A Review of the 1st International Conference on Molluscan Shellfish Safety

Seafish Report No.452

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The Sea Fish Industry Authority

Seafish Technology

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Author: M. Boulter

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Summary

The First International Conference on Shellfish Safety was held in New South Wales, Australia on 13th-17th November 1994. It followed two successful conferences on Shellfish Depuration in 1988 and 1992 in the USA and France. By widening the scope of the conference this encouraged a range of safety related issues to be raised and provided researchers and industry with a clearer picture of the range of factors which can affect shellfish safety. This report contains the abstracts of the papers, and in most cases a brief summary of the presentations. The conference recommendations and some interpretation of these with regard to the UK industry are given. An appendix outlining some of the illness associated organisms detailed in the papers is included.

1. Introduction

This conference was held in Sydney, New South Wales, Australia on 13-17 November with approximately 70 delegates from eleven countries. It was organised by The University of New South Wales and the New South Wales Health Department. The scientific committee included representatives from Australia, New Zealand, Philippines, USA and France. The programme consisted of 21 papers and a field tour of the Port Stephens oyster fishery.

The abstracts of the papers and a report on most from a UK perspective are given. Whilst every effort has been made to ensure the reports are as accurate as possible, there may be some discrepancies. Some of the presentations were not given and so do not have a UK summary. The full conference papers and recommendations should be published by the organisers by the middle of 1995. This report provides the UK industry with an initial synopsis of the relevant issues.

2. Abstracts of Papers with Summaries from a UK Perspective

2.1 Oyster Safety - What Have We Learned in the Past 5 Years?

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ABSTRACT:
During the past 5 years, the safe consumption of raw and/or partially cooked oysters has received much attention in the United States. This attention has been focused primarily on several areas and includes: (1) the development of a new sewage pollution indicator, (2) HACCP seafood inspection systems; (3) educational programs; and (4) the bacterium *Vibrio vulnificus*. In addition, the Gulf of Mexico Oyster Industry next year faces a potentially devastating 6 month oyster ban due to deaths caused by *V.vulnificus*. The challenges facing the oyster industry remain great and solutions to these problems will not be easily solved without industry changes and proactive programs by both the industry and shellfish regulatory agencies.

UK SUMMARY:

Issue 1
An indicator organism to replace *E.coli* is needed and $25M is being sought to address this issue. In Florida 6 outbreaks of gastro-enteritis probably due to Norwalk virus have occurred in the last 5 years, with between 20 - 300 people reported per case.

Issue 2
Under the Inspection of Seafood Bill the adoption of HACCP (Hazard Analysis Critical Control Point) will be required at all seafood plants within the next 18 months.

Issue 3
HACCP workshops are being conducted for the industry by the governments F.D.A.

Issue 4
In the US recent attention with regard to shellfish safety has been diverted from bacterial/viral gastro-enteritis issues to the issue of *Vibrio vulnificus* due to reports of deaths associated with the consumption of raw shellfish. An estimated 6.4 deaths per year are occurring within Florida state alone. This is the only case of a foodstuff being directly correlated in killing people in 20 years in Florida State. Immunological data shows that it is mainly killing immuno-deficient people or people with liver problems. It can cause severe illness in other members of the population. *V. vulnificus* is a rare naturally occurring bacteria with a natural inhabitant of estuarine waters and sediments and which can be bio-accumulated by molluscs. The immunological problems with *V. vulnificus* seem to be exacerbated by long delays between harvesting and consumption of shellfish in the US. The state of Texas is trying to ensure all shellfish is consumed within 10 days of harvesting. All mollusc's are carrying a government health warning concerning the dangers of eating raw shellfish.

2.2 Molluscan Shellfish Sanitation and Hygiene in the Philippines

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ABSTRACT:
This is a review of existing publications and documents from government regulatory agencies and incidents of shellfish poisoning epidemics in relation to the status of molluscan shellfish sanitation and hygiene in the Philippines. Issues arising from these epidemics in relation to shellfish sanitation and hygiene are presented along with possible solutions.

Philippine policies prescribing the quality of sea waters for growing shellfish in these areas are available. Several manuals were also locally printed which teach environmental surveys, growing shellfish, and postharvest technologies. Postharvest practices of the shellfish industry, however, remain "traditional", since most products are sold on the same day of harvest. This is in spite of the market which is estimated at 660,000 kilograms of shellfish meat daily.

Several problems beset the industry, most of which reflect the inadequate attention given this productive sector, e.g. water pollution has caused severe unabated decline in the spatfall and the quality of their products from certain areas. Twenty six previous epidemics of paralytic shellfish poisoning (PSP) have dealt severe economic impacts to the industry. Most interventions focused on the shellfish industry without considering other forces impinging upon the market. Environmental decay and social pressures due to population increase should highlight the need to develop the shellfish industry and the market in the Philippines, and elsewhere.

UK SUMMARY:
Shellfish is a common foodstuff in The Philippines. It is inexpensive to grow and the consumption level is estimated at 10g per person per day. However, there have been severe problems caused primarily by growing shellfish in urban areas (sewage contamination) and the presence of PSP at certain times of the year. As most shellfish is cooked, bacterial/viral problems are reduced. However, 427 PSP cases have occurred between 1988 -94, mainly in July -August and have killed 24 people mainly children. The government intervention policy has been to confiscate and destroy mollusc's from non approved beds. This has had severe impact on the population with 20,000 people suffering starvation due to the lack of a cheap alternative food source to shellfish.

The government wants to develop a sound shellfish sanitation programme. This should be standardised to the rest of the world since GATT (General Agreement on Tariffs and Trade) will lead to increased scope for shellfish exports. The government would also like to see some research into the use of depuration technology for PSP.

2.3 Quantity of bacteria and some water quality parameters in oyster culture beds atBan Don Bay, Southern Thailand

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ABSTRACT:
Ban Don Bay is located on the eastern coast of Southern Thailand. The bay is quite shallow with a maximum depth of about 6m. The investigation of some environmental properties in oyster culture areas in the bay (bacteria and water quality) were conducted during October 1993 up to September 1994. The average level of bacteria in oyster meat *(Crassostrea belcheri)* were as follows: total coliforms 6.32 MPN/100g, faecal coliforms 1.92 MPN/100g, total marine vibrios $2.25x10^2$ MPN/100g, *Vibrio parahaemolyticus* 2.93 x 10^4 MPN/100g, and total bacteria 2.40 x 10 MPN/100g. The average bacterial content of surface sea water in culture areas were 3.53, 0.79, 2.75, 0.54 and 3.4 x 10^2 MPN/100 mL as total coliforms, faecal coliforms, total marine vibrios, *Vibrio parahaemolyticus* and total bacteria, respectively. The average bacterial content of bottom sea water of culture areas were 2.47, 0.08, 2.21, 1.05 and 3.95 x 10 MPN/100mL as total coliforms, faecal coliforms, total marine, *Vibrio parahaemolyticus* and total bacterial, respectively. Some water quality parameters were also analysed. The average values were water salinity 20.75ppt, pH 7.96, dissolved oxygen 5.3mg/L, phosphate 0.45 mg/L, ammonia-nitrogen 0.10 mg/L and hydrogen sulphide 0.03 mg/L. Some more details which concern the bacteria condition of sea water and oyster meat will be discussed.

UK SUMMARY:
Ban Don Bay is a large mollusc production area in Thailand, producing 2000 tonnes of oysters per year and substantial quantities of cockles. The government has a thorough monitoring programme in place in this area, with product sampling conducted twice per month. This is paid for by the government not industry.

2.4 Shellfish Water Quality Protection Programme on the West Coast of Canada

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ABSTRACT:
NONE RECEIVED.

UK SUMMARY:
In Canada the government implemented its own NSSP (National Shellfish Sanitation Programme) in 1992 to replace the US USSP which the Canadians had been following since 1965. This is based on surveying the water quality of harvesting areas not the shellfish (the same as the US system). On the West Coast they survey 34 harvesting areas and last year undertook 8,000 F.coli tests with a staff of 6 and two mobile laboratories. The cost of this operation was c2.5 M, which was paid for by the government.

Waters are classified into 4 categories :-

1. Approved	= fit for human consumption
2. Conditionally approved	= fit for human consumption for part of the year
3. Closed areas. (25 % of all areas)	= limited harvesting if licenced and depurated
4.Within 300m of a sewage discharge	= totally prohibited

The approved water classification is based on achieving <14 *E.coli* /100 ml with an MPN test, with only 10% of samples allowed to exceed this level.

There are 3 UV depuration facilities on the west coast of Canada. These can handle up to 900 tonnes per year. The latest plant cost $1.7M and its validation study will be starting soon. It will take a period of months before the plant passes the verification studies and is allowed to operate commercially.

The main molluscan fishery is for Geoduck clams and is worth $30M per year mainly in export to Japan. These are harvested in clean areas.

2.5 An Overview of the Gulf of Mexico Program Initiatives in Relation to Seafood

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ABSTRACT:
In the summer of 1986, the US Environmental Protection Agency (EPA) held a public meeting in Brownsville, Texas on the subject of incineration at sea. The large turnout of concerned citizens made it clear that the public was interested in the well-being of the Gulf of Mexico. The response of these citizens, coupled with indications of environmental degradation in the Gulf, prompted the EPA, Region 4, to propose a Gulf-wide strategy termed "the Gulf Initiative". The intent of this initiative was to identify Gulf-wide problems, and set goals and research needs while developing a strategy to respond to those problems. During an initial Gulf Initiative Workshop held in Gulf Shores, Alabama in August of 1986, 59 persons, representing a broad spectrum of organisations concerned with marine pollution, identified critical issues and activities causing problems in the Gulf, made recommendations for a program management structure and provided a strong consensus that such a program was needed.

In August of 1988, the Gulf of Mexico Program was established as an intergovernmental program at Stennis Space Center, Mississippi, under the sponsorship of the EPA.

On 10 December 1992, eleven federal agencies, the governors of all five gulf coastal states and the Citizens Advisory Committee (CAC) chairman, signed an agreement for cooperation to build the support and obtain the resources necessary to meet nine environmental challenges within five years and to attain the long term goal of protecting the Gulf of Mexico. Those federal agencies included the: Environmental Protection Agency (EPA); Soil Conservation Service (SCS); National Oceanic and Atmospheric Administration (NOAA); Fish and Wildlife Service (FWS); National Park Service (NPS); Food and Drug Administration (FDA); National Aeronautical and Space Administration (NASA); US Army; Department of the Navy; US Air Force; and the US Coast Guard. This symposium signatory document was termed, "A Partnership for Action".

Endorsed by the five Gulf states as well as by key Federal agencies, this document increases the level of awareness and commitment from these groups to the issues that threaten the Gulf of Mexico. As the document itself says, it helps to harmonise the diverse interests focused on the Gulf. This agreement outlines nine five-year environmental challenges. These challenges will be a major driving force in laying out the program activities for the next five years. It is a landmark document for the program, showing that state and federal agencies can work together as a team and agree upon a common set of challenges aimed at improving the environmental status of the Gulf. It also underscores the commitment of these agencies to continue to work as a team to meet these challenges.

The nine challenges are:

1. Significantly reduce the rate of loss of coastal wetlands
2. Achieve an increase in Gulf Coast seagrass beds
3. Enhance the sustainability of Gulf commercial and recreational fisheries
4. Protect human health and food supply by reducing the input of nutrients, toxic substances, and pathogens to the Gulf
5. Increase Gulf shellfish beds available for safe harvesting by 10 percent
6. Ensure that all Gulf beaches are safe for swimming and other recreational uses
7. Reduce by at least ten percent the amount of trash on beaches
8. Improve and expand coastal habitats that support migratory birds, fish and other living resources
9. Expand public education/outreach tailored for each Gulf Coast county or parish.

The goal of the Gulf of Mexico Program is to protect, restore, and enhance the coastal and marine waters of the Gulf of Mexico and its coastal natural habitats, to sustain living resources, to protect human health and the food supply, and to ensure the recreational use of Gulf shores, beaches and waters - in ways consistent with the well being of the region.

The main purpose of the Program is to develop and implement an agenda for protecting, restoring, and maintaining the health and productivity of the Gulf and assure that the waters and products of the Gulf are safe for use. Such a strategy should achieve a balance between the impacts and demands of human related activities and the preservation and enhancement of the marine resources of the Gulf.

Implementation of projects is carried out through the Program office , the committees, or the agencies and organisations involved with the program. The primary tool for identification of environmental problems and implementation of projects to remediate the associated adverse impacts are the committee action agendas.

PAPER NOT PRESENTED.

2.6 Detection of Human Enteric Viruses in Molluscan Shellfish by PCR

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ABSTRACT:

Public health controls for molluscan shellfish are hampered by the absence of methods for the detection in shellfish of the viral pathogens causing illness. The polymerise chain reaction (PCR) offers a potential way forward, however, its application to complex samples such as shellfish is hindered by the presence of potent amplification inhibitors. We describe the development of a procedure employing a combination of virus purification and nucleic acid extraction for the removal of the majority of such PCR inhibitors from shellfish. Initial developmental work used seeded polio virus as a model and showed that PCR sample tolerance ranged from 2 to 5g shellfish of highly polluted samples with overall sensitivity limits of <10 pfu poliovirus. This method was then applied to a range of naturally polluted oyster and mussel samples and the results compared to conventional enterovirus isolation by plaque assay. All enterovirus isolation positive samples were also positive by PCR. However, at one field site shellfish were positive by enterovirus PCR but negative for virus isolation. These methods have recently been applied to the detection of hepatitis A virus and Norwalk and related Small Round Structured Viruses in shellfish. Initial seeding experiments have shown that the method for removal of amplification inhibitors is equally applicable to the detection of these viruses by PCR in shellfish. Further studies on polluted field samples are in progress. These studies show that detection of human enteric viruses in molluscan shellfish by PCR is feasible and may ultimately contribute to the further development of public health controls.

UK SUMMARY:

This work is showing a significant degree of success in being able to identify viruses at very small dose levels in difficult media, ie shellfish flesh. The work has been possible due to recent breakthroughs in the field of molecular biology in recent years, especially the use of PCR to amplify DNA material. This work will hopefully lead to a standard methodology for identifying Norwalk and SRS viruses which can be used to identify the sources of food poisoning outbreaks and link them too stool samples from infected individuals.

2.7 Improved Detection of Enteric Viral Pathogens and Indicators in Oysters and their Habitat Waters

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ABSTRACT:
New methods were developed and evaluated to detect indicator viruses and human enteric viruses in oysters and their habitat waters. Procedures were developed to detect human enteric viruses (enteroviruses, hepatitis A virus and Norwalk-type viruses) in oysters by reverse transcriptase-polymerise chain reaction (RT-PCR) amplification and oligonucleotide probing. Methods were developed to detect F+ coliphages in water and shellfish and to serotype F-specific RNA coliphage isolates as indicators of human and animal faecal contamination. In field samples of water or shellfish from sites impacted by human or non-human faecal or sewage contamination, F+ coliphages were detected in the majority of samples at levels similar to or greater than the levels of faecal indicator bacteria. Serotyping of F+ RNA coliphage isolates from water and shellfish correctly identified known faecal contamination sources as being either human or animal in origin or combinations of both. Culturable enteric viruses were detected in 16 of 31 (52%) of oyster samples from a site impacted by a discharge of chlorinated, secondary sewage effluent. Enteric virus detection was more sensitive by RT-PCR and oligoprobing than by cell culture infectivity. F+ coliphage and indicator bacteria levels were higher in enteric virus-positive oysters than in enteric virus-negative oysters. Levels of F+ coliphages in oysters were higher than those of the widely used faecal coliform bacteria indicator. These results suggest that F+ coliphages may be a sensitive indicator of sewage contamination in oysters and perhaps other bivalve molluscan shellfish.

PAPER NOT PRESENTED.

2.8 The Virucidal Effectiveness of Sewage Chlorination with Reference to Molluscan Shellfisheries

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ABSTRACT:
The link between the contraction of viral gastroenteritis as a result of eating sewage contaminated shellfish has long been established. In the UK the disinfection of sewage with chlorine is adopted by some water companies to aid compliance with bathing beach standards. The effectiveness of chlorine as a virucide is of major significance when this disinfectant is adopted for sewage discharges located near shellfisheries. This laboratory study was centred on the chlorination of primary treated effluent with three doses of 8, 16, and 30 mg/L of free chlorine. The results showed that the inactivation of naturally occurring E. *coli* and faecal streptococci was rapid but very poor inactivation of F+ bacteriophage (a potential 'virus indicator') was recorded at all three doses. To ensure that these results were not artificial, comparisons were made between the inactivation rates of laboratory seeded pathogens in sterilised sewage with pathogens naturally occurring in sewage. There was found to be only minor differences in sensitivity. However, initial experiments comparing the inactivation rates of laboratory grown polio virus with the vaccine strain shed in faeces have shown that there may be a difference in sensitivity, and that this may be caused by the way the virus is presented to the disinfectant. These results have significance for the effectiveness of chlorination as a sewage treatment process particularly where this impacts on molluscan shellfisheries.

UK SUMMARY:
It has been shown that chlorine disinfection of sewage reduces levels of bacterial indicator organisms such as *E.coli*. However, potential viral indicators such as F+ bacteriophage did not similarly reduce. Hence chlorine disinfection may not be the most appropriate method of sewage disinfection in areas of shellfish production if public health is to be assured.

2.9 Marinade Effectiveness Against *Listena monocytogenes* Cells in Suspension or Associated With Green Shell Mussels *(Perna canaliculus)*

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ABSTRACT:
In order to determine the listericidal efficacy of three marinades used in the production of marinated green shell mussels *(Perna canaliculus)*, decimal reduction times (D-values) were determined for a mixture of seven strains of *Listeria monocytogenes* exposed to marinades in the presence or absence of mussels. Using an acetic acid (1.5% w/v) marinade, calculated D-values in the presence and absence of mussels were 77.3 and 33.3 h, respectively. Likewise for an acetic acid (0.7%)/lactic acid (0.75%) marinade and an acetic acid (1.5%)/gluconic delta lactone (0.2%) based marinade the D-values in the presence and absence of mussels were 125.5 and 26.9 h, and 86.3 and 19.3 h, respectively. The various increases in decimal reduction in the presence of mussels indicated that there was not a simple relationship between the listericidal nature of the marinades and the presence or absence of mussels. This result suggests that difficulties may occur when trying to relate acid inhibition studies carried out in model broth systems to "real food" systems.

UK SUMMARY:
In 1990 the New Zealand mussel industry suffered problems due to the presence of Listeria monocytogenes. A large outbreak including two deaths was reported. Sample monitoring showed the presence of listeria in 35% of all cooked samples and 47% in smoked mussels. Research was undertaken to investigate the best marinade in which to store mussel meats to prevent this problem from reoccurring. The best marinade found was 1.5% acetic acid which reduced the levels of listeria by 90% in 77 hours. All product is now held for a prescribed period of time at the factories to ensure that listeria with not be a problem with the product when it leaves the factory.

2.10 Computerised Mapping of Shellfish Harvesting Areas in the UK

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ABSTRACT:

EC Directive 91/492/EEC requires shellfish harvesting areas to be classified according to the degree of faecal pollution as determined by bacterial indicator species. This has required definitive information on the location and extent of molluscan shellfisheries and, in order to select appropriate monitoring points, information on the size, type and position of point source sewage discharges. This information has been mapped using GIS software (Arc/Info, Environmental Systems Research Institute, Inc., Redlands, California, USA) to provide a management tool for classification of harvesting areas under the terms of the Directive. All major harvesting areas (76) around the coast of England and Wales have now been mapped under 6 main coverages; digitised topographic data captured from the Ordnance Survey 1:25,000 Pathfinder series in 10x10 km tiles; the position and extent of commercial shellfish beds; the extent of the classified zone; continuous and intermittent sewage discharges with details of type and flows; location of shellfish purification plants; and shellfish monitoring points. Point coverages, such as sewage discharges and shellfish monitoring points, were created automatically from databases containing attributes and grid references. Arc/Info has proved flexible for: adding additional information as separate coverages; using line work from one coverage as part of another, interrogating data points for their associated information; generating plots for different purposes containing selections of available coverages and at varying scales; quick, easy and comprehensive editing facilities. Maps are plotted as hard copy to the required size and can also be accessed via GIS viewing software such as ArcView on a notebook PC for maximum portability.

2.11 Polyunsaturated Fatty Acid Requirements of the Milk Conch *Strombus costatus* (Gmelin) During Larval Development

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ABSTRACT:

The purpose of this study is to determine the relationship between high mortality and nutritional aspects under culture of *Strombus costatus* larvae. Milk conch larvae were raised under hatchery conditions and harvested 16 days after feeding using five specific microalgal diets: *Chaetoceros muelleri* clone (CHGRA), *Thalassiosirafluviatilis* clone (THAL), *Isochrysis sp.* clone (CISO), *Tetraselmis chuii* clone (UW474), and *Dunaliella tertiolecta* clone (DUN). Microalgae were cultured semi continuously with Guillard's F/10 media at 26°C. Aerated cultures were grown in 4L culture medium in 10L autoclaved plastic carboy shelves illuminated with 75-80 E.m s white fluorescent light and periods of 12h light: 12h dark cycles. Cells were harvested towards the end of log phase. All samples from fatty acid methyl esters (FAME) and dimethyl disulphide (DDS) derivates were analysed by gas chromatography-mass spectrometry (GC-MS). Based on growth rates, THAL and CISO resulted in the optimal diet provided to S. *costatus*, followed by moderated results with CHGRA and (UW474), and poorly with DUN. The low elongation-desaturation of both families of linoleic and linolenic in S. *costatus* larvae indicated that may be primarily controlled by action of acyltransferase enzymes instead of the desaturases, and these could be the principal way to control the fatty acids composition in larvae. The larvae selectively incorporated long chains of C20 and C22 polyunsaturated fatty acids (PUFA) present on THAL and CISO diets, conversely to CHGRA and DUN. Results indicated that larvae of this species require 20:5 (n-3), but especially 20:4 (n-6) and 20:6(n-3) as shown as for many marine organisms.

2.12 The Boring Giant Clam (*Tridacna croces*) Fisheries of Coron, Northern Palawan, Philippines

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ABSTRACT:
In the Philippines, *Tridacna croces* or the boring giant clam is the only tridacnid species that can be gathered from the world for domestic and foreign trade. In 1993, 70% of frozen T. *crocea* meat (mantle and adductor muscle) that were exported to Japan came from Coron, Northern Palawan in the Southern Philippines. With the supply requirements of 1 MT/exporter/month (90,000 - 145,000 clam meat/MT), T. *crocea* harvesting in Coron waters can be assessed to have reached the level of over-exploitation. On this premise, a study was conducted to assess the status of T. *crocea* stocks in Coron waters and the existing clam meat export industry in the area. Materials for this study were obtained through land-and marine-based surveys. The land-based activities include interview with exporters and middlemen/brokers, warehouse visits and photo-documentation of T. *crocea* meat processing in one of the plants. Production/catch data were obtained from the brokers who maintain record of their daily catch and the Department of Agriculture (DA) District Office record of auxiliary invoices issued to exporters prior to shipment. For the marine-based survey, the research divers on SCUBA conducted ocular inspection of T. *crocea* collecting areas in Coron. There are 8 T. *crocea* meat exporting firms in the Philippines with Coron as source. Its distinct taste and tenderness, especially those with mantle length 5-9 cm, make it the desired species in sashimi trade. The exporters obtain their supplies from middlemen/brokers in Coron who employ Palawan antifes to collect T. *crocea* meat. Due to the continued increase in the demand and price of frozen T. crocea meat with Okinawa as the main destination, tremendous pressure was placed on the resource with the present catch rate dropping to 48-68kg/collector/day from the 1989-1991 yield of 60-175kg/collector/day. The marine-based survey verified this and showed that Coron waters are already depleted and can no longer provide for each exporter's supply requirement of 1 MT/month.

2.13 Algal Toxins in Australian Shellfish: Increased Public Awareness and Implications for Industry

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ABSTRACT:
Following a history of carefree harvesting of large quantities of wild shellfish by Aboriginal tribes (apparently without any reports of shellfish poisoning), starting in the mid 1980s the Australian shellfish industry has woken up to the potential dangers of contamination of cultured and wild shellfish with algal biotoxins. Positive test results are now available from Australian shellfish for paralytic shellfish poisons (PSP) (from the dinoflagellates *Gymnodiniurn catenatum, Alexandrium catenella, A. minutum),* diarrhetic shellfish poisons (DSP) (dinoflaggellates *Dinophysis acuminata, D. forni),* amnesic shellfish poisons (ASP) (presumably from the diatom *Pseudonitzschiapungens f.* multiseries) and cyanobacterial peptide toxins *(Nodularia spumigena).* While most of the causative algal species probably already existed in Australian coastal waters in Aboriginal times, there is evidence for increased translocation of harmful species in recent times via international and domestic shipping, as well as associated with the translocation of shellfish stocks. Furthermore, widespread coastal eutrophication is now creating a fertile nutrient medium allowing algae to reach harmful cell concentrations. Faced with increased public awareness of algal biotoxins in shellfish products, there is an urgent need for an Australia-wide National Strategy for Monitoring and Management of Shellfish Biotoxins, building on the successful shellfish quality assurance programmes (PSP, domoic acid) already in place in Tasmania and Victoria.

UK SUMMARY:
It was reported that most of the toxin producing algae are present in small numbers in most waters around the world. However in the past 10 years the blooming of these toxin producing species has been occurring with disastrous consequences for the shellfish industry all around the world. The reasons for the blooms are not clear but increased run off of fertilizers from the land, increased movement of water by ships ballast and movement of shellfish are probably implicated.

Algal blooms seem more prevalent when the water temperature drops to 12° C coincident with an increase in rainfall, also when the wind stress of the water is < 5m/sec. There are at present no easy ways of identifying toxin producing algae. However it is hoped that within the next 3 years the development of gene probes to identify different species will be successful. This will provide a relatively cheap and easy to use identification method.

For shellfish cultivation to be sustainable in the long term good quality controls must be in place including algal monitoring to ensure public safety. There is also a need for the development of standard monitoring methods for product quality to be assured worldwide.

2.14 New Zealand's Phytoplankton Programmes

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ABSTRACT:
In September 1993, a large-scale Phytoplankton Research Programme was established, to complement the information gained from both the existing Shellfish Biotoxin Monitoring Programme and other phytoplankton monitoring programmes. Seventeen sites from the north - eastern North Island were selected to represent both commercial growing areas and recreational shellfish beds affected in the summer of 1993 event. A database was developed to find seasonal and temporal trends throughout the regions sampled. Isolated NSP incidents were reported in the region, with various *Gymnodinium* species recorded at the same sites. All potentially toxic species and dominant other genera/species were recorded, allowing examination of data for regulatory, commercial or research applications.

UK SUMMARY:
The research programme found a number of interesting points. Firstly that when the NZ industry was shut down earlier in 1993 a number of false positive results could have contributed to the closing of the industry. Secondly they found an increase in algal blooms when the temperature dropped to 12°C.

It was found that levels of PSP took 7 months to drop from 400 to 80 µg/100g.

Their legal limits are :-

PSP	80 µg/100g or 400 mouse units/100g
NSP Brevetoxins	20 mouse units/100g
DSP Okadaic acid	10µg/100g or 5 mouse units/100g
ASP Domoic Acid	2 mouse units/100g

It was also reported that the levels set were cautious. The biotoxin unit of MAF is costing $NZ 3.6m per year to run with the industry bearing a proportion of that cost. (22% ($792,000), personal communication with industry by the author.) The main fishery in Marlborough sound is harvesting 45,000 tonnes per year of green lipped mussels.

2.15 Shellfish Contamination by Marine Biotoxins in the South China Sea

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ABSTRACT:
Harvest from the wild and culture of shellfish are major sources of food and livelihood along the coasts of the South China Sea. This growing industry, however, has been negatively impacted by blooms of harmful microalgae. Paralytic shellfish poisoning (PSP) from the blooms of *Pyrodinium bahamense* var. *compressum* has been the major cause of health and economic problems from these contaminations. Presence of other harmful algae producing other types of biotoxins has also been detected in some shellfish growing areas.

This paper is a review of the reported events of shellfish contamination by marine biotoxins along harvest and culture sites in the South China Sea from 1976 to the present. Management schemes to cope with these contaminations are likewise discussed.

UK SUMMARY:
The Philippines has had a major problem with PSP for a number of years (See Dr Pastors paper no. 2.2.) The adoption of expensive monitoring operations for PSP are difficult to fund. The need for cheap effective methods of monitoring for PSP is needed and research into the possibility of depuration methods for PSP.

2.16 PSP Toxin Monitoring in Tasmania

RAY BROWN

Tasmanian Shellfish Quality Control Program,
Hobart, Tasmania

ABSTRACT:

NONE RECIEVED.

UK SUMMARY:
Tazmania has a shellfish industry worth $A 12m p.a. at the farm gate, employing 300 people mainly growing oysters. They grow Pacifics, Sydney Rock's and a few Flat Oysters. There are 160 farms in 45 areas. In 1985 the first outbreak of PSP was reported after a large rainfall, with levels reaching >8000 µg/100g. Testing is both by mouse test and HPLC and cost $A 20,000 in the first year of monitoring 1986.

Research has found that there is a good correlation between the mouse test and the HPLC results for PSP.

Most PSP outbreaks have occurred subsequently in April and May (Autumn) when rainfall increases and there is calm weather and a temperature of 12 C. In 1993 40 farms in the industry were shut down for 7 months. The blooms are mainly in river run off areas and there is a consistent time lag between rainfall and the blooms reaching the farms. During the bloom season sampling occurs every week and out of season sampling occurs every two weeks.

The cost of sampling and testing has been $A2-3m since the outbreaks occurred.

2.17 Identification of Domoic Acid in Scallops from Australian Coastal Waters

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ABSTRACT:
In Canada during 1987, a large outbreak of human poisoning (now known as Amnesic Shellfish Poisoning, ASP) occurred when mussels *(Mytilus edulis)* contaminated with domoic acid were ingested. Further investigations demonstrated that the source of domoic acid was a diatom *Pseudonitzschia pungens forma multiseries.* In 1991, when the diatom *P. pseudodlicatissima* was detected in Port Phillip Bay, analysis of shellfish for the presence of this potent neurotoxin began. High performance liquid chromatography was used to examine shellfish throughout the period of the algal bloom. No domoic acid has been detected in shellfish from Port Phillip Bay. Later, Gippsland Lakes mussels (M. *galloprovincialis)* and Bass Strait scallops *(Pecten fumata),* from locations about 330 km east of Melbourne, were also tested. In June 1993, the edible adductor muscle and roe of a sample of scallops was found to contain an unknown compound with the same retention time as domoic acid. A photodiode array detector gave similar spectra for both the unknown and domoic acid. Subsequently, gas chromatography-mass spectrometry confirmed the identity of the unknown as domoic acid at 1.2g/g. This was the first time that domoic acid had been found in shellfish taken from Australian waters. The source of domoic acid has not been identified. However, further analyses of scallops over the ensuing 5 months showed that domoic acid ranged from 0.2 to 1.2g/g in the edible portion and 1.8 to 26.2g/g in the viscera. Even though the edible portion of scallops did not exceed the regulatory limit of 20 g, health authorities in Victoria banned the sale of viscera and whole scallops as a precautionary measure. Monitoring for domoic acid is an integral part of Victorian Fisheries ' Shellfish Quality Assurance Program.

UK SUMMARY:
In Victoria when an algal bloom occurs it is rapidly identified to see if it is a toxin or non toxin producing species. This is part of the Victoria State shellfish quality assurance program. In 1993 the levels of domoic acid found in scallops from the Bass strait off shore fishery were very low but one reading from a whole scallop was above the legal limit of 20 µg/g. The authorities subsequently banned all sales of scallops in the whole form to safeguard public health.

2.18 Toxicity Variations in Alexandrium *minutum* and Gymnodinium *caternatum*

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2 Murdoch University, Perth, Western Australia

ABSTRACT:
There is an urgent need to increase scientific knowledge of the physiology and ecology of Paralytic Shellfish Poison-producing algae. Only with an improved understanding of environmental effects on toxicity, can serious attempts be undertaken to combat and reduce the impact of toxic algal blooms. Changes in Paralytic Shellfish Poison toxin content in two dinoflagellates *Alexandrium minutum* and *Gymnodinium cateratum* have been investigated under variable batch culture conditions. Nutrient limitation effects on cell toxicity, cell toxicity variations over the growth cycle, and diurnal cycling in toxicity are evaluated. The toxins produced by the dinoflagellates were analysed by high performance liquid chromatography, using post-column oxidation and fluorescence detection. The toxin content per cell was shown to decrease under limiting nutrient conditions and be at a maximum concentration during early exponential growth. It is proposed that cell toxicity is a function of available nitrogen (nutrients) after growth requirements have been satisfied, and that the purpose of toxin production is as a nitrogen store. Diurnal cycling of cell toxin content by up to 300% was identified during a period of low cell division, which indicates that toxin cycling is another example of circadian rhythms in dinoflagellates.

UK SUMMARY:
This laboratory study has shown that the toxicity of PSP algae *Alexandrium minutum* and *Gymnodinium cateratum* both decrease if there is a low nutrient content in the water. The following results show the effect of nutrients in the water and the toxicity of the algae.

	Toxicity Level
Control (High Nutrients)	300
Low Phosphates	160
Low nitrates	55
Both low	42

2.19 Shellfish Biotoxins and Algal Blooms in Southern New Zealand (1994)

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ABSTRACT:
During 1994, extensive closures of the South Island coastline have been necessary due to the contamination of shellfish with aqueous and lipid soluble biotoxins. On most occasions good correlations have been found between the results of toxicity bioassays and the abundance of potentially toxic dinoflagellate species in the plankton. A predictive capability involving routine screening of plankton samples is possible. On a few occasions positive bioassays have occurred in the absence of identifiable causative planktonic organisms and important questions about the validity of the methods used in the biotoxin monitoring program have arisen.

UK SUMMARY:
It has been found that by taking plankton samples and screening to the algal cell size (80 microns) it is possible to easily identify toxic algae by microscope. It was found that the plankton levels correlated quite well with bioassay tests on shellfish. Since the large closure in early 1993 all subsequent closures have been more localised due to the level of knowledge obtained regarding the algal blooms.

The industry itself is closely monitoring harvesting areas by both plankton and bio-assay methods and will close areas before legal limits are reached. It has been found that it takes three weeks for levels to reduce in the shellfish post bloom.

Another major problem for the industry has been the presence of false positive results. On some occasions the industry's own results have been negative however the Ministry results have been positive, using the same test procedure, and the industry has been closed even though no toxic algae can be found in plankton samples.

Methods of reducing the number of false positive results with lipid soluble toxins need to be investigated.

2.20 Movement of Mussel Spat within New Zealand: The Risks of Associated Toxic Microalgal Introductions

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2 MAF Fisheries, Wellington, New Zealand

ABSTRACT:
An abundant source of green shell mussel spat *(Perna canaliculus)* is found attached to seaweed which is washed up sporadically on to Ninety Mile Beach, northern North Island, New Zealand. This spat supplies most of the needs of New Zealand's mussel industry. Cysts of the toxic dinoflagellate *Alexandrium ostenfeldii* have been identified on the weed. This has led to the implementation of controls on the movement of the spat (through the Ministry of Agriculture and Fisheries under the Biosecurity Act). Recently cysts of A . *ostenfeldii* have been found on mussel ropes on South Island farms. Strains of this species isolated from different sites around New Zealand are known to have different paralytic shellfish toxin profiles and toxin analyses are being carried out to ascertain whether "Sounds" and "Northland weed" strains have comparable toxicities.

UK SUMMARY:
Care must be taken when moving bivalve mollusc's for re-immersion in other areas as it has been found that cysts of certain species of toxin producing algae can be amongst the shellfish. Research must be carried out to identify what levels, if any, of algal cysts are acceptable within batches of shellfish being moved for re-immersion.

2.21 Algae Blooms in New South Wales: Monitoring and Management

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ABSTRACT:
This paper discusses the occurrence of toxic algal blooms in NSW and the monitoring and management thereof.

New South Wales experiences nuisance algal blooms in both inland and coastal waters. The increasing occurrence of blue-green algal blooms in the State's freshwaters and their impact on water supply, led the NSW Government to establish a formal system of management and monitoring of blue-green algae throughout the State. Similarly, the occurrence of potentially toxic marine red tides appears to be increasing in frequency in coastal and estuarine waters. Because of their possible impact on aquaculture, fisheries and public health, a government/industry group has been formed to develop a management contingency plan for red-tide blooms modelled on the successful blue-green algal management plan. Essential elements in the management of marine biotoxins are early warning monitoring of phytoplankton populations, toxin testing of mollusc tissue, harvest closures and community information releases.

UK SUMMARY:
The NSW Environmental Protection Agency has experience in dealing with freshwater toxic blue-green algae and is using this experience to facilitate a management plan to cope with marine toxic algae if they occur. Two important parts of their strategy are plans to deal with the press and the subsequent public concern and implementation of a plankton monitoring programme, which may require industry assistance.

2.22 Problems of Surveillance and Laboratory Methodology in a Marine Biotoxin Crisis

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ABSTRACT:

In the summer of 1993 New Zealand was subject to a major toxic shellfish poisoning (TSP) event. The outbreak was the first recorded event of TSP, resulting in extensive human intoxication. It was later shown to have potentially involved at least four types of TSP including paralytic shellfish poisoning (PSP), diarrhoeic shellfish poisoning (DSP), amnesic shellfish poisoning (ASP), and neurotoxic shellfish poisoning (NSP). To effectively cover the extensive New Zealand coastline of both recreational and commercial shellfish harvesting, up to 250 sampling points per week were required to be sampled and analysed. During the crisis the evolving epidemiological picture and species of phytoplankton identified suggested the potential for PSP or NSP, and in some areas DSP. The unknown nature of the NSP toxins and the newness of any TSP episodes in New Zealand required a rapid and quantitative toxicity testing regime. The key feature for testing lipid soluble toxins such as DSP and NSP is the method of extraction of the toxins from the sample, with the then currently accepted procedure being the extraction of NSP toxins with ethyl as a solvent. To efficiently and safely process the large number of samples required (up to 250 per week) and to account for the unknown character of the NSP toxins involved, a generic extraction procedure was developed using acetone and dichloromethane to isolate lipid soluble toxins. The large number of samples and the rapidity required for the introduction of a surveillance scheme, coupled with the unknown character of the toxins, required the use of a mouse bioassay. The difficulty of obtaining over 2,000 mice per week for bioassay was answered in the interim by the importation of mice from overseas. The ongoing program of surveillance involving up to 140 coastal sampling points has resulted in further modifications which now cover a coupled NSP/DSP bioassay, the standard PSP bioassay, and high performance liquid chromatography (HPLC) for ASP.

UK SUMMARY:

The communicable diseases centre (CDC) in NZ had to rapidly gear up to cope with monitoring hundreds of samples in early 1993 due to the algal toxin outbreak. This rapid increase in samples for analysis required a rationalisation of the test procedures to make the best use of resources, both staff and mice. A procedure was established where 400g of shellfish was required to analyse for all four toxins. All sites are tested weekly for all four toxins.

From 400g			
100g	PSP	Bioassay	3 mice
150g	DSP/NSP	Bioassay	3mice
100g	ASP	HPLC	
50g	Lost in Processing		

2.23 Using Depuration To Add Value To Shellfish Harvested From Approved Waters

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ABSTRACT:
Atlantic LittleNeck ClamFarms (Atlantic Farms) is the world's largest producer of the "quahog" or hard clam, *Mercenaria mercanaria,* the only clam species routinely served on the half-shell in North America. Atlantic Farms uses a "SeaPolishing" process in its market preparation of product which includes a standard 48-hour depuration. Although all clams are grown to market only on approved shellfish growing areas, they are depurated after harvest in an attempt to add additional safety and value to the product.

Results from SeaPolishing performed regularly over the last three years shows that even shellfish harvested from approved grow out areas can benefit from depuration. Total heterotrophic bacteria plate counts, and coliform bacteria MPN's all decreased during depuration. In addition, depuration allows the shells, which have been heavily etched by anaerobic substrates, to oxidize producing a lighter more attractive shell. Sand and silt present in the mantle cavity are also purged during depuration. Finally, tasting tests indicate that depurated clams maintain a fresher, cleaner flavour than undepurated product during long term cold storage (≥ one week).

Although a significant additional cost is incurred with depuration, the product quality is greatly improved and the marketing advantages appear to justify costs.

PAPER NOT PRESENTED.

2.24 Strategies for Removing Indigenous Bacterial Pathogens from Shellfish in Northern New England, USA.

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ABSTRACT:
In the Great Bay Estuary of northern New England, oysters *(Crassostrea virginica)* are harvested commercially in Maine and recreationally in New Hampshire. Most of the oyster resources occur in restricted waters, thus requiring relaying or depuration prior to marketing or consumption. Studies have identified suitable relay sites and conditions in the estuary for removing both faecal-borne and indigenous bacterial pathogens. One site is Spinney Creek, ME, where Spinney Creek Shellfish Inc. (SCS) is located. SCS is a commercial facility that has developed a consistent and reliable system for depurating and relaying shellfish. The other site is Great Bay, NH. The effectiveness of relaying and depurating oysters was assessed by analysing tissue, harvest water and relay water samples for faecal coliforms, *Escherichia coli, Clostridiurn perfringens, V. parahaemolyticus* and V. *vulnificus. Vibrio vulnificus* and V. *parahaemolyticus* are present only from June through October in a spatially heterogenous manner that is influenced by salinity and nonpoint source pollution. V. *vulnificus* has not been detected in Spinney Creek and only rarely and at low numbers in Great Bay. At SCS, a container relay system involving a 7 d exposure to Spinney Creek water pumped into flow-through relay lagoons was consistently effective in reducing *V. vulnificus* from oysters during August through October in 1990,1991 and 1993. Relaying oysters to Great Bay resulted in elimination of detectable *V. vulnificus* after 4 weeks, in contrast to its presence for 8 weeks in oysters that remained at the harvest site. Consistent reductions of faecal bacteria have been achieved in the relay studies and in the SCS UV-depuration system for >6 years. These results show that it may be possible to reduce or eliminate indigenous bacterial pathogens from oysters by relaying them to waters with low levels or an absence of the target bacteria. Ongoing studies continue to define environmental conditions that may be conducive to removing indigenous pathogens from shellfish.

UK SUMMARY:
At SCS bivalve molluscs are depurated as they are harvested from a restricted water classification similar to our class B waters. However, due to the increase in problems elsewhere in the US, Stephen Jones has been investigating what conditions are required to depurate *Vibrio vulnificus.* It has been shown that a longer time period is required than the 48 hours for normal depuration.

The presence of *V. vulnificus* in the water would not seem to be as prevalent in Maine as in the southern US probably due to the water temperature. The presence of *V. vulnificus* in the Great Bay estuary would also seem to be linked to the water salinity.

2.25 An Assessment of Optimum Environmental Criteria for the Successful Purification of *Ostrea edulis, Tapes decussatus, Tapes philippinarum, Cerastoderma edule,* and *Mytlus edulis* in The UK

MARK BOULTER and PETER WILSON

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ABSTRACT:
The successful operation of a purification system for bivalve molluscs requires them to be in good condition and their optimum physiological requirements to be maintained during the period they are immersed. These include mollusc density, seawater quality, salinity, temperature and dissolved oxygen level. Seafish have over a period of several years investigated the purification of both mussels *(M.edulis)* and oysters *(C.gigas)* and as a result introduced changes to the established design criteria for these species. During the 1990s this work has been extended to include investigative work on *O.edulis, T. decussatus, T. philippinarum* and C. *edule.* Most experiments were carried out using small scale purification tanks built to replicate the traditional single layer systems or were carried out in commercial purification tanks. For oysters and clams the results have allowed for recommendations regarding maximum and minimum water temperatures and stocking densities. For cockles, a species which has traditionally not been purified in the UK, it has been possible to show that purification is possible, subject to a defined operating criteria, given gentle harvesting methods. For mussels recent results have shown it is possible to purify in deep layers in pallet bins and hence adopt mechanical handling orientated purification systems which allows for less handling of the product. To conclude it is vitally important that the physiological parameters are suitable in a purification tank for the species being held if successful purification is to occur.

UK SUMMARY:
The successful operation of a depuration system for bivalve molluscs requires them to be in good condition and their optimum physiological requirements to be maintained during the period they are immersed. These include mollusc stocking density, seawater quality, salinity, temperature and dissolved oxygen level. In the UK requirements were mainly established for mussels *(Mytilus edulis)* in the 1920's and 50's and native oysters *(Ostrea edulis)* in the 1960's, based upon scientific research at that time. As other species have become commercially viable and have required depuration there has, until recently, been only limited further research, with existing requirements being used or adopted from elsewhere. In the late 1980's Seafish investigated some aspects of the depuration of mussels and pacific oysters *(Crassostrea gigas)* as part of the development of their standard design depuration systems. It became clear that some of the scientific research had been limited in its extent by the constraints of technology and low investment potential of the industry at the time. As a result some changes were introduced to established conditions for these species, notably the container stacking of mussels and seawater temperature requirements for pacific oysters.

More recently Seafish extended this work to include investigative work on native oysters, cockles *(Cerastoderma edule)* and three species of clam, hard shell *(Mercenaria mercenaria),* manila *(Tapes philippinarum)* and native *(Tapes decussatus).* The possibility of depurating mussels in a deeper layer than currently specified was also considered as this would have a number of practical advantages for companies handling large quantities of product. This work was carried out with part funding from the UK Ministry of Agriculture Fisheries and Food (M.A.F.F.).

Over several years now Seafish have carried out research into the depuration of bivalve molluscs in the UK and have been responsible for several changes to established methods. The aim has not been to disprove existing requirements for depuration but to show that with improved understanding of bivalve mollusc physiology and effective means of monitoring, that for those species currently depurated, there is scope to improve effectiveness. The work on oysters and clams is an example of this. The range of species of bivalve mollusc harvested in the UK has in recent years begun to expand but further research to establish their depuration requirements has been limited. The work on cockles is original and has allowed a market to continue and potentially expand into Europe. Our improved understanding has also enabled us to look again at some of the traditional techniques used for depuration. The bulk bin is again an example, where the reduced handling is of benefit to both processor and mussels.

2.26 Effects of temperature and salinity on *Vibrio vulnificus* and *Escherichia coli* depuration from Sydney Rock Oyster, *saccostrea commercialis*

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ABSTRACT:
The survival of *V.vulnificus* was compared with that of *E.coli* in an ultraviolet-sterilised estuarine water with temperatures of 15, 20 and 25°C, and water salinities of 20, 30 and 40 parts per thousand. The persistence of these two microorganisms in Sydney rock oysters depurated at the same water temperatures and salinities was also monitored for 48h in an ultraviolet-assisted depuration system. Survival of *V.vulnificus* cells in ultraviolet sterilised estuarine water was greater as the water temperatures increased from 15 to 25°C while *E.coli* survival changed inversely under the same water temperatures. Survival of both microorganisms in sterilised estuarine waters varied inversely to increasing salinity. Depuration of *V.vulnificus* from oysters at a water temperature of 25°C was found to be more effective than at 20°C after 48h. Substantial reduction in numbers of the pathogen in oysters was recorded in shellstocks depurated at 15°C probably due to cold stress. More effective depuration of *V.vulnificus* and *E.coli* from oysters at salinities >20 parts per thousand was demonstrated

UK SUMMARY:
This paper described some interesting points. *E. coli* survives better in cold water, *V. vulnificus* in warmer water. The greater seawater salinity lowers the survival of both organisms. The warmer water provides better depuration of oysters, as they are more active. The higher the salinity the greater the depuration, also probably activity related.

2.27 The Relationship Between Levels of *E.coli* in Shellfish and in Seawater with Reference to Legislative Standards

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ABSTRACT:
EC Directive 91/492 stipulates that shellfish harvesting areas are classified into 1 of 3 categories according to the degree of faecal pollution as determined by *E.coli,* or faecal coliform, counts in shellfish. These legislative standards can form an effective statutory framework for shellfish but do not directly correlate with *E.coli* counts in water which are much more widely used indices of water quality. This is problematical where predictions of shellfish quality based on available water quality information are required. In this study we examine the correlation between *E.coli* levels in shellfish, taken for monitoring for Directive 91/492, and seawater samples taken concurrently. More than 600 samples comprising 3 different species were tested at 42 different monitoring points over a 2 year period. When all data were aggregated, there was a strong relationship between the average levels of *E.coli* in shellfish and seawater at each monitoring point. However, a high level of data variability implies that this relationship is of limited value for prediction of compliance with legislative standards based on individual observations. Instead, *E.coli* compliance criteria for category B shellfish, as specified in Directive 91/492, were modelled as a function of levels in seawater. This model was developed to establish targets for geometric mean levels of *E.coli* in seawater compatible with category B compliance in shellfish. Predictions were tested against actual shellfish classifications at each of the monitoring locations with satisfactory results. This data shows that a relationship exists between *E.coli* levels in shellfish and in water and that data aggregation at the geometric mean level is highly predictive.

UK SUMMARY:
During routine sampling of shellfish for classification purposes water samples were also taken from a number of harvesting areas and the correlation between the two monitoring methods assessed. In raw form this data was difficult to correlate. However after subdividing the results by different EU classification areas, (A,B and C), and by different species of shellfish the correlation between water and shellfish coliform levels can be identified. This could be used to relate the EU and the US regulatory systems. It also allows the potential for growers to survey a site before shellfish are present to identify the likely EU classification category.

2.28 Industry Self Regulated Oyster Quality Assurance

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ABSTRACT:
In 1990, following outbreaks of food poisoning caused by contaminated oysters, the NSW Health Minister established a Committee to set up oyster quality assurance programs (QAP) for NSW estuaries. Quality assurance programs were to be self-regulating and funded by oyster farmers and be based on individual estuaries. The aim was to provide long term quality assurance for commercial oysters.

A model QAP was developed which provided minimum standards of quality assurance. It required an environmental survey to identify pollution sources, monitoring of growing areas, a pollution early warning system, and effective communication among oyster farmers.

The concept of quality assurance was widely supported by the oyster industry resulting in programs being established on some 24 estuaries. Each local estuary group of oyster farmers has elected a Committee, Monitor and Coordinator and implements voluntary harvesting and purification closures depending on bacterial results of oysters and or water.

To date the improvement in the public health safety of NSW oysters has been substantial. The self-regulatory approach to quality assurance by oyster farmers is unique in Australia and may serve as a model for other shellfish industries.

UK SUMMARY:
This paper described how peer pressure and industry self regulation methods have improved Oyster standards without a great deal of government intervention. The advisory committee for the Minister For Health Services Management, incorporating industry representatives, produced an Oyster Quality Assurance Programme and a Guide for Programme co-ordinators. These have then been used by the industry, through regional coordinators to improve the standards achieved by the industry across the whole state. An overall coordinator was placed on secondment from the Department of Fisheries, funded by the Health department, for the first year of the programme.

2.29 International Shellfish Trade and Food Safety Controls

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ABSTRACT:
The development and implementation of effective food safety controls for shellfish continue to challenge regulatory authorities. This is mainly due to the nature of environmental contaminants and post harvesting handling problems. Shellfish is a high risk food that unfortunately continues to be associated with food poisoning incidents. Consequently consumer groups and the shellfish industry urge governments to improve the food safety controls for shellfish. With international trade, the regulatory food safety controls for shellfish must reach beyond national boundaries to protect public health. Generally, food regulatory agencies must be satisfied that the food safety controls for imported shellfish applied in the exporting country, will adequately protect public health or be equivalent to domestic controls. Australia has a small but growing export trade in shellfish to many countries including France, Hong Kong, Japan, Malaysia, Singapore and USA. Australia also imports shellfish from Korea, Japan and New Zealand. Much of this trade is in shellstock that is consumed raw and has a limited shelf-life. Inspection by sampling, as practised by a number of countries including Australia, provides limited assistance in making a decision regarding the acceptability of a shipment. Hence this approach leaves much to be desired. Under the General Agreement on Tariffs and Trade (GATT) the Agreement on Sanitary and Phytosanitary Measures recognises the concept of equivalence of inspection systems between countries and supports the use of harmonised sanitary measures between contracting parties on the basis of international standards, guidelines and recommendations developed by the Codex Alimentarius Commission. Formal government to government agreements have been signed that are based on a recognition of equivalence between inspection systems. Ongoing efforts to harmonise inspection systems coupled with the current recommendations of the Codex Alimentarius Commission are encouraging the international use of the HACCP system approach.

UK SUMMARY:
Due to the international nature of the seafood industry and with the finalisation of the GATT talks the need for standardisation and recognition of equivalence between different countries shellfish assurance programmes and legislation is imperative. This is most likely to come through the adoption of Hazard Analysis Critical Control Point (HACCP) programmes. Until shellfish consumption stops being associated with food poisoning outbreaks the pressure for more control measures will be inevitable.

2.30 Marketing and Consumer Concerns with Shellfish

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ABSTRACT:
The increasing variety of bivalve molluscs available and recurring problems with the quality of some shellfish have added to the consumers uncertainty about how to handle and cook these seafoods and the seafood trade and consumers are more frequently questioning the quality and safety of bivalves. Nevertheless the concerns and wants of consumers are often ignored or overlooked by government agencies and industry groups despite the fundamental position they hold in the marketing chain.

The public needs to be better informed about the origins and uses of the various shellfish if consumption and trade are to continue growing. However, researchers, government regulatory agencies, growers and marketers should take greater care in communications, particularly in briefing the media on their work so that consumers are not being unduly alarmed about the safety of shellfish. Communication between government, industry and consumers is vital but it must be simple, clear, honest and relevant if it is to be effective.

UK SUMMARY:
The consumer needs to be educated in a clear and simple manner regarding shellfish, their origins, uses, nutritional values and the consumer protection measures in place to assure product safety and quality. The media need to be educated in an unbiased, clear and simple manner, not baffled with science and jargon. The media should also be cultivated and fed with good news stories, as the shellfish industry is a colourful, photogenic industry which can easily lend itself to a good story and a nice business lunch! This will then give some unbiased reporting and damage limitation when any bad news stories come along.

3. Summary of Conference Recommendations

The following are the recommendations from the Conference in draft form. The final recommendations will be inserted when they are obtained.

Networking

- Industry/Researchers/Regulators →.Email/Bulletin/Newsletter

Education WHO? HOW? (through retailers and handlers)

- Regional Communications (eg. Institute of Food Technology)
- Local QAP Committees
- Pamphlets (eg. FDA)
- Regular stories → Press
- Schools/Regions

Water Quality/Biotoxin Monitoring/Co-ordination

- Early warning systems (prioritise risks)
- Detection methods (workshops?)
- Water quality/coastal management → pressure on Governments to lessen costs
- Ballast water controls
- Contingency plans

Regulations

- Standardised divisions
- Interpretation/Implementation of Limits
- Uniformity (International)

Depuration/Quality Assurance

- Improved technology
- ($Ozone_3$) optimisation (viruses, *V. vulnificus*), toxic algae, biotoxins
- Total post harvest handling

Industry Involvement

- Practical issues
- Less scientific issues/papers
- Workshops

Research

- Australian Research Council (ARC) grants
- Lobbying
- Detection limits

Another Conference (SICMSS)

- Key personnel/regional committees to develop programme
- Regional (government/industry ?) support
- Where/When: Philippines - Nov/Dec 1996

Priorities

- Detection limits, types and standards
- Early warning systems (ie satellites, Norwegians technology, geographically based computer systems)
- Looking ahead - develop systems of handling problems before they happen
- Organising industry (ie. finance) Place pressure on government via conference (Phillip, Dr Theophanous). Sewage pollution (coastal management)
- Newsletter to be initiated

4. A View of the Recommendations from the Conference In A UK Context

4.1 Networking

The transference of information between industry, regulators and research bodies should be encouraged and be achieved by the most applicable means.

4.2 Education

It is important that the industry, regulators and researchers are well informed regarding the range of factors affecting shellfish safety, through such a forum as this conference and it's subsequent publications. It is important to educate any person who handles shellfish of the safety issues and nutritional values of shellfish. This knowledge should also be used, by informed persons, to feed unbiased information and also good news stories to the press and consumers, to educate the consumers regarding shellfish nutrition and safety. The good news stories earn some credit with the press, which can be very useful for when the bad news stories come along.

4.3 Water Quality/Biotoxin Monitoring/Co-ordination

Pressure to improve the treatment and disposal of sewage and to reduce the discharge of nutrients must be maintained. Development of early warning systems and the improvements in methods of detecting toxic algae should be encouraged. A review of contingency plans, by both industry and regulators, for when an outbreak occurs, in light of the experiences found in New Zealand and other places where unexpected outbreaks have occurred, may be necessary. Control of ballast water dumping should be encouraged.

4.4 Regulations

Standardisation or Memorandums of Understanding (MOU's) regarding the equivalence of regulations are seen as being a key factor for the exportation of product especially into the USA or Europe. The New Zealand mussel industry which uses the US system, has shown equivalence to EU Category A standard. Implementation of regulations and their common interpretation must be achieved, if trust is to be gained and product safety to be safeguarded. The aim of common standards for toxins, coliforms and ultimately viruses must be a goal.

4.5 Depuration/Quality Assurance

Development of valid indicators for detecting pollution of water and shellfish by human viral pathogens must be encouraged and then adopted. Further investigation to develop techniques to eliminate or reduce levels of pathogenic or toxin producing species in raw shellfish should continue. Research into the use of ozone (O_3) and it's effects with Viruses, Vibrio bacteria and toxic algae should be implemented. All product handlers should be made aware of good post harvest handling care methods and the problems which can occur due to bad handling.

5. Conclusions

Faecal pollution can contribute viral and bacterial contaminants and is the primary source of infection. However the natural aquatic environment supports bacterial pathogens such as Vibrio's and toxin producing algae, which can cause serious illnesses.

If growing, harvesting or catching molluscs the potential for risks to food safety needs consideration. Several factors need to be taken into consideration, including host risk factors, sources and types of microorganism, reduction potential by depuration or heat treatment procedures and processing and handling methods which can allow microorganisms to survive and grow prior to consumption.

The industry and also possibly the public should be made aware of all these factors. Products have differing levels of risk which must be assessed in order to control these risks effectively. The largest number of illnesses are clinically suggestive of Norwalk virus. Although relatively common these illnesses tend to be relatively mild with no associated mortalities. Other naturally occurring marine pathogens are responsible for fewer reported cases of infections but can have more serious consequences and so should not be overlooked.

The industry should not be complacent even when operating within the extensive current legal framework. It should be prepared to increase sampling regimes to include viruses and toxic algae in the future and be aware that short or longer term harvesting closures and/or changes to depuration systems and/or an increase in depuration times may become inevitable if the protection of the public and hence the survival of the industry is to be assured.

Appendix 1

A Summary of the Illness Associated Organisms which can affect Molluscan Shellfish Safety

1. Introduction

Bivalve mollusc's have the potential to transmit disease from viral and bacterial microorganisms and from toxin producing algae . These agents are acquired from three sources:

1. Faecal pollution of the aquatic environment.
2. The natural aquatic environment.
3. Post harvest contamination/abuse.

A brief description of the main causative agents of molluscan associated illness and the testing and control measures routinely adopted to protect the public are outlined in the following summary.

2. Sewage Pollution Related Organisms

2.1 Bacterial Pathogens

Human illness caused by the consumption of molluscs contaminated by seawater polluted with bacterial pathogens has largely been stopped by a general decrease in the incidence of bacterial illness within the human population, the adoption of depuration technology to cleanse shellfish grown in mildly polluted waters and the closing of heavily polluted harvesting areas.

The coliform indicator used to define water and product quality is an indicator of bacterial and possibly viral human enteric pathogens. A problem with the coliform indicator is that it does not indicate the presence of non-sewage related naturally occurring bacterial pathogens such as *Vibrio*'s, also it does not correlate well with the presence of human enteric viruses, which are the pathogens now most commonly associated with sewage contamination of molluscs.

Another potential problem is that the microbial quality of harvest waters does not appear to be a good indicator of some bacterial pathogens such as nontyphoidal *Salmonella* contamination. In the US oysters removed from closed and open beds show the same level of contamination (4%), and no correlation was observed between the presence of *E.coli* and *Salmonella* in shellfish. However, *Salmonella* do not grow well in oysters during storage and are not retained during depuration or relaying so contaminated oysters can be purified.

The EU currently use the US end product standard to define our clean water category rather than the US water standard. The US standards are:

A mean level of 14 *F.Coli* MPN/100ML with <10% of samples exceeding 43 *F.Coli* MPN/100ML and an end product standard of 230 *E.Coli* MPN/100g of meat.

2.2 Viral Pathogens

The largest number of illnesses are reported from unknown etiologies clinically suggestive of Norwalk and Norwalk-like agents of human enteric viral gastroenteritis. The vast majority of these cases are associated with the consumption of raw mollusc's taken from harvest waters contaminated with raw or poorly treated human sewage.

Classification of molluscan growing waters based on valid human enteric virus indicators, as well as proper treatment and disposal of sewage are required to deter raw shellfish associated viral infections. Currently there is no accepted indicator for human viruses in shellfish or growing waters.

Depuration and relaying remove enteric bacterial pathogens and indicators from mollusc's. However, the depuration of some enteric viruses may be slow. *Hepatitis A* virus (HAV) persists far longer in oysters and clams than *E.coli*. Depurated mollusc's have been responsible for outbreaks due to enteric viruses.

Vital research is needed to develop an adequate viral indicator and (if possible) to develop enhanced depuration technology to reduce viral pathogens.

3. Naturally Occurring Marine Microorganisms

3.1 Bacterial Pathogens

The main pathogenic, free living, natural bacteria are of the *Vibrio* species. These are not generally associated with faecal contamination and their numbers tend to be higher in the warmer summer months. *Vibrio*'s are responsible for fewer cases of illness than viruses. However, as well as causing gastroenteritis some species are associated with high mortalities in people who are immunocompromised or have liver disease. *Vibrio vulnificus*, which is common in waters warmer than 20 C, has, in the US, killed 39 people between 1978 - 1987. Currently all shellfish harvesting in the southern US states is prohibited due to the severity of the problem with the death toll standing at 6.4 people per year in Florida state alone.

Testing for *Vibrio*'s is not a problem and research into removal of *V.parahaemolyticus* and *V.vulnificus* by depuration and relaying is ongoing. Depuration maybe possible but not within the 42 hour period currently used for bacterial removal.

3.2 Toxin Producing Algae

A number of species of algae produce toxins of one form or another. These algae are bioaccummulated by molluscs when algal blooms occur and the shellfish become toxic and remain toxic for several weeks after the bloom subsides. A number of different forms of poisoning have now been recognised :

PSP	Paralytic shellfish poisoning
DSP	Diarrhetic shellfish poisoning
NSP	Neurotoxic shellfish poisoning
ASP	Amnesic shellfish poisoning

There are currently no depuration methods for combating toxic algal bioaccummulation. However, there is scope for research in this field. The standard control measure is to not harvest/catch the shellfish until levels of the toxin have subsided naturally. This requires regular routine monitoring to ensure that the algal toxins are not present.

3.2.1 PSP

Paralytic shellfish poisoning results from the consumption of mollusc's that have bioaccummulated toxigenic dinoflagellates. The species of concern are *Gonyaulax catenella, G.tamarenses, Alexandrium minutum* and *Gymnodinium cateratum.* The cause of PSP is a complex of toxins known as saxitoxins which are neurotoxins. These are potentially life threatening however more usually cause tingling and numbness.

PSP can be tested for by Mouse bioassay, HPLC (high performance liquid chromatography), and ELISA (Enzyme linked immuno absorbent assays) tests. The latter method is not yet fully developed.

3.2.2 NSP

Neurotoxic shellfish poisoning or brevetoxic poisoning is caused by the red tide organism *Gymnodinium breve.* This causes respiratory irritation/gastroenteritis and is not usually fatal.

There is a mouse bioassay for NSP.

3.2.3 DSP

Diarrhetic shellfish poisoning is caused by *Dinophysis fortii, D.acuminata* and *Procentrum lima.* A number of toxins have been identified most commonly okadaic acid. Symptoms include diarrhoea, nausea and vomiting. This is not life threatening.

There is a mouse bioassay for DSP, HPLC can be used for okadaic acid determination.

3.2.4 ASP

Amnesic shellfish poisoning is the name given to the illness caused by consumption of domoic acid which is present in the diatom *Nitzschia pungens* and *N.pseudodelicatissima.* The symptoms of domoic acid poisoning are vomiting, diarrhoea, disorientation and memory loss. There have been reports of death in older people.

This can be tested for by HPLC.