifuged, supernatant filtered and kept at 4°C until inoculation into healthy abalone
Herpes-like virus transmission trial 1

Treatment Groups
(12-15 abalone/group)

1) Co-habitation with sick abalone
2) Injected with frozen virus (100uL)
3) Injected with fresh virus (100uL)
4) Injected with DMEM only (100uL)

Experiments were carried out in 100 L plastic tanks containing 80 L aerated salt water (sea). Water flow rate through the system was 3 Litres/hour
An extra tank contained healthy naïve abalone as negative (no treatment) control.

Health status and mortality was recorded daily.

Histological examination and electron microscope analyses were performed on some moribund animals.
Herpes-like virus transmission trial 1: Results

Cumulative mortality in abalone

- Co-habitation
- Fresh virus injected
- Frozen virus injected
- DMEM injected
- Untreated

Days post-exposure to virus

% mortality
Conclusions

- The virus is transmitted through the water column from sick abalone to the healthy abalone.
- The virus is highly pathogenic, killing abalone within a few days of infection.
- Injection of the virus in the abalone foot causes disease and mortality.
- The virus remains virulent and pathogenic after being frozen at -80°C.
Herpes-like virus transmission trial 2: Water dilution and virus transmission

Objective:
To determine if dilution of contaminated water is a sufficient means of disease control
Method:

- Sample 200L of farm water (on-going abalone mortalities) and transport to AAHL
- Expose healthy abalone to various dilution levels (100%, 10%, 1%, 0.01%, 0.001%) of farm water for a 48 hour period
- Record morbidity and mortality
- Perform histological examination of moribund animals
Herpes-like virus transmission trial 2: Water dilution and virus transmission

Results:

- No disease or mortality in any of the experimental groups

- Positive control group (virus-injected abalone) demonstrated typical clinical signs and mortality
Conclusions:

1) There was no (or little) virus present in the water at the time of sampling (virus titres may fluctuate in a farm setting, particularly in flow-through systems)

and/or

2) The transportation of the water to AAHL affected the viability of the virus present in the water

and/or

3) The virus in the water was present at a titre that was too low to cause a productive infection in the abalone
Herpes-like virus transmission trial 2: Water dilution and virus transmission

Control Experiment:

- Co-habitation of healthy abalone with diseased animals (held in basket) until approx. 50% mortality

- Expose healthy abalone to various dilution levels (100%, 10%, 1%, 0.01%, 0.001%) of simulated farm water from the co-habitation group for a 48 hour period

- Record morbidity and mortality

- Perform histological examination of moribund animals
Results

Cumulative mortality in abalone exposed to virus infected water

- Co-habitation
- 100% water
- 10% water
- 1% water
- 0.01% water
- 0.001% water
- Untreated control
Herpes-like virus transmission trial 2: Water dilution and virus transmission

Conclusions

The virus remained infectious to animals even after a 1 in 100 dilution (although only one of the duplicate tanks was affected at this dilution compared with both tanks affected at 100% and 10% levels)

Even though the dilution factor at the outlet of the farm is greater than 1 in 100 and while dilution reduces infectious dose, it remains possible that wild mollusc species could become infected by virus released into the environment.
Abalone Herpes-like virus: Summary

What we know:

- The disease can be transmitted horizontally
- The virus is transmitted through the water column – direct contact is not required for transmission
- The virus is highly pathogenic, killing abalone within a few days of infection
- Injection of the virus in the abalone foot causes disease and mortality
- The virus remains virulent and pathogenic after being frozen at -80°C
- Cannot be grown in fish cell lines
- Oyster herpes virus PCR negative
Abalone Herpes-like virus
Research Needs

What we need to know:

- Geographic range
- Is it the same as the Taiwanese virus?
- Host range
- Distribution and prevalence
- Infectious dose
- Sensitivity to physico-chemical conditions
- Stability in the environment
- Routes of transmission
- Tissue distribution
- Mechanisms of resistance, if any
- Control methods