

Nutritional management of sea-caged Southern Bluefin Tuna (*Thunnus maccoyii*)

R.J. van Barneveld^{1,6}, A.R. Smart^{2,6}, S.M. Clarke^{5,6}, C.G. Carter^{3,6}, B.J. Davis¹, D.R. Tivey⁴ and J.D. Brooker⁴

¹SARDI-Pig and Poultry Production Institute, Nutrition Research Laboratory, The University of Adelaide, Roseworthy Campus, Roseworthy, SA 5371

²SARDI-Tuna Research, Lincoln Marine Science Centre, PO Box 2023 Port Lincoln, SA 5606

³University of Tasmania, Department of Aquaculture, Launceston Campus, PO Box 1214 Launceston, TAS 7250

⁴Department of Animal Science, University of Adelaide, Waite Agricultural Research Institute, Glen Osmond, SA 5061

⁵SARDI Aquatic Sciences Centre, PO Box 120 Henley Beach, SA 5022

⁶Co-operative Research Centre for Aquaculture, University of Technology, Sydney, PO Box 123 Broadway, NSW 2007

Summary

The farming of Southern Bluefin Tuna (SBT) in sea-cages in Boston Bay, South Australia, is one of Australia's fastest growing and most valuable aquaculture industries. The fish are fattened for sale on the Japanese sashimi market with the industry having the potential to produce 3 000 tonnes of tuna in 1996/97 worth about \$A90 million. At present, the industry relies heavily on the use of local and imported 'trash fish' such as pilchards, mackerel and herring for the nutrition of the sea-caged SBT. If the industry is to be sustainable, however, there is a need to reduce the nutritional reliance on trash fish. The development of a manufactured feeds for SBT is the highest industry priority. Research to date has quantified the potential for manufactured feeds to replace trash fish and significant advances have been made in the development of research methods necessary to assess the performance of SBT fed these feeds. Despite this, a manufactured feed that can promote SBT growth and flesh quality equivalent to pilchards still eludes the industry, and many questions pertaining to the nutritional management of sea-caged SBT are still to be answered.

Introduction

Pressure on wild stocks of fish, global population growth, and research suggesting a need to increase the proportion of seafood in western diets has resulted in the rapid development of many aquaculture systems. Csavas (1994) has suggested that seafood consumption will reach 84 million tonnes by the year 2000. This is a 17% increase since 1990. In contrast, the FAO (1994) estimate that global fisheries landings peaked in 1989 at 87 million tonnes and have since fluctuated near this level. This suggests that fisheries stocks are being harvested at close to their maximum sustainable yield. As a consequence, market demand for aquaculture products is expected to increase from 19.3 million tonnes,

worth \$US32.5 billion, in 1992 to about 22-24 million tonnes by the end of this century (Chamberlain and Rosenthal, 1995). In addition, the health benefits of consuming seafood in terms of reduced **cardio-vascular** disease are becoming widely documented and will result in an increase in the proportion of seafood in western diets. This will place further pressure on global seafood supplies and demands on aquaculture systems.

The farming of SBT has been a major success story in the expansion of the Australian aquaculture industry, although it has not been without its problems. The industry, started in 1990, produced approximately 2 000 tonnes of tuna with a market value of \$60 million in 1994/95 and has the potential to produce about 3 000 tonnes worth about \$90 million in 1996/97. The industry also has a significant economic multiplier effect because of its **labour** intensiveness and infrastructure requirements. It is significant because of the employment it has created in a regional area facing economic decline, and because of the impetus it has provided in South Australia for the development of associated service industries (infrastructure construction and maintenance, research and technical services, etc.) and of industry sectors (mussel farming and longer term holding of rock lobster).

Tuna farming has developed in the last few years as Australia's fastest growing aquaculture industry. Its initial success was due to the adoption of technologies from Japan. However, some of these technologies are not compatible with ecologically sustainable development. In particular the development of feeds that do not rely on imported frozen pilchards and that reduce the nutrient input into the water are seen as urgent priorities both by industry and government regulatory agencies.

Compared to more traditional animal production systems, there is great scope to improve our knowledge of the nutrition of many aquaculture species including SBT. Fish nutrition, however, presents many new challenges to the researcher due to the nature of the

industry, and the difficulty associated with conducting nutrition experiments under water.

The aims of this paper are to:

- Describe the development of SBT farming in South Australia;
- Outline the current commercial nutritional management of SBT;
- Discuss the development of manufactured feeds and nutrition management strategies for SBT;
- Suggest future research directions for SBT nutrition research.

Background to Southern Bluefin Tuna farming in South Australia

Description of Southern Bluefin Tuna

Southern Bluefin Tuna (*Thunnus maccoyii*) are one of 13 species of tuna in the Scombridae family. Close relations are butterfly mackerel and billfishes such as swordfish, marlins and spearfish. Its closest relative is the Northern Bluefin Tuna (*Thunnus thunnus*). The SBT is a large fish that can reach weights of up to 200 kg and lengths of up to two metres. They are pelagic living near the surface of the ocean. The lifespan of a SBT is in excess of 20 years, and they reach maturity at approximately 8 years.

Tuna are adapted to maximise feeding success in an environment where food is sparse and patchy, by being able to locate, capture and process food rapidly. Thus, tuna have very high energy demands associated with continuous swimming, gill ventilation and anaerobic swimming during feeding and an aerobic capacity that exceeds that of most other fishes (Korsmeyer et al. 1996).

Breeding occurs in the warm Indian Ocean, south of Indonesia. As the tuna grow, they move southwards towards major feeding grounds in the Southern Ocean. The tuna migrate from the spawning ground, around the Western Australian coast to the Great Australian Bight.

Development of the sea-caged Southern Bluefin Tuna industry

The sea-caged SBT industry began in response to the decrease in the quotas for ocean caught tuna. It became increasingly more difficult for the Port Lincoln fishing community to make a profit from canned ocean caught tuna sold on the domestic market. By enhancing product quality through supplementary feeding in sea cages, the Japanese sashimi market became a very profitable target while still allowing the tuna industry to work within the reduced quotas.

The tuna farming process begins with a catch of wild fish between December and February each year.

Commercial tuna fishing fleets track schools of tuna west of the Eyre Peninsula in South Australia. Once a suitable catch is located 'purse seines' are used for capture of the tuna destined for the sea-cages. Purse seines are nets that are towed around the school of tuna. The bottom of the net is then closed off, like a purse string, and in general, the tuna are then swum from the purse seine net through an opening into an attached 'Bridgestone' type towing cage. The tuna are towed slowly (1 to 2 knots) back to Boston Bay, Port Lincoln, with towing sometimes required for several hundred kilometres. Once in the bay, the tuna are swum from the towing cage into moored sea-cages being counted during the process using underwater video. Fish swim and feed normally through the towing process and enter the moored cages in a relaxed and healthy state. The non-handling of the tuna during the capture process contributes to the minimisation of stress and injury which occurs if the tuna are poled during capture.

The sea-cages used in the farming process are made of high density black polyethylene plastic (HDPE) usually with plastic-moulded stanchions. The diameter of commercial cages averages 40 metres. Two mesh nets are suspended from the floating pontoon. The inner net contains the fish, and the mesh size ranges from 60 – 90 mm. The outer net is a net to prevent access by predators, with a mesh size of 150 to 200 mm. The inner net drops approximately 10 metres, and the predator net falls to the sea floor, weighted by a chain (Evans, 1992). It is important that the nets have some fouling by marine growths, so that the SBT can clearly distinguish them and avoid entanglement.

It initially was planned that the fattening of tuna would occur over a period of six months. It soon became apparent, however, that marketable fish could be produced in three months, dependent on their initial stocking size. This, and earlier initial stocking dates, has led to the possibility of two farming cycles per year.

Current nutritional management of sea-caged Southern Bluefin Tuna

The current commercial nutritional management of sea-caged SBT is very basic. It essentially involves providing approximately \$20 million (15–20 000 tonnes) worth of 'trash fish' to about one hundred thousand sea-caged fish distributed across the commercial farms.

The 'trash fish' is predominantly pilchards but also includes jack mackerel, blue mackerel and herring. Approximately 50% of the 15–20 000 tonnes of trash fish used annually is from overseas. The smaller trash fish such as pilchards are fed whole while larger species are chopped prior to feeding. In some cases, the trash fish have been coated with vitamin and mineral premixes in an attempt to maximise the performance of the SBT. The benefits of this practice are hard to quantify, however, and as most of the vitamins and minerals are washed from the pilchards prior to consumption by the

tuna, it is unlikely that this supplementation strategy is cost-effective.

In some instances, **frozen** trash fish are thawed prior to being manually shovelled into the pens. Other delivery methods have been tried with the most recent and common practice being the provision of frozen blocks of pilchards into mesh floating containers that deliver feed as they **defrost**.

The industry recognises a number of issues associated with their current feeding methods that could affect their sustainability. These include:

- The natural stocks of the trash fish are limited in Australia and shipments of such fish from overseas imposes a quarantine risk;
- International supplies of pilchards are variable in volume and quality (Japanese supplies have, for example, declined markedly and the fat content of pilchards used in feeds varies from 1–22%);
- As the industry develops and operating costs need to be reduced, it will become increasingly important to reduce feed costs through **mechanisation**.
- A trash fish diet may not allow the tuna to grow to their full potential.
- Poor utilisation of the trash fish diet by SBT may be resulting in significant amounts of waste nutrients being released into Boston Bay.

In addition, the current feeding practices are costly, **labour** intensive and inefficient in terms of feed conversion and wastage. As a consequence the development of a manufactured feed and alternative feeding strategies has been deemed the highest industry priority. It is perceived that:

- Manufactured feed can be better matched to the nutritional requirements of farmed tuna thereby enhancing growth and fish health, which translates into increased farm production levels;
- Manufactured feed will provide the potential for improved product quality (in particular fat content, colour and texture) as they are more stable in storage than trash fish and can be altered to better meet the requirements of fish farming and the markets;
- Manufactured feed will reduce industry feeding costs as its generally lower moisture content and promotion of better feed conversion ratio will reduce the quantities required and also, therefore, costs associated with feed storage and transport;
- Manufactured feed will greatly reduce environmental concerns associated with the present use of trash fish, including reducing the overall requirement for pilchards, minimising risks

of importing and dispersing undesirable diseases and pests, and reducing organic wastes in the **farm** environment which can harbour and promote diseases as well as detrimentally affect water quality.

- Development of a manufactured feed will allow the selection and incorporation of more cost-effective feed ingredients and a reduction in the quantity of **fresh** fish, fish meal and fish oils in the feed, supplies of which are rapidly diminishing.

The economic benefits of the development of a suitable manufactured feed has been estimated to be as high as \$9.5 million/annum to the Tuna Boat Owners of **Australia** and \$5 million/annum to successful feed manufacturers. Additional economic benefits would be expected to flow **from** ongoing research leading to further enhancement of these feeds.

Development of manufactured feeds and nutrition management strategies for sea-caged Southern Bluefin Tuna

The process of developing a suitable manufactured **feed** and alternative feeding strategies for SBT has been difficult. One of the main reasons for this is that all research conducted with these fish is pioneering and every research technique must be developed before valuable results can be obtained. In addition, we have a poor knowledge of the behaviour of the SBT in a caged environment and it takes time to understand their physical needs.

Special considerations for nutrition research with sea-caged Southern Bluefin Tuna

Compared to traditional nutrition research with terrestrial animal species, there are many physical impediments to conducting nutrition research with SBT. These include:

- The ability to maintain an experimental diet underwater with minimal nutrient loss;
- Difficulties associated with measuring growth or nutritional parameters (e.g. little potential for routine faeces or **digesta** collection).
- The high value of the experimental fish;
- The inability to maintain a constant experimental environment;
- High accommodation costs resulting in a limited ability to replicate experimental treatments;
- Difficulties associated with getting SBT to accept experimental diets.

A nutritional parameter that requires special consideration in aquaculture systems is feed conversion ratio (FCR). Difficulties associated with measuring growth rate, collection of wasted feed and associated leaching will all contribute to inaccurate estimates of FCR. When interpreting results from aquaculture systems, one must ask whether FCR is an appropriate measure of fish performance during nutritional studies.

The high value of experimental animals can limit the number used in experiments, and the ability to conduct flesh analysis as part of nutritional studies. Farmed SBT can attract more than \$A54 per kilogram on the Japanese sashimi market.

The effects of environment on the nutrient requirements of terrestrial production animals is well established. In addition, when conducting nutrition experiments with terrestrial species, the environment can often be closely controlled. This is far more difficult when conducting nutrition experiments with SBT in an ocean environment.

Digestive physiology of Southern Bluefin Tuna

When developing a manufactured feed and feeding strategy, a knowledge of the digestive physiology and natural feeding habits of the target species is a logical starting point.

Digestive anatomy

A detailed investigation into the comparative anatomy and systemics of the tunas (genus *Thunnus*) was completed by Gibbs and Collette (1966). The oesophagus merges indistinguishably into the stomach which forms a blind sac posteriorly (Figure 1). The intestine rises from the anterior end of the stomach, and a very large caecal mass is attached to its origin by several ducts that are not externally apparent. The intestine proceeds caudad for half or more the length of the body cavity (straight intestine), forms a loop, runs cranial (ascending portion) almost to the pylorus, then forms another loop and continues in a nearly straight line (descending portion) to the anus. The spleen is located between the straight and ascending portions of the intestine. The gall bladder is a long, tubular sac rising from the right lobe of the liver, attached to the dorsal wall of the left side of the straight intestine (Figure 1).

The specific anatomy of the SBT has a number of nutritional implications. The blind stomach has a massive capacity. Individuals are easily able to consume 10% of their body weight in pilchards in one feeding period. This may indicate an ability to feed infrequently while still maintaining a constant flow of digesta, and hence nutrient absorption, through the intestine. The lower intestine of the SBT is very short, with little difference in the morphology of the intestinal segments defined above. The functions of the whole SBT intestine

are likely to resemble the small intestine of terrestrial animals. Anatomically, there are few digestive sites that would be suited to the fermentation of high fibre diets.

Digestive enzymes

Homogenates of pyloric caeca have been tested at the University of Adelaide for pancreatic proteolytic enzyme profiles (D.Tivey *et al.* unpublished data). The presence of major proteolytic enzymes, similar to those found in terrestrial animals, has been observed. In particular, a high activity of a dipeptidyl peptidase IV-like enzyme has been demonstrated. It appears that diet type (pilchards vs manufactured feed) has little effect on the resident proteolytic enzymes in the pyloric caeca.

The results of Tivey *et al.* (unpublished data) indicate that the expression of enzymes in the digestive tract is not rate limiting to protein digestion *in vivo*. Factors which may influence the digestion of protein include protein type, differences in the mechanical processing of the diet prior to feeding, the residence time of the food in the pyloric caeca, the rate of pancreatic secretion in response to food in the digestive tract and the effect of microenvironment (mucin type and quantity) of the pyloric caeca on luminal enzyme activities and absorption of digestion end products.

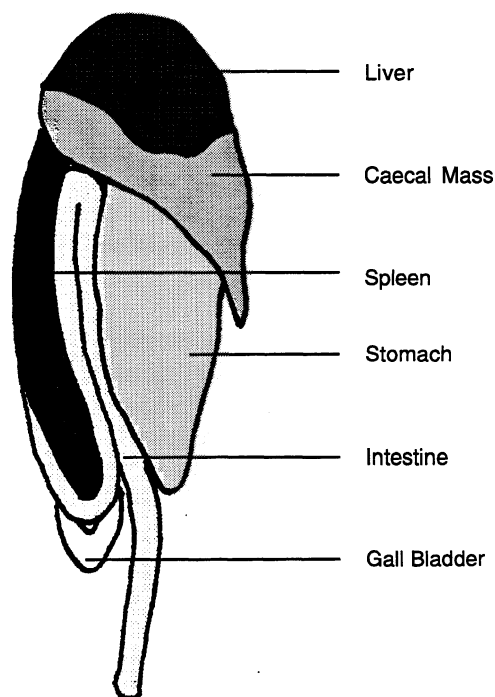


Figure 1 Schematic representation of the digestive tract of a Southern Bluefin Tuna (*Thunnus maccoyii*)

Digestive microbiology

The bacterial population of the digestive tract of SBT has been isolated and characterised at the University of Adelaide by J. Brooker et al. (unpublished data). Up to 10^8 bacteria/ml were detected in samples collected from the pyloric caeca and intestine. There was a diverse range of microorganisms present and these appeared to be a resident population in the digestive tract rather than derived from the feed. The populations differed depending on whether pilchards or manufactured diets were fed, but were highest in the pyloric caeca.

Brooker et al. (unpublished data) found that bacteria isolated from the pyloric caeca were mainly proteolytic with a small number being amylolytic as well. In contrast, many bacterial isolates from the intestine were either lipolytic or proteolytic and lipolytic. This suggests that there is a separation of functions in the digestive tract with bacteria in the intestine contributing to the digestion of lipid in the diet. Lipase expressing bacteria were more prominent when SBT were fed pilchards compared with those fed a manufactured feed.

Scanning electron microscopy has revealed different modes of action of the microbial populations in the digestive tract of SBT (Brooker et al. unpublished data). The pyloric caeca contains populations that are mainly attached to small particles of feed in the lumen. In contrast, the intestine contains microbial populations that appear to be attached to the intestinal wall. Bacterial attachment to feed in the pyloric caeca may have a role in the ability of SBT to digest fibrous feed ingredients.

What are the nutritional requirements of Southern Bluefin Tuna?

The initial strategy for the development of a manufactured feed for SBT was to match the diet specifications to the requirements of the SBT with the aim of reducing feed costs, minimising environmental pollution, ensuring even growth rates, maximising carcass quality and exploiting desirable carcass traits whilst allowing the selection of the most cost-effective ingredients. Unfortunately, a number of these objectives are conflicting.

Unlike most terrestrial and aquaculture production systems, the optimum farmed SBT carcass contains a significantly higher level of intramuscular fat compared to wild caught fish. Other systems specifically target a reduction in carcass fat. Deposition of fat is an inefficient process. It takes approximately five times as much energy to deposit a gram of fat compared to a gram of lean meat (van Barneveld et al. 1997). In addition, carcass fat deposition is increased when the diet specifications do not match the requirements of the fish for optimum growth. Excesses of protein, amino acids and energy or an imbalance of these respective nutrients can result in increased fat production. Hence, to maximise carcass quality traits, it is unlikely that feeding efficiency and growth rates will be maximised and

environmental pollution may be higher than if the diets were tailored for maximum lean growth. For this reason, it is difficult to define a basis for diet formulation for SBT.

As there is no existing information on the nutritional requirements of SBT, best estimates have been made to facilitate diet formulations based on the requirements of other pelagic fish. These estimates have been used to ensure that there is no deficiency of any one nutrient, rather than to accurately match diet specifications to nutrient requirements. We must also consider that diet form appears to have a major impact on the nutritional value of manufactured feeds, and we have no knowledge of the sensitivity of caged SBT to changes in the protein and energy content of their diet. For this reason, highly tuned diet specifications may not result in improved performance of the fish. In addition, due to the difficulties associated with conducting nutrition research with SBT, experiments to specifically define requirements for certain nutrients are unlikely in the near future.

An 'ideal' dietary amino acid profile has been defined from the amino acid balance of red and white muscle in wild tuna (van Barneveld, 1996; Table 1). Comparison of this profile with the balance used in diet formulations for SBT prior to 1997 indicates that there are excesses of methionine, valine, leucine, phenylalanine, and arginine while there is a notable deficiency of histidine. If diets formulated for SBT contain high levels of protein, the amino acid balance is likely to have little relevance due to the large amount of amino acids that will be wasted and deaminated. As the protein content of these diets is reduced, however, and the quantity of fresh fish and fish meal is replaced through the use of alternative protein sources, the ideal amino acid profile will have greater relevance.

Progress in the development of a manufactured diet for sea-caged Southern Bluefin Tuna.

Four large-scale feeding experiments have been conducted with sea-caged SBT since 1994 through the Cooperative Research Centre for Aquaculture. Due to the time of harvest and the duration of experiments, only one large scale feeding experiment is possible per year. Smaller, more intensive experiments such as digestibility and transit time studies are conducted following the large scale growth experiments using the remaining fish.

Feeding and growth experiment 1(1994)

Southern Bluefin Tuna are extremely selective about the form of a manufactured feed they will accept. They appear sensitive to shape, colour, texture, odour, moisture content, oil content and fish or fish meal content. As a consequence, the number of potential diet presentation options and diet ingredients is limited.

The acceptance of semi-moist 'sausage-type' pellets and extruded pellets was compared with pilchards using SBT held in three cages anchored in Boston Bay, SA (van Bameveld *et al.* 1995). This experiment was also used to refine experimental methodology for the handling, tagging and performance monitoring of SBT in research cages.

The tuna could be handled, measured and tagged with only 10% mortality. Tuna would not consume extruded pellets (>75% DM) but slowly accepted semi-moist 'sausage-type' pellets (52–70% DM). An extensive weaning period of 42 days was required before the SBT would readily consume the pellets resulting in substantial weight loss. Feed conversion ratios (FCR) of 7:1 were recorded for SBT consuming pellets compared to 13:1 by those fed pilchards. The growth and meat quality of SBT fed pellets appeared comparable to those fed pilchards. Capture, weighing, tagging and transfer of fish was shown to have a negative impact on the performance of SBT, and hence care must be taken when interpreting these results for use in a commercial environment.

The results suggested that there was potential for manufactured feeds to be offered as a semi-moist 'sausage-type' pellet to replace pilchards as the diet for caged SBT. In addition, the long time taken for the fish in this experiment to take the pellets as their sole diet may have been due to initial low water temperatures and the fact that the SBT had previously been fed pilchards for prolonged periods.

Feeding and growth experiment 2 (1995)

The second feeding and growth experiment evaluated the time to wean recently caught SBT held in warmer waters onto manufactured feeds and compared the growth performance of SBT fed pilchards or manufactured diets (van Barneveld *et al.* 1995). A diet of known composition was formulated to contain a variety of animal proteins, fish proteins and cereals and to meet the estimated requirements of SBT. During manufacture, however, this diet was significantly extended with water and flour to facilitate processing into sausage skins. The experiment used six cages containing SBT allocated following tagging, and measurements of weight and length. The SBT were fed to satiety and the weight of feed added to each cage was recorded daily. After 105 d, the SBT were harvested, identified and re-weighed. Tuna mortality was high initially in cages with unfouled nets and through handling when the fish were in poor condition. In addition, poaching resulted in significant losses of fish from some cages and made the interpretation of the results difficult (determination of FCR etc.).

The SBT were successfully weaned onto artificial diets within 14 days. SBT fed the manufactured feed had lower weight gains and higher feed conversion ratios than SBT fed pilchards (Table 2). The variability associated with the performance of SBT fed the manufactured feed was also higher than those fed pilchards. There was no consistent relationship between

Table 1 Essential amino acid content of red and white muscle in wild SBT (g/kg, dry matter), dietary amino acid balance (used prior to 1997) and proposed ideal amino acid balance (from van Bameveld, 1996).

Amino acid	Red muscle	White muscle	Dietary balance ¹	Ideal balance ¹
Methionine	11.0	10.9	49	35
Threonine	18.2	17.2	56	56
Valine	20.4	19.2	74	63
Isoleucine	18.7	17.1	61	57
Leucine	29.3	27.6	110	90
Phenylalanine	15.1	14.9	63	47
Lysine	32.5	30.7	100	100
Histidine	30.5	30.3	37	96
Arginine	23.9	22.3	93	73

¹Balance on a DM basis.

Table 2 Growth performance of SBT fed pilchards and a manufactured feed (from van Bameveld *et al.* 1995).

Cage	Pilchards		Manufactured feed	
	1	2	1	2
Number of SBT	15	5	16	15
Average final weight (kg)	28.4	37.9	22.0	30.0
Average length (cm)	106.4	117.1	102.0	111.1
Average weight gain (kg)	6.8	8.7	0.9	4.0
Average daily gain (g)	65	83	9	38
Feed conversion ratio	26	23	110	27

stocking density and the growth performance of the SBT. The growth response from one cage of SBT fed the manufactured feed suggests that there is a high potential for a manufactured feed to replace pilchards in this aquaculture system. The dilution of nutrients in this diet during pelleting may be one reason for the lower than anticipated performance.

Results from the above experiments had significant outcomes in terms of improving experimental methods and **identifying** gaps in our knowledge. In particular, the effects of nutritional history on the performance of SBT on manufactured feeds, the role of attractants and flavour enhancers in manufactured feeds, the nutritive value of alternative protein sources for SBT, and the specific nutrient requirements of SBT are all areas where our knowledge is limited.

Feeding and growth experiment 3 (1996)

The use of sausage skins was a slow and inefficient way of making manufactured feeds for SBT. The aim of the third large-scale feeding and growth experiment was to assess the value of a spray-coated pellet formulated to meet the nutritional requirements of SBT and to accommodate water and binder addition at the time of manufacture. This experiment was also designed to achieve a higher level of replication than previous experiments using three cages per experimental treatment.

The formulated feed contained a combination of Chilean fish meal, blood meal, Antarctic krill meal, DL-methionine, squid oil, Jack Mackerel oil, wheat gluten, pre-gelled starch, water, vitamins, minerals and squid flavouring. The pellet was well accepted after a short period of introduction, but growth performance of the SBT on this diet was not determined due to sudden SBT deaths that occurred across the whole industry in April, 1996 following a storm. A report of this incident (Clarke, 1996) concluded that the storm stirred sediment on the bay floor resulting in gill irritation, subsequent mucous production and asphyxiation of the SBT. The research **farm** lost 75% of its fish while the commercial farms lost in the vicinity of \$50 million worth of fish. The majority of commercial **farms** have now been moved to deeper waters outside the bay.

Feeding and growth experiment 4 (1997)

The previous growth experiments showed some potential for manufactured feeds, but growth performance of the SBT was always substantially lower than pilchard-fed fish. Results **from** morphology studies and digestibility experiments in conjunction with the growth experiments suggested that ingredient particle size and diet form may play an important role in the utilisation of nutrients (Davis, 1997). It appears that when diets are offered in sausage skins or spray-coated pellets **they** rapidly break down in the stomach. Due to the small particle size of the ingredients, they are quickly passed from the stomach into the pyloric **caeca** and

intestine. Rapid transit time of the small particles through the short intestine results in poor utilisation. In contrast, pilchards and well bound diets are slowly broken down in the stomach, and the constant flow of small quantities of nutrients through the intestine results in better absorption and subsequent utilisation. Further evidence of this phenomenon is demonstrated when SBT are fed whole pilchards or pilchards chopped and included in sausage skins. Despite **an** identical nutritional value, SBT fed pilchards in skins had poorer growth performance than fish fed whole pilchards (Davis, 1997).

The objectives of this experiment were to compare the growth performance of SBT fed one of the following feed forms. The feed forms were:

- Pilchards;
- Cooperative Research Centre (CRC) for Aquaculture Mash: 'Mash' feeds are defined as feeds that contain a proportion of **fresh** trash fish in combination with a concentrate. The advantage of this feed is that acceptance is still high, while the overall requirement for trash fish is reduced;
- Northern Bluefin Tuna Mash: This diet was based on fresh trash fish and a concentrate of unknown composition that has been imported **from** Japan where it has been used successfully with Northern **Bluefin** Tuna;
- Extruded pellet: This is a high quality, low moisture feed form that has been proven for use in other aquaculture systems. It is highly flexible, hygienic and extruders can produce large quantities of feed quickly and efficiently. It could only be produced in limited quantities due to the remote location of the equipment and there was insufficient to assess growth performance;
- Two commercial diet preparations: Composition of these diets was unknown.

All feeds were well accepted **after** a short introduction period. To enhance acceptance, SBT were fed an alginate bound pellet **from** the time of capture and during towing into Boston Bay.

The growth response with a manufactured feed was the best so far achieved (Table 3). The CRC Aquaculture Mash promoted mean growth rates that were only 45% of those achieved with pilchards, however, the variation surrounding the growth of the fish fed manufactured feeds was significantly less than that observed on pilchards. It appears that exceptional growth of one fish fed pilchards has greatly increased the mean growth rate of this treatment. The condition of fish fed the CRC Aquaculture Mash was similar to those fed pilchards. The growth rate and condition of fish fed the Northern Bluefin Tuna Mash and proprietary manufactured feeds was poor in comparison with pilchards and the CRC Aquaculture Mash, and in some instances, the fish lost weight and condition (Table 3).

The CRC Aquaculture Mash was based on pilchards, Chilean fish meal, krill meal, free amino acids, squid oil, vitamins, minerals, anti-oxidants and water. On an 'as-fed' basis, the CRC Aquaculture Mash had a higher nutrient density than pilchards (Table 4), but despite a slightly reduced feed intake, was unable to support growth rates similar to those observed with pilchards.

Evaluating feed ingredients for use in manufactured diets

If a suitable trash fish replacement is to be found for use in SBT manufactured feeds, a number of other protein sources (animal proteins, grain legumes, cereals) need to be evaluated. A knowledge of the nutritional

value of feed ingredients other than trash fish and fish meals will allow us to cost-effectively replace these protein sources while maintaining the required balance of amino acids and lipids.

Experiments have been made to assess the *in vivo* digestibility and transit time of feed ingredients, manufactured feeds and pilchards fed to SBT (Davis, 1997; van Bameveld *et al.* unpublished data). The digestibility of dry matter, nitrogen and energy along the digestive tract has been determined for Peruvian fish meal fed to SBT in 'sausage skins' (Table 5). Negative digestibilities for nitrogen were calculated in the stomach and pyloric caeca. This is likely to be due to large contributions of endogenous N in these regions. Endogenous N contributions are also likely to have resulted in an underestimate of the nitrogen digestibility in the distal intestine.

Table 3 Mean growth performance (and standard deviation) of Southern Bluefin Tuna fed pilchards and manufactured feeds with varvina forms.

	Pilchards		CRC Mash		NBT Mash		Man A	Man B
	Cage 1	Cage 2	Cage 1	Cage 2	Cage 1	Cage 2		
Fish sampled	9	8	9	6	10	10	10	10
Weight gain (kg)	12.95 (10.18)	7.46 (1.89)	5.62 (1.60)	3.68 (1.29)	1.33 (1.00)	1.59 (0.53)	-0.85 (1.09)	0.44 (1.07)
Length gain (cm)	5.39 (2.07)	5.69 (1.87)	4.39 (2.41)	3.92 (1.16)	2.05 (1.91)	1.85 (1.45)	1.20 (0.82)	1.30 (1.46)
Condition index*	24.42 (0.93)	23.50 (0.84)	22.55 (1.26)	21.35 (0.70)	18.84 (1.46)	19.87 (1.06)	17.40 (1.43)	18.89 (1.09)
Condition index gain	11.83 (12.40)	4.77 (1.44)	3.21 (1.52)	2.04 (1.81)	0.57 (1.66)	0.85 (1.01)	-1.85 (1.61)	1.29 (1.62)

* Condition index is a subjective measurement based on the weight and length of the fish.

Man A, Commercially manufactured diet A; Man B, Commercially manufactured diet B; NBT, Northern Bluefin Tuna; CRC, Cooperative Research Centre.

Table 4 Composition of CRC Aquaculture Mash and pilchards (g/kg, as-fed) fed to Southern Bluefin Tuna during the 1997 large-scale feeding and growth experiment in Port Lincoln, SA.

Nutrient	CRC Aquaculture Mash	Pilchards
Crude protein	363.00	210.00
Dry matter	615.00	340.00
Fat	152.00	92.00
Lysine	27.32	7.60
Threonine	15.61	3.54
Methionine	10.68	1.30
Isoleucine	31.68	34.99
Leucine	28.73	4.00
Valine	19.61	3.75
Phenylalanine	17.72	3.84
Histidine	10.49	4.48
Arginine	26.05	6.43

A range of problems was encountered with experiments conducted to determine the digestibility of nutrients in the distal intestine of SBT fed manufactured feeds and pilchards. Poor mixing and a wide range in ingredient particle size is thought to have resulted in negative amino acid digestibility estimates being obtained in this experiment. **Digesta** transit time was assessed in SBT fed manufactured feeds and pilchards using a marker dilution technique (Davis, 1997). The manufactured diet was estimated to have a transit time of 5.3 hours. Sampling of the fish fed pilchards was not long enough to dilute the marker to 80% of the equilibrium concentration and hence an exact transit time could not be assessed, yet it was longer than that observed with the manufactured feed. The significant difference in transit times for manufactured feeds compared to pilchards may help explain the poorer utilisation of the former.

Future nutrition research directions

Having identified a manufactured feed that is accepted by the SBT as well as promoting about 70% of the growth and condition achieved by using a trash fish diet, the opportunity exists to use this feed as a research tool and enhance our knowledge of SBT nutrition. The following research program is proposed for the SBT research farm in Boston Bay, SA from July 1997.

- Utilise the CRC Aquaculture Mash feed to assess the ability of caged SBT to respond to changes in the protein and moisture content of their diet;
- Assess the potential of natural colour enhancers, such as astaxanthans, in manufactured feeds;
- Assess the influence of **frequency** of feeding on the growth performance of SBT. In the short term, the ability to feed less frequently while maintaining performance represents massive cost savings to the tuna farms.

Table 5 Dry matter, nitrogen and energy digestibility in the stomach, pyloric caeca, proximal intestine and distal intestine, in a Peruvian fish meal based diet fed to caged Southern Bluefin Tuna, determined 2.5 and 5.0 hours after feeding.

	Collection time (hours)		Statistics		
	2.5	5.0	SEM (2.5h)	SEM (5.0h)	Time
Dry matter digestibility					
Stomach	0.00	0.06	0.021	0.021	NS
Pyloric caeca	-0.08	-0.24	0.024	0.029	**
Proximal intestine	0.17	0.06	0.065	0.088	NS
Distal intestine	0.42	0.35	0.048	0.048	NS
Region*Time	NS				
Region	***				
SEM	0.044				
Nitrogen digestibility					
Stomach	-0.13	-0.07	0.020	0.019	NS
Pyloric caeca	0.02	-0.05	0.076	0.082	NS
Proximal intestine	0.11	-0.01	0.062	0.077	NS
Distal intestine	0.38	0.33	0.062	0.059	NS
Region*Time	NS				
Region	***				
SEM	0.047				
Energy digestibility					
Stomach	0.12	0.20	0.023	0.022	*
Pyloric caeca	0.21	0.19	0.068	0.073	NS
Proximal intestine	0.41	0.29	0.049	0.060	NS
Distal intestine	0.62	0.59	0.041	0.039	NS
Region*Time	NS				
Region	***				
SEM	0.039				

NS, not significant; * P<0.05; *** P<0.001; SEM, standard error of the mean (after Davis, 1997).

- Develop suitable extruded feed **forms** and trial these forms on a large scale;
- Assess the potential to reduce the quantity of fresh fish, fish meal and fish oils in manufactured feeds.

These experiments will be run in conjunction with experiments aimed at modelling the energetic expenditure of SBT in sea-cages, improving the flesh quality of SBT by pre- and post-harvest handling and storage techniques and the identification of growth correlates to improve the efficiency of research experiments.

Conclusions

A manufactured feed that can promote growth and flesh quality characteristics in SBT similar to that currently achieved with trash fish is likely to be developed soon. Research conducted over the past 4 years has revealed some potential for manufactured feeds in this aquaculture system but progress has been slow due to wide ranging difficulties associated with researching a large, valuable, pelagic species. Enhanced research techniques and diet manufacturing technology (e.g. extrusion) combined with a collaborative research program will greatly assist development of a manufactured feed in the future. Together with specific feeding strategies, advanced nutritional management of SBT will reduce the dependence on trash fish, ensuring the viability of this industry in the long term.

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