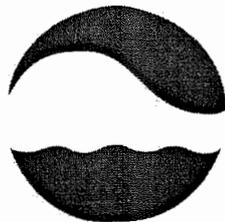


Aquafin CRC

**Aquafin CRC - Southern Bluefin tuna
Aquaculture Subprogram:
A risk assessment of factors influencing
the health of southern bluefin tuna**

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Project No. 2001/253

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2003

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ISBN 1 86295 099 7

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OBJECTIVES:

1. To provide a qualitative fish health risk assessment for the SBT aquaculture industry in Australia.
2. To review SBT health information from the industry (including their database), research organisations and scientific literature.
3. To identify areas of highest risk and propose management control measures for the industry, as well as research priorities.
4. To disseminate the results of this SBT health risk assessment project.

NON TECHNICAL SUMMARY:

The rapidly developing international tuna aquaculture industry started with a joint Japan/Australia experiment in 1991. Since then it has grown into the largest finfish aquaculture in Australia with an export value of \$290 million. It is based on the capture of wild fish and subsequent fattening of these fish in pontoons over a period of 3-6 months. Continuous husbandry improvements ensure very low mortality. This project was developed to review all available information on SBT health, assess current and potential risks in this area and provide the basis for future research and development in the area of SBT health.

1. To provide a qualitative fish health risk assessment for the tuna aquaculture industry in Australia

Under the current production systems, the fish captured from the wild for grow-out are generally three years old, with assumed natural mortality levels in the wild of 20-30% p.a. up to that stage. Therefore, the tuna captured for farming have already survived three years or more of health challenges and built up immunity to some diseases. This disease resistance, together with best management practices, results in extremely low mortalities in SBT industry. Additionally, SBT as a species appears to be relatively resistant to infectious diseases. Currently, overall risk for any disease ranges from negligible to low. Under the current culture system, where no therapeutic agents are used, no pressure on organisms to develop into more pathogenic forms is being applied. However, this risk assessment should be updated as farming practices and conditions change and more information on tuna health and physiology becomes available.

2. To review tuna health information from the industry (including their database), research organisations and scientific literature.

An extensive literature review was conducted to provide background information for the risk assessment. In general, there is a lack of published information on SBT health and tuna health in general. Publication of some of the information already

available on SBT would be advantageous to SBT farmers and scientists. For example the review of tuna parasites, compiled by the Tuna Boat Owners Association of South Australia (TBOASA) in 2000, should be published either as a CDROM or SARDI report. TBOASA and IDEXX-VPS has a collection of tuna pathology specimens. This collection can provide a historical record of the state of tuna health at the time of collection. There is a real need to establish an industry-driven health database, based on a surveillance and monitoring program. Further research is needed to provide more knowledge and understanding of tuna health.

3. To identify areas of highest risk and propose management control measures for the industry, as well as research priorities.

Existing parasites were identified as potentially highest risk (but still ranking only as low overall risk), particularly if grow-out time was extended. This risk can be minimised by mitigation, particularly as the parasite life cycles and reservoirs become well known. One of the highest priorities in the area of tuna health is to establish a well designed, transparent health surveillance and monitoring program, to ensure that there is an early warning if the disease risk increases. Furthermore, the literature review identified a general lack of information about SBT health and physiology. In particular, cardiorespiratory physiology of tuna is not well understood. Additionally, there is little information about life cycles of parasites and host-parasite interactions. Effects of environmental factors, for example harmful algal blooms, should also be evaluated.

4. To disseminate the results of this SBT health risk assessment project.

The results have been disseminated at stakeholders meetings and in this report.

OUTCOMES ACHIEVED

As a result of the literature review and risk assessment we have achieved greater ability to anticipate and guard against fish health related incursions in tuna farms and hatcheries. It was determined, based on the methods used and the limited information available, that currently overall disease risk ranges from negligible to low. Changes in overall risk due to potential changes in farming practices were also considered. The TBOASA fish health database was reviewed and categorized as a collection of pathology specimens and not a database. This has increased our knowledge of the extent and broader usefulness of the existing TBOASA fish health collection. Finally, research needs were determined and prioritized. This project forms the basis for further tuna health research as well as fish health monitoring and surveillance of the SBT industry.

KEYWORDS: southern bluefin tuna, SBT, aquaculture, health risks.

Acknowledgments

The SBT Aquaculture Subprogram's role in the management, coordination and review of this project is gratefully acknowledged. AusVet Animal Health Services was subcontracted and provided the review of the scientific literature and the review of the TBOASA health database, and also proposed the fish health surveillance and monitoring program and research needs. Some sections of their report, in particular the review of TBOASA database and background information for risk assessment, are part of the main report, other sections provided information only or are included as appendices. We are very grateful to AusVet Animal Health Services for their time and commitment to this project. We thank Dr Barry Munday and Dr Ewan Sergeant for providing literature review and other background information.

The SBT aquaculture industry provided helpful and constructive comments on this report, mostly during stakeholders meetings. We are grateful for their interest and time, in particular we thank Brian Jeffriess and Daryl Evans. We would like to acknowledge Dr Marty Deveney for providing expert information on fish parasites, Dr Ruth Reuter for providing information on SBT specimens collection at IDEXX-VPS and Dr Hidemasa Kawakami for providing scientific information on tuna health. We also thank Dr Sarah Kleeman, Biosecurity Australia, for her advice on risk assessment, in particular risk evaluation matrices.

This is an Aquafin CRC research project.

Background

The aquaculture of southern bluefin tuna (SBT) has been a major success story in the expansion of the Australian aquaculture industry. The industry started in 1991 and increased from 17 tonnes of SBT into the farms initially, to 5,185 tonnes in 2002. Despite the fact that most of the available tuna quota is already entering the aquaculture sector, continued industry growth is expected due to further technical development. This includes: improved survival of SBT, holding of SBT for longer periods of time; improvements in product quality leading to better market prices; and advances in feeds, feeding strategies and farming technologies leading to reduced operating costs.

Under the current production systems, the fish captured from the wild for grow-out are generally three years old, with assumed natural mortality levels in the wild of 20-30% per annum up to that stage (CCSBT 2002). Therefore, the tuna captured for farming have survived three years or more of health challenges. While the SBT industry has not been affected by disease outbreaks, further intensification of tuna industry and closing of the tuna life cycle may increase the risk of health problems.

Diseases can be prevented only when the risks are recognised and managed before the diseases occur. Risk analysis is a structured process providing help to decision-makers (MacDiarmid 2001). It is a process comprising hazard identification, risk assessment, risk management and risk communication (Wooldridge 2001). The identification of risk factors (hazard identification or a risk register) is crucial for developing preventative measures (Thrusfield, 1995). There is a wide range of information from tuna health surveys available in the industry, however most of this information has not been fully utilised. Published scientific literature also reports problems in the aquaculture of scombrids and other relevant health risks. There is therefore a clear need to review all the information available.

During the first few years of commercial operations of tuna aquaculture, low levels of mortalities were recorded from an unexplained cause. This condition was called swimmer syndrome and the cause was subsequently identified by Dr Barry Munday, University of Tasmania as the ciliate protozoan *Uronema nigricans*. Successful AusIndustry funding through their Graduate Employment Program led to the Tuna Boat Owners Association of South Australia (TBOASA) appointing a tuna health officer to address the swimmer syndrome issue, refine pathology techniques for SBT, and obtain baseline information on parasites, haematology, blood chemistry and histology in association with the University of Tasmania, University of Queensland and South Australian Research and Development Institute (SARDI). The FRDC soon after funded a project through SARDI/University of Tasmania to enhance detection of *Uronema nigricans* using immunofluorescent staining techniques (Project No 95/083).

The tuna farmers also sought to improve work practices by developing an Industry Code of Practice and reduce mortality of tuna through improvements in capture and towing techniques, improved understanding of the fish health implications of higher quality feeds, mixed feeds and targeted feeding programs as well as improved understanding of the health impacts of integrating husbandry with environmental management. These changes resulted in minimisation of the stress of capture, transport and holding as well as a significant reduction in overall

mortalities. The modern SBT aquaculture industry has total capture to harvest mortalities around 2-4%.

While a range of parasites has been identified in wild species of tuna (Langdon 1990, Oldewage 1993, Muruges and Madhavi 1995, Petter and Cabaret 1995, Moravec et al 1999, Kohn et al 2001), few caused health problems in a culture situation (Munday et al 1997, Cribb et al 2000, Colquitt et al 2001, Munday et al 2003). Under the current culture system, no pressure on organisms to develop into more pathogenic forms is being applied. However, it is well known that an intensification of other aquaculture industries resulted in an emergence of new health problems. In particular, marine hatcheries often suffer from disease outbreaks, due to the susceptibility of early life stages to pathogens and the potential for vertical transmission of diseases. Additionally, it has been suggested that some environmental factors, for example blooms of raphidophyte flagellate *Chatonella marina*, may contribute to tuna mortalities during farming (Hallegraeff et al 1998, Munday and Hallegraeff 1998).

The main objectives of this project are to provide qualitative fish health risk assessment for the tuna industry in Australia; to review tuna health information available from the industry, research organisations and scientific literature; to identify areas of highest risk and propose risk management strategies including control measures for the industry and research priorities for tuna health. This project is critical as it provides, for the first time, a comprehensive review of the potential SBT health issues based on relevant published information and the TBOASA existing database.

This project will provide a sound basis for future SBT fish health research and development activities. The risk assessment process should be considered dynamic and reviewed on a regular basis as more comprehensive information becomes available.

Need

While the SBT aquaculture industry has not been substantially affected by disease outbreaks to date, further development and possible intensification of not only the tuna industry, but aquaculture in general in the tuna farming regions, will increase the risk of tuna health problems in the future. Significant disease related mortalities are best prevented by recognising and managing the risks before they become a major issue.

While the SBT industry has been proactive in their attempts to document tuna health, the TBOASA tuna health database and information from tuna health surveys have never been externally reviewed. The extent and usefulness of their contents should be evaluated and reported. Published scientific literature also contains reports on SBT health issues, as well as other finfish health risks related to SBT aquaculture and there is a clear need to review the information available and identify the need for future research.

Objectives

1. To provide a qualitative fish health risk assessment for the tuna aquaculture industry in Australia.
2. To review tuna health information from the industry (including their database), research organisations and scientific literature.
3. To identify areas of highest risk and propose management control measures for the industry, as well as research priorities.
4. To disseminate the results of this SBT health risk assessment project.

Methods

Hazard identification

For the purpose of this study, a potential hazard is defined as:
A condition that has the potential to negatively impact tuna health.

A wide range of potential hazards to tuna health were identified either from the published literature, from expert opinion or from survey of stakeholders, including the SBT industry. Potential hazards were classified as either infectious or non-infectious. In addition to the specific hazards to tuna health, the impacts of a large number of contributing factors were considered as part of the risk assessment process.

Risk assessment

This risk assessment was based on information provided by the SBT industry (Appendix 3) and the literature review (including causation webs) provided by AusVet Animal Health Services (Appendix 5). A web of causation was developed for each hazard regarded as potentially significant, identifying the various contributory effects and their likely magnitude for the occurrence of the hazard. Based on these webs, and other information on the epidemiology, the likely effects of these hazards and the tuna industry, risk assessment was carried out for each potential hazard. In the face of uncertainty and lack of information, conservative judgements were made regarding the expected impact or significance of disease establishment. Risk assessment was performed using a risk evaluation matrix (Figure 1) and followed the process described by AFFA (2001). Risk is the product of probability and consequences. When interpreting the risk estimation matrix it must be remembered that although the descriptors for each axis are similar (negligible, very low, low, moderate, high, extreme), the vertical axis refers to probability and the horizontal axis to consequences (AFFA, 2001). This means that the matrix cannot be symmetrical as a "negligible" probability combined with "extreme" consequences is not the same as "extreme" probability combined with "negligible" consequences. Additionally, "risk" is expressed in the same units as are used to estimate consequences (AFFA, 2001).

Key factors used in classifying significance of disease were:

- biological effects on SBT
- availability, cost and effectiveness of control
- economic effects at an enterprise/industry/national level

Terms used to describe severity of impact (consequence of entry, establishment and spread) were:

- negligible - no biological consequences (no mortality or morbidity and no significant pathological changes), transient or easy to control or eradicate, low economic effects at individual enterprise level and insignificant at a regional level
- very low - very mild biological consequences (extremely low mortality or morbidity and very little of significant pathological changes), normally easy to control or eradicate, may affect economic performance at enterprise level but no significance at industry level

- low – mild biological consequences (low mortality or morbidity and low prevalence of significant pathological changes), can be controlled or eradicated, may affect economic performance at enterprise level but negligible significance at industry level
- moderate – some biological consequences (some mortality or morbidity and some significant pathological changes), can be controlled or eradicated but only at significant cost, significant harm at enterprise level but not national level
- high – serious biological consequences (high mortality or morbidity and significant pathological changes), prolonged effect on industry (greater or equal to a normal production cycle), not easy to control or eradicate, significant harm to economic performance at industry level
- extreme – catastrophic biological consequences, total mortality within short period of time, having catastrophic effect on the industry, significant effect on economic performance at national level

Terms used for likelihood (probability of entry, establishment and spread) were:

- extreme – the event would be extremely likely to occur
- high – the event would be very likely to occur
- moderate – the event would occur with even probability
- low – the event would be unlikely to occur
- very low – the event would be extremely unlikely to occur
- negligible – the event would almost certainly not occur

Risk Estimation Matrix

Probability of entry, establishment and spread	extreme	negligible	very low	low	moderate	high	extreme
	high	negligible	very low	low	moderate	high	extreme
	moderate	negligible	negligible	very low	low	moderate	high
	low	negligible	negligible	negligible	very low	low	moderate
	very low	negligible	negligible	negligible	negligible	very low	low
	negligible	negligible	negligible	negligible	negligible	negligible	very low
		negligible	very low	low	moderate	high	extreme

Consequence of entry, establishment and spread

Figure 1. Risk estimation matrix (based on AFFA, 2001).

This risk assessment was performed for the tuna industry assuming no change in its current operation. To assist with industry planning for future development, risk assessment was also performed according to three alternative scenarios for industry structure in the future. These scenarios were:

1. Longer grow-out

This scenario assumes that the industry remains much the same as at present, but that fish are grown out for longer periods for example 12 to 15 months.

2. Polyculture

This scenario assumes farming of other species in close proximity to the existing industry.

3. Propagation

This scenario assumes the development of a tuna hatchery and nursery in close proximity to the existing wild-caught tuna farms. For this scenario, risks were subdivided into:

- risks to the hatchery
- risks posed by the hatchery to the existing wild-caught industry
- risks associated with grow-out of juveniles from the hatchery

By their very nature, these scenarios are hypothetical. These scenarios are therefore intended to highlight areas of potential concern, rather than make definitive assessments of likely events in the case of various changes within the industry. Information for these scenarios was provided by AusVet Animal Health Services and the SBT industry.

Review of TBOASA tuna health database

The TBOASA database was reviewed by Dr Angus Cameron, AusVet Animal Health Services through interviews and discussions with David Ellis, Kirsten Rough and Dr Ruth Reuter as well as examination of available materials, mainly at the IDEXX-VPS laboratories in Adelaide and at the TBOASA office in Port Lincoln. Electronic records were also examined together with a number of reports and data tabulations provided by the TBOASA and SARDI.

Research needs

Research needs and priorities were first identified by AusVet Animal Health Services and further discussed and prioritised at a stakeholder workshop on tuna health risks held in Port Lincoln on 4 November 2002. This included an interactive exercise, where each stakeholder present could decide how to divide research funding among different research priorities as well as add new research priorities. Results were collated and presented as percentage of total and rank. Research needs were determined by the authors of this report with input from a stakeholder workshop (tuna health risk workshop on 4 November 2002) and the report from AusVet Animal Health Services (Appendix 4).

Results/Discussion

Risk register – hazard identification

A wide range of potential hazards to tuna health were identified and classified as either infectious disease or non-infectious condition. The following specific hazards to tuna health were identified in accordance with the definition above:

Infectious

- (i) Viral
 - Vacuolating encephalopathy and retinopathy
 - Red sea bream iridovirus infection
 - Yellowtail ascites and viral deformity
- (ii) Bacterial
 - Pasteurellosis
 - Mycobacteriosis
- (iii) Protozoan:
 - Scuticociliate
- (iv) Metazoan
 - Acanthocephala
 - Arthropoda
 - Amphipod unspecified
 - Isopod unspecified
 - Aschelminthes
 - Myxozoa
 - Kudoa* sp.
 - Platyhelminthes

Non-infectious

- Hypoxia
- Nutritional
 - Vitamin E depletion
 - Vitamin C depletion
- Toxicological
 - Microalgal toxicosis
- Predation
- Suspended sediments

It is worth noting that a number of other potential hazards were considered in the hazard analysis, but they did not meet the definition presented. In some cases, this was because, while posing a threat to the SBT *industry* (for example the presence of chemical residues), they did not pose a specific threat to the health of the fish. Such potential hazards have been omitted because they are beyond the scope of this report.

Risk assessment

Current SBT industry

A detailed description and the results of the risk assessment for each of the potential hazards is provided in Appendix 4. The results are summarised in Table 1. This risk assessment is based on the assumptions that are listed in Appendix 3 and 4, and is relevant to the SBT industry as it is today. All hazards were ranked as low, or very low or negligible risk.

The overall risks ranged from negligible to low. While this is an excellent result, supported by the history of no health problems and low health-related mortalities in the SBT industry, any risk greater than negligible can be minimised. The risks which are rated at the highest level (low) include: gill fluke, blood fluke and hypoxia.

Gill fluke infection

This gill fluke is an unidentified capsaline (M Deveney pers. com.) monogenean (Rough, 2000, type 3). It has been reported only from wild and farmed SBT in southern Australian waters. Under current culture conditions the prevalence seems to decrease with time in captivity, however number of parasites may increase on gills of an individual fish. It is not known to directly cause mortality. The parasite produces focal lamellar fusion which is seen macroscopically as white patches on the gill surfaces and has been linked to respiratory distress. This parasite is already present in SBT in Australia. The probability of exposure has been assessed as high based on Rough's (2000) observations. Because this parasite usually occurs in heavy infestations in only small numbers of fish, the consequences of entry, establishment and spread was assessed as low. Thus, the overall risk is low. Further research is required into the biology and epidemiology of this parasite, as little is currently known about it.

Blood fluke infection

Cardicola forsteri (*Digenea Sanguinicolidae*) has only been recorded from SBT in southern Australian waters (Rough 2000, parasite incorrectly named as *Cardicola smithii*). Colquitt (1999) found no evidence of infection in newly captured fish but three months later 66% had eggs embedded in the myocardium and another month later 100% had eggs in the myocardium and 50% had eggs impacted in the afferent filamentary arteries. Rough (pers. com.) reported that it is common for harvest fish to have up to 35% of individual batches with typical lesions when harvested between May and August. As the post 60 day mortality is very low in farmed tuna and blood fluke tends to peak later (Rough 2000), this parasite in itself must be a small contributing factor in mortalities. The gills of infected fish have multifocal, pale lesions often extending in an arc across the gill arch. These lesions are attributable to the granulomatous reaction resulting from the presence of eggs and developing miracidia in the gill filaments. It is reasonable to presume that these lesions affect the respiratory efficiency of the gills. Probably more important is the effect on the heart where the host reaction leads to myocardial damage and the "back-pressure" from the occluded afferent filamentary arteries leads to ventricular hypertrophy (Colquitt et al. 2001).

Until more is known about the life cycle of this parasite it is not possible to make meaningful recommendations on control. Because this infection is a known parasite of SBT in Australia, the probability of entry, establishment and spread was

assessed as extreme (eg, 100% prevalence in June 1999 (Colquitt, 1999)). It is possible that this parasite may occur at high prevalence in farmed fish and may contribute to reduced fish performance. However, there is no evidence that this occurs, so the consequences of infection have been assessed as low. Further research is required into the biology and epidemiology of this parasite. A survey of other fish species in the vicinity of the tuna pontoons for the presence of adult flukes should be undertaken to ascertain which, if any, of these are infected. Identification of the intermediate hosts is essential for the development of control measures.

Hypoxia

Hypoxia may arise due a number of different environmental factors including algal blooms, environmental conditions, gill damage and increased SBT oxygen requirements under stress or other conditions. Prevention depends upon choosing appropriate sites for pontoons, ensuring that management practices do not contribute to hypoxia (i.e. ensure good water flow through nets) and, where appropriate, by avoidance, for example by towing nets away from algal blooms or other causes of de-oxygenation. Feed should be withheld during times of danger. The regulation of a maximum stocking rate of 4 kg/m³ of pontoon area appears to address the potential hypoxia problem. Furthermore, after 10 years of tuna farming there is no evidence of oxygen depletion. The only time there had been a problem was when some farmers continued to feed during dodge tides. Currently, this is not done. The dissolved oxygen values obtained in government monitoring programs have always been above 6.0 mg/L. Because many of the causative factors have been identified and are effectively managed, the probability of exposure has been assessed as very low. Because of the presumed high susceptibility of SBT to hypoxia, the consequences of exposure have been assessed as extreme. Overall risk is low. There is a need for research on the physiology of SBT.

Table 1. Summary of risk assessment findings and suggested additional research for potential fish-health hazards for the farmed tuna industry in Australia.

Hazard	Probability of entry and spread	Consequence of entry, establishment and spread	Overall Risk	Occurrence in SBT	Comments & additional research
<i>Viral diseases</i>					
Vacuolating encephalopathy and retinopathy	moderate	negligible	negligible	No	Further research is required to clarify the susceptibility of SBT to VER viruses.
Red sea-bream iridovirus infection	very low	low	negligible	No	Japanese report that only 0-1 year class of tuna are susceptible to RSIV. Baitfish need to be tested for susceptibility to RSIV. Local snapper, yellowtail kingfish and other susceptible fish could be surveyed for the RSIV.
Yellowtail ascites and viral deformity	high	very low	very low	Not known to be present in Australian waters	Maintain contact with appropriate Japanese researchers regarding possible cases in Pacific bluefin tuna. Also, local yellowtail kingfish could be surveyed for presence of the viruses.
<i>Bacterial diseases</i>					
Pseudotuberculosis	low	low	negligible	No	Maintain an overview of the pertinent literature.
Mycobacteriosis	moderate	negligible	negligible	Yes	Mycobacteriosis is unlikely to be a problem in the present SBT industry unless baitfish species and/or sources are changed without examination of the fish for mycobacteriosis.
<i>Protozoan diseases</i>					
Scuticociliate infection	low	very low	negligible	Yes	Improved husbandry has reduced the prevalence of "swimmers" in cultured SBT from low to rare but scutociliate infections will be a major risk to larval/juvenile tuna if a hatchery is established.
<i>Metazoan diseases</i>					
<i>Kudoa</i> infection	low	very low	negligible	Yes	Further research is required into the epidemiology and control of this parasite, particularly if a propagation and grow-out facility is planned.
Gill fluke (capsaline) disease	high	low	low	Yes	Further research is required into the biology and epidemiology of this parasite.

Hazard	Probability of entry and spread	Consequence of entry, establishment and spread	Overall Risk	Occurrence in SBT	Comments & additional research
Blood fluke infection	extreme	low	low	Yes	Further research is required into the biology and epidemiology of this parasite.
<i>Caligus elongatus</i> infection	moderate	low	very low	Yes	Should be monitored, especially if longer grow-out is instituted.
<i>Non-infectious conditions</i>					
Vitamin E depletion	low	low	negligible	N/A	Further research is required into the vitamin E status for juvenile tuna if hatchery rearing is proposed.
Vitamin C depletion	low	low	negligible	N/A	Collaboration with Japanese on cause(s) of deformed opercula in hatchery fish which may be due to vitamin C deficiency.
Microalgal toxicosis	moderate	low	very low	N/A	Further research is required into the likelihood of occurrence and management of toxic blooms, as well as their potential effect on farmed SBT.
Toxicants (methane and hydrogen sulphide)	negligible	low	negligible	N/A	No research required at present.
Hypoxia	very low	extreme	low	N/A	Determination of the normal cardiorespiratory physiology of SBT is an essential requirement.
Suspended sediments	low	low	negligible	N/A	No research required at present.
Predation	low	moderate	very low	N/A	Current deterrent methods as presently used by farmers seems to have reduced pinniped interactions.

Scenario 1: Longer grow-out of wild-caught tuna

The main effects under this scenario would be:

- wild-caught SBT would be held in pontoons for a longer period of time
- there may be less opportunity to fallow sites

However, as stated previously, the tuna in longer grow-out will still be generally 3 years old, which means that they have survived three or more years of health challenges and have acquired resistance as well as innate resistance to at least some diseases. Current mortality patterns (majority in the first 60 days) suggest that longer holding is not causing any problems.

The risk assessment is presented in Table 2. The overall risk increased for blood fluke (low to moderate) and predation (very low to low) - compared to the current situation, (see Table 1). This is a result of the assumption that, with greater holding time, there will be more opportunities for formation of parasite reservoirs in the farming area, for example in net fouling. This may require changes in farming practices, for example changing nets *in situ* and mitigation against fouling of the nets. If the parasite reservoirs were reduced it would lower the risk. Scuticociliate infection occurs only in low temperatures and extended tuna grow-out would still only include one winter, so the period of temperatures below 15°C (May-June) would be the same as in current grow-out. Prevalence of metazoan parasites, other than blood fluke, seems to decrease with holding time, while *Caligus elongatus* infestation is solely a problem during tow.

Longer grow-out would result in longer exposure to the environmental conditions, which may increase risks of algal bloom, suspension of sediments and predation. It is highly unlikely, however, that any reduction in the “natural” fallowing period (current grow-out is only for 3-6 months per year) would not be managed by regulation and farm practices. For example, fallowing might be required every 12-15 months, rather than the present 2 years. Farmers can move pontoons during the grow-out. Maximum stocking density could also be lowered. Any potential change in sediment toxicity can be managed through adjustments in regulations and farming practices.

Furthermore, the Tuna Research Farm held significant number of SBT continuously from 1991 to 1996 with very limited mortalities. This has been overcome by a whole range of mitigation devices, which reduced seal interactions to a rare occurrence. Modern tuna farming is even more advanced and improved, with an acute awareness of avoiding rancidity in imported feeds. The general grow-out conditions have improved across the industry due to improvement in the farming practices and diet as well as better environmental conditions at current lease sites.

Table 2. Summary of risk assessment findings for extended tuna grow-out.

Hazard	Probability of entry and spread	Consequence of entry, establishment and spread	Overall Risk	Occurrence in SBT
<i>Viral diseases</i>				
Vacuolating encephalopathy and retinopathy	moderate	negligible	negligible	No
Red sea-bream iridovirus infection	very low	moderate	negligible	No
Yellowtail ascites and viral deformity	high	very low	very low	Not known to be present in Australian waters
<i>Bacterial diseases</i>				
Pseudotuberculosis	low	low	negligible	No
Mycobacteriosis	moderate	negligible	negligible	Yes
<i>Protozoan diseases</i>				
Scuticociliate infection	low	very low	negligible	Yes
<i>Metazoan diseases</i>				
<i>Kudoa</i> infection	moderate	very low	negligible	Yes
Gill fluke (capsaline) disease	high	low	low	Yes
Blood fluke infection	extreme	moderate	moderate	Yes
<i>Caligus elongatus</i> infection	moderate	low	very low	Yes
<i>Non-infectious conditions</i>				
Vitamin E depletion	low	low	negligible	N/A
Vitamin C depletion	low	low	negligible	N/A
Microalgal toxicosis	high	low	low	N/A
Toxicants (methane and hydrogen sulphide)	negligible	low	negligible	N/A
Hypoxia	very low	extreme	low	N/A
Suspended sediments	moderate	low	very low	N/A
Predation	moderate	moderate	low	N/A

Scenario 2: Polyculture

The main influences polyculture may have on affecting the risk of disease in the tuna industry are:

- introduction of species carrying disease agents which can affect SBT
- increasing the biomass in the culture areas

The actual risk of disease due to the first mechanism depends on the species that are cultured with SBT. The only impact of introducing polyculture species which are not capable of sharing infectious disease with SBT is through increasing the biomass. However, this is not a necessary consequence of polyculture, and depends on management policies.

Polyculture species

In the short term, finfish species that may be cultured in proximity to SBT are yellowtail kingfish, *Seriola lalandi*, and snapper, *Pagrus auratus*. Both these species are susceptible to exotic iridoviruses which can infect tuna. They are also affected by skin and gill parasites, however these are host-specific (M. Deveney, pers. com.) Other polyculture species that have been mentioned include molluscs. Molluscs may be able to harbour certain viruses, and act as intermediate hosts for parasites. Molluscs are not known to play a role in the lifecycle of any of the parasites that have been identified in the risk register. However, until, for instance, the identity of the intermediate host for the blood fluke, *C. forsteri*, is known, and the full host range of potential viral pathogens is identified, they cannot be entirely discounted as potential threats.

Clearly, the manner in which polyculture is conducted may have an impact on disease risk. For instance, if closely related species were cultured in the same pontoon, there would be a greater opportunity for the spread of disease than if the new species were cultured in separate pontoons. Some polyculture schemes may also increase the risk of trauma, but it is assumed that these concerns would be addressed by management interventions.

Hazards

Increases in risk of disease due to increased biomass in polyculture systems would be similar to those described for the previous scenario of a longer grow-out period. The main effects would be related to increase sediment build up below pontoons (if indeed the polyculture system resulted in this) and the resultant effects. No change in disease risk ranking from the current situation is suggested for this, as it is assumed that any such problems could be adequately resolved through management changes.

If the species introduced were capable of sharing disease with SBT (but did not result in the introduction of new disease), then again, there would be no significant change in risk ranking. The other species would play a similar role to the presence of other SBT under the current system. Higher stocking densities would offer greater opportunities for disease spread, but again, it is assumed that this would be controlled through good management.

Yellowtail polyculture

As indicated in the Risk Assessment the expert opinion is that *Benedenia seriolae*, a parasite of yellowtail kingfish, does not infect SBT.

Yellowtail in Japan suffer significant mortality as a result of infection with the bacterium *Lactococcus garviae*. This organism causes significant mortalities in rainbow trout in Australia from time to time. Indeed, in the early days of mariculture of salmonids in Tasmania it caused significant mortalities in ocean trout but not Atlantic salmon. However, it has ceased to be a problem, presumably as a result of improved management. As *Lactococcus garviae* only causes disease in some species, and has never been reported from tuna, it may not constitute a risk to SBT even if an outbreak occurred in yellowtail nearby. In relation to spread in the marine environment, experience in Tasmania was that spread only occurred between pontoons in close association.

YAV or VDV may already be present in Australian waters, but are only likely to result in disease in intensive culture. It is currently believed that SBT is not susceptible to infection; however, if yellowtail were intensively cultured in close proximity to SBT, this assumption may be tested. The very low risk of susceptibility does not warrant a change in risk ranking for YAV and VDV for this scenario.

Scenario 3: Propagation

Based on the results of the risk analysis, a number of risks and research issues were identified that are specifically applicable to the propagation of SBT in Australian waters. These are issues of varying importance to the existing industry, but which have the potential for a serious impact on SBT health if propagation facilities were established in Australia as an alternative source of fish for the existing tuna industry. These problems could have a significant effect at company level but not on the part of the industry still relying on wild-caught SBT.

Hatchery risks

The risk analysis results relevant specifically to a hatchery are summarised in Table 3. Key issues of concern are scuticociliate infection and collision trauma.

The larval form of the disease caused by scuticociliates (as opposed to swimmer syndrome) is caused by an as yet not fully identified species. It is very likely that the causal agent(s) are present in Australian waters and that, based on experience elsewhere, infections would occur in a hatchery. The high morbidity and mortality rates are offset by the potential to control and the limitation of effects to company level, meaning that the overall risk is moderate in this scenario.

By analogy with *Pentacapsula* sp. in striped trumpeter, *Kudoa* sp. infection may cause greater problems than are currently seen if a hatchery was established. In wild caught tuna, infection rates are very low, but hatchery reared tuna, due to their high stocking rates and the decreased effect of natural selection to weed out weaker fish, are likely to develop a much higher rate of infection prior to grow out. This may not result in any overt disease within the hatchery, but there is a significant risk that a higher proportion of fish reared from hatchery stock may show carcass damage. This would be manifested through an increase in the risk of exposure. While the risk would increase over the risk posed by the current culture system, this risk would still remain negligible due to the very low consequences of infection.

Other conditions of concern to the existing industry may also be of concern in a propagation facility, including RSIV, microalgal toxicosis, blood fluke infection and capsaline gill fluke disease, although the risk is likely to be similar to that experienced in the current situation.

Table 3. Summary of risk assessment findings and suggested additional research for a tuna hatchery in Australia. The overall risk refers to effects on the whole SBT industry (see definitions in Methods).

Hazard	Probability of entry and spread	Consequence of entry, and spread	Overall Risk	Comments & additional research
Vacuolating encephalopathy and retinopathy	moderate	negligible	negligible	
Red sea-bream iridovirus infection	very low	moderate	very low	0-1 year class tuna susceptible in Japan, mortality increases with temperature over 25°C.
Yellowtail ascites and viral deformity	high	very low	very low	
Pseudo tuberculosis	low	low	negligible	
Mycobacteriosis	high	low	low	Opportunistic organism capable of surviving in the environment. Not reported by Japanese researchers in their hatcheries.
Scuticociliate infection	extreme	moderate	moderate	Opportunistic environmental organism. Hatchery not likely to add significantly to total environmental population (cf under pontoon organic residues). Can be controlled by generating copper ions in the water.
<i>Kudoa</i> infection	moderate	very low	negligible	Further research is required into the epidemiology and control of this parasite.
Gill fluke	high	low	low	
Blood fluke	extreme	low	low	
<i>Caligus elongatus</i> infection	moderate	low	low	
Vitamin E depletion	moderate	moderate	low	Further research is required into the Vitamin E requirements of juvenile tuna if hatchery rearing is proposed.
Vitamin C depletion	moderate	moderate	low	Collaboration with Japanese on cause(s) of deformed opercula which may be due to hypovitaminosis C would be desirable.
Algal toxicity	moderate	low	very low	
Toxicants	negligible	low	negligible	Methane and hydrogen sulphide
Hypoxia	very low	extreme	low	
Sediments	low	low	negligible	Suspended sediments
Predation	low	moderate	very low	
Collision-trauma during grow-out	extreme	moderate	moderate	Maintain contact with the Japanese in regard to control measures.

Risks posed by the hatchery to the existing wild-caught industry

The main potential risk that a hatchery could pose to the existing wild-caught industry relates to the spread of contagious disease direct from the hatchery, or from hatchery reared fish in grow-out. This could occur due to:

- the introduction of new contagious diseases into the area through the hatchery
- a change in the frequency of existing (possibly currently inapparent) diseases in hatchery reared fish, leading to increased exposure of wild-caught fish

Introduction of new disease

As the source of broodfish for any hatchery would be from the stocks used for “normal” SBT aquaculture, the spectrum of infectious diseases in the hatchery will logically reflect those of the population being fattened. Thus, if locally sourced broodstock were used, there will be no source of “new” diseases from a hatchery through this source. Another source of new infections is through the feed, and if different sources of feed from different origins were used in the hatchery, this would pose a different risk of the introduction of exotic diseases. It is assumed that only locally sourced broodstock would be used in the hatchery and that the same precautions for imported foodstuffs would be used as are applied by the wild-caught industry. Based on these assumptions, there would be no significant increase in the risk of the introduction of diseases exotic to Australia and/or the Port Lincoln area due to the establishment of a hatchery.

Change of frequency of existing pathogens

Due to high stocking densities, difficulties in maintaining optimal nutrition, and a decrease in the pressure of natural selection that is exerted in the wild (resulting in low survival rates but only the healthiest fish being available for capture), there are greater opportunities for the spread of infectious disease within a hatchery. Potential pathogens that are either currently unrecognised, or are considered to be non-pathogenic, could multiply within the hatchery, resulting in high prevalence in the stock produced. This high burden amongst hatchery-reared fish may act as a heavy source of contamination for nearby wild-caught fish, with the result that they become affected by the disease.

As no such pathogens are currently recognised, this possibility is based solely on conjecture, and must be considered to be relatively unlikely. Furthermore, effluent discharge regulations for hatcheries and translocation policy controlling release of stock to grow-out sites should reduce the risk of pathogens entering the natural environment from hatcheries either in effluent or carriers.

Spread of disease from the hatchery

A final possibility is that multiplication of pathogens with the more susceptible population within the hatchery could result in a heavy pathogen load in water discharged from the hatchery, leading in turn to increased infection rates in wild-caught fish. It is assumed that measures would be taken to prevent this happening. Filtration followed by ozonation of waste water from the hatchery should be sufficient to minimise any risk of the hatchery as a source of disease for wild-caught fish.

Conclusion

Each of the risks discussed above are largely theoretical. If they did prove to be a reality, it is likely that hatchery management measures would be able to control them. Provided any hatchery is properly managed it should not pose a significant

disease risk to the older tuna caught from the wild for fattening. Regulations controlling effluent discharge and relocation of stock to grow-out as well as availability of reliable and fast diagnostic tests would further reduce the risks.

Risks associated with grow-out of fish from the hatchery

Grow-out fish from a hatchery would face different disease risks to wild-caught fish under the current culture system due to three main factors:

- they represent a potentially more susceptible population
- they may have a higher prevalence of disease at the start of the grow-out period
- they require a longer grow-out

As previously mentioned, the population of fish produced from a hatchery is likely to be different to the population of fish that survive to grow-out in the wild. This is because hatchery conditions are designed to minimise mortality, resulting in a higher proportion of less robust or immunologically less competent fish in the population. This may result in a generally higher level of disease problems during grow-out.

Related to the first point, but also due to the ease of disease transmission within a hatchery, it is likely that the stock produced will have higher prevalence than would be present in the wild-caught population. For instance, *Kudoa* sp prevalence (initially at very low numbers of parasites) may be much higher, resulting in significantly higher production losses after harvest.

However, the effect that is probably the most important is the fact that hatchery reared fish require a longer grow-out period, and thus suffer all the same increases in risk of health problems that have already be described for longer grow-out of wild-caught fish.

TBOASA tuna health database review

Provided as part of AusVet Animal Health Services report to Aquafin CRC

Introduction

A database, in its usual usage, refers to a computerised collection of data, organised in such a fashion as to enable the convenient storage, retrieval and analysis of the data. In a broader sense, it may be applied to a similarly organised collection of materials, rather than just data. An important feature of a database is that it has a value greater than the sum of its parts. For instance, individual records in a diagnostic database have obvious value in providing a diagnosis, which may help respond to a disease problem. However, the cumulation of information on all diagnoses has an added value of providing an industry-wide picture of the types of disease problems encountered, useful for surveillance, monitoring, future planning and priority setting.

The form of the database, its contents and its outputs depend largely on the objectives that it aims to meet. A number of potential objectives for the tuna health database were considered, discussed in more detail below:

- surveillance and monitoring
- pathology reference collection
- reference for biochemistry / haematology normal values
- examples for teaching
- historical record

The approach used to review the Tuna Health Database was based on an examination of available materials, mainly at IDEXX-VPS laboratories and at TBOASA in Port Lincoln. Electronic records were examined, as well as reviewing a number of reports and data tabulations.

Materials Available

IDEXX-VPS

Information on tuna submissions held by the IDEXX-VPS Laboratory in Adelaide is derived from two sources:

- the TBOASA submissions (mainly through Kirsten Rough)
- SARDI research

Specimens submitted by TBOASA are almost entirely made up of mixed organ (liver, spleen, kidney, gill, brain) formalin-fixed specimens for histopathological examination and have been submitted with no history. The vast majority of these have had no pathology identified. A smaller number of blood samples have been received.

Specimens submitted by SARDI have included a small number of whole carcasses, as well as tissue and blood samples.

Haematology and biochemistry reports have been produced for the blood samples, and the quantitative results of these are stored in the IDEXX-VPS computerised database, making extraction feasible. Tissue samples have been processed and examined histopathologically. All reports, fixed specimens, blocks and slides from TBOASA samples have been returned to TBOASA in Port Lincoln.

Histopathology reports from IDEXX-VPS are created as MS Word documents. Recently, some of the changes observed have been coded, and these codes have been included in the report. However, details of the reports have not been stored in a computerised database. None of the reports include details of history or presenting complaint, and few are able to provide a diagnosis.

Data management systems at IDEXX-VPS only allow the number and date of submissions from particular clients to be reported. As most of the submissions have consisted of multiple specimens, they are not able to report the number of specimens received. This would require manual examination of all text reports.

Specimens have been received from three wide-scale disease events over the past decade. The total number of specimens submitted is not currently available, as IDEXX-VPS only records submissions, and the number of specimens per submission

has been very variable. This can only be determined by manually examining the reports.

TBOASA specimens

TBOASA holds the blocks and slides for all specimens sent to IDEXX-VPS. Formalin fixed samples were also held, but these have been discarded as leakage was creating a serious storage problem.

TBOASA also has hard copies of all the IDEXX-VPS pathology reports for specimens submitted.

Slides and blocks are labelled with identification numbers, and these are generally referenced on the written pathology reports. However, there is no information available about the origin of the specimens, their location, history or signs.

Other materials and outputs

There have been a number of outputs based on the information and material described. These include:

- industry reports
- Kirsten Rough's Master's thesis (Rough K.M. (1998). Haematology of Farmed and Wild Southern Bluefin Tuna, M App Sci thesis, University of Tasmania)
- a limited distribution Atlas of Parasites by Kirsten Rough (Rough K.M. (2000) An Illustrated Guide to the Parasites of Southern Bluefin Tuna, *Thunnus maccoyii*. Tuna Boat Owners Association of South Australia, Eastwood)
- a number of published and unpublished papers
- "Tuna health summary" tables prepared by Kirsten Rough
- tables of observed values for a range of metabolic parameters in tuna blood, prepared by Kirsten Rough

The reports in particular contain extensive environmental data, for example algal monitoring; however they are not organised for easy retrieval or summary. While all these outputs are valuable in their own right, they cannot be construed to form part of a database (as discussed above) and will not be discussed further.

Conclusions

In summary, the available material consists of:

At TBOASA:

- blocks
- slides
- hard copy reports

At IDEXX-VPS:

- MS Word format reports
- electronic records of haematology and biochemistry results

In general, reports can be associated with blocks and slides, but the origin, history and signs of individual fish from which the specimens were taken is not known for any of the materials.

Using the definition provided above, it is not possible to refer to the materials held by TBOASA or IDEXX-VPS as a database, as they are not organised for retrieval or

analysis. It would be more accurate to describe them as a collection of pathology specimens. The main feature that is missing, and which prevents the collection being considered as a database, is the presence of structured data allowing the classification of specimens. Ideally this structured data would provide information to classify the specimens with respect to:

- date of collection
- location of collection / owner
- description of animal (eg age etc)
- presenting signs
- history
- specimens submitted
- changes identified
- diagnosis

Such data would be stored on a computerised database, and include a reference number enabling convenient retrieval of the original stored material.

Potential Application

A properly structured tuna health database, consisting of the specimens and a computerised database containing some or all of the above information, would have two main applications. The first is for surveillance and monitoring. Analysis of the diagnosis, date and location of collection of the samples would provide an indication of the distribution of different diseases in the population, and may provide clues as to the seasonal or growth-cycle occurrence of particular diseases. Base-line data from the database would provide a base-line reference to assess whether any particular disease event should be considered normal, or whether it represents an unusual and potentially serious problem.

If used as a surveillance tool, a specimen / laboratory based database suffers from a major limitation. The information contained does not refer to the diseases present in the population, but the diseases present in the specimens submitted. If the reporting / submission rate for disease problems is high, the database may provide a relatively accurate picture of the overall population. In many cases, however, the reporting rate is low, and subject to many biases, limiting the inferences that can be made from the database to the general farmed population.

The second main application for a properly structured database is as a pathology reference collection, to support future research and diagnostic work. Taking history and signs into account, it would be possible to develop an indication of the range of normal findings (for haematology / biochemistry, and histopathology), as well as reference specimens showing the types of pathology that can occur related to certain diseases. This would have great potential to support future research and diagnostic work.

Neither of these applications of the collection is feasible, due to the lack of background information about the specimens.

One potential use of the collection is to provide examples of particular types of pathological changes, for reference or teaching purposes, but even this would require manual matching of the pathology reports to slides, and the real benefit of this must be questioned. Another is to provide a historical record of the state of

health of the tuna at the time of collection, which may be of assistance in determining the date of arrival of future pathogens. For example, stored fixed blocks can be examined with PCR to identify previously unrecognised pathogens. One subset of the collection which may be able to provide some useful data, in the absence of supporting data, is the biochemical analysis of blood specimens. At the individual specimen level, interpretation is very difficult. However, at the population level, it may be possible to plot distributions of particular values. These distributions are likely to be made up of mixed populations of normal and abnormal animals. Mixture population modelling techniques enable computerised modelling of multiple underlying distributions in a mixed distribution, and may reveal the normal and abnormal ranges for future reference. In order to undertake this research, the data owners (TBOASA) would need to request an electronic copy of the data from IDEXX-VPS in a format suitable for analysis.

Recommendations

Current specimen collection

It is recommended that the current specimen collection continue to be stored, in conjunction with the pathology reports. Although its potential value is limited, long term storage will not be expensive, and it will be available for future research projects in which manual linking of the reports to the specimens may be warranted. It would be of great benefit to the industry, researchers and for future training if information currently available only as internal reports or corporate memory was published. This would increase its availability and ensured that the corporate knowledge is not lost with individuals leaving the industry.

Future disease specimen collection

It is recommended that an industry driven health database be developed to ensure that the maximum benefit can be obtained from specimens collected and analysed in the future. This database would record all the necessary information referred to previously, to enable the retrieval, interpretation and analysis of information and specimens.

Analysis of haematological and biochemical parameters

A summary analysis of the haematological and biochemical parameters has been undertaken by Kirsten Rough. If further detailed analysis beyond this is required, it is recommended that TBOASA request an electronic copy of all biochemical and haematological measurements stored in their database.

Research Needs

Significant gaps in our knowledge of tuna health were identified during the literature review and risk assessment. This means that some of the risk estimates do not have enough information input and the likelihood and consequence of the risks may change as the information becomes available. In particular, there is very little known about normal cardiorespiratory physiology of tuna, effects of nutrition on health, potential effects of viral pathogens, tuna parasitology, effects of toxic substances and harmful algal blooms.

During the tuna health workshop on 4 November 2002 research priorities were discussed in detail. The most important research areas from the point of view of the tuna industry were tuna physiology and effect of nutrition on health, while nonindustry stakeholders saw effect of nutrition on health and tuna parasitology as the two main research areas. Nonindustry stakeholders further divided nutrition research into the effects of manufactured diets on health and the effects of vitamin E on tuna health. These two areas ranked almost equal (Table 4 contains pooled results). Additionally, parasitology research needs were further divided into life cycles of parasites, diagnostic tests and others. Here, other parasitology research ranked first with 42%, parasite life cycles ranked second (35%) and diagnostic tests ranked third (23%). This may reflect the participants' lack of background as development of a diagnostic test would be needed to determine life cycles.

Only nonindustry stakeholders identified additional research priorities including:

- effects of environment on tuna health
- risks related to interannual variability and climate
- husbandry stress management
- availability of treatments

Table 4. Research priorities as determined by vote at the tuna health risk workshop on 4 November 2002. Each person attending the conference had an equal vote, which could be either divided between the priorities or used to support one priority. The results are presented separately for industry and nonindustry workshop participants. Nonindustry stakeholders included researchers and government employees.

Research priorities	Industry Rank (%)	Nonindustry Rank (%)	Average rank (%)
Physiology	1 (35.00)	4 (13.33)	2 (24.2)
Nutrition	2 (22.50)	1 (35.83)	1 (29.2)
Virology	3 (16.25)	3 (16.67)	4 (16.5)
Toxicology	4 (13.75)	5 (8.33)	5 (11.0)
Parasitology	5 (12.50)	2 (25.83)	3 (19.2)

On the basis of the literature review, risk assessment and stakeholder's feedback we determined that the research priorities are:

Normal cardiorespiratory and stress physiology of southern bluefin tuna

Currently, there is no information available for cardiorespiratory physiology and very limited information on stress physiology. While mortality of tuna held in pontoons is minimal, suggesting best husbandry practices, information on tuna physiology may help to reduce early mortalities, which account for most losses in the industry. It will also provide information crucial to understand tuna's tolerance to hypoxia and risks posed by such threats as harmful algal blooms and other diseases that affect respiration. Knowledge of pathophysiology of gill and cardiac diseases is essential to fully understand and minimise the impact of these diseases on fish.

Tuna parasitology

As tuna currently captured for aquaculture are in general 3 years old, some individuals carry parasites. The parasites identified in the risk assessment as being of low risk are blood fluke and gill fluke. The significance of these and other parasites may increase if the fish are held in captivity for significantly longer periods of time. Management and control of these conditions requires knowledge of parasites life cycles and host-parasite interactions. Parasite reservoirs should be identified so that they can be controlled. Existing diagnostic tests should be optimised and standardised and new tests should be developed. Close collaboration should be maintained with researchers developing diagnostic tests for the same species or genera of parasites.

Tuna virology

Currently there are no tuna cell lines available and as a result our knowledge of tuna viral pathogens is very limited. FRDC project 2001/200 (also part of Aquafin CRC Health Program) is addressing this issue. Once the cell lines become available, a survey of tuna viral pathogens should be undertaken to investigate the presence of viruses in tuna. We have very little information on susceptibility to viral diseases (although it appears to be low in older fish, judging by the low mortality record in the tuna industry). If any viral pathogens are significant, their reservoirs (for example mussels, wild fish, biofouling) should be determined to minimise future risks. Diagnostic methods for detection of significant viral pathogens should be developed. Susceptibility of tuna to viral diseases should be further investigated.

Toxicity of algal toxins and other compounds

It has been suggested that tuna may be susceptible to microalgae. While the industry experience suggests low susceptibility to algal species and densities normally encountered in the tuna farming areas, it is important to determine potential effects of harmful algal blooms on tuna aquaculture. The fish are currently seasonally exposed to reasonably high level of microalgae, with some evidence of compromise but no mortalities. More detailed information could reduce this compromise and help to reduce the risk due to unusual blooms of harmful algae.

While effects of nutrition on tuna health were ranked high by both industry and nonindustry stakeholders, we believe that current tuna nutrition research covers

effects of nutrition (both manufactured and traditional diets) on tuna performance and product quality, including effects on the health of fish. If there is interest in exploring health effects in more detail, this should be done in collaboration with nutrition research, not as a stand alone project.

In parallel to a research program, a health training program should be developed for the tuna industry. In particular, disease emergency exercises and training in recognition of health problems on farms should be a priority. The industry should have Emergency Health Response protocols. The farm workers should be able to recognise major parasites and other disease signs. Training would result in capacity building and would be important for the SBT industry human resources.

There is also a significant potential for collaboration between environment and health areas, both for training and research purpose. This potential should be fully explored and utilised to increase use of resources and avoid overlap.

Benefits

Southern Bluefin Tuna aquaculture industry (TBOASA) is the ultimate beneficiary of the health risk assessment. The economic value of this project is difficult to quantify, but the extensive tuna mortalities in April-May 1996 highlight the potential impact and cost of major mortalities. In 1996, 70% of the tuna held by the industry died within a few weeks, this equating to a 1999/2000 market value of \$141 million. Such a loss would result in serious economic and job loss to a major regional area of South Australia, with flow on effects elsewhere.

The main beneficiaries of this project are tuna farmers, who can gain improved access to current knowledge (included in this report). The SBT industry may benefit economically from this report as well as from implementation of the recommendations. The SBT industry managers and researchers as well as development fund providers have improved up-to date knowledge of tuna health and therefore capacity to invest/allocate the tuna industry's research and development funds.

Further Development

Tuna health surveillance

There is a need for an on-going, well designed and properly documented tuna health surveillance and monitoring program. The suggested components include: targeted surveillance for known exotic disease threats (e.g. RSIV), targeted monitoring of key endemic disease threats (e.g. parasites), environmental monitoring for early warning of disease (for example microalgae and oxygen levels), general monitoring for assessing endemic disease and general surveillance for exotic or emerging disease threats. It is proposed that the program is developed and tested as an externally funded project and then becomes a fully industry-driven continuous program.

Tuna health database

The materials held by TBOASA and IDEXX-VPS are not organised for retrieval or analysis and as a result they are only a collection of pathology specimens with mainly historical value. The well illustrated review on tuna parasites (Rough, 2000) should be published and made available to the SBT industry, researchers and for educational purposes. Other information, currently available only as unpublished reports or not available in a written format, should be published or recorded in confidential industry documents, to ensure the retention of corporate knowledge, even as individuals leave the industry. An industry driven tuna health database should be developed, desirably as a part of tuna health surveillance program.

Further research and training

Future research on tuna health was prioritised and the most important areas included were tuna physiology, tuna parasitology and virology. Toxicity of algae and other agents should be assessed. Additionally, risk assessment should be reviewed as new knowledge becomes available and if farming practices and conditions change.

Training in the area of health, for example on disease emergency and recognition of health problems should be provided to the SBT industry. This training could be structured as informal communications and workshops. While good communication channels already exist in the SBT industry and have been essential for dealing with health issues, a more structured approach could add another dimension. Fish health training should be directed at different levels, including general farm staff, farm researchers, SBT scientists and fish health professionals. Tuna health training should include provision of suitable industry-related projects for Masters and PhD students. Training for external participants, such as fish health professionals and academics is crucial for successful and efficient collaboration with the industry and development of good relationship between scientists or veterinarians and the industry.

Planned outcomes

The planned outcomes of this project were to achieve greater ability to anticipate and guard against fish health related incursions in tuna farms and hatcheries and to gain knowledge of the extent and broader usefulness of the existing TBOASA fish health database. The project's outputs include this report, together with the bibliography and summary of relevant literature, report assessing qualitative health risks, report outlining a proposed farmed SBT health surveillance project, as well as other communications. Oral presentations at industry workshops and scientific conferences and individual meetings with tuna industry increased the industry's understanding of tuna health risks and their awareness of potential health problems and their likelihood. This report provides all the relevant and updated scientific and practical information on southern bluefin tuna health. Additionally, this report reviews existing TBOASA database and provides recommendations for tuna health surveillance and database set-up.

Conclusion

Objective 1

To provide a qualitative fish health risk assessment for the tuna aquaculture industry in Australia:

- a qualitative fish health risk assessment based on single risk evaluation matrix was provided for the SBT aquaculture in Australia
- the overall disease risk ranged from negligible to low for hazards considered
- a change in farming practices or environmental conditions could increase risks
- these increased risks can be lowered by control and mitigation, particularly as information becomes available

Objective 2

To review tuna health information from the industry (included their database), research organisations and scientific literature:

- tuna health information was reviewed, including TBOASA database, scientific literature and information from research organisation
- the material currently available at TBOASA was classified as a pathology specimens collection
- there is a general lack of published scientific information on SBT health and physiology

Objective 3

To identify areas of highest risk and propose management control measures for the industry, as well as research priorities:

- the areas of highest risk were identified
- risks which ranked highest (low) included: gill fluke, blood fluke and hypoxia
- tuna health surveillance and monitoring program was proposed
- research priorities were proposed and included SBT physiology (in particular cardiorespiratory and stress physiology), SBT parasitology (in particular blood fluke and parasite life cycles and reservoirs), SBT virology (in particular viral pathogens carried by SBT) and toxicity of algae and other compounds to SBT

Objective 4

To disseminate the results of this SBT health risk assessment project:

- the results of this SBT health risk assessment have been disseminated at industry meetings, scientific conferences and in the form of this report

Summary

In summary, this project has shown that:

1. SBT as a species appears to be relatively resistant to infectious diseases.
2. Current best farming practices should guarantee negligible to low disease risks.
3. Any changes to farming practices such as extended grow-out time, culture of other species in a close proximity or propagation could increase risks.
4. A well designed and executed tuna health surveillance and monitoring program will further lower the risks and provide early warning system.
5. There is a general lack of information on tuna health and SBT physiology.
6. Increased knowledge of parasitic diseases and exotic diseases would greatly benefit the industry in the future.
7. There is a need for training in the area of fish health.
8. There is a need to preserve and document corporate knowledge of tuna health.
9. Preventions of disease spread should be achieved through best farming practices and appropriate regulations.
10. Risk assessment should be reviewed as new knowledge becomes available and if farming practices and conditions change.

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Appendix 1

INTELLECTUAL PROPERTY

The intellectual property and valuable information arising from this report are:

1. Copyright in this report

Appendix 2

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Appendix 3

EVOLUTION OF SBT FARMING

(Including Health Parameters)

provided by Brian Jeffriess TBOASA

Brief History of Industry

Globally, modern tuna farming began with a joint Japan/Australia experiment in Port Lincoln in 1991. Up until that time:

- (1) SBT had never been farmed.
- (2) NBT (*Thunnus thynnus*) had been farmed:
 - In Japan, by wild capture of 200g. fish and grow-out to about 8kg with high mortality in pontoons in bays.
 - In the Mediterranean, by trapping post-spawning large fish (eg. 150kg.) in bays and then short-term grow-out (eg. 3 months).

In 1991 and 1992, the capture/transport of SBT in the Port Lincoln trials was catching by pole into the boat deck, and then transport to Port Lincoln in the bait tanks of the boat (OFCF, 1994). The issues were:

- (1) Because of the boat tank size, catching was restricted to small fish (eg. 12kg), and to less than 100 fish/trip.
- (2) In the initial trials (ie. 1991), mortalities were very high at 40-70% (OFCF, 1994).
- (3) By mid-1992, some boats were consistently catching 80-100 fish/trip, with very low mortalities in transport of around 10% (OFCF, 1994). However, the length of the trip (eg. 80 hours), the limited number of boats, and the stress on the fish - all restricted the fish quality and grow-out numbers. For example, the maximum tonnage which could have been harvested under this system was below 500 tonnes.

Modern capture/transport technology was developed from 1993. This technique is circling the fish with a purse seine net, transferring them through underwater net panels to a towing net, and then towing them to Port Lincoln at one knot or less (ie. 10-20 days). The fish are then transferred to the farming pontoon through underwater net panels. The fish are never handled.

The number/tonnes of fish into farms has increased to around 99% of the current Australian SBT catch quota. The data is:

(Dec/Nov)	Tonnes Into Farms (tonnes)	Total Australian Quota (tonnes)
1990/91	17	5,265
1992/93	722	5,265
1994/95	908	5,265
1996/97	2,498	5,265
1998/99	4,991	5,265
2001/02	5,185	5,265

Source: AFMA, 2002

Health Implications

The Port Lincoln technology results in:

- (1) The capacity to catch larger fish. Operators are now able to target any size fish present in the fishing grounds. They choose to target around 17-20kg (3 years old) for marketing reasons. This average size (compared with 10kg under the old systems), means the captured SBT have survived 3 years of wild health challenges with an assumed natural mortality of 20-30% p.a. (CCSBT 2002). This is a significant (default) health management practice (Rough, pers. com.).
- (2) The system minimises the stress of capture, transport and storage. The technology has been considerably refined over the years, (eg. slower towing, better pontoon bracing, slower transfers, smaller purse seine shots, larger towing pontoons, reduced numbers in the tow pontoon, better understand of the GAB/Gulf temperature effect). It is now unusual to see a significant number of fish damaged from the purse seine shot and/or the tow.

Feeding Systems

The evolution has been:

- (1) In 1991 to 1994 most feed (frozen pilchards/mackerel) was imported from Japan, with a small SA pilchard catch quota.
- (2) From 1995 to 1998 the dominant feed was Californian pilchards, supplemented by a low level (ie. up to 5,000 tonnes) of local pilchards.
- (3) From 1999 to 2003 the industry has gradually moved to a more diversified feed structure.
 - Californian pilchards have moved from about 50% of total feed to around 20% in 2003.
 - European herring, fed January/May to achieve maximum growth, have increased to around 20% in 2003.
 - The SA pilchard quota has increased to over 21,000 tonnes in 2003, supplemented by an expected Tasmania red bait supply of up to 10,000 tonnes.

- High quality European pilchards (Ellis, 2002) have gained to around 15% of total feeds.

(4) From 1997 an increasing proportion of feeding has been via frozen blocks thawing in a small cage in the pontoon before being made available to the tuna. About 75-80% of feed is now by this method.

There is now a much greater understanding of feed quality, and the benefits of feed mix.

Health Implications

The improvements from feed quality and mix have been directly reflected in improved fish health. The fish health proxies are lower mortalities and higher prices (see both below).

Grow-out/Harvesting Structure

The SBT are mainly captured January/February with small catches also in December and March. Grow-out is 3-6 months, depending on the marketing strategies of individual companies.

In 2001/02, the trend to super low temperature (SLT) frozen product - harvested mid-June to mid-August - has continued.

	Tonnes	
	Fresh	Frozen
1998	4,510	1,746
1999	2,300	4,687
2000	2,767	5,065
2001	2,543	5,679
2002	2,500	6,050

Source: 1998-2001 (Japan import data); 2002 (TBOASA estimate)

Frozen farm SBT prices/volumes are negotiated around February each year. They are fixed prices, guaranteed by the Japanese buyer, at ex-pontoon. The buyers are able/prepared to do this because of the consistency of product quality in the pontoons and across the companies (ie. the guaranteed price is the same for all farmers).

To show the increase in prices, the following is the tonnage/price of Australian SBT into Japan.

	Tonnes	Yen/kg (landed Japan, ie. c&f)
1998	6,256	1,974
1999	6,987	2,342
2000	7,832	2,173
2001	8,223	2,383
2002	8,550	2,450

Source: 1998-2001 (Japan Import Data); 2002 (TBOASA estimate).

These increases are despite frozen product being lower c&f value than fresh, and frozen product increasing its share of the harvest from 20% in 1998 to 75% in 2002.

Health Implications

The health risk parameters in this trend are:

- (1) The shift to frozen product means fish are being held longer to get the best fatness/colour mix in mid-winter, ie. the risks are theoretically greater from predators, parasites, net problems, environmental impacts, disease (eg. VHSV transfer is a "colder water" problem).
- (2) The SBT are, in theory, more exposed to uronema which is traditionally a cold water problem.
- (3) The market confidence in the quality and consistency of the product has improved and reflected in health proxies (eg. higher price, lower mortalities).

Mortalities

Unaggregated industry data indicates declining mortalities since 1997. This is due to:

- (1) Improved catching/towing/transfer techniques which have reduced mortalities during those processes. More important the reduction in stress has substantially cut the so-called 10-day and 20-day and 60-day mortalities resulting from breakdown of immune systems.
- (2) Better understanding of mixing feeds.
- (3) No/reduced feeding during predictably stressful events, eg. dodge tides, algae.
- (4) Generally better husbandry.
- (5) Virtual elimination of predator mortalities and stress from predators.

Official mortality data from AFMA is now available since the start of the CCSBT Trade Information Scheme (TIS) in 1999. This requires AFMA to reconcile fish transferred into farms/mortalities/harvesting for all farms. These are then aggregated into industry data for the CCSBT.

The AFMA data - example attached - is:

Dec/Nov	Mortality Rate Through Farming (%)
1999/00 ⁽²⁾	6.89
2000/01 ⁽²⁾	4.83
2001/02 ⁽¹⁾	2.20

⁽¹⁾ Interim data from returns to AFMA, as aggregated by TBOASA.

⁽²⁾ AFMA 2001, and AFMA 2002.

It is doubtful whether the 2001/02 level can be significantly improved. The reasons are:

- (1) The 2002 summer was the coolest ever recorded in SA.
- (2) Towing weather conditions are outside industry's control.
- (3) In the future, grow-out periods are likely to be longer, with greater risks.

However, husbandry, etc. improvements are more likely to be reflected in greater weights gains.

Improvements in Prospect

Improvements likely to occur in the next 3 years include:

- (1) Continuous husbandry improvements.
- (2) Less fouling on nets, resulting in:
 - Better water flow
 - Less parasite reservoirs
 - Less risk of bottom net drag
 - Possibility of bigger pontoons/lower stocking rates.
- (3) Continuous reduction in stocking rates as an industry practice. The average in a 40m. diameter pontoon has already fallen from average 3,000 fish in 1997 to below 1,800 fish in 2002 with some stocking around 1,400 fish (Source: TBOASA).
- (4) Deeper and more oceanic sites from 2003 - providing scope for deeper nets, lower stocking rates, and better water flows.
- (5) The local pilchard catch quota is increasingly structured to provide fresh pilchards to the farms.
- (6) Real-time bait analysis is improving bait quality. Bait diversity is also improving with increased use of red bait and European pelagics.
- (7) Mixed bait (pellet) feeds provide a possible option to offset any problems with wet feeds.

Appendix 4

Tuna health risk analysis and literature review

Based on information from tuna industry and scientific literature review and additional information provided by AusVet Animal Health Services.

Risk analysis performed by the authors of this report.

Infectious Agents

Viral diseases

The only reported viral disease involving *Thunnus* spp is red sea bream iridoviral infection (Kawakami and Nakajima 2002). Red sea bream is very similar to our snapper species. Also, a number of viral diseases of other pelagic fish could possibly involve tuna and will be considered here. The diseases to be considered are:

- Vacuolating encephalopathy and retinopathy
- Red sea-bream iridovirus infection
- Yellowtail ascites and viral deformity

Vacuolating encephalopathy and retinopathy (VER), viral nervous necrosis (VNN) or encephalomyelitis (after Munday et al. 2002)

Agent name and taxonomy

The viruses are classified as genus Betanodavirus within the family Nodaviridae. Official virus species names are *barfin flounder nervous necrosis virus* (BFNNV), *Dicentrarchus labrax encephalitis virus* (DIEV), *Japanese flounder nervous necrosis virus* (JFNNV), *Lates calcarifer encephalitis virus* (LcEV), *redspotted grouper nervous necrosis virus* (RGNNV), *striped jack nervous necrosis virus* (SJNNV), *tiger puffer nervous necrosis virus* (TPNNV) and the tentative species names are Atlantic halibut nodavirus (AHNV), Malabar grouper nervous necrosis virus (MGNNV).

Geographic and host range

Betanodavirus infections have been reported from most parts of the world, including Australia, but with the notable exception of Africa. Clinical disease has been reported in 32 species from 16 families. However, it is notable that larval/juvenile northern bluefin tuna (*Thunnus orientalis*) held in waters where VER occurs in other species have not been affected (Y Sawada pers. com.). As the various betanodavirus strains show low species specificity this observation suggests that *Thunnus* spp. may not be susceptible.

Morbidity and mortality rates

For larval and juvenile fish, mortality rates are usually in the range of 50-100%.

Transmission

Initial transmission is known to be vertical for striped jack and European sea bass and is probable for most other species. Lateral transmission from infected to non-infected fish occurs readily.

Agent stability

Resistant to pH 2-9 and stable in seawater at 15°C for more than a year.

Tissue tropism

In larval and juvenile fish, betanodaviruses have particular tropism for tissues of the central nervous system and retina. However, subclinically-infected adult fish have virus present in a range of tissues including the ovary which is usually invaded shortly before spawning.

Brief description of major pathological and biological effects

Pathology and clinical effects are only seen in fish which have significant involvement of the central nervous system and retina. At the light microscope level this is characterised by extensive vacuolation of the nervous tissues and/or retina, sometimes accompanied by necrosis. This is most often seen in larval or juvenile fish, but older fish, especially when under heat stress, may show such lesions. The clinical signs are very much related to the pathology, ie the fish may be blind (retinal damage) and/or show swimming abnormalities such as spiralling, and looping swimming patterns. Hyperinflation of the swim bladder is seen in some species. Mass mortality is commonly seen in larval infections but older fish may show a less dramatic mortality pattern.

Key diagnostic features and overview of diagnostic methods

In endemically-infected populations the clinical signs are sufficiently characteristic to enable a presumptive diagnosis to be made, especially if supported by histological evidence. However, definitive diagnosis depends on isolating or identifying the virus, detecting the viral genome and/or detecting specific anti-betanodavirus antibodies. Although various combinations and permutations of these techniques have been described, the method now favoured for routine diagnosis is an appropriate nested PCR, possibly combined with culture and/or an ELISA for specific antibodies (Munday *et al.* 2002).

*Disease management procedures (Munday *et al.* 2002)*

At the hatchery level control of betanodavirus infections relies on selection of uninfected broodstock and/or decontamination of eggs. Although the nested PCR to detect carrier fish is a very sensitive the technique has to be optimised for each virus strain and this has yet to be achieved. In particular, this technique may fail if applied to ovarian biopsies prior to transfer of infection to the ovary. In those circumstances testing of broods for antibodies against betanodaviruses should also be undertaken. Eggs may be disinfected using ozone, but again, the methodology needs to be tailored to the specific circumstances.

In the past, certification testing of larvae and juvenile fish has relied on histological examinations which are not sufficiently sensitive. Use of a nested PCR and/or culture in SSN-1 or E-11 clone of this cell line is suggested as better methods. Grotmol *et al.* (2000) reported that the RT-PCR was able to detect 100 to 1000 copies of *in vitro* transcribed RNA. Dalla Valle *et al.* (2000) found that a two-step nested PCR was 100 times more sensitive than the standard RT-PCR and a one-step nested PCR increased sensitivity tenfold. Thus, for the nested PCR the best result is that it can detect one copy of transcribed RNA and the worst result is that it only able to detect 100 copies. Putting these sensitivity limits in the context of infectivity it is reasonable to consider the sensitivity of the nested PCR as 100% for certification purposes. As Arimoto *et al.* (1993) have shown that SJNNV does not spread by cohabitation when the prevalence of infected fish is 1% or less it is suggested that certification testing should be at an intensity sufficient to detect a 1% prevalence, ie 300 fish to provide 95% confidence of detecting at least one

infected fish (Simon & Schill 1984). Obviously, such a large sample would require pooling of fish to keep the procedure within economical bounds. In deciding on a pooling strategy it must be remembered that, "The confidence intervals bounding estimates were generally smaller when larger numbers of groups were used and samples had few fish per pool" (Williams & Moffit 2001).

Risk of exposure of larval tuna to betanodavirus

Barramundi nodavirus (LcEV) is widespread in Australia and exposure of artificially-raised SBT larvae/juveniles would be possible. As barramundi are raised at higher water temperatures, the barramundi nodavirus is acclimated to higher temperatures such as the body temperature of southern bluefin tuna and, therefore, the latter is not likely to be a limiting factor.

Susceptibility of SBT to betanodaviruses

The Japanese experience where NBT are raised in proximity to a number of species with endemic VER without clinical disease occurring suggests that *Thunnus* spp. may not be susceptible. Ideally, this should be tested by experimental infection trials (probably in Japan).

Survey of SBT for evidence of nodaviral infection

It would be appropriate to test SBT for evidence of current infection. If found, this would at least forewarn of potential problems with VER if a hatchery were to be established.

Hypothesised pathways of infection

Hypothesised pathways for occurrence of VER in farmed tuna are shown in Figure 1.

Probability of entry, establishment and spread

Considering the presence of at least one species of this virus in Australian waters, the probability of release is assumed to be high. Release could be through the introduction of other farmed species (e.g. barramundi) or from infection of wild tuna at spawning sites in the Indian Ocean. If VER was released in Australian tuna fishery waters, exposure of farmed fish will depend on the source (farmed or wild fish) and the proximity to farmed tuna. Therefore, the probability of entry, establishment and spread is assessed as moderate.

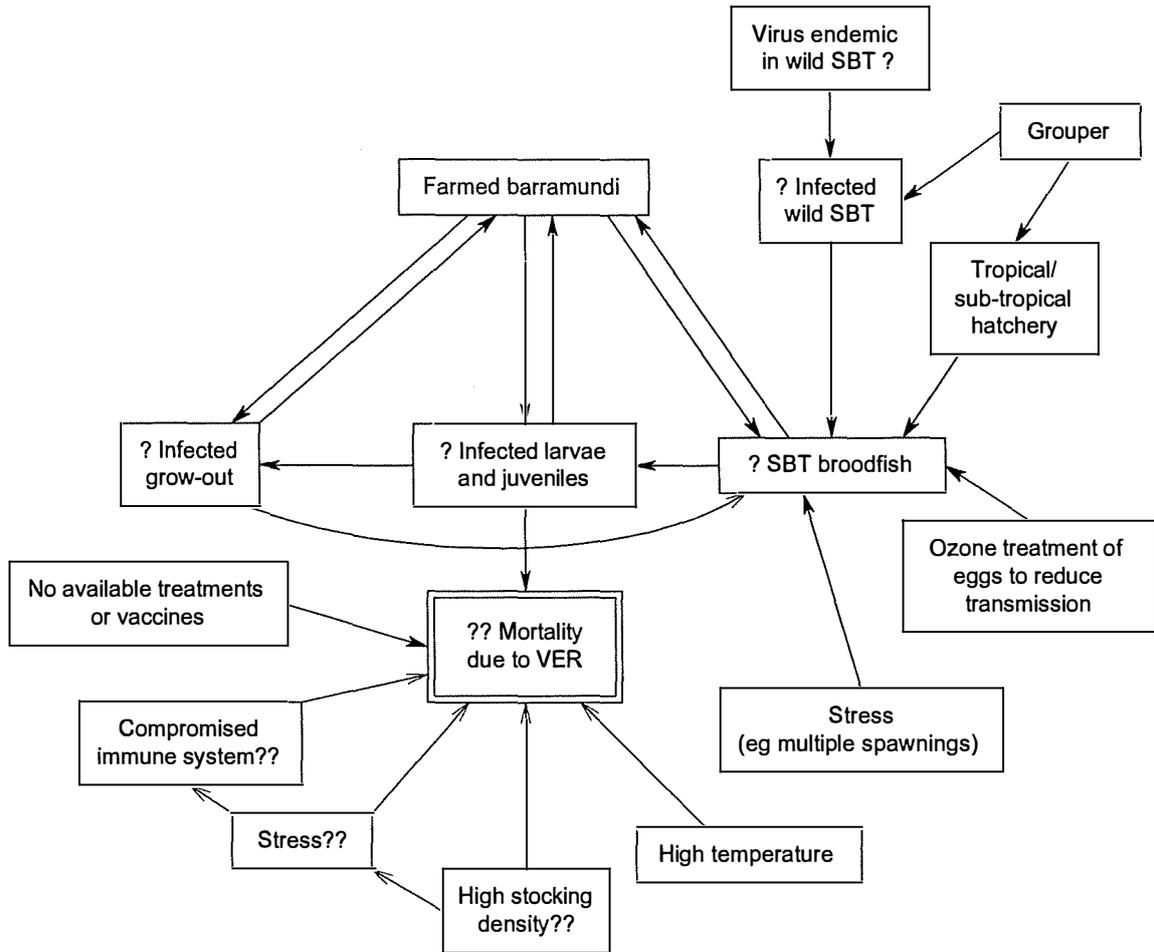
Consequence of entry, establishment and spread

Based on Japanese experience, SBT are probably relatively resistant to VER infection. Even if they are not resistant, the main clinical effects of VER are usually on larvae and juvenile fish, which are currently not farmed in Australia. Therefore, the consequences of exposure have been assessed as very low.

However, in the event of a tuna breeding and grow-out facility being considered, it would be essential to establish the level of resistance of juvenile SBT to VER with more certainty, and this assessment may need to be revised after consideration of any new data.

Further research

Further research is required to clarify the susceptibility of SBT to VER viruses. This could possibly be undertaken in Japan.



Red sea-bream iridoviral infection

Agent name and taxonomy

Red sea-bream iridovirus (RSIV). This discussion also includes closely related iridoviruses which cause disease in a number of other pelagic fish. In contradistinction to epizootic haematopoietic necrosis virus (EHNV) these viruses are not members of the genus *Ranavirus* (Hyatt et al. 2000).

Geographic and host range

These iridoviral infections have been reported from Japan and throughout the Asia-Pacific region but not from Australia. The host range is quite wide, encompassing at least 24 fish species including yellowtail and groupers (Nakajima et al. 1999), with at least one report of disease in *Thunnus thynnus* (Kawakami and Nakajima 2002). Note that the Pacific population of northern bluefin tuna is often referred to as albacore in Japan (as is the case in the paper by Kawakami and Nakajima 2002). Also, this has now been reclassified as *Thunnus orientalis*.

Morbidity and mortality rates

Losses have been reported mainly in fish during warmer months of the year. Nakajima et al. (1999) characterise the disease outbreaks as "mass mortalities". Inouye et al. (1992) reported losses of 20-60% in red sea-bream.

Transmission

There are no reports of vertical transmission of RSIV and the presence of high titres of virus in many tissues suggests that lateral transmission through the water column is the usual means of spread.

Agent stability

The virus is sensitive to pH 3.0.

Tissue tropism

These viruses have tropism for the spleen, liver, kidney and gills (Oshima et al. 1998).

Brief description of major pathological and biological effects

Diseased fish are lethargic and have severe anaemia, petechiation of the gills and splenic hypertrophy. Typically, enlarged, basophilic cells are present in the spleen, heart, kidney, liver and gills. From their locations and morphology it is likely that these cells are leucocytes (Inoue et al. 1992)

Key diagnostic features and overview of diagnostic methods

The histopathological picture of enlarged cells in the spleen, liver, kidney and gills which stain strongly with Giemsa is very characteristic of RSIV infection. Further confirmation can be obtained by demonstrating the iridovirus virions by electronmicroscopy, culture of the virus in RTG-2, CHSE-214, FHM, BF-2 or KRE-3 cells at 20-25°C, detection of specific antigen by immunofluorescence using monoclonal antibodies and detection of specific genomic sequences by PCR (Inouye et al. 1992, Nakajima et al. 1998, Oshima et al. 1998).

Disease management procedures

Apart from normal hygienic precautions there are no current specific control procedures being used in endemic areas. Recently, Nakajima et al. (1999) reported promising results from vaccination trials.

Comments

RSIV and related viruses are exotic to Australia and, if introduced, could severely compromise a number of emerging aquaculture industries (e.g., snapper, yellowtail) as well as the tuna industry. The controls on non-viable fish products imposed by AQIS (1999) are appropriate and the main danger would be from illegal importation of whole round fish of susceptible species or live fish.

Hypothesised pathways of infection

Hypothesised pathways for occurrence of RSIV in farmed tuna are shown in Figure 2.

Probability of entry, establishment and spread

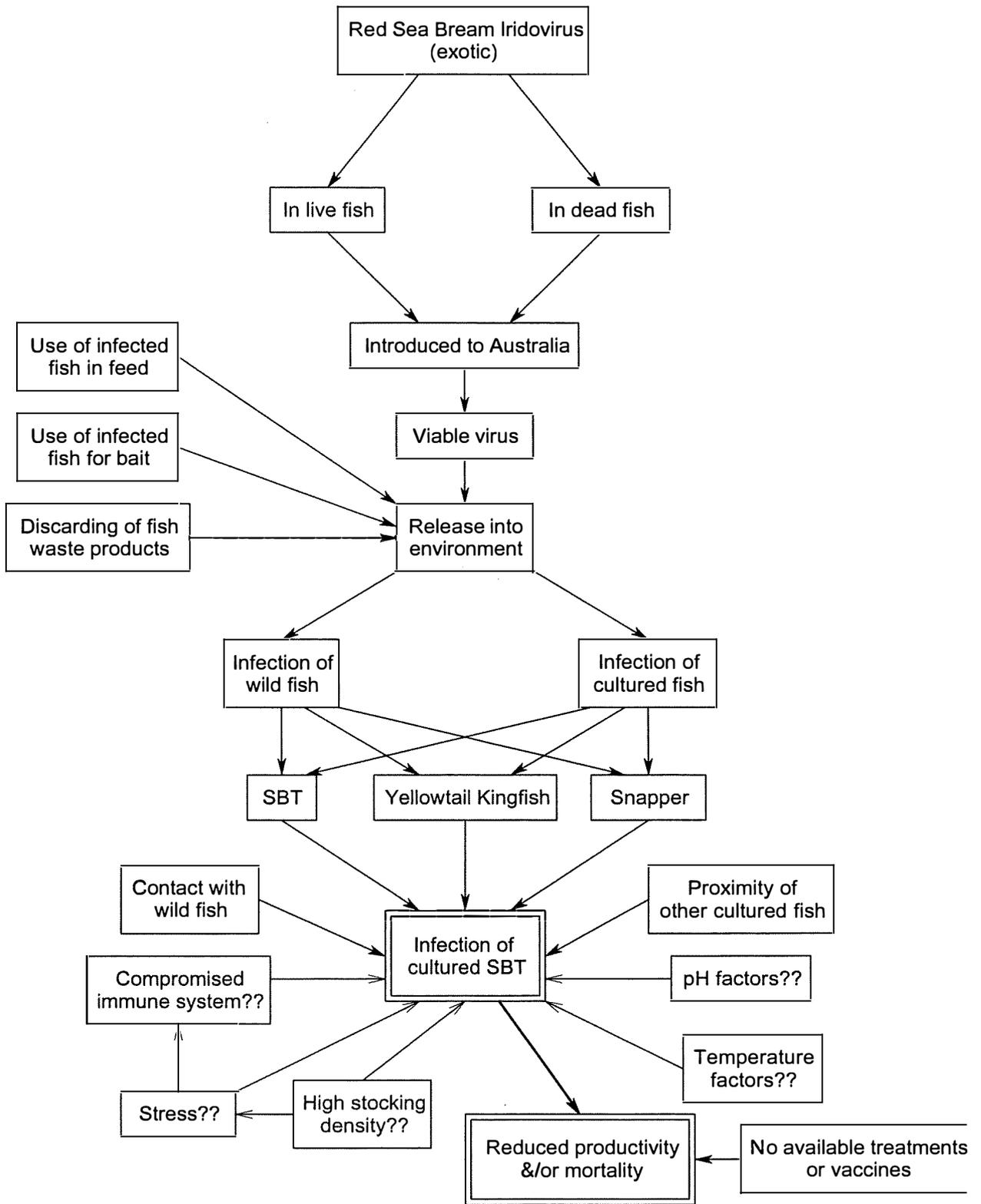
At present the only likely source of the virus would be red sea-bream, yellowtail or other table fish imported from Japan, or SE Asia. Current biosecurity controls are likely to be adequate to protect against introduction of RSIV. The probability of entry, establishment and spread from importation of whole fish for use as baitfish or fish feed would be low (AQIS 2000). However, it is possible that some baitfish, such as Japanese mackerel, could carry the virus. However, Japanese mackerel have not been used as tuna feed since 1996. Also, it is possible that infection could become established in species such as snapper and yellowtail kingfish in the vicinity and ensure the continuity of the disease. Consequently, probability of entry, establishment and spread has been assessed as very low.

Consequence of entry, establishment and spread

On the basis of Pacific bluefin tuna being susceptible to RSIV, SBT are also assumed to be susceptible, and production effects are likely to be substantial. However, Japanese researchers suggest that only fish 0-1 years old are susceptible. In Japan mortalities occur only in 0 class and are temperature dependent, increasing above 25°C (H. Kawakami, pers. com.). Therefore, the consequences of entry, establishment and spread of RSIV have been assessed as low.

Further research

Baitfish need to be tested for susceptibility to RSIV. Local snapper, yellowtail kingfish and other susceptible fish could be surveyed for the presence of RSIV.



Yellowtail ascites and viral deformity (after AQIS 1999)

Agent name and taxonomy

Yellowtail ascites virus (YAV) and viral deformity virus (VDV) are aquabirnaviruses, which belong to a new genogroup.

Geographic and host range

These diseases have not been reported outside Japan and have only caused disease in yellowtail. However, inapparent infections with YAV have been reported in gold-striped amberjack, greater amberjack and three-line grunt in Japan.

Morbidity and mortality rates

In one study wild yellowtail were found to have a 15% carrier rate. Both diseases can cause high mortality rates (up to 90%).

Transmission

Vertical transmission is thought to be important in the spread of YAV because the virus is readily isolated from gonadal products. Horizontal transmission is suspected but not proven. Information is lacking on the mode of transmission of VDV.

Agent stability

In general, aquabirnaviruses are relatively resistant to environmental conditions. YAV and VDV have characteristics similar to infectious pancreatic necrosis virus (IPNV) - they are stable at pH range 3-11 and are stable at 56°C for 30 minutes.

Tissue tropism

YAV occurs in visceral organs (especially kidney) and gonadal tissues whereas VDV has tropism for the brain.

Brief description of major pathological and biological effects

YAV is a disease of juvenile yellowtail and is characterised by swelling of the abdomen due to accumulation of ascitic fluid. There is also catarrhal enteritis, haemorrhages in the liver, stomach and pyloric caeca and pallor of the gills. Histologically, there is necrosis in the liver and spleen and vacuolar degeneration of the renal tubules. VDV infection in yellowtail fingerlings under 10g bodyweight is characterised by marked scoliosis and swimming abnormalities. The brains of these fish show congestion and haemorrhage.

Key diagnostic features and overview of diagnostic methods

Both diseases have characteristic clinical signs but definitive diagnosis depends on culture of the viruses, or the use of genomic probes (PCR). The viruses grow in most common fish cell lines including CHSE-124.

Disease management procedures

No information is available on control procedures.

Comments

Aquabirnaviruses are widespread in the marine environment and YAV and VDV are possibly already present in Australian waters, but would only be likely to be apparent under intense mariculture of such species as yellowtail kingfish.

Hypothesised pathways of infection

Hypothesised pathways for occurrence of aquabirnaviruses in farmed tuna are shown in Figure 3.

Probability of entry, establishment and spread

These viruses may already be in Australian waters or could be introduced. Once the virus is present in Australian waters, exposure of farmed tuna is also likely. The probability of entry, establishment and spread is assessed as high.

Consequence of entry, establishment and spread

SBT are assumed to be resistant to these viruses, based on Japanese experience. Therefore, the consequences of introduction of YAV or VDV have been assessed as very low.

Further research

Maintain contact with appropriate Japanese workers regarding possible cases in Japanese tuna. Also, local yellowtail kingfish could be surveyed for presence of the viruses.

Bacterial diseases

Southern bluefin tuna seem to be remarkably resistant to bacterial diseases. There are anecdotal reports of *Vibrio* spp. infections associated with trauma incurred by being towed in pontoons and Rough et al. (1999) reported opportunistic aeromonad infections in fish with *Caligus elongatus* infestations.

Two bacterial diseases have been reported in northern bluefin tuna, namely pasteurellosis and mycobacteriosis, and these are discussed below.

Pasteurellosis or pseudotuberculosis

Agent name and taxonomy

The disease is caused by *Photobacterium damsella* subsp. *piscicida*, previously ascribed to the Pasteurellaceae.

Geographic and host range

Pasteurellosis is essentially a warm-water (20-25°C) disease affecting a wide range of wild and cultured fish (AQIS 1999) in many countries but not Australia. Peric (2002) reported lesions consistent with this disease in northern bluefin tuna cultured in the Mediterranean where this disease is endemic. Other fish which are affected and which occur in Australia include *Seriola* spp., *Pagrus* spp., *Epinephelus* spp. and *Mugil cephalus*.

Morbidity and mortality rates

Peric (2002) does not provide a morbidity rate for northern bluefin tuna but it is apparently low, without specific mortalities being recorded. Hamaguchi et al. (1992) were able to experimentally produce mortalities in northern bluefin tuna which were challenged with *Photobacterium phosphoreum*. High mortalities (up to 50%) have been reported in other farmed species such as yellowtail and European sea bass.

Transmission

P. damsella subsp. *piscicida* does not appear to survive in seawater for more than 3-5 days and, therefore, transmission is presumed to be lateral between fish through the water column (AQIS 1999).

Agent stability

See above.

Tissue tropism

The organism can be found in the internal viscera, especially the kidney and spleen.

Brief description of major pathological and biological effects

Acute infections are characterised by non-specific septicaemic changes such as congestion and haemorrhage. Chronic infections ("pseudotuberculosis") are characterised by the granulomatous reactions in visceral organs, especially the kidney and the spleen.

Key diagnostic features and overview of diagnostic methods

Although the granulomatous lesions are suggestive of pasteurellosis such changes are seen in other diseases such as mycobacteriosis. The organisms in Gram stain are stout Gram -ve rods which sometimes show bipolar staining with Giemsa stain.

Definitive diagnosis is achieved by culturing the organism followed by biochemical and/ or serological identification. The organism is not fastidious except for a requirement for 1-3% sodium chloride in the medium.

Disease management procedures

In the past antibiotics have been the mainstay to control the disease but the appearance of R-plasmids conferring transferable resistance to all commonly used drugs has restricted their efficacy. While experimental vaccines have shown some promise, control is very much related to careful management, including avoidance of overcrowding (Roberts 2001).

Comments

Without doubt pasteurellosis/pseudotuberculosis would be a major concern to marine aquaculture if it was introduced into Australia. On the limited information available it would not appear to pose a major threat to tuna culture.

Probability of entry, establishment and spread

Because there have been no reports of widespread outbreaks of pasteurellosis in baitfish (Roberts 2001) and, therefore, it is unlikely that large numbers of infected fish would be fed to tuna, the probability of entry is low. However, the bacterium is capable of infecting a wide range of warm-water finfish and this assessment may need to be reviewed at a later date. The probability of establishment and spread was also assessed as low, because not many tuna would be exposed to the probable low dose of bacterium in any infected baitfish. This means that the overall probability of entry, establishment and spread was assessed as low.

Consequence of entry, establishment and spread

Based on the limited information available from the Mediterranean in regard to infection of NBT, the consequences of introduction of pseudotuberculosis have been assessed as low.

Further research

Maintain an overview of the pertinent literature.

Mycobacteriosis or piscine tuberculosis

Agent name and taxonomy

Mycobacteriosis of fish is caused by a range of mycobacteria which are probably inhabitants of the aquatic environment but which are able to infect and spread between fish. Mycobacteria are acid-fast organisms, which characteristically produce chronic infections in infected fish.

Geographic and host range

Mycobacterial infections of fish occur worldwide and occur in a vast array of species. Mycobacteriosis is particularly problematic in aquarium fish. Biavati and Manera (1991) reported a probable case in a northern bluefin tuna.

Morbidity and mortality rates

Inapparent and clinical infections vary widely. For instance, in Tasmania mycobacteriosis is rarely seen in rainbow trout but is a major problem in Western Australia where environmental mycobacteria are often present and rainbow trout are subjected to heat stress. The disease appears to be rare in *Thunnus* spp.

Transmission

It is probable that initial transmission is from the environment, but that fish then act as amplifying agents and spread is from fish to fish through the water column or by cannibalism. Most authors rule out vertical transmission (Austin and Austin 1987).

Agent stability

Mycobacteria are among the most resilient bacteria, living for long periods in the environment and being killed by only the most powerful disinfectants.

Tissue tropism

In mycobacteriosis, almost no tissues are spared although liver, kidney and spleen appear to be predilection sites.

Brief description of major pathological and biological effects

Clinically there are multiple manifestations including cachexia, ascites, colour changes, exophthalmos and ulceration. Internal lesions are characterised by granuloma formation in many organs especially the liver, kidney and spleen.

Key diagnostic features and overview of diagnostic methods

The presence of granulomas in visceral organs is suggestive of mycobacteriosis which must be confirmed by demonstration of acid-fast bacteria using a modified Ziehl-Neelsen stain. The bacteria can be grown on a number of media but this is usually only undertaken to provide cultures for precise identification (ie speciation).

Disease management procedures

It is imperative not to feed infected fish to tuna. As over 150 species of fish have been reported with mycobacterial infections the possibility always exists that baitfish fed to tuna could be infected although there is no evidence that this has been the case to date. Control at the farm level consists of reducing stocking and stresses. Eradication requires slaughter of all fish and cleaning and disinfection of all equipment.

Comments

There is nothing to suggest that mycobacteriosis is likely to be a problem in the present tuna industry unless baitfish species and/or sources are changed without examination of the fish for mycobacteriosis. The situation with propagation is quite different as the water temperatures used and stocking densities are conducive to the disease establishing and spreading. In particular, the use of minced fish to feed 20-120 day old tuna (Miyashita et al. 2000) could be one possible source of mycobacteria.

Hypothesised pathways of infection

Hypothesised pathways for occurrence of mycobacteriosis in farmed tuna are shown in Figure 5.

Probability of entry, establishment and spread

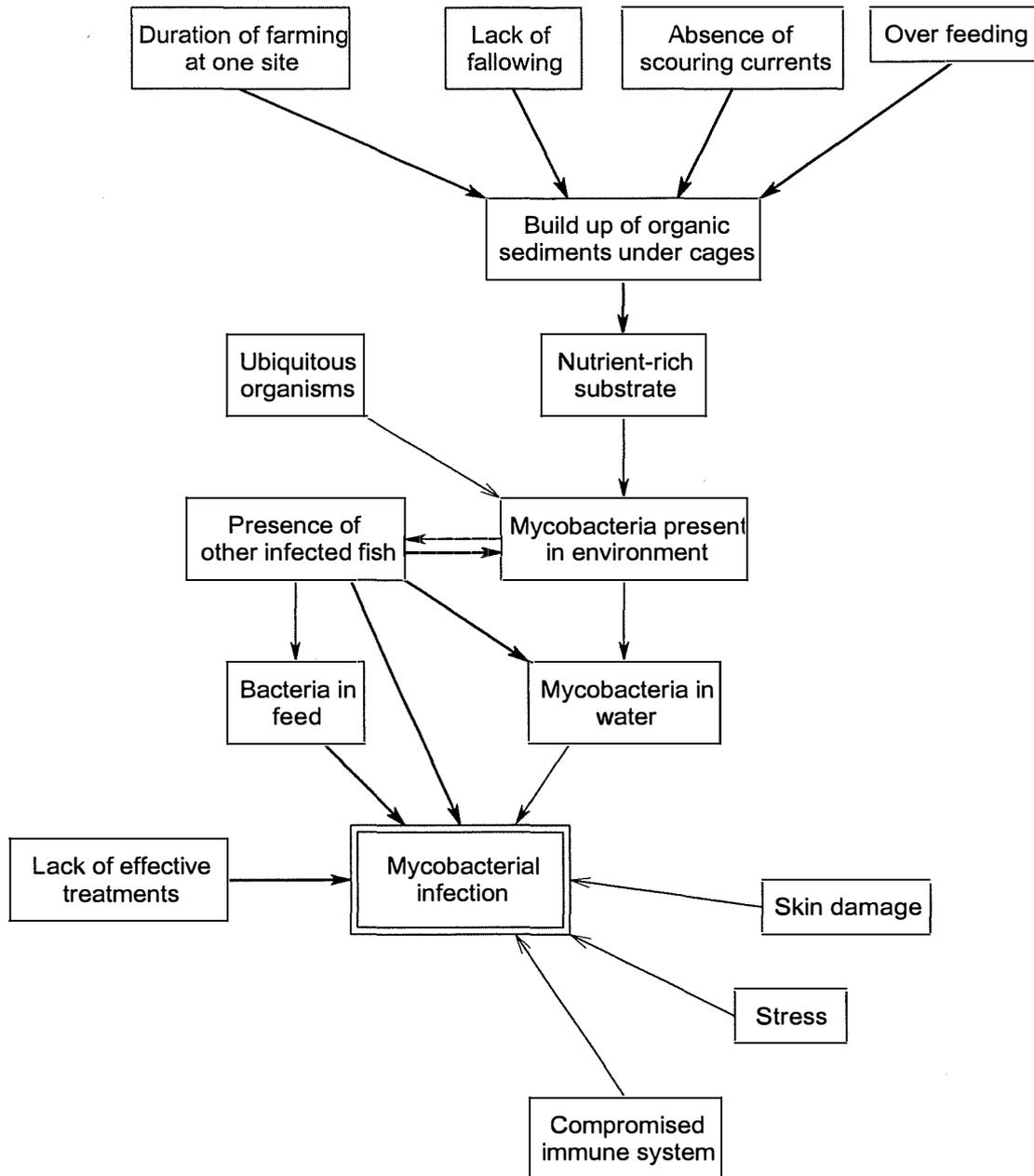
Because mycobacterial infections of fish are already known to occur in Australia, but mainly in the aquarium industry and freshwater aquaculture, the probability of entry has been assessed as moderate. Considering the ubiquitous nature of these organisms, the probability of establishment has been assessed as moderate to high, especially in a hatchery situation. The overall probability of entry, establishment and spread was assessed as moderate.

Consequence of entry, establishment and spread

Mycobacteriosis has not been reported as a significant pathogen of farmed or wild tuna. Therefore, the consequences of introduction of mycobacteriosis have been assessed as negligible.

Further research

None required at present.



Protozoan diseases

As the Myxozoa are now considered to be metazoans they will be considered under that category. As a consequence, very few true protozoans have been reported in *Thunnus* spp. and even some of those apparently are unable to infect southern bluefin tuna.

The coccidian *Goussia auxidis* occurs in the livers of albacore and yellowfin tuna but not southern bluefin tuna (Jones 1990) so it will not be considered in this document.

The only condition which will be considered here is scuticociliate infection.

Scuticociliate infections

Scuticociliate infections occur as two distinct disease syndromes, namely, scuticociliate infection of larval northern bluefin tuna (Y Sawada pers. com.) and swimmer syndrome in cultured southern bluefin tuna (Munday et al. 1997).

Agent name and taxonomy

These parasites belong to the Order Scuticociliatida within the Phylum Ciliophora (the ciliates) and are characterised by the presence of a scutica, a hook-like field of kinetosomes (Lom and Dykova 1992). The parasites infecting larval northern bluefin tuna have not been fully identified but the organism causing the swimmer syndrome has been identified as *Uronema nigricans* (Munday et al. 1997).

Geographic and host range

Scuticociliates have been reported as a cause of disease in many species of larval and juvenile finfish and a smaller range of traumatised and/or compromised adult fish (Munday et al. 1997). The swimmer syndrome has only been reported from Australia (Munday et al. 1997) and then only from southern bluefin tuna except for one case in a yellowtail kingfish (Rough 2000).

Morbidity and mortality rates

Morbidity and mortality rates in larval fish with scuticociliate infections are usually high and frequently reach 100% in both instances. In 1993 mortality due to the swimmer syndrome in cultured southern bluefin tuna in South Australia was estimated to be 5-10% (Munday et al. 1997) but in recent years the prevalence has been lower. Very little is known about morbidity due to *Uronema nigricans* in cultured southern bluefin tuna although a limited study by Munday and Rough (unpublished data) did not suggest that there were many infected fish which did not progress to clinical disease.

Transmission

Scuticociliates are free-living ciliates which are facultative pathogens. Infection is through the water column from, in the first instance, colonies of the ciliates in organic material in culture vessels or under pontoons. The portals of entry in larval and juvenile fish are believed to be integumentary damage, the gills and, possibly, the gastrointestinal tract. In the case of the swimmer syndrome the ciliates colonise the olfactory rosette and then invade up the olfactory nerves to the brain. Once some fish become infected they serve as reservoirs for the parasites – this applies particularly to larval fish. In the case of larval fish, infections have been noted over a very wide range of temperatures (Munday unpublished) in contrast to the

swimmer syndrome which is mainly seen at temperatures below 18°C (Munday et al. 1997).

Agent stability

Very little is known on this point. As far as is known the scuticociliates do not form cysts and therefore are unlikely to survive long out of water. Crosbie and Munday (1999) reported that *U nigricans* would not grow at salinity 3.5 ppt and growth was marginal at temperatures of 10 and 25°C.

Tissue tropism

Scuticociliates have a particular predilection for muscle and nervous tissues but may invade any part of the body – indeed some larvae literally become colonies of scuticociliates (Lom and Dykova 1992). As has been indicated above, in the swimmer syndrome the infection is limited to the olfactory rosette and the central nervous system. Indeed, it is notable that southern bluefin tuna with damaged integuments do not often suffer invasion of their muscles by scuticociliates which is a feature of such damage in many other species (Munday unpublished).

Brief description of major pathological and biological effects

Larval fish are usually found dead or moribund whereas juvenile fish may be noted ill with external ulceration and/or white patches on the skin. Adult fish (apart from tuna) usually show ulceration following damage to the integument. Southern bluefin tuna may die without displaying clinical signs but typical “swimmers” turn blue and swim erratically at the surface (Rough 2000).

Histologically, infected tissues usually show lysis by the numerous ciliates with minimal host response except for the olfactory rosette and olfactory nerves in cultured tuna where a lymphocytic response is usually seen (Munday et al. 1997).

Key diagnostic features and overview of diagnostic methods

Scrapings from affected tissues usually show the typical pyriform scuticociliates. This is normally adequate for diagnosis but can be supplemented by histological demonstration of the organisms in lesions and/or immunofluorescence (Watts et al. 1996).

Disease management procedures

Control of these diseases depends upon good management and hygiene, ie. As far as possible organic material should not be allowed to accumulate in the environment, the fish should not be stressed or traumatised, etc. Therapy is not practicable for treatment or prevention of the swimmer syndrome although Crosbie and Munday (1999) reported that 100-200 ppm formalin, 250-2000 ppm hydrogen peroxide and 1-8 ppm malachite green were lethal for *U nigricans*. Larval northern bluefin tuna have been successfully treated with copper ions (Y Sawada pers. com.).

Comments

It appears that improved husbandry has reduced the prevalence of “swimmers” in cultured southern bluefin tuna. However, scuticiliate infections would be a major risk for a hatchery, if one is established.

Hypothesised pathways of infection

Hypothesised pathways for occurrence of scuticociliates in farmed tuna are shown in Figure 6.

Probability of entry, establishment and spread

Because scuticociliate infection already occurs widely in Australia, the probability of entry is high. However, since scutociliate infection is now effectively controlled in farmed SBT, the overall probability of entry, establishment and spread was assessed as low.

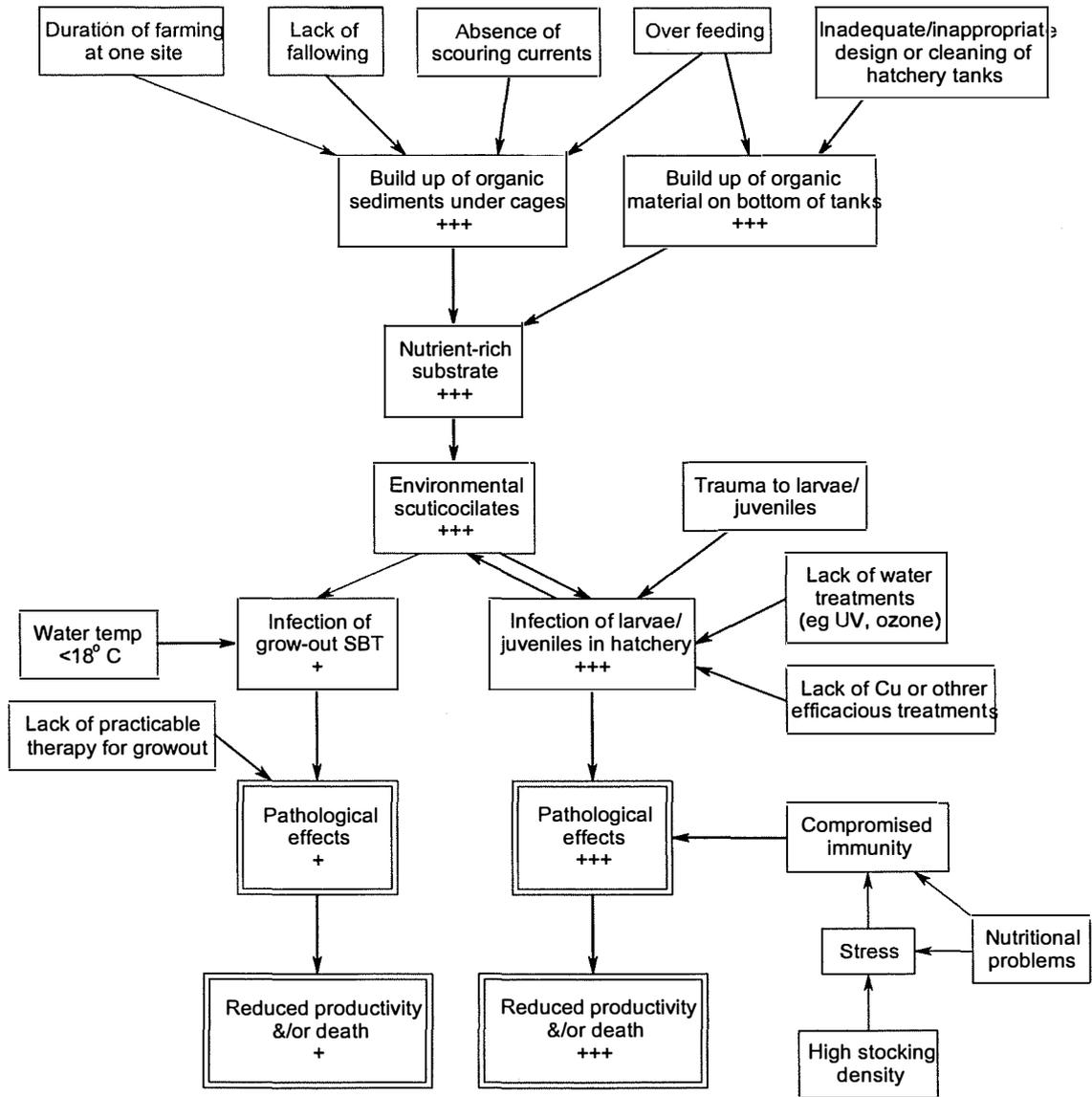
Consequence of entry, establishment and spread

Because management systems have evolved that mainly control scutociliate infection in farmed tuna, the consequences of infection have been assessed as very low.

Because of the very high mortality rates experienced in larval/juvenile fish, the consequences of scutociliate infection for a hatchery facility have been assessed as extreme.

Further research

Not required at present.



Metazoan infections

As the metazoan infections of southern bluefin tuna have been so well-documented (Rough 2000) there is no point in considering parasites of other *Thunnus* spp. which have not been found in southern bluefin tuna. Also, it has been suggested that farmed yellowtail kingfish and snapper may pose a threat to tuna as far as metazoal infections are concerned. However, these species already occur in association with wild and farmed southern bluefin tuna and any parasites capable of transfer will have been reported in Rough's (2000) comprehensive publication. There has been a report of *Benedenia seriolae* in tuna but this requires more verification before being accepted as valid (see footnote).¹ A modified version of Rough's (2000) table of parasites of southern bluefin tuna is attached (Table 1).

Kudoa infection of southern bluefin tuna

Agent name and taxonomy

Langdon 1990 suggested that this parasite was *Kudoa nova* which causes post-mortem liquefaction in the muscles of bigeye tuna (Williams and Bunkley-Williams 1996). As suggested by Rough (2000) *Kudoa* sp. is probably a better designation until the parasite is subjected to genomic identification. It is distinct from *Kudoa thyrssites* which occurs in mahi mahi, pilchards, couta and salmonids in Australia.

Geographic and host range

This parasite has only been described in southern bluefin tuna in Australian waters.

¹ Comments from Dr M Deveney

Regarding the report of *B. seriolae* as a parasite of *Thunnus*. The reports in Kohn and Cohen (1998) and Kohn and Paiva (2000) are based on direct citation of a report by Baeza and Castro (1975), who reported a single specimen of '*Benedenia melleni*' (= *Neobenedenia m.*) from the gills of *Thunnus thynnus orientalis* captured with specimens of *Seriola mazatlana* (= *S. lalandi* although several authors specifically mention the confusion over the status of South American *Seriola* spp.). Both the host and parasite identification are questionable. As you have pointed out, the tuna I.D. is less important. Baeza and Castro did not deposit any specimens in a museum or collection, but Oliva (1986) collected similar material from *S. mazatlana* which Whittington and Horton (1996) identified as *Benedenia seriolae*. Kohn and Cohen's comment that "Whittington and Horton proposed the synonymy of *Benedenia melleni* of Baeza and Castro, 1975 and *Neobenedenia melleni* of Oliva, 1986 with *B. seriolae*" is misleading in the extreme. Whittington and Horton pointed out that these records were misidentifications. The size of the specimens described by Baeza and Castro makes their I.D. as *N. melleni* impossible. It is most likely that they found a specimen of *B. seriolae* which dislodged from a kingfish and either attached to a nearby tuna or which fell off after it died and became lodged in the gills of the tuna. The problems of deriving host-parasite data for capsalids from fish specimens of different species that have been stored together was discussed by Whittington *et al.* 2001. Alternatively (and I don't doubt that this is possible), the specimen from *Thunnus* may have been a capsaline similar to those that we identified from Kirsten Rough's material and which Baeza and Castro misidentified.

Morbidity and mortality rates

Mortality is not associated with this infection. Morbidity rates are not well-documented although fish caught off the NSW coast for canning were reported to have a prevalence of about 1% as determined by the presence of visible cysts (Munday unpublished). Rough (2000) reported that the prevalence decreased with increasing fish size. The prevalence appears to be lower in farmed fish than in wild tuna (about 3%). This may be because bait freezing and storage appears to reduce/eliminate the problem. The Japanese marketing reports to SBT farmers identifies reductions in sale value due to infection with this organism.

Transmission

Nothing is known about transmission although it is presumed to be similar to other myxosporeans and involve an invertebrate alternative host. It is pertinent that Langdon (1990) examined cysts 1-3 mm in size from southern bluefin tuna caught for processing off the Albany, WA coast indicating that at least some fish are infected well before they get to Port Lincoln.

Agent stability

No information is available.

Tissue tropism

Macroscopically, the condition appears as multiple white nodules scattered throughout the muscles. These constitute a blemish that downgrades the carcass. Langdon (1990) provided evidence that suggested that the cysts were actually in peripheral nerves. However, this requires verification as Rough (2000) illustrated what she interpreted as sporulating plasmodia in muscle fibres.

Brief description of major pathological and biological effects

Unlike *Kudoa nova* in bigeye tuna *Kudoa* sp. in southern bluefin tuna does not produce postmortem myoliquefaction. Its only effect is to produce blemishes in the muscle.

Key diagnostic features

The macroscopic lesions of multiple white nodules in the flesh of southern bluefin tuna is quite characteristic of the infection. Confirmation can be made by examining stained or unstained smears from the lesion which will reveal the spores with four polar capsules arranged in a stellate pattern.

Disease management procedures

None available.

Comments

The present prevalence level is tolerable but could increase if propagation is undertaken (cf *Pentacapsula* sp. in striped trumpeter).

Hypothesised pathways of infection

Hypothesised pathways for occurrence of *Kudoa* infection in farmed tuna are shown in Figure 7.

Probability of entry, establishment and spread

Kudoa sp. is a known parasite of SBT in Australia, implying the existence of a local life cycle for this parasite. Thus, there is a reasonable probability of exposure. However, the apparent low incidence indicates that the probability is low.

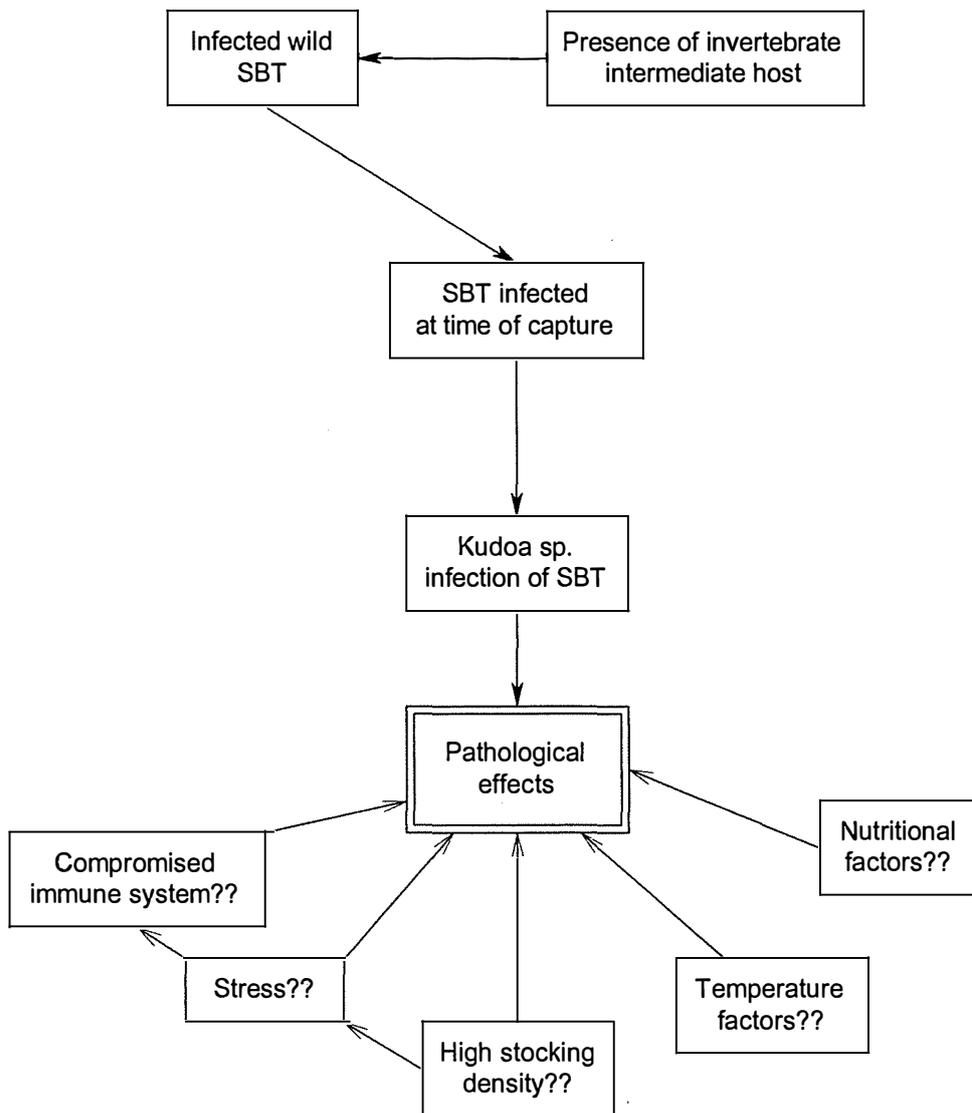
Exposure to *Kudoa* sp. may be higher if propagation and grow-out is undertaken, as has occurred with *Pentacapsula* sp. in striped trumpeter (S Battaglione pers. com.), increasing the probability to moderate.

Consequence of entry, establishment and spread

As this parasite causes some blemishes in muscle tissue of the carcass, the consequences of infection in terms of product quality are significant. Even though the prevalence of lesions is only about 1% the carcasses are usually not fully inspected until they reach Japan and the discovery of an infected fish can lead to quite serious repercussions for the supplier concerned. However, due to the lack of health impact, the health consequences of infection with *Kudoa* must be assessed as very low.

Further research

Further research is required into the epidemiology and control of this parasite, particularly if a propagation and grow-out facility is planned.



Monogeneans

Neohexostoma sp. is common on the gills of farmed and wild tuna but cause minimal damage. *Caballerocotla* sp. occurs on the tongue of wild southern bluefin tuna but is not a problem in farmed tuna (Rough 2000).

Gill fluke infection

Agent name and taxonomy

Unidentified capsaline (M Deveney pers. com.) monogenean (Rough, 2000, type 3).

Geographic and host range

Only reported from wild and farmed southern bluefin tuna in southern Australian waters.

Morbidity and mortality rates

Rough (2000) states "It is more common in farmed fish but its distribution is often confined to only a few pontoons, and heavy infections to individual fish". It is not known to directly cause mortality.

Transmission

Is presumed to be direct, ie not involve an intermediate host.

Agent stability

No information, but not likely to survive for long off host and, especially, out of water.

Tissue tropism

The flukes attach to the gills.

Brief description of major pathological and biological effects

The parasite produces focal lamellar fusion which is seen macroscopically as white patches on the gill surfaces. Rough (2000) states "heavy infections can lead to respiratory stress in tuna".

Key diagnostic features

The presence of white patches on the gills together with the presence of monogenean flukes permits presumptive diagnosis. As no description of the parasite is available a definitive diagnosis is not possible.

Disease management procedures

Therapeutic treatment would not be practicable nor warranted. Control would most likely be achieved by reducing stocking densities for both pontoons and fish in pontoons.

Comments

It would be useful to identify this parasite so more could be learnt about its biology.

Hypothesised pathways of infection

Hypothesised pathways for occurrence of monogenean gill flukes in farmed tuna are shown in Figure 8.

Probability of entry, establishment and spread

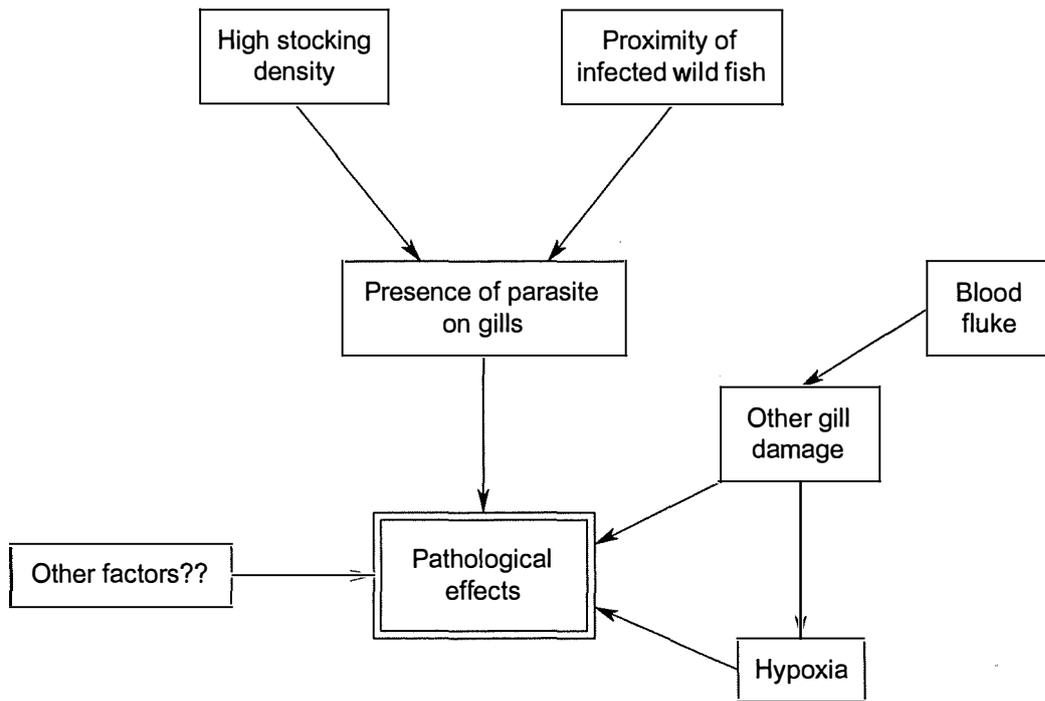
This parasite is already present in SBT in Australia. The probability of exposure has been assessed as high based on Rough's (2000) observations.

Consequence of entry, establishment and spread

Because this parasite usually occurs in heavy infestations in only small numbers of fish, the consequences of entry, establishment and spread was assessed as low.

Further research

Further research is required into the biology and epidemiology of this parasite, as little is currently known about it.



Digeneans

Digeneans are flukes that have both definitive and intermediate hosts, except for didymozoids which may be able to complete their life cycle in one host (Lester 1980).

Digeneans of southern bluefin tuna causing no or minimal disease, apart from minor blemishes, include *Atalostropion sardae*, *Cetiotrema crassum*, *Colocyntotrema* sp., *Didymocystis thynni*, *Didymocystis wedli*, *Didymocystis* sp., *Hirudinella ventricosa*, *Koellikera bipartia*, *Koellikera* sp., *Syncoelium filiferum*, and five unidentified digeneans (Rough 2000). One digenean that produces significant pathology is *Cardicola forsteri* (Cribb et al. 2000).

Blood fluke infection of southern bluefin tuna

Agent name and taxonomy

Cardicola forsteri (Digenea Sanguinicolidae).

Geographic and host range

Only records are for southern bluefin tuna in southern Australian waters (Rough 2000, parasite incorrectly named as *Cardicola smithii*).

Morbidity and mortality rates

Colquitt et al. (2001) found no evidence of *C. forsteri* infection in wild southern bluefin tuna, but Rough (2000) stated that the parasite was “found in wild and farmed tuna but more commonly in farmed fish”. Colquitt (1999) found no evidence of infection in newly captured fish but three months later 66% had eggs embedded in the myocardium and another month later 100% had eggs in the myocardium and 50% had eggs impacted in the afferent filamentary arteries. Rough (pers. com.) reported that it is common for harvest fish to have up to 35% of individual batches with typical lesions when harvested between May and August. In some instances increased mortalities have been associated with severe *C. forsteri* infections (Munday unpublished) but it was not clear whether the sanguinocolid was solely responsible. Furthermore, farming practices and conditions improved since this observation was made. As the post 60 day mortality is very low in farmed tuna and blood fluke tends to peak later (Rough 2000), this parasite in itself must be a small contributing factor in mortalities.

Transmission

Logically, the tuna are infected by cercariae released from the intermediate host. It has been suggested that wild tuna show little evidence of infection, and infection increases along with the length of time fish are held in captivity. This may mean that one of two mechanisms may be involved (either singly and/or in combination):

- The few recently-captured, infected fish which arrive in Port Lincoln produce miracidia which infect the intermediate hosts (eg gastropods, bivalves, polychaetes) which then release cercariae to infect other tuna and so on to produce a “vicious cycle” because of the relative immobility of the fish; or
- The cycle is already established in the vicinity of Port Lincoln in some other definitive host(s) and the tuna become involved because they are restrained in the area.

Interestingly, anecdotal evidence suggests that “fresh” sites are associated with more severe disease than “old” sites. This suggests that the parasite may be pathogenic for both the definitive and intermediate hosts.

Agent stability

No information is available but it would be expected that all stages would be labile outside the hosts.

Tissue tropism

Between one and three small adults occur in the hearts of infected fish where they do not produce any direct pathology as a result of being anchored by tegumental spines. However, the eggs they produce become embedded in the myocardium and the afferent filamentary arteries of the gills where they produce pathology.

Brief description of major pathological and biological effects

The gills of infected fish have multifocal, pale lesions often extending in an arc across the gill arch. These lesions are attributable to the granulomatous reaction resulting from impaction of eggs and developing miracidia in the gill filaments. It is reasonable to presume that these lesions affect the respiratory efficiency of the gills. Probably more important is the effect on the heart where the host reaction leads to myocardial damage and the “back-pressure” from the occluded afferent filamentary arteries leads to ventricular hypertrophy (Colquitt et al. 2001). The significance of these cardiac abnormalities can be appreciated when it is realised that the ability of tuna to withstand reduced ambient oxygen is limited by their relative inability to increase the stroke volume of blood expelled from the heart under hypoxic conditions when bradycardia occurs (Brill 1996). If the capacity of the heart is reduced by the presence of hyperplastic tissue this constraint is further compounded.

Key diagnostic features and overview of diagnostic methods

Definitive diagnosis of *Cardicola forsteri* infection can only be made by collection and identification of adults. However, the presence of typical gill lesions, together with demonstration of eggs/miracidia in gill scrapings (Rough 2000) is adequate for provisional diagnosis. Histological examination of heart and/or gills also permits provisional diagnosis. As infected tuna develop a serological response to *C. forsteri* infection (Colquitt 1999) present/past infection can be confirmed by this means.

Disease management procedures

Until more is known about the life cycle of this parasite it is not possible to make meaningful recommendations on control.

Hypothesised pathways of infection

Hypothesised pathways for occurrence of *C. forsteri* in farmed tuna are shown in Figure 9.

Probability of entry, establishment and spread

Because this infection is a known parasite of SBT in Australia, the probability of entry, establishment and spread was assessed as extreme (eg, 100% prevalence in June 1999 (Colquitt, 1999)).

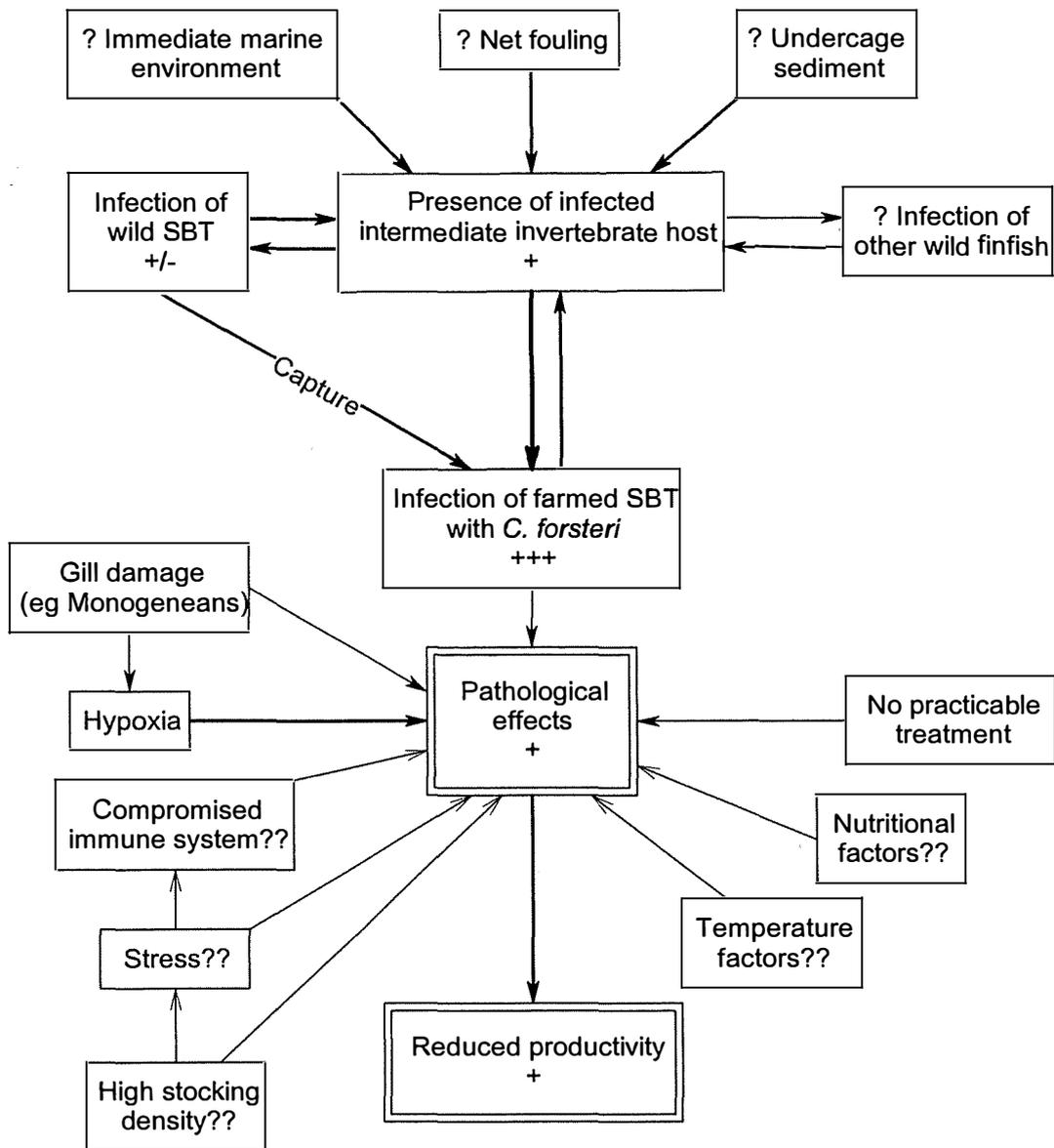
Consequence of entry, establishment and spread

Because this parasite may occur at high prevalence in farmed fish, and may contribute to reduced fish performance, but there is no evidence that it does happen, the consequences of infection have been assessed as low.

Further research

Further research is required into the biology and epidemiology of this parasite. A survey of other fish species in the vicinity of the tuna pontoons for the presence of adult flukes should be undertaken to ascertain which, if any, of these are infected. More information is needed on the prevalence of *C. forsteri* in wild southern bluefin tuna. Based on Colquitt's (1999) studies an ELISA for specific antibodies should be developed and used to screen wild southern bluefin tuna for evidence of infection.

Identification of the intermediate hosts is essential for the development of control measures. The type of strategies will be different if the intermediate host is present on the mesh of the pontoons rather than the seafloor. It may be necessary to develop a molecular probe to achieve this aim.



***Caligus elongatus* infection**

Aetiology

This condition is caused by infection by the copepod *Caligus elongatus*.

Clinical signs

Rough *et al.* (1999) reported that *Caligus elongatus* grazes on the integument of southern bluefin tuna and may produce grazing trails on the skin and ocular tissues.

Pathology

Lesions due to the above copepods are related to their grazing behaviour.

Epidemiology

Caligus elongatus has multiple hosts so tuna may become infected from a variety of sources. In the case of *Caligus elongatus* infecting captive southern bluefin tuna, capture trauma and high stocking densities are believed to predispose to heavy infections (Rough *et al.* 1999).

Diagnosis

Experienced diagnosticians can make a presumptive diagnosis of this copepod infection based on the morphology of the parasite and the types of lesions induced by their activities. However, definitive diagnosis is only possible by a scientist skilled in identifying the parasite.

Treatment

Although a number of therapeutants are capable of killing copepod parasites it is impracticable to use these agents under current tuna aquaculture conditions. In addition, at the present level of loss of production such treatments would be uneconomical.

Prevention

As Rough *et al.* (1999) have suggested that trauma may predispose to *Caligus elongatus* infections then reduction of damage due to capture, towing and harvesting should simultaneously reduce the level of infestation/damage caused by this copepod. Also, this parasite is carried by other species of fish so it would be appropriate to keep other forms of aquaculture separate from tuna farms.

Hypothesised pathways of infection

Hypothesised pathways for occurrence of *C. elongatus* in farmed tuna are shown in Figure 10.

Probability of entry, establishment and spread

The probability of the parasite being in the vicinity of tuna pontoons and tuna actually being exposed to the parasite has been assessed as moderate.

Consequence of entry, establishment and spread

The consequences of tuna being attacked by *Caligus elongatus* has been assessed as low.

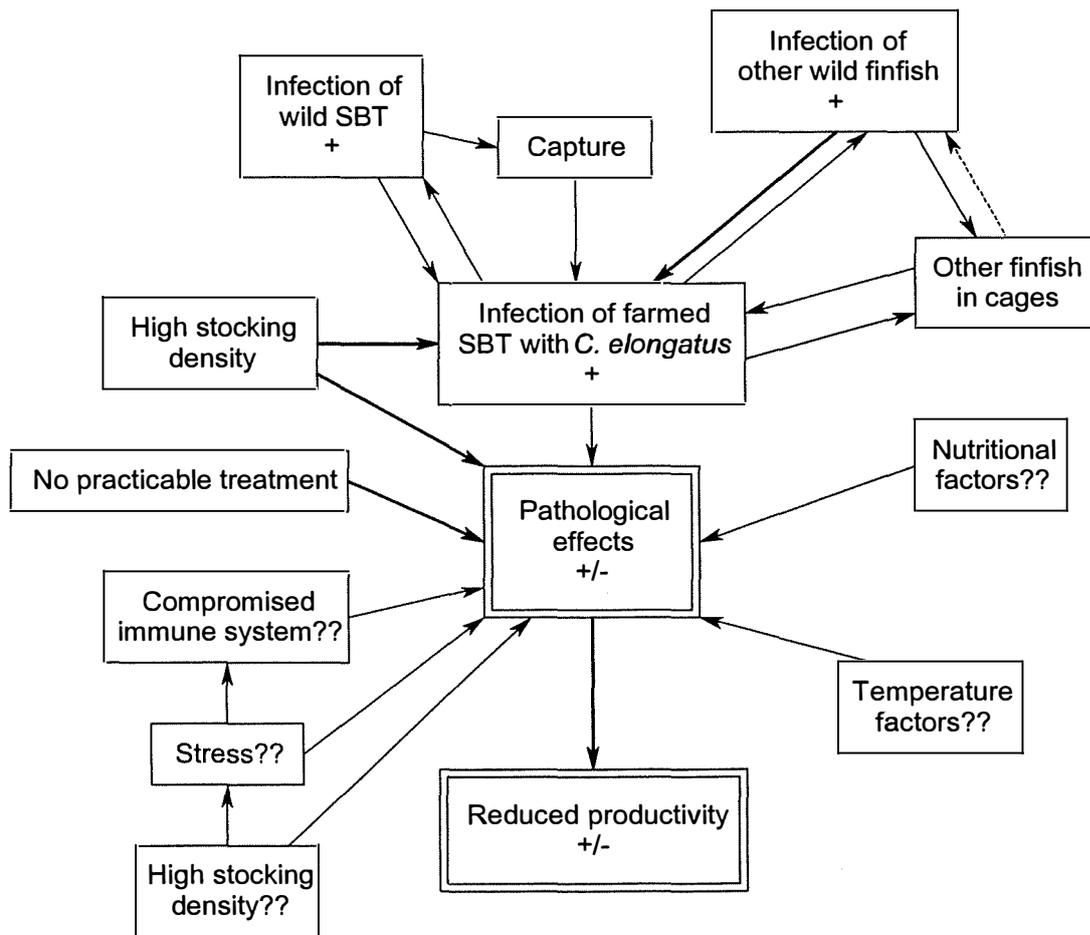


Table 1. Metazoan parasites of southern bluefin tuna *Thunnus maccoyii*

Myxozoa

Kudoa sp. Langdon 1990, Rough 2000

Acanthocephala

Rhadinorhynchus pristis Humphrey 1995

Arthropoda

Amphipod unspecified Rough 2000

Brachiella thynni Rough 2000

Caligus elongatus Rough et al. 1999

Elytrophora brachyptera Hewitt and Hine 1972

Euryphorus brachypterus Rough 2000

Isopod unspecified Rough 2000

Pseudocynus appendiculatus Rough 2000

Aschelminthes

Capsularia marina Humphrey 1995

Contacaecum legendrei Humphrey 1995

Hysterothylacium cornatum Humphrey 1995

Spirurida sp. Rough 2000

Platyhelminthes

Atalostropion sardae Rough 2000

Caballerocotyla sp. Rough 2000

Callitetrarhynchus gracilis Rough 2000

Cardicola forsteri Cribb et al. 2000

Cetiotrema crassum Manter 1970

Colocyntrotrema sp. Rough 2000

Didymocystis sp. Rough 2000

Didymocystis thynni Rough 2000

Didymocystis wedli Rough 2000

Hirudinella ventricosa Olsen 1980

Koellikeria biparita Schmidt and Roberts 1989

Koellikeria sp. Rough 2000

Neohexostoma sp. Rough 2000

Nybelinia lingualis Rough 2000

Otobothrium dipsacum Rough 2000

Pterobothrium heteracanthum Rough 2000

Syncoelium filiferum Rough 2000

Non-infectious Conditions

Many non-infectious diseases of fish fall into the category of “pathophysiology”, ie they are characterised by a significant perturbation of the normal physiology of the fish. For instance, tunas have high oxygen requirements: for example, juvenile northern bluefin tuna have an oxygen consumption four times the value for red sea-bream (Miyashita et al. 1999). Also, unlike other teleosts, tuna cannot reduce activity in response to low dissolved oxygen but must increase their swimming speed and/or gape in order to obtain adequate oxygen from the water, something which cannot be sustained over a long period (Bushnell and Brill 1991). Indeed, in his 1996 review article Brill states, “This inability to tolerate lowered ambient oxygen appears, in turn, to significantly influence tunas’ depth distributions and may limit their vertical movements”. Up to that time studies had been mainly confined to skipjack and yellowfin tuna. However, in 2000 Lowe *et al.* reported that blood from bigeye tuna had a significantly higher oxygen affinity than blood from yellowfin or skipjack tunas or kawakawa. As there are no comparable data for southern bluefin tuna we are left with the quandary as to which species they most closely resemble. This lack of information on the normal physiology of southern bluefin tuna makes it extremely difficult to precisely define such factors as minimum dissolved oxygen requirements and is something that needs to be urgently addressed.

Further research

Determination of the normal cardiorespiratory physiology of southern bluefin tuna is an essential and urgent requirement.

Nutritional problems

There are no reports from Japan concerning confirmed nutritional problems in northern bluefin tuna culture. This is probably related to the high quality of baitfish used to feed the cultured tuna in Japan (B Munday pers. obs.). However, Sawada (pers. com.) has noted opercular abnormalities in juvenile Pacific bluefin tuna which could be a sign of vitamin C deficiency.

Australian tuna farmers have to rely to a large extent on imported baitfish which may be substantially oxidised (Hansen et al. 1977). At present, the relatively short time that southern bluefin tuna are kept in captivity probably ensures that clinical deficiency signs due to depletion of antioxidants has not been reported.

Furthermore, when tuna were held in captivity for prolonged period of time (1991-1996) there was no obvious sign of vitamin deficiency, even though the fish were fed frozen baitfish for 5 years.

Vitamin E depletion

Aetiology

Two causes of vitamin E depletion are possible: absolute deficiency of vitamin E in the diet, and induced vitamin E deficiency. In the case of aquaculture, absolute deficiency of vitamin E is uncommon. The more usual form of vitamin E depletion is produced by feeding a diet high in oxidised lipids which dramatically increases the requirement for biological antioxidants such as vitamin E (Roberts 2001). Such highly oxidised lipids have been demonstrated in oil-rich baitfish both overseas (Hansen et al. 1977) and in Australia (Fitz-Gerald and Bremner 1994).

Clinical signs

No clinical evidence of vitamin E deficiency has been reported for *Thunnus* spp. However, there are numerous examples of these diseases in many species worldwide and almost always associated with feeding oxidised lipids (Roberts 2001). Most commonly the signs relate to loss of muscle function and muscle wastage but sometimes all that is apparent is general lethargy and /or darkening (Roberts 2001). Experimentally, sea bass given a diet low in vitamin E and high in oxidised lipid were found to have lowered immune function (Obach et al. 1993).

Clinical pathology

Fish with myopathy have elevated plasma creatine kinase levels (Messenger et al. 1992). Although rainbow trout on a low vitamin E diet were shown to have low plasma vitamin E levels (Bell et al. 1985) most assays for vitamin E in spontaneous occurrences of vitamin E deficiency have used liver as the preferred tissue. In that context it should be noted that in a report on vitamin E-responsive myopathy in rainbow trout, McLoughlin et al. (1992) found that affected fish had liver vitamin E levels of 40 µg/g and unaffected, supplemented fish had liver levels of 196 µg/g. It is particularly interesting that tuna in captivity for only a month and fed pilchards had a mean liver vitamin E level of 33µg/g, comparable to that of the diseased trout, whereas tuna fed pellets had a mean liver vitamin E level of 490µg/g (Munday unpublished)

Macroscopic pathology

Fish with myopathy show muscle wasting. Fish with lipoid liver syndrome (associated with high levels of oxidised lipids in the feed) have enlarged livers, bronze hearts and anaemia (Roberts 2001).

Histopathology

Roberts (2001) describes the myopathy associated with vitamin E deficiency as bland without inflammation, whereas McLoughlin et al. (1992) reported mineralisation and active regeneration in the vitamin E-responsive myopathy they investigated. This variability is similar to that found in terrestrial animals and complicates diagnosis. The livers of salmonids with lipoid liver disease are heavily infiltrated with fat and may also have infiltrates of macrophages containing ceroid.

Species/ages affected

Many species of fish have been described with vitamin E-responsive diseases. It is notable that younger, rapidly-growing fish seem to be the most susceptible and, in that context, hatchery-reared, juvenile tuna being fed "fish mash" would probably be most at risk.

Management effects

Obviously, feeding diets containing high levels of oxidised lipids will lead to depleted vitamin E reserves. Also, management practices leading to high growth rates tend to exacerbate the problem by increasing the requirement for vitamin E. In terrestrial animals unaccustomed exercise can precipitate clinical disease in animals with incipient lesions.

Diagnosis

Definitive diagnosis can be difficult and usually depends upon demonstration of the typical lesions of myopathy or lipoid liver disease supported by low tissue

vitamin E levels. Presumptive diagnosis can be made on the basis of histological findings and confirmed by a response to vitamin E supplementation.

Treatment

Treatment is by increasing the vitamin E content of the food and, if possible, removing oxidised lipids from the diet. Merely coating oxidised baitfish with gels containing vitamin E is not adequate because the vitamin E will be immediately oxidised (Fitz-Gerald and Bremner 1994).

Prevention

Ideally, prevention is by ensuring that fish diets do not contain oxidised lipids. In addition, in practice it is usual to supplement the diet with vitamin E and other antioxidants.

Comments

Under the present management system for southern bluefin tuna it is unlikely that the fish will show signs of vitamin E deficiency. This situation is likely to change if longer fattening periods are instituted. However, it would be interesting to sample fish sequentially over the period of captivity to ascertain the changes in vitamin E status. If a hatchery is established then close attention will need to be paid to the levels of oxidised lipids and vitamin E in the diet.

Hypothesised pathways of occurrence

Hypothesised pathways for occurrence of Vitamin E deficiency in farmed tuna are shown in Figure 11.

Probability of entry, establishment and spread

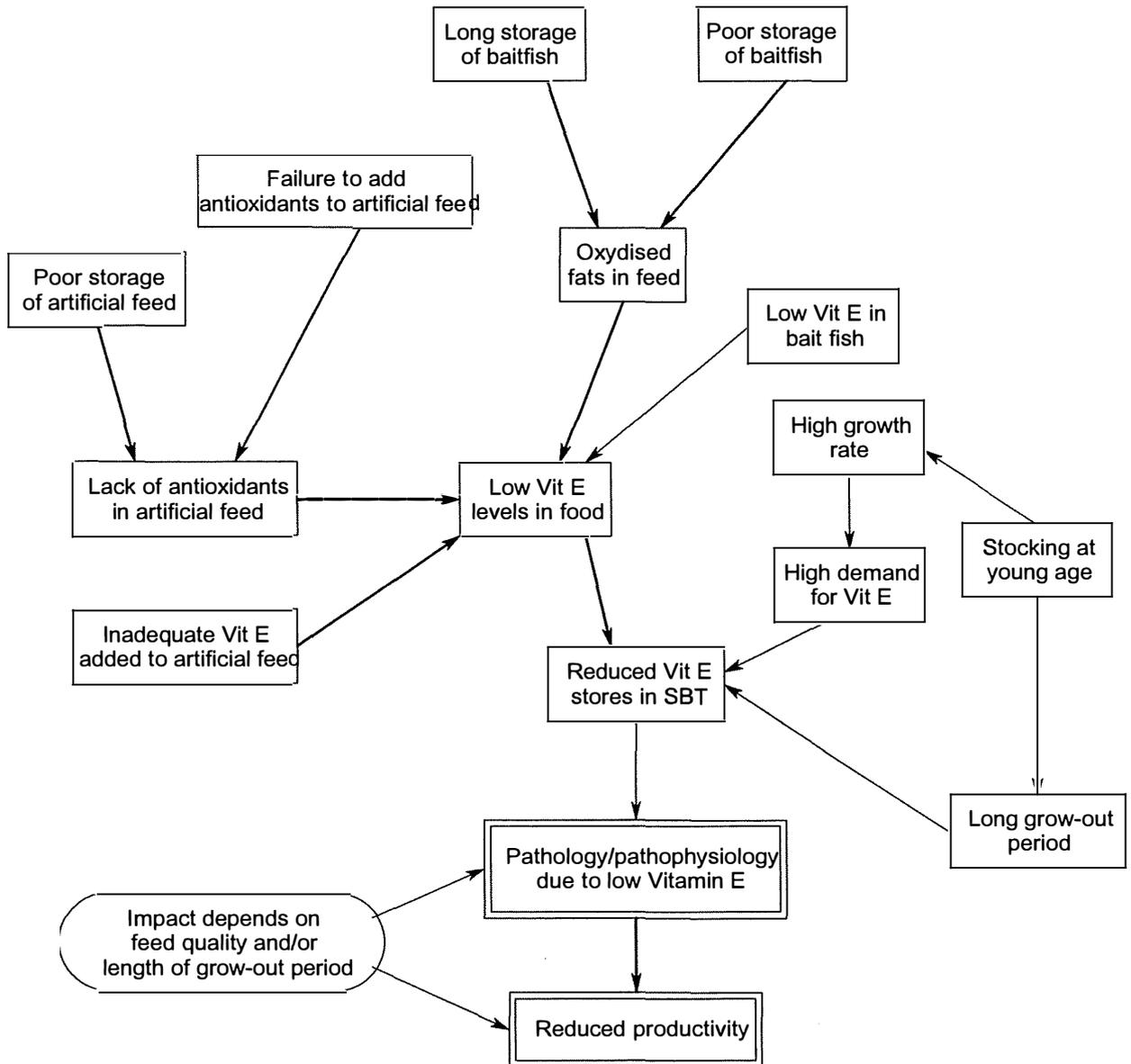
Under the present management system for southern bluefin tuna in Australia, modern tuna farming has developed ways to avoid oxidised bait, so the probability of exposure has been assessed as low. This assessment would need to be reconsidered for hatchery or longer-term grow-out facilities.

Consequence of entry, establishment and spread

Because this condition can be managed through close attention to diet and feed quality especially the use of compounded feedstuffs with adequate vitamin E supplements, the consequences have been assessed as low under current circumstances. The consequence for a hatchery would be moderate.

Further research

Further research is required into the vitamin E status of farmed tuna, and particularly for juvenile tuna if hatchery rearing is proposed. Also, research on more acceptable forms of manufactured diets would be useful.



Vitamin C depletion

Aetiology:

Because vitamin C is very labile as a result of its tendency to oxidise, it is not unusual for levels of this vitamin to be depleted in feedstuffs stored for long periods and/or inappropriately, giving rise to the potential for an absolute deficiency of vitamin C. This problem has been overcome to some extent by the use of stabilised forms of vitamin C which, in some circumstances, may be less biologically-available than the native vitamin. Also, in some fish, inanition coincident with weaning from a natural diet to a manufactured diet can lead to hypovitaminosis C. As with vitamin E, vitamin C is readily depleted as a result of reacting with oxidised lipids in the diet, leading to an induced deficiency of vitamin C.

Clinical signs

Clinical signs are mainly related to the skeletal and other supporting tissues. Frequently there are spinal fractures leading to lordosis or scoliosis (Roberts 2001). Deformities of the gills are frequent as are abnormalities (eg foreshortening) of the opercula. It may be pertinent that opercular deformities occur at a high prevalence in Pacific bluefin tuna larvae/juveniles (Y Sawada pers. com.).

Clinical pathology

It is difficult to perform meaningful analyses of blood and tissues for vitamin C and there are no other reliable clinical pathological assays.

Macroscopic

The presence of skeletal deformities, possibly associated with abnormalities of gill and opercular structure, is highly suggestive of vitamin C deficiency.

Histopathology

The histological lesions in bone have been described as deforming diatheses of cartilage, accompanied by osteoid replacement of bony tissue (Roberts 2001).

Species/ages affected

Hypovitaminosis C usually affects young fish although residual lesions may stay with the fish for life. Many species have been reported to be affected and it would be expected that juvenile tuna raised under hatchery conditions would be at risk.

Management effects

Great care must be taken to preserve adequate levels of vitamin C in the diet of young fish and feeding of "fish mash", as is done with juvenile northern bluefin tuna, is one practice which is likely to reduce the intake of this vitamin at a crucial time.

Diagnosis

A diagnosis of vitamin C deficiency depends upon combining a number of observations to arrive at a presumptive diagnosis. That is, typical lesions coupled with a history suggestive of dietary depletion of the vitamin is adequate for a provisional diagnosis of hypovitaminosis C. Response to treatment (ie cessation of new cases) would be confirmatory of the diagnosis.

Treatment

Supplementation of the diet together with withdrawal of oxidised lipids should encourage the development of normal bone but will not resolve already-established lesions.

Prevention

Prophylaxis depends on ensuring that feedstuffs for juvenile fish are adequately fortified with vitamin C and are not stored for long periods, especially at high temperatures. Highly oxidised feedstuffs should be avoided.

Comments

Between-species differences in susceptibility to vitamin C deficiency have been observed (Munday unpublished) and the fact that this condition has not unequivocally been reported from hatchery-reared Pacific bluefin tuna does not mean that southern bluefin tuna might not suffer from skeletal scurvy.

Hypothesised pathways of occurrence

Hypothesised pathways for occurrence of Vitamin C deficiency in farmed tuna are shown in Figure 12.

Probability of entry, establishment and spread

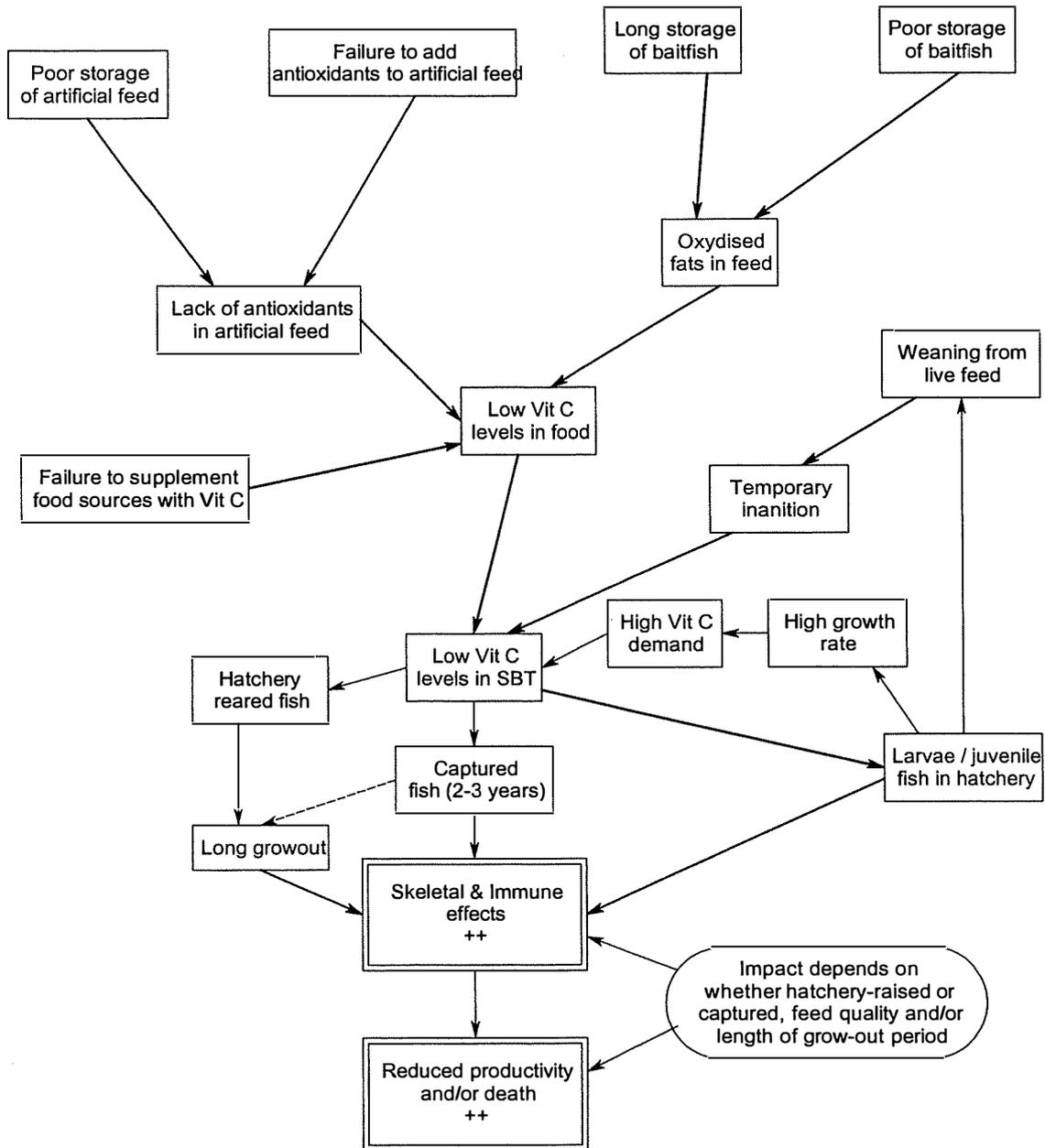
Under the present management system for southern bluefin tuna in Australia, modern tuna farming has developed ways to avoid oxidised bait, so the probability of exposure has been assessed as low. This assessment would need to be reconsidered for hatchery or longer-term grow-out facilities.

Consequence of entry, establishment and spread

Because of the present short period of captivity of culture SBT the consequence has been assessed as low. If this condition occurred in a hatchery or longer-term grow-out facility for SBT in Australia, the consequences have been assessed as moderate.

Further research

Collaboration with Japanese on cause(s) of deformed opercula.
Develop more acceptable manufactured feedstuffs.



Toxicological problems

Microalgal toxicosis:

Aetiology

Exposure to a variety of toxic marine microalgae. A number of these, such as *Chattonella marina* and *Chaetoceros* spp have been reported in the Port Lincoln area.

Clinical signs

Although the toxic mechanisms related to Australasian strains of *Chattonella marina* are not fully known it appears that, as in other parts of the world, both neurotoxins and reactive oxygen species are involved. Neurotoxins lead to abnormal behaviour such as uncoordinated swimming while reactive oxygen radicles are very damaging to the gill epithelium leading to excessive production of mucus and signs of respiratory distress (Ishimatsu et al. 1996). *Chaetoceros* spp. Can produce physical damage to the gills.

Clinical pathology

Ishimatsu et al. (1990) reported a number of clinical pathology changes in fish experimentally exposed to *Chattonella*. However, none of these are suitable for routine diagnostic use. Exposure to, and accumulation of lipophilic toxins such as brevetoxins can be determined by assays of fatty tissues (Munday and Hallegraeff 1998).

Macroscopic pathology

The main pathology which has been reported is copious amounts of mucus flowing from the gills. The mass mortality of southern bluefin tuna at Port Lincoln in 1996 (Clarke 1996) showed similar lesions, but these may have had other causes.

Histopathology

Ishimatsu et al. (1996) and Hishida et al. (1997) described epithelial swelling and lifting together with the presence of excessive mucus in experimental *C. marina* exposure of yellowtail. However, this is not specific for microalgal toxicosis. Very similar lesions were reported by Munday and Hallegraeff (1998) in southern bluefin tuna which died in South Australia in 1996.

Species/ages affected

Many different species of fish of different ages have been reported as being affected by toxic algal blooms worldwide (Hallegraeff et al. 1995). Based on the findings of Hishida et al. (1998) it would be expected that tuna would be exquisitely susceptible to *Chattonella marina* blooms. Those authors exposed yellowtail, red sea-bream and Japanese flounder to 4000 *Chattonella marina* / mL and all yellowtail were dead in a mean time of 83 minutes. Two of six bream died at 165 and 253 minutes and none of the flounder died. The authors explained these observations on the basis of a number of physiological variables, especially ventilation volume which was 1099.6, 486.5 and 384.0 mL/kg/min for yellowtail, red sea-bream and Japanese flounder respectively. Ventilation volume for yellowfin tuna is 3900 mL/kg/min (Bushnell et al. 1990).

Level of exposure

Okaichi et al. (1989) reported that 500 *Chattonella marina* cells/mL are lethal to yellowtail. In view of the above information it is reasonable to suggest that the toxic level for tuna would be much lower.

Management/environmental effects

Obviously, anything which reduces available oxygen to fish exposed to toxic algal blooms will increase the mortality rate. Such factors include high pontoon stocking density, a high degree of net-fouling, poor water exchange rates and excessive under pontoon sediment leading to oxygen depletion in the water column.

Diagnosis

Definitive diagnosis of microalgal toxicosis is often difficult and in the instance of the tunas this is further complicated by the lack of experimental data. Basically, diagnosis is dependent upon the occurrence of typical clinical and pathological findings, together with the presence of potentially toxic levels of microalgae in the water column and elimination of other likely causes of mortality.

Treatment

There are no practical treatments for algal toxicosis in tunas.

Prevention

For practical purposes prevention equates to avoidance, eg by not placing tuna pontoons inside Boston Bay and towing pontoons away from potentially-toxic blooms. There is some evidence that certain clays can be used to flocculate the algae and the use of skirts on pontoons to exclude algae has also been suggested (Rensel, 2000).

Comments

Obviously, much more research is required to clarify the potential for toxic algal blooms to cause mortalities in farmed tuna but the magnitude of losses in other species worldwide emphasises the need for such research.

Hypothesised pathways of occurrence

Hypothesised pathways for occurrence of microalgal toxicity in farmed tuna are shown in Figure 15.

Probability of entry, establishment and spread

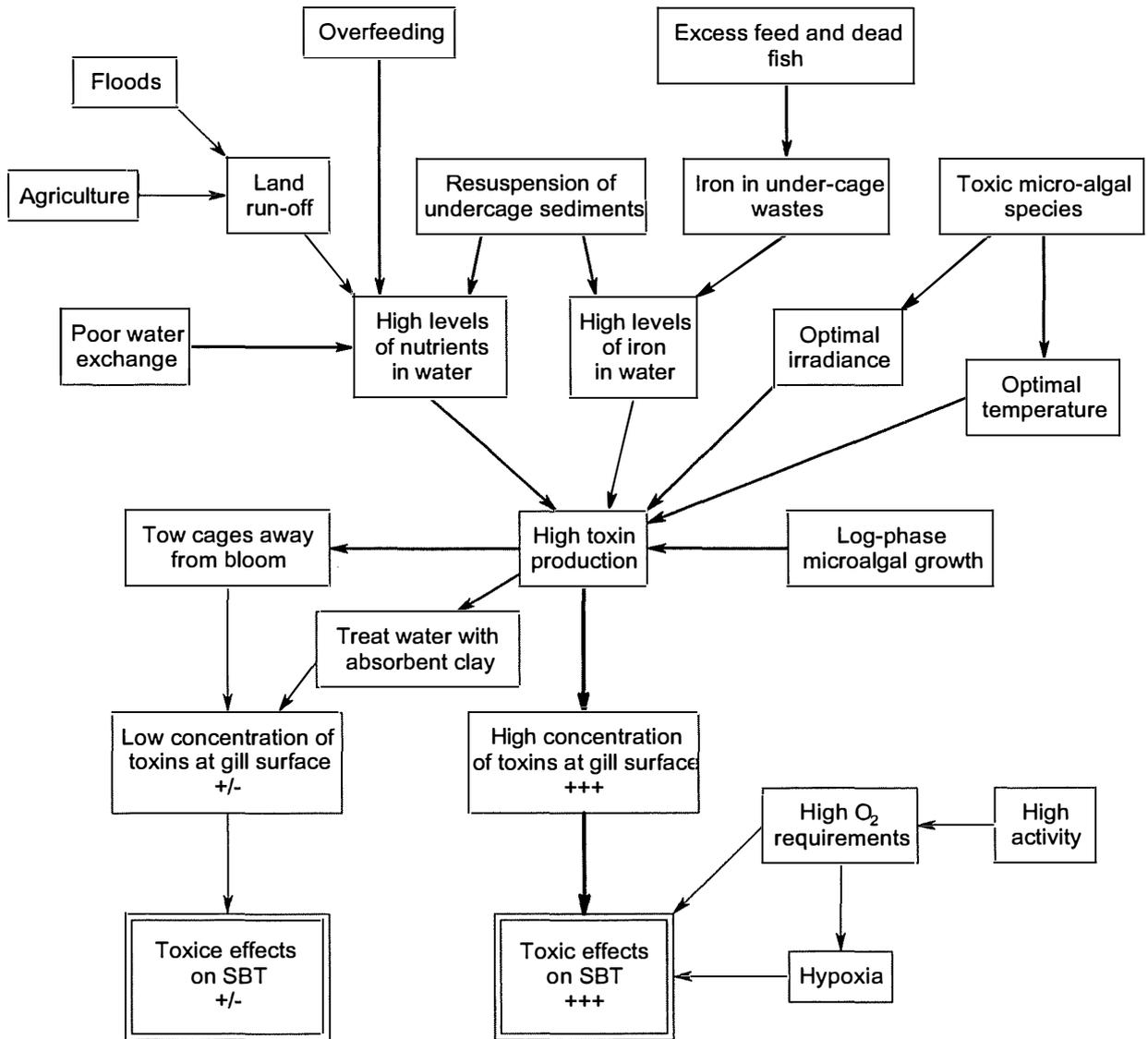
Algal blooms occur in tuna farming areas and some species present have been reported to be toxic. However, the farm nutrient input does not appear to create significant increase in microalgal activity. The probability of entry, establishment and spread is moderate.

Consequence of entry, establishment and spread

Tuna have been consistently exposed to reasonably high level of microalgae, with some evidence of compromise but with no evidence of abnormal mortalities. If a bloom is present, farmers reduce/stop feeding and can tow the pontoons to other sites. The availability of these mitigation strategies and lack of history of algal toxicity to tuna result in the consequence being assessed as low.

Further research

Further research is required into the occurrence and prevention of toxic blooms, as well as their potential effect on farmed tuna.



Therapeutic agents

Aetiology

It is unlikely that in the present circumstances that southern bluefin tuna will be treated with therapeutic agents. Also, other cultured species which are treated will only be treated according to NRA guidelines or "off-label" under veterinary supervision so tuna are unlikely to be exposed to significant levels of therapeutic agents from these sources. Consequently, the only likely problem is when fish in hatcheries are treated for disease. In the latter case a range of antimicrobials may be used. In most instances adverse reactions are likely to be due to inadvertent overdosage.

Clinical signs

A range of clinical signs may occur but the most common are "sudden death" and respiratory impairment.

Pathology

Lesions are variable but most often are in the form of acute gill damage.

Epidemiology

As stated above overdosage is the most frequent cause of morbidity/mortality due to therapeutic agents. This may be due to errors in calculation and/or determination of sizes of tanks, etc. Also, if therapeutants are poorly mixed in the water column or feed "spot" toxicity can occur. In some instances the use of low grade products and/or inappropriately stored products can lead to toxicity.

Diagnosis

Diagnosis can be difficult and relies on a good history, knowledge of the type of toxicity produced by the particular toxicant and elimination of other causes.

Treatment

Treatment will depend upon the type of toxicity but is often not very successful.

Prevention

It is important to test any new therapy with a few fish before subjecting the whole population to the treatment.

Hypothesised pathways of occurrence

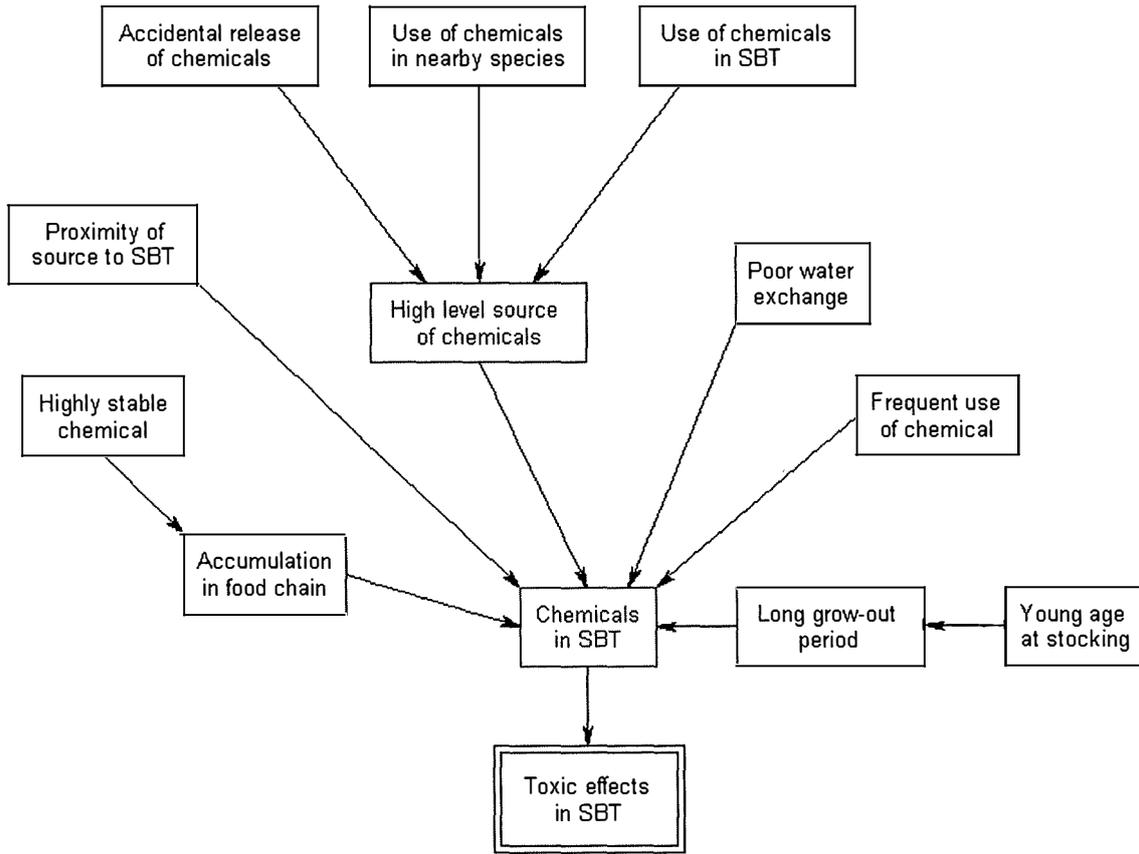
Hypothesised pathways for occurrence of toxicity of therapeutic agents in farmed tuna are shown in Figure 16.

Probability of entry, establishment and spread

Currently no therapeutic agents are used in tuna farming and there is no other aquaculture activity which could result in release of significant volume of therapeutic agent, so the probability is negligible. For hatcheries the probability of release has been estimated as high, however the probability of exposure has been estimated as low.

Consequence assessment

While there is no scientific information on toxicity of therapeutants to tuna, it is assumed that because of their age and size at the time of capture they would not be very sensitive. For hatcheries the consequences of exposure have been assessed as high.



Toxicants (methane and hydrogen sulphide)

Aetiology

These gases are produced from excessive accumulations of organic material under pontoons.

Clinical signs

In modern aquaculture it is unusual for gaseous discharges to reach the level at which they cause clinical signs. However, in association with other causes they may lead to respiratory embarrassment and/or general lethargy and failure to thrive.

Pathology

Lesions are likely to be non-specific although hydrogen sulphide exposure can lead to liver degeneration.

Diagnosis

Diagnosis is more by observation/measurement of excessive gas formation than by tests on the fish themselves.

Treatment

Treatment is inappropriate.

Prevention

Appropriate site selection and good farm management will ensure that gas formation is not a problem.

Hypothesised pathways of occurrence

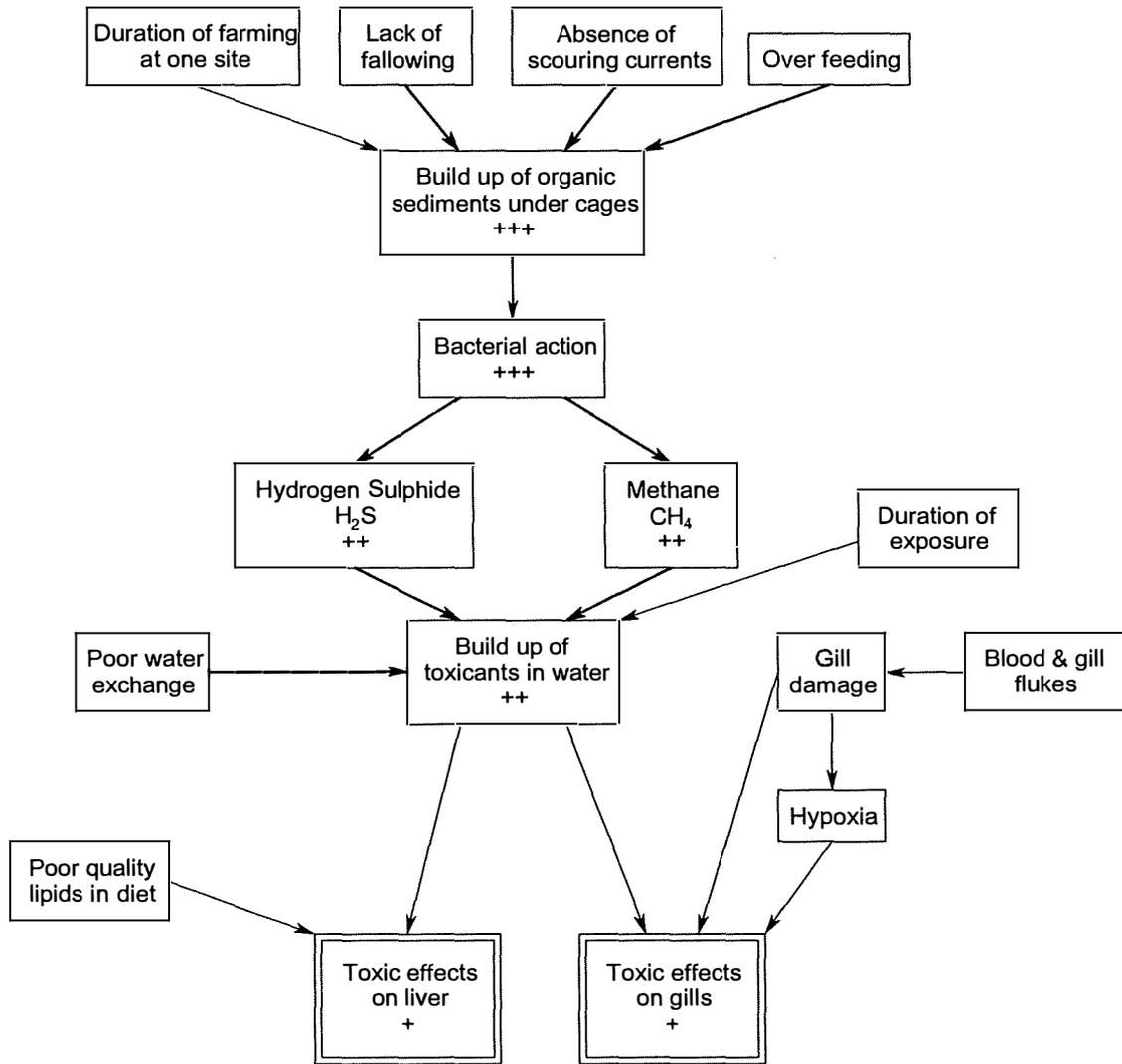
Hypothesised pathways for occurrence of methane and hydrogen sulphide toxicants in farmed tuna are shown in Figure 17.

Probability of entry, establishment and spread

The probability of entry, establishment and spread is negligible as under the current best tuna farming practices the gassing is highly unlikely. This is supported by environmental research (Svane et al 2002).

Consequence of entry, establishment and spread

Except under exceptional circumstances the consequences of gassing are likely to be low (L). Thus, the consequence has been assessed as low.



Trauma

Trauma is an inevitable part of capture, transport, transfer and harvest of southern bluefin tuna and as operators in South Australia have become expert in ameliorating this problem it will not be discussed here.

The one condition which will be considered is collision of cultured, juvenile northern bluefin tuna with the sides of the tank or net wall.

Mortality of Pacific bluefin tuna due to trauma caused by collision during grow-out culture (Miyashita et al. 2000).

Aetiology

Believed to be related to a number of factors such as responses to disturbances such as flashes of light and vibrations in fish with an underdeveloped steering and braking mechanisms in these fish (Miyashita et al. 2000).

Clinical signs

The fish have been observed to respond to stimuli by panicking and colliding with the sides of the tanks or pontoons.

Pathology

There is considerable damage to the vertebral column and parasphenoid bones.

Epidemiology

This condition appears to be entirely related to confinement of a pelagic fish in a limited space.

Diagnosis

This condition can be diagnosed by observation and post-mortem examination.

Treatment

There is no treatment.

Prevention

Miyashita et al. (2000) recommend a 24 hour light schedule which reduces panic incidents.

Comments

As captive southern bluefin tuna appear to be easier to handle than Pacific northern bluefin tuna this problem may not be as severe in the former if propagation is attempted.

Hypothesised pathways of occurrence

Hypothesised pathways for occurrence of collision-trauma in larval/juvenile farmed tuna are shown in Figure 18.

Probability of entry, establishment and spread

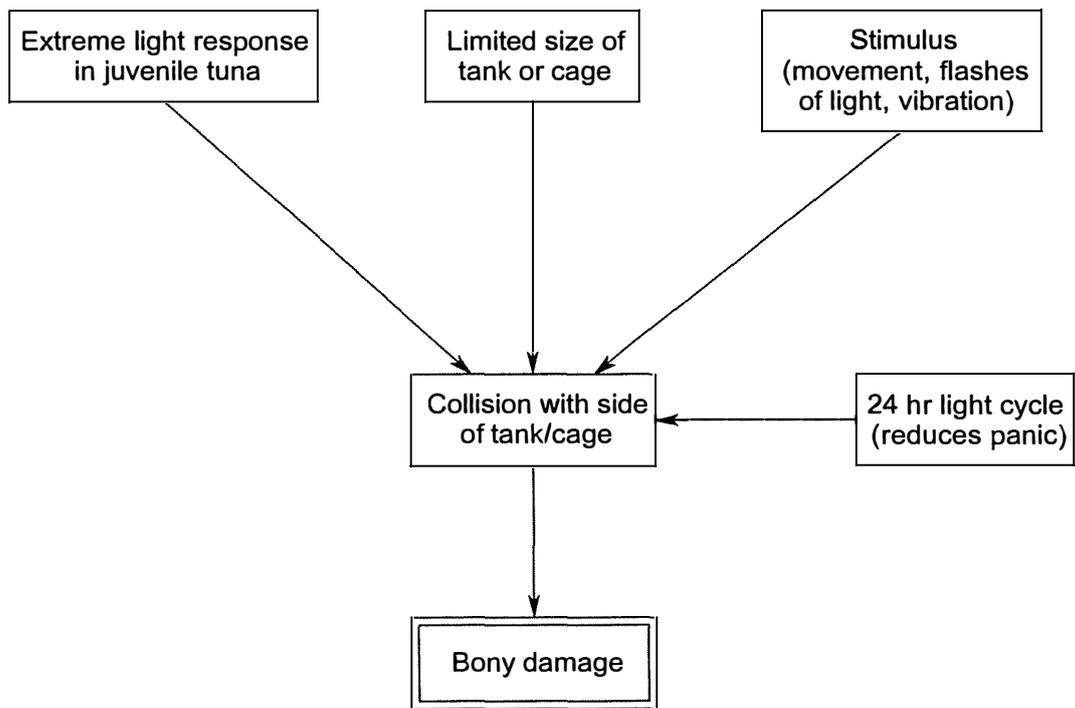
If a hatchery and grow-out facility for SBT is developed the probability of release for this condition should be included in the assessment. In the absence of grow-out of fish this condition is not applicable. Because this condition occurs on a regular basis in Pacific bluefin tuna hatcheries and grow-out facilities, the probability of exposure has been assessed as very high.

Consequence of entry, establishment and spread

Because the number of affected fish is relatively large (only because most have died at younger ages from other causes) the consequences of exposure have been assessed as high.

Further research

Maintain contact with the Japanese in regard to control measures.



Environmental problems

Hypoxia

Aetiology:

Hypoxia may arise due a number of different environmental factors:

- **Inadequate aeration of culture tanks.** Because of the high oxygen requirement of larval tuna aeration of the water column is of crucial importance.
- **Deoxygenation of the water column due to algal blooms.** When algal blooms respire during overcast weather or overnight they can readily deplete the oxygen in the water column. In 1997 a huge bloom of *Trichodesmium erythracum* in Spencer Gulf put the tuna industry on high alert (Holden 1997).
- **Impedement of water exchange in pontoons due to net fouling and/or poor tidal flows.** Both these factors are known to occur in relation to tuna culture and could contribute to mortalities due to hypoxia.
- **Deoxygenation by under pontoon sediments.** The studies by Cronin (1995) suggest that this could be a contributory factor in the less well-scoured sites around Boston Island.

In addition, a number of animal factors can influence the impact of relative hypoxia:

- **Oxygen requirements.** Under certain conditions, the requirements of fish for oxygen are increased, making them more susceptible to environmental oxygen depletion.
- **Gill damage.** A number of other disease problems (eg blood fluke, microalgal toxicosis) can lead to gill damage, decreasing the ability of the fish to absorb oxygen and increasing their susceptibility to low dissolved oxygen levels.

Clinical signs

Affected fish usually tend to come to the surface and/or seek water inlets. Gasping behaviour may be seen and usually the opercula are flared.

Pathology

Other than the presence of flared opercula there are no significant pathological findings.

Species/ages affected

As previously emphasised juvenile tuna have very high oxygen requirements and, therefore, are highly susceptible to hypoxia (Miyashita et al. 1999). In general terms, tuna are as sensitive to hypoxia as other teleosts. However, as indicated earlier, tuna are not able to adjust their activities under hypoxia because they must continue to swim in order to respire. Bigeye tuna are tolerant of lower dissolved oxygen levels than yellowfin tuna (Bushnell et al. 1990), but the situation with southern bluefin tuna is unknown.

Management practices

As previously indicated, net maintenance, pontoon siting etc. can be very important in maintaining adequate oxygen levels in the water column. If a period of hypoxia is predicted the fish should not be fed until the danger period has passed.

Diagnosis

Diagnosis should be simple by observation of the fish and measuring dissolved oxygen in the water column at different depths. Bushnell et al. (1990) suggested that the mean incipient oxygen response threshold for tunas was about 5 ppm.

Treatment

Depending on circumstances, treatment can be by supplementary aeration/oxygenation, the use of current movers (paddle wheels, outboard motors etc.).

Prevention

Prevention depends upon choosing appropriate sites for pontoons, ensuring that management practices do not contribute to hypoxia (eg ensure good water flow through nets) and, where appropriate, by avoidance, ie by towing nets away from algal blooms or other causes of de-oxygenation. Feed should be withheld during times of danger.

Hypothesised pathways of occurrence

Hypothesised pathways for occurrence of hypoxia in farmed tuna are shown in Figure 19.

Probability of entry, establishment and spread

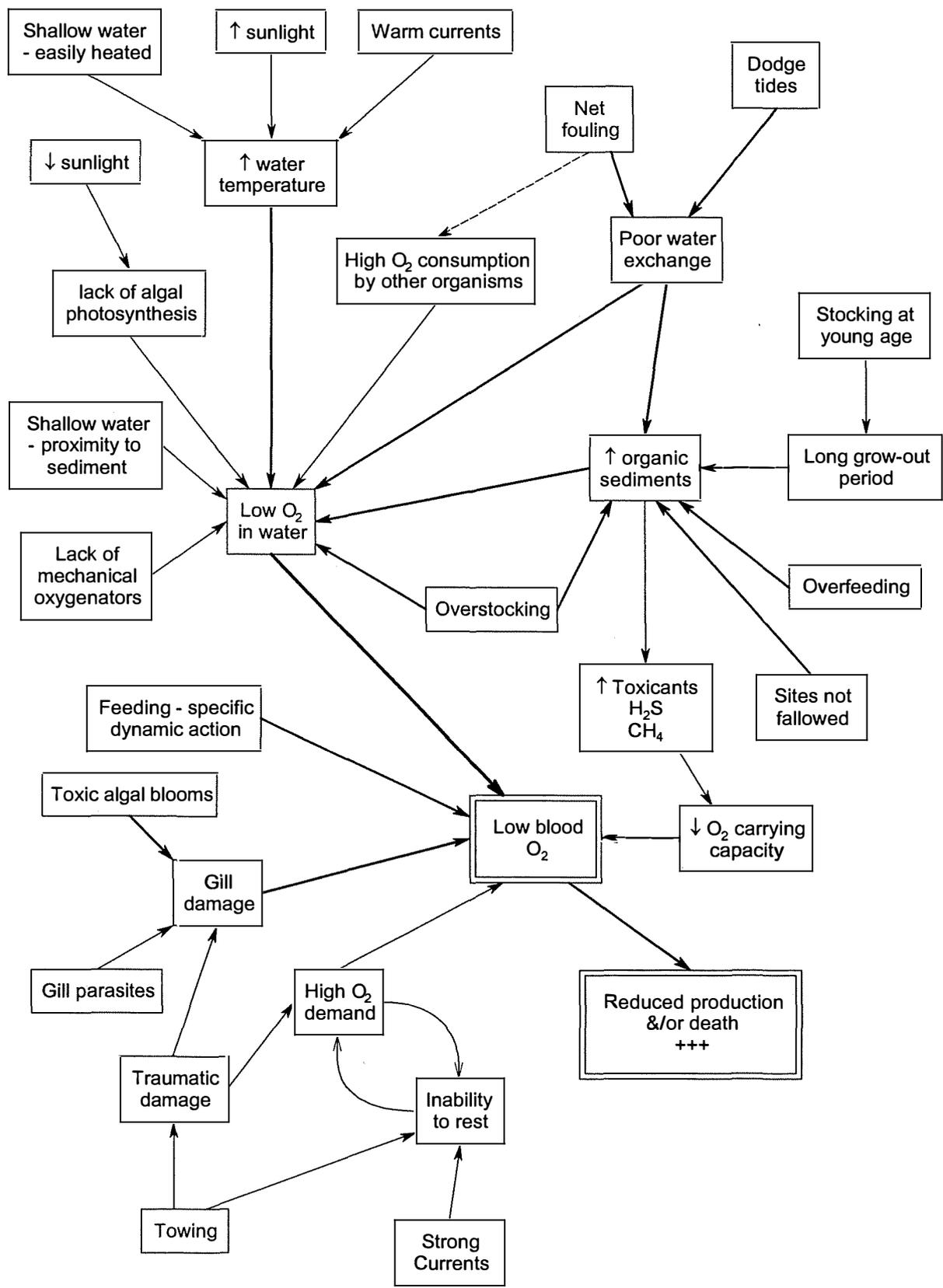
The regulation of a maximum stocking rate of 4 kg/cm² of pontoon area appears to address the potential hypoxia problem. Furthermore, after 10 years of tuna farming there is no evidence of oxygen depletion. The only time there had been a problem was when some farmers continued to feed during dodge tides. Currently, this is not done. The dissolved oxygen values obtained in government monitoring programs have always been above 6.0 mg/L. Because many of the causative factors have been identified and are effectively managed, the probability of exposure has been assessed as very low.

Consequence of entry, establishment and spread

Because of the presumed high susceptibility of SBT to hypoxia, the consequences of exposure have been assessed as extreme.

Further research

There is urgent need for research on the physiology of southern bluefin tuna.



Smothering by suspended solids

Aetiology

It is important to distinguish non-toxic suspended solids from toxic suspended solids, The latter are derived mainly from mining activities and are high in heavy metals such as copper and zinc. As tuna farms are unlikely to be sited where runoff from mines would occur the latter will not be considered here.

In his review of the available literature Waters (1995) reported that for small salmonids suspended solids in the range of 500 to 1,500 mg/L could lead to reduced survival. He did note that some workers had exposed fish to levels as high as 250,000mg/L for short periods without losses. Similarly, Stern and Stickle (1978) suggested that turbidities equivalent to 175,000-225,000mg/L were necessary to induce mortalities in freshwater fish.

In experiments with "Deep Ocean Mining Environmental Study (DOMES)" Barry (1978) found that yellowfin tuna would feed in waters with turbidities up to 7.4 NTU (equivalent to 10mg/L of DOMES mud).

Clarke (1996) attributed the 1996 mass mortality of southern bluefin tuna at Port Lincoln to the effects of suspended solids even though the highest turbidity reading quoted in that report was 2.0 NTU, several days after the peak mortality. However, as with all data related to that incident, no readings were available at the time of the main mortality and the diagnosis remains open.

Clinical signs

At lethal concentrations of suspended solids Stern and Stickle (1978) reported the following set of clinical signs:

- Momentary swimming at the surface and gulping of air and water.
- Leaning towards one side while remaining at the surface for several minutes.
- Floating on one side for up to 30 minutes with an occasional swimming movement.
- Floating with only occasional feeble opercular and pectoral fin movements until death.
- At sublethal concentrations the fish exhibit "coughing", apparently in an effort to clear their gills of sediment.

Pathology

Stern and Stickle (1978) reported that in fish which died the opercular cavities and gill filaments were clogged with sediment.

Type of suspended solids

Servizi and Martens (1991) found that the larger the particle size, the more deleterious the effects.

Temperature

The above authors also found that temperatures outside the optima for coho salmon decreased the tolerance of the fish to suspended solids.

Species/ages of fish

Smaller fish are more susceptible to the deleterious effects of suspended solids. In general, more active fish are less tolerant of suspended solids (Stern and Stickle

1978). Clarke attributed the mass mortality of southern bluefin tuna in 1996 in Boston Bay to suspended solids and cited a personal communication from Dr Peter Montague concerning a similar event in the Mediterranean involving northern bluefin tuna.

Diagnosis

Diagnosis of mortalities due to suspended solids should be relatively simple based on the presence of large quantities of sediment impacted in the opercular cavity, together with appropriate data on turbidity levels.

Treatment

If fish are transferred to clear water they successfully expel the accumulated solids.

Prevention

Prevention of deleterious effects of suspended solids in respect of tuna really means avoidance by towing pontoons away from the affected area.

Hypothesised pathways of occurrence

Hypothesised pathways for occurrence of smothering in farmed tuna are shown in Figure 20.

Probability of entry, establishment and spread

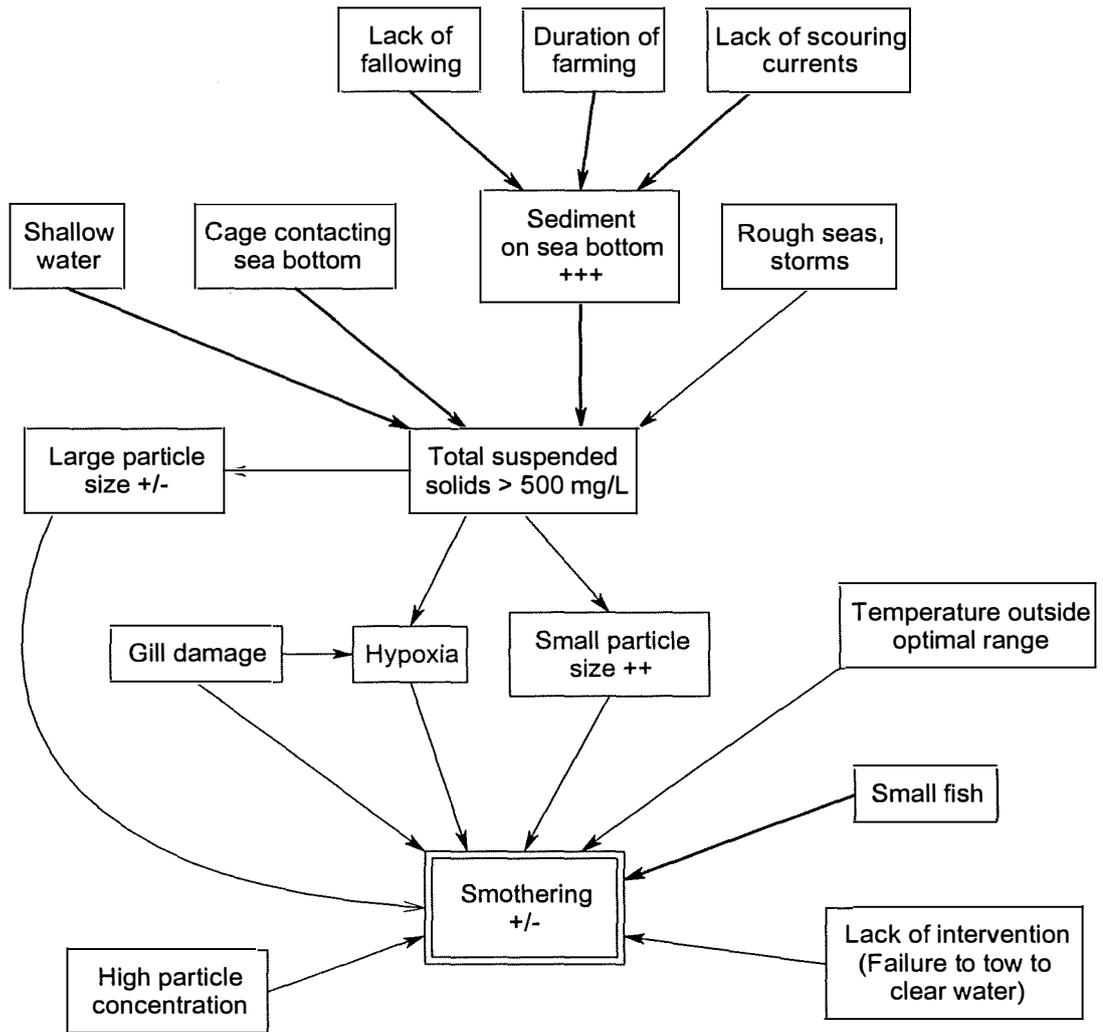
Because many of the causative factors have been identified and are effectively managed (eg by towing away from the affected area), the probability of exposure has been assessed as low.

Consequence of entry, establishment and spread

Because of the resistance of yellowfin tuna (Barry 1978) to smothering, the consequences of exposure have been assessed as low.

Further research

Not required at present.



Predation

Aetiology

Predation may be by man (poaching), seals or sharks.

Clinical signs

If the fish are completely removed or eaten there will be no clinical signs. Signs which may be seen are tooth or gaff marks and a general stress response with depressed feed intake.

Pathology

The pathology reflects the clinical signs.

Epidemiology

Basically predation is the result of having predators and fish in association in the absence of adequate protection.

Diagnosis

Sometimes predation is not detected until a full harvest has been conducted and a shortfall of fish has been revealed. Sometimes the predators are observed actually attacking/taking the fish or typical damage to the fish and /or the nets is obvious.

Treatment

Treatment is symptomatic eg reduce other stressors, mend damaged nets, etc.

Prevention

Reducing poaching involves regular patrolling of farms and installation of barriers. Although a number of strategies can be used to reduce seal and shark predation the modern approach is to use well-tensioned heavy-gauge nets and electric 'fences'. Of course attractants such as blood, carcasses etc should not be released near the pontoons.

Comment

Electric fences and other measures, as presently used by farmers, seem to have reduced predation problems.

Hypothesised pathways of occurrence

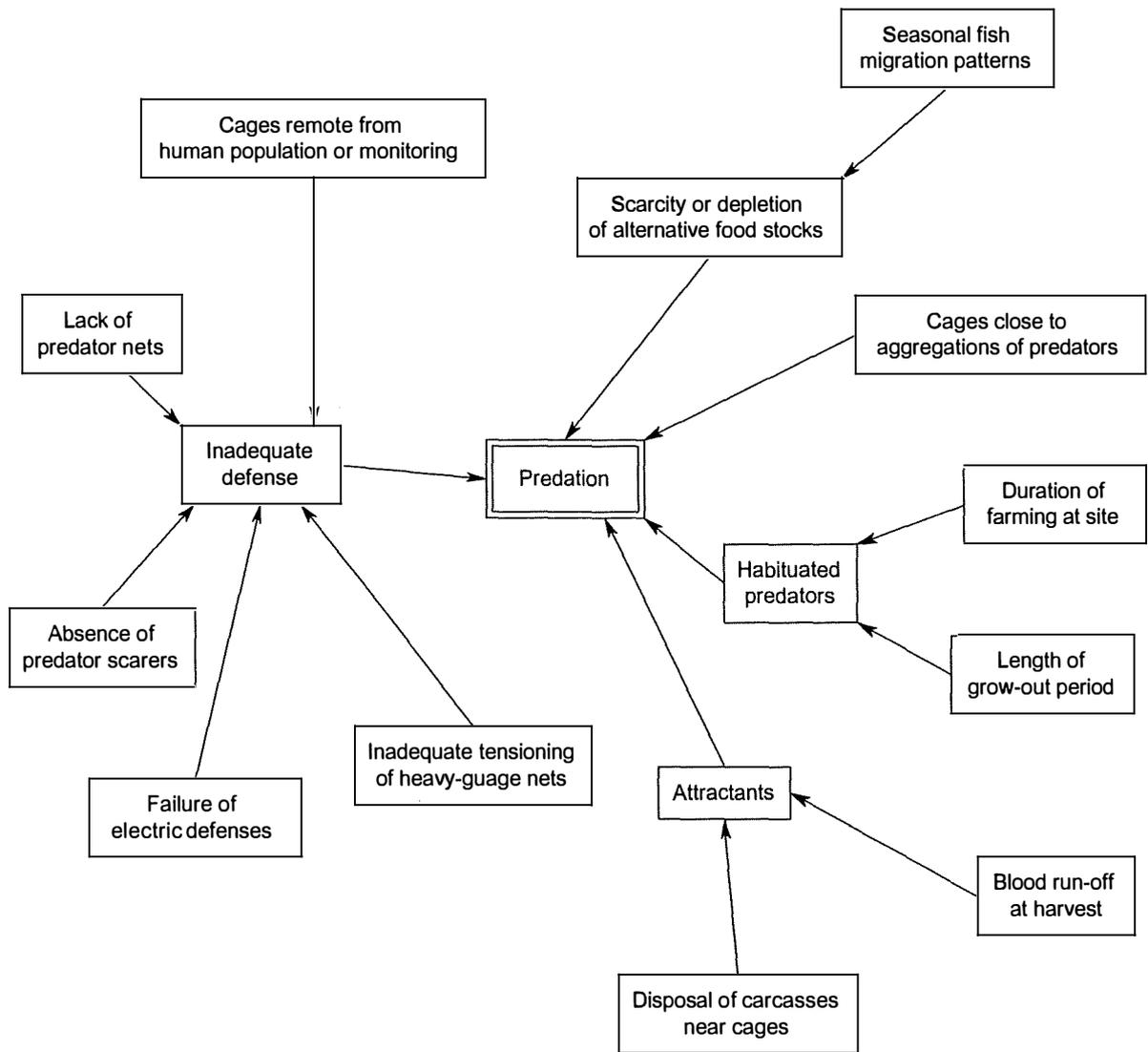
Hypothesised pathways for occurrence of predation in farmed tuna are shown in Figure 21.

Probability of entry, establishment and spread

Under normal circumstances, the likelihood of predation by predators is likely to be low. The likelihood of predation has been reduced by mitigation devices, for example electric fences, higher side netting and better main net husbandry. This assessment assumes best farming practices, including the use of mitigation devices.

Consequence of entry, establishment and spread

Consequence of entry, establishment and spread was assessed as moderate.



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Appendix 5

TBOASA TUNA HEALTH TABLE 1994-2000

TUNA HEALTH SUMMARY (prepared by Kirsten Rough for TBOASA) - 1994

MONTH	WILD SAMPLES ^a	BASILINE FARM SAMPLES <input type="checkbox"/>	HARVEST SAMPLES	TARGETED SAMPLES
Feb	200 (gross examination only to get familiar with normal size, shape and colour of organs; and parasites, 11 different types noted - <i>Neohexastoma</i> , <i>Didymozoid</i> 2, 3, 4, 5, 6, 7, <i>Cestoda</i> 1, 2, <i>Acanthocephala</i> 1, <i>Euryphorus</i>)	2 (meshed during transfer, a further 3 types of parasites found - <i>Cetriotrema</i> , <i>Didymozoid</i> 1, <i>Cestoda</i> *)		
Mar	750 (gross examination only to get familiar with normal size, shape and colour of organs; and parasites, all previous 14 types present plus and extra 5 - <i>Caballerocotyla</i> , <i>Kudoa</i> , <i>Nybelinia</i> , <i>Pseudocycnus</i> , <i>Cestoda</i> *)	5 (moribund post tow)	2 (brood stock 93 - NSF)	
Apr	70 (gross examination only to get familiar with normal size, shape and colour of organs; and parasites, no new types found)	3 (moribund post tow)	2 (brood stock 92 - NSF)	
Jun			15 (New parasite - <i>Pterobothrium</i>)	1 (swimmer haematology - depression to absence of eosinophils; <i>Uronema</i> demonstrated wet preparations & histology, no other significant findings)
Aug			5 (NSF)	
Sep			17 (NSF)	15 (nutrition trial, 5/treatment)
Oct				
Nov			10 (NSF)	20 (nutrition trial, 5/treatment)

LEGEND: ^a on the fishing grounds; at weight check or baseline sampling

*Cestoda** turned out to be different stages of development of the same tapeworm

MORTALITY EVENT signifies >200 mortalities from a single pontoon or lease site within a 48hour period

TUNA HEALTH SUMMARY (prepared by Kirsten Rough for TBOASA) - 1995

MONTH	WILD SAMPLES ^a	BASELINE FARM SAMPLES <input type="checkbox"/>	HARVEST SAMPLES	TARGETED SAMPLES
Jan			10 (brood stock 92/93, NSF)	
Feb	18 (majority NSF, inshore specimens had marked infiltration of lymphocytes in filament tips, new parasite in stomach lumen - <i>Hirudinella ventricosa</i> like, new parasite in pectoral fin recess - <i>Brachiella thynni</i>)	2 (moribund post tow, elevated CPK, chronic inflammation of filaments, no obvious cause)	10 (brood stock 91, NSF) 10 (94 carry-overs, NSF)	
Mar	10 (muscle only residue survey)		21 (New parasites - <i>Isopoda</i> 1, <i>Nematoda</i> 1, <i>Digenea</i> 3)	12 (eye damage (specimens sent to VPS had varying degrees of panopthalmitis and keratitis, gill filament tips and lamellae had chronic infiltration of lymphocytes) (cataract specimens sent to Uni Tas no report received, but probably later stage of the specimens sent to VPS) A new parasite <i>Caligus elongatus</i> was found to be consistently associated with the eye trauma; and over subsequent years this parasite was found to be only a problem in farms where fish diet was suboptimal, or practices induced severe stress to tuna)
May			3 (New parasites - <i>Digenea</i> 5, 6)	3 (residual eye damage)
Jun			9 (New parasite - <i>Digenea</i> 7)	2 (residual eye damage)
Jul			8 (NSF)	
Aug			15 (NSF)	24 (nutrition trial, fish condition well below normal for this late stage in the cycle, extreme neutrophilia and lymphopaena, mild anaemia, increased melanomacrophage centres in haematopoietic organs, mucus cell hyperplasia and infiltration of lymphocytes in gill lamellae and filaments)

TUNA HEALTH SUMMARY (prepared by Kirsten Rough for TBOASA) - 1996

MONTH	WILD SAMPLES ^a	BASILINE FARM SAMPLES <input type="checkbox"/>	HARVEST SAMPLES	TARGETED SAMPLES
Jan			8 (brood stock 91, NSF)	
Feb				2 (Immunology trial, fish injected (antisera and adjuvant) and reinjected, killed and serum sent to UTas. Both anaemic) 10 (Anaesthetic trial, 5x treated 5x controls, treated had 3 fold increase in blood cortisol, glucose and lactate; organs NSF)
Mar	10 (New parasite - <i>Copepoda</i> 5)		10 (NSF)	
Apr	6 (NSF)		10 (95-carryovers, NSF) 9 (New parasite - <i>Didymozoid</i> 8)	MORTALITY EVENT 655 Mortality autopsies (suitable for gross examinations only); 120 live fish and fresh mortalities (blood and histological samples collected where appropriate); All fish had flared opercula, extreme quantities of mucus on the gills, mucous smear microscopy demonstrated large quantities of fine sand, silt and clay particles sufficient to give a brown discoloration to the mucus. Presence of some phytoplankton in mucous, <i>Dinophysis caudata</i> and <i>Ceratium furca</i> . Spleen mildly to extremely congested, stomach in most cases very full, all other organs NSF. Histologically, mucous sloughed off during transport and processing, swollen epithelial cells on secondary lamella, large quantities of "nematode eggs" later identified as <i>Sanguinicolid</i> cercaria present in filaments, spleen extremely congested, staining suggested no evidence of haemolytic anaemia, liver, brain, kidneys, heart and other organs all NSF. Blood NSF. New parasite <i>Didymozoid</i> 10.
May			2 (brood stock 91, NSF)	2 sampled for Vet Lab 13 replicates for insurance case, elevated CPK 3 (swimmers depressed eosinophils, elevated neutrophils; <i>Uronema</i> demonstrated wet preparations & histology, no other significant findings)
Jul			10 (NSF)	1 (swimmer haematology - depression to absence of eosinophils; <i>Uronema</i> demonstrated wet preparations & histology, no other significant findings)

MONTH	WILD SAMPLES ^a	BASILINE FARM SAMPLES <input type="checkbox"/>	HARVEST SAMPLES	TARGETED SAMPLES
Aug			6 (NSF)	
Sep			10 (NSF)	

TUNA HEALTH SUMMARY (prepared by Kirsten Rough for TBOASA) - 1997

MONTH	WILD SAMPLES ^a	BASELINE FARM SAMPLES □	HARVEST SAMPLES	TARGETED SAMPLES
Jan		18 (NSF)		
Feb		15 (NSF)	20 (NSF) 20 (muscle for structure, fat deposition, and glycogen)	
Mar		20 (NSF)		
Apr	25 (NSF)		27 (NSF)	20 (pontoon experienced mortality - Algae bloom <i>Gymnodinium mikimotoi</i> like: histology demonstrated swollen epithelial cells in gut and intestine, gills had high levels of <i>Pseudocycnus appendiculatus</i> and amphipods (new parasite) in wet preparations, chitinous material associated with lesions in histological sections of primary and secondary lamellae)
May			21 (New parasite <i>Didymozoid</i> 9)	2 (Algae bloom <i>Gymnodinium mikimotoi</i> like: no significant findings) 11 (pellet fed: haematology – some anaemia, neutrophilia and lymphopenia; gross pathology - higher incidence of didymozoid, kudoa and caligid parasites, histology – increased melanomacrophage centres in haematopoietic organs, mucus cell hyperplasia and infiltration of lymphocytes in gill lamellae and filaments)
Jun			19 (NSF)	5 (pellet fed: haematology – some anaemia, neutrophilia and lymphopenia; gross pathology - higher incidence of didymozoid, kudoa and caligid parasites, histology – increased melanomacrophage centres in haematopoietic organs, mucus cell hyperplasia and infiltration of lymphocytes in gill lamellae and filaments)
Jul			12 (NSF)	
Aug			27 (NSF) 30 (muscle for structure, fat deposition, and glycogen)	6 (swimmers: haematology – depression to absence of eosinophils; <i>Uronema</i> demonstrated wet preparations & histology, no other significant findings)

MONTH	WILD SAMPLES ^a	BASELINE FARM SAMPLES <input type="checkbox"/>	HARVEST SAMPLES	TARGETED SAMPLES
Oct			10 (NSF) 10 (muscle for structure, fat deposition, and glycogen)	
Nov				30 (gross examination) + 6 (blood and histological samples) Pontoons across this lease showed generally low productivity and persistently high mortalities, autopsy: malnutrition and internal and gill haemorrhaging evident, scoliosis and lordosis, atrophy of spleen and digestive organs, corneal oedema and slight cataract formation; histology: depression of haematopoietic tissue {kidneys and spleen} increased melano-macrophage aggregates {kidneys liver and spleen}, irregularities in cartilage formation, haemorrhage and aneurysms (some surrounded by epithelial hyperplasia), vascular congestion and leakage {gills}, sections of perivascular haemorrhage {brain} - additional tissues sent for viral and bacterial culture, virology negative, microbiology mixed <i>Pseudomonad's</i> and <i>Vibrio's</i> (including <i>V. Harveyii</i>) full review of farm practices submitted to operator

TUNA HEALTH SUMMARY (prepared by Kirsten Rough for TBOASA) - 1998

MONTH	WILD SAMPLES ^a	BASELINE FARM SAMPLES <input type="checkbox"/>	HARVEST SAMPLES	TARGETED SAMPLES
Jan		2 (bilateral corneal opacity, organs - typical tow damage, Sanguinicolids + normal parasites) 5 (NSF)		12 (eye damage, <i>Caligids</i> present, post tow trauma evident in kidney and spleen, moderate quantities of "nematode eggs" later identified as <i>Sanguinicolid</i> cercaria present, no reaction by gills)
Feb		1 (tow trauma) 12 (baseline for commercial farms' feed supplement trial, health plus muscle structure, fat deposition, and glycogen level and vitamin testing, new parasite, <i>Syncoelium</i> on tongue)		10 (same tow pontoon as above, persistent eye damage, <i>Caligids</i> present, blood; 1 x lipaemic, 5 x slightly anaemic, histology; depression of lymphomyeloid tissue increased melano-macrophage aggregates) 10 mortalities, 5 gaffed (eye damage, numerous <i>Caligids</i> present (>50/fish), post tow trauma very evident in gross examinations and histology of kidney and spleen, low quantities of "nematode eggs" later identified as <i>Sanguinicolid</i> eggs present in lamellae, but no cellular reaction)
Mar			10 (full sampling, NSF) + 20 (muscle tissue for structure, fat deposition)	13 (Algae bloom, <i>Trichodesmium</i> been present from Liguana Island east since mid February, now also <i>Gymnodinium breve</i> and <i>Chattonella</i> sp. in passage and tuna farm area fish slow - elevated mortality from two tow pontoons (supplied 4 farmers) corneal oedema, gills moderately mucousy, lymphomyeloid tissue depressed, liver congested pooled samples sent for virology all negative. New parasite - <i>Monogenea</i> 3) 12 (mortalities; gross pathology - seal attacks) 2 (pontoon experiencing mortality; gross pathology - fish thin, pasty white plaques of mucus on gills, atrophy of spleen, residual tow damage evident; haematology - depressed haematocrit, elevated leucocrit, elevated eosinophils and neutrophils; histology - lymphoid depression, <i>Sanguinicolid</i> eggs present in lamellae, mild cellular reaction) 35 (mortalities from pontoon experiencing increased deaths; gross pathology - post tow trauma + gills very mucousy in some <i>Chattonella</i> and amphipods present in mucus, 30 fish had fibrous patches on ventricle, 2 fish looked anaemic)

MONTH	WILD SAMPLES ^a	BASELINE FARM SAMPLES <input type="checkbox"/>	HARVEST SAMPLES	TARGETED SAMPLES
Apr			<p>10 (elevated haematocrit)</p> <p>10 (depressed haematocrit, all with <i>Sanguinicolids</i> in gill screens but no tuna mortality)</p> <p>63 (gill screens for pellet material, mucus, lesions etc)</p>	<p>10 (pellet supplemented pontoon with increased mortality; fish generally in pontoon look stressed, gross pathology – gills very mucousy, atrophy of spleen and digestive organs, livers bright orange; haematology – mild anaemia, neutrophilia; histology - lymphoid depression, depressed activity in pancreas)</p> <p>2 (mortalities; gross pathology – thin fish with excessive Caligids, atrophy of haematopoietic and digestive organs)</p> <p>84 (7/pellet and pilchard treatment research farm; blood only for haematocrit and leucocrit)</p>
May			<p>35 (gills and ventricle for sanguinicolids)</p> <p>38 (NSF)</p>	
Jun				7 (fed dry pellet, research farm; blood only for haematocrit and leucocrit)
Jul			<p>60 (gills and ventricle for sanguinicolids)</p> <p>10 (NSF)</p> <p>10 (muscle and liver for vitamin E)</p>	
Aug			<p>14 (parasite check only)</p> <p>10 (NSF)</p>	
Sep			<p>10 (muscle and liver for vitamin E)</p> <p>7 (NSF)</p>	

TUNA HEALTH SUMMARY (prepared by Kirsten Rough for TBOASA) - 1999

MONTH	WILD SAMPLES ^a	BASELINE FARM SAMPLES □	HARVEST SAMPLES	TARGETED SAMPLES
Feb			10 (NSF)	
Mar	25 (NSF)		100 (gills and ventricle for sanguinicolids) 10 (NSF)	
May			100 (gills and ventricle for sanguinicolids) 20 (NSF)	<p>MORTALITY EVENT</p> <p>45 (gross examination, histological samples collected where appropriate)</p> <p>Rapid onset of mortalities no environmental abnormalities, autopsy: moderate to excessive amounts of mucus on gills, atrophy of spleen and haematopoietic organs, muscle pale and soft; histology: patchy swelling of lamella epithelial cells, excess mucus with collections of bacterial rods, atrophy and depression of haematopoietic tissue (kidneys and spleen), acute degeneration of kidney tubular epithelial cells, marked swelling and fragmentation of myofibres some resembling cystic structures of variable size, congestion and degenerate products between affected myofibres, complete absence of glycogen in all muscle sections, <i>Uronema</i> present in brain sections of 1 fish - additional tissues sent for alpha-tocopherol, parasitology, viral and bacterial culture, muscle alpha-tocopherol levels low, muscle parasitology NSF, virology negative, microbiology growth of <i>Pseudomonas fluorescens</i> (brain and kidney) and <i>Ps. aeruginosa</i> (kidney) from some fish</p>
Jun				5 (at harvest, follow up from May: elevated levels of blood haematocrit, haemoglobin, neutrophils, CPK, protein, sodium, potassium, cortisol, glucose and lactate; histology: collections of mucus on filaments, atrophy and depression of haematopoietic tissue (kidneys and spleen), swelling, fragmentation, and haemorrhage in and around myofibers)

MONTH	WILD SAMPLES ^a	BASELINE FARM SAMPLES <input type="checkbox"/>	HARVEST SAMPLES	TARGETED SAMPLES
Jul			100 (gills and ventricle for sanguinicolids) 20 (NSF)	MORTALITY EVENT (same fish as May) 30 (gross examination) + 10 (blood and histological samples) Rapid onset of mortalities coincide with dodge tide following 3+days 100% cloud cover and flat calm seas, autopsy: sediment plus moderate to excessive amounts of mucus on dark, congested gills, gill parasites all dead, atrophy of spleen and haematopoietic organs, liver and muscle pale and soft; histology: large collections of mucus and bacteria on filaments, atrophy and depression of haematopoietic tissue (kidneys and spleen), swelling, fragmentation, haemorrhage in and around myofibers - additional tissues sent for alpha-tocopherol, viral and bacterial culture, muscle alpha-tocopherol NSF, virology negative, microbiology, mixed culture of <i>Vibrio's</i> , <i>Aeromonads</i> , <i>Pseudomonads</i> , <i>Serratia</i> and <i>Proteus</i> sp
Aug			150 (gills and ventricle for sanguinicolids)	

TUNA HEALTH SUMMARY (prepared by Kirsten Rough for TBOASA) - 2000

MONTH	WILD SAMPLES ^a	BASELINE FARM SAMPLES <input type="checkbox"/>	HARVEST SAMPLES	TARGETED SAMPLES
Jan				<p>Cultured Kingfish mass mortality - hypoxia</p> <p>35 (mortalities; gross pathology - seal attacks)</p> <p>3 (mortalities; gross pathology - spleen extremely congested, haematopoietic kidney pale swimbladder very distended, all other organs NSF)</p>
Feb		4 (Muscle and liver for residues only)		20 (mortalities from various farms; gross pathology - gill and buccal cavity congestion)
Mar			<p>130 (gill and buccal cavity inspection only)</p> <p>27 (Muscle and liver for residues only)</p> <p>10 (NSF)</p> <p>8 (NSF)</p>	<p>57 (Pontoon experiencing mortality; Anaemia, rectified by dietary adjustments)</p> <p>5 (pellet fed tuna, mortalities; gross pathology - very thin with intense infestations of Caligids, anaemic, atrophy of spleen and digestive organs, high incidence of didymozoids)</p> <p>13 (Lease experiencing mortality; gross pathology - gill and buccal cavity congestion, sediment and phytoplankton present in mucus mixed algae bloom extending across lease (<i>Skeletonema</i> dominant species, no toxic species present))</p>

MONTH	WILD SAMPLES ^a	BASELINE FARM SAMPLES <input type="checkbox"/>	HARVEST SAMPLES	TARGETED SAMPLES
Apr				<p>40 (mortalities; gross pathology – seal attacks)</p> <p>7 (mortalities from various pontoons same lease; gross pathology – gill and buccal cavity congestion others seal attack)</p> <p>3 (Pontoon increased mortality: haematology - neutrophilia, lymphopaena; gross pathology - fish thin to moderate condition, aneurysms and congestion of gills, amphipods associated with inflammation and necrotic patches, atrophy of spleen)</p> <p>5 (Pontoon increased mortality: gross pathology - fish moderate condition, excess mucus on gills, amphipods associated with inflammation, atrophy of haematopoietic organs, congestion of spleen)</p> <p>10 (Lease increased mortality: pontoons grossly overstocked; gross pathology – fish quite thin, high prevalence of <i>Caligus</i>, congested gills with excessive mucus, atrophy of haematopoietic organs, low dissolved oxygen, mixed algae bloom extending across lease (<i>Skeletonema</i> and <i>Thalassiosira</i> dominant diatoms $150+KL^{-1}$, <i>Gymnodinium</i> dominant dinoflagellate $16KL^{-1}$, no <i>Chattonella</i> present)</p>
May			20 (NSF)	10 (at harvest, follow up from March dietary deficiency: slightly elevated neutrophils, and spleen mildly congested)
Jun			20 (NSF)	
Jul			20 (NSF)	

Appendix 6

SURVEILLANCE AND MONITORING RECOMMENDATIONS

Provided as part of AusVet Animal Health Services report to Aquafin CRC

Introduction

This section aims to outline a number of recommendations for a SBT health surveillance and monitoring system, based on the findings of this risk assessment. Note that it does not purport to detail all the requirements of an industry surveillance program, nor attempt to comprehensively address all issues that may be of concern to the industry. The scope is limited to surveillance of diseases that impact on the health of cultured SBT in Australia. It is anticipated that, if adopted, these recommendations will be integrated with other aspects of a surveillance system addressing other issues such as product quality, market support, animal welfare and environmental management.

Surveillance and monitoring are terms which are often confused and for which a variety of inconsistent definitions exist. In this report, surveillance is defined as a systematic process of data collection, management, analysis and reporting, targeted at the detection of diseases which are not currently known to occur in Australia. Surveillance therefore deals with exotic and emerging diseases, and its purpose is to alert the industry to a new disease event. Monitoring on the other hand is a systematic process of data collection, management analysis and reporting, targeted at detecting changes in the distribution of level of diseases known to occur in the population.

Objectives

An important prerequisite for an effective surveillance and monitoring system is a clearly stated set of objectives for the system. This should be formulated by industry in collaboration with other stakeholders. However, as an example, the objectives of a tuna health surveillance system may be:

- The early detection of exotic or emerging diseases;
- The early warning of environmental or other conditions likely to result in disease problems;
- The detection of changes in the level or distribution of identified priority endemic diseases;
- The provision of data to support claims of freedom from specified diseases.

Note that the objectives of surveillance are distinct from the objectives of the research priorities identified above. It is possible that the outputs of a surveillance system may either provide data which can be used for research, or indicate further research needs.

Types of surveillance and monitoring

In order to meet the stated objectives, a number of different approaches to surveillance and monitoring are available.

Targeted surveillance and monitoring

This approach is based on the targeted identification of animals with a specific disease. Targeted surveillance may, for instance, have the aim of detecting RSIV, and involve the routine testing of fish for evidence of the presence of the virus, using a specific test (eg PCR). The key characteristic of targeted surveillance is that, while it provides information about the disease being targeted, it provides no information about any other disease. For instance, a fish may be PCR negative to RSIV, but have blood fluke problems. A system targeted at detecting RSIV would not provide any information about the blood fluke.

General surveillance

The other approach to surveillance that can be used is general surveillance. This is not targeted at any disease in particular but aims to collect information about all diseases (or at least all significant diseases). A general surveillance system is unable to use specific diagnostic tests such as PCR, but instead depends on broader indicators of disease, which may then be followed up with a specific disease investigation. Laboratory diagnostic submissions are a good source of general surveillance data. Another example of general surveillance is a system involving the systematic recording of a general disease indicator (eg number of morts). Changes in the indicator indicate a change in the disease status, alerting producers to a potential problem. The surveillance system does not necessarily have to identify what the problem is, but simply indicate that a problem exists, and trigger a more detailed investigation. The main advantage of the use of general surveillance is that no other system is able to detect incursions or emergence of previously unrecognised diseases. As the tuna industry is relatively young, there is a significant risk that new diseases will continue to emerge for some time, so general surveillance will be required to detect them and respond to them in a timely manner.

Environmental surveillance

Another approach to surveillance or monitoring is not to measure the level of disease in fish, but to monitor key risk factors for that disease, in an attempt to predict the likelihood of disease occurrence and provide early warning. While this may be used in a number of different contexts, environmental monitoring gives one example. In particular, monitoring the environment to predict the risk of microalgal blooms could provide early warning for microalgal toxicosis.

Structure of a surveillance and monitoring system

An effective tuna health surveillance and monitoring system is likely to require a combination of approaches to achieve its objectives. In addition, as indicated in the definitions given above, data collation, management and reporting systems are essential components of a surveillance and monitoring system, as without them, the information collected is not able to assist the industry.

A comprehensive surveillance and monitoring system may have many different components. While these may all operate independently (which is often the case with systems that have evolved over time, and taken on new components to address new risks), there are often distinct advantages to combining all the surveillance and monitoring activities into a single system with coordinated data management, analysis and reporting functions. This is because much of the supporting data required to appropriately analyse data from one component of a

surveillance system, (eg farm population data), is required by all components of the system. Integration of multiple data sources into the same system may result in significant increases in efficiency.

Any surveillance and monitoring system is likely to involve contributions from a number of different sources, however the most important of these is the producers themselves. As a result, success of the system requires the cooperation of most, if not all, of the producers. This cooperation should extend to a willingness to contribute farm level data to the surveillance system, and to make, record and submit a limited number of extra observations to the system.

A model for an industry-level monitoring and surveillance system that has been effective in other industries is the use of an Internet-based centralised database with secure access to authorised (industry users). The database manages the submission of monitoring and surveillance information by all data providers (including producers), performs automated analysis, and acts as the main source for reporting of surveillance information, on demand to authorised users. This is one model that may be worth considering for the SBT industry.

Recommended components

These recommendations are based on the outputs of the risk assessment, as well as an understanding for the need for general surveillance to support industry protection against incursions of exotic or emerging diseases. They are based on a combination of the three types of surveillance listed above – targeted, general and environmental. The recommended components are:

- Targeted surveillance for known exotic disease threats (e.g. RSIV)
- Targeted monitoring of key endemic disease threats (e.g. parasites)
- Environmental monitoring for early warning of disease (e.g. microalgal toxicosis and hypoxia)
- General monitoring for assessing endemic disease
- General surveillance for exotic or emerging disease threats

These are discussed in more detail below. Note that this discussion is limited to a broad outline of the recommended areas for surveillance. A detailed surveillance plan would require further discussions with stakeholders and investigation of the most appropriate and practical approach to achieving the objectives, including issues of stakeholder responsibilities, data collection methods, data recording systems, data recording frequency, analysis and reporting procedures and funding options.

Targeted surveillance for known exotic disease threats (RSIV)

The risk analysis identified red sea-bream iridovirus as pathogen with a potential to cause significant losses in future SBT hatcheries, it is recommended that a suitable surveillance system be developed to provide early warning of any incursion of the virus into Australian waters. The exact operation of the surveillance system should be developed in light of the findings of the recommended virological research. For instance, if the host range for RSIV is found to include other species, or if evidence of the virus is found in Australia, this may alter the best choice of species and/or tissues for sampling as part of the surveillance system. In addition, virological

research of the other viruses listed in this risk analysis may indicate that they too should be included in the surveillance system.

Depending on the research findings, one example of a surveillance system for RSIV may involve a combination of regular testing of randomly selected samples from frozen imported baitfish (using, for example, a rapid inexpensive specific test such as PCR), along with routine testing of suspect morts sent to the laboratory for diagnosis using a more general tissue culture approach (ie a combined targeted surveillance system and enhanced general surveillance system).

Targeted monitoring of key endemic disease threats (parasites)

Two parasites were identified as posing increased risks: blood fluke and gill fluke. Because of the potential impact of these parasites on individual health and the industry as a whole (through the downgrading of carcasses), it is suggested that a monitoring system be introduced to track changes in the prevalence and distribution of these parasites within the industry.

Monitoring of both gill fluke and blood fluke could be relatively easily achieved through the systematic collection of observations of tuna gills at harvest time. A quick examination is able to identify either evidence of the parasite, or lesions typical of blood fluke infection with reasonable accuracy. If systematically collected across the industry, this would provide valuable information on the true magnitude of the disease problems, as well as any changes that occur over time.

Environmental monitoring for early warning of disease (microalgal toxicosis and hypoxia)

The potential impact of microalgal toxicosis on the industry means that it is important to maintain and enhance the capacity to provide early warning of potential problems. The type of program instituted by the TBOASA for microalgal monitoring should be continued or further enhanced, through the regular collection and identification of microalgae from a representative number of sites in the culture area at regular intervals. This would allow a damaging bloom to be rapidly identified and evasive action, such as towing pontoons to other sites, to be implemented. In addition, this type of surveillance information may be combined with locally collected or remotely sensed environmental data (eg sunshine, water surface temperatures, nutrient concentrations, oceanographic parameters) to develop a model for use in forecasting the occurrence of toxic blooms.

This system should include not only potentially toxic algae, but a coordinated reporting and monitoring system for blooms of any algae, due to the role they can play in causing problems with hypoxia. As with microalgae, a combination of field observations of algal blooms, oxygen meter readings, and remotely sensed data on temperature or cloud cover may enable the development of a predictive model, acting as an early warning system for hypoxia.

General monitoring for assessing endemic disease

While the monitoring and surveillance systems discussed to this point may be effective at detecting, or even predicting, the occurrence of the high and very high risk diseases identified in this study, there remains the problem of detecting either new disease threats, or existing diseases, currently ranked with low risk, but which may increase in importance due to a change in environment, management or other factors. In order to rapidly identify the potential emergence of endemic

diseases, or changes in the frequency of current disease problems, it is recommended that a general monitoring system be developed. Practical models for such a system employed in other industries involves the collection of data on a limited number of readily available key indicators of disease, and the continuous monitoring of these indicators across the entire industry. One common example is the use of morts as a key indicator. Systems exist whereby each farm could confidentially report the mortalities to a centralised system. No farm would be able to access another farm's details, but the system would report and analyse the industry total and average mortalities. This could be used for benchmarking (allowing a farmer to determine if they are suffering more mortalities than the current industry average), as well as identifying industry level events where all farms suffer an unusual increase in the level of mortalities. While mortalities are used as an indicator in this example, a number of other indicators could be recorded and reported within the system. Such a system may be in addition to, but integrated with, any other specific surveillance activities undertaken as a result of the risk assessment described in this document.

General surveillance for exotic or emerging disease threats

The use of simple, inexpensive key indicators at the farm level provides one approach to identifying unusual disease events. However a simple increase in mortalities would not be adequate to identify the incursion of an exotic disease. Such a system would need to be supported by another system, able to carry out more detailed disease investigations and make definitive diagnoses of disease problems. Typically, this is based on the use of data from diagnostic laboratories, as discussed previously in the report on the industry health database.

It is recommended that a systematic approach be used to submit a representative number of moribund and dead fish for laboratory examination, in order to identify the cause of any unusual disease events, as well as provide on-going assurance of the absence of key known disease threats. Examination of such specimens would initially involve the use of general tests, capable of detecting a range of diseases and disease processes (such as a combination of gross pathology, histopathology, and, if indicated, viral tissue culture). Specific tests could then be used to characterise any agents. As with other components of the surveillance system, but also highlighted in the review of the tuna health database, it is critically important to ensure that the data generated through such a surveillance system are captured, managed, analysed and reported in such a way as to achieve the objectives of the system.

Conclusions

The components of a SBT health surveillance and monitoring system discussed in this section could be achieved through the following activities:

Centralised surveillance database development

Ideally, a secure centralised database, accessible by all producers, and responsible for the management, analysis and reporting of surveillance data would be set up. Such a database would allow the confidential submission of individual farm data, which would then be used to present the aggregate industry picture without compromising private information. All data generators identified below would be responsible for submitting data to the database for surveillance and monitoring purposes.

On farm data collection

Producers would provide the core of the surveillance data through on-farm data collection. Data items to be recorded and submitted to the monitoring system would include:

- Total population
- Total morts
- Oxygen levels
- Other environmental variables such as water temperature and turbidity
- Observations of algal blooms
- Specimens of algae for laboratory identification
- Submission of moribund fish, or fish from an unusual disease incident to the laboratory for diagnosis
- Recording observations of gill lesions from all fish at harvest

Remote data collection

Depending on the results of research into the development of models for early warning, a number of remotely collected environmental variables may be important for integration into a surveillance and monitoring database. These may include interpolated temperature data from the Bureau of Meteorology, and a range of satellite data including ocean currents, surface water temperature and cloud cover.

Laboratory support

The diagnostic laboratory would provide general diagnostic information which would be submitted to the surveillance and monitoring database, including details of the disease events that prompted submission, findings, and diagnosis. In addition, specific testing results for RSIV and any other viruses included in the surveillance system would be included.

Laboratory identification of algae would also be submitted to the database.

Feed monitoring

If research indicated that the approach was suitable, the laboratory would also provide information on PCR results from routinely sampled imported frozen baitfish, for target viruses.

Appendix 7

TUNA HEALTH RISK REGISTER INDUSTRY SURVEY RESULTS

Results collated and provided as part of AusVet Animal Health Services report to Aquafin CRC

Introduction

As part of the process of building a risk register and prioritising risks to the tuna health, a survey was developed and distributed amongst the industry. A copy of the questionnaire is shown in Appendix 1.

Responses were received from 17 individuals representing 14 producers and 2 government organisations (SARDI and PIRSA). Two individuals divided their responses into two groups, potential versus actual risk, and current risk versus risk if factor present.

Respondents were invited to add any significant risks not already listed. Eight environmental, three disease, 12 husbandry and two other risks were added to the list.

Summary of Responses

The following table summarises the responses to each of the risks.

1) Environmental	Responses	Mean	Max	Min	Median	Rank
Microalgae	19	5.6	9	2	6	6
Low dissolved oxygen	19	4.9	9	0	5	9
Accumulation of farms wastes on seafloor	19	3.5	8	1	2	23
Resuspension of sediments through net contact with the seafloor	19	3.1	9	0	2	26
Resuspension of sediments through storm activity	19	4.3	10	0	3	14
Sediments acting as a reservoir of disease-pests	17	3.3	8	0	2	24
High H2S levels in sediments	17	2.2	10	0	1	28
High nutrient levels	18	3.1	8	0	2	27
Predation	19	6.1	10	1	7	3
Net fouling affecting water flow and dissolved oxygen levels	19	4.7	10	1	5	13
Net fouling affecting pontoon flotation	17	3.2	10	0	2	25
Net fouling affecting the transparency of the structure to currents and waves and this making it more likely to be damaged, moorings to drag, etc	17	3.9	10	1	3	20

Net fouling increasing the occurrence of fish health issues by providing a substrate for disease/pests	19	4.0	10	0	3	19
Birds acting as a reservoir/host for disease/pests	19	3.6	10	0	3	22
Poaching	2	6.0	8	4	6	
Availability of lease sites	7	7.7	10	3	7	2
Ballast water	3	8.3	10	7	8	1
Water temperature	1	2.0	2	2	2	
Water quality	1	6.0	6	6	6	
Government resource allocation policy	1	7.0	7	7	7	
Storm damage	1	7.0	7	7	7	
Seals	1	6.0	6	6	6	
2) Disease						
Bacterial pathogens	18	4.3	10	1	4.5	15
Viral pathogens	18	4.3	10	0	5	16
Swimmer syndrome	19	5.1	10	0	6	8
Other parasites	18	4.8	10	1	5.5	10
Emergent disease	1	9.0	9	9	9	
Cateracts	1	2.0	2	2	2	
<i>Kudoa</i>	1	6.0	6	6	6	
3) Husbandry						
Feed - baitfish	18	4.8	10	0	5	11
Feed - manufactured	17	3.8	8	0	4	21
Propagation (hatchery)	17	5.4	10	1	5	7
Other species culture	17	4.8	10	0	4	12
Time in captivity	16	4.3	10	0	4	17
Source of aquaculture SBT	18	4.2	10	0	4	18
The tow experience - net damage, lack of feeding, sunburn	18	5.9	10	1	7	4
Transfers - stress, net damage	18	5.7	10	1	6.5	5
Continuous culture	1	9.0	9	9	9	
Ships striking pontoons	2	7.0	10	4	7	
Ballast water	6	8.0	10	6	8	1
Government	2	10.0	10	10	10	
Infrastructure cleaning on site	1	6.0	6	6	6	
Infrastructure cleaning on shore	1	2.0	2	2	2	
Oil and fuel spills, septic systems overflow etc	1	6.0	6	6	6	
Feeding on dodgy tides	1	7.0	7	7	7	
Public perception	1	10.0	10	10	10	
Storm water run off	1	8.0	8	8	8	
Heavy industry residues						
Proximity of other finfish/mussels	1	10.0	10	10	10	
4) Other						
Politics	1	8.0	8	8	8	
Sewage from ships	1	7.0	7	7	7	

Summary of Participants' Response Patterns

The differences in the perceptions by different participants of risk is illustrated in the following table.

Respondents' Patterns	Responses	Mean	Max	Min	Median
Respondent 1	27	1.4	8	0	1
Respondent 2	29	7.6	10	1	8
Respondent 3 (Potential Risk)	25	8.9	10	5	10
Respondent 3 (Actual Risk)	22	2.7	6	0	3
Respondent 4	26	3.2	8	2	3
Respondent 5	28	5.2	10	1	5
Respondent 6	27	3.6	8	1	2
Respondent 7	28	3.6	10	0	4
Respondent 8	28	5.9	10	2	5.5
Respondent 9	27	3.4	10	1	2
Respondent 10	27	2.4	10	1	1
Respondent 11	27	6.8	10	0	7
Respondent 12	28	1.9	7	0	1
Respondent 13	37	5.5	9	2	6
Respondent 14	25	4.2	8	2	3
Respondent 15	27	6.2	8	3	6
Respondent 16	26	3.8	8	1	4
Respondent 17 (Risk if factor present)	27	7.0	10	1	8
Respondent 17 (Risk using current practices)	18	2.7	10	0.5	1.25

Ranked Responses

Risks were ranked on the basis of the average risk level of all responses. Factors identified as risks by fewer than 7 respondents were excluded from the ranking. The ranked risks are shown in the table below:

Risk	Responses	Mean	Max	Min	Median	Rank
Ballast water	9	8.1	10	6	8	1
Availability of lease sites	7	7.7	10	3	7	2
Predation	19	6.1	10	1	7	3
The tow experience - net damage, lack of feeding, sunburn	18	5.9	10	1	7	4
Transfers - stress, net damage	18	5.7	10	1	6.5	5
Microalgae	19	5.6	9	2	6	6
Propagation (hatchery)	17	5.4	10	1	5	7
Swimmer syndrome	19	5.1	10	0	6	8
Low dissolved oxygen	19	4.9	9	0	5	9
Other parasites	18	4.8	10	1	5.5	10
Feed - baitfish	18	4.8	10	0	5	11
Other species culture	17	4.8	10	0	4	12
Net fouling affecting water flow and dissolved oxygen levels	19	4.7	10	1	5	13
Resuspension of sediments through storm activity	19	4.3	10	0	3	14
Bacterial pathogens	18	4.3	10	1	4.5	15
Viral pathogens	18	4.3	10	0	5	16
Time in captivity	16	4.3	10	0	4	17
Source of aquaculture SBT	18	4.2	10	0	4	18
Net fouling increasing the occurrence of fish health issues by providing a substrate for disease/pests	19	4	10	0	3	19
Net fouling affecting the transparency of the structure to currents and waves and this making it more likely to be damaged, moorings to drag, etc	17	3.9	10	1	3	20
Feed - manufactured	17	3.8	8	0	4	21
Birds acting as a reservoir/host for disease/pests	19	3.6	10	0	3	22
Accumulation of farms wastes on seafloor	19	3.5	8	1	2	23
Sediments acting as a reservoir of disease-pests	17	3.3	8	0	2	24
Net fouling affecting pontoon flotation	17	3.2	10	0	2	25
Resuspension of sediments through net contact with the seafloor	19	3.1	9	0	2	26
High nutrient levels	18	3.1	8	0	2	27
High H ₂ S levels in sediments	17	2.2	10	0	1	28

Discussion

The risks identified can be divided into a number of broad classes:

- Infectious disease
- Trauma
- Nutritional problems
- Predation
- Toxins, pollution and water quality
- Oxygen and smothering
- Damage to infrastructure
- Political risks

A number of the risks listed can contribute to several different classes. For instance sea floor sediments can provide a substrate for infectious diseases, produce toxins (H_2S), decrease O_2 levels and make smothering more likely.

It is important to note, that SBT industry saw baitfish as a hazard but from the point of view of lack of supply and reduced quality at the time of low supply (potentially a nutritional problem), not as a source of pathogens.

The last two categories (political problems and damage to infrastructure) can be disregarded for the purposes of this study, as they do not represent direct risks to tuna health, but risks to the industry or farm profitability more generally.

It is interesting to note that all other classes of risk are represented in the top 11 ranked risks, indicating that producers recognise that there are a very wide range of factors that have a high risk of impacting negatively on the health of their stock.

All the identified risks that pertain to tuna health will be included in the risk analysis.

Appendix 8

SBT Aquaculture Industry Risk Register Survey Questionnaire

The establishment of a risk register is the first step in the newly funded Aquafin CRC southern bluefin tuna (SBT) risk assessment project. The partners involved, the TBOASA, TAFI and SARDI request that all stakeholders participate in compiling the risk register. The register should be based not only on scientific evidence but also include industry experience, personal opinions and anecdotal information. Without your involvement this will not happen!

Please indicate the level of risk which you believe is associated with each aspect of SBT culture. Write a number from 0 to 10 in the boxes provided to indicate the level of risk you believe is appropriate for assigning to the factor, where 0 is equivalent to no risk and 10 is the highest risk possible. You can assign the same risk level to a number of factors but each factor should have only one risk level, unless you differentiate one factor into more. For example, if you believe that one species of microalgae can be a greater risk than others, provide a risk level for all microalgae and then identify the microalgal species of concern and give this a risk level as well. Some of the risks are potential rather than present risks, for example what is the risk of SBT propagation to SBT health.

Add as much explanation and information to this survey as possible, for example if you perceive the culture of some other fish species more risky than others in near proximity to SBT aquaculture operations, include this information giving these species names (eg. snapper).

Thank you for your participation.

Company name:

Name of the person filling the form:

1) Environmental

- Microalgae
- Low dissolved oxygen (DO)
- Accumulation of farm wastes on seafloor
- Resuspension of sediments through net contact with the seafloor
- Resuspension of sediments through storm activity
- Sediments acting as a reservoir of disease/pests
- High H₂S levels in sediments
- High nutrients levels
- Predation
- Net fouling affecting water flow and dissolved oxygen levels
- Net fouling affecting pontoon flotation

Net fouling affecting the transparency of the structure to currents and waves and this making it more likely to be damaged, moorings to drag, etc

Net fouling increasing the occurrence of fish health issues by providing a substrate for disease/pests

Birds acting as a reservoir/host for disease/pests

Other
(Please describe)

2) Disease

- Bacterial pathogens
- Viral pathogens
- Swimmer syndrome.....
- Other parasites
- Other
- (Please describe)

3) Husbandry

- Feed - baitfish
- Feed - manufactured
- Propagation (hatchery)
- Other species culture
- Time in captivity
- Source of aquaculture SBT
- The tow experience - net damage, lack of feeding, sunburn
- Transfers - stress, net damage
- Other
- (Please describe)

4) Other (please list with the number reflecting risk)

Appendix 9

STAKEHOLDER FEEDBACK

Introduction

Risk assessment cannot be successfully conducted without stakeholder input. This includes information on practical aspects of tuna farming, providing unpublished industry information and stakeholder's assessment of risks and their significance. As risk assessment is a subjective process it does require continuous input from stakeholders to be useful. Stakeholder feedback was invited on a number of occasions:

- November 2001 – project proposal – industry meeting
- January 2002 – risk register – industry meeting
- May 2002 – meeting with consultants and industry, Adelaide
- July 2002 – presentation of risk assessment results at industry workshop in Port Lincoln
- August 2002 - Milestone report August 2002 contained full AusVet Animal Health Services report and input was invited from the tuna industry at this point
- September 2002 - Meeting with industry and FRDC during Aquafin CRC Conference
- October 2002 - After corrections based on comments from project investigators, this draft was reviewed and final draft provided for comments in October 2002
- November 2002 – Tuna health risk workshop Port Lincoln

Written comments from TBOASA were received on two occasions: an email (later sent as a letter) dated 1.10.2002 and a letter dated 6.01.2003. All comments and corrections, both verbal and written, were discussed further with the stakeholders and incorporated in this report.

Risk assessment

Risk assessment process was demonstrated to the industry at the tuna health risks workshop on 4 November 2002. The industry representatives present at the workshop were asked to use two step matrix risk evaluation method (exposure assessment and consequence assessment) and were provided with examples before the exercise. Each person could choose a hazard, which risk they wanted to estimate, they were asked for justification of their risk choices. Eight workshop participants completed the exercise. Four participants chose to assess risk of *Kudoa*, the overall risk ranged from very low (2 participants) to very high (one participant), the remaining participant ranked risk as high. Two participants chose to estimate risk for algal toxicosis, the results ranged from high to very high, one person assessed risk of hypoxia as very high and another as moderate. The exercise aimed to achieve feedback from the industry while demonstrating the subjective nature of risk assessment.

Research Priorities

Significant gaps in our knowledge of tuna health were identified during the literature review and risk assessment. This means that some of the risk estimates do not have enough information input and the likelihood and consequence of the risks may change as the information becomes available. In particular, there is very little known about normal cardiorespiratory physiology of tuna, effects of nutrition on health, potential effects of viral pathogens, effects of toxic substances, including harmful algal blooms and tuna parasitology.

Table 4. Research priorities as determined by vote at the tuna health risk workshop on 4 November 2002. Each person attending the conference had an equal vote, which could be either divided between the priorities or used to support one priority. The results are presented separately for industry and nonindustry workshop participants. Nonindustry stakeholders included researchers and government employees.

Research priorities	Industry Rank (%)	Nonindustry Rank (%)	Average rank (%)
Physiology	1 (35.00)	4 (13.33)	2 (24.2)
Nutrition	2 (22.50)	1 (35.83)	1 (29.2)
Virology	3 (16.25)	3 (16.67)	4 (16.5)
Toxicology	4 (13.75)	5 (8.33)	5 (11.0)
Parasitology	5 (12.50)	2 (25.83)	3 (19.2)

The most important research areas from the point of view of the tuna industry were tuna physiology and effect of nutrition on health, while nonindustry stakeholders saw effect of nutrition on health and tuna parasitology as the two main research areas.

Nonindustry stakeholders further divided nutrition research into research on manufactured diets effects on health and research on vitamin E effects on tuna health. These two areas ranked almost equal (Table 4 contains pooled results). Additionally, parasitology research needs were further divided into research on life cycles of parasites, diagnostic tests and other. Here, other parasitology research ranked first with 42%, parasite life cycles ranked second (35%) and diagnostic tests ranked third (23%). This may reflect the participants' lack of background as development of a diagnostic test would be needed to determine life cycles.

Only nonindustry stakeholders identified additional research priorities including:

- effects of environment on tuna health
- risks related to interannual variability and climate
- husbandry stress management
- availability of treatments

Surveillance Priorities

Surveillance priorities were discussed with the stakeholders at the tuna health risk workshop on 4 November 2002. The industry's preferences were distributed very evenly among general surveillance (3 votes), targeted surveillance (3 votes), microalgal toxicosis (3 votes), hypoxia (3 votes), development of centralised coordinated surveillance database (3 votes), blood fluke (2 votes), gill fluke (2 votes). However, nonindustry stakeholders saw general surveillance as a priority (11 votes), followed by general monitoring and information system (each 5 votes), targeted surveillance and blood fluke (each 4 votes), and microalgal toxicosis (2 votes).

Appendix 10

Presentations made in relation to this report

1. Tuna health risks. B. Nowak (SBT Industry Workshop, Port Lincoln 13 November 2001)
2. Tuna health risks project – risk register. B. Nowak (Port Lincoln SBT Industry Meeting, 24 January 2002)
3. Tuna health risks. B. Nowak (SBT Industry Workshop Port Lincoln, 29 July 2002)
4. Evaluation of health risks to the farmed southern bluefin tuna B. Nowak (International Symposium on Aquatic Animal Health, New Orleans, Louisiana, USA, 1-6 September 2002)
5. Aquafin CRC Health Program – research highlights. B. Nowak (Aquafest, Hobart, Tasmania, 22 September 2002)
6. Evaluation of health risks to the farmed southern bluefin tuna. B. Nowak (Aquafin CRC Conference, Hobart, Tasmania, 22-24 September 2002)
7. Evaluation of health risks to the farmed southern bluefin tuna. B. Nowak and A. Cameron (SBT Industry Workshop, Port Lincoln, 4 November 2002)